Detoxification of phenols in sugarcane syrup for improved yeast fermentation

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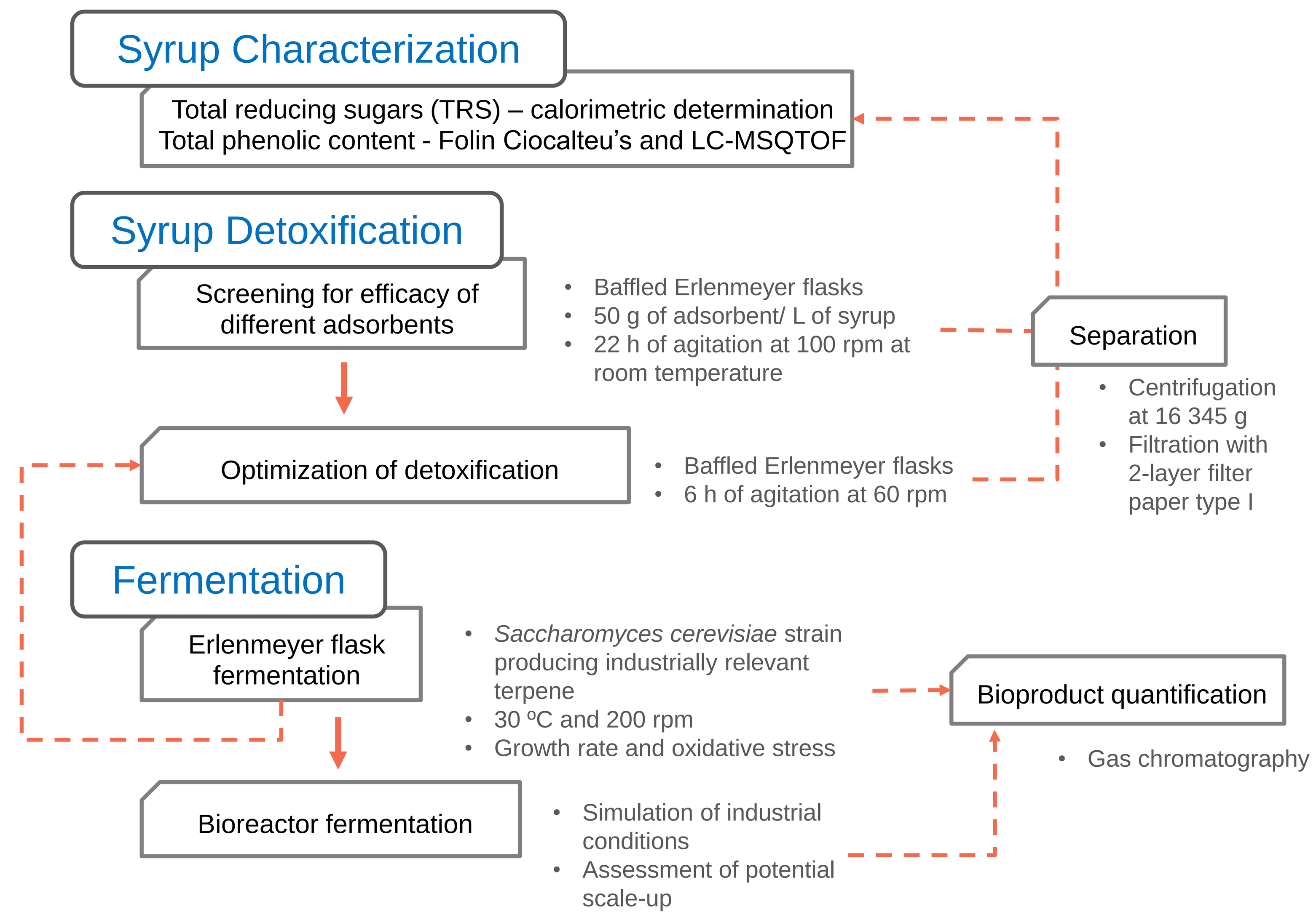
Introduction

