Heat stress and harvest heroes: cultivating tolerance in wheat through innovative genetic breeding for enhanced grain fill

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Take home message

- This research tackles the threat of rising temperatures to wheat production, focusing on the vulnerability of the critical grain fill growth stage during milk and dough development
- Elevated daytime temperatures shorten the grain-filling period, while high night-time temperatures disrupt respiration, induce rapid maturity and compromise photosynthesis
- This project involves an adaptive approach towards screening for heat tolerance, rigorous phenotyping, creating new heat-tolerant varieties, and deepening genetic understanding to equip wheat growers with genetic tools to remain productive when faced with extreme temperatures during the growing season
- Tolerant commercial varieties like LRPB Hellfire⦶, LRPB Flanker⦶, and LRPB Reliant⦶ express small yield variability in various conditions, and the genetic stability of Mace⦶ and Scepter⦶ indicates consistent yield performance
- Imported and locally developed genetic material in Australia demonstrates promising potential as a robust defense against extreme heat events
- A deeper understanding of leaf waxiness (glaucousness) in environments like Kununurra, predicted to occur in the future, can expedite the identification of wheat lines with superior heat tolerance.

Rationale behind research

As climate change intensifies, extreme heat events are becoming more frequent, posing a significant threat to wheat production. Wheat is particularly sensitive during the grain fill (milk and dough) growth stage, and extreme heat during this period can severely impact both crop yield and quality. Such heat events disrupt the translocation of nutrients and the synthesis and deposition of starch, negatively affecting kernel size.

Daytime high temperatures during grain fill can impact a wheat plant though:

- **Reduced grain filling duration**–- elevated daytime temperatures shorten the critical grain-filling period, limiting the wheat plant's ability to store nutrients and resulting in lower yield.
- **Kernel weight reduction** – elevated daytime temperatures during grain filling significantly reduce the weight of individual wheat kernels, impacting overall yield and quality.

Night-time high temperatures during grain fill can impact a wheat plant by:

- **Impairing respiration** – night-time high temperatures interfere with the plant's respiration, hindering the efficient use of stored sugars for grain development, affecting overall quality and yield.
- **Inducing senescence** – prolonged exposure to high night-time temperature causes premature aging in wheat plants, shortening the grain-filling period and impacting final yield.
- **Compromising photosynthesis** – elevated night-time temperature disrupts the balance between photosynthesis and respiration, reducing the plant's ability to produce and store energy, further contributing to reduced grain quality and yield.

Combating increasing temperatures across Australia

To ensure wheat producers across Australia can combat rising temperatures, our research employs a multi-stepped approach:

1. Screening for heat tolerance: genotypes are assessed under high temperature environmental conditions using a three-tiered technique. This involves a combination of large-scale field trials where a staggered time of sowing approach is used to induce a natural heat stress. A small subset of the most promising genetic material is further assessed under controlled conditions such as in-field controlled environment chambers and standard glasshouse facilities.
2. Phenotyping and validation: the performance of 200 of the most promising lines is evaluated at key sites across Australia in conjunction with Intergrain's Multi-Environment Trial (MET) network, considering phenology, UAV imagery, yield, grain weight, and screenings.
3. Creating new genotypes: crosses among lines with high genetic potential are conducted, contributing to ongoing efforts to develop heat-tolerant wheat varieties.
4. Genetic understanding: by 2025, we aim to figure out the unique code (haplotype structure) in wheat's genetic makeup, which acts like a special pattern to help understand how different traits for heat tolerance are connected in the DNA.

What did well under high temperature stress in 2022 and 2023?

Building on the findings of the GRDC funded initiative US00081, which initially focused on heat tolerance during anthesis, the current pre-breeding project extends its scope to scrutinize grain filling more closely. By leveraging the successful three-tiered phenotyping technique developed in the previous project, we can effectively assess the performance and physiological responses of wheat genotypes in hot environments. Unlike traditional single-tiered approaches conducted in controlled environments like a glasshouse, this research prioritises field conditions for assessment. Wheat varieties that exhibited success in the field were further evaluated in both glasshouse and in-field controlled environment chambers for a comprehensive analysis.

