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Formulation development and physicochemical characterisation of model beverage emulsions stabilised by guar gum and carboxymethyl cellulose

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RESEARCH ARTICLE

Abstract

The potential of two functional hydrocolloids of guar gum (GG, 0.3-0.5%) and carboxymethyl cellulose (CMC, 0.2-0.4%) to design an optimum and stable formulation for corn oil-water emulsions was studied using response surface methodology. The physicochemical attributes including pH, turbidity, size index, apparent viscosity and physical stability as a function of two independent variables were investigated. The results showed that empirical second-order polynomial models with high R^2 (>0.94) can be used to fit the experimental data. The best formulation (0.5% GG and 0.2% CMC) resulted in pH of 4.02, turbidity of 0.706, size index of 0.900, apparent viscosity of 996 cP and stability of 93.5%, and the models ensured a good fitting of the observed data. A highly positive correlation was found between viscosity and turbidity values (P<0.01; r^2=0.977).

Keywords: cellulose gum, emulsion size index, emulsion stability, guar gum, rheology

1. Introduction

A food emulsion is consisting of a suspension with small droplets of oil in an aqueous medium (O/W, e.g. milk, beverage) or vice versa (W/O, e.g. butter, margarine) (McClements, 1999). These emulsion systems rapidly or slowly separate into two immiscible phases during a time period because they thermodynamically are unstable. Many biopolymers were used to create stable emulsion types due to the resistance enhancement of emulsion droplet against physical changes through the balance between attractive (Van der Waals and osmotic) and repulsive (electrostatic, steric and hydration) forces between these droplets (Gharibzahedi *et al.*, 2013a).

Polysaccharides as emulsifiers or stabilisers were applied to prepare food emulsions because of two possible effects including the viscosity increase of the dispersing phase and the surface adsorption (Gharibzahedi *et al.*, 2013b). Carboxymethyl cellulose (CMC) or cellulose gum is the most utilised cellulose ether in the emulsion systems. Contrary to pure cellulose, CMC is highly water-soluble, but retains the biodegradability of its original natural macromolecule. This component is used as a thickener, binder, stabiliser and suspending and water-retaining agent in the pharmaceutical, food and bio-industries (Pilizota *et al.*, 1996). Guar gum (GG) as a practical polysaccharide is obtained from the seed endosperm of *Cyamopsis tetragonolobus*. GG is highly viscous at low concentrations and has useful thickening, stabilising and water-binding attributes (Casas and Garcia-Ochoa, 1999). Based on these functional characteristics, it is applied to improve mixing tolerance, prolong the shelf-life of the end product through its moisture retention property and prevent syneresis phenomenon in frozen food products (Benichou *et al.*, 2002).

Optimisation of experimental parameters is usually evaluated by systematic variation of one parameter while the others are maintained constant. However, this approach is unable to predict the best conditions of the process (Gharibzahedi *et al.*, 2013a). Response surface methodology (RSM) is a collection of mathematical and statistical techniques based on the fit of a polynomial equation to the experimental data, which must describe the behaviour of a data set with the objective of making

statistical previsions. It can be well applied by reducing the number of experimental trials when a response or a set of responses of interest is influenced by several variables (Gharibzahedi *et al.*, 2012a).

Mirhosseini *et al.* (2008a) evaluated the effect of pectin (1.5, 3.0 and 4.5%) and CMC (0.1, 0.3 and 0.5%) on physical stability, turbidity loss rate, cloudiness and flavour release of orange beverage emulsions during six months storage. They found that the CMC-based beverage emulsions exhibited a significantly higher degree of cloudiness as compared to the ones containing pectin during the storage. A decrease in the release content of aldehyde volatile compounds also appeared to be in parallel with the decrease in emulsion stability and turbidity. Rahmati *et al.* (2015) by optimising influence of GG and xanthan gum (XG) on the characteristics of reduced fat mayonnaise using RSM found that the more physical stability with smaller size of droplets were observed in samples with higher concentrations of used hydrocolloids. Gharibzahedi *et al.* (2013c) studied the effects of gum arabic (GA) and XG on the stability, size index, turbidity and pH of walnut oil (5.09% w/w)/ water emulsion using RSM. They reported that the highest stability and turbidity, and the lowest size index and pH can be obtained with 10.0%, w/w GA and 0.13%, w/w XG. These researchers also optimised the various properties of walnut oil/water emulsions formulated with GA such as turbidity loss rate, density, size index, viscosity and stability (Gharibzahedi *et al.*, 2012a,b). RSM generally showed that an increase of GA content in the studied range and initial concentration of walnut oil were associated with high emulsion stability and minimum droplet size.