- In **industrial fermentations**, plant biomass, such as **sugarcane syrup**, is used to provide the carbon source for the yeast to grow and **biosynthesize** the desired added-value compounds **[1]**.
- Sugarcane syrup contains several **polyphenols** that may influence the fermentation process, exerting **antimicrobial activity**. Ferulic acid is being mentioned as one of the most toxic phenolic compounds which is present in the syrup **[2]**.
- **Reducing** the **polyphenol** concentration may help to **improve** the yeast **growth** rate, yield or **productivity [3]**.
- Detoxification of syrup through using **activated charcoals** or specific **bead resins** has the potential to **drastically reduce** phenols content **[4, 5]**.
- This work aims at **extracting the phenols** present in sugarcane syrup by using solid adsorbents to assess the **effect** of phenols removal on yeast **fermentation**.

Planification



Results & Discussion

SYRUP CHARACTERIZATION

- Characterization of sugarcane syrup, used in industrial fermentations, was performed.

Table 1 – Characterization of sugarcane syrup used in industrial fermentations.

Parameter	Concentration
Total reducing sugars	714.8 g/L
Total phenols (Folin Ciocalteu's)	2080 ± 119 mg/L
Sum of identified phenols (LC-MSQTOF)	52.35 ± 1.05 mg/L
Hydroxybenzoic acids	13.70 ± 0.27 mg/L
Hydroxycinnamic acids	27.07 ± 0.41 mg/L
Flavones	11.59 ± 0.12 mg/L
pH	5.58 ± 0.01

SYRUP DETOXIFICATION WITH DIFFERENT ADSORBENTS

- A large screening was performed to assess the efficacy of different materials in the detoxification of phenolic compounds from sugarcane syrup.
- A clear difference between the charcoals and the bead resins was observed: the resins were less effective, presenting lower phenols adsorption capacity.
- The material with the best performance (highest resulting detoxification) was the charcoal "Carbone Superattivo CP" (Proenol, Portugal). This was selected as the best adsorbent for polyphenols and used for the following studies.

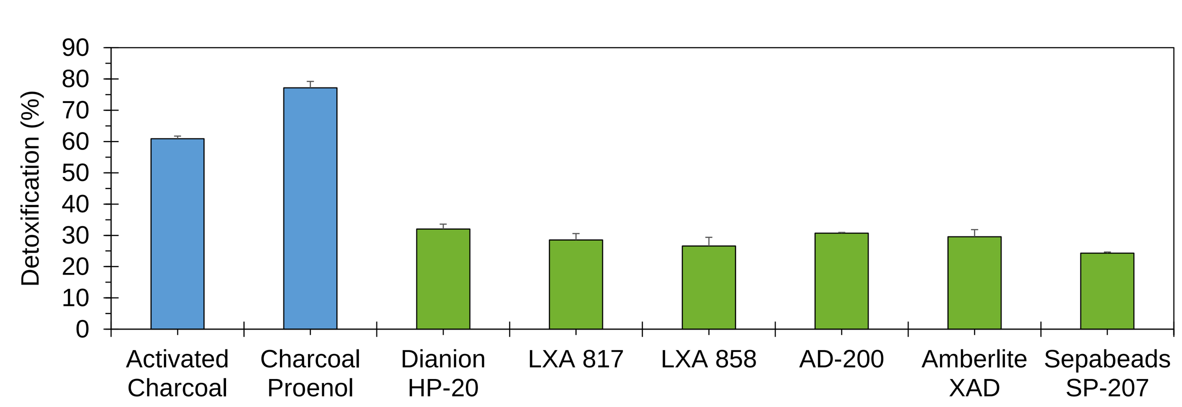


Figure 1 – Percentage (%) of detoxification of total phenols, according to Folin Ciocalteus's methodology, obtained for each material tested. Two charcoals (**blue**) and six bead shaped resins (**green**) were tested.

