Office Use Only	
Project Code	
Project Type	

FINAL REPORT 2022

Applicants must read the *SAGIT Project Funding Guidelines* prior to completing this form. These guidelines can be downloaded from www.sagit.com.au

Final reports must be submitted by email to admin@sagit.com.au as a Microsoft Word document in the format shown **within two months** after the completion of the Project Term.

PROJECT CODE	UA619
PROJECT TITLE	(10 words maximum)
Revealing the basis for head loss in barley	

PROJECT DURATION *These dates **must** be the same as those stated in the Funding Agreement.*	
Project start date	**1/04/2019**
Project end date	**1/02/2022**

PROJECT SUPERVISOR CONTACT DETAILS *(responsible for the overall project)*		
Title:	**First Name:**	**Surname:**
A/Prof	Matthew	Tucker
Organisation:	**University of Adelaide**	

ADMINISTRATION CONTACT DETAILS *(responsible for all administrative matters relating to project)*		
Title:	**First Name:**	**Surname:**
Ms	Chelsea	Dubois
Organisation:	**University of Adelaide**	

PROJECT REPORT: *Please provide a clear description for each of the following:*

Executive Summary (200 words maximum)

A few paragraphs covering what was discovered, written in a manner that is easily understood and relevant to SA growers. A number of key dot points should be included which can be used in SAGIT communication programs.

This project was undertaken to assess the use of plant growth regulators (PGRs) in managing barley head loss, and to develop new screening tools that can assist in the delivery of new tolerant varieties.

- Using a series of check cultivars (Schooner, Compass, Planet and Spartacus) we developed a variety classification system for head loss that is relevant across multiple SA environments. Environmental and genetic factors that influence plant hormone (gibberellin) levels contribute to head loss susceptibility. Importantly, head retention does not directly relate with lodging or brackling tolerance.

- We demonstrated that Trinexapac ethyl (Moddus Evo) was effective at managing barley head loss across multiple environments. The decision to use a PGR management intervention should be considered if growers are likely to save more than 26 heads/m^2 (economic threshold) at approximately 14 days after optimal harvest date.

- Differences in barley peduncle composition correlate with head retention in moderately susceptible or susceptible such as Compass and Schooner, respectively, and these can be detected using non-destructive assays. These differences may be useful to assist breeders in making selections for new genetic solutions.

Project objectives

A concise statement of the aims of the project in outcome terms should be provided.

- Reduce statewide yield losses to environmentally induced head loss in barley
- Fine tune the effectiveness of plant growth regulators (PGRs) as a management strategy for growers
- Develop a new screening method for selection of head retention in breeding programs
- Provide growers with up to date information on head loss sensitivity in current barley cultivars.

Overall Performance

A concise statement indicating the extent to which the project objectives were achieved, a list of personnel who participated in the Research Project including co-operators, and any difficulties encountered and the reasons for these difficulties.

The project objectives were broadly successful. From a research perspective, the trials were outstanding and provided a significant amount of data for analysis and application.

- Importantly, the field data allowed us to refine a SA barley head loss susceptibility index (to be presented at the Australian Agronomy Conference, 2022).

- The cost-effectiveness of PGR application was clearly demonstrated across multiple sites, and we highlighted a pressure point where it should be considered as a management option.

- We developed a new screening method, combining compositional data and X-ray CT, to screen for new genotypes showing improved genetic head-retention.

- Wind-tunnel trials and development of structural engineering models gave promising results, but require further optimization to inform breeding strategies.

- Although the objective to "reduce statewide barley losses to head loss" was broad, the core data and outcomes provide information that should assist growers to make informed decisions that reduce head loss.

Personnel

- **UA (AFW)** – Matthew Tucker, Caterina Selva, Haoyu Lou, Ghazwan Karem, Vincent Bulone
- **UA (ECMS)** – Maziar Arjomandi, Navid Freidoonimehr, Azadeh Jafari, Harry Rowton
- **SARDI** – Melissa McCallum, Rhiannon Schilling, Kenton Porker

Difficulties

- Dr Kenton Porker accepted a new position at Field Agronomy Research prior to the end of UA619. However, he remained involved in the project and contributed significantly to the interpretation of data and the extension into UA721.

