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Optimization of Fish Based Extruded Product by Twin Screw Extrusion using Response Surface Methodology and Shelf life Evaluation

C. K. Kamalakanth, C. G. Joshy, Jones Varkey[#], C. N. Ravishankar and T. K. Srinivasa Gopal*
ICAR-Central Institute of Fisheries Technology, P. O. Matsyapuri, Cochin - 682 029, India

Abstract

Effect of extrusion conditions like temperature, screw speed, and moisture content of the fish incorporated extruded snack on specific mechanical energy (SME) and texture were evaluated and optimized by using response surface methodology. A central composite design with 20 runs was formulated for the experiment and the quadratic model was most suitable for explaining the changes of dependent variables with respect to independent variables. It was found that as the temperature increased, SME and texture decreased; whereas, SME was directly proportional with screw speed. There was a decrease in the SME and increase in texture as the moisture content increased. Maximum desirability value of 0.873 revealed that the screw speed of 380 rpm at a temperature of 140°C and 17% moisture content were optimum for the development of fish incorporated extruded snack and values for SME and texture were 251.26 KJ kg^{-1} and 9.86 N, respectively. Storage study of fish-incorporated extruded snack developed at optimized conditions revealed that thiobarbituric acid and texture were increased with storage period. Similarly, redness of the sample increased while whiteness and yellowness of the sample were decreased with storage. At ambient temperature (28±1°C), the product had a shelf life of 4 months.

Keywords: Fish, twin screw extrusion, response surface methodology, specific mechanical energy, texture, storage

Introduction

Demand for fish and fishery products are increasing in India and reduction in post harvest losses can make a major contribution to satisfying this demand. In recent years, consumers prefer ready to eat products with intricate shapes and small sizes which is beyond the capabilities of single screw extruder; while, twinscrew extruders are capable to fulfill these needs. They give more flexibility and better control over the product and process parameters. The term twin screw applies to extruders with two screws of equal length placed inside the same barrel. Twin screw extruder is a better choice for producing a wide variety of high value products at low volume because the screw speed is an influential factor while extruding. Due to the high residence time and good conveyance, twin-screw extruders are better choice for the smooth extrusion of high moisture content materials. Tropical fishes comprising of several species in varying sizes impose several problems in their handling and preservation and this is one of the reasons for substantial wastage of landed fish. High level of fish meat inclusion, ultra small product size and shape etc. are some advantages of the production of value added products with twin-screw extruders. Fish protein is considered as a good source of lysine with high biological value (Balachandran, 2001). Use of fish mince with cereals for extrusion process will enable production of shelf-stable products at ambient temperature. Co-extrusion of fish with cereals offers a possibility of producing shelf stable foods with good nutritional quality (Clayton & Das, 1982). Similar works reported on successful extrusion of fish and fish powder with cereal flour as reported by Gogoi et al., 1996; Suknark, 2001; Pansawat et al., 2008. Extrusion cooking technology has limitless application in the processing of cereal based foods and other materials.

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* *E-mail: tksgopal@gmail.com*
Present Address:
[#] *Export Inspection Council of India, Mumbai - 400 004, India*

Modeling of extrusion processing involves consideration of process and system parameters, product properties and explores the variability in the experimental data (Filli et al., 2013). Response surface methodology (RSM) explores the relationships between explanatory variables and response variables (Montgomery, 2001). RSM helps to control the cost and time of the experiment by reducing the number of experimental runs needed to provide sufficient information for statistically acceptable result and also optimized process parameters for multiple responses. In the present study, the extrusion parameters of fish incorporated extruded product was standardized with parameters like temperature of extrusion (°C), moisture content of the feed material and the screw speed (rpm) of the extruder on specific mechanical energy (Jg^{-1}) and texture (N) measured in terms of shear strength of the product and to evaluate the shelf life of the developed product at optimized condition were evaluated.

Materials and methods

Minced fish meat from pink perch (*Nemipterus japonicus*), rice flour, and chickpea flour or besan (Bengal gram flour) in the ratio of 15, 75 and 10% were the ingredients used for the flour mixture prior to extrusion. These ingredients were mixed in a bowel chopper. Moisture was varied between 15 to 19% by addition of water and conditioned for 30 min at 24°C.

