Wheatbelt Orchid Rescue Project

Final Report 8

Translocation of Orchids in Wheatbelt Nature Reserves

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Wheatbelt Orchid Rescue
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Our environment, our future
The University of Western Australia
Friends of Kings Park
W.A. Native Orchid Study & Conservation Group

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Wheatbelt Orchid Rescue Project Final Reports

Brundrett M. 2011a. Wheatbelt Orchid Rescue Project Final Report 1. Objectives, Outcomes and Overall Conclusions. Wheatbelt Orchid Rescue Project, University of Western Australia. *Link 1*

Brundrett M. 2011b. Wheatbelt Orchid Rescue Project Final Report 2. Population Size and Vital Statistics Data for the Granite Spider Orchid (*Caladenia graniticola*). Wheatbelt Orchid Rescue Project, University of Western Australia. *Link 2*

Brundrett M. 2011c. Wheatbelt Orchid Rescue Project Final Report 3. Population Size and Vital Statistics Data for the Ballerina Orchid (*Caladenia melanema*). Wheatbelt Orchid Rescue Project, University of Western Australia. *Link 3*

Brundrett M. 2011d. Wheatbelt Orchid Rescue Project Final Report 4. Population Size and Vital Statistics Data for the William's Spider Orchid (*Caladenia williamsiae*). Wheatbelt Orchid Rescue Project, University of Western Australia. *Link 4*

Brundrett M. 2011e. Wheatbelt Orchid Rescue Project Final Report 5. Population Size and Vital Statistics Data for the lonely Hammer Orchid (*Drakaea isolata*). Wheatbelt Orchid Rescue Project, University of Western Australia. *Link 5*

Brundrett M. 2011f. Wheatbelt Orchid Rescue Project Final Report 6. Population Survey Data for Southern Populations of the Western Underground Orchid (*Rhizanthella gardneri*). Wheatbelt Orchid Rescue Project, University of Western Australia. *Link 6*

Brundrett M and Ager E. 2011. Wheatbelt Orchid Rescue Project Final Report 7. Seed Collecting, Soil Baiting and Propagation of Orchids. Wheatbelt Orchid Rescue Project, University of Western Australia. *Link 7*

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Brundrett M. 2011. *Wheatbelt Orchid Rescue Project: Case Studies of Collaborative Orchid Conservation in Western Australia.* University of Western Australia, Crawley, Western Australia.

Note: Appendix 1 contains location data for Declared Rare Flora that is not included in publicly available versions of this report.

1. Introduction and Objectives

The Wheatbelt Orchid Rescue (WOR) project is a Lotterywest funded collaboration between the Western Australian Native Orchid Study and Conservation Group (WANOSCG), the School of Plant Biology at the University of Western Australia (UWA), the Friends of Kings Park and the Department of Environment and Conservation (DEC). This project aims to help conserve Critically Endangered orchids in the Western Australian Wheatbelt by obtaining knowledge required for sustainable management and directly contributing to recovery actions. Please refer to WOR Report 1 for overall objectives and outcomes of the project.

This report primarily concerns translocation of three rare orchids, the ballerina orchid (*Caladenia melanema*), the granite spider orchid (*Caladenia graniticola*) and William's spider orchid (*Caladenia williamsiae*). These species were chosen for translocation following a comprehensive investigation of their ecology and biology presented in WOR Reports 2-4. These reports also provided detailed information on the status of orchid populations required to support the translocation approval process.

The overall objective of WOR project translocations was to reduce the risk of extinction for 3 endangered *Caladenia* species by increasing the size and extent of local populations using seedlings propagated with local provenance seed and fungi. A secondary aim of translocations was to gain knowledge required for future translocations by investigate the comparative effectiveness and efficiency of different methods for orchid propagation and outplanting. Specific objectives of this experiment translocation trial are to:

1. Develop successful translocation protocols for terrestrial orchids for use in relatively arid sites in the eastern wheatbelt.
2. Compare existing methods of orchid propagation and establishment for effectiveness in the field and glasshouse.
3. Augment existing populations by increasing the area of occupied habitat and number of orchids.
4. Compare the reproductive potential of rare species with co-occurring common orchids.
5. Investigate the role of mycorrhizal fungi as factors limiting orchid recruitment in harsh habitats.
6. Provide data on outcomes and recommend future recovery actions and translocation methodologies to land managers.

