# THE ABILITY OF WINE YEASTS FERMENTING BY THE ADDITION OF EXOGENOUS BIOTIN

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## **ABSTRACT**

Research is focused on the increase of the field of obtaining the wine yeast, under physical and chemical conditions. Study of different influences on yeast production is very important for the promotion of new cultivation methods for increasing both the fermentative and conservation capacity.

The present article deals with the study of biotin activity on the biotechnological properties of the wine yeast.

Our results showed that addition of biotin can offer beneficial conditions for improving the fermentation, being also an important factor of stability for wine yeast *Saccharomyces ellipsoideus*.

#### **KEYWORD**

Biotin - Saccharomyces ellipsoideus – fermentation - physical and chemical conditions

## INTRODUCTION

Because the enzyme equipment, yeast cell produces alcoholic fermentation using an enzymatic complex that catalyzes at different stages of oxidation-reduction processes of carbohydrates which can be fermented, ultimately resulting alcohol. The most important enzymes are dehydrogenates - glycerat and alcohol dehydrogenate, which has as coenzyme the nicotine-amide-dinucleotide which is important in the hydrogen transfer in catabolic reactions. Fermentation speed is an exponential function, being influenced by the number of cells / ml medium and starter culture of micro-organisms (Dan, 2001).

Most strains of yeast ferments substrates containing hexose and oligosaccharides: sucrose, maltose, raffinose, lactose and celobiose. Most vigorous cells and with high fermentation power are young cells compared with that aging cells which would ferment more slowly the same substrate.

Instead, biomass can be raised through various methods and additives, these being practically the theme of this paper.

By studying the accumulation of biomass, there is a good propagation of cells, reaching values of  $108 \div 1010/\text{cm}3$ , with a reduction in the percentage of autolysates cells (Banu, 2008).

An important aspect in the growth of multiplication rate of yeast cells is to determine the optimal conditions for cultivation. Mono-and multifactorial kinetic dependencies were established, which describes the influence of concentration of the basic components of the nutrient medium, temperature, culture medium pH, mixing intensity on the rate of multiplication of yeast.

Many research aimed the possibility of increasing returns of wine yeast obtaining, both under the influence of physical and chemicals factors. The study of these factors lead to

the need to promote new farming methods aimed at increasing the activity of fermentation and storage extension (Tita, 2001).

## **MATERIALS AND METHODS**

Our study focused on monitoring the activity of three strains of fermentation wine yeast *Saccharomyces ellipsoideus* obtained from indigenous wine varieties Feteasca royal, namely: DVF12, DVF25, and DVF28. These three strains of yeast (20g) were subjected to alcoholic fermentation in a Laboratory bioreactor of 5 l (4 l useful volume) type Biostat A, B. Braun Biotech International, equipped with a computerized system for monitoring, control and registration, equipped with sensors for temperature, dissolved oxygen, free oxygen, carbon dioxide and optical sensor for recording the biomass. The transformation of the values read by the biomass optical sensor in mass units (g / l) is done with a calibration curve specific for wine yeast (Oprean, 2010).

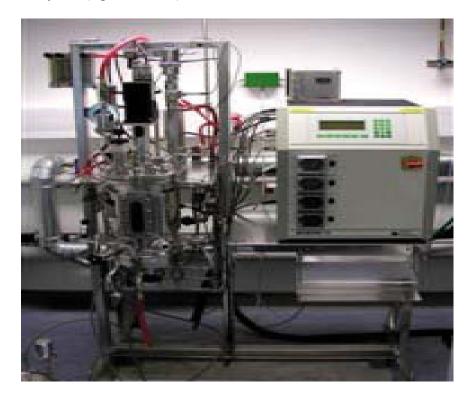


Fig. 1. Laboratory bioreactor type Biostat A, B. Braun Biotech International

- malt wort culture medium used join to the range produced by the Scharlau Chemie SA firm, Spain and contains malt extract 15 g / l, peptone 1 g / l maltose 12.5 g / l dextrin 2.5 g / l , dipotassium phosphate 1 g / l ammonium chloride 1 g / l, pH 4.8.
- fermentation was performed in the presence of biotin-2-oxohexahydro --thieno (3, 4-d) imidazole-6-YL with chemical formula:  $C_{10}H_{16}N_2O_3S$ , which was added successively in the following proportions: 1,5,10 mg / l.
- fermentation period was set at 10 days, the parameters observed being stored in a database.

## **RESULTS AND DISCUSSION**

After studying the results has been observed that addition of biotin increases the capacity of multiplication of *Saccharomyces ellipsoideus* wine yeasts, thus decreasing the

fermentation time. The biomass accumulation is more substantial in the culture medium in which an amount of biotin of  $5\mu g$  / l was added, the values being the same to higher added amounts. One of the explanations is that biotin promotes the formation of protein bonds in a form that can't be assimilated by the *Saccharomyces ellipsoideus* yeast.

