

Original Research Article

Essential Amino Acid composition of Noodles Analogue from Aerial Yam, Rice, and African Yam Bean flour blend using Response Surface Methodology

ABSTRACT

Response surface technology was used to evaluate the essential amino acid of noodles analogue from Aerial yam (*Dioscorea bulbifera*), Rice (*Oryza sativa*), and African yam bean (*Sphenostylis sternocarpa*) flour mixture. Flours from Aerial yam, rice, and African yam bean were individually produced blended in the ratio of 50%: 25%: 25%, respectively and extruded using Brabender single screw laboratory extruder (Duisburg DCE 330 Model) filled with 3.0mm die nozzle diameter. A central composite rotatable design (CCRD) with three variables, namely barrel temperature, feed moisture content and screw speed and fire level coded -a, -1, 0, +1, +a, was used in the experimental of design to obtain 20 samples of noodles analogue. Essential Amino Acid profiling was done on the noodles analogue and data analyzed by regression analysis. Result showed that lysine content ranged from 4.55 to 7.55mg/100g, valine content ranged from 3.81 to 5.06mg/100g, methionine content ranged from 0.86 to 2.24mg/100g, phenylalanine content ranged from 5.06 to 5.45mg/100g, histidine content ranged from 3.09 to 4.20mg/100g, tryptophan content ranged from 0.83 to 1.96mg/100g, leucine content ranged from 6.87 to 7.90mg/100g, isoleucine content ranged from 2.91 to 4.91mg/100g, threonine content ranged from 3.55 to 4.44mg/100g. The coefficients of determinations (R^2) were high and ranged from 0.8987 to 0.9916 at 5% level. The response surface plots suggests that the models developed had a good fit and the CCRD was effective in explaining the effect of the process condition on noodles analogue as influenced by barrel temperature, feed moisture content and screw speed of the extruder. The data obtained from the study could be used for the control of product characteristics. The study indicated improved noodles analogue from Rice, Aerial yam, and African Yam Bean can be produced as noodles analogue.

Key Words: Noodles analogue, Extrusion, Response Surface Methodology

1. INTRODUCTION

Adequate nutrition is essential for proper growth and cognitive development which has a positive correlation with high productivity for individual as well as every country round the globe. In Nigeria, most person's even children can adequately meet the recommended daily allowance for calories given the nature of our staple foods, which are largely of starch origin; starchy foods like rice and aerial yam (*Dioscorea bulbifera*) an unpopular and underutilized yam are primarily high energy calorie food consisting of carbohydrate within the range of 72 to 75%; and requiring simple boiling to form a meal. However; rice and aerial yam are low in lysine but may contain appreciable amount of methionine. Legumes protein such as African yam bean are adequate in lysine but deficient in methionine, all the same, total protein intake is usually just half of the recommended level (Akinyele, 1987). Recognizing the risk in neglecting attention on quality protein and amino acids in diets with respect to global health agenda (Semba, 2016) calls for a rethink. Consequently, there is a need to enrich our staple foods by way of nutrient complementarities with effective processing technology into a value-added product that can create a niche for itself in human nutrition. One possible approach to this is the supplementation of starchy food crops with food legumes such as African yam bean, whose edible portion contains protein twice that of cereals (Yilma and Adamassau, 2019), through extrusion process. Extrusion cooking affects the nature of many food constituents, including starches and proteins by changing their physical, chemical and nutritional properties (Iwe, 2001)

African yam bean is a lesser known legumes in the tropical and is important source of protein for human body although with low true protein digestibility of about 62.9% as reported by Kalu *et al.* (2019). African yam bean protein content ranges between 21 and 29% with high metabolic energy (Kalu *et al.*, 2019). Although proteins from legumes are not equal in quality with animal protein, nonetheless, they can be an adequate substitute if they are eaten in combination with other foods.

The essential amino acids are defined by World Health Organization (WHO) and Food and Agriculture Organization (FAO) as indispensable nutrients (World Health Organization, 2007). These are amino acids that

cannot be synthesized or produced by mammals, including humans, therefore, they must be taken in as food nutrients or dietary supplements.

