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## **OPTIMIZATION OF OSMOTICALLY DEHYDRATED BEETROOT CANDY USING RESPONSE SURFACE METHODOLOGY**

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### **ABSTRACT**

Beetroot being an alkaline food having pH 7.5 to 8.0 contains higher antioxidant compounds. It has the potential to be used as a healthy food. Recent findings rank beetroots among the ten most potent antioxidant vegetables. The beetroot can be used as a treatment for fevers and constipation, amongst other ailments. For the optimization of osmotic dehydration by response surface methodology, the experiments were conducted according to Central Composite Rotatable Design (CCRD) with three variables at five levels. The low and high levels of the variables were 35 and 55°C for osmotic solution temperature, 30 and 60°Brix for sucrose solution concentration, 30 and 180 min for duration of dipping in osmotic solution, respectively. The fruit to solution ratio was kept 1:4 (w/w) during all the experiments. The optimum conditions for osmotic solution concentration, temperature and process duration were 60°Bx, 55°C and 180 min, respectively.

**Keywords:** Optimization, osmotic dehydration, beetroot, response surface methodology, sucrose.

### **INTRODUCTION**

In recent years increased attention has been focused on utilization of healthy foods. The beetroot (*Beta vulgaris*) being an alkaline food with pH from 7.5 to 8.0. has been acclaimed for its health benefits, in particular for its disease fighting antioxidant potential, significant amount of vitamin C and vitamins B<sub>1</sub>, B<sub>2</sub>, niacin, B<sub>6</sub>, B<sub>12</sub> whilst the leaves are an excellent source of vitamin A (Peter *et al.*, 2011). Sushanr *et al.* (2006) reported that at pH slightly above 7.4, cancer cells become dormant and at pH 8.5, cancer cells will die while healthy cells will live as alkalizing agents which will neutralize harmful acids. Chen *et al.* (2005) has shown that red beetroot pigments boost levels of proteins, called phase II enzymes that help detoxify potential cancer-causing substances and purge them from the body. Appel *et al.* (1997) reported that betanins obtained from the beetroots are used industrially as red food colorants, e.g. to improve the color of tomato paste, sauces, desserts, jams and jellies, ice cream, sweets and cereals. Nowadays, the people are more health conscious and they want to know the pros and cons of the food which they are consuming. Instead of curing a disease, people prefer in preventing the initiation of any disease. Out of these, the beet roots being alkaline in nature can be utilized as economical medicinal foods necessary for good health (Anonymous, 2010).

In India, this highly nutritious and medicinal valued crop is sown during months of September-November in northern plains while it can be sown from first week of March to July only in hilly areas having cool weather. Beet root is mainly cultivated in Haryana, Uttar Pradesh, Himachal Pradesh, West Bengal and Maharashtra (Anonymous, 2010). The beet root is consumed directly as fresh in form of salad as well as juice and can also be processed into beetroot candy, beetroot juice powder etc. Due to the beneficial effects, the crop should be consumed regularly in one or the other form. Therefore, there is a need of proper processing and preservation techniques so as to get the maximum benefits from beetroot. The various methods available for preservation of fruits and vegetables are canning, freeze drying, and vacuum drying, hot air drying and osmotic dehydration. Among different methods of food dehydration a well-known process to achieve good-quality product is freeze drying, but it is an expensive method of food preservation relative to the value of that product. Therefore, there is a need for a simple and inexpensive alternate process, which has low capital investment and offers a way to save highly perishable products and make them available for the regions away from production zones. Osmotic dehydration is one of these methods (Shi and Le-Maguer, 2002).

