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Studies on the Addition of Hydrocolloids to Tomato-Carrot Juice Blend

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Abstract

The study determined the optimum combination of tomato-carrot juice and hydrocolloid blend. Tomato-carrot juice and hydrocolloids (carboxymethylcellulose and xanthan gum in ratio 2:1) were mixed together in various ratios to obtain the blends using Response Surface Methodology (RSM). Experimental runs were coded at levels of -1, 0 and+1 for the low, centre and high levels of the factorial points. RSM was applied to establish and exploit the relationship of response (pH, oBrix, Viscosity and Overall acceptability) to the design variables (tomato juice, carrot juice and hydrocolloids) using Box-Behnken design with seventeen treatments including five center points. PlotIT software package was used to generate four second-order polynomial equations for the responses. The measured and predicted values, significance of regression coefficient, analysis of variance, canonical analysis and contour plots for the responses were determined to check the adequacy of fit and used to obtain the optimum values for the variables. The results showed that the blend of 160 ml tomato juice, 90 ml carrot juice and 0.60 g hydrocolloid was optimized for pH; 130 ml tomato juice, 120 ml carrot juice and 0.44 g hydrocolloid for oBrix; 118 ml tomato juice, 132 ml carrot juice and 0.42 g hydrocolloid for viscosity and 131 ml tomato juice, 119 ml carrot juice and 0.60 g hydrocolloid for overall acceptability. Results further showed that the regression models were significant and had R2 values in the range of 0.838-0.997. The mathematical models for the response were considered adequate as the predicted values were close to the experimental values. The research concluded from the response surface methodology that the most acceptable quality and optimum combination of tomato-carrot juice blend was the combination of the variables designated as 160 ml tomato juice, 90 ml carrot juice and 0.60 g hydrocolloid.

Keywords: Tomato-carrot juice; Hydrocolloids; Response surface methodology

Introduction

Research on Vegetable juices attracts more attention due to the nutritional and phytochemical value of many vegetables [1]. While tomato is highly esteemed as a source of vitamin C and a significant source of vitamin A and B. It has about 20-25 mg ascorbic acid per 100 g [2]. Lycopene, a carotenoid found in tomato products, prevents oxidation of low density lipoprotein (LDL) cholesterol and reduces the risk of developing atherosclerosis and coronary heart disease. Carrot (Daucus carota) is one of the traditional root crops of northern Nigeria. It is very rich in carotene, which is a precursor of vitamin A and contains appreciable amounts of thiamine and riboflavin. Carrot has long been a component of tomato blends [3]. Tomato juice and blends based on tomato have long been popular and account for over 90 percent of the non-fruit juice trade [1].

Hydrocolloids are water-soluble, high molecular weight polysaccharides that find wide application in food industry because of their ability to improve the rheological and textural characteristics of food systems and often used as food additives for enhancing viscosity, creating gel-structures and lengthening the physical stability [4-8]. Examples of hydrocolloids are carboxymethylcellulose (CMC), guar gum, starch, xanthan gum, pectin, gelatine etc. [9]. CMC is a modified cellulose gum with excellent water retention properties and improves storage stability and juiciness in food products over an extended period of time [10]. Xanthan gum is an extracellular polysaccharide secreted by *Xanthomonas campestris* [11]. It is soluble in cold water and solutions exhibit highly pseudoplastic flow. Its viscosity has excellent stability over a wide pH and temperature range and the polysaccharide is resistant to enzymatic degradation. Xanthan gum is used in combination with other hydrocolloids [12].

Response Surface Methodology (RSM) is the statistical tool used extensively for optimizing processes in the tropical fruit juice production [13,14]. It usually uses an experimental design to fit a first- or –second-order polynomial by a least significance technique [15]. This graphical optimization technique has been used in other juice treatments such as mango juice [15]. One of the major problems encountered in the preparation of fruit juices is cloudiness due primarily to the presence of pectin [16].

The objective of the present work was to determine the optimum combination of tomato-carrot juice and hydrocolloid in juice blend using some physico-chemical parameters (pH, obrix, viscosity and overall acceptability) as indices of the blend quality and develop mathematical models of response for the physico-chemical parameters as a function of independent variables (tomato-carrot juice and hydrocolloids) using RSM.