In 2022, a mild year, trials nationwide experienced minimal heat stress, except for Kununurra. In contrast, 2023 proved to be a challenging year, characterised by elevated temperatures and prolonged periods of high temperature during grain filling across the country. This resulted in a discernible impact on harvest yields. Table 1 illustrates the temperature variances for each year in relation to their respective harvest yields for Narrabri in north-west NSW. The average yield decrease in the challenging year (2023) amounted to 1.8 t/ha, while in the favorable year it was 1 t/ha (2022).

Table 1. Planting dates, average trial yield (t/ha), and maximum and minimum in-season temperatures for each time of sowing (TOS) at Narrabri in 2022 and 2023.

Year	TOS	Planting date	Max in-season temperature (°C)	Min in-season temperature (°C)	Average trial yield (t/ha)
2022	1	27/05/2022	32.8 on 27/11	-2.1 on 10/07	6.8
	2	19/07/2022	33.8 on 19/11	-1.6 on 19/07	5.8
2023	1	30/05/2023	32.4 on 2/10	-3.5 on 20/07	4.6
	2	19/07/2023	36.8 on 13/11	-3.5 on 20/07	2.8

Table 2 illustrates the yield decline between normal and late planting for current commercial lines at Narrabri, NSW. Lines with a low yeild reduction between years, such as Valiantⅆ, Rockstarⅆ, Maceⅆ and Scepterⅆ showcased their versatility and ability to withstand different years and envirnoments under stress while maintaining yield stability.

Table 2. Yield reduction (t/ha) between normal and late planting and yield stability (YS) ranking across years for current commercial lines at Narrabri, NSW in 2022 and 2023.

Genotype	Yield reduction (t/ha)		YS Rank
	2022	2023	
Valiant CL Plusⅆ	2.13	1.21	1
Rockstarⅆ	2.17	1.95	2
Maceⅆ	1.66	1.56	3
Scepterⅆ	1.45	1.48	4
Borlaug 100ⅆ	1.67	1.89	5
Catapultⅆ	1.21	1.75	6
Cutlassⅆ	1.08	1.90	7
Suntopⅆ	0.93	1.89	8
Sheriff CL Plusⅆ	1.31	2.28	9
Coolahⅆ	0.13	1.15	10
Condoⅆ	0.60	1.74	11
Vikingⅆ	0.49	1.68	12
Beckomⅆ	1.10	2.45	13
LRPB Stealthⅆ	0.49	1.89	14
Havocⅆ	0.47	1.99	15
LRPB Hellfireⅆ	-0.35	1.31	16
Sunchaserⅆ	0.30	2.09	17
Vixenⅆ	0.64	2.43	18
LRPB Flankerⅆ	-0.19	1.60	19
Sunmaster	1.17	3.20	20
LRPB Reliantⅆ	-0.56	1.69	21
LRPB Mustangⅆ	0.52	2.98	22