2. Materials and Methods

2.1.Approvals Process

Collection of seed and mycorrhizal fungi was covered by a Permit to Take Rare Flora issued to Dr Mark Brundrett, School of Plant Biology (Permit No. 203). The stated purpose of this permit was for "research into evolutionary biology, soil fungi, and pollination requirements to assist in recovery actions". No whole plants were taken under this permit. The author also had a licence to "take flora for scientific purposes within DEC lands" and flora collection Licence for "scientific and other prescribed purposes".

Three draft translocation proposals (written by WOR and DEC) were submitted to DEC in April 2009 (Brundrett and Edgley 2009abc). After a lengthy period of consultation, comments, reviews and revisions the final version of the translocation proposals were approved in late July 2009 (Table 1). Translocation trials were established in August 2009 and July 2010.

2.2. Species and Site Selection

Three of the most threatened orchids that occur in the West Australian wheatbelt were chosen for translocation trials in early 2009. These were (i) the ballerina orchid (*Caladenia melanema*) that occurs near Pingrup, (ii) Dragon Rocks Nature Reserve populations of the granite spider orchid (*Caladenia graniticola*) and (iii) the William's spider orchid (*Caladenia williamsiae*) near Brookton. For each species, two translocation sites were selected in suitable habitat that was currently unoccupied with a fairly dense canopy and sparse understorey so competition from other species was minimal. These sites were 100-500 m distant from known existing plants of the same species. Habitat vegetation for *C. graniticola* consisted of tall *Allocasuarina* woodland adjacent to granite rocks. For *C. williamsiae*, translocation sites were in lateritic shrubland (heathland). Habitats for *C. melanema* are tall shrublands of *Melaleuca lateriflora* near salt lakes.

Table 1. Timing of WOR fieldwork and approvals processes required for translocations.

Tasks and locations	Dates
Seed collection	Nov. 23-24, 2007
Seed collection	Dec. 2-3, 2008
Propagation of orchids	Feb.-July 2009
Preliminary status reports submitted for 5 species	April 2009
Translocation proposals submitted for 3 species	April 30 2009
Reviewing and revision of translocation proposals	May-July 2009
Approval of translocation proposals granted	July 21
Translocation trials established near Brookton, Pingrup and Hyden	Aug. 6-7, 2009
Seed collection	Oct. 26 2009
Seed and soil collection	March 17, 2010
Propagation of orchids	Feb.-June 2010
Translocation trials established near Hyden	July 19-20, 2010
Soil collection for glasshouse trials	July 19-21, 2010
Seed collection	Oct. 13, Dec. 13, 2010

2.3. Translocations

Seed and soil collecting is described in WOR Report 7. Seed was sourced from naturally pollinated plants using multiple parents. Seedlings were raised by a modified seed baiting technique in pasteurised potting mix inoculated with organic material from the natural habitat of each orchid, with the exception of *C. williamsiae*, which grew best in non-symbiotic sterile culture (see WOR Report 7). Soils used to raise seedlings consisted of pasteurised potting mix amended with fungi contained in organic matter sourced from the same site where translocations later occurred and therefore can be considered free of pests and diseases. Plants were raised from seed by germination in an incubator at 21°C in the glasshouse facilities area of the University of Western Australia. For each orchid, 28 seedlings were planted in total at 4 plots at 2 locations in 2009, as shown in Table 2.

Outplanting trials for orchid seedlings differed considerably from methods used in Perth (e.g. Batty et al. 2001, Scade et al. 2006) due to the relatively dry environments were orchids were translocated in the wheatbelt. For both experiments, seedlings were protected by 10 x 10 cm semi-permeable translocation pouches developed for the WOR project by the author (MB). These pouches protect seedlings in a humid environment to reduce the impact of desiccation and also reduce grazing by invertebrates (WOR Report 7). These pouches also reduced the need for seedlings to be acclimatised before translocation and provided room for tuber formation (see Fig. 1). Each pouch initially contained 7 seedlings spaced 1 cm apart (Figs. 3,4). The aboveground and belowground development of individual seedlings was photographed and measured through transparent pouches (see Figs. 2-4).

Relatively large seedlings of orchids (with a 1-2 cm long leaf) were used. Each rare orchid species had a common orchid planted at the same locations for comparison (Table 3). There were 2 small subplots a few m apart at 4 locations for each species (Table 3). Seedlings were protected from grazing by larger animals under small cages as shown in Figure 1.