Osmotic dehydration is a water removal technique applied to horticultural products such as fruits and vegetables for long storage period by reducing the water content and increasing soluble solid content by immersing in aqueous solutions of high osmotic pressure such as sugar and salts (Wanasundara *et al.*, 1996, Lee, W.C. *et al.*, 2006). The most commonly used osmotic agents are sucrose for fruits and sodium chloride for vegetables (Mudahar *et al.*, 1989). Osmotic dehydration as a pretreatment to many processes improves nutritional, sensorial and functional properties of food without changing its integrity. It is effective even at ambient temperature, so heat damage to texture, colour and flavour of food is minimized (Kaymark-erteki *et al.*, 2000). Beetroot mainly used in salad, juice preparations and in form of pickles. Considering the nutritional as well as medicinal importance of beetroot it should be the part of our diet. Moreover, concerning its availability throughout the year, beetroot can be processed into beet root candies. In present investigation an attempt has been made to prepare beetroot candy by using osmo-dehydration technique. The aim of this work was to optimize the osmotic dehydration process of beetroot candy as a function of sucrose concentration, osmotic temperature, and time, using response surface methodology with the purpose of achieving maximum possible water loss, solute gain, and sensory scores.

## MATERIAL AND METHODS

### EXPERIMENTAL DESIGN

For the optimization of osmotic dehydration by response surface methodology, the experiments were conducted according to Central Composite Rotatable Design (CCRD) with three variables at five levels. The independent variables were osmotic solution temperature, solute concentration, and duration of osmotic dehydration process. The low and high levels of the variables were 35 and 55°C for osmotic solution temperature, 30 and 60°Brix for sucrose solution concentration, 30 and 180 min for duration of dipping in osmotic solution, respectively (Ade-Omowaye *et al.*, 2002). The fruit to solution ratio was kept 1:4 (w / w) during all the experiments to minimize problems related to the management of the osmotic solutions like reconcentration, microbial contamination, reutilization, and discharge of the spent solution (Torreggiani & Bertolo, 2002). The relationship between levels of different coded and uncoded form of independent variables is given in Table 1. The experiments plan in coded and uncoded form of process variables along with results is as given in Table 2.

The experiments were conducted randomly to minimize the effects of unexplained variability in the observed responses because of external factors.

### PREPARATION OF SAMPLES

Fresh, well graded beetroot were collected from local market of Sirsa, Haryana on daily basis prior to each set of experiment. They were thoroughly washed with water to remove adhering soil and other debris. Then the fruits were cut into cubes (1cm ×1cm×1cm) using clean knife. No blanching was done prior to osmosis as it is detrimental to the osmotic dehydration process due to loss of semi-permeability of cell membranes (Ponting, 1973). Sugar, the osmotic agent, was purchased from a local market. The osmotic solution is prepared by mixing the sugar with proper amount of pure water.

### OSMOTIC DEHYDRATION

Beetroot cubes were weighed and then placed into stainless steel containers containing calculated volume of osmotic solution of different concentrations at pre set at desired temperature in hot water bath. The temperature of the osmotic solution was maintained by hot water bath agitating at the rate of 50 oscillations per min. In each of the experiments fresh osmotic syrup was used. All the experiments were conducted in triplicate and the average value was taken for calculations. Agitation was given during osmosis for reducing the mass transfer resistance at the surface of the fruit and for good mixing and close temperature control in osmotic medium (Chopra, 2001). The beetroot cubes were removed from the container at the specified time and rinsed with fresh water to remove the excess solute adhered to the surface. The osmotically dehydrated beetroot cubes were then spread on an absorbent paper to remove the free water present on the outer surface. Then out of the total osmotically dehydrated beetroot, about 15–20 g sample was put in the pre-weighed Petri dish for determination of dry matter by oven method. The remaining part of the sample was dried to final moisture content of 10% (wb) in hot air dryer at 60°C air temperature (Mundada *et al.*, 2010). The dried samples were packed in high density polyethylene (HDPE) (80 micron) bags and kept at ambient temperature for further quality analysis.

