PHYSIOLOGICAL STUDY OF YEAST STRAINS INVOLVED IN TEQUILA FERMENTATION

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Key world: Tequila, alcoholic fermentation S. cerevisiae and K. africana yeast strains.

The fermentation process plays a main role in the elaboration of tequila. In this stage, ethanol and several volatile compounds are formed; it contributes to the unique characteristics of tequila. The tequila fermentation process involves a large variety of yeast strains, belonging to different genera and species; among which, Saccharomyces and Kloeckera (or Hanseniaspora) are the most common (1). Despite the popularity of tequila, the industry has very limited knowledge of native yeast characterization and parameters that affect agave juice fermentation (2). The aims of the present study were to determine the fermentative capability and volatile compound productions by yeast strains isolated from agave tequilana Weber juice, as well as establish strategies to increase growth rate, fermentative efficiency and the synthesis of volatile compounds desirables. Also these strategies will permit to obtain distillates with sensory profiles satisfactory. Finally, in this study will present an innovative alternative to improve the productivity of the fermentation stage.

![Fig. 1. Cluster analysis on the basis of the kinetic parameters and percentage of the total concentration of volatile compounds produced by five native yeast family strains in pure culture.](image)

The results showed a clear difference between fermentative abilities (2) and the volatile compounds production (3) of two different genres during the fermentation of agave juice (Fig. 1). The genus Kloeckera showed a low growth rate and higher synthesis of esters than Saccharomyces yeasts strains. In the case of Saccharomyces cultures, higher fermentative capability was presented. However, supplementation with an organic nitrogen source greatly stimulated the growth and fermentative capacity of Kloeckera (Table 1). This result suggested that, in contrast to S. cerevisiae, K. africana required other growth factors not supplied by the agave juice (4). The effects of dilution rate, temperature and micro-aeration on growth, fermentation and synthesis of volatile compounds of native yeast belong of two genus, cultured in continuous fed with Agave tequilana juice were studied.

In continuous cultures, the temperature and aeration improved biomass production and consumption of reducing sugars of two species of yeasts (Fig. 2). The aeration also significantly augmented ethanol production ($P < 0.05$).

![Fig. 2. Influence of operational conditions in the fermentative capability of Kloeckera africana and S. cerevisiae.](image)

### Table 1. Kinetic parameters on the bases of biomass

<table>
<thead>
<tr>
<th>Yeast</th>
<th>Nitrogen sources</th>
<th>$\mu_{\text{max}}$ (h$^{-1}$)</th>
<th>$Y_{x/s}$ (g/g)</th>
<th>$X_t$ (g/l)</th>
</tr>
</thead>
<tbody>
<tr>
<td>K1</td>
<td>Organic</td>
<td>0.306</td>
<td>0.032</td>
<td>3.20</td>
</tr>
<tr>
<td></td>
<td>Inorganic</td>
<td>0.126</td>
<td>0.062</td>
<td>0.75</td>
</tr>
<tr>
<td>S1</td>
<td>Inorganic</td>
<td>0.310</td>
<td>0.037</td>
<td>3.56</td>
</tr>
<tr>
<td></td>
<td>Inorganic</td>
<td>0.340</td>
<td>0.036</td>
<td>3.62</td>
</tr>
</tbody>
</table>

Different aromatic profiles were found in each condition studied, indicating that they significantly affected the metabolism of the S. cerevisiae and Kloeckera yeast strains. The continuous agave juice fermentation was scale up. The results obtained showed that continuous systems can be used at a pilot level.

Acknowledgement. Thank to SEP-CONACYT for the financial support to project 24547.

Bibliografía.

1. DM Díaz-Montaño & J Ramirez. The fermentative and aromatic ability of Kloeckera and Hanseniaspora yeasts In Yeast Biotechnology: diversity and applic. 281-305.