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Applying the seed use chain to identify research priorities for the rehabilitation of *Spinifex* (*Triodia spp*.) in the Pilbara, Western Australia

Blake Wood [a,*], Simone Pedrini [b], Shane Turner [b], Roy Wittkuhn [a], Ebony Cowan [a], Pamela Chester [a], Pedro Pañella [b], Micol Orengo [b]

[a] *Rio Tinto, Australia*

[b] *School of Molecular and Life Science, Curtin University, Australia*

Abstract

Rio Tinto has an active progressive rehabilitation program centrally coordinated across multiple iron ore mining operations, ports and rail networks in the Pilbara region of Western Australia. Spinifex (Triodia spp.) grassland is a dominant vegetation type in the region and makes up a large component of the seed mixes broadcast as part of rehabilitation projects. Despite its importance, industry research and reviews have highlighted that Triodia is challenging to establish in rehabilitated areas. Whilst there has been important research done by institutions such as The University of Western Australia and Kings Park and Botanic Garden (Kings Park) in understanding Triodia recruitment, knowledge gaps still exist in how to test, store and treat Triodia seed.

To address these knowledge gaps, in 2025, Rio Tinto and the Native Seed Technology and Innovation Hub at Curtin University (Curtin) developed a research partnership. To inform the direction of research, the Triodia seed use chain was mapped, with business-as-usual practices and current knowledge gaps detailed. This step permitted a collaborative prioritisation process, with the most significant knowledge gaps being the focus for initial research. Curtin and Rio Tinto's initial research program concentrated on seed testing, cleaning seed for storage and the alleviation of dormancy. Early findings have identified the ability to clean Triodia batches to pure florets, which will significantly reduce seed storage volumes, as well as identifying potential pathways for dormancy alleviation. Research will continue, with a goal to develop a scalable dormancy-breaking method that ultimately leads to increased recruitment of Triodia in the field. Research will be scaled from the laboratory to the field, adding confidence that improvements are practical and scalable across species and the Pilbara region.

Keywords: *Triodia, seed use chain, rehabilitation, Pilbara, seed dormancy, seed cleaning*

1 Introduction

Rio Tinto operates an integrated network of iron ore mines, rail corridors and port infrastructure across the Pilbara region of Western Australia. Although finite, the long operational life spans of these assets necessitate early and sustained planning for closure and rehabilitation. Progressive rehabilitation is implemented across multiple sites to inform practices and methodologies that will enable successful rehabilitation outcomes, and reduce disturbance footprints and closure liabilities.

Improving the density and cover of *Spinifex* (*Triodia spp*.) in rehabilitated environments is a challenge across the Pilbara and broader mining industry (e.g. Erickson et al. 2017). This is a significant issue, as *Triodia* is a dominant vegetation genus for many of the operations Rio Tinto progressively rehabilitates. Furthermore, *Triodia* seed currently comprises 70% by weight of the seed mixes used for direct seeding across

* Corresponding author. Email address: blake.wood2@riotinto.com

rehabilitation sites. Due to seed supply constraints, there is a need to both improve the seed use efficiency of *Triodia* as well as increase the success of *Triodia* recruitment from seed.

To help address the recruitment problem with *Triodia* outlined above, Rio Tinto sought to understand:

1. Why is the recruitment of *Triodia* poor in rehabilitation environments?
2. What are the factors that can be changed or improved to increase the rate of recruitment?
3. Which of these improvement factors can be both scaled for rehabilitation programs and justified economically?

In 2023 Rio Tinto undertook a desktop review and established a research partnership with the Native Seed Technology and Innovation Hub at Curtin University (hereafter Curtin) in 2025 to begin answering these questions and provide scalable solutions to the sustainable use of *Triodia* seeds for mine site rehabilitation.

2 Methodology

2.1 Why is the recruitment of *Triodia* poor in rehabilitation?

To answer this question, Rio Tinto initially undertook a desktop review of the literature related to *Triodia* recruitment and mine site rehabilitation in the Pilbara. The following key success factors were identified as part of this review:

1. *Triodia* seed is physiologically dormant, with dormancy largely imposed by the floret – the covering structure surrounding the caryopsis (Kildisheva 2019; Erickson et al. 2016).
2. *Triodia* typically has low seed fill rates, which may be an evolutionary response to varied rainfall, and predator activity (Wright 2021). When stored *Triodia* seed batches at Rio Tinto were tested, only 15 to 50% of the florets contained a caryopsis. This has implications for how many pure seeds are stored and distributed when seeding rehabilitation projects.
3. Fundamental research, particularly by the University of Western Australia and Kings Park and Botanic Garden (Kings Park), found that recruitment of *Triodia* can be improved by undertaking the following
   a. alleviation of dormancy, for which there are a range of techniques including cleaning to the pure caryopsis and treating with karrikinolide (KAR_1), an active component in smoke that promotes germination, flaming of the floret, dry after ripening (DAR), and rapid wetting and drying (Erickson et al. 2017; Lewandrowski et al. 2017; Berto et al. 2020)
   b. seeding *Triodia* at the correct depth in the soil, with 5 mm being optimal (Masarei et al. 2020)
   c. providing sown *Triodia* with an ecological rainfall event, with greater than 120 mm over a week found to be optimal (Erickson et al. 2023).

