

Optimization And Its Characterization Of Legumes Based Milk Chocolate To Enhance Its Folic Acid Content

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Abstract— The objective of this work was to enhance Folic acid content in Legumes based Milk Chocolate, and to investigate the effect of incorporation of Legumes and Peanut milk by analyzing its Physico-Chemical and Organoleptic characteristics. The chocolate was made-up with different ratios of Legumes milk (Black-eyed pea, Cow pea, Chick pea and Yellow pea) and Peanut milk, with essential components of Cocoa powder, Butter and Jaggery. Sensory attributes of chocolates such as Flavour, Texture, Taste, Appearance and Colour were evaluated using 9 point hedonic scale, and found Yellow pea milk:Peanut milk of 2:1 ratio was most acceptable. Significant components of chocolate were optimized using Response Surface Methodology (RSM) based on Central Composite Design (CCD) experiments. The Analysis of Variance (ANOVA) data obtained from RSM was analyzed using a second degree polynomial equation and the contour plots obtained were used to study the interaction among the variables of legumes based milk chocolate. The optimum conditions for the developed nutritious chocolate was found to be Yellowpea = 80.52 g; Cocoa powder = 2.23 g and Butter = 5.06 g with the maximum Folic acid concentration of 2.0561 mg/g.

Index Terms— Legumes milk chocolate, Folic acid, Peanut, Yellowpea

I. INTRODUCTION

Dairy milk have been dominating the market as a good source of Protein, Minerals, Vitamins, and also numerous nutrients (Calcium, Magnesium, Selenium, Riboflavin, Vitamin B5 and Vitamin B12) with a significant contribution to the body's needs. However, dairy milk does not contain enough Iron and Folate to meet out the needs of growing children. Therefore the development and fortification of vegetable alternative to dairy milk in formulation of human foods has been increased due to their rich nutritional content. Hence the recent interest in plant based legumes used in vegetable milk and products made the growing awareness of the nutritional benefits of plant based foods by health conscious consumers. Legumes such as Black beans, Black-eyed peas, Chick peas, Cow peas, Kidney beans, Pinto beans, Split peas, Yellow peas and etc., provide many nutrients found in both vegetable and protein rich food groups, so they can count and satisfy our daily needs. And also they are naturally low in fat [1] and could be a healthy alternative to many animal-based proteins which are generally higher in saturated fats and cholesterol. Various legumes milk obtained from Black-eye peas, Cow peas, Chick peas, Yellow peas and blended with Peanut milk would be an ideal dairy milk substitute [2] in chocolate preparation. Legumes milk chocolate [3] made using the above composite blends have adequate sources of nutrients such as Protein, Minerals, Vitamins Dietary fiber

and Folic acid might be an alternative to dairy milk chocolate.

The objective of this work was to extract the milk from legumes that would be acceptable for the production of milk chocolates. The ingredients were optimized using Response Surface Methodology to produce best quality chocolate.

Quality and Effects of processing on nutritional value and mineral compositions of the legumes milk were studied.

II. MATERIALS AND METHODS

A. Raw materials

Black-eye pea (Vigna unguiculata), Cow pea (Vigna catjang), Chick pea (Cicer arietinum), Yellow pea (Pisum sativum) and Peanut (Arachis hypogaea) were purchased from the local market in Chidambaram, India. Care was taken to ensure that good quality and mould free legumes were selected. The Milk powder, Cocoa powder, Butter and Sugar were purchased from the local super market.

B. Sample pretreatments

Weighed quantities of the beans and nuts were blanched by submerging in boiling water for 1 and 5 minutes respectively, and then drained and dehulled. The dehulled

beans and nuts were soaked in 2%NaHCO₃ for 3 h and 18 h respectively and washed in clean water, to ensure the removal of bean flavor [4] in the final product and to soften the Peanuts.

C. Extraction of legumes milk

Soaked beans and nuts were grounded separately using blender in the ratio of 2:1 (Legumes:Water). It was blended to obtain a smooth, fine, homogenized liquid and filtered with muslin cloth to obtain legumes milk.

D. Preparation of chocolate

Legumes milk was heated below its boiling point and then specific quantities of Cocoa powder [5], Butter and Sugar were added with continuous stirring to get a smooth fine pastelike texture. Then it was poured in a mold and kept in refrigerator to obtain a desired shape.

E. Experimental design

Beans:Nuts milk ratio of 1:1, 2:1, 3:1 and 4:1 for all four beans were used for making chocolate with all other parameters kept as constant and 16 sets of chocolate were prepared totally. The optimum concentration of the variables and their interactions were studied by Response Surface Methodology (RSM) using Central Composite Design (CCD). The significant medium components were studied at 'n' levels, keeping other less significant components at constant level. The design of experiments was formulated using the statistical software Minitab. All experiments were carried out in triplicate and their mean values are presented.