Materials and Methods

Materials

Ripe and fresh tomato fruits (*Lycopersicon lycopersicum*) and fresh carrots (*Daucus carota L*) were purchased from the local market in Ile-Ife, Osun-state. The hydrocolloids (carboxyl methylcellulose, xanthan gum and guar gum), sodium benzoate, citric acid, aspartame and other chemicals were of analytical grade obtained from Captain Investment Limited, Surulere, Lagos.

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Experimental design

RSM was used to investigate the main effect of tomato-carrot juice blend. Based on preliminary results, desirable ranges of the independent variables selected for the investigation were 30-70% for tomato juice (X1), 30-70% for carrot juice (X2) and 0.4 g-0.6 g for hydrocolloids (XG and CMC) (X3). The main effect of each variable on pH, oBrix, viscosity and overall acceptability was estimated as the difference between both averages of measurement at the high (+1), low level (-1) and the mid-value (0) of the independent variables. The uncoded values for tomato-carrot juice at the high level were 70%, 30% for low value and 50% for mid-value for the independent variables while hydrocolloid was 0.6 g for high value, 0.4 g for low value and 0.5 g for the mid-value.

The Box-Behnken design was adopted with three factors at three levels as shown in Table 1, including five replicates at the center point, which was used for fitting a second-order response surface [17]. The complete design consisted of 17 experiments as indicated in Table 2.

The variance for each factor assessed was partitioned into linear and quadratic components and were represented using the second order polynomial function as follows:

Y=βo+β1 X1+β2 X2+β3 X3+β12 X1 X2 +β13 X1 X3+β23 X2 X3+β11 X12+β22 X22+β33 X32

where: Y is the predicted response, β0 model constant, X1, X2 and X3 are independent variables; β1, β2 and β3 are linear coefficients; β12, β13, β23 are the cross product coefficient and β11, β22 and β33 are the quadratic coefficients.

	Coded levels and real values			
Independent variables	Variable code	-1	0	+1
Tomato juice (%)	X_1	30	50	70
Carrot juice (%)	X_2	30	50	70
Hydrocolloids (XG&CMC)(g/250ml)	X_3	0.4	0.5	0.6

XG = Xanthan gum

CMC= Carboxymethylcellulose

Table 1: Coded level and real value for the Box-Behnken experiment.

	Independent coded variables		
Trials	X_1	X_2	X_3
1	-1	-1	0
2	1	-1	0
3	-1	1	0
4	1	1	0
5	-1	0	-1
6	1	0	-1
7	-1	0	1
8	1	0	1
9	0	-1	-1
10	0	1	-1
11	0	-1	1
12	0	1	1
13	0	0	0
14	0	0	0
15	0	0	0
16	0	0	0
17	0	0	0

Table 2: Box-Behnken design.

Analyses for RSM

The regression analysis, analysis of variance (ANOVA) and canonical analysis of the Plot IT (version 3.2) software package were used to fit the second order polynomial equations for the response variables. Lack-of-fit test of each model was calculated. R2 values, Standard Error (SE) estimate, significance F-test and the derived p values were the criteria used for eliminating a variable from the full regression equation. The fitted polynomial equations were expressed as surfaces and were performed on the fitted models. Coefficients for the linear and quadratic terms contour plots by using the Plot IT package.

Preparation of tomato-carrot juice blend

Tomato-carrot juice blend was produced as shown in Figure 3a. The fresh tomatoes were washed with portable water and sorted (blemished fruits were discarded). The carrot roots were washed thoroughly; both ends were removed and peeled with a sharp knife. The sorted fruits and the cleaned carrot roots were separately blanched in hot water containing (90°C for 3 mins) and the juice extracted using a Juice extractor (JM300, Salton R, Juiceman, UK). The juice for each fruit was filtered using muslin cloth.

Hydrocolloids (carboxyl methylcellulose and xanthan gum), aspartame and preservatives (sodium benzoate and citric acid) were dissolved in 50 ml of water using a mixer (YT518WB, Gazab, Kachan, Hong Kong). Tomato-carrot juice, hydrocolloids, aspartame and preservatives were filled into the glass bottles, corked and pasteurized at 85°C for 15 minutes. The pasteurized juice was allowed to cool.

