THE STATISTIC EXPERIMENTAL SHAPING OF INFLUENCE OF MEDIUM FACTORS UPON THE QUANTITY OF BIOMASS PRODUCED BY *SACCHAROMYCES* YEASTS

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Rezumat

În derularea oricărui proces fermentativ se impune o optimizare a condiţiilor fermentative. Prin modelarea procesului de stimulare a mediului fermentativ se înţelege obţinerea unei relaţii funcţionale între variabilele dependente şi o serie de factori experimentali.

Scopul experimentărilor a fost de a stabili un model matematic pentru a vedea dacă concentraţia de germeni de grâu şi de extreact de malţ sunt factori determinanţi pentru biomasa de drojdie.

Résumé

On imposeune optimization des conditions fermentatives dans le déroulment de tout processus fermentatif. Par modeler le processus de stimulation du milieu fermentatif on comprend l'obtention d'une rélation fonctionnelle entre les variables dépendantes et une série de facteurs expérimentause.

Le but des expériments a été d'établir un modèle mathématique pour voir si la concentration des germes de blé et de l'extrait de malt sont des facteurs déterminants pour le biomasse de leyure.

Abriss

In der Abwicklung jedem Gärungsprozess ist sehr wichtig eine Optimierung der Gärungsbedingungen durchzuführen. Durch die Modellierung dem Stimulierungsprozess des Gärungsmilieus versteht man die Erlangung einer funktionellen Beziehung zwischen de abhängigen Variabeln und eine

Serie experimentellen Faktoren. Der Zweck der Forschungen war die Bestimmung einem mathematischen

Muster um zu sehen ob die Konzentration der Weizenkeime und des Malzextraktes sind entscheidende Faktoren für die Hefenbiomasse.

Introduction

The main invertase producers are yeasts Saccharomyces cerevisiae, cultivated on fermentative medium, which contain different sources of carbon, nitrogen, growth factors and minerals.

In biology it is very difficult to obtain a functional relationship between the dependent variables and a series of experimental factors, and when we obtain that it is a very complex equation.

The experimental factors can be quantitative factors (temperature, concentration of one nutrient, etc.) or qualitative factors (different source of carbon or nitrogen)

In order to establish the correlation between the essential nourishing components for the efficiency of biomass biosynthesis, the factorial experiments were carried out and the results were performed through the regress equation and the analysis of respond surfaces.

Statistic processing has in view to establish some correlation, between quantity of yeast biomass and simultaneous action of the two chosen quantitative factors: the quantity of wheat germs and the quantity of wort extract.

For this purpose a series of culture medium were conceived, through concentration variation of two nutritive sources chosen as studying factors like independent variables.

Materials and methods

As a source of yeast we used baker yeast (*Saccharomyces cerevisiae)* from ROMPAK S.A. with 32,5% dry matter, and 46,54% protein content (N x 6,25).

As essential medium for the yeast's growing we used industrial medium adapted to the laboratory conditions: 40cm^3 wort, 0,08g $(NH_4)_2HPO_4$, 0,08g KH_2PO_4, 0,02g $Mg(NO_3)_2$ and 0,02g KNO_3.

Starting from this medium a series of culture medium were conceived through concentration variations of the two nutritive sources chosen (wheat germs and wort extract).

Yeast growth was studied in the same conditions of time, temperature, *p*H and stirring.

The general work scheme was: the baker yeast sizing at 10^6 cells / cm^3 was suspended in $40cm^3$ nutritive medium, in aseptic conditions. The tests were maintain on mechanical agitator (230 rot/min), at 30^0C, *p*H= 4,5, for 24hours.

Then the tests were centrifuged 25 minutes at 4000 rot/min for obtaining the biomasses.

The obtained biomasses were studied following the determination of humidity and dry substance.

In order to appreciate the degree of increasing of commercial strain of yeast in the work conditions we determinated growth by global evaluation of biomasses forming with determination of optical density at λ = 600 nm, initial and at the end of cultivation.