F. Chemical analysis

The proximate composition of chocolates was determined using standard methods. Protein, Total Carbohydrate, Folic acid and Minerals (Na, Ca and K) were analysed using Lowry's method, Phenol-Sulphuric acid method, High Performance Liquid Chromatography (HPLC) and Flame Photometer respectively.

G. Sensory evaluation for acceptable legumes milk chocolates

Sensory attributes of Flavour, Texture, Taste, Appearance and Colour were evaluated using 9 point hedonic scale [6] (where 9-Like Extremely, 8-Like Very Much, 7-Like Moderately, 6-Like Slightly, 5-Neither Like nor Dislike, 4-Dislike Slightly, 3-Dislike Moderately, 2-Dislike Very Much, 1-Dislike Extremely) by using 15 panelist.

A. Proximate analysis of legumes and legumes milk chocolate

The proximate composition of legumes shown in table I illustrates that the moisture content of the four types of legumes are varied from 9.0 to 12.87% as well as their milk moisture varied from 80 to 85%. The protein values varied from 7.14 to 23.4 g/100g contrasting their milk chocolates protein varying from 8.8 to 25.0 g/100 g. This result showed a significant value in protein [7] during conversion process from legumes to legumes milk chocolate. The Folic acid content of various legumes are Black-eye pea: 444 mg, Cow Pea: 639 mg, Chick pea: 557 mg, Yellow pea: 2.74 mg and Peanut: 2.46 mg. Whereas in different ratios of legumes and peanut milk chocolates, the Folic acid content varied from 1.551 mg to 2.733 mg [8].

Table I: The proximate composition of legumes

Sample	Moisture (%)	Protein (g/100g)	Fat (g/100g)	Carbohydrate (g/100g)
Black-eyed pea	12.0	22.9	1.4	60.6
Cow pea	13.4	24.1	1.0	54.5
Chick pea	9.8	17.1	5.3	60.9
Yellow pea	16.0	19.7	1.1	56.5
Peanut	3.0	25.3	40.1	26.1

The level of preference of chocolates made using the legumes milk samples as well as a control was obtained using 9 points hedonic scale. The legumes milk 2:1 had scored maximum sensory score with respect to taste and overall acceptability. The chocolate acceptance was evaluated based on product appearance, colour, glossiness and smooth texture [9]. The results of sensory analysis of legumes milk chocolates prepared with different combination of legumes and peanut revealed that the yellow pea and peanut in the ratio 2:1 was best among all the samples with respect to overall acceptability. The yellow pea with peanut milk chocolates was rich in mineral, protein and folic acid content. Table II clearly indicates that chocolate prepared with legumes milk was nutritionally higher than the milk chocolates.

Table II: Proximate composition and Nutritive values of legumes milk chocolates

S. No	Legumes milk	Protein (g/100g)	Total Carbohydrate	Total Ash	Folic acid	Minerals (mg/100g)
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III. RESULTS AND DISCUSSION

	chocolate samples		drates (g/100g)	%	(mg)	Na	Ca	K
1	Black-eye pea milk	23.0	54.0	1.6	3.280	36	80	640
2	Cow pea milk	18.8	63.6	4.5	4.345	32	75	740
3	Chick pea milk	18.0	56.0	0.5	2.546	48	100	520
4	Yellow pea milk	19.0	70.5	1.5	3.083	56	65	870
5	Black-eye pea milk: Peanut milk (2:1)	25.0	67.0	3.8	1.551	67	174	690
6	Cow pea milk: Peanut milk (2:1)	28.0	72.0	5.7	1.738	56	150	790
7	Chick pea milk: Peanut milk (2:1)	21.0	65.8	0.6	2.733	72	180	778
8	Yellow pea milk: Peanut milk (2:1)	35.0	75.6	1.4	2.060	78	290	876

9	Control	15.0	55.8	2.0	1.711	64	240	440
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B. Optimization of parameters involved in the production of Yellow pea milk chocolate using Central Composite Design (CCD) and Response Surface Methodology (RSM)

The independent variables Yellow pea, Cocoa powder and Butter were optimized by CCD experimental design. Table III shows the range and levels of the independent variables selected for the preparation of legume based milk chocolate.