The lag phase decreases and the exponential multiplication phase increases so we can clearly say that biotin affects the yeast cell's life cycle.

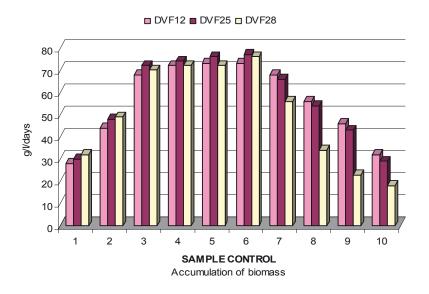


Fig.2. Biomass accumulation of the three control strains: *Saccharomyces ellipsoideus* DVF12, DVF25, DVF28

Figure 2 shows the variation of biomass accumulation of the three strains of yeast *Saccharomyces ellipsoideus* DVF12, DVF25, DVF28 considered blank, fermented in malt wort for 10 days .It can be seen that the peak period of fermentation is 3-6 days, the most active yeast being DVF25.

Figures 3-5 show the alcoholic fermentation of wine yeasts in the presence of biotin 1, 5,  $10\mu g/l$  and biomass accumulation.

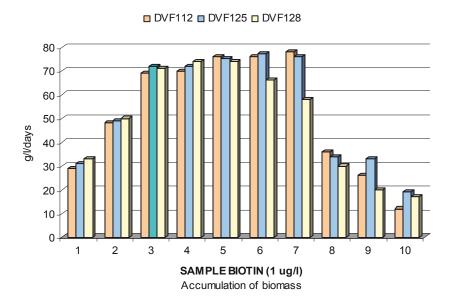


Fig.3. The biomasse accumulation of the three strains: *Saccharomyces ellipsoideus* DVF112, DVF125, DVF128 activated in the presence of 1µg/l biotin.

Note that the fermentation is prolonged by about one day, DVF228 and DVF225 being the most active yeast. It can say that the biotin added in the culture medium at this value serves to activate and to extent the fermentation and the amount of biomass increases.

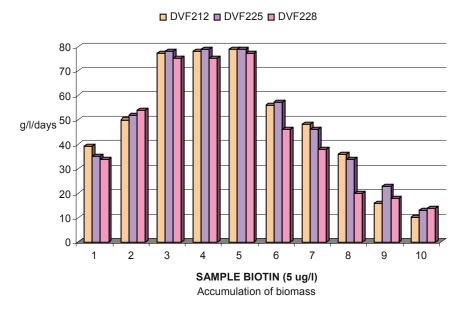


Fig.4. Biomass accumulation of the three strains: *Saccharomyces ellipsoideus* DVF212, DVF225, DVF228 activated in the presence of biotin 5µg / l.

If we added  $5\mu g$  / l, yeast activity increases rapidly in the early days, but then sudden drop in the 6 days, followed by a slow fermentation, but constant until the end of the monitoring period.

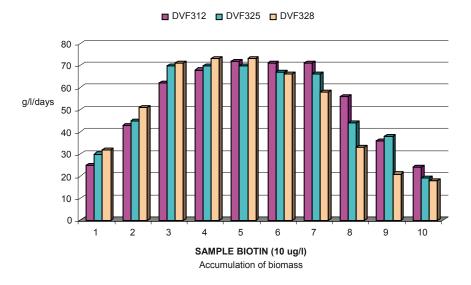


Fig.5. Biomass accumulation of the three strains: *Saccharomyces ellipsoideus* DVF312, DVF325, DVF328 activated in the presence of biotin 10μg / 1.

In figure 5 is seen a prolonged period of fermentation and constant up to 7 days, the amount of biomass is higher and the period of decrease is sudden and brief compared with the first two presentations.

## **CONCLUSIONS**

It appears that the addition of biotin can offer considerable benefits in terms of fermentation process and is a stabilizing factor for *Saccharomyces ellipsoideus* wine yeasts. Is apparent that biomass accumulation is inversely proportional to time, the most significant results, ranging from the 3 to 7 days. The decline period already started from day 5, at figure 5, and the optimal ratio of accumulation / time is shown in figure 4.

We conclude that biotin added to culture medium in controlled amounts (5µg / l) is a stimulating factor for the fermentation process, having a positive influence on the biomass accumulation. If it is added in excess (10µg / l) it is a hindrance in yeast multiplication, so biomass accumulation is reduced and the effects are less favorable. These results may be a prerequisite for obtaining biotechnological preparations with multiple practical utilities.

#### **AKNOWLEDGEMENTS**

This research paper was supported under the project "Increasing the role of the Phd studies and the competitivity of the Phd students, in an united Europe", POSDRU/6/1.5/S/26, ID 7706, coordinator Phd Prof..eng.Ovidiu Tita.

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