In reviewing the literature, it was unclear which of these factors could feasibly be implemented, both practically and economically, at scale. For example, provision of 120 mm in an irrigation event to a newly rehabilitated site is in many cases not practical, given the scale of the projects over vast landform structures and limited water supplies.

2.2 What are the factors that can be changed or improved to increase the rate of recruitment?

By mapping the current seed use chain for *Triodia*, deficiencies could be compared against the literature and knowledge gaps identified at each step, from a rehabilitation operational perspective (Figure 1).

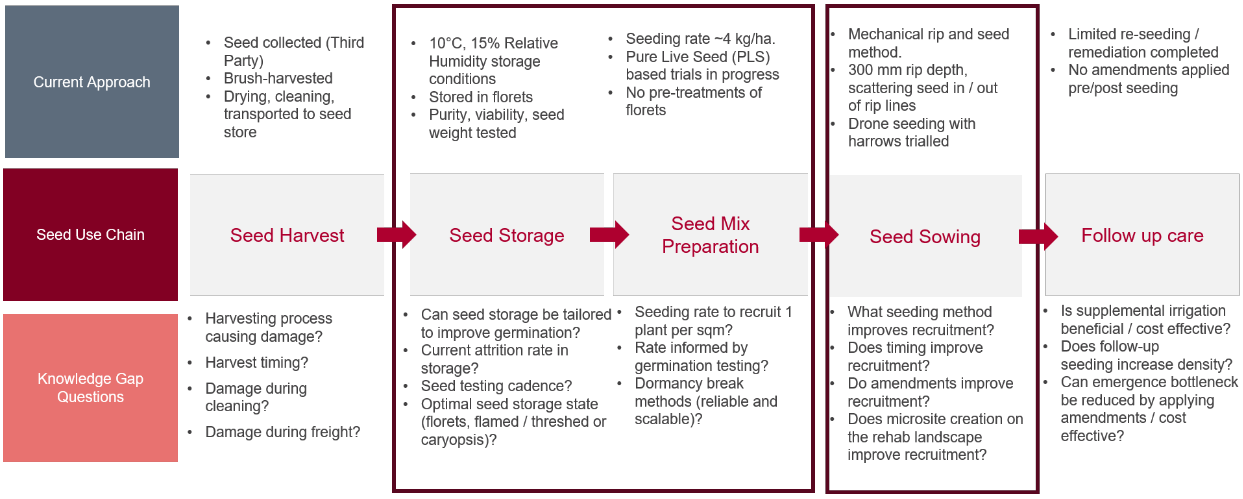


Figure 1 The Rio Tinto seed use chain of *Triodia*, with current approaches and knowledge gap questions for each stage identified. Two broad research areas were identified as the most important to pursue, as shown in 2 boxes: the seed storage and mix preparation, and the seed sowing of *Triodia*

From this exercise, most of the management activities and knowledge gaps were found to be associated with how *Triodia* seed was stored and prepared for seeding, and how it was seeded across rehabilitation sites.

To address the seeding method knowledge gaps, desktop research and field trials began in 2023 to evaluate the current seeding method (ripping and mechanical seeding) against other seeding methods, such as drone seeding and the use of agricultural seeding equipment. This research culminated in a head-to-head field trial of seeding methods in 2025 and a design process for a single-pass seeding solution commenced.

To progress targeted research on improving seed storage and preparation, a partnership began with Curtin. Curtin to develop an initial research program based on the current seed use chain and identified knowledge gaps for *Triodia*.

