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Twelve new QTLs for phenological traits in *Vitis vinifera* L.

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Changes in phenology are a major response to climate change in present cultivars. Therefore, we need to account for these traits in breeding programs to maintain yield and quality.

As in other perennials, many phenology QTLs have already been detected in grapevine, most often explaining a low proportion of total variance, but occasionally up to 40-70% (Costantini et al 2008, Duchêne et al 2012, Grzeskowiak et al 2013, Zyprian et al 2016). Very few colocalized between studies.

Our objective was to better understand genetic determinism of phenological stages in *V. vinifera* L., by searching for QTLs and candidate genes in three unrelated populations.

Material and methods

Plant material

This study was based on three pseudo-F1 mapping populations, MTP3140 (N=139), MTP3234 (N=174) and SG (N=191), planted at the Montpellier SupAgro Chapitre Research Vineyard, Hérault, France.

Phenotyping

Six developmental stages were recorded in 3-6 years, according to the modified E-L system (Coombe 1995): budburst (BB), flowering beginning (FB), flowering end (FE), veraison beginning (VB), véraison end (VE), ripening (R). Each date and interval between dates was converted into day of the year (DOY, the number of days from January 1st). Heat sums were also calculated using the Grapevine Flowering Veraison (GFV) model (Parker et al 2011).

Statistical analyses

The linear mixed model best fitting the data was selected according to the Baysian Information Criterion (BIC). All tested models included a random genotypic effect, and possibly some fixed effects and their interaction with genotype. Broad-sense heritability was estimated for each trait from variance components.

QTL detection

It was performed on each consensus and parental map (Doligez et al 2006, 2010, 2013) with the R/qtl package, using the Best Linear Unbiased Predictors (BLUPs) of genotypic effects. Multiple QTL regression was carried out with the "stepwiseqtl" function following Huang et al. (2012), using a genome-wide error rate of 0.05.

Search for candidate genes

Positional candidate genes located in the confidence interval of QTLs was obtained from RefSeq NCBI 12X.0-based *Vitis vinifera* Annotation Release 101, mapped on the PN40024 12X.2 reference genome sequence.

Phenotypic data

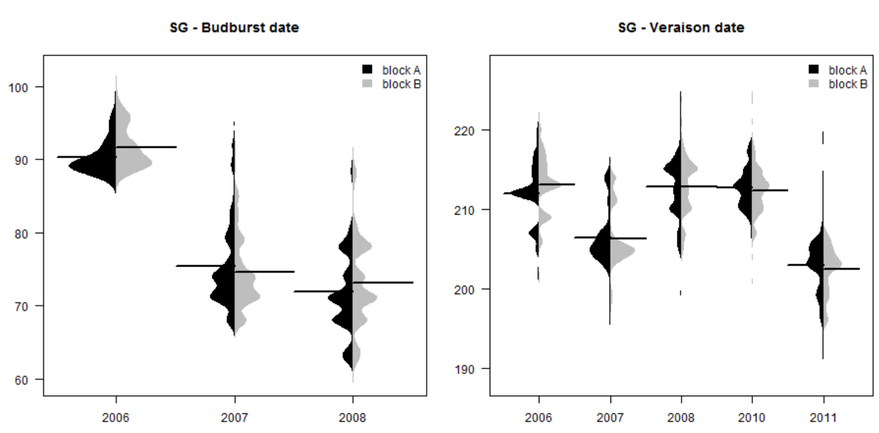

Fig 1. Distribution of some raw phenotypic data in population SG (Syrah x Grenache). Dates are in DOY.

Large phenotypic and genotypic correlations (≥ 0.6) were observed between the DOY and GFV models for most dates and intervals.

Table 1. Broad-sense heritability on an inter-annual genotype mean basis.

Trait (in DOY)	MTP3140	MTP3234	SG
Budburst	0.66	0.50	0.75
Flowering	0.71	0.68	0.77
Veraison	0.85	0.76	0.93
Ripening	0.66	0.66	0.69
Budburst - Flowering	0.52	0.35	0.39
Flowering - Veraison	0.85	0.77	0.72
Veraison - Ripening	0.40	-	0.36
Budburst - Veraison	0.73	0.65	0.72
Budburst - Ripening	0.62	-	0.54
Flowering - Ripening	0.62	-	0.62

12 new repeated QTLs

A total of 18 QTLs were found in at least 2 years (14 on consensus maps and 4 additional ones on parental maps) in 2 of the 3 populations, mainly for budburst and veraison dates, explaining up to 35% of total variance. Twelve of them were new QTLs and 6 were confirmed ones.