KEY PERFORMANCE INDICATORS (KPI)

*Please indicate whether KPIs were achieved. The KPIs **must** be the same as those stated in the Application for Funding and a brief explanation provided as to how they were achieved or why they were not achieved.*

KPI	Achieved	If not achieved, please state reason.
2019 Field trials sown (3 sites, 4 genotypes, 3 treatments)	Yes ☒ No ☐	
2019 Glasshouse trials sown (4 genotypes, 3 treatments)	Yes ☒ No ☐	
Wind tunnel run 1 completed	Yes ☒ No ☐	
Field and glasshouse measurements/compositional assays completed first year – Experiment 1/2	Yes ☒ No ☒	GH plants were not suitable for analysis – all experiments subsequently focused on field material
SAGIT progress report	Yes ☒ No ☐	
2020 Field trials sown	Yes ☒ No ☐	
2020 Glass house experiments sown	Yes ☐ No ☒	See above
Wind tunnel run 2 completed	Yes ☒ No ☐	
Field and glasshouse measurements completed second year – experiment 1	Yes ☒ No ☐	
Statistical analysis and head loss model development	Yes ☒ No ☐	
Final report	Yes ☒ No ☒	Report was delayed due to staff transitioning to new positions and covid delays (inability to access infrastructure)

TECHNICAL INFORMATION (Not to exceed **three** pages)
Provide sufficient data and short clear statements of outcomes.

Field Experimentation Summary

We investigated the single application of a plant growth regulator (Moddus Evo @400ml/ha) across multiple environments to reduce the incidence of head loss in a range of commercially grown barley cultivars. A total of 8 field experiments from 2019 – 2021 were sown under best district practice including optimal sowing dates, and were managed for pests, disease and nutrition. A core set of four genotypes that ranged in plant architecture and head loss susceptibility were grown in all four environments (**Figure 1**) and treatments were designed in factorial experiments to manipulate plant morphology and head loss. Repeated measurements of head loss occurred at 7 – 14 day intervals post-harvest ripe. Detailed measurements of plant morphology and growth characteristics such as crop height, internode length, peduncle length, sterility, and yield were assessed and are being prepared for peer review publication. Here we present a summary of the head loss data, and expected yield loss ranges from different cultivar and plant growth regulator treatments across all environments.

2019	2020	2021*
Cooke Plains (Mallee)	Cooke Plains (Mallee)	Riverton (Mid North)
Minlaton (YP)	Minlaton (YP)	Millicent (SE)
Riverton (Mid North)	Riverton (Mid North)	

Treatments all sites in 2019/20

- **Untreated** – An untreated control grown under normal conditions
- **GA (Gibberellic acid)** – Gibberellins are known to influence stem extension. GA was applied at 20g/ha at Zadoks growth stage 37 – 39.
- **PGR 2** - (Plant growth regulator) in the form of Moddus Evo (Trinexepac Ethyl) at 400ml/ha was applied at Zadok's growth stage 37 – 39 for head loss suppression as per label

Treatments 2021 (*SAGIT UA721 investment)

- **PGR 1** - Moddus Evo applied at 400ml/ha at the onset of stem elongation (Zadoks Growth Stage 31) for lodging control as per label
- **PGR 3** - Moddus Evo applied at 400ml/ha at the onset of stem elongation (Zadoks Growth Stage 31) and 400m/ha at GS37 (*Millicent only*)

Genotypes:

All sites: Spartacus CL (short erect plant type), RGT Planet (vigorous high yield potential), Compass (vigorous growing and susceptible to lodging and head loss), Schooner (historical cultivar with poor head retention).

