

A Case Study on Maximizing Aqua Feed Pellet Properties Using Response Surface Methodology and Genetic Algorithm

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ABSTRACT

Aims: The present case study is on maximizing the aqua feed properties using response surface methodology and genetic algorithm.

Study design: Effect of extrusion process variables like screw speed, L/D ratio, barrel temperature and feed moisture content were analyzed to maximize the aqua feed properties like water stability, true density and expansion ratio.

Place and Duration of Study: This study was carried out in the Department of Agricultural and Food Engineering, Indian Institute of Technology, Kharagpur, India.

Methodology: A variable length single screw extruder was used in the study. The process variables selected were screw speed (rpm), length-to-diameter (L/D) ratio, barrel temperature (°C) and feed moisture content (%). The pelletized aqua feed was analyzed for physical properties like water stability, true density and expansion ratio. Extrusion experimental data was collected by based on central composite design. The experimental data was further analyzed using response surface methodology (RSM) and genetic algorithm (GA) for maximizing feed properties.

Results: Regression equations developed for the experimental data has adequately described the effect of process variables on the physical properties with coefficient of determination values (R^2) of > 0.95 . RSM analysis indicated WS, ER and TD were maximized at L/D ratio of 12-13, screw speed of 60-80 rpm, feed moisture content of 30-40% and barrel temperature of $\leq 80^\circ\text{C}$ for ER and TD and $> 90^\circ\text{C}$ for WS. Based on GA analysis a maximum WS of 98.10% was predicted at a screw speed of 96.71 rpm, L/D ratio of 13.67, barrel temperature of 96.26°C and feed moisture content of 33.55%. Maximum ER and TD of 0.99 and 1346.9 kg/m^3 was also predicted at screw speed of 60.37 and 90.24 rpm, L/D ratio of 12.18 and 13.52, barrel temperature of 68.50 and 64.88°C and medium feed moisture content of 33.61 and 38.36%.

Conclusion: The present data analysis indicated that WS is mainly governed by barrel temperature and feed moisture content, which might have resulted in formation of starch-protein complexes due to denaturation of protein and gelatinization of starch. Screw speed coupled with temperature and feed moisture content controlled the ER and TD values. Higher screw speeds might have reduced the viscosity of the feed dough resulting in higher TD and lower ER values. Based on RSM and GA analysis screw speed, barrel temperature and feed moisture content were the interacting process variables influencing maximum WS followed by ER and TD.

Keywords: Extrusion cooking, aqua feed properties, response surface methodology, genetic algorithm

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1. INTRODUCTION

Extrusion is a very popular and important processing operation in the food industry to produce novel products (Raiz, 2012). It is a continuous process where different food ingredients are mixed, plasticized, cooked and formed in the presence of moisture, temperature, pressure, and mechanical shear. It is a high temperature short time process used in many industries for production of multi-components products like expanded snack foods, ready-to-eat cereals, animal feeds and pet foods (Chinnaswamy and Hanna, 1988). Corn snacks were commercially produced first using extrusion cooking just after the World War-II using high shear extruders. During this past half-century food extrusion has developed from a simple pressing and forming technology into sophisticated cooking processes, which have replaced many conventional technologies in food processing (Ilo et al., 2000).

1.1 Feed Extrusion

Extrusion processing is an important feed processing operation for making novel feed products (Paton and Spratt, 1984). The advantages of pelletization of aqua feed are a) improves the digestibility due to gelatinization of starch, b) prevents separation of the ingredients during handling and transit, and c) makes pellets stable for extended periods preserving their physical integrity and chemical composition (Harper 1981; Paton and Spratt 1984; Chinnaswamy and Hanna, 1988). Singh et al. (2007) reviewed the nutritional aspects of extruded food products and concluded that beneficial effects include reduction of anti-nutritional factors, gelatinization of starch, increase in soluble dietary fiber and reduction of lipid oxidation whereas the harmful effects include changes in amino acid profile, carbohydrates, dietary fiber, vitamins and mineral content. Extrusion process variables like moisture, pressure and mechanical shear partially denature the protein and gelatinize the starch components of the aqua feed and alter the texture resulting in physiochemical changes (Friesen et al., 1992; Bhattacharya and Prakash, 1992; Camire et al., 1990). Dar et al. (2012) and Athar et al. (2006) indicated extrusion temperature significantly impacts the microstructure, textural and functional attributes of extrudate. Reactions like gelatinization of starch and denaturing of protein of the multi-component aqua feed are important for producing aqua feed pellets with high water stability, expansion ratio, bulk density and durability (Harper, 1986 and Lim and Cuzon, 1994).

1.2 Feed Physical Properties

Knowledge of the physical properties and process variables influencing them are essential for producing high quality aqua feed pellets, designing equipment's for processing facilities, and optimizing the unit operations (Rosentrater, 2006). Some key quality attributes of aqua feed pellets are water stability, bulk density, moisture content, water activity, yield stress, apparent viscosity, thermal conductivity, thermal diffusivity, heat capacity, drying behavior, and mass, bulk, and unit densities (Rosentrater and Muthukumarappan, 2006). Extrusion helps to manufacture pellets from multi-component aqua feed mixes with both animal (fish meal, squid meal, shrimp head meal and low cost fishes) and plant sources (de-oiled soy meal, de-oiled rice bran and wheat flour) with desirable physical properties like high water stability (WS), expansion ratio (ER) and true density (TD). According to Wood (1995) selecting right process conditions and inclusions of ingredients play a major role on the quality attributes of the aqua feed pellets. WS is influenced by the binding ability of the ingredients, size of the ingredients, and the extrusion process conditions. According to Ali et al. (2005) aqua feed water stability is achieved mainly through use of appropriate binding material coupled with suitable processing technology. Effiong et al. (2009) indicated that feed ingredients have a great impact on the water stability of the pellets. Rout and Bandyopadhyay (1999) and Bandyopadhyay and Rout (2001) in their studies indicated that extrusion processing of aqua feed mix produce pellets with high WS values. Reducing the size of the ingredients during extrusion can also result in high WS as the air space between

the particles is reduced and prevent water or moisture to seep through the fine cracks of the pellets. In general lower WS values results in disintegration of pellets quickly and facilitate faster leaching of the nutrients and micronutrients, which undergo oxidation-reduction process causing water pollution and deposition of peat and muck in the fish pond bottom (Lim and Cuzon, 1994 and Ali, 1988).