OPTIMIZATION OF SYRUP DETOXIFICATION

- Adsorption was conducted by reducing the agitation time and speed (Fig. 2-C). After 4 hours of agitation at 60 rpm, the polyphenols detoxification reached maximum value of 72.73 %.

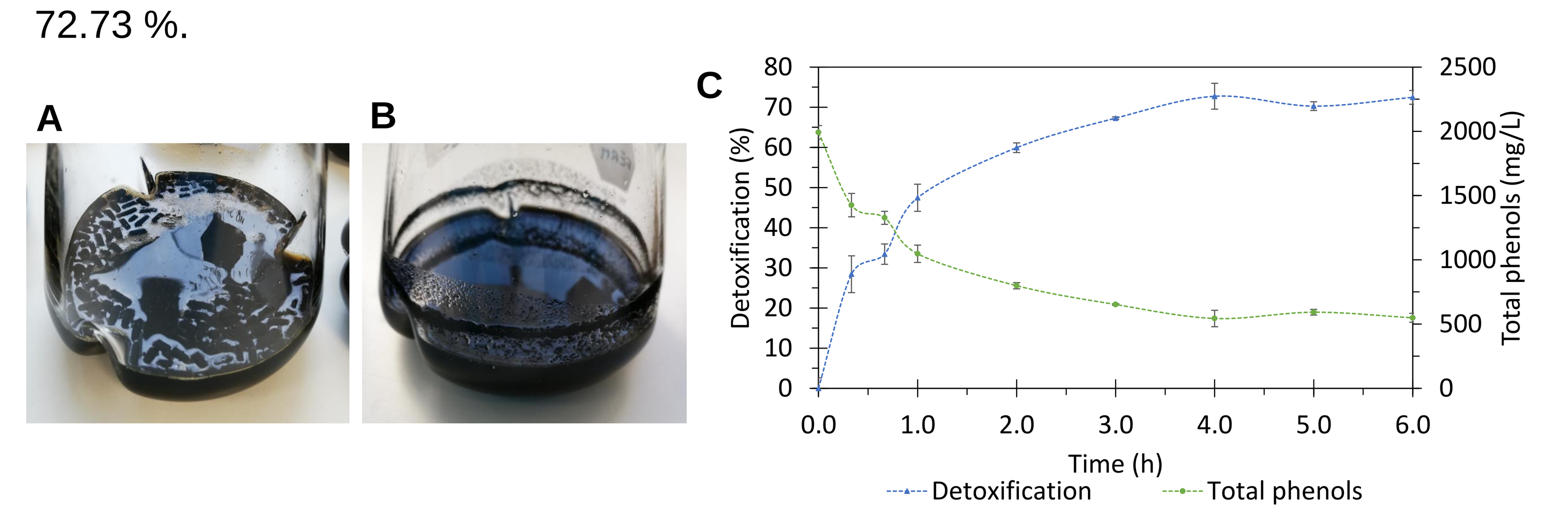


Figure 2 – Optimization of sugarcane syrup detoxification using charcoal Proenol: pictures of syrup in mixture with charcoal before (**A**) and after detoxification (**B**); total phenols, according to Folin Ciocalteu's method, and phenol detoxification percentage throughout incubation time (**C**).

- To improve detoxification, the concentration of charcoal was increased from 50 to 100 g/L of syrup (Fig. 3). With the highest charcoal concentration, 96.42 % of detoxification was reached. The compounds still remaining in syrup were gentisic acid, hydroxybenzoic acid and hydroxybenzoic-4-B-glucoside at 0.18, 0.41 and 1.28 mg/L, respectively, according to LC-MSQTOF.