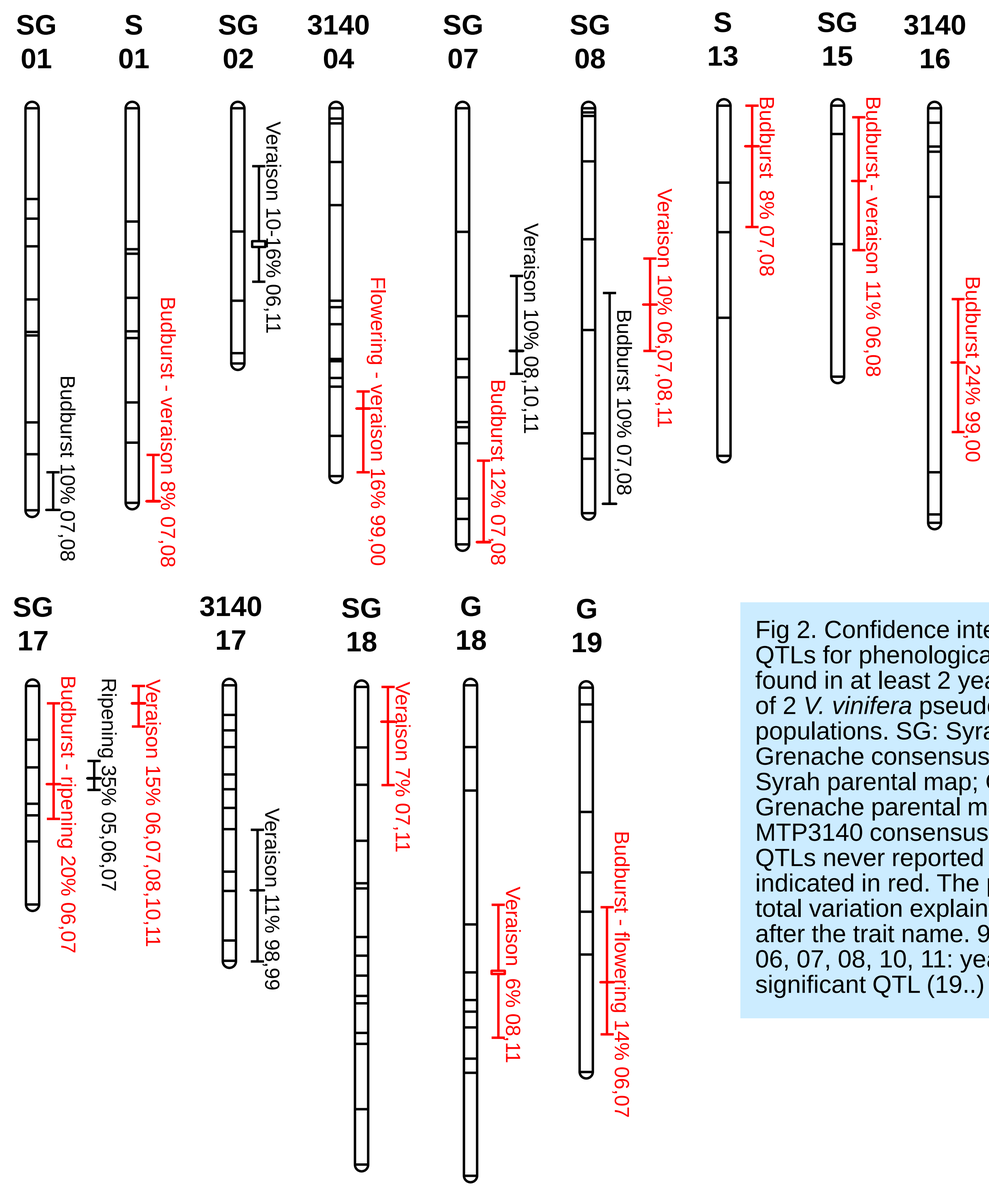

Fig 2. Confidence intervals of QTLs for phenological traits found in at least 2 years in one of 2 *V. vinifera* pseudo-F1 populations. SG: Syrah x Grenache consensus map; S: Syrah parental map; G: Grenache parental map; 3140: MTP3140 consensus map. QTLs never reported before are indicated in red. The percent of total variation explained is given after the trait name. 98, 99, 00, 06, 07, 08, 10, 11: years with significant QTL (19..)

Candidate genes

Several candidate genes potentially involved in phenology-related trait variations were found near LOD peaks of these QTLs. In particular, for budburst-related traits, based on the review by Abbott et al (2015) for perennials, we found a protein homolog of timeless (involved in circadian rhythm), a CONSTANS-LIKE zinc finger protein (transcription factor promoting flowering in long days), and an indole-3-acetic acid-amido synthetase GH3.17-like (involved in biosynthesis of auxin). For veraison, we found a bidirectional sugar transporter SWEET recently shown to play an important role in berry maturation (Charles Romieu, personal communication).

Our results confirm the highly polygenic nature of phenology-related traits, show that some QTLs are stable across years, and yield potentially very interesting candidate genes for further studies on the molecular determinism of variations in phenological stages.

Ackowledgements

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