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Optimization of Drying Process of *Zea Mays* Malt to Use as Alternative Source of Amylolytics Enzymes

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ABSTRACT

This work aimed to study the drying process optimization of maize (Zea Mays) malt for obtaining maize malt, without affecting enzymatic activity of α e β -amylases from maize malt. Results showed that dryer operation must occur in zone at 54°C and 5.18-6 h process time. The maize malt obtained had good enzymatic properties.

Key words: α and β -amylases enzymes, maize (*Zea mays*) malt; drying process, optimization

INTRODUCTION

Drying process is used with the aims to keep the enzymatic activity of agricultural products. It involves removal of volatile substances (commonly, but not exclusively, water) from a solid product, or it is a process in whith the water activity of a product is decreased due to removal of water by vaporization. Thus, the drying process is the conjuct of science and technology that needs of experiments on several phenomenons that occur in this process (Jesus, 2002). Some considerations about the initial and moisture content of product are used to justify the drying phenomenon. These considerations are the form of the water transport into solid structure and in the solid surface (Brod, 2003).

In 1998, a transporter type dryer straw mat was made by Freire and Sartori with aim to analyze its behavior in the drying process of the *Brachiaria brizantha* grass seeds. They evaluated the drying kynetics and the process effect on the seed quality (germination power). They concluded that the dryer was viable and the quality test showed that the grass seeds dryed at temperatures more or equal to 48.5 ºC had low germination power.

Of late, the use of enzymes in feeding, pharmaceutics, textile and others industries is increased for being economicaly viable. They are also used in the manufacture of alcoholic drinks, detergent, biosensors, diagnostic kits and in the management of environment polluents (Jesus, 2002). The α and β - amylases are commercial enzymes used its many applications, mainly in the starch hydrolyses (Fogarty and Kelly, 1979; Wiseman, 1987). They are obtained commonly from barley malt or microorganisms.

α-amylase (EC 3.3.1.1; α-1,4 glucan, 4 - glucanhydroxilase) is extracellulas enzyme that hydrolyses of the α-1,4 bonds, of the amylose, amylopectin, glycogen and dextrin molecules, but can not hydrolyses α-1,6 bonds. It has molecular

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weight about 50 kDa, with isoelectric point 5.4, very good enzymatic activity about pH 4.7 and 75 °C (prorerties enzymes dependes from source) (Reguly, 1996; Wiseman, 1987).

β-amylase (EC 3.2.1.1, α-1,4, glucanmalthydrolase) is a extracellulas enzyme that hydrolyses of amylopectin and glycogen, breaking each second α-1,4 bond. It has molecular weight about 50 kDa, with isoelectric point 5.4, very good enzymatic activity about pH 4.5-6.5 and 55-57 °C, it inactived after temperatures above of 60 °C (Reguly, 1996; Wiseman, 1987).

Maize *(Zea mays)* is a agricultural product very popular in Brazil that has low cost price. Thus, the maize malt obteining will go aggregate valor to maize culture. This work aimed study the drying process optimization of maize malt obtaining by response surface methodology (RSM).

MATERIALS AND METHODS

Maize malt obtaining

The maize seeds were selected, their weight was measured, washed, the seeds were carried to water absorption until 40- 45 % (w/w) and germinated in laboratory. The germination time was between 4-5 days. The maize malt was dried as show in Table 2 and storaged at 5 °C (Santana, 2003).

Drying process methodology

Convective dryer with air circulation was used in maize malt drying process. Dryer was operated in constants condictions of temperature, relative moisture (64 ± 1%) and air flux. Tree drying temperatura were utilized 54, 64 e 76°C. The germinated seeds (malt) were placed in watch glass (previously dried and its weight measured). The initial weight (${\boldsymbol{X_0}}$) were measured and they were dried for 12 h into dryer. Dry weight (${\boldsymbol{X_i}}$) were measured after each hour. The moisture content ($\boldsymbol{U}$) is given to equation 1 and experimental weight (${\boldsymbol{X_{exp}}}$) is given to equation 2 (Freire e Sartori, 1998; Jesus, 2002), bout are sown at following:

$$U = \left(\frac{X_0 - X_i}{X_0} \right) * 100 \qquad (1)$$

$$X_{\exp} = \frac{X_i}{X_0} \qquad (2)$$

Determination of protein total concentration

Protein concentration in samples was measured by Bradford method (Bradford, 1976).