Response surface methodology (RSM) is a collection of statistical and mathematical techniques useful for developing, improving and optimizing process in which a response of interest is influenced by several variables and the objective is to optimize this process. RSM has important application in the design, development and formulation of new product, as well as in the improvement of existing product design (Myers and Montgomery, 2002). It defines the effect of independent variables, alone or in combination, on the processes. In addition to analyzing effects of the independent variables, a mathematical model which describes the process is generated (Myers and Montgomery, 2002)

RSM consists of a group of mathematical and statistical techniques that can be used to define the relationships between the response and the independent variables. RSM defines effect of the independent variables, alone or in combination, on the process and also generates a mathematical model. The graphical perspective of the mathematical model has led to the term Response Surface Methodology. The RSM approach has widely been applied in extruded food product development (Seth and Rajamanickam, 2019; Kalu *et al.*, 2022) Cereal and tubers foods lack some of the essential amino acids. Complementing rice and aerial yam with African yam bean in the production a more balanced single noodle meal using extrusion process might be the needed improvement in production of cheap, affordable enriched noodles analogue. However, this has not been studied. To this extent, the amino acid profiles of African yam bean will complement those of rice and Ariel yam, hence the objective of this work is to evaluate the essential amino acid of noodles analogue from Ariel yam, rice and African yam bean mixture.

2. MATERIALS AND METHODS

2.1 Procurement of Materials

The Aerial yam was bought form Onueke market in Ebonyi State and identified at National Root Crop Research Institute (NRCI), Umudike, Abia State, Nigeria. The Rice and cream colored African yam bean were bought from Akaeze market in Ebonyi State and identified and bought at National Institute of Horticulture (NIHOT) Mbato sub zone, Okigwe, Imo state. Xanthan gum (G 1253, sigma – Aldrich USA) was procured from pharmaceutical shop in Aba, Dangote iodized table salt was purchased from a Super market in Afikpo, Ebonyi state, Nigeria.

2.2 Sample preparation

The flour samples used in this research were prepared in the Food Processing Laboratory, Department of Food Technology, AkanuIbiam Federal Polytechnic, Unwana Ebonyi State, Nigeria.

2.2.1 Preparation of Aerial yam flour

Aerial yam flour was prepared according to the method described by Kalu *et al.* (2022) on water yam with slight difference. Aerial yam bulbs were washed, sorted, and peeled manually with a stainless steel knife. The peeled bulbs were sliced manually in 10mm thickness and dried at 60°C for 12 h in an air convection oven. Dried chips were cooled for 4h at room temperature under air current and milled using hammer mill. The flour sample was sieved through 600µm mesh size, packaged and sealed in polyethylene bag for further use.

2.2.2 Preparation of rice flour

Rice grain were sorted, and cleaned in an aspirator (Model: OB 125Bindapst Hungary), washed with potable water and allowed to drain under air current, the grains were dried at 60°C for 12h, milled using hammer mill. The flour sample was sieved through 600µm mesh size, packaged and sealed in polyethylene bag for further use.

2.2.3 Preparation of African yam bean flour

African yam bean flour was prepared according to the method described by Kalu *et al.* (2022) with a slight difference. Cream colored African yam bean grain were sorted and washed with potable water. Cleaned grains were soaked for 3h at room temperature and dehulled in a dehulling machine. Dehulled grains were dried at 60°C for 12h in an air convection oven and milled with hammer mill. The flour sample was sieved through 600µm mesh size, packaged and sealed in polyethylene bag for further use.

2.3 Flour Blending Ratio

Flour samples were blended in the ratio of 50% Aerial yam, 25% rice, and 25% African yam bean based on preliminary result

2.4 Experimental Design

A central composite rotatable design (CCRD) for three variable was employed to examine the response pattern of the effects of barrel temperature, BT ($^{\circ}$ C), feed moisture content, FMC (%) and screw speed, SS (rpm) on amino acid composition of the noodles analogue. Each variable was evaluated as shown in Table 1. Each variable were at five levels, namely $-\alpha - 1$, $+0$, $+1$ and $+\alpha$ gave 15 variable combinations in which the 15th combination was replicated 5 time at the center point (0, 0, 0) of the design to generate a total of 20 experimental runs used.

2.5 Noodles analogue formulation

Every one hundred grams (100g) of flour was mixed in the desired water level according to the experiment design (Table 1). 1g Iodized salt and xanthan gum 0.5 g each respectively was added for thickening and stability (Gambuset *al.*, 2007) and thoroughly mixed using Hobart mixer (Model: A:200; English). Thereafter, the mixture was subjected to extrusion cooking.