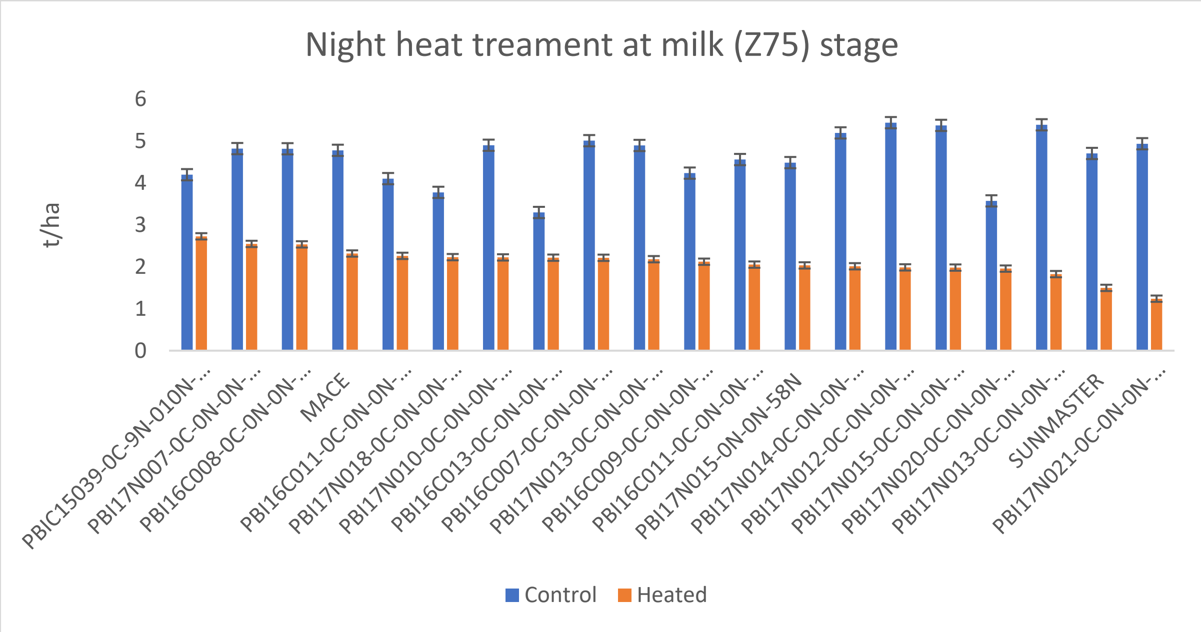
The use of Intergrain's MET network is crucial for assessing the heat tolerance of wheat varieties because it provides a comprehensive evaluation across diverse environmental conditions. Table 3 depicts the best-performing wheat varieties across 24 locations in 2022. This extensive trial network considers variations in temperature, precipitation, and other environmental factors, providing a more realistic representation of the challenges faced by wheat crops in various regions.

Table 3. The yield performance based on average ranking (Av rank) of 14 candidate lines with high Genomic Estimated Breeding Values (GEBVs) for heat tolerance across all trial sites for each state and the 24 sites nationally in Intergrains (IG) multi-environment trials, and at 3 times of sowing (TOS) at Kununurra in 2022 expressed as % Sunmasterⓓ (best performing check nationally in IG's 2022 MET).

	Average yield ranking					**Yield at Kununurra (% Sunmasterⓓ)**			
	NSW	**SA**	**VIC**	**WA**	**All sites**	**TOS1**	**TOS2**	**TOS3**	**Average all TOS**
Genotype	**4 sites**	**4 sites**	**5 sites**	**11 sites**	**24 sites**				
ZWB11-132	8.4	11.0	9.8	10.1	9.8	130.8	108.4	141.8	127.0
ZWB13-171	5.6	5.8	6.2	9.3	7.5	123.7	111.3	138.9	124.6
ZWB13-261	12.7	19.0	17.2	11.5	14.5	117.3	108.6	145.8	123.9
PBI17N008-0N-0N-35N	9.2	11.3	6.6	10.7	10.0	115.0	112.5	131.3	119.6
ZWW11-061	8.4	13.8	12.2	8.9	10.6	115.4	98.7	140.9	118.4
ZWB13-238	13.4	14.0	14.8	13.6	13.7	112.4	101.8	139.4	117.9
Maceⓓ	10.7	5.3	16.8	9.8	11.1	111.5	106.0	136.0	117.8
Vixenⓓ	9.9	7.8	12.2	8.2	9.8	98.3	106.4	146.2	117.0
PBI17N006-0N-0N-96N	7.9	11.0	6.0	9.8	8.9	114.8	103.2	131.0	116.3
ZWW10-069	10.0	12.8	9.2	12.4	11.5	115.5	110.2	121.9	115.9
ZIZ12-045	6.4	13.0	8.0	11.1	9.8	122.4	103.1	118.7	114.7
PBI17N015-0N-0N-55N	9.3	10.8	7.4	8.5	8.9	111.6	105.8	125.7	114.4
ZWB12-009	8.2	9.5	5.6	9.3	8.1	106.1	113.0	123.0	114.0
PBI19N007-0N-6N	15.0	13.8	15.2	16.8	16.2	106.0	106.0	129.5	113.8
PBI17N009-0N-0N-46N	5.9	3.8	5.0	10.1	7.0	111.0	94.4	123.7	109.7
Scepterⓓ	10.5	5.3	10.4	7.3	8.3	101.2	97.7	105.4	101.4
Sunmasterⓓ	4.4	2.0	4.8	6.2	4.8	100.0	100.0	100.0	100.0