Enzymatic activity analysis

In maize malt samples the enzymatic activity was determined by Wohlgenuth method, modified by Sandstedt, Kneen and Blish (1959), showed of according to Reguly (1996). A SKB is amongh of starch content dextrinized for one hours and one gram of amylase.

Drying process optimization

The influence of the time (***t***) and temperature (***T***) on drying process on the enzymatic activity (***AE***) of the maize malt was studied by 2^2 experimental planning with hexagon design (showed in Table 2) and response surface methodology (RSM) for optimization of drying process. The enzymatic activity was shown in ***ln AE*** form in the models. Linear, hiperbolic and square models were tested. The least square were used to estimate the madel parameters and the model fitting were made by analysis of variance methodology (ANOVA), all showed in Barros Neto et al., 1995 and 2001). The following variables codifications were used:

$$x_1 = \frac{t_i - 8}{2} \qquad (3)$$

$$x_2 = \frac{T_i - 65}{11} \qquad (4)$$

RESULTS AND DISCUSSION

Drying process

Table 1 showed the eperimental weight of the malted seed at different drying time and temperatures. Results showed that the moisture content was between 40 at 43 % (w/w), and more high moisture desorption rate was shown at 75°C.

Drying ishotems showed in the Fig. 1 were obtained with the data of Table 1. Dry weight began to become constant between 3-4 hours. In this moment, the drying curves showed paralels to the time axias (Brod, 2003; Freire e Sartori, 1998; Jesus, 2002).

Table 1 -Experimentals data of drying process of maize malt.

Assays	Drying temperature					
	54°C		65°C		76°C	
	t (h)	X_{exp}	***t*** (h)	X_{exp}	***t*** (h)	X_{exp}
	0	1	0	1	0	1
1	1	0.6822	1	0.6663	1	0.6950
2	2	0.6493	2	0.6037	2	0.6408
3	3	0.6227	3	0.6000	3	0.5844
4	4	0.6136	4	0.5946	4	0.554
5	5	0.5854	5.15	0.5775	5	0.5789
6	6	0.5638	6	0.5635	6	0.5854
7	7	0.5891	7	0.5833	7	0.5586
8	8	0.5915	8	0.5796	8	0.6322
9	9	0.5667	8	0.5787	9	0.5338
10	10	0.5860	8	0.5802	10	0.5648
11	11	0.6067	9	0.5781	11	0.5469
12	12	0.5773	10	0.5802	12	0.5888
13			10.85	0.5900		
14			12	0.6027		

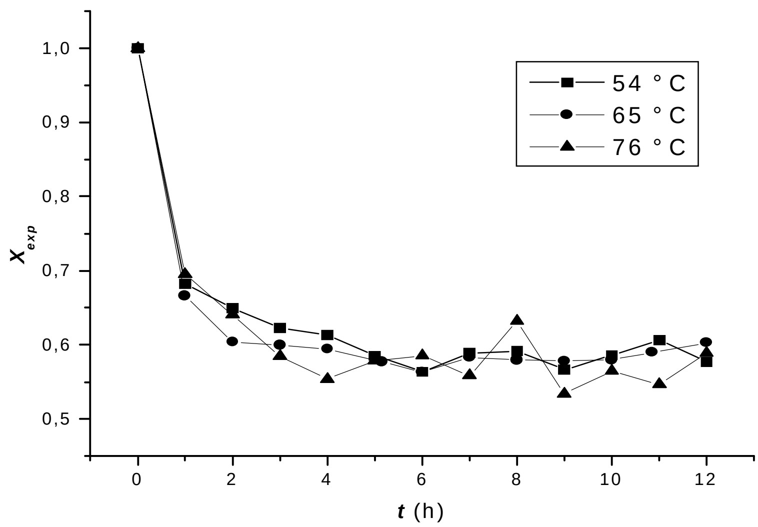

Figure 1 - Drying isotherms of the maize malt.

