#### FOOD PRODUCT DESIGN - CHOCOLATE FORMULAS WITH ADDITION OF BREWER'S YEAST POWDER

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## Abstract

The purpose of this study was to use the beer yeast as a substitute for cocoa powder in the chocolate manufacturing recipe with high antioxidant properties. It is known that beer yeast is rich in high antioxidant compounds. To preserve these properties, the beer yeast has been conditioned at a moderate temperature ( $65^{\circ}$ C) to ensure its stability. Then, chocolate formulas, simple and with addition of beer yeast powder in different percentages, was obtaining. The total antioxidant capacity was measured by FRAP assay and to assess the total polyphenol content was used the Folin–Ciocalteu method. By incorporating the beer yeast powder into the chocolate recipe, an increase in the total phenolic content and total antioxidant capacity has been recorded compared to the control sample. Therefore, the total polyphenol content vary from 2.71 mM GAE/100 g DM in the control sample to 6.05 mM GAE/100 g DM in the chocolate formula with 40% yeast. Based on the results obtained from this study, we can affirm that the beer yeast can be used as an ingredient with high added value for designing innovative food products; in this case the chocolate with a more intense taste and flavor.

## Introduction

Chocolate is characterized as solid dispersion of cocoa mass, sugar, additives, cocoa butter, lecithin, and flavonoids and there may be variations between types [6].

Yeasts have a rich history and a bright future in biotechnology. Their involvement and importance in traditional food fermentations are unparalleled by other organisms of biotechnological relevance. Most yeast species are nonpathogenic to humans and animals and it is likely that yeasts will be increasingly used in traditional processes as their safety is more extensively established [5]. *Saccharomyces cerevisiae* is the principal yeast utilized in biotechnology worldwide, due largely to its unique physiology and associated key roles in many food fermentations and other industrial processes [10] including wine, beer, coffee and chocolate During wine fermentation, each strain of yeast releases distinctive metabolites, which contribute to flavor and aroma. So it, the beer yeast utilization in chocolate manufacturing recipe changes the chocolate taste and smell [9].

## Experimental

## Beer yeast conditioning

Fresh beer yeast was taken from the Ursus Breweries brewery from Timisoara- Romania, being by-product of the primary fermentation of brewer's wort. The beer yeast was kept in refrigerated condition (4-5°C) for 12 hours after which, was dehydrated in the laboratory condition. The dehydration process was performed with Food Dehydrator by Heinner/Germany at the working temperature of  $65^{\circ}C$  for 12 hours and at the fin of this process the moisture of beer yeast was 5.83%. The initial moisture of beer yeast was 64.34% wet basis.

#### **Obtaining chocolate formulas**

It was obtained, in manufacturing condition, the simple chocolate (no added) – C (control sample) and the chocolate formulas with addition of beer yeast powder in different percentages: 10, 20, 30 and 40% of total cocoa weight – which were noted: C1, C2, C3 and C4. Recipes for chocolate manufacturing formulas are shown in Table 1. It is noted that the cocoa powder content decreased proportionally with the percentage of yeast powder added in the chocolate formulas.

Ingredients	C	C1	C2	C3	C4
sugar [g]	420	420	420	420	420
butter with 80% fat [g]	200	200	200	200	200
milk power [g]	255	255	255	255	255
cocoa power [g]	150	135	120	105	90
beer yeast powder [g]	0	15	30	45	60
water [mL]	135	135	135	135	135

Table 1. The ingredients used to make chocolate formulas

## Chocolate manufacturing recipe

The sugar and the water are boiled for 10 minutes until the sugar is melted. After melting the sugar, added the butter. Mixed until the butter melts, remove from heat and add the mixture of milk powder and cocoa and stir continuously until smooth. The beer yeast is added in chocolate in different percentages: 10, 20, 30 and 40% of cocoa weight. Pour the melted chocolate on a silicone molds. After 2-3 hours checked if the chocolate is cool, remove from the molds, packaged and remained at 18°C.

## Chemical analysis

1 g of chocolate samples were grinded and were dissolved in 20 mL ethanol-water solution 45:55 (v/v), at room temperature. After 30 minutes, the samples was filtered and centrifuged for 10 min at 5000 rpm with Mikro 200 Microliter Centrifuges by Hettich Lab Technology / Germany. The supernatant were analyzed to determine the total phenolic content and the total antioxidant activity.

The *total antioxidant capacity* was measured by FRAP assay (Benzie and Strain 1996) [1]. FRAP reagent was prepared freshly by 10 mM TPTZ (2,4,6-Tris(2-pyridyl)-s-triazine) solution (diluted in HCl 40 mM), 20 mM FeCl<sub>3</sub>·6H<sub>2</sub>O solution and 300 mM sodium acetate buffer at pH 3.6 in the ratio of 1:1:10. Was added 0.5 mL hydroalcoholic extract samples diluted in the ratio 1:100 (v/v) in distilled water and 2.5 mL FRAP reagent. Absorbance was read at 593 nm, after 30 minutes, using an aqueous solution of FeSO<sub>4</sub> as standard. Correlation coefficient for calibration curve was:  $r^2 = 0.9972$ . Total antioxidant capacity was expressed as mM Fe<sup>2+</sup>/100 g DM (dry matter).