There is little information in the field of formulation optimisation of O/W food-grade emulsions containing GG and CMC according to their physicochemical characteristics such as physical stability, viscosity, pH, turbidity and droplet size using RSM. The present study was thus conducted to characterise and optimise the main and interaction effects of critical structural components (GG and CMC) on the physicochemical properties of a model beverage emulsion based on corn oil (CO) using RSM. The main objective of this research was to determine optimum levels of independent variables leading to: (1) the highest emulsion stability, viscosity and turbidity; and also (2) the least droplet size and pH.

2. Materials and methods

Chemicals and materials

GG and CMC were purchased from Merck Chemical Co. (Darmstadt, Germany). Sodium azide (SA) as a preservative agent was obtained from Sigma-Aldrich Chemical Co. (St. Louis, MO, USA). CO was provided by Oila Food Co. (Tehran, Iran).

Emulsion preparation

The method of Gharibzahedi *et al.* (2013a) with minor modification was used for preparation of O/W emulsions. These emulsions were manufactured based on the following formula: CMC (0.2-0.4%, w/w), GG (0.3-0.5%, w/w), CO (12%, w/w), SA (0.05%, w/w) and deionised water. In first, SA (0.15%, w/w) was dispersed in deionised water (60±2 °C) using a heater-stirrer (Stuart CB162; Bibby Scientific Ltd., Stone, UK) in order to prepare the water phase. Then, GG and CMC were gradually added to this solution and kept at room temperature (23±1 °C) overnight to facilitate hydration. After the full hydration, CO (7%, w/w) was slowly added to the water phase during continuous mixing to prepare an initial coarse emulsion. Consequently, the obtained coarse emulsions to produce the fine-disperse emulsions with small average droplet size and narrow particle-size distribution were homogenised using a Ultra Turrax (T25 Digital; IKA, Staufen, Germany) in 12,500 rpm for 2.5 min and then sonicated for 4 min at 40 °C with 30% of nominal power using an 35 kHz ultrasonic homogeniser (TI-H-10, transonic; Elma, Singen, Germany).

pH analysis

The pH value of emulsions was measured using a glass pH electrode (MP230; Mettler Toldo, Greifensee, Switzerland).

Turbidity determination

The turbidity was assayed based on the method described by Taherian *et al.* (2006). The turbidity of diluted emulsions (1:1000) was triptically evaluated based on the absorbance at 660 nm by an UV-visible spectrophotometer (DR/4000U-HACH; Hach, Loveland, CO, USA).

Size index measurement

The measurement of this index was done by an UV-visible spectrophotometer (DR/4000U-HACH; Hach) using the method applied by Gharibzahedi *et al.* (2013b). Briefly, the model emulsions were diluted with deionised water at a ratio of 1:100. The absorbance readings were triptically measured at wavelengths of 800 and 400 nm. The size index was defined as the ratio of absorbance at 800 nm over 400 nm. Deionised water as a blank for absorbance was used.

Evaluation of apparent viscosity

The method of Mohagheghi *et al.* (2011) was applied to determine the viscosity of model emulsions at 25 °C using a steady-stress viscometer equipped with a ULA spindle (Brookfield DV-II+ programmable viscometer; Brookfield, Middleboro, MA, USA). About 16 ml of each formulation at different rotational speeds depending on the torque values was analysed. Preliminary experiments showed that

the appreciate value of torque in order to obtain reliable results was between 10 and 100% of the measuring range. Analyses were immediately carried out in triplicate after the production of samples.

Physical stability assay

Creaming values for the emulsions were calculated from the ratio of cream volume over total volume of emulsion samples up on standing. Duplicate samples containing 15 ml of prepared emulsion were stored in 25 ml-tube for 14 day at 25±1 °C. The results of creaming index were expressed as percentage of the total height of the emulsions in the tube (Gharibzahedi *et al.* 2013c).

Experimental design and statistical analysis

The effect of GG (0.3-0.5%, x_1), and CMC (0.2-0.4%, x_2) concentrations on the pH (Y_1), turbidity (Y_2), size index (Y_3), apparent viscosity (Y_4), and physical stability (Y_5) was evaluated using RSM method based on central composite rotatable design. Fourteen treatments were conducted based on the design, each at five coded levels: -1.41, -1, 0, 1 and 1.41 (Table 1). Experiments were randomised in order to minimise the effects of unexplained variability in the observed responses due to extraneous factors. Our preliminary studies showed that the selected levels for GG and CMC in the emulsion formulations can lead to the desirable amounts of physicochemical characteristics.

