

TOWARDS FINE MAPPING OF THE QFT.CRI - 3B.1 QTL IN WHEAT USING NEW GENIC MARKERS

Zbyněk Milec ¹, Kateřina Pánková ¹, Simon Griffiths ², John W. Snape ²

¹ Crop Research Institute, Drnovská 507, Prague 6, CZECH REPUBLIC

² John Innes Centre, Norwich Research Park, Colney, Norwich, UK

Motto: "Consciousness means choice...plants possess it in right, but...have renounced it...has not been said that plants are earth parasites?"
H. Bergson: *L'energie spirituelle*

Introduction

Exploiting variation for genes controlling differences in flowering time is the main method for adapting common wheat (*Triticum aestivum* L.) to different environmental conditions. Genes of small effect, commonly referred to as earliness *per se* genes, can be used for the „fine-tuning“ of flowering time. A recent study (Pánková et al., 2008) identified the effect of such a QTL, *QFt.CRI-3B.1* on chromosome 3B in Czech wheat alternative variety Česká Přesívka. This flowering time QTL was mapped on the long arm of chromosome 3B distal to marker *Xbarc164* and proximal to marker *Xcfa2170* (nearest flanking markers) in two populations of recombinant substitution lines (RSL) derived from crosses of Sandra x Sandra (CP3B) and Zlatka x Zlatka (CP3B). To narrow the QTL region for fine mapping, near isogenic lines (NIL) are being produced and new Conserved Orthologous Sequence (COS) markers are being designed from ESTs located within the region.

Results

Initially, we designed 49 COS marker primers and screened them for polymorphism using the parental varieties Sandra, Zlatka and Česká Přesívka (CP). Unfortunately, none of these markers were polymorphic between these closely related parents. However, other mapping populations could be used to map some of these markers – doubled haploid (DH) populations for the UK wheat varieties Spark x Rialto, Charger x Badger, Avalon x Cadenza and the ITMI RIL population, Opata x Synthetic.

Out of 49 markers, 3 were found polymorphic and mapped. Two of them were mapped in the Opata x Synthetic population and were distal to *Xbarc164* and one was proximal to *Xbarc164* in the Avalon x Cadenza population. These markers were used as anchor points for designing new COS markers using rice chromosome 1 sequence for the target parents. A colinearity table was compiled (Figure 1) to determine suitable rice LOCs on chromosome 1 for primer design. *Brachypodium* super-contig sequence was also used to confirm the proper order of wESTs.

Another 40 COS markers were designed. Only two of them were polymorphic for the parental varieties. Unfortunately, they were not mapped to the region of interest i.e. distal to marker *Xbarc164* and proximal to marker *Xcfa2170*. We also used the insertion site-based polymorphism (ISBP) markers published by Paux et al. "A Physical Map of the 1-Gigabase Bread Wheat Chromosome 3B" (2008). *Cfp* primers from the respective region on 3B were selected and screened for polymorphism using the parental varieties. To try to increase marker density further in the QTL region, we also used *gpw* primers, kindly provided by Catherine Feuillet and Pierre Sourdille (INRA, Clermont -Ferrand).

Unfortunately, only one ISBP marker - *Xcfp1632* - was polymorphic, for the Sandra population only. It mapped between markers *Xbarc164* and *Xcfa2170*. Out of 23 *gpw* markers, 4 markers were polymorphic for both the Zlatka and Sandra populations. These 4 markers *Xgpw7148*, *Xgpw4431*, *Xgpw4310* and *Xgpw3254* have been mapped between our QTL flanking markers (Figure 2). QTL analysis using QTL café http://www.biosciences.bham.ac.uk/labs/kearsey/qtl_ns/qtl.html was performed using the new map and previous flowering time data, Figure 3. A statistically significant QTL was found for the new marker *Xcfp1632* inside the interval.

Conclusions

The QTL region has been slightly narrowed by mapping marker *Xcfp1632* distal to marker *Xbarc164*. QTL analysis showed statistical significance for the flowering time QTL with an additive effect of nearly 4 days. Apparently the completion of a large set of NILs is necessary to obtain a higher level of recombination within the QTL region between flanking markers *Xbarc164* and *Xcfa2170*. Pánková et al. (2008) also found that vernalized substitution lines flowered later than their respective donors while non-vernalized substitution lines were earlier than their respective donors. The possible presence of a temperature-sensitive gene is under investigation by growing plants with and without vernalization, and at normal and high temperatures.