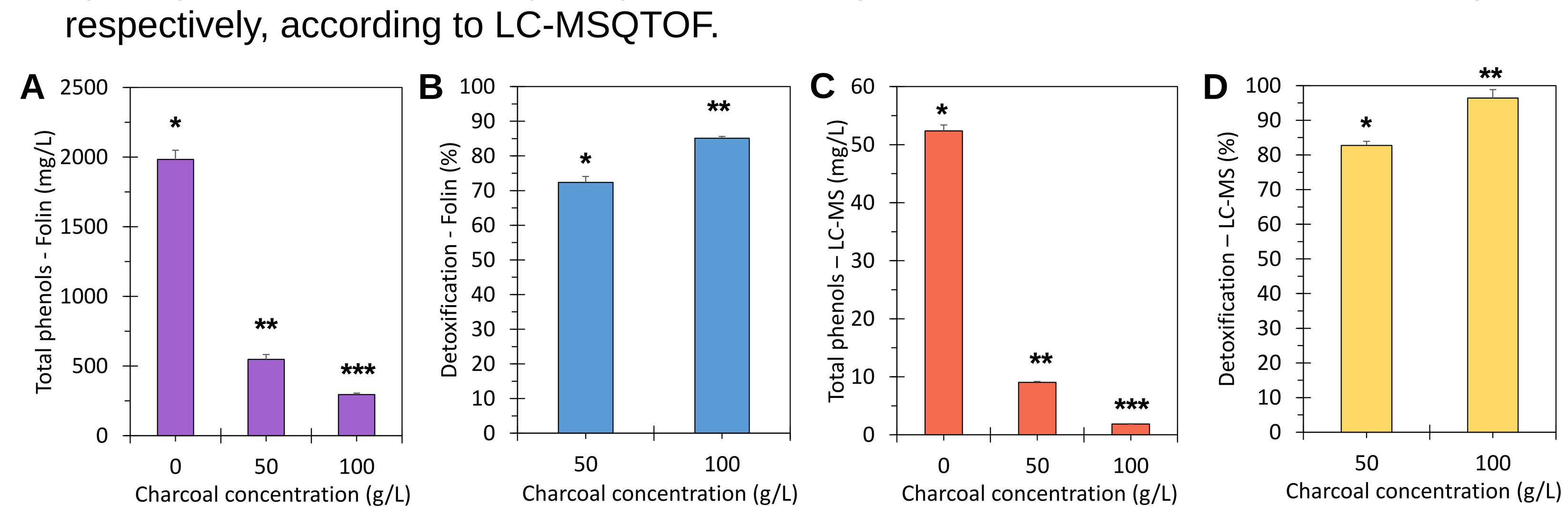


Figure 3 – Quantification of total phenols, according to Folin Ciocalteu's methodology (**A**) and sum of individual quantification by LC-MSQTOF (**C**). Percentage of polyphenols detoxification (%), according to Folin Ciocalteu's (**B**) and LC-MSQTOF methodologies (**D**). Higher number of asterisks represents higher statistically significant difference.

Conclusions

- **Charcoal** (Carbone Superattivo CP) was **more effective** than bead resins in the adsorption of polyphenols from sugarcane syrup.
- **Agitation** speed and time were tested, and it **did not influence** phenols detoxification by charcoal.
- **Very high detoxification** was reached (up to **96.42 %**, measured by **LC-MSQTOF**) using 100 g of charcoal per L of syrup.
- **Gentisic acid**, **hydroxybenzoic acid** and **hydroxybenzoic-4-B-glucoside** remained in the syrup after detoxification.

Future Work

- The **detoxification** process will be further **optimized** to obtain **complete removal** of phenols and **minimize** charcoal **usage**.
- **Shake-flask** fermentations will be performed to evaluate yeast **performance** in **phenols-free** syrup, as compared to its performance in **non-purified** syrup.
- **Bioreactor** fermentations, simulating **industrial conditions**, will be implemented to validate the results found in flasks, and the **potential** for **scaling-up** syrup detoxification will be assessed.

References

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