For statistical calculations, the relation between the coded values and actual values are described by Equation 1:

$$x_i = \frac{X_i - X_0}{\Delta X} \quad (1)$$

Where x_i = the coded value of the variable; X_i = the actual value of the variable; X_0 = the actual value of X_i at the centre point; and ΔX = the step change value of the variables.

The results of the experimental design were fitted with a second-order polynomial equation by a multiple regression technique. The quadratic equation to predict the optimal point was explained as follows:

$$y = b_0 + b_1x_1 + b_2x_2 + b_{12}x_1x_2 + b_{11}x_1^2 + b_{22}x_2^2 \quad (2)$$

The coefficients of the polynomial were represented by b_0 (constant term), b_1 and b_2 (linear effects), b_{11} and b_{22} (quadratic effects), and b_{12} (interaction effects).

The quality of the fit of polynomial model was expressed by the coefficient of determination (R^2) and the adjusted R^2 (R^2_{adj}), coefficient of variation (CV), the prediction error sum of squares (PRESS) and adequate precision (ADP) as previously stated by Gharibzahedi *et al.* (2012c, 2013c).

For regression analysis of the data obtained and to estimate the coefficient of the regression equation, a statistical program package Design Expert (trial version 7.1.6; Stat-Ease Inc., Minneapolis, MN, USA) was used. The equations

Table 1. Response surface methodology matrix and experimental data obtained for the response variables.

Run	Independent variables		Response variable				
	GG[1] (%, w/w; x_1)	CMC[2] (%, w/w; x_2)	pH (Y_1)	Turbidity (Y_2)	Size index (Y_3)	Viscosity (cP; Y_4)	Stability (%; Y_5)
1	0.30	0.20	3.98±0.02	0.645±0.023	1.143±0.019	442±11	73.7±1.1
2	0.50	0.20	4.03±0.04	0.716±0.012	0.912±0.025	983±21	95.2±2.2
3	0.30	0.40	4.08±0.06	0.722±0.016	0.832±0.003	626±13	78.2±2.3
4	0.50	0.40	4.51±0.00	0.080±0.003	1.090±0.009	1,071±8	92.6±3.0
5	0.26	0.30	3.90±0.01	0.602±0.002	1.010±0.011	378±17	73.1±2.2
6	0.54	0.30	4.20±0.08	0.712±0.026	0.981±0.028	1,094±12	94.8±4.6
7	0.40	0.16	4.03±0.06	0.687±0.017	1.023±0.031	760±23	81.9±1.0
8	0.40	0.44	4.43±0.02	0.799±0.007	0.921±0.036	916±32	87.1±0.7
9	0.40	0.30	4.06±0.05	0.789±0.019	0.902±0.021	549±1	78.2±0.2
10	0.40	0.30	4.06±0.04	0.745±0.045	0.895±0.008	603±4	77.2±0.6
11	0.40	0.30	4.14±0.06	0.755±0.002	0.912±0.061	588±6	78.1±1.3
12	0.40	0.30	4.08±0.02	0.761±0.009	0.924±0.026	612±12	80.3±2.6
13	0.40	0.30	3.99±0.08	0.750±0.025	0.918±0.045	565±22	79.6±2.4
14	0.40	0.30	3.95±0.09	0.749±0.010	0.935±0.075	577±15	79.3±5.0

[1] GG = guar gum.
[2] CMC = carboxymethyl cellulose.

were validated by the statistical tests called the analysis of variance (ANOVA) analysis. The significance of each term in the equation is to estimate the goodness of fit in each case. Response surfaces were drawn to determine the individual and interactive effects of test variable on percentage extraction of chromium. The optimal values of the test variables were first obtained in coded units and then converted to the un-coded units. Correlation analysis was also performed employing Pearson's test using SPSS 13 (SPSS Inc., Chicago, IL, USA) software.