The program proposed the following focus areas:

1. Quality testing: across 5 *Triodia* species, seed would be tested to get a baseline of thousand seed weight and pure live seed (PLS) measurements (viability and purity). Batches of each of the species would also be germination tested to estimate the current germinability of each batch without pre-treatment.
2. X-ray testing: batches from each of the 5 *Triodia* species would be tested in an X-ray machine with software developed by Curtin to measure viability and seed weight. These tests would evaluate whether X-ray was a viable alternative to the traditional cut test to determine seed viability.
3. Seed processing: processing techniques were trialled to evaluate whether unfilled florets could be successfully removed from 5 *Triodia* seed batches as well as cleaned to the caryopses. Furthermore, any cleaned batches (whether florets or caryopses) would be tested for germination to understand if seed processing was having a deleterious effect. Processing techniques would include threshing or acid digestion with threshing and aspiration of non-viable floret material.
4. Seed dormancy: a range of dormancy alleviation techniques would be used on 5 batches of *Triodia* to evaluate germinability against untreated florets. Dormancy treatments would include caryopses and florets treated with KAR_1, smoke water and acid digestion. Separate long-term experiments would be established to evaluate whether DAR and exposure to dry heat leads to increased germination. The mechanism of germination suppression by the floret would also be investigated through a series of tests, with saturated oxygen environments and leaching of the floret designed to answer whether or not the floret restricts oxygen to the embryo or a chemical present within the floret causes the dormancy.

2.3 Which of these factor improvements can be both scaled for mine site rehabilitation programs and justified economically?

Critical to each of the 4 research themes was an analysis of the economics of any proposed change to the current seed use chain. For example, if a method was identified to remove unfilled florets from a seed batch, the cost of doing this processing step would be weighed against the potential benefits, which would be seed storage space saved. Such hypotheses need to be experimented with first by identifying the proper equipment.

Whilst the first year of the research partnership was unlikely to completely answer any of the targeted knowledge gaps, the aim of the research was to incorporate new findings and process improvements into the company Seed Management Plan (SMP) so that practices were formalised and *Triodia* was consistently treated the same way. The research team was provided access to the SMP to make agreed updates as new findings were made.

3 Results and data

3.1 Quality testing

Five *Triodia spp*. were provided to Curtin for the first year of the research. Curtin performed the initial quality seed testing, following the methods described in the Society for Ecological Restoration international standards for native seed in ecological restoration (Pedrini & Dixon 2020), which included assessing the purity (%), viability (%) and the seed weight (g).

The PLS percentage was calculated by:

$$\frac{purity\ (\%) \times viability\ (\%)}{100} \qquad (1)$$

The seed quality test results indicated that whilst purity is high (very little non-seed material in the collections), viability (proportion of filled seed) is low for the *Triodia* species, ranging from 28.3 to 55.7% (Table 1).

Table 1 Seed quality testing of 5 *Triodia* species

Species	Thousand seed weight (g)	Purity (%)	Viability (%)	Pure live seeds (%)
Triodia basedowii	1.92	98.4	38.1	37.4
Triodia epactia	1.44	96.5	41.3	39.9
Triodia longiceps	1.01	99.8	55.7	55.6
Triodia pungens	0.88	99.9	29.0	28.9
Triodia wiseana	0.84	99.7	28.3	28.2

Initial germination testing confirmed the presence of dormancy for *Triodia* seed contained within the floret. Both *T. longiceps* and *wiseana* had negligible germination when contained in the floret, with or without dormancy pre-treatment (data not shown). Similarly, *T. b*asedowii in the floret had negligible germination without dormancy pre-treatments, and less than 5% germination with any pre-treatment (Figure 2). However, after 9 days, this species recorded 40% germination with the floret removed (caryopsis). *T. pungens* in the floret recorded 10% germination after 10 days, with dormancy pre-treatments (KAR_1) applied at the start of the germination test increasing the germination to 60% after 14 days. By removing the floret and applying no other dormancy pre-treatment, germination of *T. pungens* increased to 60% within 4 days. The baseline germination testing showed the role the floret plays in restricting germination and, by removing it, germination increased across all the species tested.

