Is Vrn-H1 a missed opportunity for southern Australian barley growers?

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Abstract

Over the last decade there has been a trend to earlier sowing of cereals. Growers are seeking varieties that develop slower to match needs of minimising reproductive frost risk, avoid high grain-filling temperatures and terminal water stress. Historically barley breeders focused on developing cultivars with a short mean duration to flowering through direct and indirect selection of photoperiod sensitivity (*Ppd*) alleles and insensitive vernalisation (*Vrn*) alleles. This paper discusses the concept that the lack of winter *Vrn-H1* alleles in Australian cultivars may be a missed opportunity for southern Australian barley growers and presents the history of winter barleys in Australia, and the merits of re-introducing winter *Vrn* alleles into breeding programs. Based on preliminary data it is possible to achieve a similar flowering date, and competitive yields with different combinations of phenology genes including winter *Vrn* alleles from earlier sowing.

Keywords

Phenology, sowing time, vernalisation, photoperiod.

Introduction

Matching crop phenology with availability of resources and avoiding stress events during flowering and grain-filling are key factors influencing yield and crop adaptation (Richards 1991). In Australia, climatic conditions in the temperate cereal production areas define the periods for sowing and the phenological events which follow, such as the transition from the vegetative to reproductive phase and flowering time. Various genes control the timing and duration of developmental phases, mainly attributable to photoperiod (*Ppd*), low temperature vernalisation (*Vrn*) response genes, and earliness *per se* (*Eps or Eam*) loci (Campoli and von Korff 2014). Yield improvements have been achieved through direct selection of yield based on traditional May sowing dates and an appropriate flowering time, resulting in indirect selection for phenology gene combinations favouring this farming system environment. The recent decade trend of earlier sowing dates using current varieties could result in undesirable early flowering and slower developing varieties with new developmental gene combinations may be necessary to fit this farming system. This paper aims to discuss the merits of utilising *Vrn* alleles from winter barleys to improve yield in early sown crops.

What is a winter barley?

Vernalisation is the requirement for a period of exposure to low temperature before the plant apical meristem will transition from vegetative to reproductive development. Vernalisation alters the length of the vegetative phase, and hence floral initiation which indirectly affects the duration of subsequent pre-heading phases. Genotypes differ in low temperature requirement in the duration and intensity of effective vernalising temperatures from no requirement in "traditional spring types" to 3-12°C for an extended period in winter types (Garcia del Moral et al. 2002). The winter type is predominantly controlled by the three vernalisation genes *Vrn-H1*, *Vrn-H2*, and *Vrn-H3*. Different *Vrn-H1* genes are the primary driver of vernalisation response (Trevaskis et al. 2007), and interact with *Vrn-H2* and *Vrn-H3* vernalisation genes and the photoperiod pathway. Unlike spring barley, true winter types require adequate cold stimulus for the *Vrn-H1* gene to induce development of the reproductive meristem, and *Vrn-H2* overrides *Ppd-H1* (photoperiod response) if exposed to sufficiently low temperatures (Distelfeld et al. 2009). Spring barleys do not have a recognised vernalisation requirement associated with *Vrn-H1* and *Vrn-H2* so flowering is dependent on photoperiod (*Ppd-H1*) and *earliness per se* genes.

Breeding and evaluating winter barleys

The first 'modern' winter barley in Australia was Ulandra, selected and released in 1987 by NSW DPI (Read and Macdonald 1987) followed by Urambie in 2005, a semi-dwarf feed barley aimed at both dual purpose and grain only situations suited to early March to mid-May sowing in NSW. Prior to this, most adapted Australian cultivars and spring types introduced from Europe, Canada, and Japan have either no or a minimal vernalisation requirement (Boyd et al. 2003). A study revealed there is limited variation in *Vrn-H1* in spring cultivars grown in southern Australia, and the majority have a deletion of the winter type at *Vrn-H2* (Porker unpublished). Boyd et al. 2003 concluded best adapted barley cultivars for Australian low-medium rainfall environments are early-maturing spring types, that combine a short vegetative phase, exhibit a high photoperiod response and limited vernalisation requirement. This plant ideotype was easy to select in summer selection nurseries utilised by Australian breeding programs, based on selection for early flowering and limited tillering under long hot days. However, this traditional ideotype breeding approach is challenged by the knowledge that a recent release, Compass, carries a photoperiod insensitive allele, but has achieved a short mean duration to flower in winter plantings by combining other developmental genes. It is therefore possible to achieve desired flowering dates with phenology gene combinations other than those previously explored. True winter types sown in summer selection nurseries fail to flower and produce viable seed, meaning any lines possessing winter *Vrn-H1* alleles do not progress to yield evaluation trials. The lack of intogression of the winter *Vrn-H1* allele may be a missed opportunity for southern Australian barley growers.

