

## DEVELOPMENT OF TEXTURED DEFATTED SUNFLOWER MEAL BY EXTRUSION USING RESPONSE SURFACE METHODOLOGY

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Sunflower (PSH 569) was used to obtain textured defatted meal. Proximate analysis, water absorption index (WAI), water solubility index (WSI), fat absorption capacities (FAC), foaming capacity (FC), and bulk density (BD) were determined. The objective of the study was the optimization of extrusion conditions for production of textured defatted sunflower meal using response surface methodology (RSM) by evaluating functional properties. It was dried, grinded, and sieved to eliminate hull and fibre. Numerical optimization provided eight solutions with desirability value varying from 0.81 to 0.82. Range of predicted values of FAC (80.96–90.49), WHC (1.95–2.12), WSI (3.22–3.36), WAI (2.84–3.08), bulk density (0.31–0.36), and foaming capacity (14.39–16.30) were used for numerical optimization. Best extrusion conditions were 16.36% feed moisture, 300 r.p.m. screw speed, and 149.40 °C barrel temperature. Textured sunflower defatted meal was prepared using the above optimized conditions.

**Keywords:** extrusion, sunflower meal, texturization, response surface methodology

Sunflower (*Helianthus annuus* L.) is one of the world's leading oilseed crops, second only to soybean for total oil production. Industrial sunflower meal cannot be directly used as a food because of their high fibre content, and because of the presence of some undesirable constituents (such as hulls, polyphenolic pigments, etc.), or precursors of toxic compounds (glucosinolates, etc.). Defatted flours, a by-product of the oil industry, constitute an important source of proteins. The sunflower defatted meal is underused, being almost exclusively employed for animal feeding in spite of its high content of highly digestible proteins with an important content of essential amino acids (except for lysine and sulphur-containing amino acids). The high concentration of phenolic compounds of which the majority is chlorogenic acid with small amounts of caffeic acid (GONZALEZ PEREZ & VEREIJKEN, 2008) is the main reason for the underutilization of sunflower oil cake. In addition, these compounds reduce protein solubility and cause unwanted organoleptic characteristics (PRASAD, 1990). Recently, sunflower meal was used to develop protein concentrates and isolates, but both are deficient in fibre content and the cost is higher than of textured defatted meal. RSM has important application in the design, development, and formulation of new products, as well as in the improvement of existing product design. It defines the effect of the independent variables, alone or in combination, on the processes.

Providing safe, nutritious, and wholesome food for poor and undernourished populations had been a major challenge for the developing world. More specifically, protein-energy malnutrition is among the most serious problems developing countries are facing today (BOYE et al., 2010). It had been estimated that 800 million malnourished people exist in least developed countries. Development of nutritious foods has been suggested by FAO to combat malnutrition among children. Consumption of high quality proteins is essential for maintaining good health. Due to the price and relative scarcity of food obtained from animals, it is

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necessary to find or develop new alternative products that offer both better quality and a greater quantity of proteins. An important reason for the increased acceptance of vegetable proteins, such as textured soy protein (TSP), is their low cost (SINGH et al., 2008). In addition to retexturing and restructuring vegetable food proteins, the extrusion cooking system performs several other important functions, i.e. it denatures proteins. Proteins are effectively denatured during the moist, thermal process of extrusion. Denaturation of protein lowers solubility, renders it digestible, and destroys the biological activity of enzymes and toxic proteins. The objective of the present study was to obtain a textured protein from defatted sunflower meal by extrusion, to optimize the process using response surface methodology, and to evaluate the functional properties of the product produced.

## 1. Materials and methods

### 1.1. Raw material

For this study, the short duration cultivar of sunflower PSH 569, developed by Plant Breeding and Genetics Department of Punjab Agricultural University, maturing within 90–100 days and widely grown in Punjab, India, was selected.

### 1.2. Chemical composition

*1.2.1. Moisture.* Two grams of sample was dried in a clean, dry, and pre-weighed moisture dish at  $130 \pm 1$  °C for 1 h, cooled in desiccator and weighed. The moisture loss was calculated and expressed in percentage (AACC, 2000).