Figure 1. Head loss trial sites at Riverton, Minlaton, Cooke Plains and Millicent.

Outcomes:

The probability curves describe the variability and response to treatments (**Figure 2**). The differences between treatments were insignificant at harvest ripe. However, in 50% of cultivar x environment combinations after 7-day harvest delay, untreated samples lost 6.6 heads/m^2, and PGR treated 1.1 heads/m^2 respectively. After 14 days the differential was 24.3 heads/m^2 lost in untreated, and 7.2 with a PGR treatment. After 21 days, the 50% probability line was 36.4 heads/m^2 when untreated, and 15.2 when PGR treated. After 28 days the untreated was 58.7 heads/m^2 and 24.4 when PGR treated. GA treatments significantly enhanced the probability of head loss in all cases.

Treatment effects begin to differentiate after 7 days post-harvest ripe and demonstrate that PGRs can improve harvest logistics and reduce head loss relative to the untreated control. The experimental treatment of applying gibberellic acid (GA) exacerbated head loss and can be used to understand the differences in susceptibility of cultivars. Given it is not commercially relevant, this will not be discussed in more detail.

Using the 50% probability lines for untreated and PGR treated and plotting them against time, this data reveals that for every day harvest was delayed when left untreated, 1.9 heads/m^2 were lost, whereas only 0.75 heads/m^2 were lost per day when a PGR was applied across all cultivars on average (**Figure 3**).