### STATISTICAL ANALYSIS AND OPTIMIZATION

The second order polynomial equation was fitted to the experimental data of each dependent variable as given below

$$Y_k = \beta_{k0} + \sum_{i=1}^n \beta_{ki}x_i + \sum_{i=1}^n \beta_{kii}x_i^2 + \sum_{i=1}^{n-1} \sum_{j=i+1}^n \beta_{kij}x_i x_j$$

Where  $Y_k$  = response variable ;  $Y_1$  = water loss (g) per 100 g fresh beetroot ;  $Y_2$  = solute gain (g) per 100 g fresh beetroot ;  $Y_3$  = sensory score ;  $x_i$  represent the coded independent variables ( $x_1$ = solution concentration ,  $x_2$  = process duration ,  $x_3$  = process temperature) ; where  $\beta_{k0}$  was the value of the fitted response at the centre point of

**Table 1: The level of different process variable in coded and uncoded form for osmotic dehydration**

Independent variables	Units	Symbols		Levels				
		Actual	Coded	-1.68	-1	0	1	1.68
Concentration	°Bx	X <sub>1</sub>	x <sub>1</sub>	19.77	30	45	60	70.23
Temperature	°C	X <sub>2</sub>	x <sub>2</sub>	28.18	35	45	55	61.82
Time	min	X <sub>3</sub>	x <sub>3</sub>	21.13	30	105	180	231.13

**Table 2: Central Composite Rotatable Design with experimental values of response variables**

Conc (°Bx)	Temp (°C)	Time (min)	Conc (°Bx)	Temp (°C)	Time (min)	Water loss (g per 100 g of fresh beetroot)	Solute gain(g per 100 g of fresh beetroot)	Sensory score
-1.000	-1.000	1.000	30.00	35.00	180.00	40.55	7.59	7.3
0.000	0.000	1.682	45.00	45.00	231.13	47.78	8.63	9.5
1.000	1.000	-1.000	60.00	55.00	30.00	45.09	7.02	8.0
1.000	-1.000	1.000	60.00	35.00	30.00	31.89	6.24	5.2
0.000	0.000	0.000	45.00	45.00	105.00	37.68	6.65	6.5
0.000	0.000	0.000	45.00	45.00	105.00	35.76	6.45	6.5
-1.000	1.000	1.000	30.00	55.00	180.00	46.99	7.98	8.5
1.682	0.000	0.000	70.23	45.00	105.00	42.03	7.16	7.5
-1.000	1.000	1.000	30.00	55.00	30.00	42.23	6.56	8.4
1.000	1.000	1.000	60.00	55.00	180.00	47.65	8.01	10
0.000	0.000	0.000	45.00	45.00	105.00	36.24	6.46	6.5
-1.682	0.000	0.000	19.77	45.00	105.00	35.12	6.22	7.0
0.000	0.000	0.000	45.00	45.00	105.00	36.78	6.45	6.5
1.000	1.000	1.000	60.00	35.00	180.00	45.67	7.78	8.0
0.000	0.000	0.000	45.00	45.00	105.00	37.14	6.28	6.5
0.000	0.000	-1.682	45.00	45.00	21.13	35.78	6.25	7.0
0.000	1.682	0.000	45.00	61.82	105.00	47.45	7.68	9.0
0.000	0.000	0.000	45.00	45.00	105.00	36.98	6.45	6.5
0.000	-1.682	0.000	45.00	28.18	105.00	30.78	6.09	5.5
-1.000	-1.000	-1.000	30.00	35.00	30.00	27.87	5.07	5.7

the design, i.e. point (0,0,0),  $\beta_{ki}$ ,  $\beta_{kii}$ , and  $\beta_{kij}$  were the linear, quadratic, and cross product regression coefficients, respectively. The analysis of the experimental data was carried out to observe the significant effect of various process variables on the various responses. The  $\beta$  coefficient is that the magnitude of these values helps to compare the relative contribution of each independent variable in the prediction of the dependent variable. Higher the positive value of  $\beta$  of a parameter, higher would be the effect of that parameter and vice versa.