*1.2.2. Protein.* Sample (weighed) was digested in Kjeldahl flask with digestion mixture (copper sulphate and potassium sulphate in 1:10 ratio) and concentrated  $H_2SO_4$  (20 ml) till light green colour and cooled. Ammonia released by distillation of digested sample with saturated NaOH (80 ml) was captured in 0.1 N HCl to calculate percent nitrogen ( $N_2$ ). The protein content was calculated as  $\% N_2 \times \text{factor}$ . The factor of 5.7 was used for calculation (AACC, 2000).

*1.2.3. Fat.* Weighed sample was taken in thimbles and extracted using Soxhlet apparatus with petroleum ether for 16 h. Ether was recollected and the round bottom flask was weighed after fat extraction. The results were expressed as percent fat (AACC, 2000).

*1.2.4. Crude fibre.* Crude fibre of sample was estimated using Fibertec (Foss company). The sample was cooled and 1 g was weighed in capsules. To the large extraction cup 250–275 ml of 1.25%  $H_2SO_4$  was added, and the stand was immersed into the beaker. Acid extraction was done by boiling for 30–40 min followed by washing with hot water. Then alkali washing was done with 1.25% NaOH for the same duration followed by hot water washing. Finally, acetone washing was carried out, and the capsules were dried in an oven for 2 h at 130 °C. The sample was cooled and weighed for crude fibre estimation.

### 1.3. Oil extraction

The sunflower was cleaned and defatted using laboratory oil expeller. The meal was dried and milled into grits using Super Mill (Perten Instruments, Sweden). After that, the sample was sieved using 200 mesh screen to separate out the large particles of the seed coat.

#### 1.4. Experimental design for sunflower

Texturization of sunflower was carried out by using Clextral BC 21 twin screw extruder (Clextral, Firminy, France). RSM was used to optimize the texturization conditions with DesignExpert version 7.0 (Statease, Minneapolis, USA). The levels of each variable were established according to preliminary trials. Central composite design was to optimize the process. The operating conditions were 14–20% feed moisture, 300–500 r.p.m. screw speed, and 120–180 °C barrel temperature. Extrusion process variables (feed moisture content, screw speed, and temperature) were coded to the level of –1, 0, +1, such that one factor at a time of experimental design was as given in Table 1 (MYERS, 1971). Twenty different combinations were studied using response surface methodology to investigate the effect of these process and component variables on response variables. The statistical analysis was performed using DesignExpert software. The main advantage of RSM was that it reduced the number of experimental runs needed to provide sufficient information for statistically acceptable results. The results were analysed by multiple linear regression method, which describes the effects of variables in the models derived. Experimental data were fitted to the selected models and regression coefficients were obtained. The analysis of variance (ANOVA) tables were generated for each of the response functions. The individual effect of each variable and also the effects of interaction term in coded levels of variables were determined. Textured samples were milled into flour using cyclotec mill (Newport Scientific, Australia) and packed in suitable packaging material for further study.

Table 1. Coded levels for independent variables

Extrusion parameters	–1.682	–1	0	+1	+1.682
Moisture content (%)	11.954	14	17	20	22.046
Screw speed (r.p.m.)	231.800	300	400	500	568.200
Barrel temperature (°C)	99.54	120	150	180	200.460

#### 1.5. Functional properties of textured defatted sunflower meal

*1.5.1. Water absorption index (WAI).* First, 1 g of defatted flour was placed in a previously weighed 50 ml centrifuge tube containing 10 ml of distilled water, stirred homogeneously with a glass rod till it mixed properly, and centrifuged at 3000 r.p.m. for 10 min at room temperature (22 °C) using a Model T-8BL Laby™ centrifuge (Laby Laboratory Instruments, Ambala Cantt, India). The residue was weighed together with the centrifuge tube after removing water. The WAI values were expressed as gram of water absorbed/gram of defatted flour (STOJICESKA et al., 2009).

$$\text{WAI (g g}^{-1}\text{)} = \frac{\text{Weight of residue}}{\text{Sample taken}}$$

Water solubility index (WSI): The supernatant obtained from WAI method was transferred to previously weighed dish, which was placed in hot air oven for the evaporation of water. The residue was weighed.

$$\text{WSI (\%)} = \frac{\text{Weight of dry matter in supernatant} \times 100}{\text{Dry weight of sample}}$$