Extrusion was carried out in co-rotating twin screw extruder (Model EB-10, M/s Basic Technology Pvt. Ltd., Kolkata, India). Temperature of extrusion (100 to 140°C), moisture (15 to 19%), screw speed (340 to 380 rpm) were the independent variables and it effect on the dependent variables like specific mechanical energy (Jg^{-1}) and texture of the product in terms of shear strength (N) on the extrusion was evaluated. These dependent variables are two important properties for extruded products (Meng et al., 2010). Optimization of process parameters was carried out by Response Surface Methodology using Design Expert software. The following parameters with ranges selected as independent variables: Temperature of extrusion (X_1 °C: 100 to 140); Moisture (X_2%: 15 to 19) and Screw speed (X_3rpm: 340 to 380). A central composite design with 20 runs was formulated for the experiment.

Statistical Modelling was carried out by second order response surface regression model to predict and optimise the response variables (Specific mechanical energy and Texture) in terms of temperature of extrusion, moisture content of the feed material and optimum screw speed of exturder. The quadratic model given below was used to break up the total variability into variability due to linear, quadratic and interaction effect of process parameters and error (Myers & Montgomery, 2002).

$$Y = \beta_o + \sum_{i} \beta_i x_i + \sum_{ii} \beta_{ii} x_{ii}^2 + \sum\sum_{i\ j,i<j} \beta_i \beta_j x_i x_j + e,\ i \neq j \quad (1)$$

Where, "Y" is response variable, "β_o" is intercept, "β_i" is linear regression coefficients, "β_{ii}" is quadratic regression coefficients, "β_{ij}" is interaction regression coefficients and "*e*" is error term. Ridge analysis was carried out to predict the response variable at different radius of the design region. The goodness of fit of the model was evaluated based on the R^2 value. The changes in the dependent variables were evaluated based on the evaluation of ridge score and response surface plot of the response variables. Desirailbty function score was computed for multiple response optimization and based on the desirabilty score an optimum combitaion of temperature of extrusion, moisture and screw speed was selected. All the statistical analysis was done using Design Expert 7.

The hardness of the product was determined by measuring shear force required to break the extrudates was carried out using a Food texture Analyzer (Lloyd Instruments, Model LRX Plus) which is referred to as texture here. The probe used was Warner-Bratzler shear attachment. Uniform samples were used for the analysis with a trigger force of 0.5 kg with a load cell of 50 N. Texture analysis were performed using Nexygen Software (Gogoi, et al., 1996).

Specific mechanical energy (SME) was calculated by using the following formula according to the method described by Gogoi, et al., (1996). SME= [net torque x 3.95] KJ kg^{-1}. Around 25 g of the product was packed in 12 μ metallised polyester film laminated with 60μ polythene. The pouches of the dimension 15 cm X 10 cm were vacuumised, flushed with nitrogen, heat-sealed and were kept at ambient temperature (28±1°C) for storage studies. The physical properties of the packaging material were also carried out. The tensile strength and elongation at break was determined using Universal Testing Machine (Lloyd instruments LRX plus, UK)

(IS: 2508-1984). The heat seal strength was determined by measuring the force required to pull apart the pieces of sealed film (ASTM, 1973). WVTR was analyzed by the method of IS: 1060 Part II (1960). Oxygen permeability of the film and tray was carried out using gas permeability apparatus (Gas and steam permeability, AtsFaar, Societa' Per Azioni, Milano, Italia) (ASTM, 1982). The shelf life of the product was determined by analyzing parameters like instrumental colour, texture, thiobarbituric acid (TBA) value (Tarladgis et al., 1960) and sensory analysis (Meilgaard et al., 1999) to determine the shelf-life of the product. Sensory analysis was carried out using 10 point hedonic scale with a score of less than 4.0 as margine for acceptability. Storage studies were conducted up to 6 months at ambient temperature (28 ± 2°C).

Results and Discussion

The experimental results of SME (KJ kg^{-1}) and Texture (N) along with the actual values of independent variable is given in Table 1. It was found that the specific mechanical energy ranged between 221.19 and 324.62 KJ kg^{-1} where as Texture ranged between 5.38 and 18.78 N.