Determination of pH

The pH of the samples was determined by using a pH meter with a glass electrode (Model H198130, Combo PH/EC/TDS, Hanna Instruments, Italy). Buffer 4.0 and 7.0 were used to standardize the electrode and it was cleaned and dipped into samples after stirring with a glass rod. Readings were taken in triplicates and mean values were calculated and recorded.

Degree brix (o Brix)

Degree brix (o Brix) was determined using Hand Refractometer (M 300002, Super Scientific, USA). The sample was placed on the prism of refractometer then the daylight plate was closed, and the scale where the boundary line intercepts was read as oBrix. All measurements were performed in triplicates and mean values obtained.

Viscosity

The viscosity of each sample was determined at room temperature by using a Brookfield digital viscometer (NDJ-85, Niryn Intelligent Company limited, Shanghai). A suitable spindle (spindle 2) and rotational speed (60 rpm) was selected for this study. This device gives the viscosity of Newtonian fluids directly (mPa.s) after calibrating with Newtonian oil [18].

Sensory analysis

Sensory evaluation was carried out on the samples for overall acceptability using 5-point Hedonic scale, where a score of 1 indicated poor sensory attribute and a score of 5 indicated excellent sensory attribute. A panel of 15 judges familiar with tomato-carrot juice were selected and presented with the coded samples. Panelists were instructed to rinse their mouth between samples test to avoid effects of residual flavours [19,20].

Results and Discussion

Table 3 shows the measured and the predicted values for the seventeen treatments performed for the responses. The regression coefficients for each response and the analysis of variance are shown in Table 4 and 5. The closer the value of R is to 1 the better is the correlation between the measured and predicted values [17].

Model Fitting and Response Surface Plotting

Four second-order polynomial equations were obtained. The four fitted models are shown as follows:

YpH=5.5+(-0.15) X1+0.1 X2+1.11ε10-16 X3+0.0275 X12+(-0.0725) X22+(-0.025) X1X2+0.0275 X32+0.025X1X3 -0.025 X2X3.

R2=0.928, X1= 2.41, X2=0.28, X3=0.97, Y=5.4.

YBrix=5.467+(-0.475) X1+0.3625X2+0.1375X3+0.1825X12+(-0.1425) X22+

(-0.1)X1X2+0.1075X32+(-0.25) X1X3+0.025 X2X3.

R2=0.838, X1= 1.13, X2=0.82, X3=-0.58, Y=5.37.

Yviscosity=73.32+19.3X1-1.8X2+32.9X3-30.79X12+17.11X22+

17.8X1X2+32.16X32+18.3X1X3+27.95X2X3.

R2=0.997, X1= 0.24, X2=0.62, X3=-0.85, Y=61.10.

Yoverall acceptability=2.566 -0.00625 X1-0.11X2+0.01125X3+0.197X12+0.2745 X22 -

0.4275X1X2+0.375X32 -0.115X1X3 -0.0725 X2X3.

R2=0.976, X1= 4.39, X2=3.76, X3=1.03, Y=2.35.

	Independent variables			Response variables							
Trials	X_1	X_2	X_3	pH		TSS (° Brix)		TSS (° Brix)		Overall acceptability	
				M	P	M	P	M	P	M	P
1	-1	-1	0	5.45	5.50	5.40	5.51	60.10	59.95	2.70	2.73
2	1	-1	0	5.26	5.25	4.40	4.76	24.80	20.75	3.61	3.57
3	-1	1	0	5.68	5.75	6.80	6.44	58.90	62.95	3.32	3.36
4	1	1	0	5.39	5.40	5.40	5.29	94.80	94.95	2.52	2.49
5	-1	0	-1	5.79	5.75	5.60	5.84	37.60	40.80	3.05	3.02
6	1	0	-1	5.42	5.40	5.40	5.39	43.80	42.80	3.20	3.23
7	-1	0	1	5.69	5.70	6.60	6.61	69.00	70.00	3.30	3.27
8	1	0	1	5.43	5.45	5.40	5.61	148.40	145.50	2.99	3.03
9	0	-1	-1	5.32	5.35	5.30	5.16	122.50	119.45	3.23	3.24
10	0	1	-1	5.58	5.60	5.50	4.95	59.10	59.95	3.17	3.16
11	0	-1	1	5.38	5.40	5.30	5.63	130.20	129.35	3.40	3.41
12	0	1	1	5.58	5.55	5.60	5.18	178.60	181.65	3.05	3.04
13	0	0	0	5.55	5.50	5.50	5.46	73.11	73.47	2.66	2.55
14	0	0	0	5.51	5.50	5.50	5.46	72.90	73.47	2.60	2.55
15	0	0	0	5.53	5.50	5.40	5.46	74.40	73.47	2.40	2.55
16	0	0	0	5.54	5.50	5.40	5.46	73.50	73.47	2.54	2.55
17	0	0	0	5.53	5.50	5.50	5.46	72.70	73.47	2.63	2.55

