

Evaluation of a new strain of *Candida guilliermondii* resistant to environmental factors in the production of xylitol

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Introduction

In biorefineries, there is a tremendous need to decrease production costs to enhance their feasibility as businesses. Biorefinery processes would benefit from reducing the number of steps needed to generate top products.

Xylitol is one of the top chemicals to be produced through a biorefinery scheme^[1]. Usually, it is generated through fermentation of xylose-rich liquors obtained after pretreating lignocellulosic materials^[1]. Diverse yeasts are employed in the fermentation process, being *Candida guilliermondii* one of the most reported^[2]. Prior to fermentation, the liquor must be detoxified to remove inhibitors like acetic acid (AA), which are generated during the pretreatment process and that affect the productivity of the microorganism. Detoxification is achieved by treating the liquor with activated carbon (AC) in packed columns or in mixing tanks^[3]. In both cases, fixed and variable costs of the process increase due to the need of equipment (packed column or mixer), raw materials (AC), and services (heat needed to regenerate AC).

The challenge and solution

Currently, detoxification of xylose-rich streams is crucial to boost conversion yields in xylitol production^[3]. Eliminating such step without diminishing productivity, will allow to decrease fixed and variable costs. Here, a new strain of *C. guilliermondii* resistant to high concentrations of AA, is employed to produce xylitol from non-detoxified liquors.

Methods

The strain of *C. guilliermondii* was isolated from organic wastes from the Central de Abasto in Mexico City. Its tolerance to high concentrations of AA was measured by culturing in 250 mL flasks containing 50 mL of YPD broth supplemented with diverse concentrations of the acid (0,5,7.5 and 10 g/L). In all cases the initial pH was adjusted to 6. Similarly, morphologic changes were assessed by culturing in agar plates.

Fermentations for xylitol production were done in 500 mL baffled flasks with 100 mL of one of the following media: a synthetic media containing pure xylose, a media formulated with non-detoxified corn cob hydrolysate, and a media formulated with detoxified corn cob hydrolysate. Hydrolysates were prepared by processing corn cob through a thermochemical pretreatment (25% wt. solids, 2% wt H₂SO₄) for 1 hr at 120 °C and 1 bar. Then, pH was adjusted to 6 by addition of KOH. Initial concentration of AA in hydrolysates was 7 g/L. Detoxification was done by mixing the hydrolysate with 5 g/L of AC for 20 hrs at 150 rpm. All media contained 20 g/L of xylose, 1 g/L of YE, 1.7 g/L of YNB and 3 mL/L of trace element solution. Fermentations were done at 27°C and 150 rpm.

Results

Figure 1 shows growth in YPD medium containing different amounts of AA. Cellular growth was higher with bigger concentrations of AA. It is reported that concentrations of AA higher than 6 g/L inhibit growth of this yeast^[4].

Morphologic changes in the strain given by different amounts of AA are shown in Figure 2, which shows that colony size is diminished by high concentrations of AA.

Finally, Figure 3 shows conversion yields of xylose to xylitol in three different media. The highest yield is achieved when employing pure xylose, which is explained by the absence of inhibitors. The lowest yield, achieved with detoxified hydrolysate, might be given due to the complete lack of AA. As shown in Fig. 1, this strain of *C. guilliermondii* performs better with the presence of such acid.

References

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- 3) Buhner, J (2004) Effect of detoxification of dilute-acid corn fiber hydrolysate on xylitol production. *Appl Biochem Biotechnol*, 119(1):13-30.
- 4) Pereira, RS (2011) Inhibitory action of toxic compounds present in lignocellulosic hydrolysates on xylose to xylitol bioconversion by *Candida guilliermondii*. *J Ind Microbiol Biotechnol*, 38(1):71-8.

Results and discussion

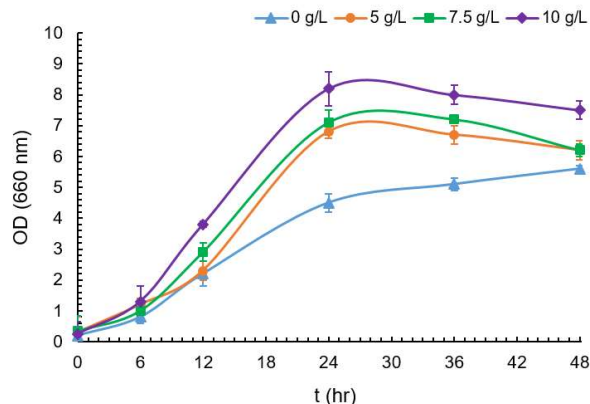


Figure 1. Yeast growth under increasing concentrations of AA (error bars mean of 2 experiments).

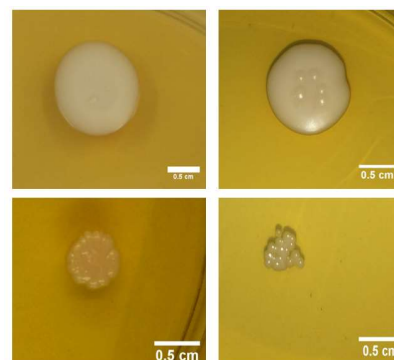


Figure 2. Morphologic changes under diverse concentrations of AA.

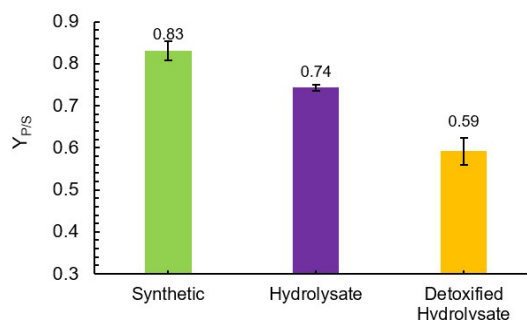


Figure 3. Average conversion yields for each media. (n=2).

Conclusions

The new strain of *C. guilliermondii* produces xylitol from non-detoxified corn cob hydrolysates with conversion yields close to the ones achieved when employing pure xylose as carbon source. This strain shows maximum cellular growth with high concentrations of AA, and a superior resistance than the reported on literature.