MODELLING FOR CONTROLLING POULTRY SLICED SAUSAGE LIPID OXIDATION AND DISCOLORATION BY RESPONSE SURFACE METHODOLOGY

Andrea Carla da Silva Barreto[1a], Elza Iouko Ida[2], Rubison Olivo[1b], Massami Shimokomaki[1b]& Rui Sergio Ferreira da Silva[2]

[1a]Department of Pharmaceutical Biochemistry and [1b]Department of Food and Experimental Nutrition, Faculty of Pharmaceutical Sciences, São Paulo University, Av. Prof Lineu Prestes, 580 CEP 05508-900-São Paulo, SP, Brazil [2.] Department of Food and Drugs Technology, The State University of Londrina, P.O.Box 6001, CEP 86051-970, Londrina, PR, Brazil.

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Background

In comparison to meat from mammals, poultry meat is composed of a relatively larger proportion of polyinsaturated fatty acids, particularly phospholipids. Therefore poultry processed meat is highly susceptible to lipid oxidation. A correlation between lipid oxidation and meat discoloration has recently been reported to exist in particular related to restructured poultry meat (1). Phytic acid prevents lipid oxidation by sequestering iron and forming a unique iron-chelate that becomes catalytically inactive (2). Tocopherols inhibit lipid oxidation by donating a hydrogen radical to free radicals and render free radicals inactive. Tocopherols are currently in use in meat and poultry industry (3). Response Surface Methodology (RSM) is a mathematical-statistical technique composed of planning, modelling and experimental analysis to relate response to quantitative factor levels which affect these response results (4). The RSM permits the development of a predictive matematical model based on the experimental data, which can be utilized for controlling purpose.

Objectives

The objective of this work is to model lipid oxidation and color changes controlling conditions using Phytic Acid (PA) and Alpha-Tocopherol (AT) as antioxidants by application of RSM in packed poultry sliced sausage stored at two commercial temperatures.

Methods

Boneless chicken thighs and drumsticks were kindly donated by CEVAL Alimentos S/A, Jundiai, SP. Samples were ground in Incomaf grinder (São Paulo, SP). Ground meat samples were mixed with other ingredients in a mixer. PA (Shikishima, Starch Co., Suzuka, Japan) kindly provided by Dr. Terao, NFRI, Tsukuba, Japan, was solubilized in distilled water and AT (Sigma) was mixed with salt before adding to the mixture. The poultry meat sausage formulation in % was as follows: Thighs and Drumsticks 84.8, Water 10.0, Starch 2.0, Salt 1.5, Sugar 0.2, Phosphate (Adicon) 0.3, Essential Oils (Adicon) 1.0, Curing Agents (Kraki) 0.3. The meat batter was packed in an impermeable and light- proof poliamide tubes. Packed samples were cooked in water bath at 70°C until internal temperature of the sausage reached 66°C. Samples were cooled down using running water to 40°C and stored at temperature app. of 5°C.

Sausage Slicing. Samples for slicing were taken after 35 days of storage. Slicing procedure was carried out with Vicris slicer and kept in a polystirene tray and covered with oxygen permeable film. To mimic commercial conditions, packed samples were stored at 7°C under luminosity produced by six Osran lamps (9 watts each) during first 14h followed by no luminosity for 10h., in sequence.

Analyses.Three sausage samples stored for 24 and 48h were randomly selected. For color analysis, triplicate measurements per sausage were obtained and averaged. Minolta Spectrophotometer (M-508d) calibrated with a standard white tile was employed.The three sausage samples used for color evaluation were also analysed for rancidity measurement (5).

Modelling. RSM was designed to test the effect of two antioxidants (PA and AT) and their combination on sliced chicken sausage lipid oxidation and discoloration. The complete factorial design 3^2 (Table 1) was applied to a total nine randomized assays. Table 2 shows the definitions of the independent variable and their levels. In every assay, the response-function (Y) was expressed in mg of TBARS/100kg of product stored at 7°C. The regression and analysis of variance were determined by SAS (SAS, 1985) and response surface by STATGRAPHICS Statistical Graphics Corporation. Results were fitted to a predictive and suitable polynomial model of the form given below:

$$Y= b_o+b_1x_1+b_2x_2+b_{11}x_1^2+b_{22}x_2^2+b_{12}x_1x_2+e$$

where Y= predictable reponses, x1,x2= coded variable levels b's=estimated coefficients and e is the random experimental error.