The response surface and contour plots were generated for different interaction of any two independent variables, while holding the value of third variable as constant (at the central value). Such three dimensional surfaces could give accurate geometrical representation and provide useful information about the behavior of the system within the experimental design. The optimization of the osmotic dehydration process was aimed at finding the levels of independent variables viz. osmotic solution concentration, temperature, and process duration, which would give maximum possible water loss, solute gain, and sensory score. When the dehydrated product has to be rehydrated before final use like dehydrated vegetables, the optimization of osmotic dehydration process is always aimed at minimum solute gain. But, in present case, the dehydrated product has to be utilized directly without rehydration. Therefore, the optimization was aimed at the maximum solute gain during osmotic dehydration process. It will also help to make the product shelf stable at ambient conditions. Response surface methodology was applied to the experimental data using commercial statistical package, Design-Export version 8.01 (Trail version; Statease Inc., Minneapolis, MN, USA). The same software was used for the generation of response surface plots, superimposition of contour plots, and optimization of process variables.

### MATHEMATICAL CALCULATIONS

Water loss and solute gain during osmotic dehydration:-

The water loss and solute gain during osmotic dehydration were calculated by the equations given by Ozen et al., 2002; Singh et al., 2007;

Water loss (g) per 100 g of fresh beetroot

$$= \frac{(W_o - W_t)}{W_o} (S_t - S_o) * 100$$

Solute gain (g) per 100 g of fresh beetroot =

$$\frac{(S_t - S_o)}{W_o} * 100$$

Where  $W_o$  is the initial weight of beetroot (g),  $W_t$  is the weight of beetroot after osmotic dehydration for any time  $t$  (min),  $S_o$  is the initial weight of solids (dry matter) in the beetroot (g) and  $S_t$  is the weight of solids (dry matter) of beetroot after osmotic dehydration for time  $t$  (min).

### ESTIMATION OF DRY MATTER AND MOISTURE CONTENT

The samples were oven dried at  $103 \pm 2$  °C with lids open until a constant weight loss (AOAC, 1965) was achieved.

### SENSORY EVALUATION OF OSMOTIC DEHYDRATED BEETROOT

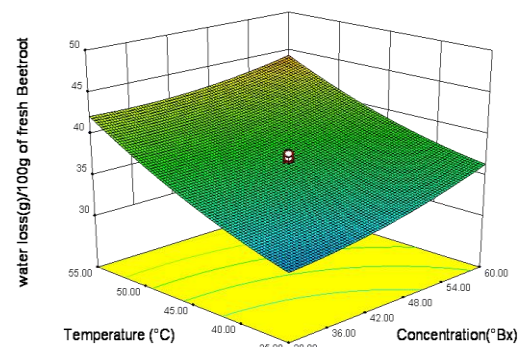
Organoleptic quality of osmotic dehydrated beetroot was determined with the help of a 10-member consumer panel, using a 9-point hedonic scale, following standard procedure. The aspects considered for osmotic dehydrated beetroot were colour, appearance, taste, flavour, and overall acceptability. The average scores of all the 10 panelists were computed for different characteristics.

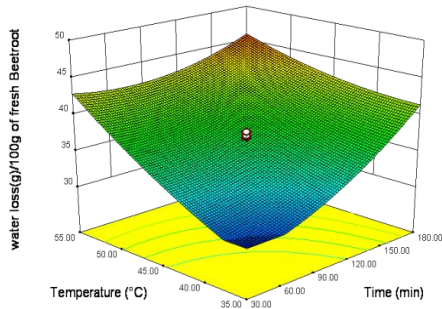
### RESULTS AND DISCUSSION

#### DIAGNOSTIC CHECKING OF FITTED MODEL AND SURFACE PLOTS FOR WATER LOSS

The results of second-order response surface model in the form of analysis of variance (ANOVA) are given in Tables 3, 4 and 5. The results indicated that the fitted quadratic models accounted for more than 90% of the variation in the experimental data, which were highly significant ( $R^2 > 0.90$ ). The magnitude of P values from Table 3 revealed that all linear and quadratic terms of process variables have significant effect at 5% level of significance ( $P < 0.05$ ) on water loss during osmotic dehydration. Further, interaction of 'temperature and time' has significant effect on water loss. The model F-value is 78.35, which implies the model is significant. The relative magnitude of  $\beta$  values (Table 3) indicates the maximum positive effect of osmotic solution temperature ( $\beta = 4.69$ ) followed by process duration ( $\beta = 3.95$ ) and concentration ( $\beta = 1.78$ ) on water loss. The quadratic and interaction terms of all the process parameters have least effect on water loss as compared to the linear terms of process variables.

