Genomic prediction to accelerate breeding potential for increased transpiration efficiency in wheat

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Abstract

Rainfall and availability of water in rain fed environments is predicted to become more variable and limited over the coming decades, highlighting the need to produce more water efficient crops to sustain or improve yields. Using structured wheat populations, multiple experiments were conducted to measure transpiration efficiency of genetically diverse families. Phenotypic performance and genotypic profile in a nested association mapping population were used to predict genetic performance of families with other genetic backgrounds. Lines from the predicted top and bottom tails for TE were phenotyped in an independent experiment. While this experiment was subjected to several heat shocks events that may have impacted genotype ranking, substantial differences between the top and bottom tails were observed in seven of the 12 studied families. Significant differences were found for three of the previously untested families. Overall, results suggest that genomic selection has potential to accelerate the breeding of wheat lines for improved water use efficiency, especially when working with related families as is the common practice in breeding.

Keywords

Wheat, transpiration efficiency, genomic selection, genomic prediction, breeding, water use.

Introduction

Water availability in rain fed environments is a critical limiting factor for grain yield of wheat (Triticum aestivum L.) in Australia (Chenu et al. 2013). Projected increases in the severity and frequency of heat- and water-stress events due to climate change increase the imperative to improve drought and heat adaptation (Lobell et al., 2015; Watson et al., 2017; Ababaei and Chenu, 2020; Collins and Chenu, 2021). A bottleneck to crop improvement is the resource intensive and relatively low-throughput techniques used for phenotyping large breeding populations for some relevant traits (Chenu et al., 2018). Increasing the efficiency with which crops utilise water can increase yields when grown in water-limited environments, especially where crops rely on stored soil moisture (Condon et al., 2002; Rebetzke et al., 2009; Leakey et al., 2019; Collins and Chenu, 2021). In such environments, crops that conserve water early during the crop cycle may retain more soil moisture for use later in the season, when water can be more effectively used to produce grain yield (e.g. Angus and van Herwaarden 2001; Collins et al. 2021; Manschadi et al. 2006). However, transpiration efficiency (TE) can be expensive or difficult to phenotype for large numbers of genotypes (Chenu et al., 2018; Fletcher et al., 2018). With recent progress in the field of plant genomics, genomic selection is increasingly used in conventional breeding programs, particularly for difficult to phenotype traits. The prediction of phenotypic performance using molecular markers has the potential to accelerate selection for complex traits such as TE by allowing selection without the need for extensive phenotyping of entire breeding populations (Bassi et al., 2016).

In this study, a genome wide association study (GWAS) was conducted using a nested association mapping (NAM) population to estimate marker effects for TE based on results from two lysimeter experiments. Genomic prediction was then used to identify high and low TE tail genotypes in both previously tested and previously untested families. The lines that were predicted to have contrasting TE were then phenotyped in a third subsequent experiment to evaluate the robustness of the approach.

Material and methods

Three experiments were conducted in a lysimeter (Chenu et al. 2018) to measure TE of wheat genotypes (Table 1) following the short-trial duration methodology described by (Fletcher et al., 2018). The first two experiments (Exp-1 and Exp-2) were used to phenotype TE of the 457 lines from a NAM population (2.32 replications per genotype across the two trials). The NAM was developed by

crossing (i) the high-TE cultivar Suntop (Fletcher and Chenu, 2015; Fletcher et al., 2018; Collins et al., 2021) that is well adapted to the north-eastern region of the Australian wheatbelt with (ii) 12 donor lines contributing heat and drought adaptations (Table 1; Fletcher 2020).

As no significant genotype x environment interactions were observed for TE between Exp-1 and Exp-2, data from both experiments were analysed together in a genome wide association study (GWAS). Additive effects of 2,446 SNP markers were estimated for TE (Fletcher, 2020).

Using genomic prediction, the phenotypic performance of lines were predicted for four families from a Suntop NAM population (training population). Predictions were also made for four un-tested families from a Scout NAM population and four un-tested families from a Mace NAM population (Table 2). For each of the 12 families, lines with the 10 highest and 10 lowest TE predictions (i.e. tails) were selected. These were then phenotyped in a third lysimeter experiment, with a randomised fully-replicated design (two replicates per genotype).

In all experiments, plants were grown in an automated high-throughput phenotyping lysimetry platform in a polyhouse (Chenu et al., 2018). The automated system watered all pots once per day at 6 am and weighed all pots every 10 minutes, using the difference in pot + reservoir weight to calculate water used. This system provided an unlimited continuous water supply to all pots. Plants were grown in 4L ANOVApots (http://www.anovapot.com) filled with the UQ23 standardised potting medium with added fertiliser to provide a non-limiting nutrient growing environment. The soil surface of all pots was covered using plastic cling film, to minimise the evaporation of water from the soil surface and to control weeds. Four seeds were sown per pot through cross-shaped holes cut in the film. Three pots with no plants were included in each experiment to measure incidental water loss as a control.

Phenotypic data were collected at harvest, which occurred between the stages of flag leaf emergence and mid-booting (Z40-45; Zadoks et al. (1974). TE was calculated as the ratio of above-ground dry biomass per cumulative kilogram of water transpired from sowing until harvest.

Table 1. Experiment characteristics including sowing date, trial duration (in days and degree days (°Cd) after sowing), environmental conditions (mean temperature, day-time vapour pressure deficit (VPD), radiation presented as mean daily radiation from sowing to harvest, and (cumulative radiation for the same period in parenthesis), and plant characteristics at harvest for the trial mean (and the reference parent, Suntop, in parenthesis).

