RESEARCH PAPER | International Journal of Agricultural Engineering | Volume 11 | Sp. Issue | April, 2018 | 41-45
ISSN–0974–2662 ■ Visit us : *www.researchjournal.co.in* ■ **DOI: 10.15740/HAS/IJAE/11.Sp. Issue/41-45**

Optimization of antioxidant rich indigenous food product "burfi" recipe using response surface methodology and its storage study

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Received : 10.04.2018; **Accepted :** 13.04.2018

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■ **Abstract :** Efforts were made to prepare the indigenous food product called burfi using underutilized ghee residue and replacing the sucrose with many health and medicinal benefits containing honey. Response Surface Methodology was used to optimize the amounts of ingredients required to prepare a burfi containing minimum amount of free fatty acids (FFA), maximum amount of Antioxidants, phenols and flavonoid contents and overall acceptability by consumer on hedonic scale. The optimized and experimental values for FFA, Antioxidant, Phenolic, flavonoid content and overall acceptability were observed similar, thus indicating reliability of the software. Thereafter, storage study analysis for a period of 30 days of prepared burfi was performed in terms of FFA and overall acceptability. FFA content increased from 1.04% at initial level to 1.42% after 30 days, which remained within desirable limits. Prepared burfi was acceptable upto 30 days on the basis of sensory score and FFA.

■ **Key words :** Ghee residue, Burfi, Honey, FFA, Antioxidant activity, Phenolic content, Flavonoid content, Sensory characteristics, Response surface methodology

■ **How to cite this paper :** Bansal, Vikas, Premi, Monica and Khan, Khursheed A. (2018). Optimization of antioxidant rich indigenous food product "burfi" recipe using response surface methodology and its storage study. *Internat. J. Agric. Engg.*, **11**(Sp. Issue) : 41-45, **DOI: 10.15740/HAS/IJAE/11.Sp. Issue/41-45.**

India is world's largest producer of milk and contributes to about 19% of world production. Being milk perishable, about 50-55% milk of the total milk produced is converted into indigenous milk products by traditional sector (www.nddb.org/statistics/milk). Throughout the country, Khoa is one of the most popular milk product commonly used as base for variety of sweetmeats particularly for burfi, gulabjamun etc. Particularly in India, burfi is most popular khoa based sweetmeat. Burfi is prepared by continuous thickening of milk in an open pan until desired (semi solid) texture product is obtained called "khoa", to which sugar is added in either forms (crystalline, powder or syrup). Due to its market demand and popularity, different types of burfi with various ingredients and flavours have been evolved having firm body, granular to smooth texture and light cream to white in color.

Public demands for sugar free products or low sugars are increasing as consumers are more concern towards health risks that are involved due to consumption of sugar rich products. Honey is natural sweetener, having medicinal and therapeutic properties. Honey is commonly used as blood purifier, eye ailments, laxative and as a medicine against cough, cold and fever. In order to overcome various health issues and to obtain therapeutic value of sweetmeat, we can replace sugar with honey.

Dairy by-products, mostly considered as industry waste, but have high nutritive value. Ghee residue is the

largest by-product of dairy industry. It is rich source of fat and proteins. It can be utilized as flavoring fat blends and also for enhancing the keeping quality (as contains natural antioxidant). Thus, ghee residue (nutritious by-product) can be utilized for manufacturing of variety of sweetmeats by virtue of its chemical composition and physical properties.

Considering the therapeutic, medicinal value of honey and nutritious value of ghee residue an efforts were made to utilize honey and ghee residue for preparation of antioxidant rich indigenous food product "burfi". Also, there was no literature available for optimization of honey and ghee residue amount in burfi. Therefore, the objective of this study was to optimize the antioxidant rich indigenous rich product "burfi" using response surface methodology and its storage study.

METHODOLOGY

Khoa, honey and cream were procured from the local market. All the reagents and solvents used were of analytical grade. Distilled water was used for the preparation of all the solutions.

Determination of TPC, TFC and AOA :

Total phenolic content (TPC) was determined using the Folin-Ciocalteu method (Slinkard and Singleton, 1977) and results were expressed as mg GAE/g. Total flavanoid content (TFC) was determined by using the Dowd method, as adopted by Arvouet-Grand *et al*. (1994) and results were expressed as mg RE/g. Antioxidant activity was determined by standard method with slight modifications (Praveen and Awang, 2007).