Table 3: Box-Behnken design and responses.

	Y_1 (pH)				Y_2 (° Brix)				Y_3 (Viscosity)				Y_4 (Overall Acceptability)			
Term	B	SE	t-value	P	B	SE	t-value	P	B	SE	t-value	P	B	SE	t-value	P
Constant	5.520				5.460				73.320				2.566			
X_1	-0.150	0.020	-7.400	0.000	-0.475	0.111	-4.280	0.004	19.300	1.174	16.440	0.000	-0.006	0.030	-0.210	0.843
X_2	-0.100	0.020	4.930	0.001	0.363	0.111	3.270	0.014	-1.800	1.174	-1.530	0.169	-0.110	0.030	-3.610	0.009
X_3	1.11E-16	0.020	0.000	1.000	0.138	0.111	1.240	0.255	32.900	1.174	28.020	0.000	0.011	0.030	0.370	0.773
$X_1.X_1$	0.028	0.028	0.980	0.358	0.183	0.153	1.190	0.272	-30.786	1.618	-19.020	0.000	0.197	0.042	4.690	0.002
$X_2.X_2$	-0.073	0.028	-2.600	0.036	0.143	0.153	-0.930	0.383	17.114	1.618	10.580	0.000	0.275	0.042	6.540	0.000
$X_3.X_3$	0.028	0.028	0.980	0.358	0.108	0.153	0.700	0.505	32.164	1.618	19.880	0.000	0.372	0.042	8.870	0.000
$X_1.X_2$	-0.428	0.043	-0.870	0.412	-0.100	0.157	-0.640	0.542	17.800	1.660	10.720	0.000	-0.428	0.043	-9.930	0.000
$X_1.X_3$	0.025	0.029	0.870	0.412	-0.250	0.157	-1.590	0.155	18.300	1.660	11.020	0.000	-0.115	0.043	-2.670	0.032
$X_2.X_3$	-0.025	0.029	-8.870	0.412	0.025	0.157	0.160	0.878	27.950	1.660	16.830	0.000	-0.073	0.043	-1.680	0.136

b, regression coefficient estimate; *SE,* standard error for regression coefficient.

Table 4: Regression coefficients of full second-order equation of physico-chemical and overall acceptability of tomato-carrot juice blend with the addition of hydrocolloids.

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Sources of Variation	d.f	Sum of Squares	Mean Squares	F	Sig
Y_{pH}					
Regression	9	0.295	0.033		
Residual	7	0.023	0.003	11.000	0.003
Lack of fit	3	0.015	0.005		
Pure error	4	0.008	0.002	2.500	
Total	16	0.318			
Y_{Brix}					
Regression	9	3.566	0.396		
Residual	7	0.689	0.099	4.000	0.040
Lack of fit	3	0.677	0.226		
Pure error	4	0.012	0.003	75.33	
Total	16	4.255			
$Y_{viscosity}$					
Regression	9	26578.070	2953.119		
Residual	7	77.184	11.026	267.830	0.000
Lack of fit	3	75.376	25.125		
Pure error	4	1.808	0.452		
Total	16	26655.254			
$Y_{Overall\ acceptability}$					
Regression	9	2.081	0.231		
Residual	7	0.052	0.007	33.000	0.000
Lack of fit	3	0.009	0.003		
Pure error	4	0.042	0.011	0.272	
Total	16	2.133			

*P < 0.05 (9,7) = 3.73; **P < 0.01 (9,7) = 6.84

*P < 0.05 (3,4) = 6.59; ** P < 0.01 (3,4) = 16.69.

Table 5: Analysis of variance for the responses.

