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Optimization attributes of fig (*Ficus carica* L.) salad dressing enrichment by *Mentha pulegium* L. extract and carboxymethyl cellulose

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Abstract

Common salad dressings led to further calories for consumers owing to high fat in their formulations. The aim of this research is to produce low-calorie salad dressing with high nutritional value and reduced fat. Box-Behnken designs were applied including figs (*Ficus carica* L.) (40 to 80%), carboxymethyl cellulose (CMC) (0.25 to 1.25%) and *Mentha pulegium* L. (MPL) extract (0.01 to 0.05%) as independent variables. The total soluble solids (TSS), pH, viscosity, stability,

peroxide index and sensory evaluations were performed; afterwards, calorie, and fatty acids (FA) were evaluated, and also scanning electron microscopy was carried out. The optimal conditions were obtained for salad dressing formulations with the highest TSS (40.3479%), viscosity (15898.75 cP), stability (94.2994%) and sensory (4.6282) and also the lowest pH (4.6032) and peroxide (0.9778 mEq/kg oil) related to fig (65.4545%), CMC (1.995%) and MPL (0.01%) concentrations, respectively. The optimal sample reduced 6-fold the calories compared to control and also demonstrated the maximum monounsaturated FAs with uniform distribution for particles. Fig salad dressing produced as a low-calorie product has the potential to be used by consumers.

Introduction

Ficus carica L. (Moraceae family), commonly known as fig, is one of the fruits cultivated in tropical regions, Asia and Mediterranean countries. Polyflavonoid and polyphenolic constituents in figs, which are distributed in skin and pulp, have antioxidant properties.¹ In different industries, fig fruit powder has been used in snacks² and toffee.³

As a natural food preservative, *Mentha pulegium* L. (MPL) has antibacterial, antifungal and antioxidant features, and is one of the best vegetables useful to improve digestive system functions, spasms and cramping of abdominal muscles.⁴ Today, due to the increasing concern about the toxic effects of using chemicals in food as additives, the role of plant extracts and essential oils as preservatives devoid of harmful effects on human health have been studied.^{5,6} As a result, in this study, MPL extract was used as a natural additive in sauce production.

Various hydrocolloids are used to increase the consistency for salad dressing such as Carboxymethylcellulose (CMC), which is a linear polysaccharide of anhydroglucose and an anionic and also water-soluble derivative of cellulose. In past research, the effect of replacing bacterial cellulose as a fat substitute in mayonnaise has been investigated.^{7,8}

The main goal of the current study is to produce a low-calorie salad dressing from natural fruit and MPL extract as a natural preservative with an optimal formulation. So, the ingredients of salad dressing including figs (40 to 80%), CMC (0.25 to 1.25%) and MPL extract (0.01 to 0.05%) were optimized using the Box-Behnken Design (BBD) and physicochemical, rheological and sensory

functions were measured. Finally, optimum sample was compared to mayonnaise as a control in terms of Fatty Acid (FA), calorie content and Scanning Electron Microscopy (SEM) images.

Materials and Methods

Material selection

Fig was purchased from local market, CMC was obtained by Sunrose Co., Nippon Paper Group in Tokyo, Japan, and phenolphthalein and potassium hydroxide with other substances were attained through Merck, Darmstadt, Germany.

Preparation of MPL extract

The obtained MPL was purchased from the local Tajrish market in Tehran (Iran) and evaporation operation was performed using a rotary evaporator.⁹

Production of fig salad dressing

Figs were crushed into small pieces and mixed with acetic acid, citric acid, salt, egg yolk, and MPL, according to the amounts defined for each formulation based on preliminary assays within specific limits by BBD. In all treatments, 2.5% acetic acid (vinegar), 1.5% citric acid, 0.5% salt, 0.5% egg yolk and obtained three concentrations of figs (2, 50, and 80%), CMC (0.25, 0.75, and 1.25%) and MPL extract (0.01, 0.03, and 0.05%) by BBD were employed. The percentage of water added to each sample varied until reaching 100%.