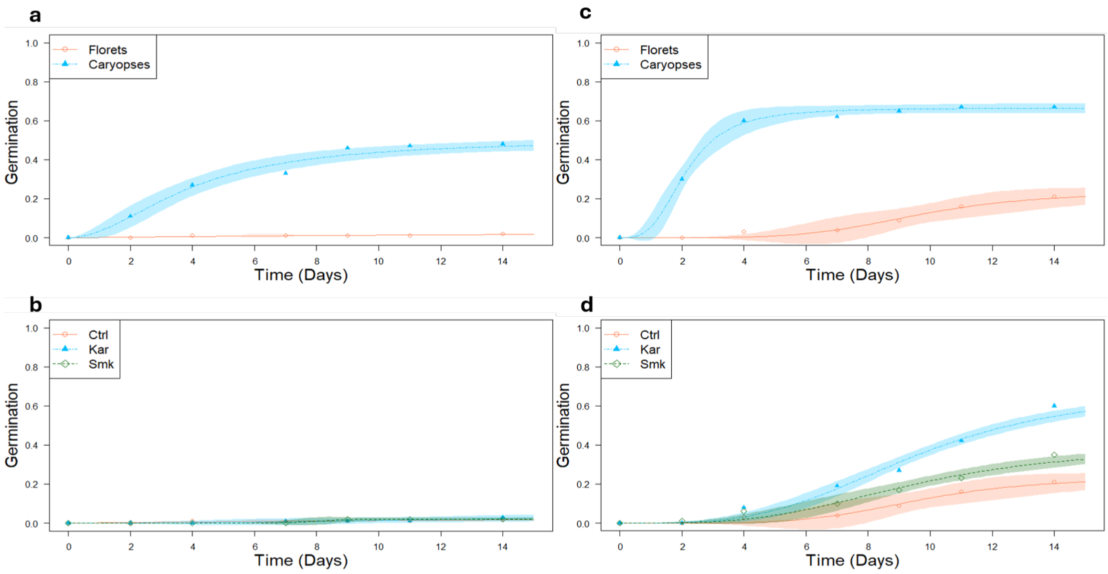


Figure 2 Proportion of germination over time (days) for: (a) *Triodia basedowii* florets and caryopses without dormancy pre-treatment; (b) *T. basedowii* florets without dormancy pre-treatment (Ctrl), exposure to 1 µM KAR_1 solution (Kar) and 10% smoke solution (Smk); (c) *T.* pungens florets and caryopses without dormancy pre-treatment; (d) *T. pungens* florets without dormancy pre-treatment (Ctrl), exposure to 1 µM KAR_1 solution (Kar) and 10% smoke solution (Smk)

Dormancy pre-treatments on the caryopses (which were exposed to KAR_1, smoke water and sulphuric acid) improved germination above no dormancy pre-treatment (Figure 3). KAR_1 exposure was the most effective dormancy pre-treatment across all species, achieving greater than 70% germination. However, the effectiveness of other dormancy pre-treatments differed between species. For example, exposure to sulphuric acid achieved less than 20% germination in *T. basedowii*, whilst when applied to *T. longiceps*, germination was approximately 60% after 4 days.

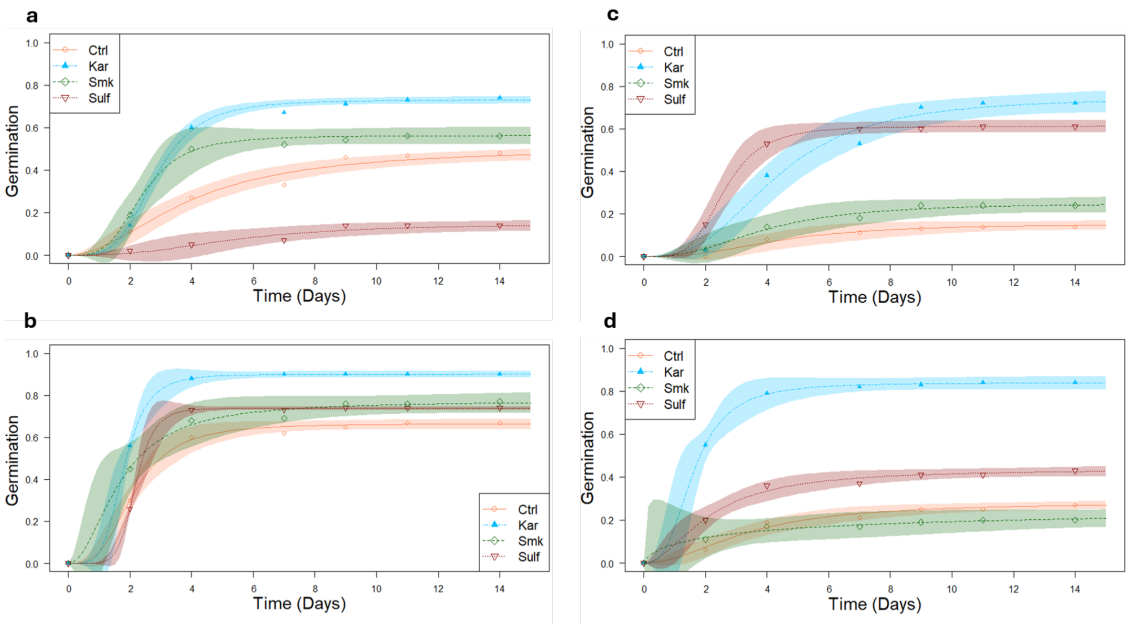


Figure 3 Proportion of germination over time (days) for: (a) *Triodia basedowii* caryopses; (b) *T. pungens* caryopses; (c) *T. longiceps* caryopses; (d) *T. wiseana* caryopses, without dormancy pre-treatment (Ctrl), exposure to 1 µM KAR1 solution (Kar), exposure to 10% smoke solution (Smk) and exposure to concentrated sulphuric acid (Sulf) for 2 minutes