2.6 Extrusion Cooking

A single screw Brabender laboratory extruder (Model DCE 330, New Jersey, USA,) located at the Food Processing Laboratory of Federal Polytechnic, Mubi, Nigeria, was used for the cooking. The extruder feed hopper equipped with auxiliary auger-screw rotating at variable speed on vertical axis was set at 60rpm for all the sample runs. The extruder was allowed to run to stabilization over a period of 30 min at screw speed of 40 to 45rpm during which time the no- load torque and temperature and pressure regimes were displayed on the control panel before the experimental runs commenced for each set of conditions. The moisture content of flours, barrel temperature and screw speed were adjusted according to the experimental design (Table 1). The feed was introduced gradually but continuously into the feed hopper and were received at the die end with of 3.00mm diameter as dried strands or pellets. The samples were allowed to cool and packaged in a polythene bag for analysis.

2.7 Amino acid profile analysis

Two grams (2.0 g) of each sample was weighed into extraction thimble and the fat extracted with chloroform/methanol mixture using a soxhlet extraction apparatus (AOAC, 2015). The extraction lasted for 6h. An aliquot (35 mg) of the defatted sample was weighed into a glass ampoule. A 7ml of 6M HCl was added and oxygen expelled by passing nitrogen gas into the ampoule (to avoid possible oxidation of some amino acids during hydrolysis). Each glass ampoule was sealed with a Bunsen flame and put into an oven at 105° C \pm 5° C for 22 h. The ampoule was allowed to cool before breaking open at the tip and the content was filtered to remove the humins. The filtrate was then be evaporated to dryness at 40° C under vacuum in a rotary evaporator. Each residue was dissolved with 5ml of acetate buffer and stored in a plastic specimen bottle, and kept in a deep freezer. The analysis was carried out using ion- exchange chromatography (IEC) as described by (Adeyeye&Afolabi, 2004). The amounts loaded, for both samples, were 5-10 ul each. These were dispensed in the cartridge of the analyzer. The Technicon Sequential Multi-sample Amino acid Analyzer (TSM) (Technicon Instruments Corporation, New York) was used for the analysis. The TSM analyzer is designed to separate and analysis free acidic, neutral and basic acids of hydrolysate. The analysis lasted for 76 min for each sample. The column flow rate was 0.50 ml/min at 60° C with reproducibility consistent within \pm 3%. The net height of each peak produced by the chart record of the TSM (each representing an amino acid) was measured and calculated. The values that were reported are averages of two exterminators.

2.8 Statistical Analysis

The data obtained from triplicate run using Central Composite Rotatable Design was analyzed statistically using Response Surface Methodology, so as to fit the quadratic polynomial equations generated using Design Expert software version 8.0.7.1 (Stat-ease Inc., USA).

A second order polynomial equation was used to fit the experimental data given in Table 2. The model proposed for the response (Y_i) was shown in equation 1 and 2

$$Y_1 = \beta_0 + \beta_1 X_1 + \beta_2 X_2 + \beta_3 X_3 + \beta_{11} X_1^2 + \beta_{22} X_2^2 + \beta_{33} X_3^2 + \beta_{12} X_1 X_2 + \beta_{13} X_1 X_3 + \beta_{23} X_2 X_3 + \beta_{123} X_1 X_2 X_3 + \dots \quad (1)$$

$$Y_1 = \beta_0 + \beta_1 BT + \beta_2 FMC + \beta_3 SS + \beta_{11} BT^2 + \beta_{22} FMC^2 + \beta_{33} SS^2 + \beta_{12} BTFMC + \beta_{13} BTS + \beta_{23} FMCSS + \beta_{123} BTFMCSS \dots\dots (2)$$

Where Y_1 is the predicted response for proximate composition, β_0 (intercept) is the value of the fitted response at the enter point of the design, β_i , β_{ii} , β_{ij} (regression coefficient term) being the linear, quadratic and cross product terms respectively and e is the random error term. In order to deduce workable optimum conditions, a graphical technique was used (Floros and Chinnan, 1988; Giovanni, 1983). To visualize the relationship between the responses and experimental levels for each of the factors the fitted polynomial equation was expressed as surface contour plots.