Exp.	Sowing date	Trial duration		Mean temp.	Mean vpd	Radiation	Leaf area	Zadoks stage	Above- ground biomass	Cumulative transpiration	
		(d)	(°Cd)	(°C)	(kPa)	(MJ m ⁻²)	(cm^2)	-	(g)	(kg)	
Exp-1	7/05/2016	60	917	19.9 (15.9)	1.23	7.85 (478.9)	1191 (1159)	41.3 (44)	8.7 (9.1)	1.72 (1.69)	
Exp-2	18/07/2016	51	793	20.1 (15.6)	1.29	10.27 (533.8)	1146 (1101)	38.7 (38)	8.1 (8.6)	1.85 (1.95)	
Exp-3	11/08/2017	50	759	25.0 (19.2)	2.36	12.63 (644.1)	909 (685)	41.6 (41)	6 (6.3)	1.92 (1.83)	

Table 2. Structure and number of lines per family for three NAM populations, with Suntop, Scout and Mace as respective reference parents. All lines from the NAM with the reference parent Suntop are presented in the first row and were previously phenotyped in Exp-1 and Exp-2 for a GWAS analysis (training population). Lines in bold font had their TE performance estimated by genomic prediction, and predicted tails for these families were then phenotyped for TE in Exp-3. Tails selected from four of the Suntop NAM families include in Exp-1 and Exp-2 (training population) were tested again in Exp-3.

	Dharwar dry	Drysdale	SB062	Seri M82	UQ114	EGA Wylie	FAC10-16	EGA Gregory	ZWB10-37	ZWW10-128	Westonia	ZWW10-50
Suntop	52	52	52	52	42	40	42	42	41	42	-	-
Scout	52	51	51	52	42	-	-	-	-	-	-	-
Mace	44	49	51	52	-	-	-	-	-	-	40	38

Results and discussion

A genome wide association study (GWAS) of the Suntop NAM population (Exp-1 and Exp-2) estimated marker effects for TE and identified 20 QTL for TE, from which 8 had an effect accounting for greater than 4% of the phenotypic variation for TE (Fletcher, 2020). The TE effects for all 2,466 markers were calculated and used in genomic prediction to select tails of genotypes in the 12 families. These genotypes were phenotyped in Exp-3. Eight families from the Mace NAM and Scout NAM had not been phenotyped previously in Exp-1 and Exp-2 (Table 1). In addition, tails for four of the previously tested Suntop NAM families were re-evaluated in Exp-3. The top tail of most families tended to have a greater, while not always significant, median TE than the bottom tail group (Figure 1). Significant differences between the top and bottom tails were found in three families (Scout x SB062, Scout x Seri M82 and Mace x Dharwar Dry). None of these three families had been previously phenotyped, highlighting the practical application and power of genomic prediction.

The eight families not tested in Exp-1 or Exp-2 each had either Mace or Scout as a parent, neither of which is represented in the training population (Suntop NAM; Table 2). Thus, lines from each of these families shared only one parent with one of the families from the training population. In conventional breeding programs, genomic prediction is typically performed with a training population that includes lines sharing the same genetic backgrounds as the target population. In such more-closely interrelated breeding populations, genomic prediction is thus expected to be more consistent with the actual phenotypes than those observed in this study. Hence, selection using GWAS prediction for TE in conventional breeding populations is likely to be even more successful than results from the current study suggest.



Figure 1. Transpiration efficiency of the predicted tail groups of the Suntop (a), Scout (b) and Mace (c) families in Exp-3. Boxes extend from the 25^{th} to 75^{th} percentile, horizontal lines represent median values of each group, vertical extending lines denote adjacent values (i.e., the most extreme values within 1.5 interquartile range of the 25^{th} and 75^{th} percentile of each group); dots denote observations outside the range of adjacent values. Significant differences between top and bottom tails of a family are indicated on the x-axis (P values: * = 0.05, ** = 0.01, *** = 0.001).

Genetic variation for TE in Exp-3 was reduced compared to other experiments, probably due to the more extreme environmental conditions. Plants in Exp-3 experienced much higher temperatures including heat-stress events than in Exp-1 and Exp-2, especially in the 3-4 weeks leading up to harvest. Heat stress may have impacted genotype ranking and partly explain unexpected results such as the failure to select tails with significantly higher TE in several of the Drysdale NAM families predicted to be high TE using genomic selection (Figure 1c). The relationship between predicted and observed performances in Exp-3 for lines of the Suntop family was positive but not statistically significant (data not shown). In contrast, a significant relationship was observed in another independent experiment with milder temperatures (Fletcher 2020).

Conclusion

Eight QTL for TE with an effect greater than 4% were identified in a NAM population phenotyped in two lysimeter experiments. The accuracy and consistency of genomic predictions using data from these two experiments were tested in a previously untested population grown in an independent experiment. For seven of the 12 studied families, the tails with top predicted TE exhibited substantially higher TE than the bottom tails. For three of the families not tested in the training experiments, the TE difference between the predicted top and bottom tails was statistically significant. These results are encouraging considering that (i) the validation experiment was subjected to several

heat shocks that may have impacted genotype ranking, and that (ii) breeders typically use genomic predictions from a training population much more representative of the whole target population than was used here. Thus, there could well be potential to use genomic selection for TE to improve adaptation of wheat to water-limited environments.

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