3.2 X-ray testing

Seed fill testing is performed at Curtin using an enclosed X-ray cabinet (Kubtec Parameter). The digital X-ray scan images are then analysed using custom-built software (XanthorRay – Ruby Edition) for assessing viability (seed fill) in all the *Triodia* species tested. The X-ray data for the species tested in 2025 will be used in the software to build parameters that can assist with automatic assessment of viability, rather than manual selection and counting of filled versus unfilled florets. By enabling automatic detection of viability, the efficiencies for viability testing using an X-ray will increase along with the speed of assessment. However, seed that is smaller than ~0.5 mm is not suitable for the current resolution of the Kubtec X-ray machine. Future stages of the research program aim to evaluate alternative X-ray machines to optimise resolution for smaller-seeded species. In the meantime, they will continue inputting seed samples into the current X-ray machine to create a robust dataset for automated viability testing of *Triodia*.

3.3 Seed processing

Initial seed quality testing indicated that 44.4 to 71.8% of the florets in *Triodia* seed batches were unfilled. Curtin therefore investigated a method to clean the *Triodia* batches down to filled florets. The seed cleaning method involved 3 processing steps. The first step was to separate the individual florets from the *Triodia* spikelets (originally part of the inflorescence) by gentle threshing (Figure 4). This was followed by sieving to isolate any spikelets remaining, which would be re-threshed. Individual florets collected from the sieve were then passed through air separation using a Petkus Selecta Zig-Zag aspirator. The cleaned sample was then verified by X-ray to confirm the presence of filled florets (Figure 5).

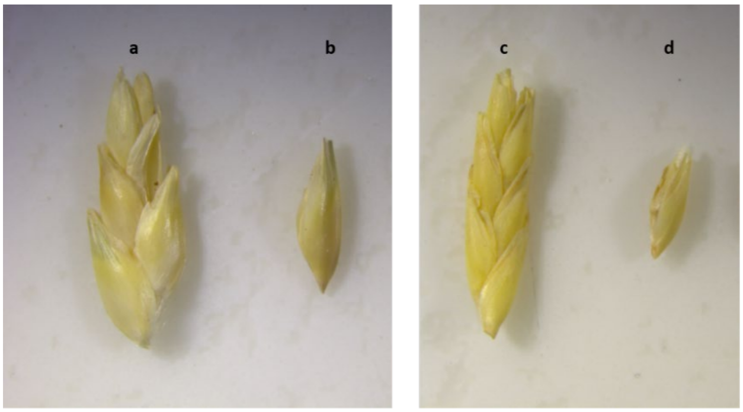


Figure 4 (a) *Triodia pungens* spikelet; (b) *T. pungens* individual floret; (c) *T. longiceps* spikelet; (d) *T. longiceps* individual floret

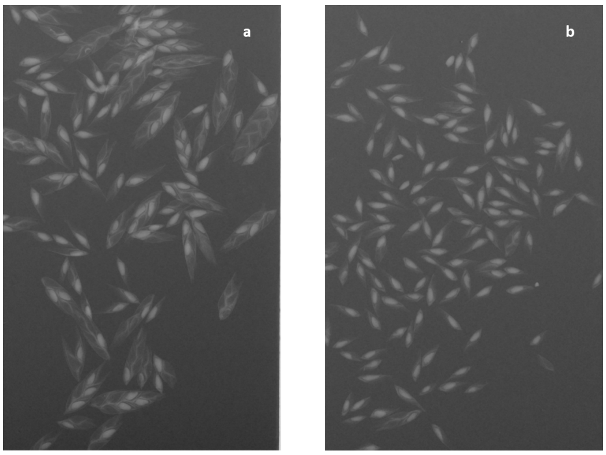


Figure 5 X-ray images of a *Triodia s*eed batch, showing filled (white bodies in the image) and unfilled florets (opaque bodies in the image). (a) Florets aggregated in spikelets; (b) Filled individual florets after cleaning

The cleaning procedure was able to increase the proportion of pure live seeds within the *Triodia* seed batches to more than 90%. In addition, pure caryopses were separated from florets in the cleaning process (Figure 6).