Other two quality attributes expansion ratio (ER) and true density (TD) are equally important as they increase digestibility and also prevents the drifting of aqua feed pellets by air or waves during feeding (Rout, 1997). Chevanan et al. (2009) has used novel ingredients like distillers dried grains with solubles and whey to improve the extrudate properties like durability and density. They have also found that feed moisture content and extruder screw speed significantly impact the extrudate properties. Vassallo et al. (2006) in their studies on physical behavior of feed pellets in Mediterranean waters indicated that length of pellets extruded affect the settling velocity. Kannadhasan et al. (2011) studied the effect of starch sources and protein on physical properties of aquaculture feed containing distillers dried grains with soluble. They concluded that extrudate properties such as expansion ratio, unit density, sinking velocity, color, water absorption and solubility indices, and pellet durability index are judging factor to check the suitability of feed for various fish species. These properties are dependent on the ingredients used in the feed preparation. Rolfe et al. (2001) in their studies concluded that moisture content of the feed, particle size and screw speed effect properties like durability and water stability. Kraugerud et al. (2011) concluded that physical properties of extruded fish feed especially the expansion ratio are strongly related to the starch content. Adding cellulose to feed diets had resulted in negative correlation with expansion ratio. Garg et al. (2010) indicated that expansion ratio, and bulk density of extruded soy-rice blend extrudates were affected by barrel temperature, feed moisture content and soy flour-rice blend ratio.

1.3 Response surface methodology and genetic algorithm

Response surface methodology (RSM) is a combination of mathematical and statistical techniques widely used in product development (Khuri and Cornell, 1987). Shankar et al. (2010a) in their studies has mentioned that RSM is good approach to summarize the trends of the process variables for either maximization or minimization of the product quality. The same authors also indicated that interpretation of the RSM results is very complex especially when optimizing a function with more than three independent variables. Many researchers like Frazier et al. (1983), Bhattacharya and Prakash (1994), Rout and Bandyopadhyay (1999), Sacchetti et al. (2004), Shankar and Bandyopadhyay (2004; 2005), Zhang and Gao (2007), Wang et al. (2009), Shankar et al. (2010a), Bhattacharya et al. (2011), Shankar et al. (2012), Lopes et al. (2012), Sobukola et al. (2012) and Minf et al. (2012) used RSM to understand the effect of process variables on product characteristics. To understand real time and complex processes like extrusion, back propagation algorithms for prediction and genetic algorithms (GAs) for optimization have gained importance (Shankar and Bandyopadhyay, 2004 and 2007; Chen, and Ramaswamy, 2002; Koc et al., 2007; Shankar et al., 2010a, Mokeddem and Khellaf, 2009; Enitan and Adeyemo, 2011; Banakar and Akandi, 2012). GAs finds extensive application where the process systems are highly complex and nonlinear (Deb, 2000; Holland, 1992). Chun et al. (1998) discussed the usefulness of heuristic algorithms as the search method for diverse optimization problems. Their studies include comparison of immune algorithms, genetic algorithms, and evolutionary algorithms on diverse optimization problems and indicated that results of genetic algorithm are superior to others.

1.4 Objective

Aqua feed formulation mix is complex as variety of raw materials from both animal and plant sources are used. Effect of extrusion process variables like barrel temperature, L/D ratio,

feed moisture content and screw speed on multi-component high protein, fat and fiber rich feed formulation are very complex and difficult to understand. Response surface methodology and genetic algorithm can be collectively used for analysis of such data. Published literature on using response surface methodology and genetic algorithm on analysis of aqua feed physical properties data is not available. The objective of the present research is to analyze the aqua feed physical properties data using response surface methodology and genetic algorithm. The specific objectives are a) to understand the effect of extrusion process variables like screw speed (rpm), L/D ratio, barrel temperature (°C) and feed moisture content (%) on aqua feed pellet properties like water stability (WS), expansion ratio (ER) and true density (TD) and b) to optimize the process conditions which can maximize the WS, ER and TD using genetic algorithm.

2. MATERIAL AND

2.1 Experimental Data

Experimental data on aqua feed pellet properties with respect to process variables like screw speed, L/D ratio, barrel temperature and feed moisture content was adapted from the studies conducted by Rout (1997) and Rout and Bandyopadhyay (1999). Figure 1 indicates the design features of the variable length single screw extruder used in their study (Rout, 1997; Rout and Bandyopadhyay, 1999 and Tumuluru et al., 2012). In their studies, extruder experiments were conducted at five levels (Table 1) based on rotatable experimental design (Rout and Bandyopadhyay, 1999 and Bandyopadhyay and Rout, 2001). Ingredients used for extrusion of multi-component aqua feed are fish meal, shrimp head meal, squid meal, deoiled soybean meal, wheat flour (whole wheat ground and bran separated), fish oil, and micro ingredients like lecithin, vitamins, minerals, and calcium di-hydrogen phosphate (Table 2). A least cost feed formula was developed based on simplex method. The raw material was initially ground to 0.3 mm particle size, and batches of dough were prepared by adding the required quantity of water to the feed mix based on the experimental design.