Physicochemical attributes

Total soluble solid (TSS) and pH of samples were determined by a handheld refractometer (Atago, N 3000E, Arnhem, Netherland) and pH meter (Metrohm 827, Herisau, Switzerland) at room temperature, respectively.¹⁰

Rheological characterization

The apparent viscosity was measured for samples through a Brookfield viscometer (model RV-DVIII, Brookfield programming Rheometry, Inc., Stoughton, USA) at $19 \pm 1^\circ\text{C}$. The flow curves were determined for samples using spindle speeds of 10, 40, 70 and 100 rpm and also viscosity levels were calculated and recorded after 60 s rotation.¹¹

Physical stability

Samples of fig salad dressing (25 mL) were weighed in tubes and centrifuged at 3000 rpm for about 10 min. In the next step, samples were placed in an oven (50°C) for 48 h and oil layer separated from salad dressing was discarded; then, emulsion stability was calculated as a percentage ratio of sediment to initial weights.¹²

Peroxide index

Briefly, 0.3 mL volume of salad dressing and 1.5 mL isooctane/2-propanol solvent (3:1, v/v) were potentially mixed thrice (about 10 s each) and afterwards centrifuged at $3600 \times g$ during 2 min (Universal 320, Hettich, Tuttlingen, Germany). The solvent evaporation was performed under nitrogen stream; after that a less layer of extracted oil was purified. The peroxide index was measured at 500 nm through a spectrophotometric apparatus (Thermo Fisher Scientific, Madison, WI, USA) for oil samples after 20 min at 25°C.¹³

Sensory evaluation

The treatments were randomly served with lettuce and water; then, sensory characteristics were evaluated by panelists based on overall quality using hedonic scales; in descriptive and also quantitative analyses, a scale of 1 to 5 has been applied for each feature.¹⁰

Calorie content assay

Approximately, 1 g sample was weighed after drying and placed in a special container; then, 2 L water was poured in bucket inside well of bomb calorimeter. The adjustment system and device were turned on and also calorie content was measured.¹⁴

Detection of fatty acid profile by gas chromatography

FA methyl esters were prepared according to the guidelines provided by International Olive Oil Council; Initially, 0.2 mL potassium hydroxide 2 N was mixed completely with 0.1 g oil extract in hexane solvent (2 mL); after stirring for 50 s and giving time to form the upper framework, this layer was removed. After 12 h at 4°C, the obtained sample was diluted with fresh hexane (1:100, v/v) for injecting into device. A Gas Chromatography (GC) system (Agilent Technologies,

Wilmington, DE, USA) consisting of 6890N GC and G1540N flame ionization detector was employed to analyze FAs.¹⁵

SEM assessment

The samples were fixed on glass by glutaraldehyde (Sigma Aldrich, Oakville, Canada) and washed several times with phosphate-buffered saline (PBS) (Merck, Darmstadt, Germany) to remove impurities. Dried samples were coated by a thin gold layer and SEM (LEO Model VP 1450, Zeiss, Oberkochen, Germany) was applied for examination.¹⁶

Statistical analysis

Statistical analysis was employed using Design-Expert 7.0 software to process data and to build model and the independent variables for fig (20, 50, and 80%), CMC (0.25, 0.75, and 1.25%) and MPL extract (0.01, 0.03, and 0.05%) were investigated at three levels. These mentioned compounds were chosen to study and optimize the effect of process variables.

