

BIODIVERSITY OF *DEKKERA BRUXELLENSIS* IN WINES a common and dangerous yeast

Yeasts belonging to *Dekkera/Brettanomyces* genus can lead to severe organoleptic defects on fermented beverages by producing unqualitative compounds such as volatile phenols. Our aim is to better define the origin of these yeasts. The genetic differentiation between *D. bruxellensis* strains isolated on grapes and those from related musts and wines allows to know whether contaminations and disseminations are of vine origin or enological origin.

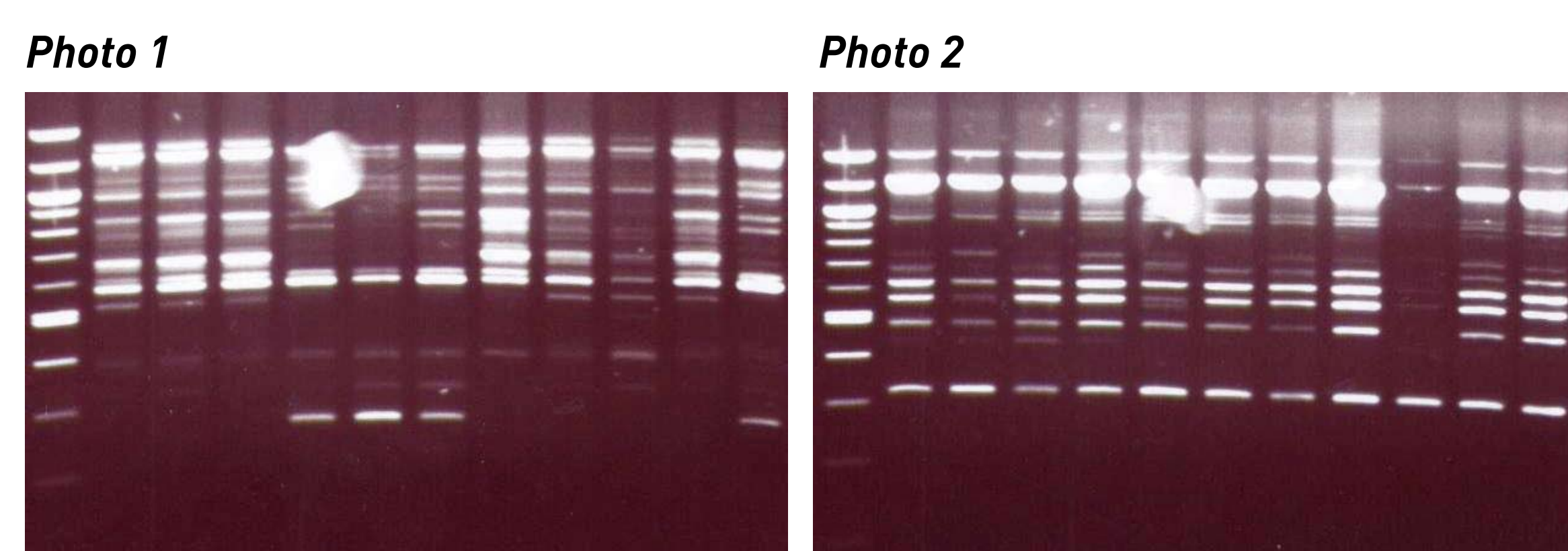


Photo 1 Photo 2
RAPD profiles after amplification with OPB15⁽¹⁾ or OPF16⁽²⁾ primers
Photos IFV - M. Coarer

Viable and cultivable yeasts from grapes and aging wines were numbered, 10 colonies by sample being isolated and purified. Specific detections using previously designed primers were then carried out and confirmed by restriction of ITS region using Hinf I and Taq I. *D. bruxellensis* strain characterization was then achieved using 3 RAPD primers (OPB15, OPD03, OPF16) giving results in accordance with REA-PFGE analysis (photos 1-2).

DEKKERA ORIGIN

One or several different *D. bruxellensis* strains may be present on the same grape, as shown for strains 06G11 and 06G12 sampled on one grape sample from Morgon in 2006. However, *D. bruxellensis* strains collected from one grape sampling are different from those isolated in the resulting wines (figure 1). If the presence of *D. bruxellensis* on grape is well known, the ability of these strains to colonize wines after fermentation remains hypothetical.

D. BRUXELLENSIS POPULATIONS EVOLUTION

Total populations of *D. bruxellensis* increase during aging of wines, between the end of the alcoholic fermentation and the completion of the malolactic fermentation. In the same time, we observed a large modification of contaminating strain biodiversity (figure 2). While 2 genotypes (07G01 and 07G03) are prevailing in November samples, they disappeared two months later giving place to numerous new genotypes, such as 07G02 present in 67 % of the wines. This strain and 07G01 are even isolated in white wines sampled in January, refuting the idea that this development is confined to red wines. However, biodiversity is more important in red wines with up to 6 different genotypes for a given wine.