(a)





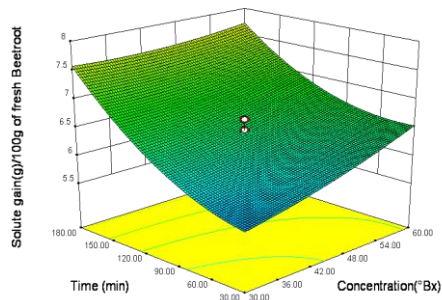
(b)

Figure-1: influence of process variables on water loss (a) sugar concentration and temperature for 105min of process duration (b) temperature and time at 45°BX, Figure 1a shows the increased water loss with increase in temperature and osmotic solution concentration. This might be because of the fact that the increase in temperature decreases the viscosity of the osmotic solution and thus reduces the external resistance to mass transfer at product surface to facilitate the outflow of water through cellular membrane (Panades et al., 2006). The increase in water loss with osmotic solution concentration is mainly because of the increase in the osmotic pressure gradient (Mundada et al., 2010, Azoubel & Murr, 2004). A similar variation in water loss with temperature and time has also been observed in Fig. 1b .which revealed that increase in water loss with time was more remarkable in high concentration than in low concentration.

#### DIAGNOSTIC CHECKING OF FITTED MODEL AND SURFACE PLOTS FOR SOLUTE GAIN

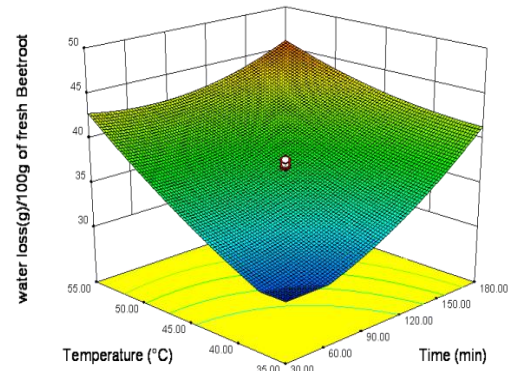
Table 4 indicates that all linear terms of process variables have significant effect ( $P < 0.05$ ) on solute gain. Further, quadratic effect of temperature and time and interaction of ‘temperature and time’ have significant effect on solute gain during osmotic dehydration ( $P < 0.05$ ). The model F-value 80.82 implies the model is significant. The magnitude  $\beta$  values indicates the maximum positive effect of process duration ( $\beta = 0.77$ ) followed by temperature ( $\beta = 0.41$ ) and osmotic solution concentration ( $\beta = 0.25$ ).

(a)

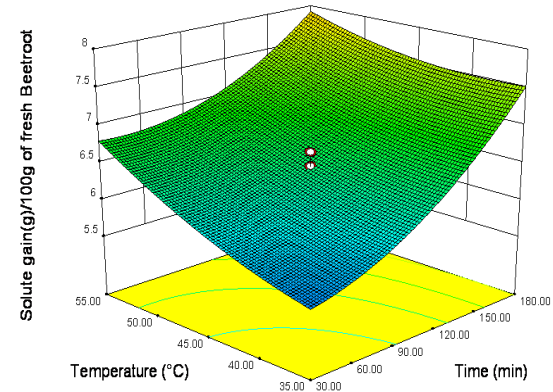


**Figure-2:** influence of process variables on solute gain (a) sugar concentration and time at 45°C of osmotic solution temperature (b) temperature and time at 45°BX of osmotic solution concentration. As shown in Fig. 2a, the solute gain increased with increase in osmotic solution concentration is mainly because of high concentration difference between the