The case for winter barleys?

Early sowing evaluation trials were conducted on the Southern Coast of WA by (Portmann and Young 1987), as they recognised that “suitable material was not being generated out of the traditional germplasm being used in the program. Our attention then started to turn to vernalisation responsive barleys.” Winter lines showed promise for early sowing in WA but were deemed unsuitable for malting (Table 1); relative to Schooner they were either similar or higher yielding. They noted the slow early vigour of winter types and reduced dry matter during winter compared to spring lines and concluded the task confronting breeders was to select less temperature sensitive winter types that could maintain growth rates similar to spring types.

Table 1. Yields (t/ha) of selected spring and winter barley lines in 1986 time of sowing trials (Portmann and Young 1987).

Line	Early May Sowing (Mt Barker 1986)	April Sowing (Esperance 1986)
Stirling	3.97	1.25
Schooner	4.86	1.52
WU35 (Winter)	5.00	2.01
WU44 (Winter)	5.11	3.10
l.s.d. (5%)	0.49	0.44

The NVT trials were interrogated for data on winter release, Urambie. Urambie has been included in 131 NVT trials across QLD, NSW and Vic but not included in South Australian NVT trials. The sowing dates have arguably been too late for adequate evaluation of Urambie. Few trials have been sown before the first week of May, in the ideal window for a winter barley, with mean sowing dates trending beyond the last week of May across regions (Table 2). Despite this, Urambie has performed close to site mean yield across many sites and seasons. Analysis of Urambie performance versus sowing dates revealed little evidence of any sowing time interaction (data not presented) suggesting trials were simply sown too late. Although late May sowing dates quickly saturate vernalisation requirements, highly photoperiod sensitive cultivars have been favoured in Australia due to reduced tillering and improved grain weight compared to Urambie.

Table 2. Trial number, earliest, latest and mean sowing dates, average Urambie yield (t/ha) and percentage of site mean yield for 131 NVT trials over 12 seasons in 7 barley growing regions (NVT online).

State	Region	No. trials	Earliest sow date	Latest sow date	Mean sow date	Urambie ave yield	Percentage of SMY
NSW	N/E	26	12-May	5-Jul	3-Jun	3.75	96
NSW	N/W	39	10-May	6-Jul	27-May	3.44	98
NSW	S/E	17	13-May	5-Jul	28-May	4.08	101
NSW	S/W	29	10-May	10-Jul	24-May	3.51	98
QLD	SEQ	2	7-Jun	7-Jun	7-Jun	4.35	106
QLD	SWQ	2	24-May	1-Jun	28-May	5.23	107
Vic	S/W	16	5-May	30-May	15-May	5.28	96

Methods

Barley near isogenic lines (NILs) with variation in vernalisation requirement and/or photoperiod sensitivity were developed by Dr Ben Trevaskis at CSIRO. Preliminary yield trials were conducted on five NILs, Commander, Compass, and Urambie in 2016 at Roseworthy and Condobolin. The NILs contained different combinations of *Vrn-H1*, *Vrn-H2*, and *Ppd-H1* development genes backcrossed to the ultra- early barley genotype WI4441, representing different facultative, winter, and spring molecular ideotypes. The NILs *B01* and *B02* were winter types with different photoperiod sensitivity genes. Whereas other lines were either spring or facultative types combining differences in photoperiod sensitivity (Table 3). Lines were sown on 5th May at Roseworthy and 28th April at Condobolin in replicated field plots. Anthesis dates were recorded at both sites and at Condobolin dissections were used to identify double ridge and awn primordia stages.

Table 3. The near isogenic lines and varieties used in field trials at Roseworthy and Condobolin in 2016, showing their major development alleles growth habit (Vrn S = Spring allele, W = Winter alleles, Photoperiod I = reduced sensitivity to day length, S = Increased sensitivity to daylength).