Experimental design :

The response surface methodology (RSM) was applied using a commercial statistical package, Design-Expert, version 8.0.2 (Statease Inc., Minneapolis, USA), to identify optimum levels of three variables of Khoa (g), ghee residue (g) and honey (g) regarding five responses: FFA (free fatty acid), AOA (antioxidant activity), TPC, TFC and overall acceptability of antioxidant rich indigenous burfi. A three-factor and three-level Box-Behnken design (Tekindal *et al.*, 2012) consisting of 17 experimental runs including five replicates at centre point was employed. In Box-Behnken design, experiments are easier to arrange and interpret as not possible in central composite designs where the corner points were very extreme. The order of experiments has been fully randomized. The data were analyzed by multiple regressions using the least-squares method. A quadratic model was used to express the responses as a function of independent variables, where A, B, and C are coded values of temperature, concentration and feed rate. The test of statistical significance was performed on the total error criteria, with a confidence level of 95%.

The significant terms in the model were found by analysis of variance (ANOVA) for each response. The adequacy of the model was checked by calculating the R^2 and adjusted-R^2. The numerical optimization techniques of the Design-Expert software were used for the simultaneous optimization of the multiple responses. The desired goals for each variable and response were chosen. All of the independent variables were kept within range, while the responses were either maximized or minimized.

RESULTS AND DISCUSSION

Present research work was undertaken to prepare an indigenous food product "burfi" using underutilized dairy industry by-product ghee residue and replacing sucrose with honey, containing appreciable amount of antioxidants, phenolic and flavonoid content with many health beneficial effects. The amount of ingredients was optimized by using box behckan design using RSM. The range of ingredients was finalized on the basis of preliminary preparation of burfi. Storage study of prepared burfi using optimized amount of ingredients was carried out to observe the effect on quality of burfi in terms of FFA content and overall acceptability.

Ghee residue preparation and its compositional analysis :

Direct cream method was used to obtain the ghee residue as by product after getting ghee as major product. The yield of ghee obtained was 132.1 g/kg of raw material used. The amount of ghee obtained was 445 g and remaining amount of weight was considered evolved as water vapor during ghee and ghee residue preparation. The prepared ghee residue was subjected to proximate analysis. The values obtained for moisture, fat, protein and ash content were 25.95%, 54.65%, 18% and 2.96% on dry basis, respectively. The observations indicated the high nutritional value of ghee residue and were in close agreement with other researchers (Deodhar, 1986

and Arumugam *et al*. (1989).

Optimization of amount of ingredients for burfi preparation :

Box-Behnken design was used to optimize the amount of ingredient required to prepare the antioxidant rich burfi. The amount of khoa, ghee residue and honey were considered as independent variable and FFA, AOA, TPC, TFC and overall acceptability were considered as dependent variables which are shown in Table 1.

The effect of independent variables (amount of khoa, ghee residue and honey) on dependent variable (FFA, AOA, TPC, TFC and overall acceptability) has been discussed as following:

Free fatty acid value :

FFA is considered as index for the shelf life of any food item containing lipids. As the FFA content increases in the food item, off flavor starts developing and at extreme level it approaches to rancidity. The effect of ghee residue was observed greatest on FFA content followd by khoa and honey had the least effect as shown in Fig. 1a. The value of FFA content varies in between 0.99-1.12% with an average value of 1.06% in the finished product (Table 1).

Antioxidant activity :

The AOA of prepared burfi varied in the range of 17 to 29 mg/g product with an average value of 21.61 mg/g product. The findings reveal that these values have a potential contribution in daily requirement of antioxidant for an adult and same had been mentioned in studies conducted on the average consumption of antioxidants by people in USA (Chun *et al*., 2005). The amount of antioxidant increases with increase in amount of honey in the burfi preparation which can be seen clearly in 3D response surface plots (Fig. 2b). The increase in antioxidant with the increase in honey concentration may