beetroot and osmotic solution (Falade & Igbeka, 2007). Figure 2b revealed that solute gain enhanced with osmotic solution temperature might be because of decrease in viscosity of the osmotic solution resulting in high diffusion rates of solute (Mundada et al., 2010, Singh et al., 2007).The increase in water loss and solute gain with time, temperature, and concentration may also be because of agitation given during osmotic dehydration process which reduces the mass transfer resistance between the surface of beetroot and osmotic solution (Panagiotou et al., 1999).



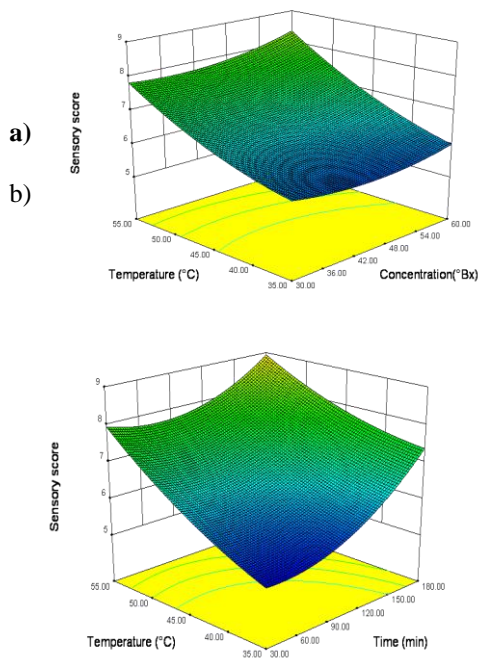
a)



b)

#### DIAGNOSTIC CHECKING OF FITTED MODEL AND SURFACE PLOTS FOR SENSORY SCORE

The magnitude of P-value from Table 5 indicates that all the linear and interaction terms have significant effect on the sensory score of the osmotic dehydrated beetroot ( $p < 0.05$ ) at 5% level of significance. The quadratic term of solution concentration and time has a non-significant effect on the sensory score, i.e. quality of osmotic dehydrated product, at the 5% level of significance ( $p > 0.05$ ). The magnitudes of  $\beta$  values indicate that osmotic solution temperature had a more pronounced effect ( $\beta = 1.07$ ) on sensory score than did process time ( $\beta = 0.78$ ) and osmotic solution concentration ( $\beta = 0.16$ ).



**Figure-3:** influence of process variables on sensory score (a) sugar concentration and temperature at 45°Bx of osmotic solution (b) temperature and time at 45°Bx of osmotic solution concentration. Figure 3 (a) shows the effect of sugar concentration and temperature on sensory score and the effect of temperature and time on sensory score shown in fig 3(b). This implies that the overall quality of the osmotic dehydrated beetroot product depends upon processing temperature, time and osmotic solution concentration.

**OPTIMIZATION OF OSMOTIC DEHYDRATION PROCESS**

To optimize the process conditions for osmotic dehydration process by numerical optimization technique, equal importance of ‘3’ was given to all the three process parameters (viz. osmotic solution of its concentration, process duration, and solution temperature). However, based on their relative contribution to quality of final product, the importance given to different responses was 4, 3, and 5 for water loss, solute gain, and sensory score, respectively as colour, flavor, firmness and appearance. Maximum importance was given to sensory score, because it included a number of parameters like colour, flavor, firmness and appearance. The optimum conditions for osmotic solution concentration, temperature and process duration were 60°Bx, 55°C and 180 min, respectively. The optimum processing conditions were experimentally verified and proven to be adequately reproducible with ±0.1% deviation.

**CONCLUSION**

Response surface methodology was effective in optimizing process parameters for the osmotic dehydration of beetroot candy in osmotic aqueous solutions of sucrose having concentrations in the range 30–60 Brix temperature 35–55 degree centigrade, and process duration 30–180 min.