Fat absorption capacities (FAC): First, 0.5 g of defatted flour was placed in a previously weighed 50 ml centrifuge tube containing 10 ml of oil and rest of procedure was the same as WAI. The FAC values were expressed as gram of water absorbed/gram of defatted flour (LIN et al., 1974).

$$\text{FAC (\%)} = \frac{\text{Weight of fat absorbed by sample} \times 100}{\text{Weight of sample}}$$

*1.5.2. Foaming capacity.* First, 1 g of defatted flour was dissolved in 100 ml of distilled water. Then the suspensions were whipped at a low speed in a blender for 1 min at room temperature (22 °C). Total foam volume was recorded and foam capacity was expressed as the percent increase in volume (KABIRULLAH & WILLS, 1983).

$$\text{FC} = \frac{\text{Final foam volume} \times 100}{\text{Initial foam volume}}$$

*1.5.3. Bulk density (BD).* The Bulk densities (g ml<sup>-1</sup>) of defatted flour were determined by volumetric method. The volume of the extruded sample was measured by using a 25 ml graduated cylinder and gently tapped for 5 times (PAN et al., 1998).

$$\text{Bulk density (g ml}^{-1}\text{)} = \frac{\text{Weight of sample}}{\text{Volume displaced by sample}}$$

## 2. Results and discussion

### 2.1. Chemical composition

The moisture, fat, fibre, and protein content of defatted textured sunflower meal were 2.56%, 2.54%, 13.07%, and 43.38%, respectively. Percent increase in protein content for textured sunflower meal was 122.86% as compared to raw sunflower (19.46%). Textured flour was added to products to improve nutritional value as it increases the protein content.

### 2.2. Functional properties

*2.2.1. Fat absorption capacity.* Defatting increased the protein's solubility and its water and oil absorption capacity. The capacity of protein to absorb water and oil is determined by its polar and non-polar amino acids composition, respectively (SATHE & SALUNKHE, 1981). The quadratic model obtained from regression analysis for fat absorption capacity (FAC) in terms of coded levels of the variables was developed as follows:

$$\text{FAC} = +91.73 - 1.57 \times A - 3.34 \times B - 6.17 \times C + 0.49 \times A \times B + 4.41 \times A \times C + 4.69 \times B \times C - 0.66 \times A^2 - 5.28 \times B^2 - 1.95 \times C^2$$

where A=feed moisture content, B=barrel temperature and C=screw speed

The analysis of variance (ANOVA) for FAC of quadratic model is given in Table 2. The value of R<sup>2</sup> was found to be 0.99. Regression analysis results showed that significant negative influence of moisture, screw speed, and temperature (P<0.05) on FAC was recorded. There

was significant interaction of feed moisture with screw speed, feed moisture with barrel temperature, and screw speed with barrel temperature ( $P < 0.01$ ) on FAC. The FAC varied from 71.11 to 104.42% in protein flour (Table 3). It was observed from the regressions analysis that during extrusion-cooking, lower moisture contents increased FAC of protein flour. The increase in FAC with increasing screw speed was consistent. Defatting increased the protein solubility and water and oil absorption capacities of the meals.

Table 2. Significance on the coefficient of estimate for sunflower meal

Factors	FAC	WHC	WSI	WAI	BD	FC
Intercept of Model	91.73	2.130	3.890	3.070	0.350	14.880
A: Moisture content	-1.57	-0.050	0.065	0.033	0.009	-0.065
B: Screw speed	-3.34	-0.090	-0.051	-0.090	0.030	-0.640
C: Barrel temperature	-6.17	-0.150	-0.290	-0.130	-0.033	0.026
AB (Moisture content×Screw speed)	0.49	0.073	-0.230	-0.059	0.002	-1.540
AC (Moisture content×Barrel temperature)	4.41	0.270	0.610	0.160	0.011	-0.750
BC (Screw speed×Barrel temperature)	4.69	0.180	0.012	0.063	0.011	4.100
A <sup>2</sup> (Moisture content) <sup>2</sup>	-0.66	0.013	0.140	-0.039	-0.018	-1.430
B <sup>2</sup> (Screw speed) <sup>2</sup>	-5.28	-0.140	-0.570	-0.120	-0.002	-0.310
C <sup>2</sup> (Barrel temperature) <sup>2</sup>	-1.95	-0.026	0.034	0.001	0.006	-0.610
P-Value for lack of fit	0.84	0.470	0.190	0.080	0.350	0.230
R <sup>2</sup>	0.99	0.97	0.98	0.96	0.94	0.98