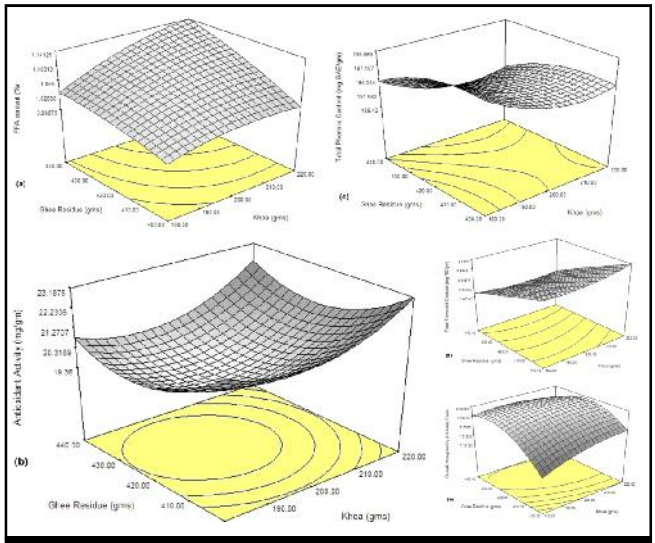

Fig. 1 : Response surface 3D plots showing effect of Khoa and ghee residue on dependent variables

Table 1 : Box Behnken design showing independent and dependent variables

Sr. No.	Independent Variables			Dependent Variables				
	Khoa (g)	Ghee residue (g)	Honey (g)	FFA (%)	AOA(mg/g)	TPC (mg GAE/g)	TFC (mg RE/g)	Overall acceptability (Hedonic Scale)
1.	200	400	170	1.04	29	210	12	7.02
2.	180	400	150	0.99	22	198	9	7.06
3.	200	400	130	1.01	18	188	7	7.12
4.	200	440	170	1.12	27	206	11	7.36
5.	200	420	150	1.07	20	192	8.5	7.98
6.	200	420	150	1.08	19	194	9	7.96
7.	220	420	170	1.09	28	208	11.7	7.68
8.	180	440	150	1.03	21	192	8.5	7.77
9.	200	420	150	1.06	19.5	196	9	7.97
10.	180	420	130	1.02	17	201	6.7	7.39
11.	180	420	170	1.04	27	210	11.8	7.48
12.	200	420	150	1.07	20	195	8.7	7.98
13.	220	400	150	1.02	23	197	9.2	7.61
14.	200	440	130	1.11	17	184	6.8	7.42
15.	220	440	150	1.14	22.5	192	8.5	7.81
16.	220	420	130	1.1	18.5	187	6.9	7.24
17.	200	420	150	1.08	19	193	8.5	7.96

be attributed to the contribution of antioxidant compounds from honey itself. Moreover, many complex chemical reactions during heat processing of burfi may also responsible for the formation of new antioxidant compounds which mask up the amount of antioxidant degraded during heat processing.

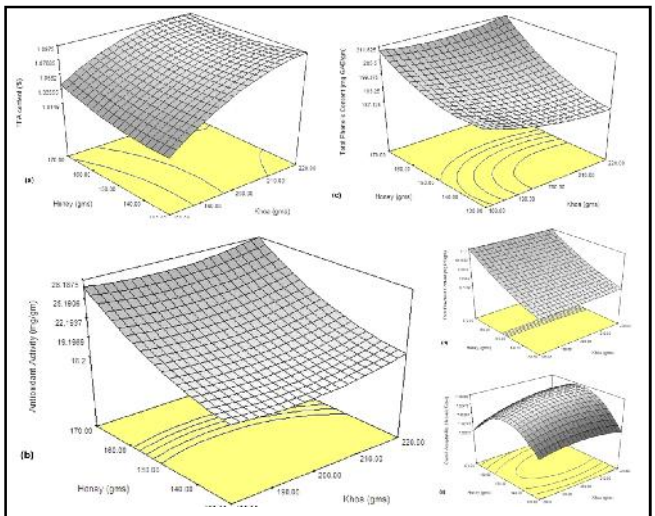

Fig. 2 : Response surface 3D plots showing effect of Khoa and honey on dependent variables

Total phenolic and flavonoid content :

Total phenolic content (TPC) of powder ranged from 184 to 210 mg GAE/g product with an average value of 196.64 mg GAE/g burfi (Table 1). Total flavonoid content (TFC) of antioxidant rich ghee residue based burfi ranged from 6.7 to 11.8 mg RE/g burfi with an average value of 8.98 mg RE/g burfi (Table 1). Regression model fitted to experimental results indicated that honey concentration had highly significant effect on both TPC and TFC which was clearly seen in 3D response surface plots (Fig. 2c and 2d). It is also evident from response surface plots that khoa has the least effect on TPC and TFC while ghee residue has negative linear effect on TPC and TFC (Fig. 1c and 1d).

