EXPLORING THE IMPACT OF ORGANIC ASSIMILABLE NITROGEN SUPPLEMENTATION ON YEAST BEHAVIOUR DURING ALCOHOLIC FERMENTATION.

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INTRODUCTION
Saccharomyces cerevisiae is known as the main agent of alcoholic fermentation (AF) in wine and selected strains are often used by winemakers to inoculate grape must. However, to complete AF, yeast need to have a sufficient amount of yeast assimilable nitrogen (YAN). In a nitrogen deficient must, mineral (diammonium phosphate) or organic YAN sources can be used, the latter being available through the use of yeast derived products and mainly yeast autolysates. Nevertheless, the exact composition of these products (lipids, vitamins, amino acids, micronutrients) and their precise effect on S. cerevisiae still remain unclear.

In this work, an organic YAN product was first finely characterized, then, several experimental designs were carried out to investigate precisely the impact of each compound on yeast behaviour as well as AF performance and enological consequences.

EXPERIMENTAL DESIGN

Example of a matrix design : Fractional factorial design
1: compound absent from alcoholic fermentation
0: compound added for alcoholic fermentation (at same concentration as the commercial product)
-1: compound added for alcoholic fermentation at mid-concentration. All compounds are present at mid-concentration for ‘0’ assay (centerpoint assay).

Fermentations were performed in sauvignon blanc grape must in which different components were added through organic YAN product reconstituted solutions, according to the experimental design. Initial YAN status was 152 mgN/L and complemented with an equivalent of 30 g/hL organic YAN reconstituted solution. Each 125 mL assay was inoculated with Zymaflore ® X5 (105 cells/mL). Fermentation kinetics were monitored through CO2 release, growth parameters by flow cytometry. End of AF chemical parameters were performed by Excell laboratories, thiols measured through CPG-SM, and H2S with Park tubes.

RESULTS

1 Impact of 39 molecules composing organic YAN yeast derived products (Hadamard matrix design)

<table>
<thead>
<tr>
<th>Riboflavin</th>
<th>GABA</th>
<th>Tryptophane</th>
<th>Arginine</th>
<th>Threonine</th>
<th>Valine</th>
<th>Lanosterol</th>
<th>C18</th>
</tr>
</thead>
<tbody>
<tr>
<td>Presence (+)</td>
<td>-Yes</td>
<td>-Yes</td>
<td>+Yes</td>
<td>+Yes</td>
<td>+Yes</td>
<td>+Yes</td>
<td>+Yes</td>
</tr>
<tr>
<td>Absence (-)</td>
<td>+Yes</td>
<td>+Yes</td>
<td>-Yes</td>
<td>-Yes</td>
<td>-Yes</td>
<td>-Yes</td>
<td>-Yes</td>
</tr>
</tbody>
</table>

Max pop (x10^8 viable cells/mL)

| Lag AF (h) | 26.2 | 26.2 | 25.6 | 26.5 | 25.6 | 26.7 | 26.8 | 27.0 | 27.5 | 27.3 | 26.2 | 28.4 | 28.1 | 26.1 | 26.2 | 26.4 | 25.9 |

T50% (h)

| 63.8 | 64.2 | 63.0 | 65.0 | 63.1 | 64.9 | 63.1 | 64.2 | 61.4 | 65.2 | 62.4 | 65.0 | 63.9 | 64.1 | 64.6 | 64.3 |

Example of the impact of riboflavin on some enological parameters

CONCLUSION

This work, to the best of our knowledge, is the first one to characterise a yeast derived organic YAN matrix used for grape must supplementation.

This enabled us to perform a synthetic reconstituted solution containing all or just a part of these compounds at the concentrations found in the product. Consequently, experimental designs were performed to study the impact of each compound alone and as well as in interaction with each other on yeast behaviour and alcoholic fermentation performance, including aromas such as thiols release.

Further work has now commenced to focus on different yeast derivatives analysis in order to find the best organic YAN formulations.