Table 1. Five level central composite design (Rout, 1997)

Independent variables	Code	Coded levels				
		-2	-1	0	1	2
Screw speed (rev min ⁻¹)	x ₁	20	50	80	110	140
L/D ratio	x ₂	8	10	12	14	16
Barrel temperature (°C)	x ₃	60	70	80	90	100
Feed moisture content (%)	x ₄	20	30	40	50	60

2.2 Extrusion of Aqua Feed

A least cost formula based on the simplex method was used to design the multi-component feed extrudate for *Penaeus monodon* from the ingredients which included fish meal, shrimp head meal, squid meal, deoiled soybean meal, wheat flour (whole wheat ground and bran separated), and fish oil (Table 2). The other micro ingredients included were lecithin, vitamins, minerals, and calcium di-hydrogen phosphate. The aqua feed is initially ground to 300µm size and is further mixed for about 20 min using a kitchen mixer grinder (Summet mixer grinder, India). Batches of dough were prepared based on the experimental design by adding the required quantity of water to the mix prior to extrusion. Further these feed mix dough were kept in a polythene bags for 10 minutes for attaining moisture uniformity. The extrusion was carried out at different levels of process variables based on the experimental design. During extrusion only the steady state output was used for the physical properties measurement. Rout (1997) and Shankar et al (2010a) has discussed in detail the feed dough preparation and further extrusion process followed for the aqua feed mixes.

168 Table 2. Composition of multi-component aqua feed mix (Rout, 1997)

Ingredients	% Inclusion
Fish meal	29.37
Shrimp head meal	10.00
Squid meal	5.00
Wheat flour	28.25
Deoiled rice bran	4.01
Deoiled soybean meal	15.00
Fish oil	2.27
Lecithin	1.00
Micro-ingredients	5.10

169 2.3 Measurement of Aqua feed Properties

170 Rout (1997) and Rout and Bandyopadhyay (1999) has discussed in detail the experimental
 171 methods followed for measurement of aqua feed properties like water stability (WS),
 172 expansion ratio (ER) and true density (TD).
 173

174
 175 Expansion ratio is defined as the ratio of diameter of the dried pellets to that of die diameter
 176 of the extruder. WS was measured using wire mesh baskets with a 0.5 mm opening. A
 177 batch of 20 g pellets were placed in these baskets and immersed in saline water. The top of
 178 the baskets was kept out of water surface whereas the feed was allowed to stay in water
 179 column for a period of 120 min. After the test period the samples were taken out of the
 180 baskets and dried in an oven at 100°C for 3 hours. WS is calculated by measuring the
 181 weight of dried sample left in the basket to the original weight.
 182

183 True density is measured using pycnometer or specific gravity bottle using toluene (Punima,
 184 1985). The procedure includes measuring the mass of a) empty specific bottle b) specific
 185 gravity bottle + feed pellets c) specific gravity bottle + feed pellets +toluene d) specific
 186 gravity bottle+ toluene and e) specific gravity bottle + distilled water. True density is
 187 calculated using Equation (1).

$$188 \rho_f = \frac{\text{Dry mass of feed particles}}{\text{Mass of toluene of equal volume}} \times \rho_t \quad (1)$$

189 where ρ_f is the true density and ρ_t is the density of toluene.
 190

191 2.4 Data Analysis

192 The experimental data generated by Rout (1997) and Rout and Bandyopadhyay (1999) was
 193 further analyzed using response surface methodology (RSM) and genetic algorithm (GA).
 194 These two are powerful data analysis techniques widely used for process modeling and
 195 optimization. RSM is a combination of statistical method whereas GA is an evolutionary
 196 algorithm, which optimizes a function based on natural selection.

197 2.5 Response Surface Methodology

198 A second-order response model (Equation 2) was chosen to develop the regression
 199 equations in original terms for WS, ER and TD for the extrusion experimental data
 200 generated by Rout (1997) and Bandyopadhyay and Rout (2001).

$$201 y = b_0 + \sum_{i=1}^n b_i x_i + \sum_{i=1}^n b_{ii} x_i^2 + \sum_{i=1}^n \sum_{j=1}^n b_{ij} x_i x_j + \varepsilon \quad (2)$$

202 where y is the dependent variable, x_i and x_j are the coded independent variables, b_0 , b_i , b_j
 203 are coefficients, n is the number of independent variables and ε is an unobservable error.
 204 The significance of the linear, quadratic and interactive terms was evaluated based on the
 205 p-value. Regression equations (Table 3) were further used for drawing the response surface

over a fitness function. A linear rank fitness selection method (Equation 3) is widely used as it behaves in a robust way and helps to prevent the stocking of the population (Shankar et al., 2010a & Sivaraj, 2011).

$$f_i = 2 - SP + 2(SP - 1) \times \frac{(Pos - 1)}{(n - 1)} \quad (3)$$

where n = the number of individuals in the population, Pos is position of an individual in this population and SP = selective pressure, which indicates the probability of selecting the best individual in the population.

Parent population of the fitted population is selected based on Roulette wheel selection technique (Equation 4). In this process, chromosomes with the highest fitness values are selected whereas the lower ones are eliminated. If the fitness of 'n' individuals in a population is $f_1, f_2, f_3, \dots, f_n$, the selection probability (P_i) of an individual is calculated using the following equation (Deb, 2000, Malhotra, 2011).

$$P_i = \frac{f_i}{(f_1 + f_2 + f_3 + \dots + f_n)} \quad (4)$$

The other two operations, which are used in genetic algorithm, are crossover and mutation. These operations are carried out on the population selected based on the Roulette wheel selection procedure. In crossover operation bit strings of the selected parent chromosomes are exchanged to generate new populations called offspring. A single point crossover method was used in the present study. The process of bit inversion mutates new offspring's generated based on crossover operation. This operation helps to maintain a) diversity within the population b) inhibits premature convergence of the population and c) prevents the population getting stuck at local points. Different steps used in genetic algorithm are given below (Shankar et al., 2010a).