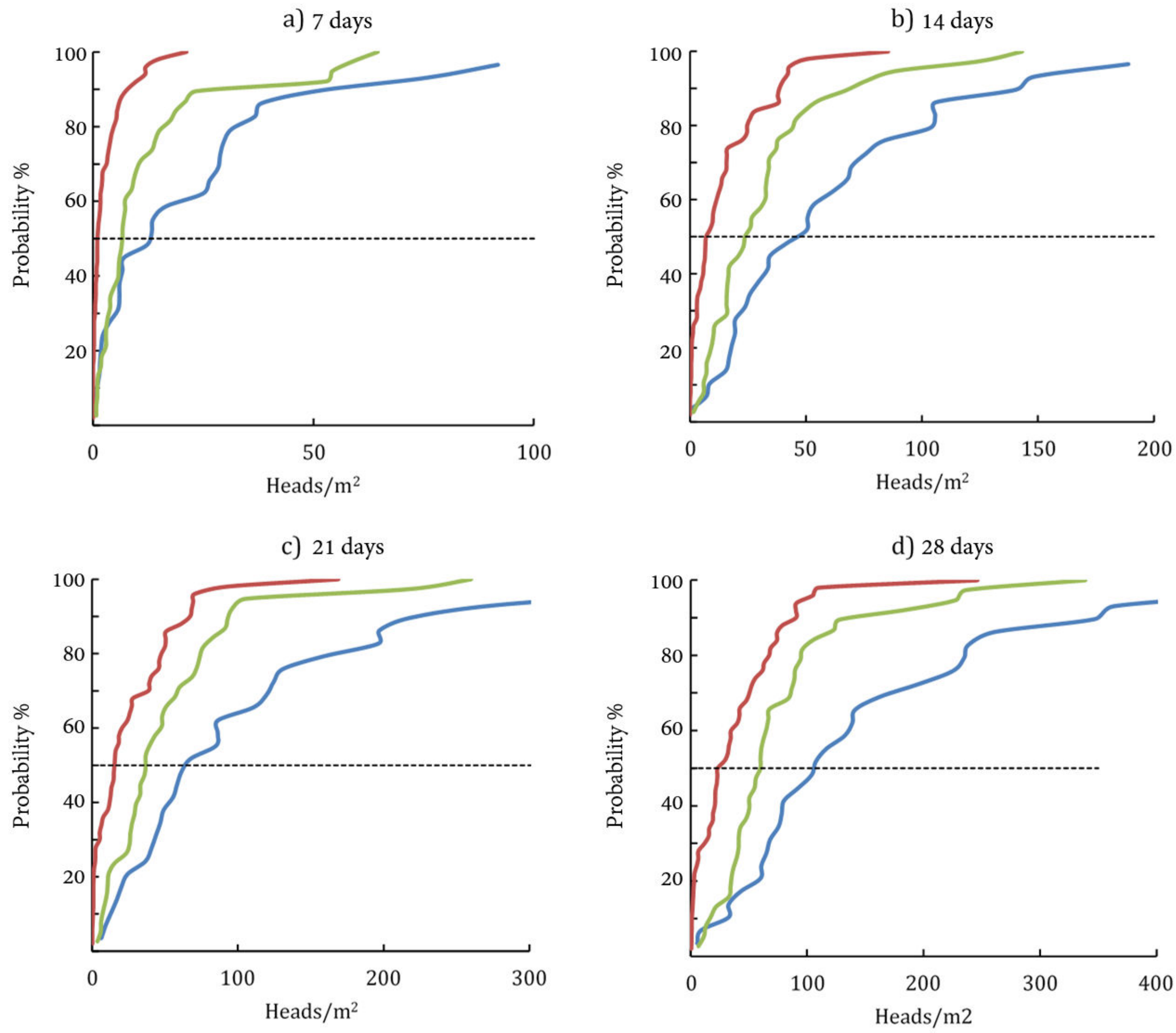


Figure 2. Probability (%) curves for head loss (head/m²) under different treatment effects Untreated control (------), GA treated (------), and PGR treated (-----) across all environments (2019 – 2021) and cultivars at a) 7 days, b) 14 days, c) 21 days, and d) 28 days after harvest ripe.

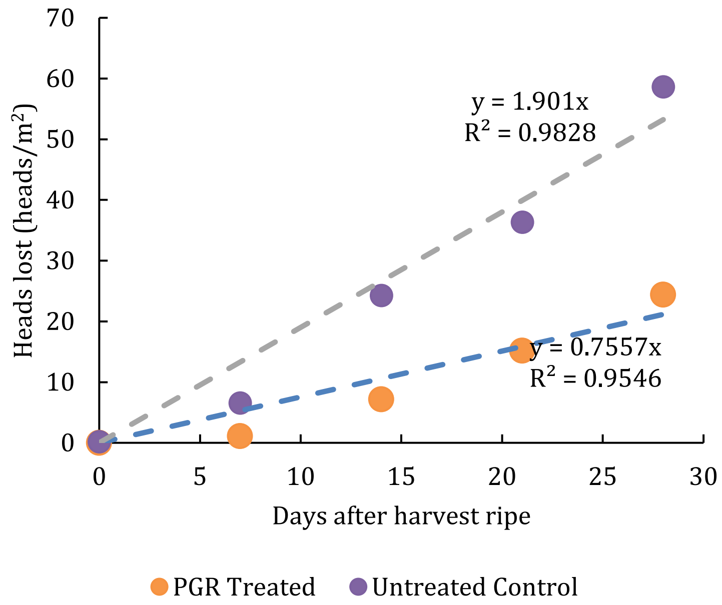


Figure 3. The 50% probability line for head loss across all environments and cultivars when left untreated or treated with a PGR.

Establishing the economic case for PGRs

Using current economic assumptions (outlined below), we found the point where PGR application was most economically viable be on average when **26 heads/m²** were saved due to intervention. This assumes each head equates to 0.7g of grain and equates to a yield loss of greater than 0.18t/ha. The analysis is based on $260/t for delivered barley price, PGR cost based on Moddus Evo at $79.30/L- ($31.72 at 400ml/ha) and an application cost of $15/ha (equating to $46.72/ha for total application). It is important to note that this is a rough rule of thumb and different varieties and finishing conditions will contribute to differences in yield components (grains per spike and grain weight). The following Table (**Table 1**) can be used to estimate differences in yield loss under different environmental conditions.

