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#### ▶ To cite this version:

Marta Avramova, Catherine Tesniere, Martine Pradal, Jean-Luc Legras, Bruno Blondin. Genetic bases of nitrogen requirement in wine yeast assessed trhrough QTL analysis. 35. International Specialised Symposium on Yeasts, Oct 2019, Antalya, Turkey. 169 p., 2019. hal-02460145

HAL Id: hal-02460145

https://hal.science/hal-02460145

Submitted on 2 Jun 2020

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# The 35<sup>th</sup> International Specialized Symposium on Yeasts

21 - 25 October 2019 Antalya, Turkey

"Yeast Cornucopia: Yeast for health and wellbeing"

## PROCEEDINGS BOOK













### Genetic Bases of Nitrogen Requirement in Wine Yeast Assessed Through QTL Analysis

#### POSTER PRESENTATION ID: 221

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In grape must, nitrogen content is often insufficient for the completion of alcoholic fermentation by yeast. For  $S_{accharomyces\ cerevisiae}$ , response to nitrogen deficiency is strain-dependent, some strains being able to complete fermentation despite nitrogen deficiency whereas others are not and result in sluggish or stuck fermentation. Thus, it is of high interest to study the mechanisms behind those different responses and exploit them to improve yeast strain for wine fermentation when nitrogen content is low. Previous study highlighted different genomic regions involved in nitrogen requirement through BSA ( $B_{ulk}$   $S_{egregant}$   $A_{nalysis}$ ), and the contributions of three genes: MDS3, GCN1, and ARG81 have been shown (1). However, many other large genomic regions were also defined for which we could not find evident candidate genes. In addition, BSA did not provide any information on possible interactions between loci.

In order to explore further the genetic bases of nitrogen requirement, we applied a QTL analysis to the fermentation rate in nitrogen deficient medium, on a population of 131 individually genotyped segregants obtained from the same cross as (1). The dense genetic map available for the segregant population (3727 markers) enabled us to perform single and multiple map QTL and thus define genomic regions which could be implied in low nitrogen requirement. In order to further validate the impact of candidate genes on the phenotype, alleles were "swapped" by CRISPR- $C_{as}\theta$  technique and phenotype was evaluated in comparison with haploid parent strains.

Several regions with high LOD scores were identified, some above the significance threshold, and others below, among which the regions containing the genes identified by (1), probably in relation with the multigenic character of the trait. In the region with the highest LOD score, two candidate genes in relation with nitrogen metabolism (namely, Target of Rapamycin (TOR) pathway and lifespan regulation) were identified. In addition, in order to reveal possible interaction between genes, strains carrying different combinations of *GCN1* and *MDS3* parental alleles (implied in TOR pathway) have been evaluated. These constructions confirm their role on the fermentation rate in low-nitrogen conditions and indicate dependence on the genetic background.

These results confirm the complexity of mechanisms involved in nitrogen requirement during alcoholic fermentation and will permit to optimise wine yeast strain selection in response to winemaking industry demands.

Keywords: Nitrogen requirement, Wine, Yeast, TOR, QTL