2.2.2. *Water solubility index.* WSI is often used as an indicator of degradation of molecular components (KIRBY et al., 1988). WSI measures the amount of soluble components released from the protein and other molecules after extrusion. The quadratic model obtained from the regression analysis for water solubility index (WSI) in terms of coded levels of the variables was developed as follows:

$$\text{WSI} = 3.89 + 0.065 \times A - 0.051 \times B - 0.29 \times C - 0.23 \times A \times B + 0.61 \times A \times C + 0.012 \times B \times C + 0.14 \times A^2 - 0.57 \times B^2 + 0.034 \times C^2$$

The analysis of variance (ANOVA) for WSI of quadratic model is given in Table 2. There was also significant interaction of feed moisture with screw speed and feed moisture with barrel temperature ( $P < 0.01$ ) on WSI. The WSI for textured defatted sunflower flour varied from 2.10 to 4.48% (Table 3). It was observed from the regressions analysis that during extrusion-cooking, higher moisture contents increased WSI of protein flour. The high mechanical shear caused breakdown of macromolecules to small molecules with higher solubility. Higher moisture content in extrusion process can diminish protein denaturation, which lowers WSI. The increase in WSI with increasing screw speed was consistent with the results reported by other researchers (DOGAN & KARWE, 2003).

Table 3. Effect of extrusion condition on product responses of sunflower meal (n=3)

Extrusion conditions			Responses					
Moisture content (%)	Screw speed (r.p.m.)	Barrel temperature (°C)	FAC (%)	WHC (ml g <sup>-1</sup> )	WSI (%)	WAI (g g <sup>-1</sup> )	BD (g ml <sup>-1</sup> )	FC (%)
14	300	120	104.42	2.87	4.20	3.267	0.346	15
20	300	120	91.46	1.98	3.42	3.12	0.333	19.92
14	500	120	87.13	2.1	4.48	3.15	0.383	8.49
20	500	120	76.79	1.63	3.05	2.7	0.380	6.2
14	300	180	73.63	1.589	2.22	2.585	0.235	9.05
20	300	180	78.96	1.92	4.14	3.01	0.270	9.91
14	500	180	75.75	1.654	2.83	2.654	0.319	17.9
20	500	180	82.39	2.15	3.54	2.91	0.356	13.65
17	400	150	91.28	2.21	3.96	3.021	0.342	14.13
17	400	150	92.87	2.17	3.96	3.12	0.340	14.53
17	400	150	91.15	2.07	3.86	3.052	0.353	14.69
17	400	150	91.60	2.08	3.75	3.06	0.334	15.46
17	400	150	91.82	2.19	3.85	3.089	0.350	15.34
17	400	150	91.65	2.089	3.95	3.09	0.372	15.15
11.95	400	150	92.88	2.21	4.15	2.834	0.288	10.89
22.05	400	150	86.90	2.12	4.43	3.05	0.330	10.81
17	231.8	150	82.56	1.85	2.47	2.91	0.308	14.32
17	568.2	150	71.11	1.61	2.10	2.513	0.402	13.7
17	400	99.54	96.72	2.266	4.45	3.266	0.432	13.32
17	400	200.46	75.78	1.84	3.54	2.84	0.319	13

Each measurement was done in 3 replicates

2.2.3. *Water absorption index.* The WAI measures the amount of water absorbed by the protein, starch, and other molecules and can be used as an index of gelatinization (ANDERSON et al., 1969). WAI depends on the availability of hydrophilic groups that bind water molecules. The quadratic model obtained from the regression analysis for water absorption index in terms of coded levels of the variables was developed as follows:

Equation in terms of coded factors:

$$\text{WAI} = +3.07 + 0.033 \times A - 0.090 \times B - 0.13 \times C - 0.059 \times A \times B + 0.16 \times A \times C + 0.063 \times B \times C - 0.039 \times A^2 - 0.12 \times B^2 + 0.0001589 \times C^2$$