Table 1. Expected yield loss range from different finishing conditions at the economic threshold of 26 heads/m²

Yield loss range (t/ha) from 26 Heads/m²		**Grain filling conditions**	
		Mild	**Harsh**
Pre - flowering Stress	**Low**	0.40	0.28
	Mid	0.30	0.21
	High	0.20	0.14

The assumptions in this calculation are based on field data under these conditions: low stress prior to flowering = 30 grains/spike, Mid stress = 24 grains/spike, High stress = 16 grains/spike. Mild grain filling conditions equate to 48mg kernel weights, and harsh conditions 35mg Kernel weight. Note, Moddus Evo application had no significant impact on grain quality (D. McBride Honours thesis, 2020/21).

Reducing head loss with harvest logistics, PGR management and cultivar selection

Harvest timing remains one of the most effective strategies to reduce head loss. PGRs were also effective in reducing head loss but the differential between untreated and PGR applied did not reach 26 heads/m² until approximately 21 days after harvest ripe. However, this did not consider the differences in genetics, or variability across environments. Considering more than 36.4 heads/m² were lost after 21 days in 50% of variety x environment combinations, we believe this is a key point where breeders could select for genetic differences in head retention. Genetic differences in head loss were evident at this time point (21 days post-harvest ripe): Spartacus (mean 20 heads/m^2) and Planet (35 heads/m^2) were the most resistant to head loss, while Compass (47 heads/m^2) and Schooner (107 heads/m^2) were the most susceptible. These data also demonstrate that more susceptible cultivars are more variable across environments, and importantly, that PGRs were effective across multiple environments (**Figure 4**).

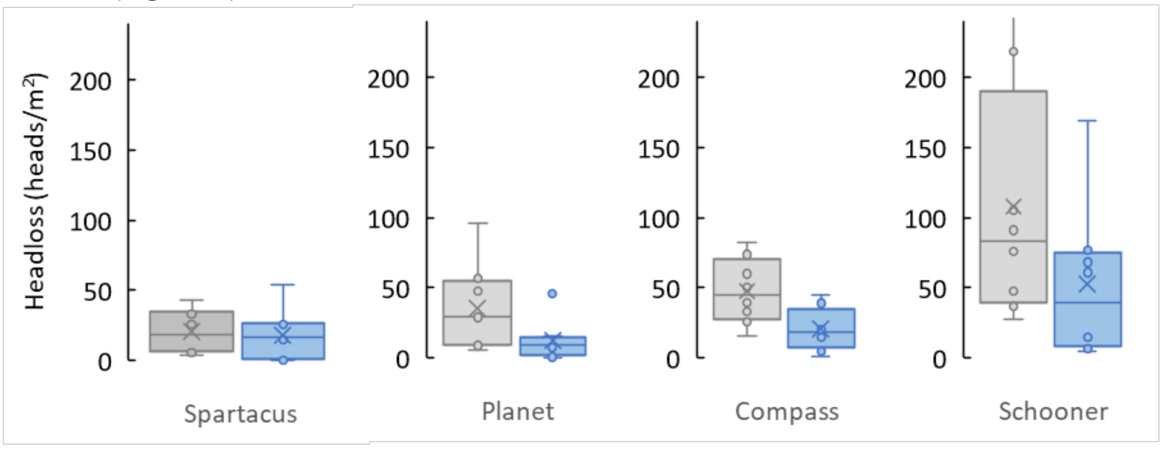


Figure 4. Variety differences in head loss across all 8 environments when left untreated (grey squares), and when treated with a PGR (blue squares) at 21 days after harvest ripe.

Based on these groupings we propose to industry a variety classification system (**See Table 2 in Conclusions below**), that uses these cultivars as benchmarks for head loss variety classifications. Head loss measurements should be taken in the period of 14 – 21 days after harvest ripe, and include moderately resistant (M) Spartacus, moderately susceptible (MS) RGT Planet, Susceptible (S) Compass, and very susceptible (VS) Schooner.