Step 1: Random populations of chromosomes, which are suitable solutions for the problem.

Step 2: Fitness values are evaluated for the population generated.

Step 3: New population is generated using the step 2 till the population is complete.

Step 4: Two parent chromosomes are selected based on the fitness values. Higher the fitness values better the chance for getting selected as a parent.

Step 5: New parents are selected with certain amount of crossover probability where new offspring's are generated. In general a crossover probabilities of 85-95 % are recommended to prevent premature convergence. No crossover will generate offspring's, which are exact copy of the parents.

Step 6: The new offspring's generated are mutated (bit inversion) with certain amount of probability. Low mutation rate of about 0.5 percent to 1 percent is recommended to obtain optimized results from genetic algorithm. Small mutation rates prevent genetic algorithms from falling into local maxima or minima.

Step 7: New population generated is placed in the current population and used in the algorithm.

Step 9: If the end condition is satisfied (where fitness does not change after certain number of iterations), the algorithm is stopped and the best solution is returned to the population.

Step 10: Go to step 2.

GA program developed by Shankar and Bandyopadhyay (2004) and Shankar et al. (2010a & b) was used in the present study. Regression equations developed for WS, ER and TD were used as the objective functions. A random population of 100 chromosomes, crossover and fitness probabilities of 0.80 and 0.98 and 100 iterations were used.

Fig 2 indicates how combination of RSM and GA is used in the present study to understand the effect of extrusion process variables on aqua feed physical properties. Shankar et al.

(2010a) has used RSM and GA in combination for optimizing the biomass flow in a single screw extruder.

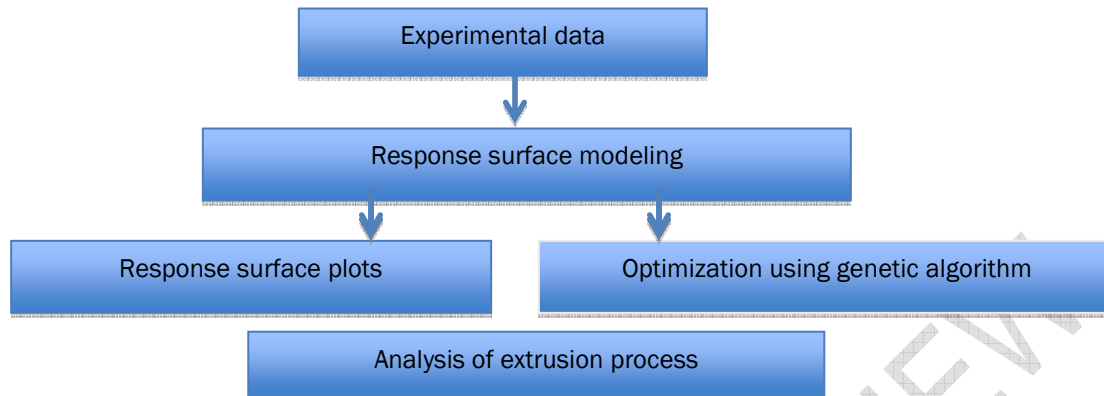


Fig. 2. Flow diagram for extrusion process data analysis using RSM and GA

3. RESULTS

3.1 Response Surface Analysis

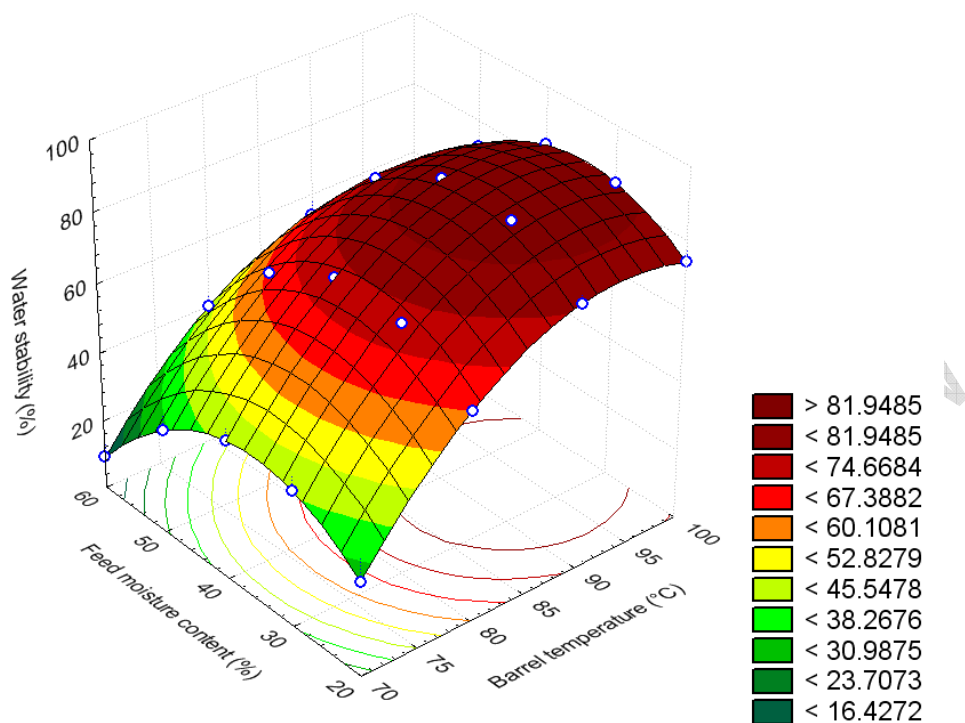
The coefficient of determination (R^2) values of the regression equations for WS, ER, and TD were 0.97, 0.95, and 0.95, respectively, indicating that they have adequately fitted. The predicted and observed plots drawn for WS, ER and TD (not shown) have corroborated with above observation.