3. RESULT AND DISCUSSION

Nine (9) responses, namely lysine content (Y_1), valine content (Y_2), methionine content (Y_3), phenylalanine content (Y_4), histidine content (Y_5), tryptophan content (Y_6), leucine content (Y_7), isoleucine content, (Y_8), and threonine content (Y_9) which are described as essential amino acid composition of the noodles analogue were evaluated. The coefficients for the actual functional relation for predicting Y_i are presented in Table 2. The non-significant terms from the model were omitted based on students T- ratio (Khuri and Cornell, 1987). The contour plots for the amino acid compositions are shown in Fig 1 to 9.

Lysine content of Noodles Analogue

The values of lysine content of the noodles analogue varied from 4.55mg/100g (at barrel temperature of 237.5°C, feed moisture content of 24% and screw speed of 150rpm) to 7.55mg/100g (at barrel temperature of 145°C, feed moisture content of 34.09% and screw speed of 150rpm, Table 1) This showed that there was decrease in lysine contents of the noodles analogue with increase in barrel temperature and decrease in feed moisture content respectively. There was no clear effect of screw speed with lysine content in this study. The lysine content in this study is higher than the range reported by Omeire (2012) on African yam bean and cassava extruded blends and Fasuanet *al.* (2021) on ready- to- eat flasks from Acha, partially deffatted sesame meal and modified corn starch additionally. The FAO/WHO (1985) stipulated daily requirement of 5.80mg/100g. The high lysine content in this study gives the food an advantage as a food product; lysine in the body can affect human behaviour. Adequate amount of lysine alongside vitamin D is responsible for the absorption of calcium from the diet, helping to support strong bones and teeth. The response surface plots for lysine content are shown in Fig 1a-c. The linear effect of barrel temperature positively and significantly affected the lysine content at 5% level. The linear effect of feed moisture content and screw speed and the interaction effect of barrel temperature negatively and significantly ($p < 0.05$) affected the lysine. The quadratic effect of feed moisture content and screw speed were positively and significantly ($p < 0.05$) related to lysine. The quadratic effect of barrel temperature negatively and significantly ($p < 0.05$) affected the lysine content. The interaction effect of barrel temperature and screw speed negatively and significantly ($p < 0.05$) affected the lysine content. The interaction effect of feed moisture content and screw speed positively and significantly ($p < 0.05$) affected (Table 2); the lysine content the first order term was significant at 5 % level. However the second order term and lack of fit were not significant at 5 % level (Table 3). The R^2 for the fit was 0.9114 (91.14%). This meant that 91.14% of the total variation in lysine content in the noodles analogue was explained by the regression model. The model equation developed for predicting lysine was shown in equation 3

$$\text{Lysine} = 25.816 + 0.53BT - 0.96FMC - 0.51SS - 3.9E-4BT*SS - 7.10E-5BT^2 + 0.02FMC^2 + 5.59E-4SS^2 (3).$$

Valine content of Noodles Analogue

The values of valine content of the noodles analogue ranged from 3.81mg/100g (at barrel temperature of 145°C, feed moisture content of 24% and screw speed of 200.45rpm) to 5.06mg/100g (at barrel temperature of 90, 120°C, feed moisture content of 18% and screw speed of 120 rpm). This showed that there was decrease in valine content of the noodles analogue as barrel temperature, feed moisture content and screw speed increased respectively. Omeire (2012) on African yam bean and cassava extruded blends reported similar trend, similarly; the range of valine content in this study was within 5.01mg/100g reference value as reported by FAO/WHO (1985). Valine appears to be important for mental alertness, muscle coordination, and emotional calm, People may use valine supplements for muscle growth, tissue repair and energy, on the other hand; deficiency may cause insomnia and reduced mental function.

The linear effect of feed moisture content and screw speed and the interaction effect of barrel temperature and screw speed were negatively and significantly ($p < 0.05$) related to valine.