Results and discussion

Lipid oxidation.Table 3 shows the obtained avaraged results of lipid oxidation (mg of TBARS/100 kg of product) and discoloration of sausage (Δa^*) from sliced sausage stored for 24 and 48h, respectively. Tables 4 &5 present analysis of variance of response function Y for lipid oxidation and discoloration, respectively. It can be observed for lipid oxidation in 24 and 48h stored samples the complete regression was significative ($p<0.05$ and 0.001, resp.). The calculated value of R indicates 88.8% and 95.74%, resp. of variability which should be explained by the model hence there is good adjustment for experimental data. The coefficient of variation (CV=4.5 and 5.57%, resp.) indicated low variabilities of experimental conditions. It can be proposed therefore the models (equation 1 and 2, resp.) to explain sausage lipid oxidation inhibition by PA and AT by SAS (Statistical Analysis System):

$$Y_{24hTBARS}= 0.1734+0.0037x_1+0.0122x_2+0.0141x_1^2+0.0017x_2x_1-0.0164x_2^2 \text{ (eq.1) and}$$

$$Y_{48hTBARS}= 0.1807+0.0085x_1+0.0358x_2+0.0131x_1^2+0.0070x_1x_2-0.0119x_2^2 \text{ (eq.2)}$$

The canonic analysis of response surface (4) indicated the stationary point as a saddle point (minimax) of value of 0.1753 mg of TBARS/kg of sausage after 24h of slicing ie. x_1=0.15 and x_2=0.36. It can be observed in Fig 1A,B the lowest lipid oxidation occurred when x_1 is located in an intermediate values (-0.6 to 0.5) ie. 39.36 to 138.6 mg of PA/100 kg and x_2=-1.0 ie. no addition of AT. In sample stored for 48h, again low values of TBARS were related to a low conc. of AT. The canonic analysis indicated the stationary point or minimax of value of 0.2012 mg TBARS/kg of sample ie. when x_1= 0.67 and x_2=1.30 (extrapolated). In can be observed in Fig 2A,B the lowest lipid oxidation occurred when x_1 values are intermediate (-0.7 to 0.6) ie. 27.72 and 147.84g of PA/100g of sample and x_2=-1.0 ie. no addition of AT. In fact, the experimental number 2 (x_1:=0 and x_2=-1 resulted in lower values of TBARS.

Discoloration. Table 4 shows results of sausage discoloration (Δa^*) after 24 and 48h storage at 7°C of sliced sausage originally kept for 35 days at 5°C.Tables 5 and 6 present analysis of variance and regression respectively, of response function Y. It can be observed regression was highly significative ($p<0.001$). R^2 was 89.66% 87.60 for samples stored at 24 and 48h, respectively, of variation should be explained by the model indicating good adjustment of experimental data. CV=6.96% and 6.36, respectively, indicated acceptable

variabilities of the experiment. Hence it can be proposed the models for samples stored for 24 and 48h, as described by the equations 3 and 4 respectively, to explain sausage discoloration under PA and AT influence:

$$Y\Delta a^{*}_{24h} = 4.1507 + 0.7867x_1 - 0.1361x_2 - 0.1078x_1^2 + 0.3233x_1x_2 - 0.2928x_2^2 \quad \text{(eq.3)}$$

$$Y\Delta a^{*}_{48h} = 5.9952 + 0.7733x_1 + 0.0161x_2 - 0.7644x_1^2 + 0.1858x_1x_2 - 0.4728x_2^2 \quad \text{(eq.4)}$$

It can be observed in Fig. 3A,B and 4A,B the response surface described by the equations 2 and 3 in relation to x_1 and x_2 for samples stored for 24 and 48h, respectively. The lowest discoloration occurred when $x_1 < -0.7$ ie. $x_1 < 36.96$g of PA/100kg of sausage and $x_2 > 0.5$ ie. $x_2 > 30.0$g of AT/100 kg of sausage and $x_1 \cong -1$ ie. no addition of PA and $x_2 \cong 1.0$, ie. $x_2 \cong 40.0$g of AT/100kg of sausage in samples stored for 24 and 48h, respectively. The experiments number 4 ($x_1=-1$; $x_2=0$ and number 7 ($x_1=-1$; $x_2=1$) confirmed the models predictions.