The *total polyphenol content* was determined by Folin-Ciocalteu method (Singleton and al., 1999) [7]. Briefly, 0.5 mL hydroalcoholic extract beer yeast and chocolate samples and diluted in the ratio 1:50 (v/v) in distilled water, 2.5 mL of Folin-Ciocalteu reagent (diluted 1:10 in distilled water) and 2.0 mL Na<sub>2</sub>CO<sub>3</sub> sol.7.5% was stirred and was kept at room temperature. After 2 hours, the absorbance was read at UV-VIS Spectrophotometer SPECORD 205 by Analytik Jena at wavelength  $\lambda = 750$  nm using gallic acid for calibration curve (r<sup>2</sup>= 0.9962). The results were expressed as mM GAE/100 g DM.

## Statistical analysis

All data are expressed as mean  $\pm$  standard deviation (SD) of three replicates (n=3). The simple linear regression analysis was performed using the computer software program Origin 8.0.

#### **Results and discussions**

Brewer's yeast power an excellent source of a variety of bioactive substances. The researchers found that brewer's yeast (*Saccharomyces cerevisiae*) contains probiotic, antioxidant and antimicrobial properties [3].

The antioxidant characteristics of the brewer's yeast power used as a functional ingredient to improve the antioxidant properties of the chocolate formulas are presented in Table 2.

Table 2. The antioxidant characteristics of the brewer's yeast power

Antioxidant characteristics	Values
Total antioxidant capacity [mM Fe <sup>2+</sup> /100 g DM]	147.22
Total polyphenol content [mM GAE/100 g DM]	59.42

The sensory properties of chocolate formulas with beer yeast are presented in Table 3.

Table 3. The chocolate sensory properties

The sensory properties	Appearance-exterior	Polished surface, without stains, scratches or air gaps	
	Appearance- interior	Chocolate with homogeneous consistency	
	Color	Dark-brown	
	Consistency	Hard and brittle	
	Taste and smell	Pleasant, taste and odor intense flavor due	
		to the yeast	
	The finesse	The product is unctuous	

Table 4 presents the total antioxidant capacity and the total polyphenol content for the different formulas of chocolate with different percentages of beer yeast powder.

Table 4. Total antioxidant capacity and total polyphenol content of chocolate formulas

The chocolate formulas	Total antioxidant capacity [mM Fe <sup>2+</sup> /100 g DM]	Total polyphenol content [mM GAE/100 g DM]
С	18.68±1.17	2.71±0.10
C1	21.97±1.24	3.54±1.14
C2	24.93±1.27	4.33±1.16
C3	28.01±1.37	5.42±0.21
C4	31.14±1.45	6.05±0.26

*Values are expressed as mean* - *standard deviation* (n = 3).

A significant difference can be noticed between control sample (18.68 mM  $Fe^{2+}/100$  g DM) and C4 chocolate formula (31.14 mM  $Fe^{2+}/100$  g DM) for the total antioxidant activity. Chocolate antioxidant properties are often claimed; however, they are frequently different from the parent natural sources due to the industry or manufacturing productions. Therefore, the antioxidant property of chocolate and cocoa are not adequately taken into consideration by consumers who use of this food as a desert for its flavor and taste properties [8]. Cocoa has long been identified as a polyphenols-rich food [4]. The content of polyphenols can vary tremendously depending on the source of cocoa beans, primary and secondary processing conditions and process of chocolates making [2]. The recently researches has been showed that chocolate, especially dark chocolate, is one of the most polyphenol-rich foods along with tea and wine [4]

According to the data presented in Table 4, we can observe that with the increasing the percentage of yeast powder in the chocolate manufacture recipe increases the total polyphenols content. Therefore,the total polyphenol content vary from 2.71 mM GAE/100 g DM in the control sample to 6.05 mM GAE/100 g DM in the chocolate formula with 40% yeast. So, the percentage of beer yeast that replaces cocoa powder in chocolate formulas, increases antioxidant properties of this products.

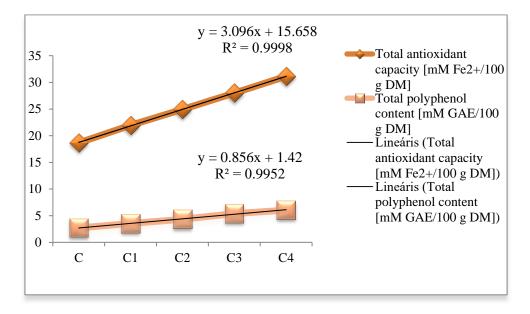


Figure 1. The relationships between the antioxidant properties chocolate formulas with beer yeast

All equations were revealed a positive relationship between the antioxidant properties of the chocolate formulas with beer yeast.

Figure 1 shows that there is a very good correlation between the antioxidant properties of the chocolate formulas. The coefficient of determination calculated is  $R^2$ = 0.9998 for the total antioxidant capacity, respectively  $R^2$ = 0.9952 for total polyphenol content.

## Conclusion

The beer yeast is a sustainable raw material available in large quantities, rich in high antioxidant compounds.

The drying at a moderate temperature of 65°C represents a simple method of beer yeast conditioning in order to preserve its antioxidant properties.

The addition of the beer yeast does in different percentage not induce negative changes in the sensory attributes of chocolate formulas, especially taste, smell, texture and consistency.

By incorporating the yeast powder into the chocolate manufacturing recipe showed an increase in the total polyphenols content and total antioxidant capacity (FRAP value) compared to the control sample.

The order of total polyphenol content in the chocolate formulas showed a similar trend as the total antioxidant capacity.

Based on the results obtained from this study, we can affirm that the use of beer yeast as a ingredient with high added value for designing innovative food products - chocolate with a more intense flavor.

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