The approximation of response variable as a function of input parameters was done using Quadratic model and fitted well to SME and Texture data with an R^2 value of 0.78 for both the responses. The linear effect of temperature and moisture was found to have significant effect on response variables at 5% level of significance. The regression coefficients of the fitted model along with R^2 value is given in Table 2.

Table 1. Results of Central Composite Experimental design in actual levels for extrusions

Run	Factor 1 Temp.(°C)	Factor 2 Moisture (%)	Factor 3 Screw speed (rpm)	Response 1 SME (kJ kg^{-1})	Response 2 Texture (N)
1	120.00	17.00	360.00	248.33	14.31
2	86.36	17.00	360.00	267.06	15.19
3	100.00	19.00	380.00	246.47	15.39
4	140.00	19.00	380.00	227.83	10.84
5	100.00	19.00	340.00	259.56	18.78
6	153.64	17.00	360.00	224.31	9.28
7	120.00	17.00	326.36	230.32	16.48
8	140.00	15.00	380.00	301.66	5.38
9	140.00	19.00	340.00	221.19	9.46
10	120.00	17.00	360.00	254.34	12.98
11	120.00	17.00	360.00	240.56	18.49
12	120.00	13.64	360.00	284.37	6.79
13	100.00	15.00	340.00	324.62	8.66
14	120.00	17.00	360.00	262.46	10.53
15	120.00	20.36	360.00	229.97	13.7
16	120.00	17.00	360.00	248.33	12.58
17	100.00	15.00	380.00	319.55	9.91
18	120.00	17.00	393.64	264.06	15.6
19	140.00	15.00	340.00	301.6	8.95
20	120.00	17.00	360.00	251.16	13.5

Table 2. Regression coefficients of fitted model with R^2 values

Response	Intercept	X_1 = Temperature	X_2= Moisture	X_3= Screw Speed	X_1* X_2	X_1* X_3	X_2* X_3	X_1^2	X_2^2	X_3^2	R^2
SME	2696.08	-4.67134	-71.03385	-7.97210	-0.05031	+7.76875E-003	-4.50000E-003	+8.78419E-003	+1.90084	+0.010114	0.78
Texture	-56.83	+0.96754	+18.33192	-0.79773	-0.030094	-1.56250E-005	+9.68750E-004	-2.30420E-003	-0.40746	+1.05814E-003	0.78

Fig. 1 shows the relationship between moisture and temperature with specific mechanical energy. It was found that as the moisture increases the SME decreased. When the temperature of extrusion increased there was a decrease in the SME. Screw speed and barrel temperature had significant effect on the SME of extruder. Fig. 2 shows the relationship between barrel temperature and screw speed of the extruder on SME. It was found that as the temperature increased the SME decreased and as the screw speed increased there was a significant increase ($p<0.05$) in the SME. Fig. 3 shows the relationship between moisture and screw speed of the extruder on the SME. There was a decrease in the SME as the moisture content increased. As the screw speed increased the SME increased slightly. The effects of ingredient properties and processing condition system parameters such as motor torque, die pressure, product temperature and SME have significant effect on final product quality (Meng et al., 2010). SME was correlated with extrudate properties such as expansion, density and texture characteristics (Altan et al., 2008; Dogan & Karwe, 2003; Onwulata, 2001).

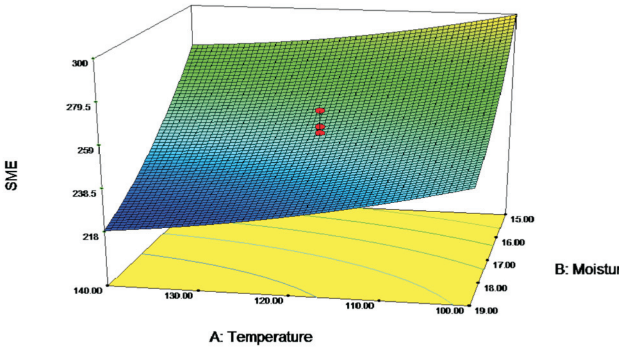

Fig. 1. Response surface plot of specific mechanical energy as a function of barrel temperature and moisture content of feed