The quadratic effect of barrel temperature and feed moisture content were positively and significantly ($p < 0.05$) related to valine content. The linear effects of feed moisture content and screw speed; were negatively and significantly ($p < 0.05$) affected the valine content. The interaction effect of all the three independent variables, positively and significantly ($p < 0.05$) affected the valine content except the interaction of barrel temperature and screw speed which were negative (Table 2). The (first and second order) terms and the lack of fit were not significant at 5 % level (Table 3). The R^2 for the fit was 0.9700 (97.00%). This meant that 97.00% of the total variation in valine content in the noodles analogue was explained by the regression model. The response surface plots for valine content are shown in Fig 2a-c. The model equation developed for predicting lysine was shown in equation 4.

$$\text{Valine} = 14.45 - 0.53 - 0.53\text{FMC} - 0.02\text{SS} + 5.34\text{E-}4\text{BT*FMC} - 7.80\text{E-}5\text{BT*SS} + 1.10\text{E-}3\text{FMC*SS} + 5.65\text{E-}3\text{FMC}^2. \quad (4)$$

Methionine content of Noodles Analogue

The value of methionine content of the noodles analogue ranged from 0.86mg/100g (at barrel temperature of 52.5, 237.5°C, feed moisture content of 24% and screw speed of 150 rpm) to 2.24mg/100g (at barrel temperature of 145°C, feed moisture content of 24% and screw speed of 150rpm). The result revealed that the methionine values of the noodles analogue decreased with increase in barrel temperature. There was no clear effect of both the feed moisture content and screw speed on the methionine content of noodles analogue. The trend was similar to the report by Omeire (2012) on African yam bean and cassava extruded blends. Generally, methionine values in the noodles were limiting as they were lower than the 2.5g/100g FAO/WHO (1991) reference pattern in all the sample just as it is limiting in most plant food systems. This observation agree with other studies on other processing methods on amino acid studies (Ikegwu et al., 2022 and Ezegebe et al., 2022) on different bio treatments studies such as germination and fermentation. Methionine play a role in the health and flexibility of skin, nails and hair. It appears to help in the proper absorption of certain metals such as selenium and zinc and the removal of heavy metals, such as lead and mercury in the human body.

The interaction effects of all the three independent variables negatively and significantly ($p < 0.05$) affected the methionine content except the interaction effect of barrel temperature and screw speed which was positive. The quadratic effect of all the three independent variables, negatively and significantly ($p < 0.05$) affected the methionine content (Table 2). The (first and second order) terms and lack of fit were not significant at 5 % level (Table 3). The R^2 for the fit was 0.9562 (95.62%). This meant that 95.62% of the total variation in methionine content in the noodles analogue was explained by the regression model. The response surface plots for methionine content are shown in Fig 3a-c. The model equation developed for predicting methionine was shown in equation 5.

$$\text{Methionine} = -6.09 - 3.71\text{E-}4\text{BT*FMC} + 6.82\text{E-}5\text{BT*SS} - 1.33\text{E-}4\text{BT}^2 - 2.40\text{FMC}^2 - 1.20\text{SS}. \quad (5)$$

Phenylalanine content of Noodles Analogue

The values of phenylalanine content varied from 5.06mg/100g (at barrel temperature of 200°C, feed moisture content of 30% and screw speed of 120rpm) to 5.45mg/100g (at barrel temperature of 145°C, feed moisture content of 24%, and screw speed of 150rpm). This indicates decrease in phenylalanine content of noodles analogue with increase in barrel temperature and feed moisture content, with a decrease in screw speed. The phenylalanine content in this study is higher than the range reported by Omeire (2012) on African yam bean and cassava extruded blends and (Ikegwu et al., 2022) on germination of lablab beans seed flour. This is possible that thermal processing may have a positive effect on phenylalanine synthesis than bio processing of food. However, the values of phenylalanine content of the samples in this study were lower than FAO/WHO (1985), who stipulated daily requirement of 6.0 mg/100g. Phenylalanine appears to help the human body use effectively other amino acids as well as proteins and enzymes. Phenylalanine is usually converted to tyrosine, which is necessary for specific brain functions in humans.

Phenylalanine deficiency, though rare, because of their natural presence in most food systems can lead to poor weight gain in infants. It may also cause eczema, fatigue, and memory problems in adults.

Phenylalanine is often in the artificial sweeteners aspartame formulation, which manufactures use to make diet sodas among other drinks. People with a rare genetic disorder called phenylketonuria (PUK) are unable to metabolize phenylalanine. As a result, they should avoid consuming foods that contain high levels of this amino acid.