Results

Optimization results of choosing the most suitable formulation for fig salad dressing

The matrix of the plan, the obtained laboratory results and the predicted responses for each experimental units are given in Table 1. After converting the results, all non-statistically significant variables were removed and the equations were changed as follows:

$$Y_{\text{TSS}} = 16.37 + 0.259 \text{ fig} + 7.58 \text{ CMC} + 0.01 \text{ fig} \times \text{CMC} + 0.06 \text{ fig} \times \text{MPL}$$

$$Y_{\text{pH}} = 16.37 + 0.259 \text{ fig} + 7.58 \text{ CMC} + 0.373 \text{ MPL} + 0.310 \text{ fig}^2 + 0.253 \text{ CMC}^2 + 0.250 \text{ MPL}^2 + 0.00 \text{ fig} \times \text{CMC} + 0.04 \text{ CMC} \times \text{MPL}$$

$$Y_{\text{Viscosity}} = 5742 + 39.4 \text{ fig} + 1244 \text{ CMC} - 52627 \text{ MPL} + 40.3 \text{ fig} \times \text{CMC}$$

$$Y_{\text{Stability}} = 12.24 + 3.01 \text{ fig} + 154.15 \text{ CMC} - 2270 \text{ MPL} - 0.029 \text{ fig}^2 - 105.71 \text{ CMC}^2 + 21611 \text{ MPL}^2 - 4.22 \text{ fig} \times \text{MPL} + 390 \text{ CMC} \times \text{MPL}$$

$$Y_{\text{Peroxide Index}} = 1.94 - 0.05 \text{ fig} - 47.27 \text{ MPL} + 0.002 \text{ fig}^2 + 0.27 \text{ fig} \times \text{MPL}$$

$$Y_{\text{Sensory evaluation}} = 1.415 + 4.141 \text{ CMC} - 14.2 \text{ MPL} - 0.001157 \text{ fig}^2 - 2.675 \text{ CMC}^2 - 17.00 \text{ CMC} \times \text{MPL}$$

According to Table 1, the amount for TSS is distinguished in the range of 22.10 to 40.03%. Figure 1a depicts the effect of increasing CMC and fig percentages on TSS for salad dressing samples with MPL concentration 0.03% in central point, which is kept constant. As observed in Figure 1a, TSS level for samples elevates with enhancement in both factors and Figure 1b indicates the effect of higher CMC and MPL percentages on this factor. The highest TSS level belonged to a sample, which had 80% figs, 1.25% CMC and 0.030% MPL.

In Table 1, the pH range was estimated from 4.11 to 5.7 in all samples; also, as it can be seen in Figure 1c, pH reduced with enhancing fig percentages, while in the other factors, the reverse trend was found. Figure 1d demonstrates the enhancing fig percentages caused pH reduction and the increase of CMC and MPL led an increase in this factor. According to Table 1, the viscosity for obtained samples is observed in the range of 6010 to 15200 cP. Figure 1e illustrates the graph effect of enhancing CMC and fig percentages on sample viscosity. The maximum viscosity belonged to a sample that had 80% figs, 1.25% CMC and 0.30% MPL.

The stability of salad dressing matrices with different levels was between 42.42 and 98.20% (Table 1). Stability developed and then declined by addition of figs; on the other hand, a completely opposite trend was observed where MPL increased (Figure 1f). The highest stability was for 80% figs, 0.25% CMC and 0.03% MPL (Figure 1g).

The hydroperoxide of lipids was determined in order to chemically evaluate oxidative intensity in fig salad dressing samples and peroxide index was estimated to be 0.41 to 1.41 mEq/kg oil (Table 1). The lowest peroxide index belonged to samples that had 80% figs, 0.25% CMC and 0.03% MPL. Figs and MPL had the greatest effect in reducing peroxide level for samples and CMC had no influence on lowering this factor (Figure 1h).

The range of organoleptic evaluation for all samples was estimated from 3.11 to 5.00 (Table 1). As it can be seen, with more CMC percentages sensory feature enhanced to a point and then reduced. Regarding to other factors, the scoring was diminished through MPL percentage (Figure 1i). The highest sensory score related to samples that had 50% figs, 0.75% CMC and 0.03% MPL.

Formulation optimization of salad dressing

In general, optimal condition for sauce formulations with the highest TSS 40.3479, pH 4.6032%, viscosity 15898.75 cP, stability 94.2994%, sensory evaluation 4.6282 and with the lowest peroxide

index 0.9778 mEq/kg oil corresponding to 65.4545% figs, 1.1995% CMC and 0.01% MPL was obtained. In this condition, the optimal formulation was employed to determine the further assays.