Results and discussion

This study supposed a regresion and correlation analisys. The regresion is studying the tipe of dependence between variables, and correlation measure the degree of this dependence on.

We had in view the following characteristics:

A = quantity of biomass

x_1 = Quantity of wheat germs

x_2 = quantity of wort extract

First, we study liniar simply dependence, like:

$$A = A(x1);\quad A = A(x2); \qquad [1]$$

The date selection we had in view are shown in table 1.

Table 1

The variation of biomass quantity depending on the two variables taken separately

Nr. crt	x1=x2	A(x1)	A(x2)
1.	0.05	2.08	2.09
2.	0.10	2.20	2.10
3.	0.15	2.39	2.15
4.	0.20	2.57	2.70
5.	0.25	2.56	2.62
6.	0.30	2.58	2.65
7.	0.35	2.57	2.64

In a rectangular axes system, the pair of experimental dates (x_i, y_i) represents a crowd of points, which can approximate a line, named regression line of Y characteristic in relation with independent variable X.

The linear dependence between A and x_1 or x_2 can be generic restore by an equation like this:

$$y = a + b \cdot x, \qquad [2]$$

Where the coefficients of regression a and b, are calculated with *the smallest square points method of Gauss.*

By analog method we can define a regression line of X characteristic in relation with independent variable Y. The equation is:

$$x = \alpha + \beta \cdot y \qquad [3]$$

The coefficient of correlation, independent of the measurement units of characteristics is a quantitative expression of a linear dependence.

Its formula is found in specialty literature:

$$r_{xx} = \frac{N\sum_{i=1}^{N} X_I Y_I - \sum_{i=1}^{N} X_I \cdot \sum_{i=11}^{N} Y_i}{\left[N\sum_{i=1}^{N} X_i^2 - \left(\sum_{i=1}^{N} X_i\right)^2\right]\left[N\sum_{i=1}^{N} Y_i^2 - \left(\sum_{i=1}^{N} Y_i\right)^2\right]}, \qquad [4]$$

The results of regression and correlation analysis are shown in table 2.

Table 2

Simple linear regression

$y = a + b \cdot x$	The coeficient a	The coeficientul b	Coeficient of correlation
$A = A(x1)$	1,7143	2.0786	0,8980
$A = A(x2)$	2,3000	1.9614	0,8573

The conclusion is that there is a certain linear dependence between the compared variables because the correlation coefficients had values around 1. The experimental data, the regression line and the spline cubic curve are shown in figure 1 and figure 2.

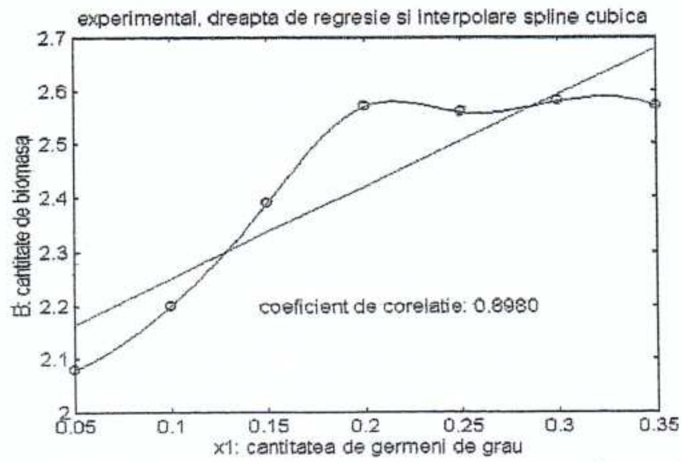

Figure 1. Mathematical model of evolution of yeast biomass quantity in relation by wheat germ quantity