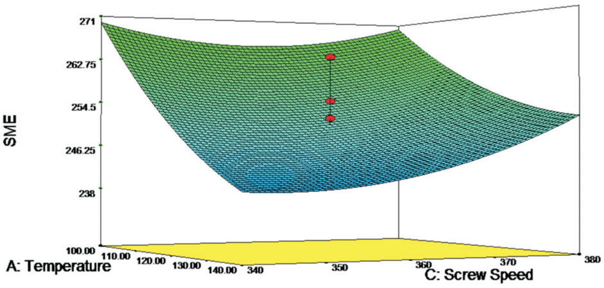

Fig. 2. Response surface plot of specific mechanical energy as a function of barrel temperature and screw speed of extruder

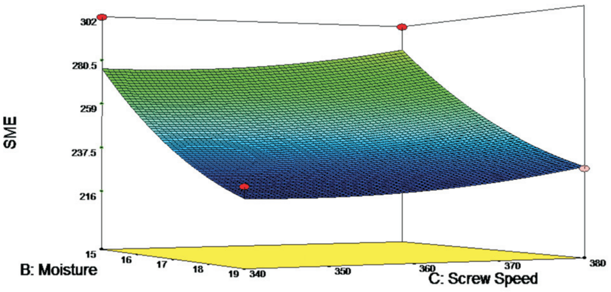

Fig. 3. Response surface plot of specific mechanical energy as a function of moisture and screw speed of extruder

Fig. 4 shows the relationship between moisture and temperature with shear strength (texture) of the extruded product. In this study, as temperature increased there is slight decrease in the texture of the extruded product. As the feed moisture increased, response surface plot showed an increase in the textural hardness in terms of shear strength. Previous studies also reported that the hardness of extrudates increased as the feed moisture content increased (Renu et al., 2015; Mayyawadee et al., 2012; Ilo & Berghofer, 1999; Faubion & Hoseney, 1982). Feed moisture and barrel temperature were found to have the most significant effect on extrudate hardness (Qing-Bo et al., 2006). Fig. 5 shows the relationship between screw speed and barrel temperature on the texture of the extruded product. As the temperature and screw speed increased there was a decrease in the texture of the product. This is because of low residence time of the feed inside the barrel of the extruder. For good expanded product, high barrel temperatures combined with low screw speeds were suitable (Oke et al., 2013). Fig. 6 shows the response surface plot of texture as a function of screw speed and moisture content of feed. It was found that as the moisture content increased, there was an increase in the texture of the product. Low moisture is usually ideal for developing puffed snack. When the screw speed increased there was a decrease in the texture of the product. This also accounts for the low residence time of the fed material inside the extruder. Feed moisture was found to be the main factor affecting extrudate density and expansion (Faubion & Hoseney, 1982).

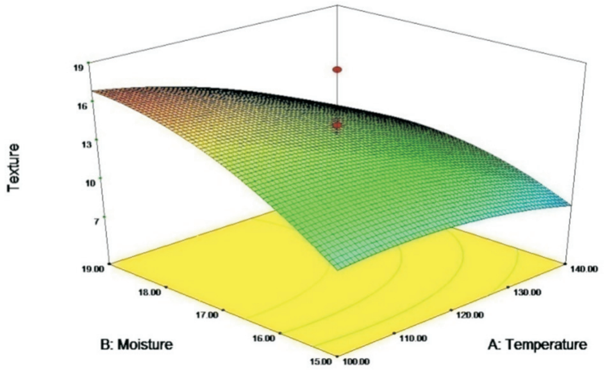

Fig. 4. Response surface plot of Texture as a function of moisture content of feed and barrel temperature

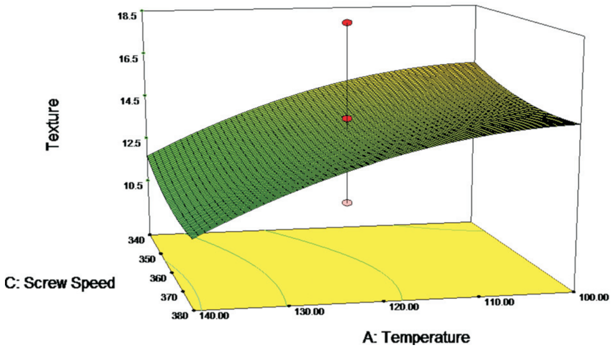

Fig. 5. Response surface plot of Texture as a function of screw speed and barrel temperature