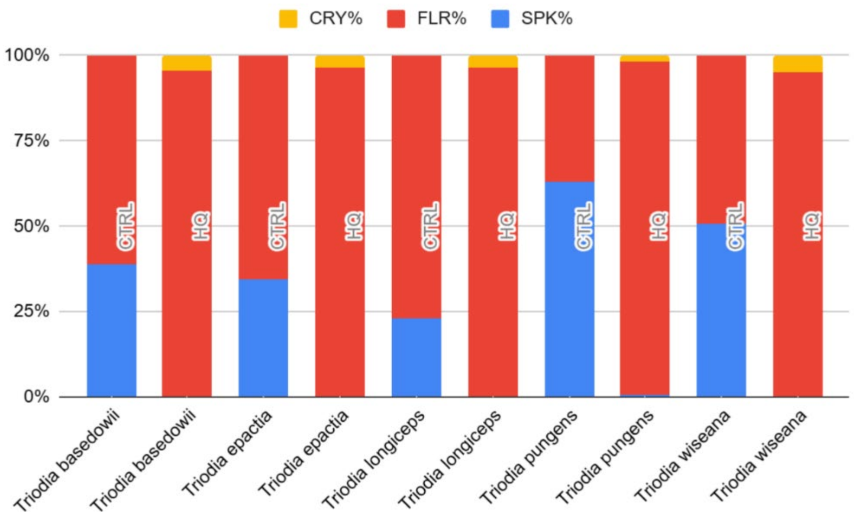


Figure 6 The proportion by weight (%) of caryopsis (CRY), filled floret (FLR) and spikelet (SPK) in the original seed batches (CTRL) and cleaned batches (HQ)

The ability to clean the *Triodia* batches to almost entirely filled batches of seed has significant seed storage savings. Depending on the species, cleaning *Triodia* seed batches to filled florets can reduce the volume occupied of the seed batch by 2.6 to 6.7 times. This volume reduction is even greater with cleaning to the caryopsis (Figure 7).

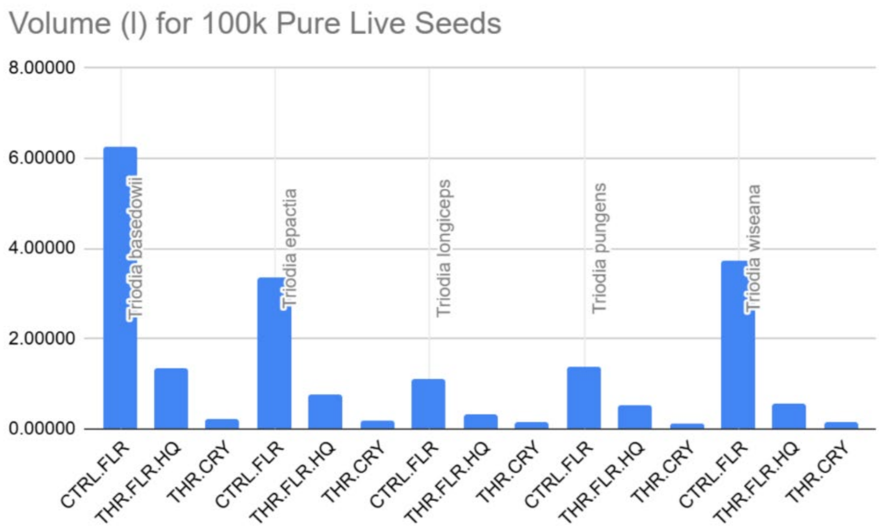


Figure 7 Volume (L) occupied for 1,000 pure live seeds across 5 *Triodia* batches, in the original batch (CTRL.FLR), the batch cleaned to pure florets (THR.FLR.HQ) and the batch cleaned to caryopses (THR.CRY)

At the time of writing, approximately 70% of the Rio Tinto seed store is *Triodia*. The volume reductions of 2.6 to 6.7 times for the *Triodia* batches tested would mean a 300% volume reduction conservatively, and up to 470%, if the entire stock of *Triodia* was cleaned to pure florets (depending on the quality of the seed batches) (Pedrini, pers. comm., 22 January 2026). The seed cleaning therefore has the potential to provide significant storage capacity without the need to expand the current controlled seed store footprint (managed at 10 °C and 15% relative humidity).