Conclusions

-RSM predicted: 1- If the aspect of sliced sausage discoloration is the first quality to be selected for the consumer, AT should be the chosen antioxidant. 2- Conversely, if the more important quality item were lipid oxidation of sliced sausage, addition of PA should be recommended.

References

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Table 1- Complete factorial design (3^2) of RSM with assays coded and original variables

ASSAYS	VARIABLES X_1	CODED X_2	VARIABLES PA(g)	AT(g)
1	-1	- 1	0	0
2	0	- 1	92,40	0
3	1	- 1	184,80	0
4	-1	0	0	20
5	0	0	92,40	20
6	1	0	184,80	20
7	-1	1	0	40
8	0	1	92,40	40
9	1	1	184,80	40

Table 2- Definition and levels of independent variables

VARIABLES	LEVELS -1	0	+1
X_1 = g Phytic Acid/100kg product	0	92,40	184,80
X_2 = d of Alpha Tocopherol/100kg product	0	20,00	40,00

Table 3- Response functions of Y expressed in mg TBARS/kg of sausage and sausage discoloration (Δa^*) after 24h of slicing stored at 7°C packed in a permeable film. Samples were originally kept for 35 days at 5°C packed in an impermeable plastic film.

ASSAYS	TBARS/kg of sausage Y_{24h}	Y_{48h}	DISCOLORATION Δa^* $Y_{\Delta a^*24h}$	$Y_{\Delta a^*48h}$
1	0.159	0.151	3.67; 3.54; 3.63	4.30; 4.15; 4.23
2	0.141	0.122	3.62; 3.59; 3.62	5.23; 4.98; 5.23
3	0.163	0.152	4.40; 4.56; 4.67	5.45; 5.61; 5.79
4	0.179	0.177	2.99; 3.15; 3.44	4.23; 4.60; 4.66
5	0.173	0.181	4.20; 4.30; 4.35	6.41; 6.08; 6.55
5	0.184	0.193	4.62; 4.74; 4.92	5.50; 5.86; 5.48
6	0.186	0.198	4.62; 4.74; 4.92	5.50; 5.86; 5.48
7	0.181	0.204	2.30; 2.46; 2.37	3.53; 3.63; 3.95
8	0.163	0.203	4.03; 3.68; 4.21	5.28; 5.63; 5.73
9	0.192	0.233	4.63; 4.32; 4.85	5.99; 5.91; 5.61

Table 4- Models F values, coefficient of determination and variation for the fitted experimental data

	non-linear regression models Y_{24h}	Y_{48h}	$Y_{\Delta a^*24h}$	$Y_{\Delta a^*48h}$
F values	6.33*	18.00**	36.42**	29.66***
R^2	0.888	0.957	0.897	0.876
C V (%)	4.50	5.57	6.96	6.36

* P< 0.05; ** P< 0.01 and *** P< 0.001

Fig 1- Response surfaces for $Y_{24hTBARS}$, $Y_{48hTBARS}$, as related to PA and AT.
A $Y_{24hTBARS}$ B $Y_{48hTBARS}$

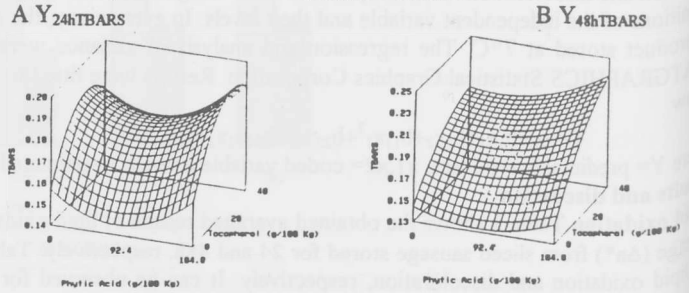

Fig 2- Response surfaces for $Y_{\Delta a^*24h}$, $Y_{\Delta a^*48h}$, as related to PA and AT.
A $Y_{\Delta a^*24h}$ B $Y_{\Delta a^*48h}$