Exploring the impact of night-time temperatures on wheat, especially during critical growth stages like grain filling, is a crucial avenue for further investigation. This is a key time when plants recover, gearing up to combat high daytime temperatures. Plants photosynthesize during the day to produce energy, and at night they respire to rest. Figure 1 depicts the impact on yield when this process is disrupted, removing the opportunity for recovery during the night. Treatments were administered during the milk (Z75) and dough (Z83) growth stages, maintaining temperatures 5°C above the ambient temperature for five consecutive nights. Most genotypes tended to be more susceptible to heat at the milk stage when the translocation of nutrients and starch building is at its highest. Interestingly, those tolerant to the milk treatment stage were not as tolerant at the dough stage, indicating the presence of distinct genetics influencing both stages. The average overnight temperature for the milk and dough growth stage treatments were 22°C and 18°C, respectively.

a)



b)

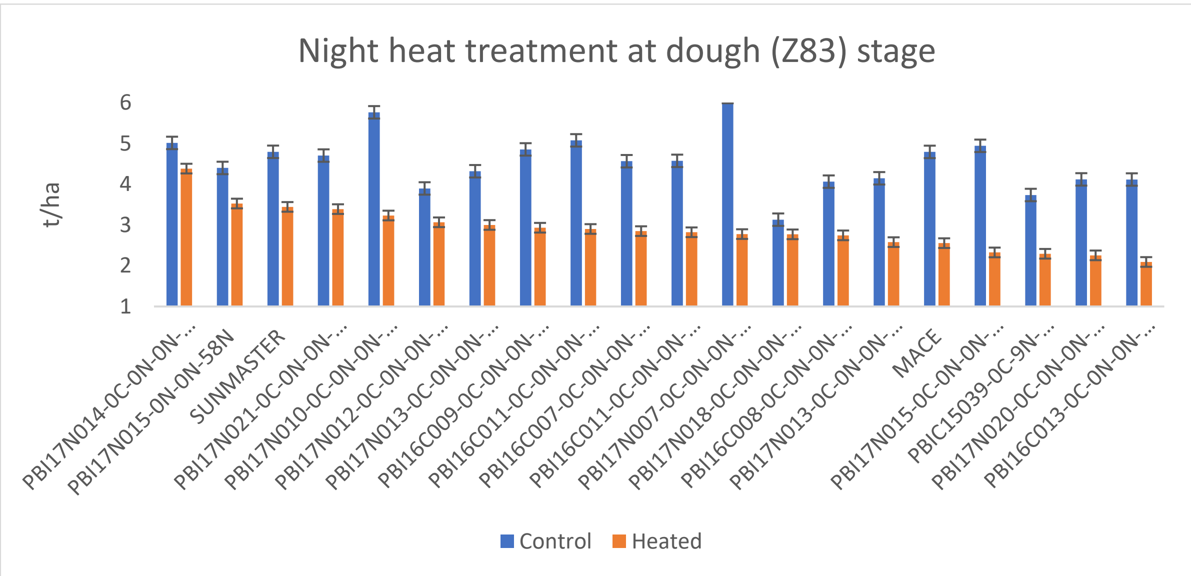


Figure 1. The grain yield (t/ha) for night-time heat treatment of 18 breeding lines, Mace(b, and Sunmaster(b at the a) milk (Z75) and b) dough (Z83) growth stages in Narrabri during 2023. Genotypes are ranked from highest to lowest yield based on performance under heat treatment. Vertical error bars represent the standard error.

Understanding the genetic basis of heat tolerance in wheat is essential for the development of sustainable and commercially viable cultivars by plant breeders. Figure 2 presents a focused exploration of Quantitative Trait Loci (QTL) associated with grain yield, as identified through a Meta-Genome Wide Association Study (MetaGWAS) analysis. Notably, the Manhattan plot reveals significant spikes on chromosomes 3A and 4A, indicating specific genomic locations strongly linked to heat tolerance. On the x-axis, each point corresponds to a genomic location, on different chromosomes. The y-axis shows the statistical significance of associations between genetic markers and heat tolerance.