Germination testing indicated that cleaning *Triodia* seed batches to caryopses and treating with KAR_1 increased germination to over 70% for all *Triodia* batches tested, which is a significant improvement compared to germination from florets. Therefore, Curtin tested the viability of using seed threshing equipment to clean to caryopses as a scalable method. However, more aggressive threshing was required to release the caryopsis from the floret compared to breaking spikelets into individual florets. Cleaning to the caryopsis therefore increased the proportion of dead and damaged seed, with the proportion differing between species (Figure 8). The increased damage of the seed was due to impacting the embryo, which sits on the side of the caryopsis of *Triodia* and is therefore quite exposed (Figure 9). Short exposure to concentrated sulphuric acid prior to threshing was found to reduce the proportion of caryopses that were dead or damaged, across the species tested, as the acid appears to have weakened the surrounding floret structure (Figure 8).

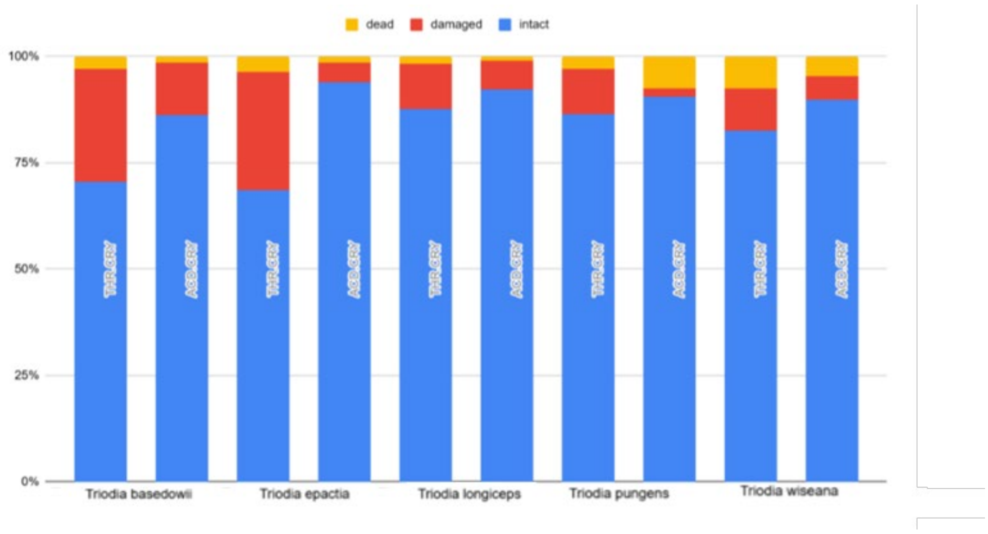
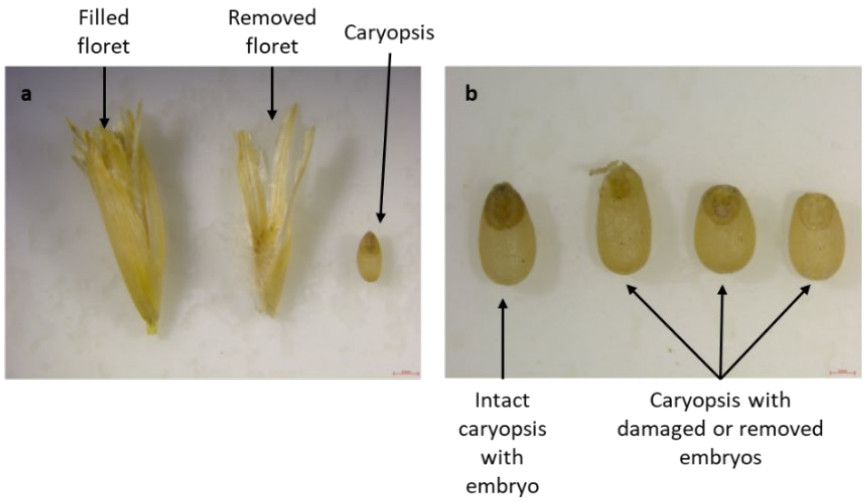


Figure 8 Proportion of seed batch (%) that contained dead, damaged and intact caryopses with threshing (THR.CRY) and acid digestion, followed by threshing (ACD.CRY), across 5 species of *Triodia* tested



(a) (b)

Figure 9 (a) Seed cleaning of *Triodia basedowii*, from the original filled floret to the extracted caryopsis; (b) Damaged or removed embryos of extracted *Triodia* caryopses

3.4 Seed dormancy

Seed dormancy was confirmed in the *Triodia* species tested, evidenced by the influence of the floret restricting germination as well as the benefit of KAR_1 application. Curtin performed experiments to understand the mechanism of the dormancy (was it mechanical, chemical or both?). Four experiments were established to understand the mechanisms. A high-temperature exposure of florets was performed at 80, 100 and 120 °C for 10, 30 and 60 minutes at each temperature, respectively. DAR of florets was also undertaken at temperatures of 40 and 60°C, at 30 and 70% relative humidity for each temperature point, respectively. These DAR environments were exposed to florets for 2, 4 and 16 weeks. In addition, cut and uncut *Triodia* florets were exposed to saturated oxygen environments (> 70% oxygen) to investigate whether the floret was restricting the access of oxygen to the embryo, thus reducing germination. Finally, an experiment was done to see if there was a chemical in the floret inhibiting germination. Floret material was macerated in either water (soluble compound extraction) or acetone (insoluble compound extraction), with the leachates applied to filter paper and caryopses plated onto the paper in germination tests.