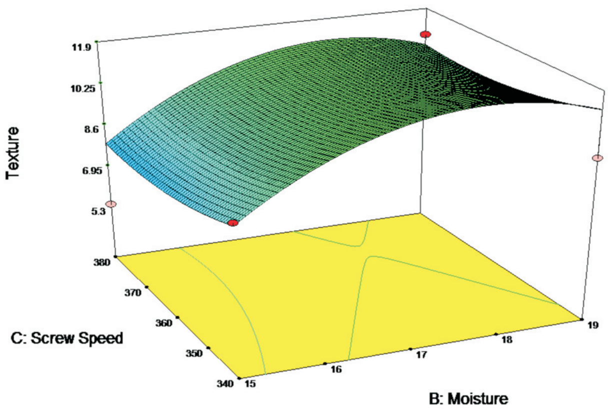

Fig. 6. Response surface plot of Texture as a function of screw speed and moisture content

Multiple responses were optimized by calculating the desirabilty function score by keeping specific mechanical energy in the range 221.19 to 324.62 KJ kg^{-1} and minimum Texture. Based on this, the optimum combination of input parameters were temperature 140°C, moisture content 17% and screw speed 380 rpm and the corresponding desirability score was 0.871. The predicted values for specific mechanical energy and Texture at the optimum combinations were 283.76 KJ kg^{-1} and 7.83 N respectively. Graphical representation of desirability score by varying temperature and moisture is given in Fig. 7.

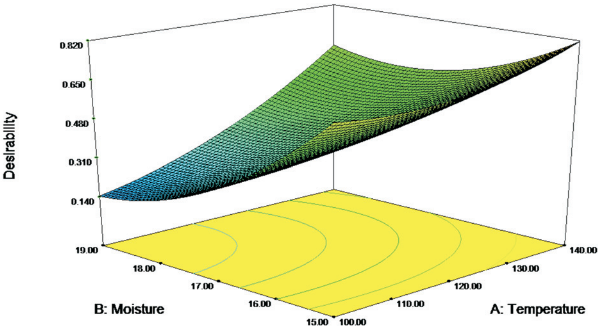

Fig. 7. Desirability function in terms of moisture and barrel temperature of the extruded product

Table 3 shows the physical properties of the packaging material. It was found that the packaging material used for the storage of extruded fish incorporated snack had good tensile and heat seal strength. It has good elongation of break at both machine as well as cross direction. The water

Table 3. Physical properties of the packaging material (12µ metalized polyester film laminated with 60µ polythene)

Physical properties	Value
Tensile strength – Machine Direction	283 ± 0.71 kg cm^{-2}
Tensile strength – Cross Direction	260 ±0.53 kg cm^{-2}
Elongation at break - Machine Direction	43±0.28 %
Elongation at break – Cross Direction	63±0.18 %
Heat Seal Strength - Machine Direction	184± 0.61 kg cm^{-2}
Heat Seal Strength – Cross Direction	172± 0.58kg cm^{-2}
Water Vapour transmission rate	1.40 ± 0.06 g m^{-2} 24 h^{-1} at 37°C and 90±2% RH
Oxygen transmission rate	5.5 ± 0.07 cc m^{-2} 24 h^{-1} atm^{-1} 20°C

vapour transmission rate of the packaging material was 1.40 g m^{-2} 24 h^{-1} at 37°C and 90±2% RH and oxygen transmission rate of 5.5 cc m^{-2} 24 h^{-1} atm^{-1} 20°C shows that the packaging material has good barrier to oxygen and water vapour.

Table 4 shows the proximate composition of the extruded product. It was found that the final product had a moisture content of 6.8%. The protein content of the sample was 23.43%. The higher protein content of the product is due to the incorporation of the fish mince to the cereal flour. Dileep et al. (2010) also reported increase in protein content of extruded product by incorporating ribbon fish and rice flour. Fat content of the developed product was 1.15%. The mineral content in terms of ash content was 2.44 %. The increase in the ash content mainly accounts for the presence of bones present in the fish mince during mincing process. Carbohydrate content was 66.18%.