3. Results and discussion

Model fitting

ANOVA confirmed that the polynomial models were highly significant for all response variables (Table 2). The very low probability ($P \leq 0.0001$) values for all regression models were indicated non-significance lack of fit ($P > 0.05$). Generally, a high R^2 shows that the variation was accounted and that the data fitted satisfactorily to the second-order polynomial equation. However, a large value of R^2 does not always imply that the regression model is a good one. Adjusted R^2 is a modification of R^2 that adjusts for the number of explanatory terms in a model. Unlike R^2, the R^2_{adj} increases only if the new term improves the model more than would be expected by chance (Ghasemlou *et al.*, 2012). Table 2 shows the all studied responses had the high R^2 (0.940-0.995), and R^2_{adj} (0.903-0.992) values. The CV is the ratio of the standard error of estimate to the mean value of observed response expressed as a percentage. It is a measure of reproducibility of the models. As a general rule, a model can be considered to reasonably reproducible if its CV is not greater than 10% (Gharibzahedi *et al.*, 2014). Results showed that the fitted models had low CV values (1.32-2.29%). The PRESS (0.007-75.77) values suggest for the adequacy of the fitted quadratic models for predictive applications (Table 2). The ADP value measured the signal to noise ratio. A ratio of greater than 4 is normally desirable (Gharibzahedi *et al.*, 2013c).The suitable ADP values (16.99-53.28) indicated that these models could be used to navigate the design space (Table 3). All these statistical parameters show the reliability of the constructed models. Figure 1 reveals the comparison between the actual response values obtained from experimental data and the predicted response values based on the polynomial regression models and proves that the models cover the experimental range of studies sufficiently.

Acidity optimisation

The effects of GG and CMC were significant ($P < 0.0001$) on the pH-values of model emulsions. Quadratic effect of CMC content and the mutual interaction between GG and CMC concentrations were found to be significant ($P < 0.05$). The linear effect of CMC followed by main effect of GG had the most significant ($P < 0.05$) effect on the pH-value of emulsions (Table 2). Figure 2A also shows that an increase in the concentrations of GG and CMC due to high number of carboxyl groups present in the solution can increase the pH-value (Casas and Garcia-Ochoa, 1999). The individual optimisation clearly reveals that an emulsion formulated with 0.30% (w/w) GG, and 0.27% (w/w) CMC led to the lowest response for pH value (Y_1=3.93). The least pH value was considered as an optimum pH region for the studied emulsions because the reduction of pH causes to maintain the functional efficiency of preservatives in the most food emulsions such as beverage emulsions. Mirhosseini *et al.* (2008b) and Gharibzahedi *et al.* (2013b) found that the stability of beverage emulsions can significantly guarantee by decreasing their pH amount. These researchers showed that GA can considerably reduce pH value of the prepared emulsions (Mirhosseini *et al.*, 2008b; Gharibzahedi *et al.*, 2013b)

Turbidity optimisation

The linear effects of two independent variables were significant on the turbidity value ($P < 0.001$; $P = 0.0001$). However, the interaction effect of GG and CMC and also the quadratic effect of CMC were non-significant. It can be seen that the variable with the largest effect on the turbidity was the quadratic effect of GG content, followed by the linear term of CMC content. In general, an increase in the levels of GG and CMC led to an increase in turbidity value (Figure 2B). The results suggested that an emulsion containing 0.40% (w/w) GG and 0.44% (w/w) CMC would result in the highest turbidity value (Y_2=0.804). Dłuzewska *et al.* (2006) explained the loss of turbidity of emulsions may be due to the aggregation of oil droplets and the changes in refractive index of oil phase and aqueous phase. Thus, increase of GG and CMC concentrations by changing refractive index of disperse phase can enhance turbidity and stability of the model emulsions. Genovese and Lozano (2001) and Abd-El-Salam *et al.* (1991), respectively, by adding 0.4-0.5 and 0.05% CMC to fruit juices found that this component can efficiently maintain their turbidity during an extended storage period. Mikkonen *et al.* (2009) also showed that use of galactomannans like GG can improve the turbidity of emulsions both immediately after preparation and after storage of up to 14 days at room temperature.