	X_1	X_2	X_3	Eigen values	Eigenvectors		
pH	0.055	-0.025	-0.025	-0.552	0.087	0.169	-0.707
	-0.055	-0.055	-0.559	-0.559	-0.239	0.971	0.000
	-0.025	0.055	0.062	0.062	0.687	0.169	0.707
°Brix							
	0.365	-0.100	-0.025	0.472	0.929	-0.090	0.357
	-0.100	-0.285	0.025	-0.291	-0.134	0.820	0.556
	-0.250	0.025	-0.215	-0.316	-0.343	-0.564	0.750
Viscosity							
	-61.580	17.800	18.300	-66.187	0.984	-0.065	0.167
	17.800	34.220	27.500	17.915	-0.144	0.843	0.518
	18.300	27.950	64.320	85.231	-0.107	-0.554	0.839
Overall acceptability							
	0.394	-0.428	-0.115	0.0120	0.757	-0.650	-0.057
	-0.428	0.549	-0.073	0.908	0.628	-0.749	-0.212
	-0.115	-0.073	0.744	0.767	0.181	0.125	0.975

Table 6: Canonical analysis of response surface.

The high values of the coefficient of determination (R2) indicate that the model p-values fit the experimental data well [15]. Table 4 illustrates the significance of regression for the responses (using Student's t-test and p-value) signify according to Ying et al. [15].

For pH, it can be observed that the variable with the largest effect was the linear term of tomato juice (X1), followed by carrot juice (X2) and the quadratic of the carrot juice (X22). The factor t-test value (-7.400) and p-value (0.000) correspond to X1, while the t-test values of 4.930 and -2.600 for X2 and X22 with the corresponding p-values of 0.001 and 0.036 which are still significant. The total coefficient of determination R2 (92.8%) implies that the sample variation is attributable to the independent variables. The optimum value for the pH of the blend was 5.4 while the corresponding optimum value for tomato juice, carrot juice and hydrocolloid were 160 ml, 90 ml and 0.60 g, respectively.

The significance of regression coefficient for o Brix is illustrated in Table 4. It can be seen that the variable with the largest effect was the linear term of tomato juice (X1), followed by the linear term of carrot juice (X2).The factor t-test value (-4.280) is small but p-value (0.004) for X1 is significant. The total coefficient of determination R2 (83.8%) implies that the sample variation is attributable to the independent variables. The optimum value for the oBrix of the blend was 5.37 while the corresponding optimum values of independent variables for tomato juice, carrot juice and hydrocolloid were 130 ml, 120 ml and 0.44 g.

In considering viscosity, all the variables had good effect except X2, although some of the variables had low t-values as seen in Table 4. The variables with the largest effect were the linear term of hydrocolloids (X3), the quadratic term of hydrocolloids (X3. X3.) and the interaction term for carrot juice and hydrocolloids (X2.X3).The t-test values (28.02,19.88 and 16.83) and p-values (0.000) for X3, X3. X3 and X2.X3. The total coefficient of determination R2 (99.7%) implies that the sample variation is attributable to the independent variables. The optimum value for the viscosity of the blend was 61.1 while the corresponding optimum value for the independent variables, tomato juice, carrot juice and hydrocolloid were 118 ml, 132 ml and 0.42 g.

For overall acceptability, all the variables had good effect except for the linear terms X1, X3 and interaction term X2.X3, although some of the variables had low t-values. The variables with the largest effect were the quadratic terms X3. X3, X2. X2 and the interaction term (X1. X2). The factor t-test value (8.87, 6.54 and -9.93) and p-values (0.000), though the t-value for X1. X2 was low. The total determination coefficient R2 (97.6%) implies that the sample variation is attributable to the independent variables. The optimum value for the overall acceptability of the blend was 2.4 while the corresponding optimum value for tomato juice, carrot juice and hydrocolloid were 131 ml, 119 ml and 0.60 g.

The statistical significance of the polynomial model was checked by F-test (ANOVA) as shown in Table 5. The values of lack of fit test for pH and Overall acceptability were significant while that of viscosity and oBrix were not significant due to high variations between readings for viscosity and the very low value of pure error for °Brix.