Response surface plots were developed based on the regression equations (Table 3) to understand the effect of extrusion process variables on WS, ER and TD. Surface plots were drawn for each of the two variables, where the other two variables were kept at the center point of the experimental design. Some of the important surface plots are given in figures 3-8. Surface plots (Figures 3 and 4) indicated that barrel temperature > 90°C, feed moisture content 30-40 %, L/D ratio of > 12 and screw speed of 80 rpm maximized the WS values. Based on surface plots 5 and 6 the ER values are maximized at barrel temperature of 70-80°C, feed moisture content of 30-40%, screw speed of 60-70 rpm, and L/D ratio of 12. Surface plots of TD (Figures 7 and 8) specify that a feed moisture content < 40%, barrel temperature < 70°C, L/D ratio of 12-14 and screw speed of 80 rpm maximized the TD values. Table 4 indicates the summary of the trends of the extrusion process variables based on response surface plots for maximization of WS, ER and TD values.

Table 3. Regression equations developed for WS, ER and TD

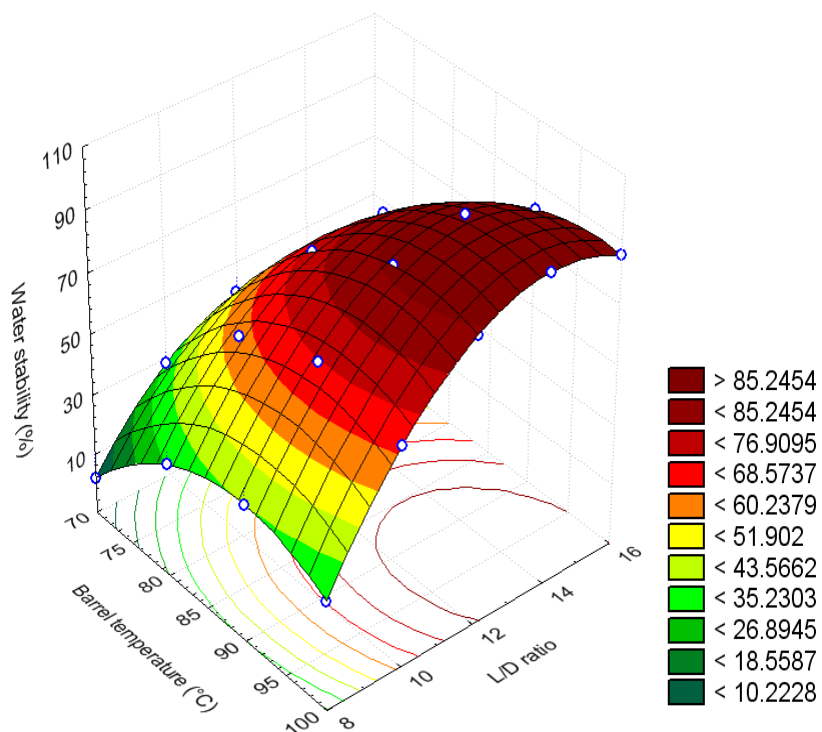
Aqua feed pellet property	Regression equations	(R^2)
WS (%)	$-914.521 + 0.566x_1 + 32.368x_2 + 14.57x_3 + 3.78x_4 - 0.0025x_1^2 - 1.78x_2^2 - 0.087x_3^2 - 0.055x_4^2 + 0.165x_2x_3$	0.97
Expansion ratio	$-0.345 + 0.0016x_1 + 0.0098x_3 + 0.015x_4 - 0.000018x_1^2 - 0.0042x_2^2 - 0.000118x_3^2 - 0.000192x_4^2 - 0.000155x_1x_2 + 0.000044x_1x_3 + 0.000253x_2x_3 - 0.000018x_1x_4 - 0.000384x_2x_4 + 0.000049x_3x_4$	0.95
True density (kg/m ³)	$560.4987 + 0.454x_1 + 57.89x_2 + 9.10x_3 + 3.62x_4 - 0.01x_1^2 - 2.83x_2^2 - 0.051x_3^2 - 0.045x_4^2 + 0.0912x_1x_2 + 0.300x_2x_4 - 0.067x_3x_4$	0.95

x_1 = screw speed (rpm), x_2 = L/D ratio, x_3 = barrel temperature (°C) and x_4 = feed moisture content (%).