Size index optimisation

Table 2 shows that all the effects of independent variables except the linear effect of GG content were significant on the size index ($P < 0.0001$; $P < 0.001$). The most significant ($P < 0.05$) effect on size index was shown to be the interaction effect of GG and CMC concentrations (Table 2). Increases in CMC content resulted in reduced size index, although this parameter sharply increased with increasing GG concentration (Figure 2C). The individual optimum location

Table 2. ANOVA table for the experimental variables as a linear, quadratic and interaction terms of each response variable and corresponding coefficients for the predictive models.[1]

Source	DF	pH			Turbidity			Size index		
		Coefficient	Sum of squares	*P*-value	Coefficient	Sum of squares	*P*-value	Coefficient	Sum of squares	*P*-value
Model	5	4.05	0.37	0.0001	0.76	0.040	<0.0001	0.910	0.089	<0.0001
Linear										
b_1 (GG content)	1	0.11	0.10	0.0004	0.039	0.012	0.0002	-	0.00002	ns
b_2 (CMC content)	1	0.14	0.16	<0.0001	0.040	0.013	0.0001	-0.035	0.009	0.0002
Quadratic										
b_{11}	1	-	0.0001	ns[2]	-0.045	0.015	<0.0001	0.043	0.014	<0.0001
b_{22}	1	0.094	0.065	0.0015	-	0.00003	ns	0.031	0.007	0.0005
Interaction										
b_{12}	1	0.095	0.036	0.0079	-	0.00003	ns	0.12	0.060	<0.0001
Residual	7		0.023			0.002			0.001	
Lack-of-fit	3		0.0006	0.9859		0.0009	0.3912		0.0008	0.3781
Pure error	4		0.023			0.001			0.001	
Total	13		0.39			0.042			0.091	
R^2		0.940			0.947			0.979		
R^2_{adj}		0.903			0.914			0.966		
CV		1.32			2.29			1.60		
PRESS		0.037			0.008			0.007		
ADP		16.99			18.02			31.27		

Source	DF	Viscosity (cP)			Emulsion stability (%)		
		Coefficient	Sum of squares	*P*-value	Coefficient	Sum of squares	*P*-value
Model	5	582.33	6.814E+05	<0.0001	78.78	692.87	<0.0001
Linear							
b_1 (GG content)	1	249.82	4.993E+05	<0.0001	8.32	554.25	<0.0001
b_2 (CMC content)	1	61.58	30,333.9	<0.0001	1.16	10.70	0.0420
Quadratic							
b_{11}	1	75.21	41,769.55	<0.0001	2.76	56.19	0.0005
b_{22}	1	126.21	1.176E+05	<0.0001	3.03	67.95	0.0003
Interaction							
b_{12}	1	-24.0	2,304.0	0.0453	-1.77	12.60	0.0305
Residual	7		3,282.1			14.64	
Lack-of-fit	3		502.8	0.8237		8.09	0.2243
Pure error	4		2,779.3			6.55	
Total	13		6.847E+05			707.51	
R^2		0.995			0.979		
R^2_{adj}		0.992			0.966		
CV		2.09			1.65		
PRESS		75.77			66.98		
ADP		53.28			26.58		

[1] GG = guar gum; CMC = carboxymethyl cellulose; R^2 = coefficient of determination; R^2_{adj} = adjusted R^2; CV = coefficient of variation; PRESS = prediction error sum of squares; ADP = adequate precision; DF = degrees of freedom.
[2] ns = non-significant.

Table 3. Optimum formulation, and predicted and experimental values of the responses at this optimal point.

Independent variables[1]	Optimum condition	
GG (%, w/w)	0.50	
CMC (%, w/w)	0.30	
Response variables	**Experimental[2]**	**Predicted**
pH	4.05±0.08	4.02
Turbidity	0.729±0.25	0.706
Size index	0.911±0.09	0.900
Viscosity (cP)	1,021±47	996
Stability (%)	94.25±0.12	93.5

[1] GG = guar gum; CMC = carboxymethyl cellulose.
[2] Mean ± standard deviation (n=5).

led to the least size index (Y_3=0.834) for the model emulsion was estimated to be achieved by a set level of 0.30% (w/w) and 0.40% (w/w) for GG concentration and CMC content, respectively. It seems that the addition of only 0.3% GG into the emulsion can relatively well cover oil droplets surface, inhibiting the droplet aggregation and forming a smaller droplet along with narrow size distribution (Gharibzahedi *et al.*, 2013c). Presence of GG and CMC polysaccharide molecules in the emulsion structure by forming external layers of film and suitable interfacial surface can lead to a decrease in the droplet size (Genovese and Lozano, 2001; Gharibzahedi *et al.*, 2013b)

Viscosity optimisation

Table 2 shows that the viscosity was directly related to the effects of linear and quadratic and interaction of GG and CMC contents (P<0.0001; P<0.05). The linear term of GG