Overall acceptability :

Overall acceptability score of antioxidant rich ghee residue based burfi ranged from 7.02 to 7.98 with an average value of 7.57 (Table 1). Ghee residue has the most significant effect on overall acceptability of the burfi followed by honey and khoa (Fig. 1e and 2e). This may be due to the synergistic effect of pleasant flavor of fat and sweet taste of honey which increases the score of overall acceptability.

Optimization :

Optimum amount of ingredients required for the preparation of antioxidant rich indigenous burfi were determined to obtain the criteria; minimum FFA content, Maximum AOA, TPC, TFC and overall acceptability. In this study, the optimization was applied for selected ranges of khoa, honey and ghee residue amounts as 180–220 g, 130-170 g, and 400-440 g, respectively. By applying desirability function method, the formulations was obtained for the optimum covering criteria with desirability value of 0.757. The formulations were obtained for the optimum amount of ingredients covering the criteria as 220 g for khoa, 405.29 g for ghee residue and 168 g for honey. At this point, FFA, AOA, TPC, TFC and overall acceptability were predicted as 1.04%, 28.56 mg/g, 210 GAE/g burfi, 11.60 RE/g burfi, and 7.5, respectively, while experimental values were obtained as 1.05%, 28.49%, 209 GAE/g burfi, 11.59 RE/g burfi, respectively. Interestingly, only small deviations were found between the experimental and predicted values. Therefore, the models obtained in this study could be used to optimize the amount of ingredients required for preparation of ghee residue based antioxidant rich burfi.

Storage study :

Storage study of indigenous antioxidant rich prepared burfi using standardized amount of ingredients was packed in polyethylene bags and glass containers. The samples were drawn at different intervals of 0, 10, 20, and 30 days and analyzed for free fatty acid value. The observations have been mentioned in Table 2.

FFA was determined during storage of ghee residue based burfi when packed under both packing materials *viz.*, polyethylene pouches and glass containers and data is presented in Table 5. Significant ($P<0.05$) increase in free fatty acid during storage was observed in burfi stored in both polyethylene pouches and glass containers. Burfi packed in polyethylene pouches and glass containers increased from 1.13 to 1.04% and 0.93 to 1.11%, respectively during 30 days storage. Significant difference ($P<0.05$) was observed in free fatty acid content of burfi stored in both polyethylene pouches and glass containers even after 10 days storage, however, significant ($P<0.05$) difference in free fatty acid was observed between free fatty acid content of burfi stored

Table 2 : Change of FFA (%) during storage of ghee residue based antioxidant rich indigenous burfi				
Storage period (Days)	0	10	20	30
Burfi Packed in Polyethylene bags (Free fatty acid %)	1.03	1.09	1.18	1.28
Burfi packed in glass containers (Free fatty acid %)	1.03	1.11	1.21	1.33

Table 3: Sensory evaluation scores of ghee residue based antioxidant rich indigenous burfi						
Number of days	Color and appearance	Flavor	Mouth feel	Taste	Texture	Overall acceptability
0	7.68	6.89	7.65	6.75	7.55	7.35
10	7.64	6.56	7.15	6.57	7.41	7.09
20	7.21	6.59	7.01	6.05	7.25	7.01
30	7.05	6.57	6.95	5.95	7.01	6.85

in polyethylene pouches and glass containers after 10 days of storage.

Increase in FFA was within desirable limits when compared to BIS (1966) as per IS: 3508 rules maximum limit for FFA is 3.0 in ghee and all three ghee residue products showed lower levels in comparison to PFA recommendations.

Sensory analysis :

The prepared indigenous antioxidant rich burfi was subjected to sensory quality for individual characteristics *viz.*, color, appearance, flavor, mouth feel, taste, texture and overall acceptability and data is presented in Table 3. A decreasing trend in sensory score was observed in all individual sensory characteristic but even after 30 days of storage, the product was liked slightly to very much. Verma and De (1978) optimized the method for the preparation of burfi from ghee residue and got a sensory score of 8.0 in comparison of 7.0 for the burfi prepared using ghee in the lab. After the preparation on 0^{th} day, the color and appearance characteristic got the highest while taste least score. The overall acceptability was decreased from 7.35 on 0^{th} day to 6.85 sensory score on 30^{th} day. Overall, the sensory data observed in Table 3 indicates that the prepared burfi was still eatable and got the highest score of 7.05 for color and appearance.

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