Contour plot for the effect of pH, °Brix., viscosity and overall acceptability on tomato-carrot juice blend

Contour plots provide a visual insight into the direction to follow in pursuit of the optimum. Canonical analysis (Table 6) determines the nature of stationary of the second order model. Eigenvalues are special set of scalars associated with linear system of equations. It is paired with corresponding eigenvectors.

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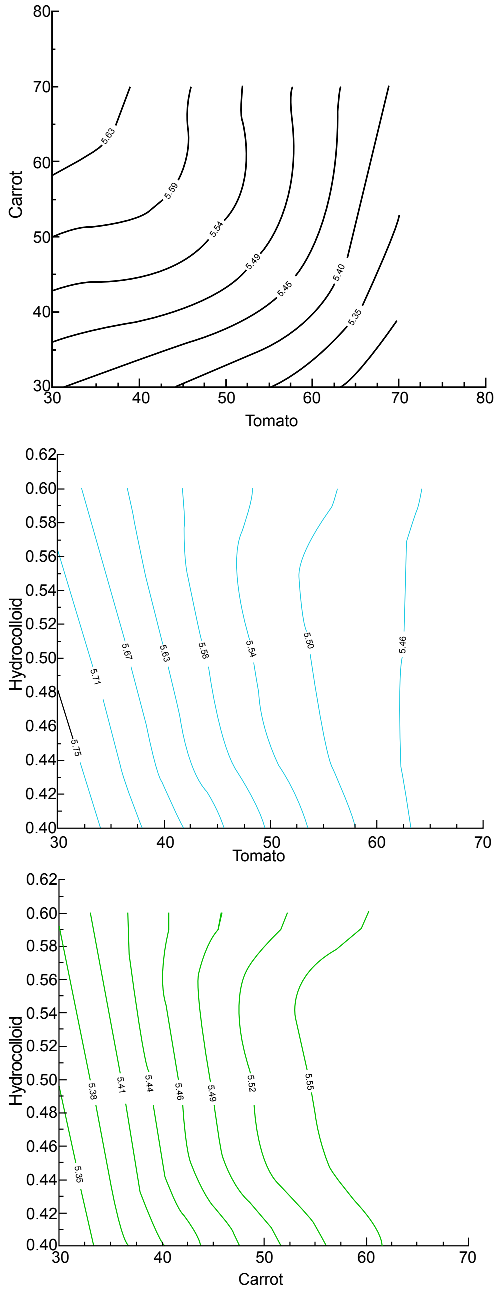

Figure 1: Contour Plot for the effect of pH on tomato-carrot juice blend.
a) Surface plot of tomato-carrot juice.
(b) Surface plot of tomato juice and hydrocolloid.
(c) Surface plot of carrot juice and hydrocolloid.

In Figure 1a, increase in tomato juice (X1) from 35% to 63% resulted in a decrease in pH from 5.53-5.35 while an increase in carrot juice (X2) from 30% - 60% led to an increase in the pH (5.45-5.53) of the juice at constant hydrocolloid (X3). In Figure 1b and 1c, increase or decrease in hydrocolloid had little effect on the juice at constant X1 and X2. The canonical analysis indicated negative coefficients for X1 and X2 while X3 had positive effect. The stationary point was a saddle point (75% negative values) and the variables were located within the experimental range. As X1 increased, a decrease in the oBrix level was observed while the oBrix level of the juice blend increased as X2 increased (Figure 2a). A decrease in X1 and increase in X3, increased the oBrix level of the blend while an increase in X2 and X3, increased the oBrix level of the juice blend (Figures 2b and 2c).

The Eigen values obtained from the canonical analysis were 75% negative and 25% positive, thus indicating that the stationary point is a saddle point. It is evident from Figures 3a-3c that increase in X1 increased the viscosity of the juice blend and reverse was the case for X2 while there was positive effect for X1 and X2 as X3 increased. The canonical analysis indicated positive coefficients for X2 and X3 while X1 was negative effect. The stationary point is a saddle point (75% positive values) and the variables were located within the experimental range. Figures 4a-4c showed that the increase in X1, X2 and X3 had positive effect on the overall acceptability of the juice blend. The