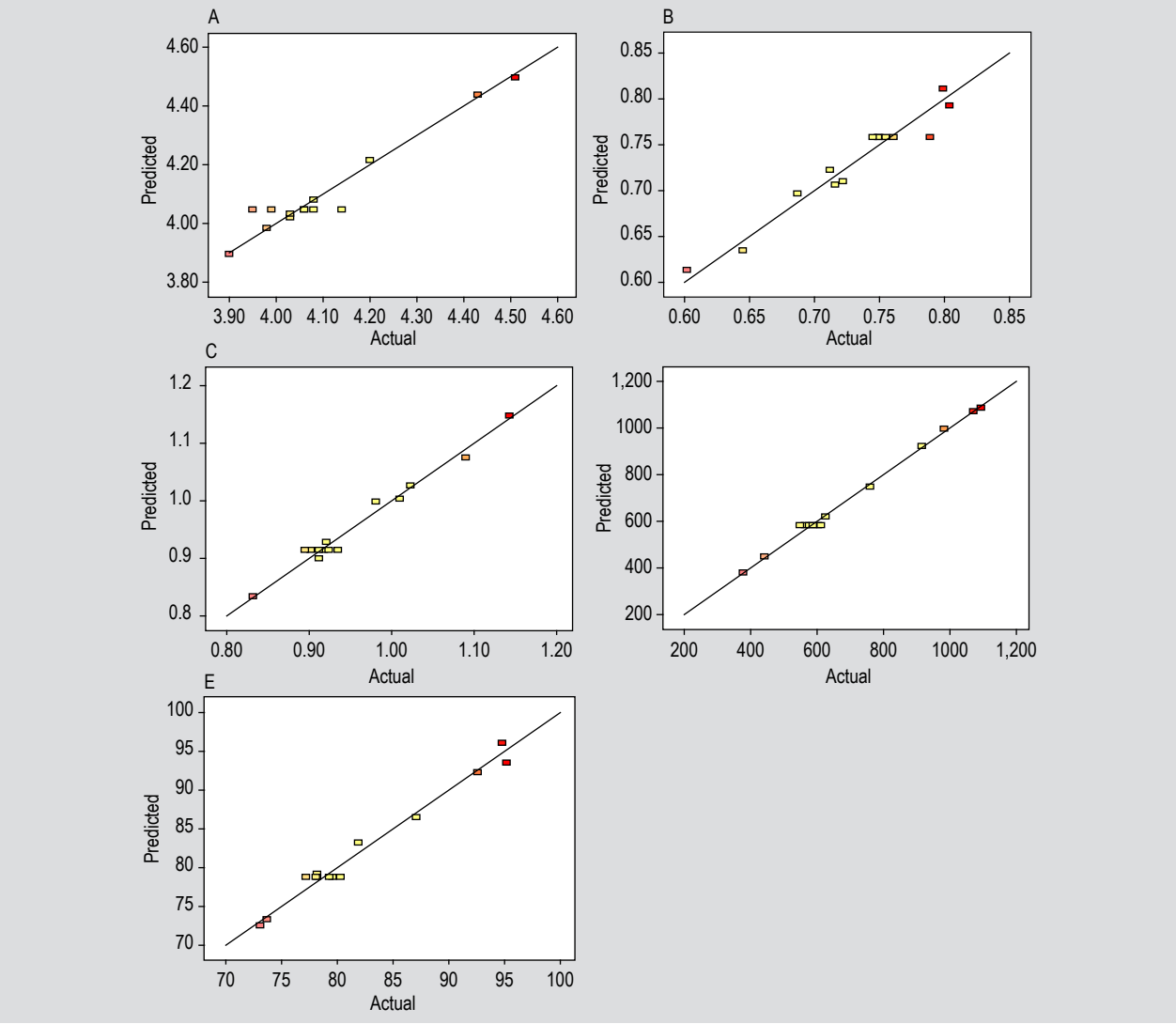

Figure 1. Comparison between predicted and actual values of (A) pH, (B) turbidity, (C) size index, (D) viscosity, and (E) stability of the developed model emulsions.