Calorie evaluation

Based on calorie measurement, mayonnaise sauce (as a control) and fig salad dressing had 500 and 80 calories, respectively. The calorie content of mayonnaise is 6.25 times that of fig salad dressing, which indicates very low-calorie content of the prepared sample.

Fatty acid profile

Table 2 demonstrates FA chromatogram for optimal sample of fig salad dressing and mayonnaise containing sunflower oil. The most FAs in produced salad dressing included polyunsaturated forms (48.3%), followed by monounsaturated FAs (45.8%) and saturated FAs (5.8%); therefore, figs and egg yolk were the source that made up salad dressing. In mayonnaise sample, monounsaturated FAs (43.1%), saturated FAs (33.9%) and polyunsaturated FAs (22.7%) were detected as dominant. As a result, harmful palmitic FAs in mayonnaise oil was higher and useful polyunsaturated FAs was much lower than fig salad dressing. Generally, FAs in mayonnaise structure are 60% more than fig salad dressing. Although no oil was applied in salad dressing due to fig presence, there was a significant percentage of mono and polyunsaturated FAs in structure. Saturated FAs were reduced remarkably in optimal treatment compared to control and also linoleic and linolenic acids.

SEM images

The images related to microscopic structure of samples are indicated in Figure 2; uniform distribution for particles, especially oil droplets, are extremely important in salad dressing stability and absence of a layer on product surface (Figure 2b). In mayonnaise sample (Figure 2a), oil drops are clearly visible and fat globules are surrounded by porous structure of protein network.

Discussion

In line with the present results, for prepared pepper sauces containing only starch and xanthan, TSS was obtained from 17.04 to 37.61%, which was reported as the sample with the lowest level having an undetectable yield stress, consistency coefficient and the highest flow index; so it was

the weakest network structure among studied salad dressing.¹¹ TSS were reported 37.45 and 38.05% for acerola ketchup and commercial tomato sample, respectively.¹⁷

The highest pH value was obtained for fresh figs (4.86), frozen figs (4.25) and jam (4) samples.¹ In the study conducted on tomato sauce, the pH range of 4.18-4.48 was obtained.¹⁸

In terms of rheological properties, salad dressing is a complex system and viscosity has a direct effect on consumer acceptance.¹⁰ In a study, enhancement in viscosity of aqueous phase due to xanthan gum addition had been indicated to reduce movement of oil droplets.¹¹ The viscosity reduction in mayonnaise enriched with olive leaf was probably due to physical and chemical interactions between phenolic compounds in essential oil or hydrocolloids,¹⁷ which was consistent with present results. Also, the viscosity of 1,589-2,286 cP has been reported for different samples of carrot sauce mixed with tomato.¹²

The stability of the colloidal system is due to the dominant negative electrostatic forces in the layers of ions formed on the surface of the sauce particles.¹³ The increase in MPL caused a decrease in the stability of the sauce, which is probably caused by the interaction of the compounds in the sauce with phenolic compounds and the disruption of the emulsions.⁹ CMC in concentrations less than 0.05% completely inhibits the clarification of apple juice.^{19,20} Adding guar gum, xanthan gum and CMC in two concentrations (0.1% and 0.25% w/w) in béchamel salad dressing had caused a significant enhancement in the stability of system.²⁰ In a study on mayonnaise enriched with olive leaf, it was found that phenolic constituents diminished stability.²

Figs and MPL had the greatest effect in reducing the peroxide index of the samples, and CMC had no effect on reducing this factor. MPL is a valuable natural source of bioactive compounds and phenolic compounds.²¹ Based on available results, the peroxide index analysis results for mayonnaise were reported as 5.12-20 mEq/kg oil.¹¹

In line with the results of this study, in the sample of the sauce prepared with cassava starch oxidized with sodium bicarbonate as a fat substitute, the sauce was stable and also acceptable in terms of sensory evaluation.¹⁴ In the study of salad dressing prepared with flaxseed oil, it was found that with 5% (w/w) of flaxseed oil, a higher sensory quality was obtained.¹⁰

In prepared fig salad dressing, the application of CMC hydrocolloid as a fat substitute had reduced product calories. Samples made from sorghum and corn-xanthan gums,²² whey protein concentrate²³ and cassava starch¹⁴ had been used to produce low calorie mayonnaise in similar studies.