Supplemental watering was provided as often as possible (every 2 weeks), but rainfall was much lower than normal in 2009 and 2010. There was a substantial rainfall deficit compared to average rainfall values after planting in both 2009 and 2010. A second smaller translocation was conducted in July 2009 by Mark Brundrett and Phylis Robertson (Friends of Kings Park Orchid Carers Group) to supplement numbers of *Caladenia graniticola* at Dragon Rock Nature Reserve. Unfortunately, this area had less than 1/2 of the annual rainfall in 2010 (Australian Bureau of Metrology, www.bom.gov.au).

2.3. Glasshouse Experiment

In a comparative study, seedlings of orchids that were the same size and from the same source as those used in field trials were planted into pots in the glasshouse. These pots contained soil from the sites where translocations occurred to allow the impact of soil moisture on plant establishment to be examined (see Figure 1). Each 15 cm pot consisted of soil excavated from a translocation sites with a top dressing of leaf litter from the same site. Seedlings were grown in the semi-permeable translocation pouches and watered every few days (Fig. 3). Seedling growth was measured and photographed weekly and window pots were used to measure dropper formation (Fig. 4).

2.4. Monitoring Translocations

Criteria for success and failure of translocations developed during the translocation proposal writing process are listed in Table 4. Plant survival and growth were recorded by taking comparable photographs and by measuring leaf and dropper length every week (glasshouse), or every 2-3 weeks (field). Measurements continued until plants were dead or dormant. Survival of seedlings was recorded for the first 1-2 years and further long-term monitoring is planned (Table 4). It was not possible to provide final numbers for seedling survival in 2010, due to severe drought conditions that resulted in low rates of emergence of orchids from dormant tubers, especially in the eastern wheatbelt.

Table 2. Rare and common orchid species included in translocation trials.

Year	Location	Orchid	*Caladenia*	Sites	Reps	No.	Total
2009	Near Brookton	Williams spider*	*C. williamsiae*	2	2	7	28
2009	Near Pingrup	Cowslip orchid	*C. flava*	2	2	7	14
2009	Near Pingrup	Ballerina orchid*	*C. melanema*	2	2	7	28
2009	Dragon Rocks NR	Clown orchid	*C. roei*	2	2	7	28
2009	Dragon Rocks NR	Granite spider*	*C. graniticola*	2	2	7	28
2010	Dragon Rocks NR	Granite spider*	*C. graniticola*	2	2	3	12

*Rare orchids.

Table 3. Experimental logistics for filed and glasshouse orchid establishment trials.

Orchids	3 rare vs. 3 common orchids
Levels	2 (field vs. glasshouse)
Locations	3 nature reserves (1 for each rare orchid)
Plots	2 plots several km apart
Reps	2 subplots per location with 2 pouches or 3 pots of soil from each
Seedlings	7 orchids per pouch

Table 4. Criteria for success or failure for translocations (Brundrett and Edgley 2009abc).

A. Criteria for Success

1. **Short Term:** (first year)
   - establishment and survival of translocated plants (number of seedlings remaining after the first growing season).
   - continued growth (leaf elongation rate) of transplanted seedlings.
   - germination of applied seed.
   - develop appropriate methods and gain knowledge that can be applied to future translocations of rare orchids.
2. **Medium Term:** (2-5 years)
   - The number of seedlings persisting through summer dormancy does not significantly decline
3. **Long Term:** (5-10 years)
   - The number of individuals is increased by natural recruitment in subsequent years.

B. Criteria for Failure

1. **Short Term:** (first year)
   - failure of translocated seedlings to establish and grow
   - failure to form a tuber and persist through summer.
   - failure of seed to germinate or grow to a stage where survival is possible.
   - failure to develop effective methods and gain knowledge to guide future translocations.
2. **Medium Term:** (2-5 years)
   - The number of seedlings persisting through summer dormancy significantly decline
3. **Long Term:** (5-10 years)
   - There is a significant decline in the number of the translocated orchids due to mortality and the lack of natural recruitment by seed over the next few years.

Figure 1. (Facing Page) **AB.** *Caladenia graniticola* translocation site at Dragon Rocks Nature Reserve showing two of the cages where seedlings were outplanted. **B.** There were two translocation pouches under each cage. **C.** Seedling with a dropper forming in a translocation pouch after several months of growth in the field. **D.** Seedlings of *C. melanema* planted in 2009 re-emerging in 2010. **E.** Translocation of *Caladenia williamsiae* near Brookton (Left - Nur Koshkuson, volunteer and right - Emily Ager, student). **F.** Mark Brundrett planting orchids. **G**. Erica Shedly and Marie Edgley also helped establish translocation trials in 2009. **H.** Phylis Robertson of the Friends of Kings Park Orchid Carers Group collecting soil for glasshouse experiments near Brookton in 2010.