Figure 1 : Genotype proportions in grapes and corresponding wines



Figure 2 : Genotype proportions evolution with ageing

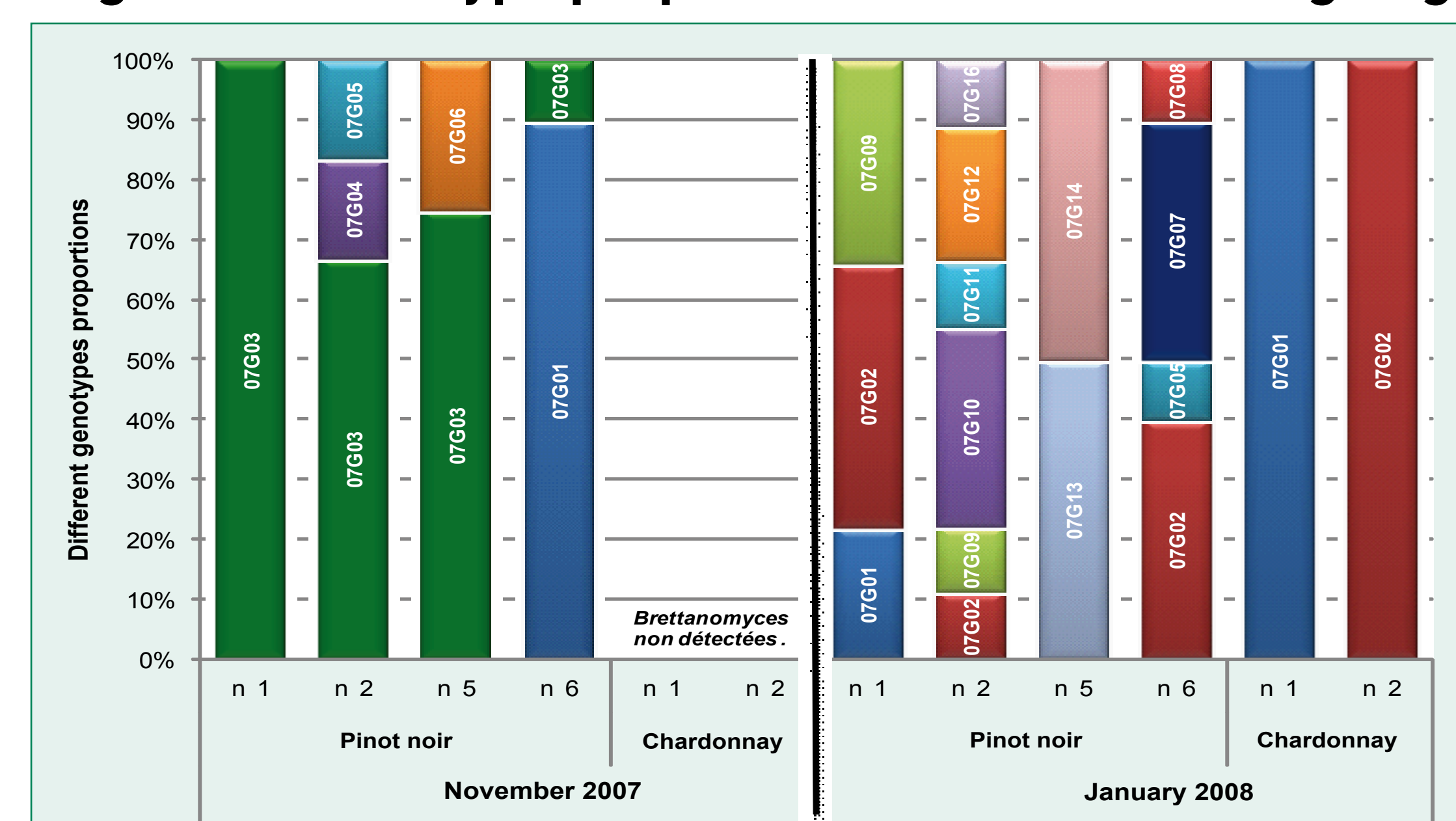
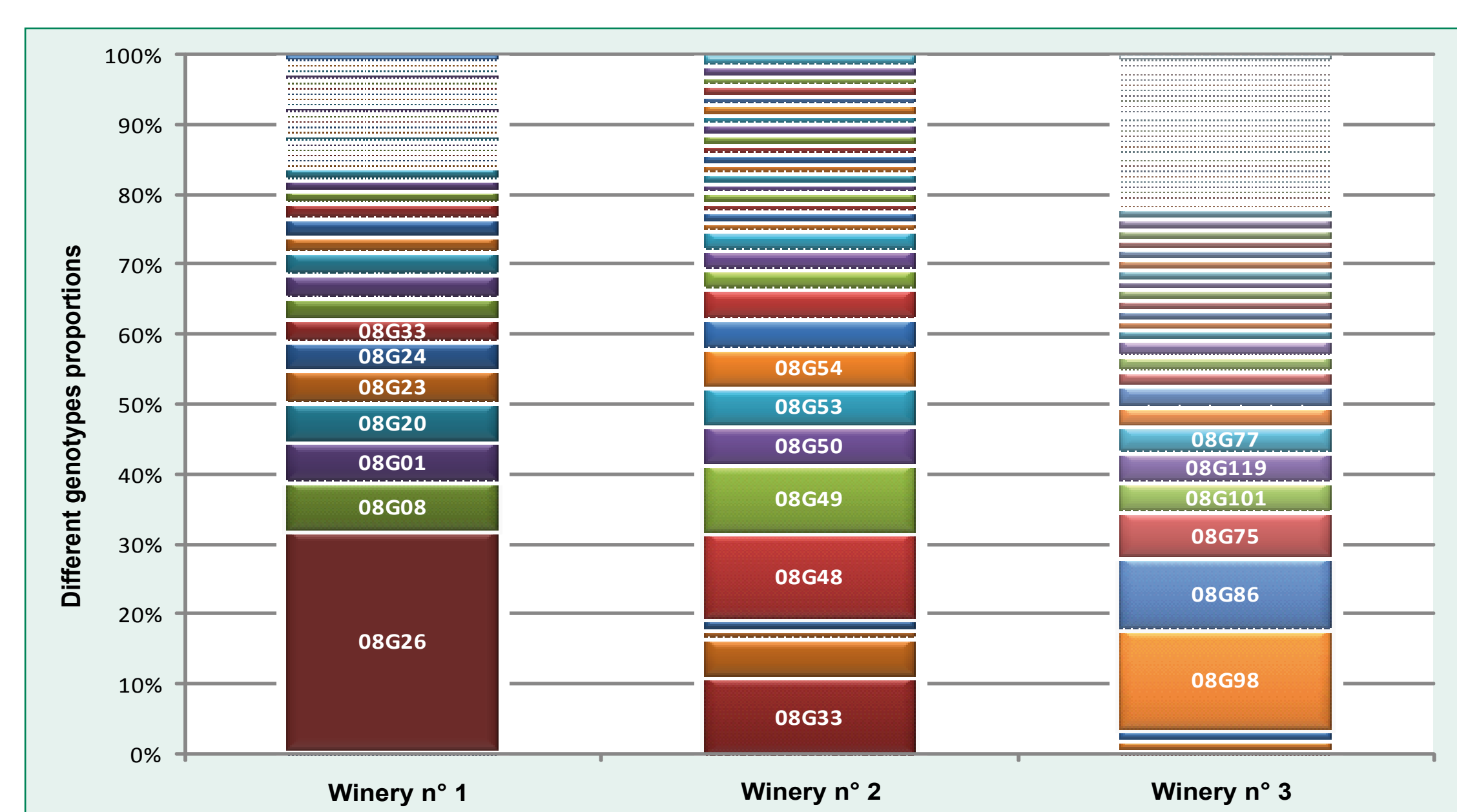


Figure 3 : Genotype proportions in different wineries



DEKKERA DISSEMINATION

Samplings were made on 18 different fermentations from 3 wineries distributed from North to South Burgundy (figure 3). 358 strains of *D. bruxellensis* were isolated and characterized. While OPB15 primer gives 42 different profiles, the concomitant use of the 3 primers leads to distinguish 123 genotypes. No strain was common to the 3 wineries and only 4 % were common to 2 wineries, such as strain 08G33 for wineries 1 and 2. Each winery has its own pool of about 5 main genotypes gathering 40 to 50 % of the isolated strains, the other 50 to 60 % being composed of numerous minor genotypes.

CONCLUSION

The large biodiversity of alteration yeast *Dekkera bruxellensis/Brettanomyces intermedius* is therefore demonstrated. Our results suggest that the strains collected on grapes are not identical to the various genotypes contaminating wines. It could be interesting to study whether this genotypic heterogeneity is correlated with phenotypic variations (resistance and/or volatile phenols production) or not.