The FAs are determined as myristic (14:0), palmitic (16:0), stearic (18:0), oleic (18:1), linoleic (18:2) and linolenic acid (18:3) in fig fruit.²⁴

Omega-3 and 6 FAs are the two main subgroups of polyunsaturated FA in figs and also are vital healthy fats for cell growth and brain function; however, human body is unable to produce essential contents in diet and phytosterol of figs was 433 mg/100 g.¹⁵

Images reported that excellent stability for product was associated not only with small oil droplets but also uniform oil droplet diameters.¹¹ All mayonnaise samples and mixed with fruit indicated a well-dispersed oil-in-water structure and globules were spherical,¹⁶ which was in line with present results.

Conclusions

Nowadays, the need to produce ultra-beneficial products with low calories has improved by increasing nutritious consumption and low-risk foods. In the present study, tests illustrated the functional and low-calorie fig salad dressing for consumer acceptance. Fig (65.4545%), CMC (1.995%) and MPL (0.01%) concentrations were the optimal conditions for salad dressing formulations. The current study has limitations such as the lack of a large statistical population as an evaluator and time restrict to perform a wide range for tests; therefore, it will be necessary to extend shelf life against preservative absence in the future.

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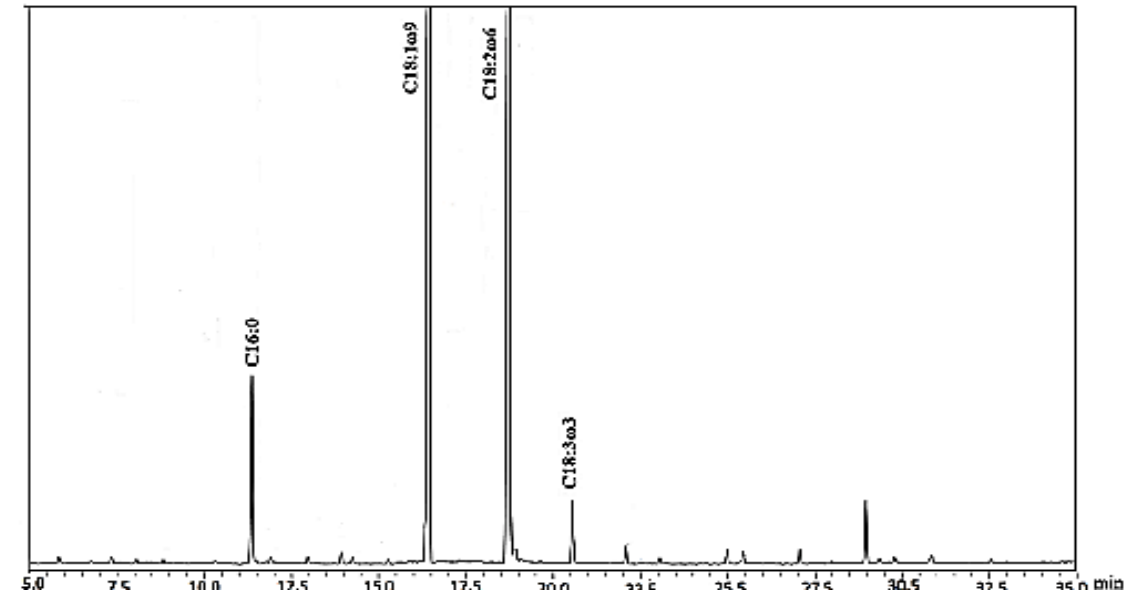
Table 1. Optimum conditions by the BBD on TSS, pH, viscosity, stability, peroxide index and sensory evaluation based on independent variables of salad dressings.