A
B
C
D
E
F
G
H

3. Results

Seedlings were transplanted to field sites in the wheatbelt in August 2009 after final approval of translocation proposals first submitted to DEC in April (Fig. 6). Unfortunately, sites in the eastern wheatbelt were exceptionally dry in the late winter and spring of 2009. Ericka Shedley (DEC) and the author (MB) watered and measured plants several times in August and September, but visits were limited by remote locations in the eastern wheatbelt (1000 km trip total). Even though translocated seedlings were at a relatively advanced stage of development (with leaf growth concluded and droppers initiated), only a few survived in 2009 or 2010. The survivors which were observed included 5 plants of *C. melanema* (Fig. 1) and a 2 of *C. graniticola* at the end of the growing season, but further observations are required before the criteria in Table 4 can be fully assessed. The main reason for seedling mortality was severe drought as seedlings continued to grow in the glasshouse where watering was more frequent. Conditions were even drier in 2010, when rainfall was the lowest on record across most of southwest Western Australia.

The use of translocation pouches was found to assist the survival of seedlings during the transition from propagation containers to field conditions. These pouches also allowed the development of seedlings to be monitored more effectively by photography and by measuring the leaf length and dropper depth of each seedling.

Figure 1 shows 5 small plants of *C. melanema* planted in 2009 that emerged in 2010. Of these, only 1 flowered and severe drought caused early senescence of these plants in September 2010, but it is anticipated that they will emerge again in 2011. Long-term monitoring is required to determine if these newly established plants will persist as a new population (*C. graniticola*) or subpopulation (*C. melanema*) due to reproduction by seed or clonal division (*C. melanema* only).

Seedling establishment was also investigated in the glasshouse using soil from translocation sites (Fig. 3). Seedling survival was better in the glasshouse then in the field in the same soils, presumably because of more frequent supplemental watering. Glasshouse trials also allowed the rate of development of seedlings to be measured more accurately by weekly observations (Fig. 2) and photographs (Fig. 4) in window pots where dropper formation was also visible. It was observed that the capacity of seedlings to form tubers was correlated with their initial size (leaf length) at the time of translocation and that leaf growth stopped once droppers were forming. Seedling tuber formation was infrequent due to rapidly increasing temperatures in late spring as a consequence of the late start of trials. Some seedlings remerged in pots in 2010.

Figure 2 provides average rates of growth of leaves and droppers of *Caladenia* seedlings in the greenhouse. Plants grew steadily in some soils but not others from the same sites and there was also considerable variation between orchid species. As shown in Figure 2, *Caladenia williamsiae* did not grow substantially after transplantation in the glasshouse or field. The average length of leaves or droppers appears to decline in some graphs in Figure 2 due to attrition of plants. One possible explanation for the slow and variable growth of transplanted orchids in Figure 2 is that only a limited amount of potting mix with fungal mycelia was transplanted with each orchid and soils were relatively low in organic matter. It is likely that mixing soils from translocation sites with more orchid potting mix containing mycorrhizal fungi would have improved soil quality and seedling growth.