The analysis of variance (ANOVA) for WAI of quadratic model is given in Table 2. Regression analysis results showed that screw speed and temperature had significant negative and moisture had positive linear ( $P < 0.001$ ) effect and significant quadratic effect on WAI. Interaction of feed moisture with screw speed and screw speed with barrel temperature had significant influence ( $P < 0.01$ ). The WAI of protein flour ranged from 2.59 to 3.27 g g<sup>-1</sup> (Table 3). WAI has attributed to the dispersion of starch in excess water and the dispersion is

increased by the degree of starch damage due to gelatinization and extrusion fragmentation. WAI increased with the increase in starch gelatinization. Increase in moisture content reduced the water absorption index. Moisture content, acting as a plasticizer during extrusion cooking, reduced the degradation of molecules that resulted in an increased capacity for water absorption (HAGENIMANA et al., 2006). WAI was higher for lower screw speed and lower temperature.

**2.2.4. Bulk density.** Bulk density is a very important parameter in the production of textured products. Density is a measure of how much expansion of the extrudate has occurred as a result of extrusion (ILO et al., 1999). The quadratic model obtained from regression analysis for bulk density (BD) in terms of coded levels of the variables was developed as follows:

$$\text{Bulk Density} = +0.35 + 0.009272 \times A + 0.030 \times B - 0.033 \times C + 0.0015 \times A \times B + 0.011 \times A \times C + 0.011 \times B \times C - 0.018 \times A^2 - 0.00163 \times B^2 + 0.005614 \times C^2$$

The analysis of variance (ANOVA) for bulk density of quadratic model is given in Table 2. The bulk density of protein flour ranges from 0.189 to 0.359 g ml<sup>-1</sup> (Table 3). Regression analyses indicated that bulk density decreased with decrease in moisture. Higher screw speeds might be expected to lower melt viscosity of the mix, increasing the elasticity of the dough, resulting in a reduction in the density of the extrudate (DING et al., 2005). Bulk density increased with decrease in moisture as higher water content produced extrudates denser than those produced with low water content. High density product is an indication of more uniform and continuous protein matrix with no air pockets and not spongy upon hydration, which is undesirable because it has low WAI, FAC, and foaming capacity. Such products are less acceptable because their texture is hard.

**2.2.5. Foaming capacity.** Foams were gaseous droplets encapsulated by a liquid film containing soluble surfactant protein resulting in reduced interfacial tension between gas and water. The quadratic model obtained from the regression analysis for foaming capacity (FC) in terms of coded levels of the variables was developed as follows:

$$\text{Foaming Capacity} = +14.88 - 0.065 \times A - 0.64 \times B + 0.026 \times C - 1.54 \times A \times B - 0.75 \times A \times C + 4.10 \times B \times C - 1.43 \times A^2 - 0.31 \times B^2 - 0.61 \times C^2$$

The analysis of variance (ANOVA) for foaming capacity of quadratic model is given in Table 2. The value of R<sup>2</sup> is found to be 0.98. Interaction (P<0.05) of feed moisture with screw speed, feed moisture with barrel temperature, and screw speed with barrel temperature were found significant. The foaming capacity of protein flour ranged from 6.20 to 19.92% (Table 3). Foaming capacity of textured defatted sunflower flour was lower than of soybean protein (McWATTERS & CHERRY, 1977). Regression analysis indicated that foaming capacity decreased with the increase in moisture. Foaming capacity was low at high screw speed.

### 3. Conclusions

Sunflower oil is consumed by large population of India. Large quantity of deoiled cake produced by the oil industry is used as animal feed. It could be used as human food, because



it is rich a source of protein and fibre. Sunflower contains chlorogenic acid, which is deactivated during oil extraction and extrusion. The off-flavour of defatted meal is removed during texturization by extrusion. Textured defatted meal is safe for human consumption. It was found that functional properties of the defatted sunflower meal protein has a great potential to serve as an excellent source of edible protein, owing to its high water and fat absorbing capacity. It also had higher water holding capacity, foaming capacity, and bulk density. Defatted meal due to its high content of protein has the potential to replace other protein sources. Textured defatted sunflower protein flour exhibits satisfactory functional properties and therefore has a bright prospect for application in the food industry. Although sunflower is mainly used for its oil, the data suggest that the seeds, a by-product, could provide protein for feed and food. Such uses could significantly increase the economic value of this crop. It can be used to add food value to foods of daily used.

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