SSR marker	wEST	Rice chromosome 1 (bp)	Brachypodium chromosome 2 (bp)	Deletion bin
<i>Xbarc164</i>				3BL10-0.50-0.63
	BE490662	32330754	50678469	3BL7-0.63-1.00
	BG262582	36002778	53412117	3BL10-0.50-0.63
	BE399869	36123144	53505605	3BL10-0.50-0.63
	BE443349	36461937	30164365	3BL10-0.50-0.63
	BE499968	36665011	53827787	3BL7-0.63-1.00
	BM134465	37619905	54579224	3BL7-0.63-1.00
	BE517732	38504350	55419897	3BL7-0.63-1.00
	BE424246	39559478	56183697	3BL7-0.63-1.00
	BE517780	39955579		3BL7-0.63-1.00
	BE443288	40181440	56746533	3BL7-0.63-1.00
	BE495175	41062970	57522327	3BL7-0.63-1.00
	BE444864	42299613	58506401	3BL7-0.63-1.00
	BM137927	43010525	59004357	3BL7-0.63-1.00
<i>Xbarc344</i>				3BL7 0.63-1.00
	CD454173	37615202	54475025	3BL7-0.63-1.00
	BQ162013	37619412	54578755	3BL7-0.63-1.00
	BQ487514	37572265	54529735	3BL7-0.63-1.00
	BF146198	38381423	55315145	3BL7-0.63-1.00
	BQ167580	39561287	56185126	3BL7-0.63-1.00
	BE424246	39559478	56184137	3BL7-0.63-1.00
	BE424246	39559478	56184137	3BL7-0.63-1.00
	BQ167580	39561287	56185126	3BL7-0.63-1.00
<i>Xcfa2170</i>				3BL7 0.63-1.00

Figure 1: Colinearity table: wEST x rice chromosome 1 x *Brachypodium* chromosome 2

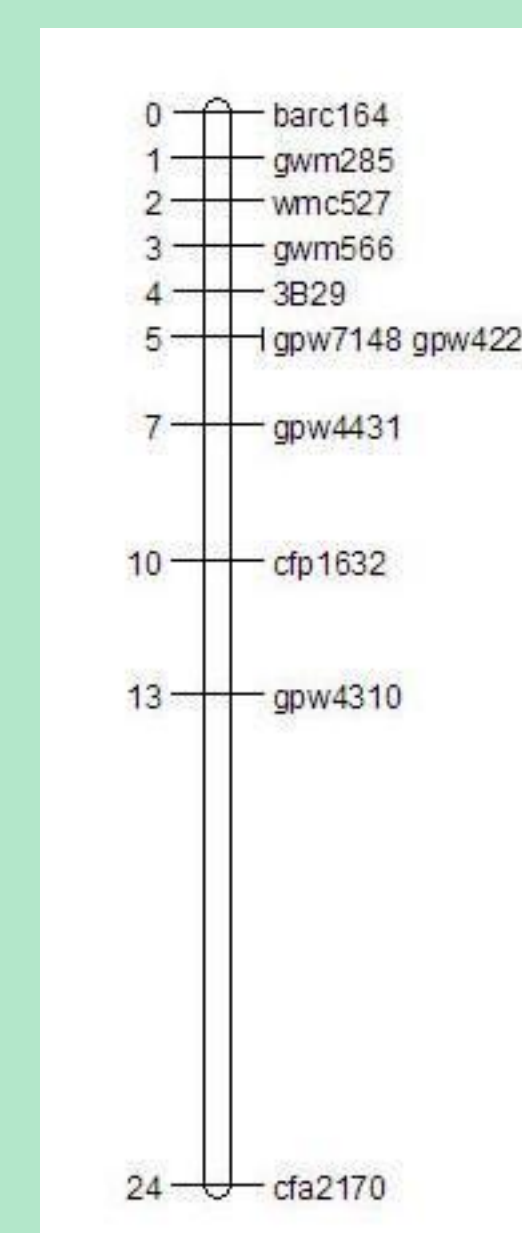


Figure 2: Sandra(Sandra3B/CP 3B) – new map

No.	Pos.	Add. eff.	mean(11)	n(11)	mean(22)	n(22)	F	P
1	0.0	-1.03	125.25	16	127.3	10	2.076	0.1625 NS
2	2.0	-1.29	125.12	17	127.7	10	3.699	0.0659 NS
3	3.0	-1.16	125.13	16	127.45	11	3.042	0.0934 NS
4	4.0	-1.45	124.5	14	127.4	10	6.207	0.0208 *
5	5.0	-1.05	125.29	17	127.4	10	2.342	0.1385 NS
6	7.0	-1.42	125.06	17	127.89	9	4.074	0.0549 NS
7	10.0	-1.85	124.08	12	127.78	9	10.06	0.0050 (**)
8	13.0	-0.42	125.94	17	126.78	9	0.325	0.5738 NS
9	24.0	-0.97	125.44	18	127.38	8	1.629	0.214 NS

Sandra3B – LOD4

1.gwm566	0cM
2.gwm285	2cM
3.barc164	3cM
4.wmc527	4cM
5.gpw7148	5cM
6.gpw4431	7cM
7.cfp1632	10cM
8.gpw4310	13cM
9.cfa2170	24cM

Figure 3: Single marker QTL analysis

References:

Pánková K., Milec Z., Simmonds J., Leverington -Waite M., Fish L., Snape J. W. (2008): Genetic Mapping of a New Flowering Time Gene on Chromosome 3B of Wheat. *Euphytica* 164:779-787.
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