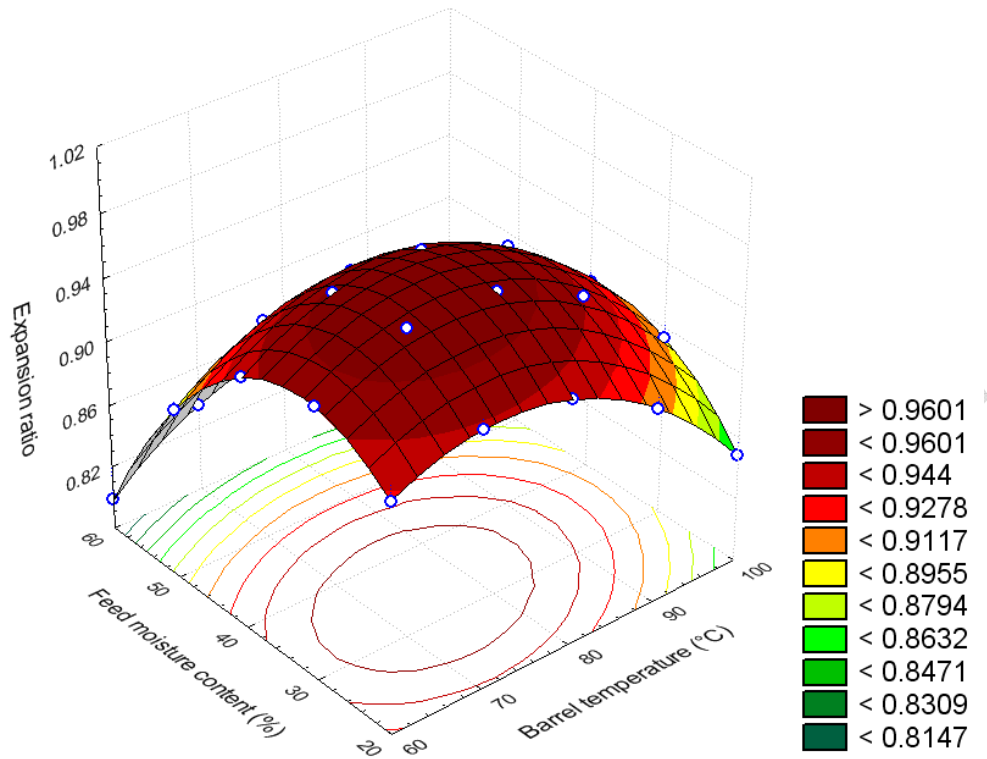
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Fig. 3. Surface plot of WS at screw speed of 80 rpm and L/D ratio of 12



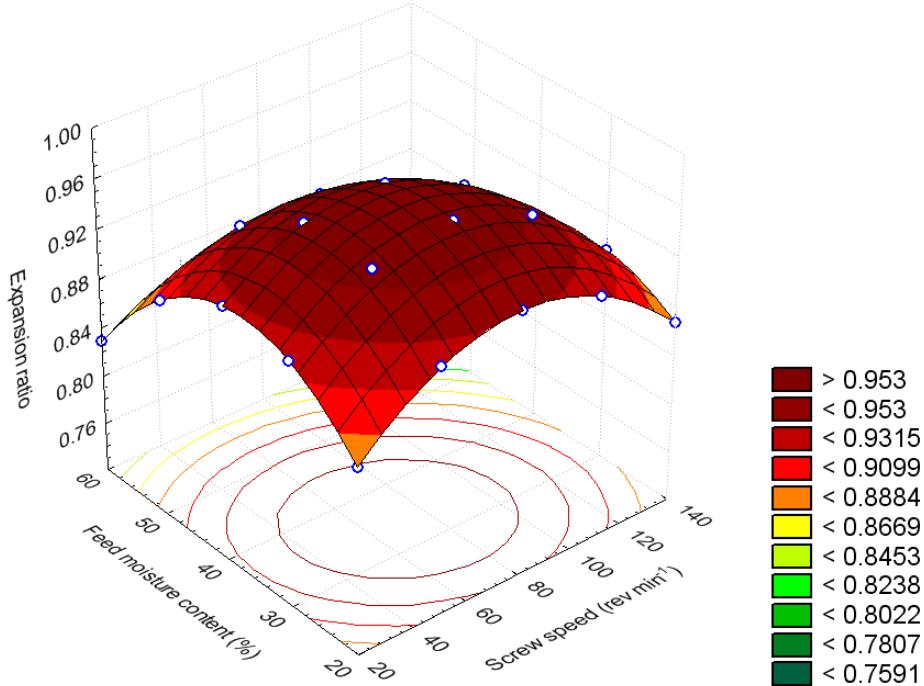
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Fig. 4. Surface plot of WS at screw speed of 80 rpm and feed moisture content of 40%



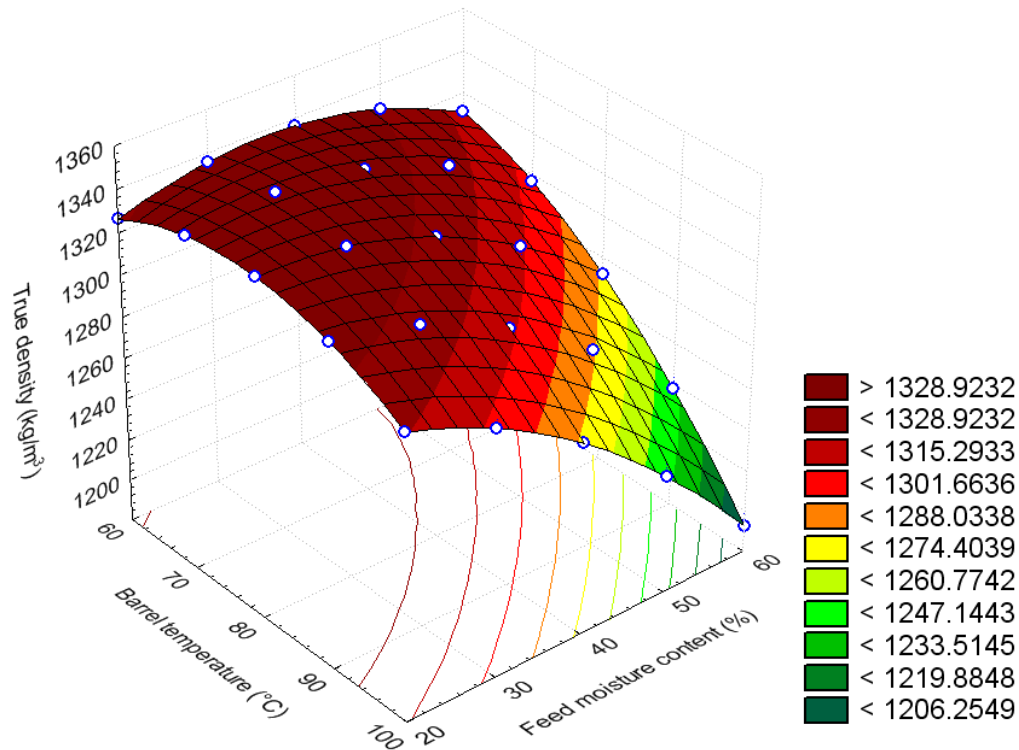
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Fig. 5. Surface plot of ER at L/D ratio of 12 and screw speed of 80 rpm