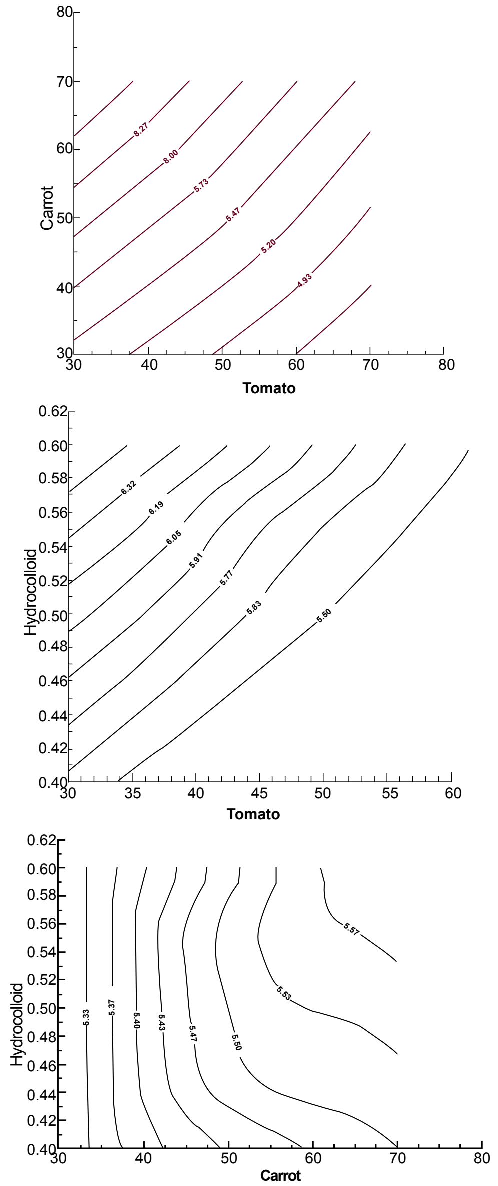

Figure 2: Contour Plot for the effect of ºbrix on tomato-carrot juice blend.
(a) Surface plot of tomato-carrot juice.
(b) Surface plot of tomato juice and hydrocolloid.
(c) Surface plot of carrot juice and hydrocolloid.

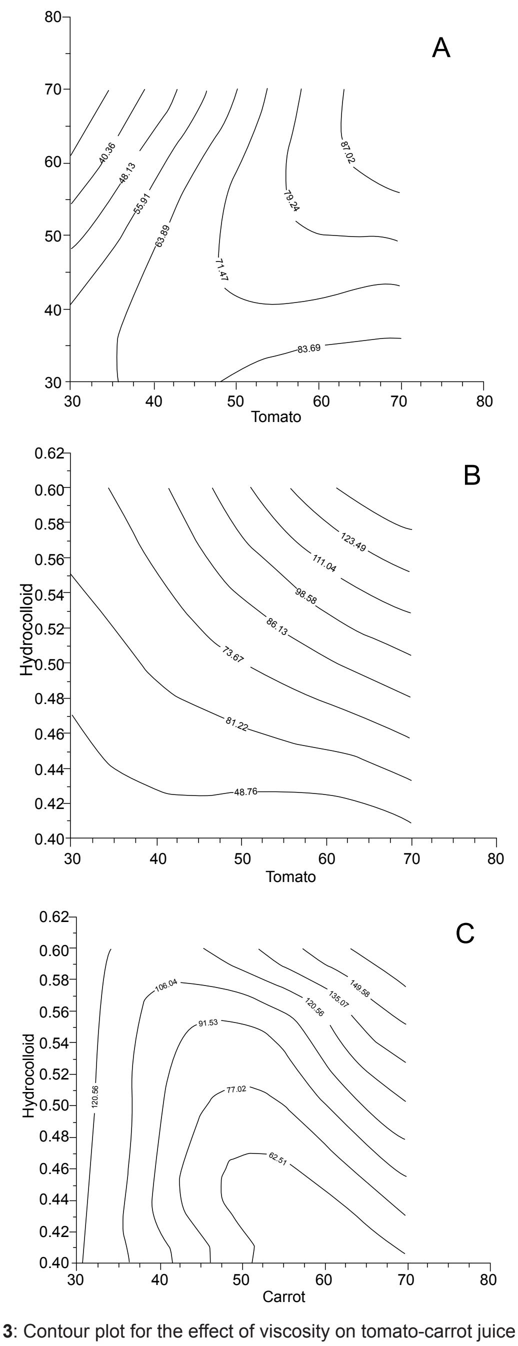

Figure 3: Contour plot for the effect of viscosity on tomato-carrot juice blend.
(a) Surface plot of Tomato-carrot juice.
(b) Surface plot of Tomato juice and hydrocolloid.
(c) Surface plot of Carrot juice and hydrocolloid.

canonical analysis indicated positive effect for X1 and X2 and X3. The stationary point was a minimum and the variables were located within the experimental range.