The linear effect of barrel temperature and feed moisture content negatively and significantly ($p < 0.05$) affected the phenylalanine. However, linear effect of feed moisture content was negative and significant at 5 % level. The interaction effect of all the variables and quadratic effect of barrel temperature and feed moisture content were

positively and significantly ($p < 0.05$) affected the phenylalanine except the quadratic effect of screw speed which was negative (Table 2). The (first and second order) terms were significant at 5 % level. However lack of fit was not significant at 5 % level (Table 3). The R^2 for the fit was 0.9956 (99.56%). This meant that 99.56% of the total variation in phenylalanine content in the noodles analogue was explained by the regression model. The response surface plots for phenylalanine content are shown in Fig 4a-c. The model equation developed for predicting methionine was shown in equation 6.

$$\text{Phenylalanine} = 6.98 - 0.16\text{FMC} - 0.02\text{SS} + 5.23\text{E-}4\text{BT}*\text{FMC} - 7.58\text{E-}5\text{BT}*\text{SS} + 2.11\text{E-}5\text{BT}^2 + 1.48\text{E-}3\text{FMC}^2 + 9.98\text{E-}5\text{SS}^2 \dots\dots\dots (6)$$

Histidine content of Noodles Analogue

The histidine content of the noodles analogue ranged from 3.09mg/100g(at barrel temperature of 90°C, feed moisture content of 30% and screw speed of 120rpm) to 4.20mg/100g (at barrel temperature of 90, 145°C, feed moisture content of 18, 24% and screw speed of 18, 200.45rpm) showing decrease in histidine value with increase in feed moisture content and decrease in screw speed and barrel temperature respectively. Ikegwu et al. 2022 reported a range of 2.90 to 3.74mg/100g on germinated lablab bean seed, which was lower than the value in this study. Similarly, FAO/WHO (1991) reference pattern of 1.9mg/100g was also lower than the values in this work. Suffice it to be suggested that thermal processing may have positive effect on histidine than germination. Growth facilitation, creation of blood cells, tissue repair, and maintenances of special protective covering over nerve cells, are functions of histidine in human. Biochemically, the body metabolizes histidine into histamine, which is crucial for immunity, reproductive health, and digestion.

The linear effect of feed moisture content and screw speed and interaction effect of barrel temperature and screw speed were negatively and significantly ($p < 0.05$) related to histidine.

The quadratic effect of all the variables were positively and significantly ($p < 0.05$) related to histidine. The R^2 for the fit was 0.9467 (94.67%). This meant that 94.67% of the total variation in histidine in noodles analogue was explained by the regression model. The response surface plots for histidine content are shown in Fig 5a-c. The model equation developed for predicting value was shown in equation 7.

$$\text{Histidine} = - 6.98 = 0.16\text{FMC} - 0.02\text{SS} + 5.23\text{E-}4\text{BT} *\text{FMC} - 7.58\text{E-}5\text{BT}*\text{SS} + 2.11\text{E-}5\text{BT}^2 + 1.48\text{E-}3 + 9.98\text{E-}5\text{SS}^2 \text{ Equation } \dots\dots\dots (7)$$