At the time of writing, the temperature and DAR experiments are still in progress. However, initial results from the saturated oxygen environment test indicated no difference in germination between cut and uncut florets in a saturated oxygen environment. In addition, initial results indicate that germination is reduced for caryopses exposed to the water leachate of florets compared to the control with no leachate present. Further experimentation is required, but this initial experiment suggests that there is a water-soluble compound in the floret that restricts germination in *Triodia*.

4 Discussion

The research undertaken in partnership with Curtin has confirmed the presence of physiological dormancy in the *Triodia* species tested, which aligns with previous work on *Triodia* (Erickson et al. 2016). Similarly, the removal of the floret and treating with KAR_1 provided the highest germination result across the species (and batches) tested. The focus for the next phase of the research with regards to the dormancy of *Triodia* will be to answer the following 2 key questions:

1. What dormancy-breaking method leads to long-term improved recruitment when applied to a field setting? Previous research has shown that, whilst germination is improved through dormancy pre-treatments, long-term survival in soil may be poorer in dormancy-treated seed compared to untreated seed in water limited environments (Lewandrowski et al. 2021). A diversification strategy may need to be employed to ensure viable seed remains in the seed bank when conditions aren’t favourable for long-term (i.e. greater than 12 months) survival (Commander et al. 2020).
2. If a dormancy pre-treatment can promote increased plant recruitment from seed, which technique can be operationalised to support the scale of mine site rehabilitation work? Furthermore, is the cost of an appropriately scaled dormancy pre-treatment justified through the increase in *Triodia* density in rehabilitated landscapes?

In addition to the findings on dormancy, the ability to clean *Triodia* batches to pure florets has the potential to significantly reduce the current storage space occupied by *Triodia* by up to 470% (Pedrini, pers. comm., 22 January 2026). This would provide redundancy in the store to cater for increased seed procurement when the opportunity arises with bumper *Triodia* harvests. Similarly to dormancy pre-treatments, future work will focus on investigating a scalable solution for cleaning *Triodia* batches to pure florets, as well as assessing the economics of investing in seed cleaning equipment for this purpose. In concert with seed processing equipment, new X-ray equipment will be investigated to assist with efficient viability testing, which will be essential during large-scale seed cleaning operations.

Whilst still ongoing, the end point of this research is to update business practices to improve the recruitment of *Triodia* in rehabilitated landscapes. To this end, any process improvements will be detailed in updates to the company’s Seed Management Plan, incorporating input from Curtin.

5 Conclusion

A research partnership established between Curtin University and Rio Tinto in 2025 has provided promising learnings in the management of *Triodia* seed for rehabilitation across Pilbara assets. Key learnings from the first year of the partnership have been the ability to clean seed batches to pure florets, which has clear benefits for seed storage space, as well as the potential for X-ray imaging as an efficient alternative to traditional cut viability testing. Both findings will be investigated for future implementation, and economic analysis to quantify the potential savings for any scalable solution found will be undertaken.

Furthermore, the dormancy findings align with previous research regarding the role of the floret in imposing dormancy. Pre-treatment techniques, such as removal of the floret and exposure to karrikinolide were found to increase the germination of tested *Triodia* batches to more than 70%. Further research will investigate the underlying mechanisms of dormancy, as well as evaluating which dormancy pre-treatment leads to increased recruitment under field (rehabilitation site) scenarios. If a dormancy treatment method can demonstrate better recruitment of *Triodia* in the field, work will then concentrate on whether the treatment can be scaled up to progressive rehabilitation programs, as well as an economic analysis of investing in the dormancy pre-treatment process.

The research partnership has been beneficial due to the collaboration between Curtin University and Rio Tinto rehabilitation and performance practitioners, particularly in clearly defining the research problem and research focus areas. By mapping the seed chain use and identifying the knowledge gaps, Curtin has been able to target research that can directly assist with business-as-usual practices. Incorporating economic analysis into the research also validates, and provides clear arguments for, the need for improvements in the current practice regarding *Triodia* seed management.

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