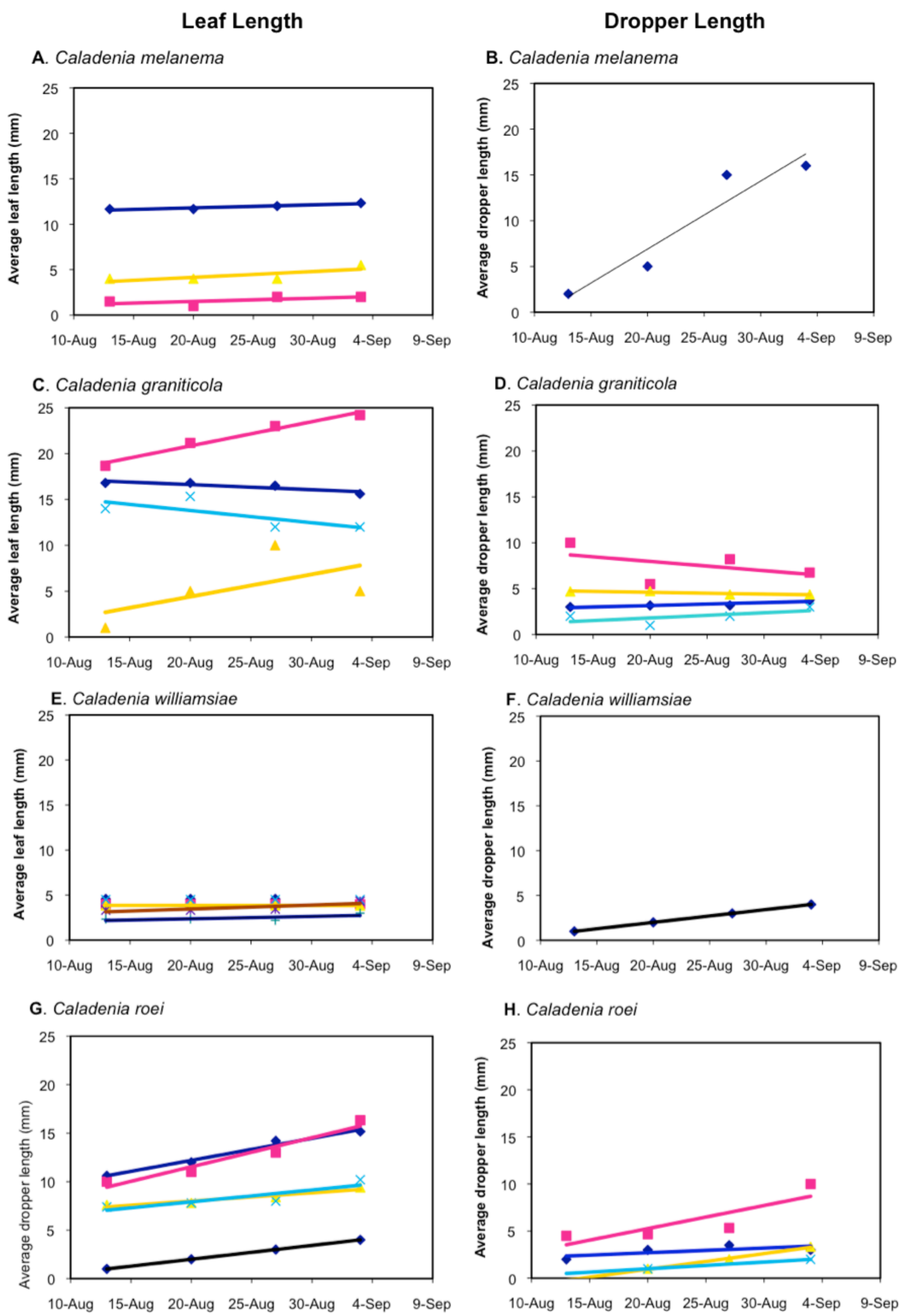

Figure 2. Growth of *Caladenia* seedlings in soil from natural habitats the glasshouse showing leaf (left) and dropper (right) growth for each species (from Ager 2009). Each line represents averages for plants in a pot. **AB**. *Caladenia melanema*. **CD**. *C. graniticola*. **EF**. *C. williamsiae*. **GH**. *C. roei*.