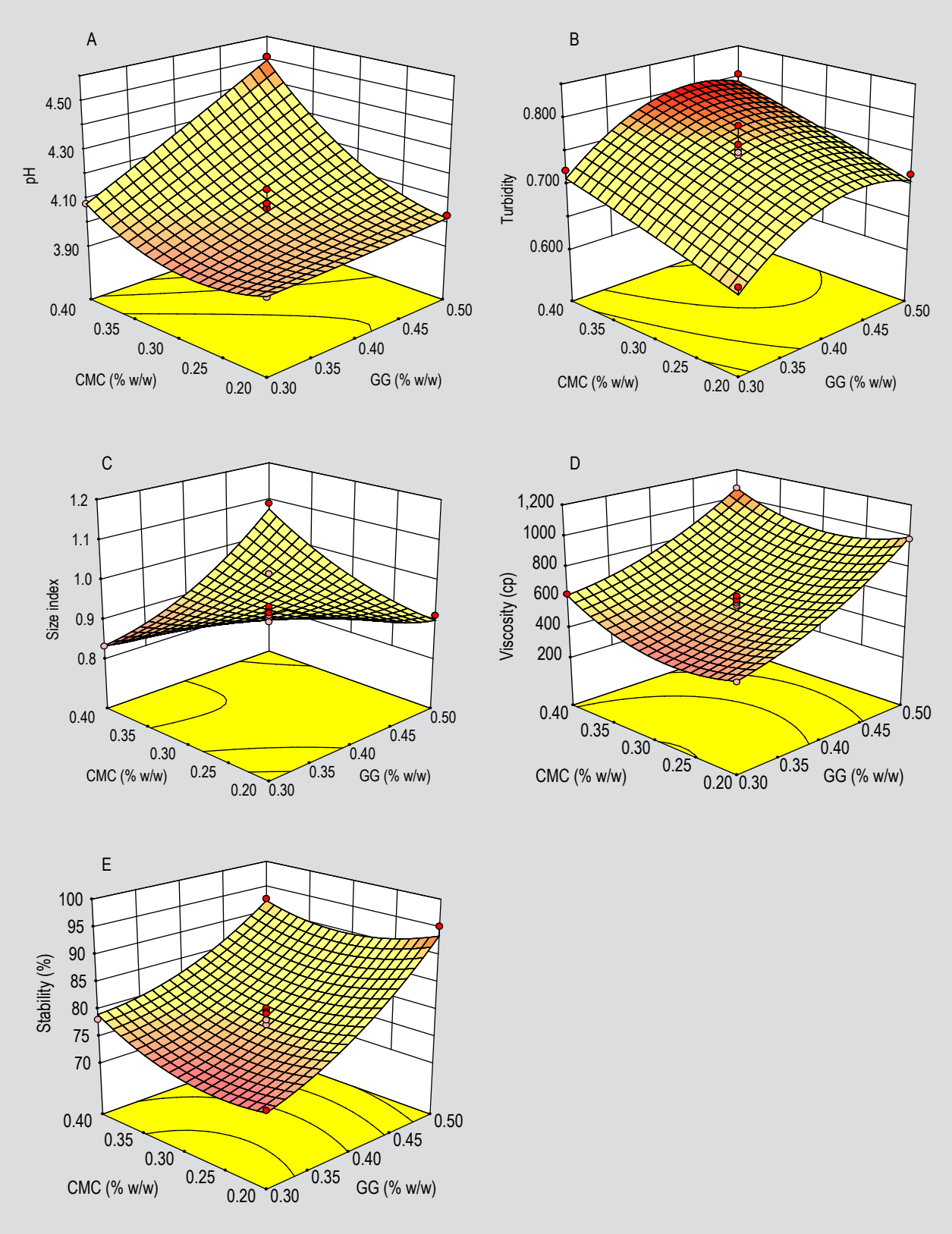

Figure 2. 3D plots showing the combined effect of guar gum (GG) and carboxymethyl cellulose (CMC) contents on (A) pH, (B) turbidity, (C) size index, (D) viscosity, and (E) emulsion stability.

concentration was the most significant factor on viscosity of the produced emulsions. The viscosity increased as the contents of GG and CMC were increased (Figure 2D). The individual optimisation procedure showed that the emulsion containing 0.50% (w/w) GG and 0.40% (w/w) CMC would provide the highest viscosity (Y_4=1,071 cP). The viscosity amounts were strongly correlated with turbidity values (P<0.01; r^2=0.977). The high viscosity of emulsions in a pseudoplastic behaviour trend by adding two polysaccharides can be related to the formation of a network structure in the aqueous phase. It was reported that when GG is solved at high temperatures there is a bigger proportion of smooth region than hairy region of the polymer with an enhancement of the number of intra and/or intermolecular bonds and hence the viscosity of the solution (Casas and Garcia-Ochoa, 1999). Polymeric chains of CMC at high concentration or molecular weight in the model emulsions become entangled, and the bulk flow becomes difficult, therefore, the viscosity significantly increases (Radi and Amiri, 2013).