Table 2 -Planning matrix of drying process optmization of the maize malt.

Assays	***t*** **(h)**	***T*** **(°C)**	x_1	x_2	***AE***
1	6	54	-1	-1	6.684
2	10	54	+1	-1	1.710
3	6	76	-1	+1	0.1292
4	10	76	+1	+1	0.2656
5	8	65	0	0	0.4822
6	8	65	0	0	0.4522
7	8	65	0	0	0.5851
8	5.18	65	-1.41	0	0.2571
9	10.8	65	+1.41	0	0.3097

Table 3 -Variance analysis (ANOVA) of the fitting model.

Souce of variance	Square sum	Free degree	Medium square sum	F_{calc}	F_{tab}
Regression	10.981	5	2.196		
Residual	0.211	3	0.070	31.269	9.01
Fitting fault	0.175	1	0.175		
Erro Puro	0.036	2	0.018	9.718	18.51
Total	11.118	8			
	% explicated variance =			98.763	
	% maximum explicated variance =			99.677	
	Determination coefficient (R^2) =			0.9876	

Drying process optimization

Table 2 showed the assays, the factors in the normal (***t*** and ***T***) and coded form (x_1 and x_2) and the response (***AE***). These data were used for obtaining models parameters by least square and for fitting model by ANOVA methodology (Barros Neto et al., 1995).

The fitting model is shown in the equation 3, that one introduces a square dependence of enzymatic activity (in ***ln AE*** form) with the drying time and temperature. There is a low influence of linear time and negative high of linear temperature, but all parameters have statistical significants.

$$\ln AE = -0.6886 + 0.0476 t_1 - 1.4522 T_2 - 0.2910 t_1^2 + 0.7437 T_2^2 + 0.5209 t_1 T_2 \qquad (5)$$

Table 3 shown the resulties of variance analysis (ANOVA) for the fitting model. According to Barros Neto et al. (1995 and 2001) R^2 must have valor about 1.0 and the variances must have valors about 100. Resulties ANOVA for the variances and R^2, showed that there was low error quantity added to the fitting model due to regression and due to the employ of the analytical methods.

Resulties of the F test, whole the first test (F_{calc}/F_{tab}) indicated that the model was statistical significant and the predict data were approaching the experimental data. The second test (F_{tab}/F_{calc}) indicated that the data were fitting and they were statiscal representing the response surface. For the F tests are statistical considerate the rate much be igual or more that 4 (Barros Neto et al., 1995). According to Barros Neto et al. (1995) bout F teste must be ≥ 4 for the model is statistical significant and preditive. Resulties showed that the model was statiscal significant, but it was not full predictive for optimize of the drying process of maize malt, in the studied condictions (Barros Neto et al., 1995 e 2001).

Figs. 2 and 3 show the 2D and 3D response surfaces for the drying process optimization. Enzymatic activity (on ***lnAE*** form) increased with reduction of the drying time and temperature. If the time factor was fixed in the minor valors (-1 at -1.41 or 5.18 at 6 h) and the temperature factor was augmented to the maximum value (+1 or 76 °C), there was a high reduction in the enzymatic activity, however, if the temperature in the minimum value (-1 or 54°C) and time was changed of minimum value (-1,41 or 5,18 h) until the maximum value (+1,41 or 10,8 h), the enzymatic activity was little loss, which shown that temperature effect was critical for the drying process of maize malt. This temperature effect on the enzymatic activity occurs due to previous inactivation the β-amylases after it exposes above 54°C. Thus, for response surface analysis may be indicated optimal operation condition for maize malt drying process was 54°C and 5.18-6 h of drying time.