Sensory evaluation

The sensory evaluation revealed the acceptability of the product in taste, flavour, colour and level of sedimentation. Presence of hydrocolloids did not depress the taste of the juice.

Conclusion

The use of RSM has revealed the optimum concentration of hydrocolloid to be added to tomato-carrot juice blend. The RSM results in combination with sensory evaluation result revealed that the combination of tomato and carrot juice in the range 30%-70% with 0.6 g hydrocolloids gave acceptable juice qualities. Four second-order polynomial models were derived by the combined use of the RSM approach and CCRD using physico-chemical parameters such as pH, oBrix, viscosity and overall acceptability as indices of the blend quality. In the pH model and oBrix model, the variable with the highest effect

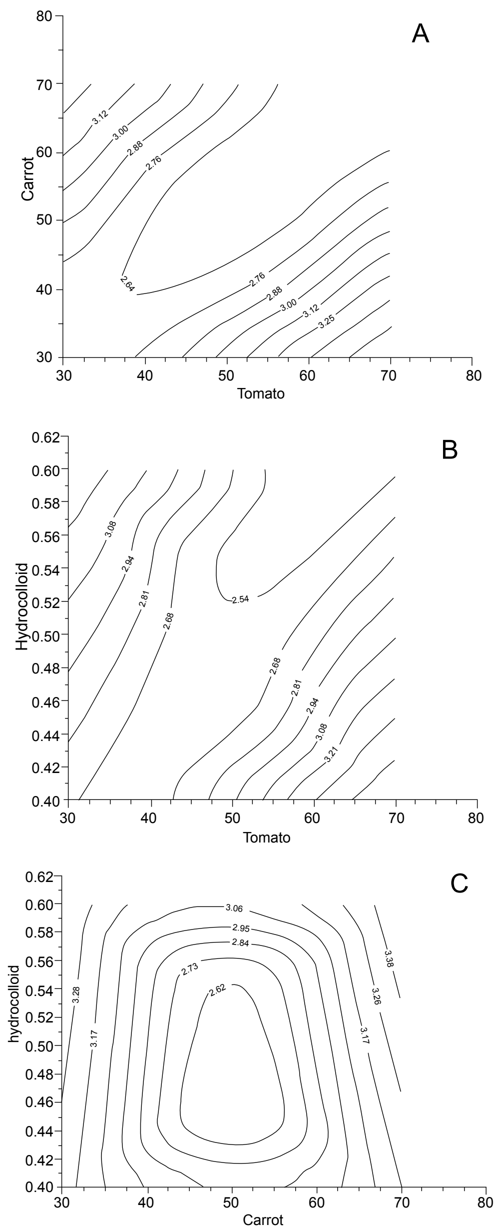

Figure 4: Contour Plot for the effect of acceptability on tomato-carrot juice blend.
(a) Surface plot of Tomato-carrot juice.
(b) Surface plot of Tomato juice and hydrocolloid.
(c) Surface plot of Carrot juice and hydrocolloid.

was the linear term of tomato juice (t-value -7.400, p-value 0.000) and (t-value 4.280, p-value 0.004) respectively. For the viscosity term of the model, the linear term of hydrocolloids had the largest effect (t-value 28.02, p-value 0.000) and in the overall acceptability model, the quadratic term of hydrocolloid (t-value 8.87, p-value 0.00) had the largest effect. The four models had high values (0.838-0.997) of the coefficient of determination (R2), which indicated that the models did fit the experimental data well. The validity tests proved that the adequacy of the above models was satisfactory. The optimum combination for tomato-carrot juice blend and hydrocolloid was 160 ml tomato juice, 90 ml carrot juice, 0.60 g hydrocolloid with pH of 5.4, obrix of 5.20 and viscosity of 169.50 mPa.s.

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