Table III: The range and levels of the ingredients selected for the preparation of yellow pea milk chocolate

Ingredients	Range and Level				
	-1.682	-1.0	0	1.0	+1.682
A – Yellow pea (g)	32.95	50	75	100	117.05
B – Cocoa powder (g)	1.159	1.5	2	2.5	2.841
C – Butter (g)	4.159	4.5	5	5.5	5.841

Table IV: The Central Composite Design matrix used in Response Surface Methodology with corresponding experimental Nutritive values and Folic acid, and predicted values of Folic acid in yellow pea milk chocolate

S.No	Coded values			Total Carbohydrates (g/100g)	Protein (g/100g)	Total Ash (%)	Minerals (mg/100g)			Folic acid (mg)	
	A	B	C				Na	Ca	K	Experimental	Predicted
1	-1	-1	-1	73.0	32.6	1.4	75	276	875	0.95667	0.9602
2	+1	-1	-1	69.5	34.0	1.6	74	280	870	0.86914	0.8563
3	-1	+1	-1	71.6	29.5	1.4	70	262	875	1.33753	1.3411
4	+1	+1	-1	73.2	33.8	1.5	48	100	720	1.54949	1.5367
5	-1	-1	+1	72.0	28.0	1.7	56	150	790	1.17954	1.1831
6	+1	-1	+1	74.0	34.0	1.4	32	175	740	0.9705	0.9577
7	-1	+1	+1	75.6	35.0	1.4	78	290	876	1.53889	1.5425
8	+1	+1	+1	74.0	33.6	1.6	74	267	868	1.62936	1.6165
9	-1.682	0	0	71.0	33.0	1.4	76	287	878	1.55112	1.5382
10	1.682	0	0	75.0	33.4	1.5	78	280	875	1.74870	1.5131
11	0	-1.682	0	72.3	30.0	1.4	72	259	860	0.71221	0.7188
12	0	1.682	0	74.3	32.2	1.4	76	284	870	1.58650	1.5931

13	0	0	-1.682	72.4	30.0	1.6	75	294	880	1.05928	1.0658
14	0	0	1.682	68.9	25.0	1.8	67	174	690	1.31384	1.3204
15	0	0	0	74.8	35.2	1.6	78	281	870	1.99179	1.9887
16	0	0	0	75.0	35.3	1.6	81	275	882	1.96013	1.9887
17	0	0	0	75.3	34.9	1.5	80	270	888	1.96936	1.9887
18	0	0	0	75.1	34.7	1.6	80	271	872	2.04090	1.9887
19	0	0	0	75.1	35.2	1.7	80	272	877	1.98217	1.9887
20	0	0	0	74.7	34.9	1.4	78	280	876	1.99007	1.9887

An orthogonal 23 factorial composite experimental design with eight star points ($\alpha = 2$) and six replicate at the center point, all in duplicates resulting in a total of 20 experiments were used to optimize the chosen key variables for the preparation of legumes milk chocolate. The experiments were carried out to study the outcome of legumes milk chocolate, using the variables such as yellow pea, cocoa powder, butter weight and their interactions on the chocolate preparation. Table IV shows the coded values for legumes milk chocolate preparation, and its experimental and predicted values of Folic acid along with its nutritional content.

C. Analysis of Variance (ANOVA) for the quadratic model

The ANOVA result for the quadratic regression model showed that the model was highly significant with good adequacy of the second-order polynomial model proposed to explain the observed yields. The coefficient of determination (R^2) for the Folic acid content in the legumes milk chocolate was 0.897, implying that only 10.3% of the total variations are not explained by the model.

The application of RSM yielded the following regression equation (1), which is an empirical relationship between response and independent variables in uncoded terms.

The regression equation was solved using MATLAB and the optimal values of the test variables were Yellow pea =

80.52 g, Cocoa powder = 223 g and Butter = 5.06 g yielding a predicted optimum Folic acid content of 2.0561 mg. To confirm the predicted response by the polynomial equation, validation experiment was carried out under optimum conditions and it was found that the experimental value of 1.9980 mg Folic acid content was near to the predicted value.

D. Optimum values of variable obtained from regression equation for the preparation of legumes milk chocolate

$$Y = 1.9980 + 0.0001X_1 + 0.0001X_2 + 0.0001X_3 + 0.0001X_1X_2 + 0.0001X_1X_3 + 0.0001X_2X_3 + 0.0001X_1^2 + 0.0001X_2^2 + 0.0001X_3^2$$

(1)

Where, X_1 , X_2 and X_3 are the coded values of Yellow pea, Cocoa powder and Butter weight respectively.