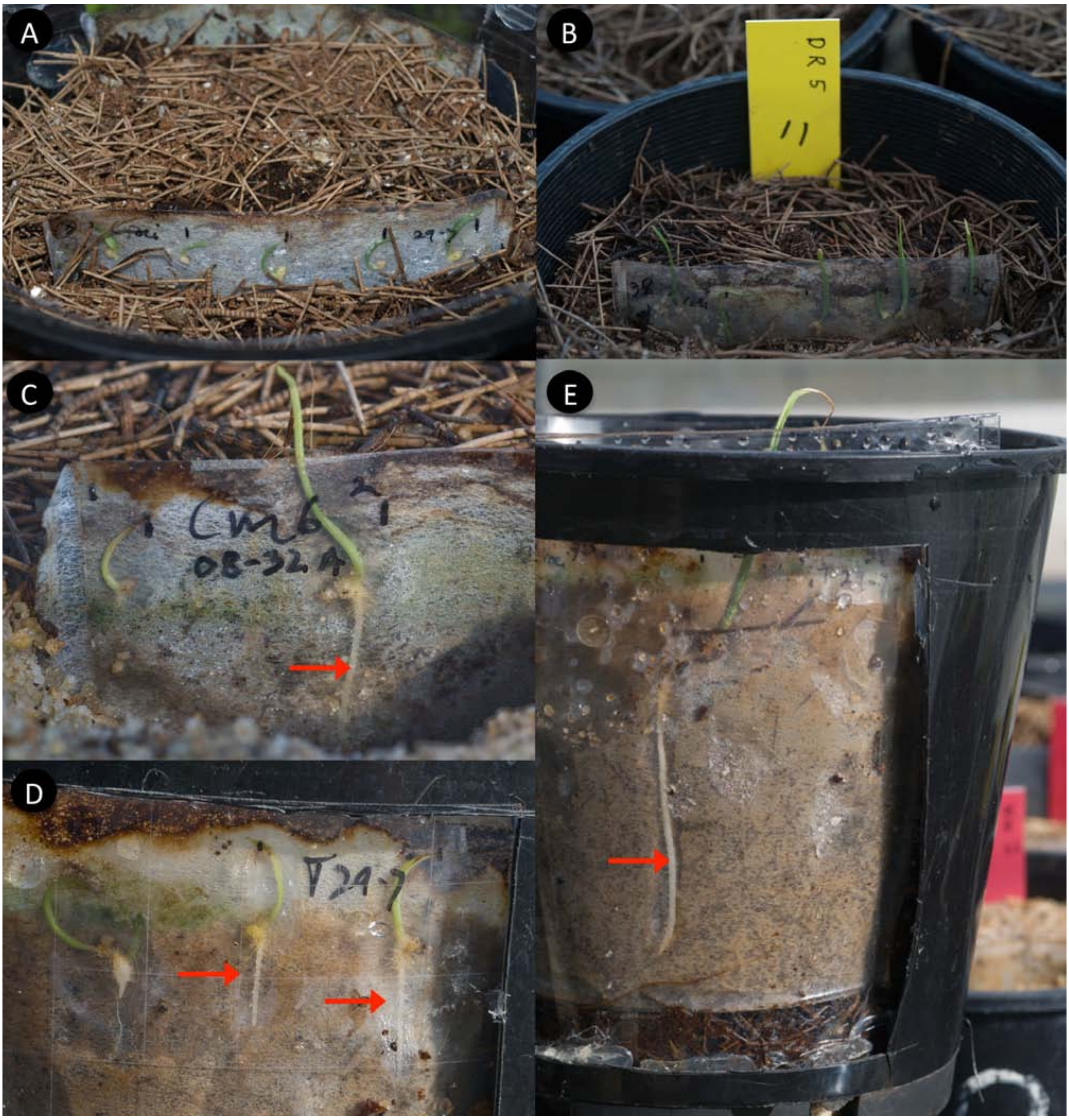

Figure 3. A. Orchid seedlings in translocation pouches in window pots in the glasshouse growing in soil from natural habitats. **ADE**. *Caladenia roei*. **B**. *Caladenia graniticola*. **C**. *Caladenia melanema*. **C-E.** Older seedling with droppers (arrows).