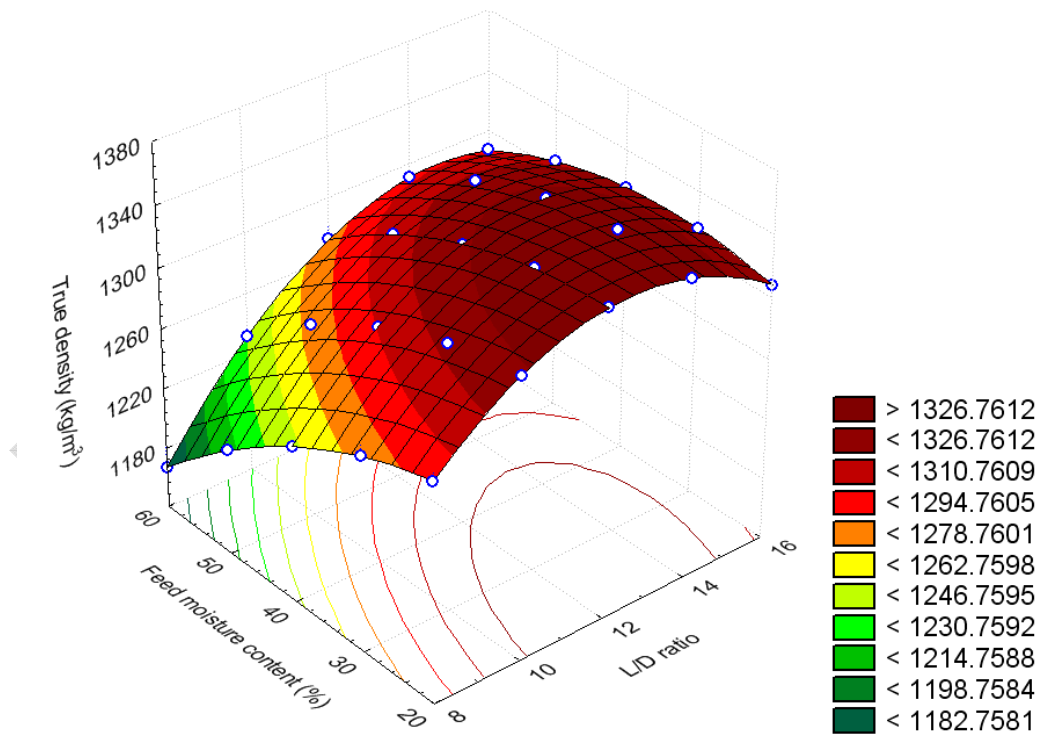
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Fig. 6. Surface plot of ER at L/D ratio of 12 and barrel temperature of 80 °C



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Fig. 7. Surface plot of TD at screw speed of 80 rpm and L/D ratio of 12



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Fig. 8. Surface plot of TD at screw speed of 80 rpm and barrel temperature of 80°C

Table 4. Summary of the extrusion process variables based on RSM

Aqua feed pellet property	Extrusion process variables				Objective
	Screw speed, rev min ⁻¹ (x ₁)	L/D ratio (x ₂)	Barrel temperature, °C (x ₃)	Feed moisture content, % (x ₄)	
WS (%)	80	>12	> 90	30-40	Maximize
ER	60-70	12	70-80	30-40	Maximize
TD (kg/m ³)	80	>12	< 70	< 40	Maximize

3.2 Genetic Algorithm Analysis

Table 5 shows the optimum process conditions obtained using GA for maximization of WS, ER and TD. Maximum WS of 98.12 % was predicted at a screw speed of 89.90 rpm, L/D ratio of 13.89, barrel temperature of 94.34°C and feed moisture content of 36.01%. Maximum ER of 0.99 was predicted at screw speed of 60.37 rpm, L/D ratio of 12.18, low barrel temperature of 68.50 and medium feed moisture content of 33.61%, whereas a maximum true density of 1346.9 kg/m³ was predicted at screw speed of 90.24 rpm, L/D ratio of 13.52, low barrel temperature of 64.88 °C and medium moisture content of 38.36 %.