LINE	*Vrn-H1*	*VRN-H2*	*Photoperiod*	Barley type
B06	S	W	I	Facultative
B15	S	S	S	Spring
Compass	S	S	I	Spring
B10	S	S	S	Spring
Commander	S	S	S	Spring
B01	W	W	S	Winter
B02	W	W	I	Winter
Urambie	W	W	I	Winter

Results

At Roseworthy the winter line *B02* flowered seven days later than Commander, at Condobolin the difference was four days (Tables 4 and 5). The effect of photoperiod was evident by delayed flowering of Commander compared to Compass. It was possible to achieve a flowering date similar to or earlier than the traditional Commander type in both environments with a phenology gene combination that was previously considered unsuitable. Despite not being selected for yield the NILs were competitive in both environments and at Roseworthy *B02* was the highest yielding line. Urambie was equal highest yielding at Condobolin. This shows the potential for utilising *Vrn-H1* winter alleles to improve yield in early sowing environments. Winter lines had a longer period to double ridge (Figure 1) and higher spike numbers. Spring lines were quickest to double ridge. The winter lines differed in time to double ridge and from awn primordia to anthesis, indicating diversity of development patterns that could be exploited within winter types. A common feature of winter lines has been a low harvest index and grain weights. However, in these trials there is little evidence to suggest a lower harvest index compared to springs although grain weights were noticeably lower in B02 at both sites (Tables 4 and 5) which may have implications for small grain screenings.

Table 4. Anthesis date, yield, harvest index, kernel weight, and ear numbers at Roseworthy 2016, sown May 5.

Genotype	Anthesis date	Yield (t/ha)	HI	K Wt (mg)	Ears/m2
B06	25-Aug	7.36	0.34	40.92	843
B15	28-Aug	6.66	0.30	44.76	519
Compass	24-Aug	7.66	0.36	49.02	578
B10	2-Sep	7.25	0.31	41.02	617
Commander	4-Sep	6.79	0.34	44.47	509
B01	26-Aug	7.48	0.42	44.52	869
B02	11-Sep	8.25	0.39	42.03	892
F pr.		<.001	0.01	<.001	<.001
l.s.d.		0.48	0.032	1.12	124

Conclusion

While more research is needed, based on traditional May – June sowing dates and the limited cultivar data there is evidence that winter vernalisation alleles show promise. Compared to highly photoperiod sensitive ideotypes proposed by many researchers in the literature, our own preliminary data suggests it is possible to achieve similar flowering dates with different combinations of *Ppd-H1* alleles and winter *Vrn1-H1* and *Vrn-H2* alleles. The yield performance or potential of these lines has never been explored in the context of early sowing or placed under any significant breeding selection for yield. It is understandable why there has been limited selection within breeding programs, given their use of summer nurseries, glasshouse or growth room environments to speed up and select early generations. However this also has implications for phenology, as plants will still require a vernalisation period which may only be partially fulfilled or not likely to be experienced in these selection environments. To avoid this breeder could adopt more expensive double haploid systems or care should be taken to ensure seedlings receive sufficient cold treatment to satisfy vernalisation requirements. Otherwise the system will be selecting genotypes with bias towards lower vernalisation requirements. We believe the time is right to reconsider the introgression of *Vrn* winter alleles into some faster developing spring cultivars as growers increasingly move their sowing dates forward.

Table 5. Anthesis date, yield, harvest index, kernel weight, and ear numbers at Condobolin 2016, sown 28 April .

Genotype	Anthesis date	Yield (t/ha)	HI	K Wt (mg)	Ears/m2
B06	18-Aug	4.89	0.33	45.5	568
B15	18-Aug	5.12	0.34	46.2	543
Compass	19-Aug	4.68	0.34	53.8	462
B10	24-Aug	4.28	0.33	42.5	601
Commander	3-Sep	4.38	0.32	41.8	574
B01	4-Sep	3.74	0.33	42.3	754
B02	4-Sep	3.95	0.32	40.7	846
Urambie	7-Sep	4.94	0.33	43.8	598
F pr.		<.001	<.001	<.001	<.001
l.s.d.		0.53	0.03	3.1	129

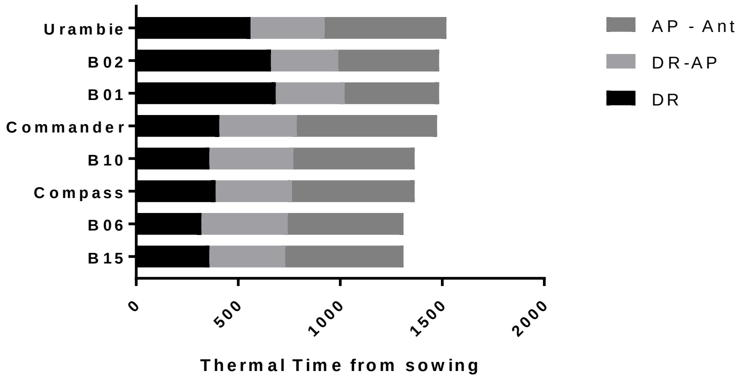

Figure 1. Phase lengths in thermal time (°Cdays) for eight barley line sown on 28 April at Condobolin 2016. The phases are sowing to double ridge (DR), double ridge to awn primordia (AP), awn primordia to anthesis (Ant).

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