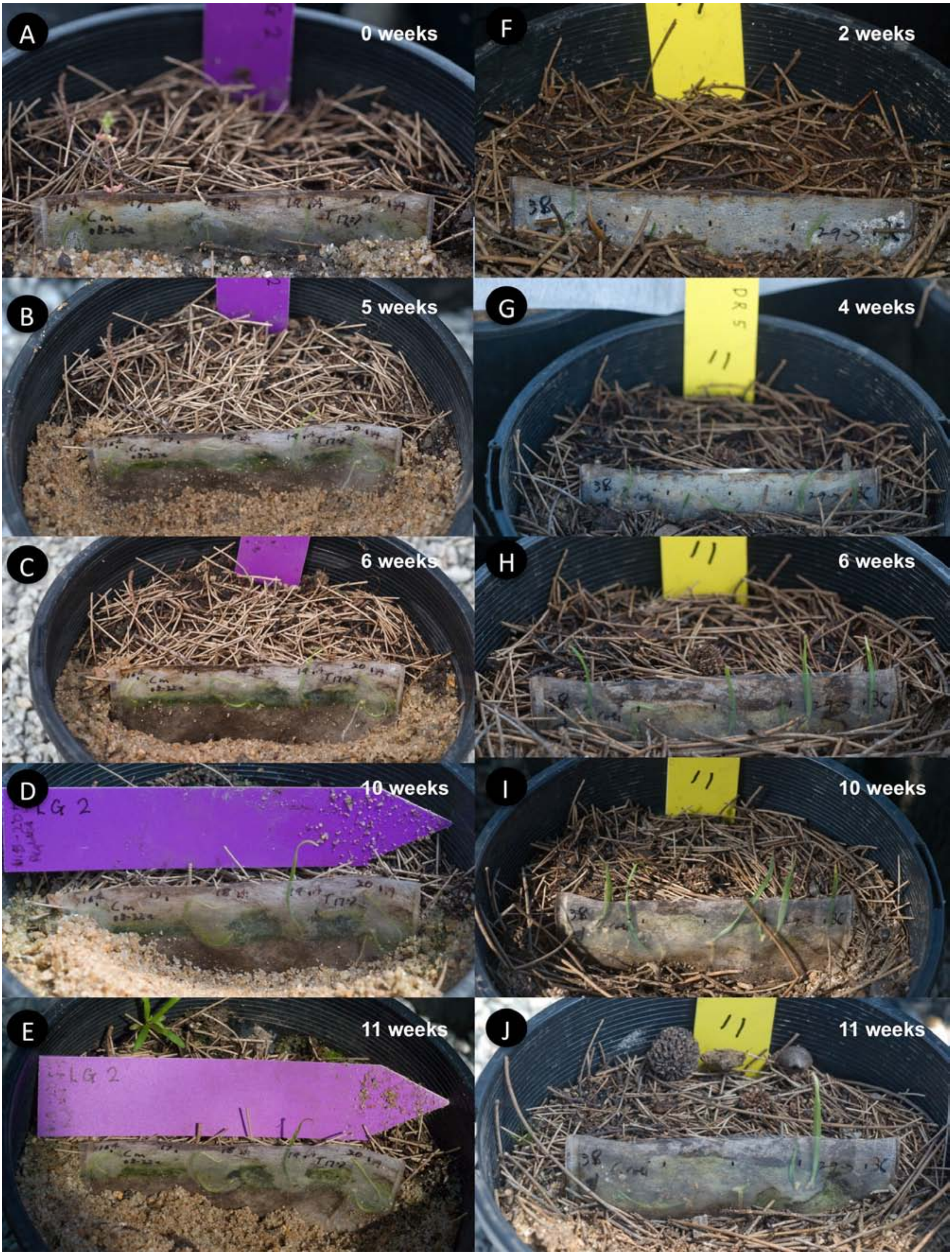

Figure 4. Growth of transplanted seedlings in pouches planted in soil from natural habitats as shown by weekly photographs of the same seedlings. **A-E**. The ballerina orchid (*Caladenia melanema*). **F-J**. The clown orchid (*Caladenia roei*).

Figure 5. Orchid seedling growth and mortality in the field. **A-F.** Translocated seedlings of the granite spider orchid (*Caladenia graniticola*) at Dragon Rocks Nature Reserve. **B**. Some larger seedlings were transplanted in 2009. **C,E**. Dropper formation was affected by severe drought. **F**. A plant that perished due to severe late winter and spring drought in 2009.

4. Conclusions and Recommendations

Translocations occurred at 3 remote locations in the central and eastern wheatbelt, necessitating a round trip of over 1000 km (Appendix 1). There was a very low survival rate of orchid seedlings, due primarily to exceptionally low rainfall in both 2009 and 2010, but a few plants did survive record-breaking drought conditions. The germination and subsequent growth of seedlings established that compatible fungi were present in some soils from these sites, but the distribution of fungal inoculum was patchy (WOR Report 7). It is hoped that there will be future opportunities to further optimize translocation protocols for orchids in dry environments in WA. The long-term survival of orchids that were established will be measured over the next few years.

These experiments provided successful preliminary trials of several new orchid propagation and translocation technologies developed by the author (MB) for orchid conservation work. In particular, translocation pouches improved seedling survival in dry soils without the need for a long acclimatisation period, provided drought conditions were not overly severe. They also allowed the growth of individual seedlings to be tracked and for dropper/tuber formation to be observed without damage to plants. The use of these pouches within window pots provided similar benefits in the glasshouse. However, there have only been preliminary trials of these new propagation methods so more work is needed before recommending their wide scale adoption.

Translocation trials in the West Australian wheatbelt provided valuable information on orchid ecology by comparing the reproductive and establishment potentials of common and rare orchids in the genus *Caladenia*. In general there were no major differences in reproductive potential between DRF and common orchids that co-occur. However, there were two exceptions; (i) one extremely rare orchid, *C. williamsiae*, germinated more rapidly than most other orchids in sterile culture, but was very difficult to establish in soil and (ii) the common orchid, *C. roei*, was easier to propagate than any other *Caladenia* species included in trials. It is not known if these species-specific differences are due to properties of mycorrhizal fungi or of the orchids themselves, or both. However, the low establishment rate of *C. williamsiae* in soils is linked to low rates of detection of compatible mycorrhizal fungi in the same soils (WOR Report 7).