Table 5. Optimum extrusion process variables based on genetic algorithm

Aqua feed pellet property	Optimum extrusion process variables				Maximum predicted pellet property
	Screw speed, rpm (x ₁)	L/D ratio (x ₂)	Barrel temperature, °C (x ₃)	Feed moisture content, % (x ₄)	
WS (%)	96.71	13.67	96.26	33.55	98.12
ER	60.37	12.18	68.50	33.61	0.99
TD (kg/m ³)	90.24	13.52	64.88	38.36	1346.9

4. DISCUSSIONS

4.1 Water Stability (WS)

RSM and GA-based analysis in the present study indicated that L/D ratio; barrel temperature and moisture content have a strong influence on WS values of the aqua feed extrudate. Mainly the gelatinization of starch and denaturation of protein present in the multi-component aqua feed mix and formation of starch-protein matrix govern WS of aqua feed pellets. Transformation of water insoluble starches into viscous gels due to gelatinization and the simultaneous conversion of soluble proteins into an insoluble fibrous network influence the WS values. Gelatinization primarily depends on the time-temperature relationship of the cooking process (Abdelrahman and Hosney, 1984; Venugopal and Keshavanath, 1984; Tacon, 1990). In the present study at high temperatures of >90°C the aqua feed mix might have undergone gelatinization of starch and resulted in the formation of starch-protein matrix. L/D ratio coupled with screw speed and feed moisture content also had a significant effect on WS. These process variables provided the necessary heating surface and residence time for cooking, and might have resulted in formation of an insoluble fibrous network. Rokey and Plattner (2003) indicated that starch gelatinization at high temperatures and moistures during extrusion are crucial because it affects feed digestibility, expansion and contributes to water stability. They also indicated that the amount of starch gelatinized during processing depends on the starch type, particle size and processing conditions. Many researchers have demonstrated that the extent of starch gelatinization is a

strong function of barrel temperature and feed moisture content during extrusion of high starch food/feed materials (Bhattacharya and Hanna, 1987; Cai and Diosady, 1993; Chiang and Johnson, 1977; Colonna et al., 1984; Diosady et al., 1985; Guy and Horne, 1988; Lawton et al., 1972; Shankar and Bandyopadhyay, 2004). Tumuluru et al. (2010) in their studies on pelleting of DDGS observed that the durability of the DDGS increased with increase in temperature and steam addition due to gelatinization of starch. Misra et al. (2002) in their studies on physiochemical parameter of aqua feeds like water stability are higher for extruded aqua feed pellets.

4.2 Expansion Ratio (ER)

ER is mainly governed by the pressure rise in the die head, which is in turn controlled by the flow rate (Botting, 1991). Medium to high moisture content in the aqua feed mix increases the flow rate by increasing the lubricating action and reducing the pressure, resulting in less expanded structure (Pillay, 1990). According to Fallahi et al. (2012) ER values of the feed extrudate are influenced by the extent of pressure developed which in turn is influenced by several factors like dough moisture content, die dimensions, and feed ingredient composition and mass flow rate, residence time, and rheological behavior of the dough. The same authors indicated that high levels of temperatures and shear forces during extrusion processing change the internal structure of the dough during cooking process. Low ER values of <1.0 observed in the present extrusion studies can be due to significant influence of lipid and protein present in aqua feed mix, which might have interfered with the gelatinization process by developing protein networks with starch and also starch-lipid complexes. Mohamed (1990) in his studies on extrusion of starch-based products concluded that the ER of the extruded product is unfavorably affected by increased moisture and protein content. Dautant et al. (2007) in their studies observed that regardless of shear rate and moisture content of the sample, high lipid and starch compositions in the aqua feed mix results in a decreased viscosity even with increase in temperature and moisture content, consequently reducing ER. In the present study lower screw speeds, lower barrel temperature, medium L/D ratio and medium feed moisture content significantly influenced the ER which might be due to reduced viscosity of the feed mix. Kim et al (1989) suggested that higher ER values could be as a result of increase in barrel temperature, which can result in more starch gelatinization and more excretion of super-heated steam

4.3 True Density (TD)

True density (TD) is influenced by screw speed, L/D ratio and barrel temperatures based on RSM and GA analysis. In general, higher screw speeds reduce the viscosity and in turn increase the density of the product. The increase in true density with increase in screw speed can be due to the increase in the non-Newtonian dough density. Also, with the increase in screw speed, the free moisture might have distributed throughout the dough more evenly and resulted in uniform puffing as the pellet leaves the die. The presence of lipid in the aqua feed mix might result in the formation of amylase-lipid complexes which, in turn, could have reduced the viscosity of the extrudate. Singh and Smith (1999) found that an increase in lipid content resulted in a decrease in viscosity and increased the TD values. The same authors observed that moisture content of the aqua feed mix alters the density to a great extent as the gelatinization of starch helps in binding the particles and improve the pellet density. At medium L/D ratio a consistent TD values were obtained in the present study, which might be due to laminar flow of the feed in the metering zone. According to Gehring et al. (2012) the density of the feed pellets decrease with increase in moisture content. In the present study low barrel temperatures and medium moisture content observed possibly might have decreased the extent of gelatinization and the content of the superheated steam, yielding a high density product as observed by (Kim et al., 1989 and Harper, 1981).

5. CONCLUSIONS

In the present study extrusion experimental data was analyzed using response surface methodology (RSM) and genetic algorithm (GA). Regression equations developed has adequately described the effect of process variables on the quality attributes with coefficient of determination values of >0.95. RSM analysis indicated that a medium screw speed of 60-80 rpm, L/D ratio of 12-13, and feed moisture content of 30-40% and barrel temperature of $\leq 80^{\circ}\text{C}$ for ER and $\text{TD} > 90^{\circ}\text{C}$ maximized WS, ER and TD. Based on GA analysis L/D ratio of 12-13.5, medium feed moisture content of 30-33% maximized the WS, ER and TD. GA analysis indicated that a maximum WS of 98.10%, 0.99 and 1346.9 kg/m^3 was predicted at a screw speed of 96.71, 60.37 and 90.24 rpm, L/D ratio of 13.67, 12.18 and 13.52 barrel temperature of 96.26, 68.50 and 64.88°C and feed moisture content of 33.55, 33.61 and 38.36%. the present data analysis indicated that screw speed, barrel temperature and feed moisture content were the interacting process variables which has influence maximum the WS values followed by ER and TD.

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