Table 4. Proximate composition of the extruded product

Parameter	Percentage
Moisture	6.8±0.07
Protein	23.43±0.48
Fat	1.15±0.12
Ash	2.44±.03
Carbohydrate	66.18±0.19

Table 5 shows the evaluation of shelf life results during each month of storage. TBA value, an index of secondary lipid oxidation, showed slight increasing trend after the initial period. The initial TBA value was 0.16 mg malonaldehyde kg^{-1}. It increased to 0.73 mg malonaldehyde kg^{-1} on 3 months of storage. On further analysis up to 6 months of storage, the value increased to 0.94 mg malonaldehyde kg^{-1} sample. This may be due to permiablity of the packaging material. However, TBA values in the range 1–2 mg malonaldehyde kg^{-1} sample is usually taken as the limit of acceptability (Goulas & Kontominas, 2007). The result is well below the limit of acceptability.

Texture is an important physical property of the extruded product. The peak force required to break the sample was 9.04 N. As the storage period increased, there was a significant increase ($p<0.05$) in the hardness of the sample. This mainly accounts for the permeability of the packaging material during storage. As the crispiness was lost during storage, the force required to break the sample also increased. The hardness values were 16.34 N on 3rd month of storage. It increased to 22.31N on 6 month of storage.

Colour is also an important factor for acceptability of any product. Initial colour values of L*(lightness), a*(redness) and b*(yellowness) values were 85.67, 0.96, 14.71, respectively. The lightness decreased during storage period. The L* value decreased to 84.03 and 83.79 on 3 and 6 months of storage study. There was a corresponding slight increase in the redness of the sample. The values were 1.26, 1.71 on 3 and 6 month of storage. The b*value (yellowness) of the sample also decreased during the storage period. The values decreased from 12.28 to 11.58 on 3 and 6 month of storage.

Organoleptic evaluation revealed that initially, there was a good overall acceptability score of 9.2. As the storage period increased, there was a decrease in the

Table 5. Shelf life evaluation result during each month of storage

Sampling interval	TBA (mg malonaldehyde kg^{-1})	Texture (Shear strength N)	Colour			Sensoryscore
			(L*)	(a*)	(b*)	
Initial	0.16±0.01	9.045±0.23	85.67±0.14	0.96±0.01	14.71±0.03	9.2±0.01
1st month	0.22±0.02	12.22±0.09	84.62±0.11	1.01±0.01	14.29±0.02	8.5±0.02
2nd month	0.62±0.01	14.69±0.03	84.22±0.18	1.04±0.02	13.82±0.03	7.6±0.01
3rd month	0.73±0.01	16.24±0.34	84.03±0.12	1.26±0.01	12.28±0.01	6.5±0.01
4th month	0.85±0.03	18.32±0.09	83.93±0.06	1.37±0.03	12.17±0.01	6.0±0.01
5th month	0.88±0.01	20.25±0.08	83.87±0.12	1.43±0.01	11.70±0.03	3.9±0.01
6th month	0.94±0.01	22.31±0.26	83.79±0.09	1.71±0.02	11.58±0.02	3.5±0.02

color as well as the texture of the product. The sensory score reduced to 6.5 after 3 months of storage. This further reduced to 3.9 on 5th month of storage. The decrease in the overall acceptability was mainly due to the decrease in the crispiness of the sample. The minimum limit of acceptability was given a score of 4. Other factor responsible for the decrease in the overall acceptability was colour and increased hardness of the sample due to storage. Permeability of the packaging material was also responsible for increased hardness. Due to loss of crispness, the entrusted product was acceptable upto 4 months.

In the present study, optimization of process parameters for extrusion of fish incorporated extruded cereal snack was carried out by response surface methodology. It was found that specific mechanical energy decreased significantly as the moisture content and barrel temperature of the extruder increased. As the barrel temperature increased, there was a slight non-significant decrease in the texture of the product. As the moisture content of the feed increased, the hardness got increased significantly ($p<0.05$). Quadratic model was best suited model for explaining the changes in feed moisture and temperature of extrusion. A desirability function of 0.873 was observed for optimum temperature of 140°C with a moisture content of 17% and a screw speed of 380 rpm. The specific mechanical energy was found to be 251.26 KJ kg^{-1} and texture was 9.86 N. Proximate composition revealed that there was high percentage of protein content in extruded product. Storage studies indicated that extruded products are acceptable up to 4 months at ambient temperature.

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