Independent variables (%)			TSS (%)		pH		Viscosity (cP)		Stability (%)		Peroxide Index (meqH ₂ O ₂ /kg)		Sensory Evaluation	
Fig	CMC	MPL	Actual value	Predicted value	Actual value	Predicted value	Actual value	Predicted value	Actual value	Predicted value	Actual value	Predicted value	Actual value	Predicted value
20	0.25	0.03	22.1	21.592	5	4.99	6458	6662	46.07	44.28	0.83	1.045	3.45	3.55
80	0.25	0.03	34.08	31.342	4.11	4.07	10178	10319	42.42	40.98	0.41	0.307	3.4	3.44
20	1.25	0.03	27.44	27.177	5.5	5.53	9061	8920	49.11	50.55	0.82	0.922	3.2	3.15
80	1.25	0.03	40.03	41.538	4.9	4.90	15200	14996	42.44	44.23	0.54	0.325	3.11	3.00
20	0.75	0.01	25.65	26.237	4.85	4.83	7613	7614	97.60	97.79	1.34	1.979	4.61	4.55
80	0.75	0.01	37.3	37.117	4.15	4.16	13020	13084	98.20	98.04	0.50	0.596	4.41	4.40
20	0.75	0.05	26.2	26.382	5.7	5.68	6010	5946	67.03	67.19	0.60	0.504	3.5	3.50
80	0.75	0.05	38	39.612	4.8	4.81	10210	10209	57.50	57.31	0.43	0.551	3.33	3.39
50	0.25	0.01	28.71	28.630	4.13	4.14	9021	8816	97.80	99.40	1.41	1.416	4.93	4.88
50	1.25	0.01	35.4	35.075	4.76	4.73	12180	12320	98.00	96.37	1.27	1.389	4.71	4.81
50	0.25	0.05	28.18	28.505	4.78	4.80	6721	6581	54.30	55.93	0.60	0.681	4.3	4.19
50	1.25	0.05	35.76	37.840	5.6	5.58	9808	10013	70.10	68.50	0.61	0.604	3.4	3.44
50	0.75	0.03	31.91	31.927	4.3	4.31	10245	9527	98.00	97.83	0.82	0.823	5	5.00
50	0.75	0.03	32	31.927	4.31	4.31	9110	9527	97.50	97.83	0.85	0.823	5	5.00
50	0.75	0.03	31.87	31.927	4.33	4.31	9226	9527	98.00	97.83	0.80	0.823	5	5.00

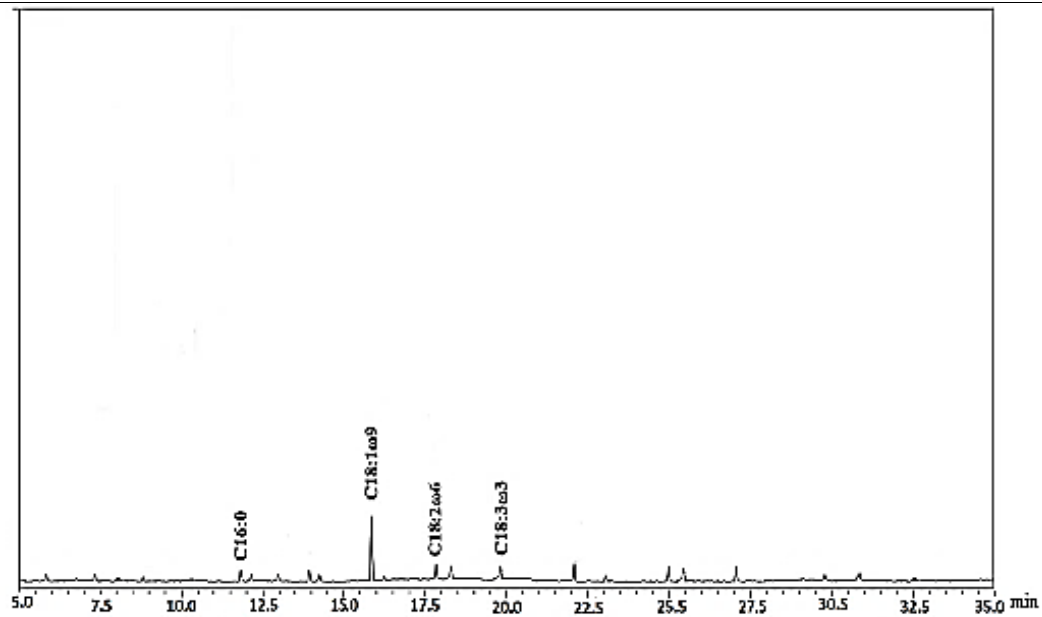
Table 2. Gas chromatography of FA profile of (a) control sample and (b) fig salad dressing to determine the concentration of FAs. DM: dry matter.