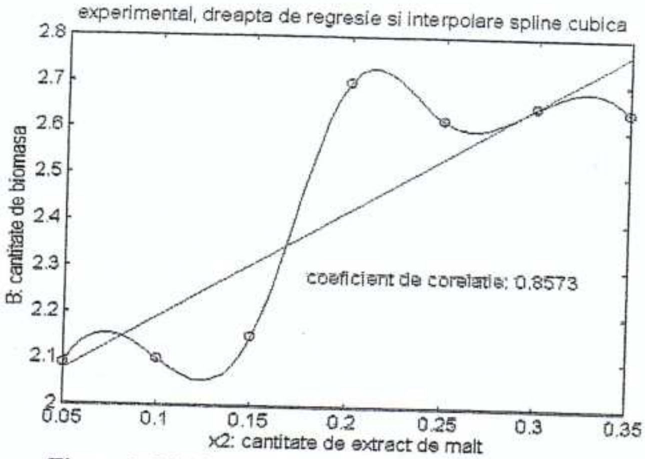

Figure 2. Mathematical model of evolution of yeast biomass quantity in relation by wort extract quantity

On base of experimental data a spline cubic interpolation in each case was realized. These data can be replaced, greatly, by effectuating a large number of experiments.

The next step was to evaluate statistic the linear multiple dependences, like:

$$A = A(x1, x2) \quad [5]$$

This study was necessary to evaluate the influence of simultaneous action of both factors. The results are shown in table 3.

Table 3

Bidimensional dependences

Nr.crt.	x1	x2	A=A(x1,x2)
1.	0.05	0.05	2.16
2.	0.10	0.10	2.45
3.	0.10	0.10	2.29
4.	0.10	0.10	2.38
5.	0.15	0.15	2.58
6.	0.15	0.15	2.25
7.	0.20	0.20	2.23
8.	0.20	0.20	2.58
9.	0.20	0.20	2.66
10.	0.20	0.20	2.85
11.	0.25	0.25	2.76
12.	0.30	0.30	2.87

Linear multiple dependence between A and (x_1 and x_2) can be related by equation like:

$$y = a + b \cdot x1 + c \cdot x2, \quad [6],$$

were the coefficients of regression a, b and c, are calculated with *the smallest square points method of Gauss*.

The results of regression and correlation analysis are shown in table 4.

Table 4

Values obtained for the coeficients of liner multiple regresion

$y = a + b \cdot x1 + c \cdot x2$	The coeficient a	The coeficient b	The coeficient c	The coeficient of multiple corelation
$A = A(x1, x2)$	2,0079	1,9532	1,0833	0,8090

We establish that respond surface variation for polynomial equation by evaluation of yeast biomass quantity changes like in figure 3.

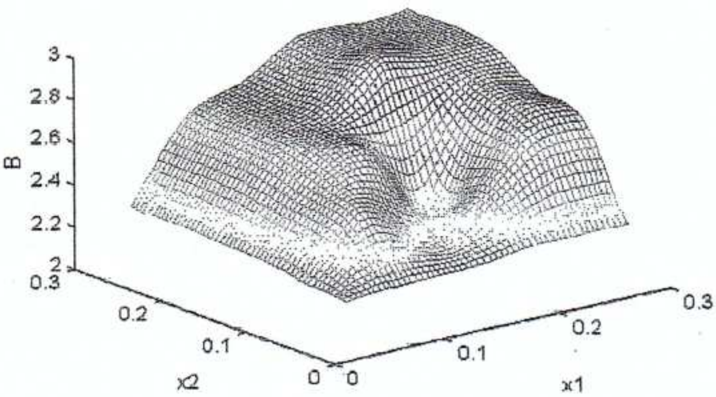

Figure 3. The evolution of correlation between wheat germs concentration and wort extract in medium composition and yeasts biomass

The respond surface presented in figure 3 permitted to establish optimal concentration of nutrients for which we obtain maximal values of biomass biosynthesis.

Conclusions

After statistical study we selected the factors which have an essential influence upon the invertase activity.

We showed the fact that the two independent variables in fermentative medium are essential for carbon and nitrogen, minerals, growing factor contributions and the fact that they have a favourable influence from qualitative and quantitative point of view upon increasing the biomass quantity of baker yeast *Saccharomyces cerevisiae.*

The results of statistical shaping show that the optimal concentration for the increase of yeast biomass quantity are 4,75 g/cm^3 of wheat germs and 3,75 g/cm^3 of wort extract.

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