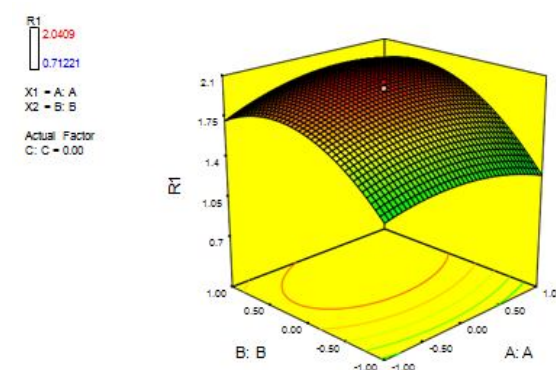


Fig. 1: Response Surface plot and Contour plot for Folic acid concentration in yellow pea milk chocolate, Yellow pea weight (A) versus Cocoa powder weight (B)

Fig. 1 illustrates the surface and contour plot effects of the varying amount of Yellow pea and Cocoa powder on the concentration of Folic acid with constant amount of Butter weight. From the plots it can be inferred that the concentration of Folic acid was higher at 80.52 g of Yellow pea and 2.23g of Cocoa powder.

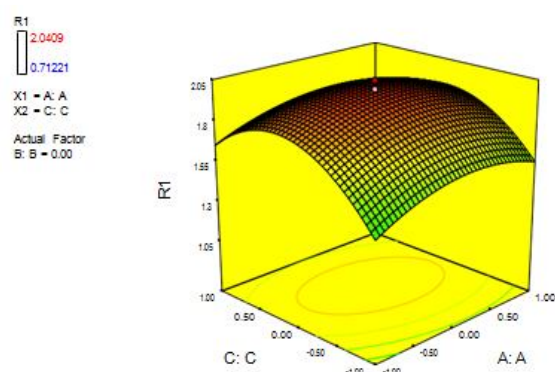


Fig. 2: Response Surface plot and Contour plot for Folic acid concentration in yellow pea milk chocolate, Yellow pea weight (A) versus Butter weight (C)

The surface and contour plot in fig. 2 indicates the varying concentration of Folic acid for different Yellow pea versus Butter weight. It was determined that the concentration of Folic acid was higher when 80.52 g of Yellow pea and 5.06 g of Butter were used.

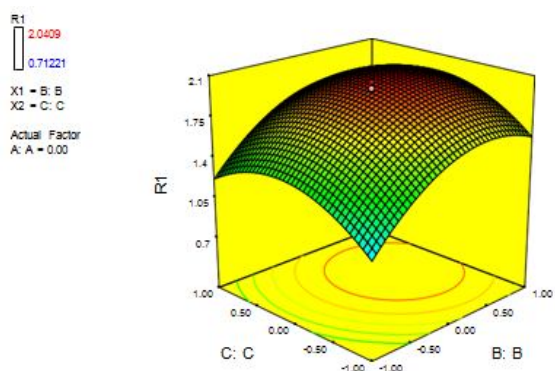


Fig. 3: Response Surface plot and Contour plot for Folic acid concentration in yellow pea milk chocolate, Cocoa powder weight (B) versus Butter weight (C)

Fig. 3 explains that when the Butter concentration was increased along with the amount of Cocoa powder used, the concentration of Folic acid present in the chocolate also increased. The maximum Folic acid concentration was observed at 2.23 g of Cocoa powder and 5.06 g of butter weight.

In this study, there were remarkable increases in both proximate and nutritional composition in the sixteen samples. The extent of solubility of the component part of legume would determine the composition of legume milk. The solubility has been improved due to the legumes pretreatment method which involves soaking, grinding and cooking. The protein content of chocolate samples

comparably have similar tendency as the initial protein content of different legume. The portions of proximate compounds in the legumes milk would be adhered to form part of colloidal solution in the milk. Carbohydrate constitutes the most predominant portion (24-26%) of the legumes that goes into milk colloidal solution. In conformity with the composition of cow milk reported in one study, moisture constitutes the major component of legumes milk, other components being present in small proportions. In terms of taste, colour, odour and mouth feel, the yellow pea milk chocolate have higher desirable attributes.

CONCLUSION

In the manufacturing of legumes milk chocolate no specialized and sophisticated equipments are needed for the extraction of legumes milk, and also high degree of nutrients are retained. From the results, it is evident that yellow pea: peanut ratio of 2:1 yielded better legumes milk chocolate with significant amount of Folic acid content. The optimum conditions for the preparation of highly accepted chocolate was found to be: Yellow pea = 80.52 g; Cocoa powder = 2.23g and Butter = 5.06 g with the maximum Folic acid concentration of 1.9980 mg which is close to the predicted value of 2.0561 mg.

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