

# Wine lees: characterization and valorization by kombucha fermentation

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#### **1. INTRODUCTION**

Winemaking generates large amounts of by-products: grape pomace, stalks, vine shoots, filtration cakes and wine lees. Although being a rich source of bioactive compounds, wine lees are the least studied and exploited by-products of the wine industry [1]. Kombucha is a traditional beverage originally prepared by the fermentation of sweetened tea leaves with a symbiotic consortium of bacteria and yeasts (SCOBY). Nowadays, Kombucha has been gaining a lot of popularity due to the health benefits associated with its consumption. Different substrates are being inoculated with tea fungus to produce novel functional beverages and to valorize agro-industrial by-products [2].

## 2. MATERIALS AND METHODS

Wine lees were diluted with distilled water (1:2 and 1:4), sweetened with 70 g/L sucrose, inoculated with the Kombucha culture (20% w/v tea fungus and 20% v/v tea kombucha). The samples were fermented for 25 days at 25°C. The pH was determined using a pH meter and the titratable acidity was analyzed according to the method stated by the OIV (Method OIV-MA-AS313-01). Brix values were measured using a refractometer. The concentration of total polyphenols was determined by following the Folin-Ciocalteu colorimetric method and the concentration of total anthocyanins by the sulfur discoloration method. The tannin content was estimated by following the Bate-Smith reaction principle [3]. The antioxidant potential was assessed by measuring the inhibition activity against DPPH (2,2-diphenyl-1-picrylhydrazyl) and ABTS (2,2'-Azino-bis (3-ethylbenzothiazoline-6-sulfonic acid)) radicals [4].

WLK-1/2 will be used to refer to wine lees Kombucha diluted to  $\frac{1}{2}$ , and WLK-1/4 will be used to refer to samples diluted to the  $\frac{1}{4}$ .

#### **3. RESULTS AND DISCUSSION**

#### 3.1. Physico-chemical characteristics

#### *3.1.1. Brix and pH*

The brix and pH measurements throughout fermentation are portrayed in figure 1. A reduction in brix values in detected throughout the fermentation, indicating a consumption of sugars by the microorgnaisms present in the SCOBY. At the end of fermentation, the decrease in brix level was estimated to be 74-77%.

The pH of wine lee Kombucha decreased during fermentation as reported in kombucha literature. This observation is mostly associated with the production of organic acids by the microorganisms in the inoculum.



Figure 1: pH and Brix values of wine lees Kombucha

#### 3.1.2. Total acidity

The evolution of total acidity during the fermentation of wine lees with the kombucha inoculum is presented in figure 2. A continuous increase in the total acidity in both samples is noticed, reaching 25.97 g/L equivalent sulfuric acid in WLK-1/4, and 17.57 g/L equivalent sulfuric acid in WLK-1/2. The increase in sulfuric acid is most probably associated to the biotransformation of simple sugars and ethanol into organic acids [5][6].



Figure 2: Total acidity of wine lees Kombucha

#### 3.2. Bioactive compounds and antioxidant activity

#### 3.2.1. Total polyphenols, anthocyanins, and tannins

The concentrations of total polyphenols and total anthocyanins during the fermentation of wine lees with the Kombucha consortium are presented in figure 3. Originally, wine lees contained abundant concentration of polyphenols (887.5 and 1302.6 mg/L GAE), and anthocyanins (54.25 and 118.56 mg/L). As shown in figure 3, the content of polyphenols and anthocyanins fluctuated during fermentation, indicating that the bioactive compounds went through structural changes as a result of the activity of the microorganisms. The highest polyphenolic content was reported on day 21 in WLK-1/2, and on days 6 and 21 in WLK-1/4. The reduction in the concentration of polyphenols during Kombucha fermentation was also reported when using winery effluent as a substrate [7] and could probably be attributed to the polymerization of phenolics into larger molecules or to their adsorption by the biofilm. The total anthocyanin content decreased in WLK-1/2 throughout fermentation, whereas the reduction occurred after day 9 in WLK-1/4.



The variation of tannin concentration during fermentation is portrayed in figure 4. The tannin content in WLK-1/2 increased to reach a maximum on day 3 (1.65 g/L), and then decreased throughout fermentation, with the exception of the slight enhancements detected on days 18 and 24. On the other hand, a decrease in the content of tannins was observed during the first 3 days of fermentation, followed by a slight increase until day 18.



Figure 3: Total polyphenols and anthocyanins in wine lees Kombucha



Figure 4: Total tannins in wine lees Kombucha

# 3.2.2. Antioxidant activity

The antioxidant activity of wine lees Kombucha is presented in figure 5. Initially, unfermented wine lees exhibited an inhibition of 75 to 82 % of DPPH radicals. An increase in the antioxidant potential was detected in both samples, reaching a maximum of 93% on day 9 in WLK-1/2, and 90% on day 18 in WLK-1/4. The improvement of the antioxidant activity after inoculation with SCOBY has been widely reported in Kombucha literature [8][9]. On the other hand, fermentation with tea fungus had a minimal impact on the inhibition of ABTS radicals. Originally, unfermented wine lees had inhibition of around 51 µmol trolox eq/ ml. A small increase was observed until day 21, followed by a decrease.





Figure 5: Antioxidant activity of wine lees Kombucha

## 3.3. Discussion and conclusion

Wine lees are a rich source of bioactive compounds and were found to exhibit a notable antioxidant activity by inhibiting DPPH radicals. Kombucha fermentation has been found to valorize wine lees, one of the by-products of the wine industry. An increase in the concentration of total polyphenols, anthocyanins, and tannins was observed, in addition to an improvement of the antioxidant activity (DPPH). Further investigation is needed though to optimize the processing parameters, in order to obtain the most optimal quality of the product. Moreover, it would be interesting the explore the organoleptic properties of the developed beverage, and to evaluate additional biological activities, as reported in the tea Kombucha literature. Finally, it would be worth to consider the valorization of other types of winery by-products by the Kombucha consortium.

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