FA	Control	Fig salad dressing
		Concentration (g/100g DM)
Palmitic acid	20.36	0.10
Oleic acid (W ₉)	25.90	0.52
Linoleic acid (W ₆)	12.01	0.26
Linolenic acid (W ₃)	1.64	0.32
Total fat	60	1.2
		Percent (W/W)
Saturated fatty acids	33.9	5.8
Monounsaturated fatty acids	43.1	43.3
Polyunsaturated fatty acids	22.7	48.3

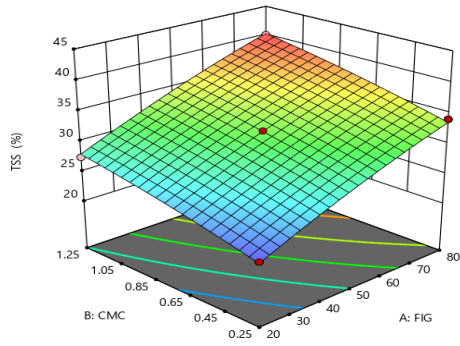
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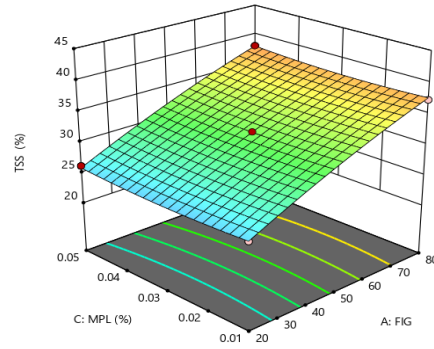
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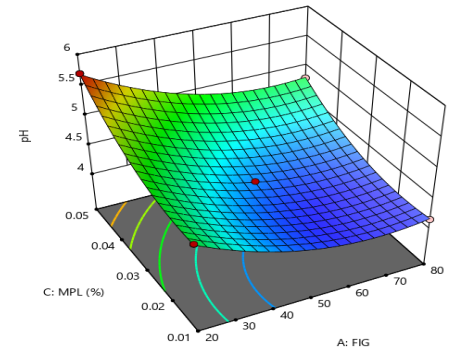
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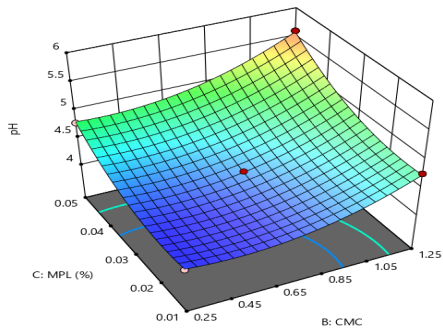
b