Use of a wind tunnel to model head loss in barley

A second goal of the project was to develop a method for aerodynamic analysis of grain crops that can be used to link high level crop loss features to the aerodynamic loads seen in the field (**Figure 5**). We showed that barley plants grown in a greenhouse did not have identical properties to the specimens grown in the field, as their head loss under similar wind speeds was significantly lower. Therefore, it was concluded that field-grown plants should be used for future analysis.

The effect of wind speed on head loss in field-grown material was investigated and the results showed that breakage of the peduncle was not simply correlated with increase of wind speed, but rather turbulence intensity influenced the likelihood of head loss more significantly. The head loss percentage for the tested specimens increased from 15% to 34% with increase of turbulence intensity from less than 1% to 16% (**Figure 5**). Wind tunnel experiments with added turbulence were therefore able to better replicate the natural wind conditions compared to the tests with uniform wind speed.

It was hypothesised that wake and vortex induced vibrations, in combination with fatigue and the heads striking each other, may collectively contribute to the observed increased failure rate of the specimens, all of which could be attributed to the increase in turbulence. It was also found that within the four tested barley types, Schooner had the highest head loss percentage with an average of 73% and the lowest head loss percentage belonged to Spartacus with 21%, both in agreement with field data reported above, demonstrating the validity of the experiment for replicating field results.

It was concluded that further studies and experiments could be made under a similar guide so as to develop a detailed model of the mechanical and geometric properties of barley crops to target when developing head loss resistance factors. Future work is required to analyse the effects of individual geometries, mechanical properties, and aerodynamic loads on the likelihood of head loss for the plants.

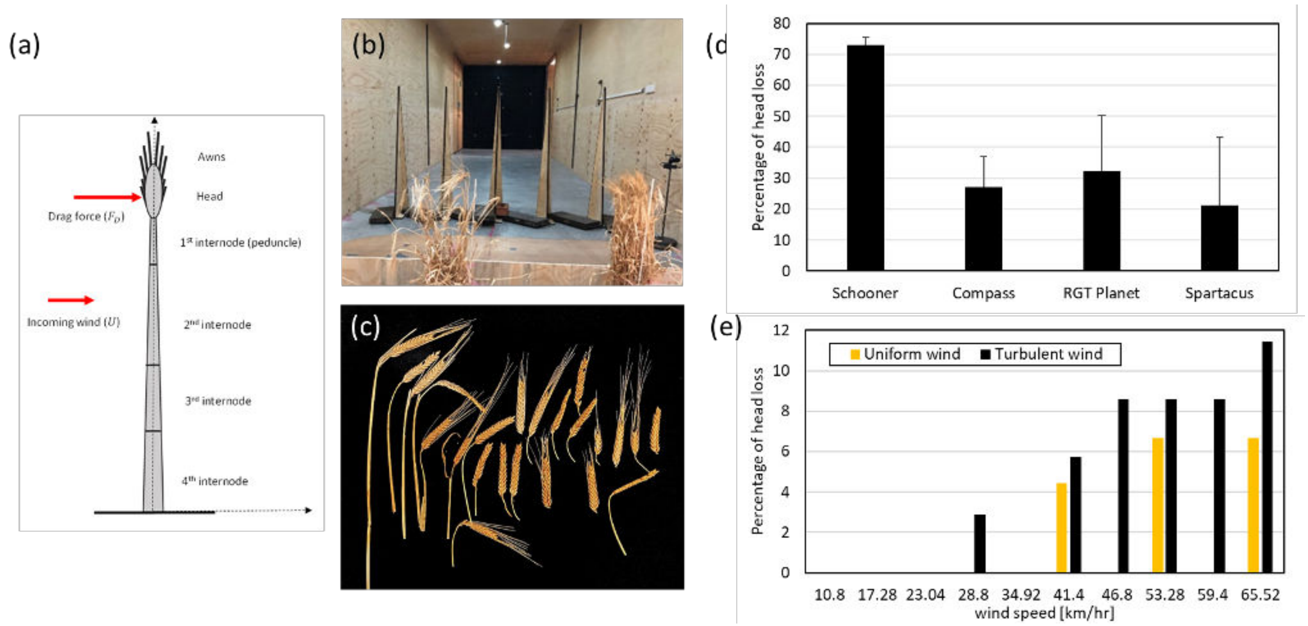
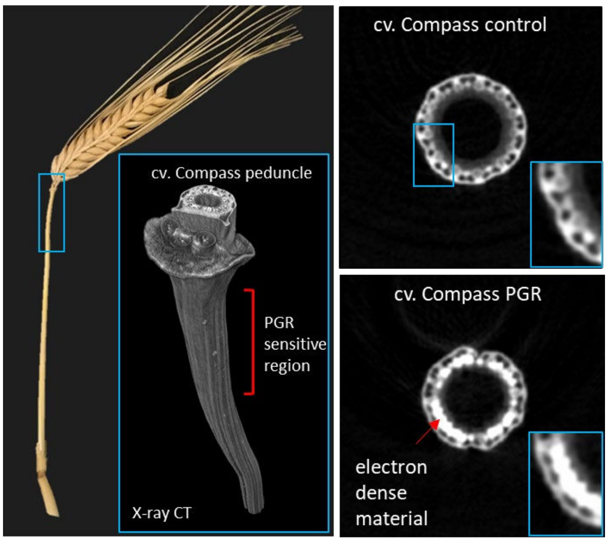