The regression equations obtained in this study can be used for optimum conditions for desired responses within the range of conditions applied in this study. Graphical techniques, in connection with response surface methodology (RSM), aided in locating optimum operating conditions, which were experimentally verified and proven to be adequately reproducible. Optimum solution by numerical optimization obtained was 60°Bx osmotic solution concentration, 55°C osmotic solution temperature, and 180 min of process duration to get maximum possible water loss, solute gain, and sensory score. The optimum processing conditions were experimentally verified and proven to be adequately reproducible with ±0.1% deviation.

**Table3 -ANOVA Table showing the variables as a linear, quadratic and interaction terms on water loss and coefficients for the prediction models**

Source	df	β	Sum of squares	F-value	P-level
Model	9	36.73	678.09	78.35	<0.0001
Conc	1	1.78	43.17	44.89	< 0.0001
Time	1	3.95	213.21	221.72	< 0.0001
Temperature	1	4.69	300.07	312.04	< 0.0001
Conc ×Time	1	-0.14	0.15	0.16	0.7000*
Conc	1	-0.70	3.95	4.11	0.0702*
×Temp	1	-2.39	45.79	47.62	< 0.0001
Time×	1	0.85	10.36	10.78	0.0082
Temp	1	1.98	56.57	58.82	< 0.0001
Conc×Conc	1	1.04	15.56	16.18	0.0024
Time×Time	10	0.9860	9.62	3.16	0.1160*
Temp×Temp	5	0.9734	7.31		
Residual	5		2.31		
Lack of Fit					
Pure Error					
R <sup>2</sup>					
Adj R <sup>2</sup>					

\*Non significant at 5%level

**Table 4: ANOVA Table showing the variables as a linear, quadratic and interaction terms on solute gain and coefficients for the prediction models**

Source	Df	β	Sum of squares	F-value	P-level
Model	9	6.46	13.78	80.82	<
Conc	1	0.25	0.86	45.49	0.0001
Time	1	0.77	8.03	423.81	<
Temperature	1	0.41	2.27	119.63	0.0001
Conc ×Time	1	-0.18	0.25	13.11	<
Conc ×Temp	1	-0.11	0.095	4.99	0.0001
Time×Temp	1	-0.21	0.34	17.96	< .0001
Conc×Conc	1	0.081	0.095	4.99	0.0047
Time×Time	1	0.35	1.73	91.14	0.0495
Temp×Temp	1	0.15	0.32	17.10	0.0017
Residual	10	0.9864	0.19	1.76	0.0495
Lack of Fit	5	0.9742	0.12		<
Pure Error	5		0.069		0.0001
R <sup>2</sup>					0.0020
Adj R <sup>2</sup>					0.2756*

\*Non significant at 5%level

**Table 5: ANOVA Table showing the variables as a linear, quadratic and interaction terms on sensory score and coefficients for the prediction models**

Source	Df	$\beta$	Sum of squares	F-value	P-level
Model	9	6.50	32.83	426.58	< 0.0001
Conc	1	0.16	0.34	39.25	< 0.0001
Time	1	0.78	8.39	981.29	< 0.0001
Temperature	1	1.07	15.58	1822.03	< 0.0001
Conc $\times$ Time	1	0.39	1.20	140.49	< 0.0001
Conc $\times$ Temp	1	0.11	0.10	11.84	0.0963
Time $\times$ Temp	1	-0.29	0.66	77.34	0.0801
Conc $\times$ Conc	1	0.26	0.99	116.34	< 0.0001
Time $\times$ Time	1	0.62	5.47	640.15	< 0.0001
Temp $\times$ Temp	1	0.26	0.99	116.34	0.0024
Residual	10		0.086		
Lack of Fit	5		0.086		
Pure Error	5	0.9974	0.000		0.2333*
R <sup>2</sup>		0.9951			
Adj R <sup>2</sup>					

\*Non significant at 5% level

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