Stability optimisation

All the effects of linear, quadratic and interaction of GG and CMC on the emulsion stability were significant (P<0.0001, P<0.001 and P<0.05, respectively). The most effective parameter on the improvement of stability was the linear effect of GG and the quadratic effect of CMC, respectively (Table 2). The highest response for emulsion stability (Y_5=93.51%) in the ranges studied was observed when the emulsion was produced with 0.5% (w/w) GG and 0.2% (w/w) CMC. Thus, an increase in GG amount and a reduction in CMC concentration led to high stability for the produced model emulsions (Figure 2E). GG stabilises different emulsions mainly by modifying the rheological properties of the aqueous phase between the dispersed particles and also its strong interfacial activity (Dickinson, 2003; Garti and Reichman, 1994). Garti and Reichman (1994) reported that relatively high GG concentrations were required for full coverage of oil droplets and emulsion stabilisation, and water dilution of those emulsions will cause fast desorption of GG and emulsion destabilisation. The negative effect of CMC on the emulsion stability can be due to the large excess presence of non-absorbed polysaccharide in the aqueous phase of emulsions prepared by high contents of polysaccharides (Dickinson *et al.*, 1999). When sufficient surface active agent is present to saturate the droplet surface, the increase of surfactant concentrations can be attributed to depletion flocculation caused by excess bulk of two surface-active materials (Mirhosseini *et al.*, 2008b). Gharibzahedi *et al.* (2013b) also attributed the destabilisation of walnut oil-water emulsion by XG to depletion flocculation since they found no evidence for adsorption of XG at the emulsion droplet surface.

Overall optimisation and validation of response surface methodology models

A stable emulsion would be considered an optimum product if the criteria applied for the optimisation resulted in the least values of pH and size index and the highest levels of viscosity, stability and turbidity. The RSM package's response optimiser determined the overall optimum region with high total desirability to be at 0.50% (w/w) GG and 0.20% (w/w) CMC. The corresponding predicted response values under the optimum conditions for pH, turbidity, size index, apparent viscosity and stability of the model emulsion were 4.02, 0.706, 0.900, 996 cP and 93.5%, respectively. The findings by conducting five replicates in the optimal point demonstrated that the corresponding experimental values for pH, turbidity, size index, apparent viscosity and stability of the desirable model emulsions were 4.05±0.08, 0.729±0.25, 0.911±0.09, 1,021±47 cP and 94.25±0.12%, respectively (Table 3). Figure 3 also shows that the increase of the emulsifier content and initial concentration of the applied CMC can lead to a high emulsion stability and minimum droplet size along with narrow size distribution. The comparison between the actual response values obtained from experimental data and the predicted response values based on the polynomial regression models proves that the models cover the experimental range of studies sufficiently.

4. Conclusions

In the present study, the contents of GG and CMC to produce a model beverage emulsion containing 7% CO were optimised to find the best physicochemical properties. RSM was found to be a key tool for optimising the proportion of emulsion structural components leading to the desirable goals of response variables studied. The

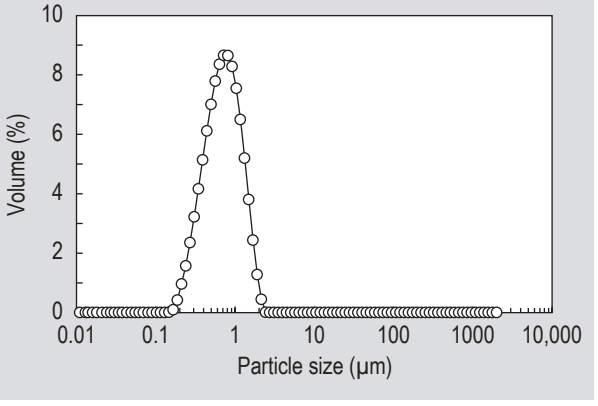

Figure 3. Droplet size distribution of the emulsion produced at the optimal point (0.50% guar gum and 0.20% carboxymethyl cellulose).

response surface analysis offered the significant regression equations with high R^2 and non-significant lack-of-fit. This observation demonstrated a satisfactory adjustment of the response surface models fitted to the experimental data. The optimum set of the independent variables was obtained graphically in order to find the desired levels of pH, turbidity, size index, viscosity and stability. The optimum composition emulsion (0.5% GG and 0.2% CMC) can considerably control the instability rate.

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