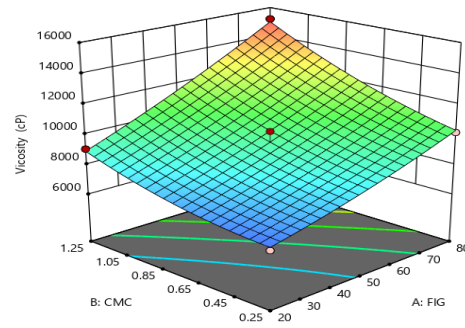
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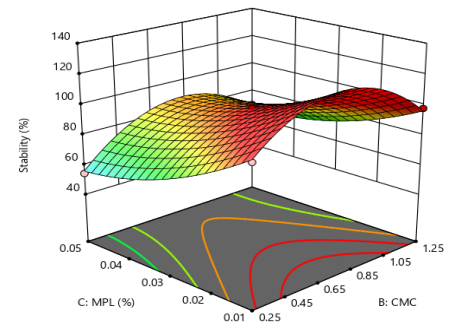
d



e



f



g

h

i

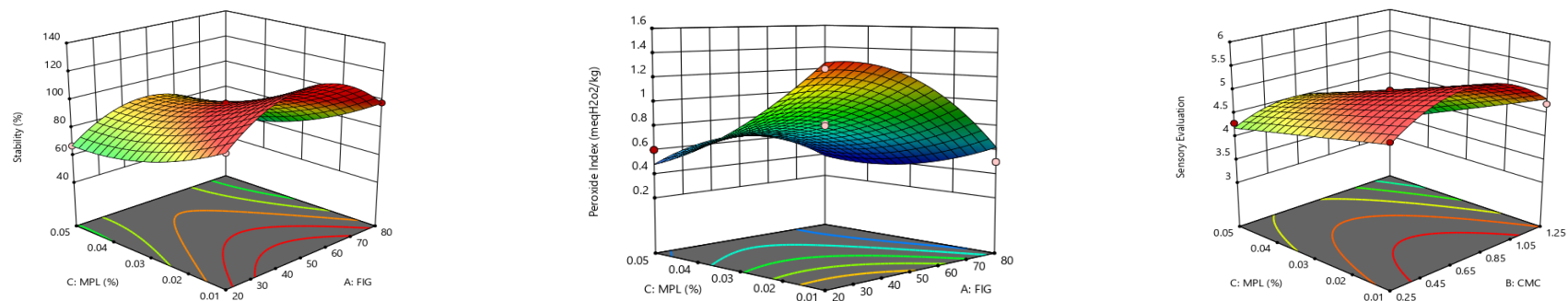


Figure 1. Three dimensional (3D) response surface plots indicating significant ($p < 0.05$) interaction effect for factors, A: Fig (20 to 80%), B: CMC (0.25 to 1.2%), C: MPL extract (0.01 to 0.05%) on (a-b) TSS (%), (c-d) pH, (e) viscosity, (f-g) stability, (h) peroxide index, and (i) sensory evaluation of fig salad dressing.

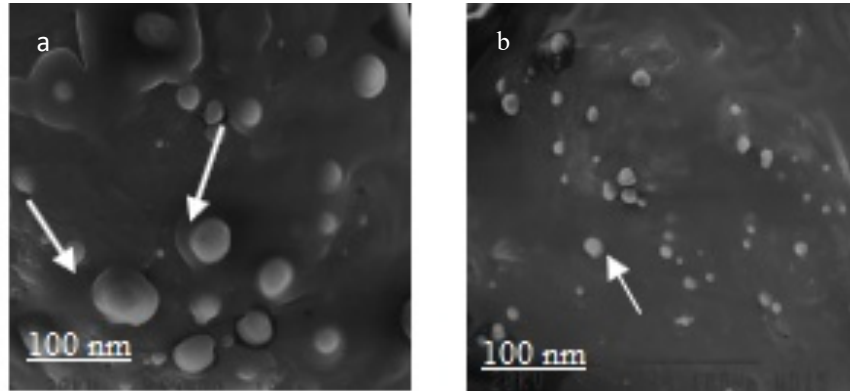


Figure 2. SEM images of mayonnaise (a) and fig salad dressing (b)