Figure 5. Field-grown barley plants were tested in a wind tunnel to establish parameters contributing to head loss. (a) Model of a barley plant stem and head. (b) Wind-tunnel showing turbulence-inducing baffles in front of field-grown plants. (c) Heads collected after turbulence-induced head loss. (d) Comparative total percentage of head loss in four barley cultivars. (e) Head loss was enhanced through a combination of wind speed and increased turbulence.

Compositional changes in the peduncle after PGR application correlate with reduced head loss

A third goal of the project was to identify biochemical markers that might be used by researchers to understand the genetic basis for head loss, or by breeders to make better selections for new varieties. As can be seen in **Figure 6**, application of PGR to Compass in field at GS37 led to the accumulation of electron dense material (bright white clumps) in the inner parts of the stem at harvest ripeness. Similar results were obtained for Schooner. This change correlated with significant reductions in head loss (after PGR treatment in the field), and is being investigated further in SAGIT UA721. This will assess whether the difference is reproducible across seasons and genotypes, and determine whether it can be used as a target for screening protocols to find new genotypes resistant to head loss.



The screening protocol is being developed further with the assistance of the Australian Plant Phenomics facility.

Figure 6: Differences in peduncle composition after PGR treatment, revealed by X-ray micro CT

CONCLUSIONS REACHED &/OR DISCOVERIES MADE (Not to exceed one page)

Please provide concise statement of any conclusions reached &/or discoveries made.

Increased peduncle length and rapid stem extension, either due to genetics or an oversupply of gibberellins, increases susceptibility to head loss.

We propose a new head loss variety classification and phenotyping method for industry, based on field observations 14 - 21 days after harvest ripe (Table 2).

Table 2. Suggested Industry Benchmarks and head loss ranges at 14 – 21 days after harvest ripe

Variety Classification	Head loss/m² Range	Suggested Benchmark cultivars
M	<10	Spartacus
MS	10 - 25	RGT Planet
S	25 - 50	Compass
VS	>50	Schooner

Gibberellin inhibition, through use of **PGR-treatment**, reduced head loss in VS and S cultivars on average by between 27 and 55 heads/m² when harvest was delayed by 21 days, and by between 3 and 23 heads/m² in MS – M cultivars

Reducing head loss by 26 heads/m² was found to be the rule of thumb where it was economically viable to apply a PGR.