Observation of seedling development in soil from natural habitats in the glasshouse suggested that it was a hostile environment for young seedlings. Even in the absence of drought, there were major problems with water repellent and hard-setting soils (due to 5-10% clay content). It is recommended that future trials include soil amendments to increase organic matter content and inoculation with additional mycorrhizal fungi if rapid growth is required. It may also be necessary to provide irrigation to supplement (or replace) rainfall. Translocation pouches provided some protection from invertebrate grazing, but some seedlings were still lost to fungus gnats in the glasshouse. Consequently, soils used for orchid growth may also benefit from improved drainage or other insect control measures, especially in the glasshouse where temperatures are higher.

The WOR project identified the slow approval process for translocation proposals for DRF plants in WA as a major factor limiting their success. In 2009 this resulted in translocation occurring several months later than was optimal, especially since subsequent rainfall was exceptionally low. There seems to be a less complex process for approval of translocation proposals elsewhere in Australia. A more streamlined approvals process is especially appropriate when; (i) the need for translocation has already been stated in the state and commonwealth approved Interim Recovery Plan for a species, (ii) the proposal is based on comprehensive knowledge of the species and (iii) there are no adverse impacts on endangered species or their habitats.

Another key recommendation of the WOR project is that a preapproved generic translocation plan be developed that that can be used for any orchid species in south-western WA. This is possible because the methodology for each species (genetic materials, monitoring, propagation of plants, experimental design, etc.) will be very similar in each case and specific information for a species has already been summarised in its IRP. Experimental methodologies will vary somewhat for each translocation as they depend on how successful propagation efforts have been and if additional treatments to protect plants from desiccation, grazing, etc., are required. However, these minor variations in experimental plans must often be decided after the plan is approved, by taking adaptive management approach to the translocation. In particular it is not possible to predict if there will be severe drought, which types of grazing will occur, exactly how many orchid plants will germinate, or how fast they will grow and it would be wasteful to propagate extra endangered orchids and then discard the extras.

For some of the DRF WA orchids studied in detail by the WOR project, overcrowding was identified as a major issue, as there were many tubers in a small area and the majority of them remain dormant each year (WOR Reports 2-5). It is recommended that in these cases, some existing tubers be translocated to new habitats to alleviate the need for laborious and expensive propagation of orchids and reduce the impacts of overcrowding (density dependent impacts on resources and pollination) in the most important Core Habitat areas for these species.

Volunteers of the Orchid Carers Group of the Friends of Kings Park provided considerable assistance to propagate rare orchids during the WOR project. Emily Ager, a research project student of the University of Western Australia, also played very important role. However, orchid propagation by volunteers or students is only possible if they are supervised by scientific staff with sufficient experience and have access to properly equipped laboratory and plant growth facilities (in this case provided by Mark Brundrett and Plant Biology at the University of Western Australia). It is recommended that dedicated facilities or resources be provided specifically for orchid conservation in WA due to the very large number of state and commonwealth listed species of high conservation concern in our state.

Perhaps the most important outcome of the WOR project was to establish means for rapid and efficient terrestrial orchid propagation by harnessing fungal activity in a semi-natural, non-sterile environment. This has the potential to make orchid conservation programs less reliant on expensive and complex laboratory facilities and highly trained personnel. Thus, it is imperative to continue development the FORGE methods for orchid conservation developed for the WOR project to investigate their applicability in other environments, especially in developing countries with many rare orchids. It is also anticipated that simular methods could be adapted for propagation of epiphytic orchids for both conservation and horticulture.

5. Acknowledgements

Translocation trials by the Wheatbelt Orchid Rescue Project were funded by Lotterywest. Erica Shedly and Marie Edgley of DEC took part in translocation trials in 2009. Phylis Robertson of the Friends of Kings Park Orchid Carers Group assisted in the 2009 trial. Orchid seedlings were produced by Mark Brundrett, Emily Ager and Nur Koshkuson in 2009. In 2010, volunteers of the Friends of Kings Park Orchid Carers Group had a major role in orchid propagation (see WOR Report 7). Facilities for orchid propagation were provided by the University of Western Australia. Hai Ngo, Rob Creasy and Margaret Collins provided invaluable advice and assistance.

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