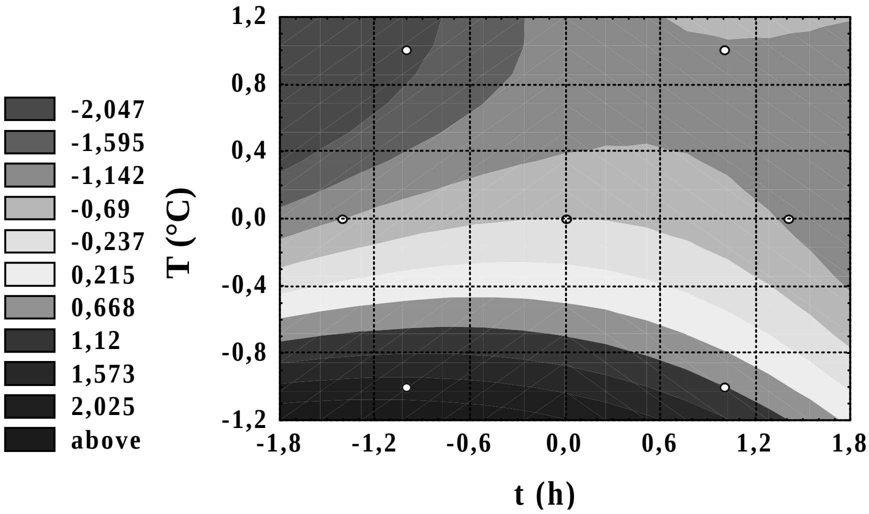

Figure 2 - 2 D response surface for understanding the enzymatic activity dependency with the time and temperature in the drying process of maize malt.

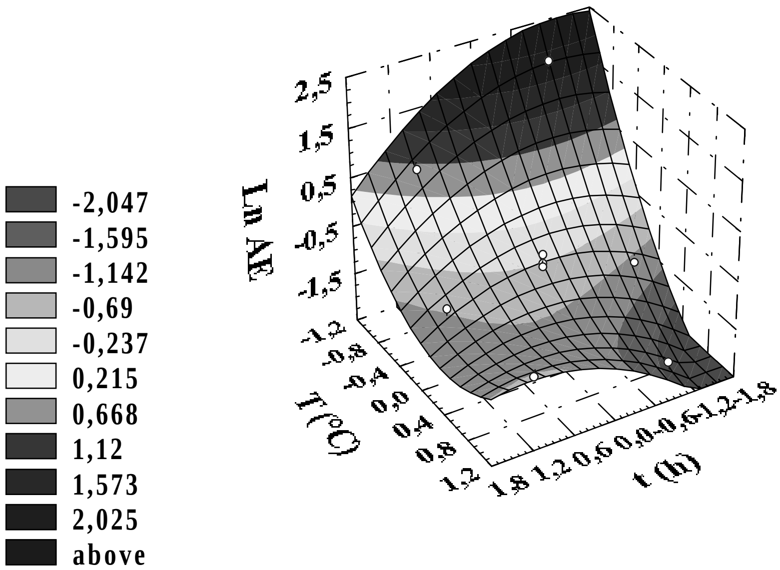

Figure 3 - 3D response surface for understanding the enzymatic activity dependency with the time and temperature in the drying process of maize malt.

CONCLUSIONS

The analysis of the results carried the following conclusions:
Drying process data showed that moisture removal from maize malt was above 40 % (w/w).
The drying isotherms were shown that moisture desorption rate was fixed after 3-4 h of drying process.
The variance analysis (ANOVA) showed that the best fitting model was the that having to square dependency of the enzymatic activity, in ***lnAE***

form, with the drying time (***t***) and the temperature (***T***).
The response surface analysis showed that influence of drying temperature on enzymatic activity was more than drying time and the best operation condition was 54°C and 5.18-6 h for the maize malt drying process.

NOMENCLATURE

AE	Enzimatic Activity (SKB/mg of protein)
F_{Calc}	Calculated F test.
F_{Tab}	Tabled F test.
i	Level of factors
T	Drying temperature (°C)
t	Drying time (h)
U	Moisture content (% w/w)
X	Drying weight (dimentionaless)
x_1 ***e*** x_2	Coded factors

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RESUMO

Este trabalho objetivou a otimização da secagem do malte de milho (*Zea Mays*) para obter um malte sem afetar a atividade das enzimas presentes neste, α e β -amilases. Os resultados mostraram que a operação do secador deve ser feita a 54°C e entre 5,18-6 h de processo. O malte obtido possuiu boas propriedades enzimáticas.

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