PGRs are most likely to be economically viable in MS-VS cultivars and are not required in M – MR cultivars unless conditions are extremely conducive to head loss

- In Schooner, a VS variety, PGR-treatment is likely to be beneficial in the majority of environments and can save up to 60 heads/m² by 7 days and 100 heads/m² by 21 days (post harvest ripe).
- In Compass, a S variety, PGR-treatment is likely to be beneficial in some environments and can save up to 50 heads/m² by 7 days, and up to 65 heads/m² by 21 days (post harvest ripe).
- In Planet, a MS variety, there is less evidence to suggest a PGR treatment is beneficial but can save up to 26 heads/m² by 14 days and 50 heads/m² by 21 days (post harvest ripe).
- In Spartacus, a M variety, there is little to no evidence to suggest PGR-treatment is required for head loss in all environments; treatment can lead to a loss of more heads in some environments compared to untreated plants

Genetic solutions were as effective as PGR-treatment to manage head loss. However, harvest logistics has a larger impact on reducing head loss than the effectiveness of PGR in most seasons.

PGR application led to physical and compositional differences in peduncle composition that correlate with head retention in VS and S cultivars. The genetic basis for these changes in stem development should pursued to find new breeding lines that show a similar response (but without PGR application).

Other positive and negative growth effects of PGRs should not be ignored and require further analysis and investigation in each cultivar.

INTELLECTUAL PROPERTY

Please provide concise statement of any intellectual property generated and potential for commercialisation.

We are still assessing whether there is any potential IP associated with UA619, particularly in terms of head loss screening protocols. This is being tested further in UA721.

APPLICATION / COMMUNICATION OF RESULTS

A concise statement describing activities undertaken to communicate the results of the project to the grains industry. This should include:

- *Main findings of the project in a dot point form suitable for use in communications to farmers;*
- *A statement of potential industry impact*
- *Publications and extension articles delivered as part of the project; and,*
- *Suggested path to market for the results including barriers to adoption.*

Note that SAGIT may directly extend information from Final reports to growers. If applicable, attach a list of published material.

- PGRs such as Moddus Evo are economically viable in most SA environments with MS-VS cultivars, but are not required in M – MR cultivars unless conditions are extremely conducive to head loss.

- PGR treatment in-season had no obvious impact on grain quality.

- Using current economic assumptions, we found the point where PGR application was most economically viable be on average when **26 heads/m^2** were saved due to intervention.

- Wind-tunnel experiments can be used to replicate head loss in barley, but only using field-grown material. In future work this approach could be used to model head loss and identify physical parameters for improved retention.

- We have identified biochemical features of barley stems that associate with head-retention. Screening protocols are being developed in partnership with breeders to screen for new cultivars that are more resistant to head loss in the field.

- Three main publications have been completed to date:
    - Detailed industry report regarding PGR use for head-retention in barley
    - Australian Agronomy Conference report “Development of a barley head loss susceptibility index for Southern Australia”
    - Honours thesis (Danielle McBride) “The impacts of plant growth regulators on grain quality in malting barley (*Hordeum vulgare* L.)”

POSSIBLE FUTURE WORK

Provide possible future directions for the research arising from the project including potential for further work and partnerships.

The work from UA619 has already been extended via several initiatives including:

1. A one-year extension to collect extra field data via SAGIT UA721

2. Pilot funding from the Australian Plant Phenomics Facility for X-ray CT scanning of peduncles (Project #0628)

3. Two-year ARDC OzBarley project aimed at consolidating phenotypic data from a panel of Australian breeder-relevant cultivars. These are being assessed for head-retention traits.

We are also negotiating with an Australian breeding company to test several of the screening assays using current and new cultivars. At an applied level, this may allow us to score the likelihood of head retention, while at a fundamental level, this could help us to identify new genes for deployment in new cultivars.

Several manuscripts based on UA619 results and screening methods will be targeted to international journals. Ideally this will form the basis for an ARC Linkage Project application with an industry partner.