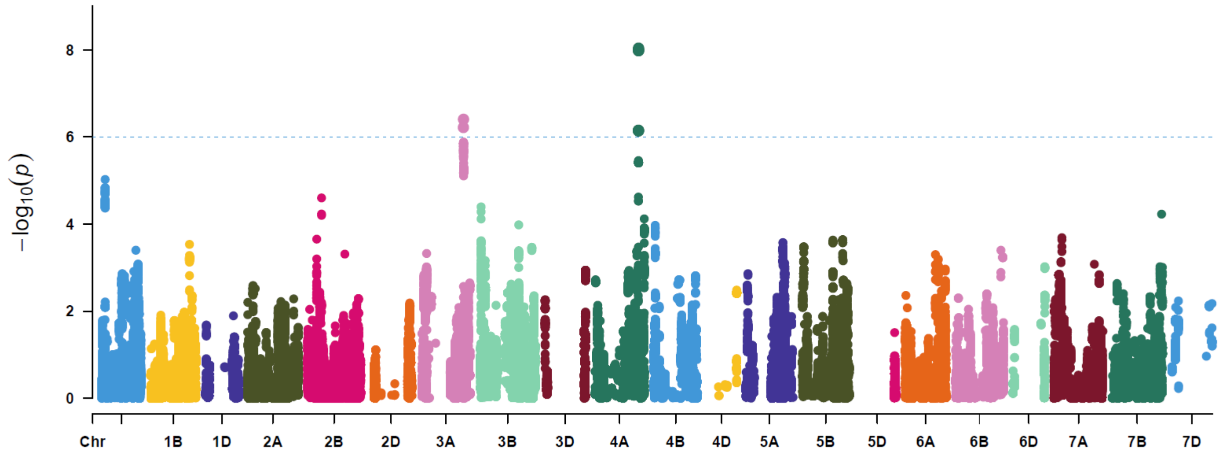


Figure 2 Manhattan plot showing the significant grain yield QTL detected with the MetaGWAS analysis that uses the late time of sowing (TOS2) in all environments. The x-axis indicates genomic locations on different chromosomes, while the y-axis represents the statistical significance of associations between genetic markers and heat tolerance.

This MetaGWAS emphasises regular sowing times across diverse environments, enhancing the robustness of its findings. Each point on the plot corresponds to a unique genomic location, strategically chosen to highlight regions of the genome influencing grain yield. By incorporating data from various environments, the analysis provides comprehensive insights into genetic markers that play a pivotal role in influencing grain yield under different conditions, all while maintaining adherence to standard sowing times. The specific identification of QTLs on chromosomes 3A and 4A suggests targeted regions for further investigation, contributing valuable knowledge to the development of heat-tolerant wheat cultivars.

Significance of the research for growers across Australia

1. With the escalating impact of climate change, more frequent extreme heat events pose a threat to wheat production. Understanding how wheat responds is essential for plant breeders to provide growers with genetically resilient varieties for a sustainable future.
2. The research has a direct impact on crop yield, aiding growers to optimise yield under challenging climatic conditions by identifying wheat genotypes that are heat tolerant during the grain filling growth stage(s).
3. Beyond quantity, the research is focused on enhancing wheat quality, ensuring the new genotypes identified maintain or improve quality attributes.
4. Equipped with insights into heat-tolerant wheat varieties that are currently available, growers can make informed decisions about resource allocation, optimising inputs like water and fertiliser.
5. Recognizing the risk extreme temperature events pose to crop production, the implementation of heat-tolerant varieties identified through this research acts as a vital risk mitigation strategy for growers.
6. Contributing to the development of wheat varieties that withstand challenging conditions, the research safeguards the economic sustainability of wheat growers through improved yield and higher-quality crops.

In summary, this research addresses the practical needs and challenges of wheat growers in Australia, providing solutions to enhance both the quantity and quality of yield, ensuring the sustainability and profitability of their operations in a changing climate.

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