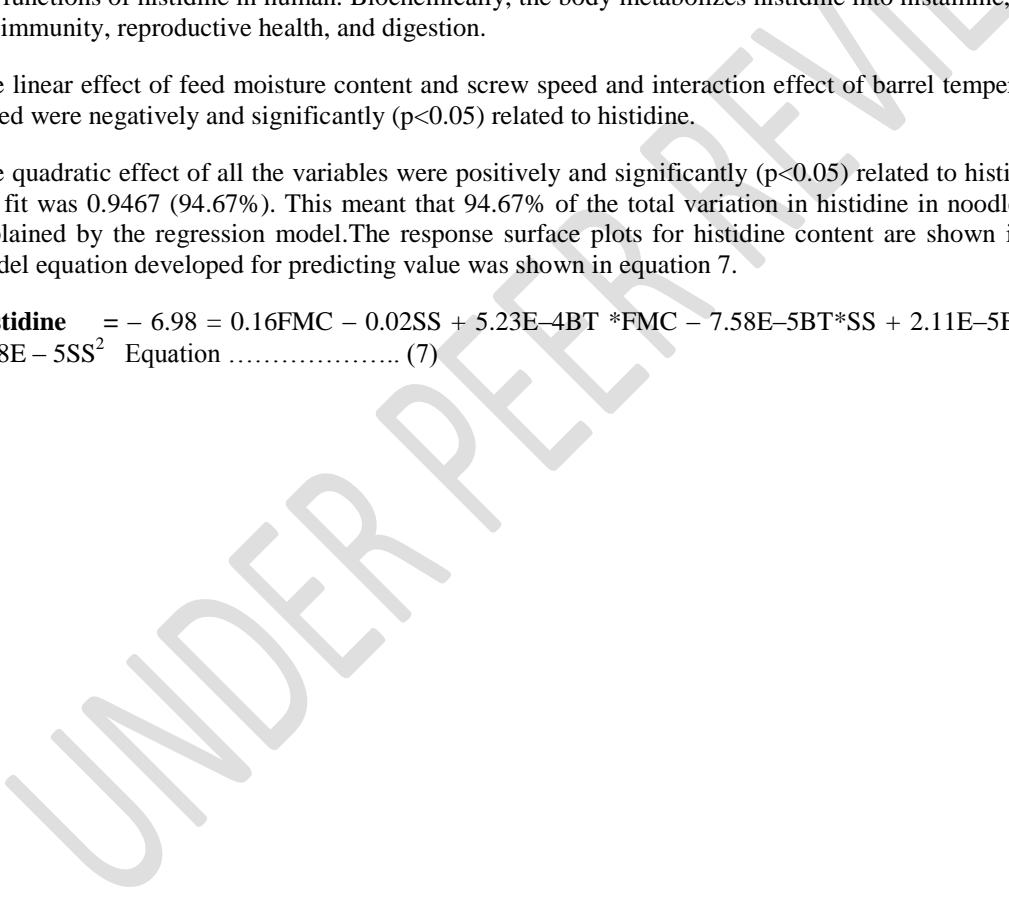


Table1: Effects of barrel temperature (BT), feed moisture content (FMC), and screw speed (SS) on the Essential Amino Acid composition of Noodles analogue

RUN	BT(°C)	FMC(%)	SCREW SPEED (rpm)	Lys	Val	Met	Phe	His	Try	Leu	Thre	Isol
1	90	18	120	6.59	5.06	1.30	5.42	3.76	1.96	6.87	3.94	3.65
2	200	18	120	6.87	5.06	1.32	5.27	3.71	0.89	7.26	4.11	3.49
3	90	30	120	6.25	4.11	1.64	5.16	3.49	1.21	7.76	4.14	3.64
4	200	30	120	7.30	4.82	1.17	5.06	4.13	1.03	7.00	4.21	3.52
5	90	18	180	6.67	4.60	1.30	5.20	4.20	1.38	7.00	4.10	3.81
6	200	18	180	4.71	4.09	1.77	5.25	3.64	1.03	7.90	4.29	3.99
7	90	30	180	7.07	4.45	1.40	5.21	4.03	1.34	7.12	3.68	3.92
8	200	30	180	5.89	4.65	1.38	5.31	4.17	1.87	7.97	3.76	4.13
9	52.5	24	150	5.31	4.88	0.86	5.27	3.84	1.28	7.36	4.23	4.87
10	237.5	24	150	4.55	5.05	0.86	5.23	3.91	1.83	7.56	4.44	4.91
11	145	13.91	150	6.85	4.85	1.78	5.45	3.74	1.44	7.20	3.82	3.06
12	145	34.09	150	7.55	4.52	1.73	5.28	3.95	1.52	7.08	3.55	3.18
13	145	24	99.55	7.52	4.35	1.60	5.07	3.70	1.44	7.03	4.18	2.91
14	145	24	200.45	6.41	3.81	1.78	5.09	4.20	1.67	7.12	3.94	3.56
15	145	24	150	5.52	3.99	2.11	5.20	3.58	1.75	6.74	4.01	3.02
16	145	24	150	5.52	4.21	2.24	5.19	3.58	1.94	6.88	4.04	3.07
17	145	24	150	5.52	4.24	1.84	5.22	3.71	1.63	6.88	3.92	3.40
18	145	24	150	5.52	3.90	1.84	5.19	3.81	1.63	6.88	4.01	3.40
19	145	24	150	4.99	4.20	1.99	5.21	3.74	1.63	6.88	4.14	3.21
20	145	24	150	6.20	4.10	2.01	5.19	3.73	1.94	6.88	4.01	3.20

Where BT = Barrel temperature; FMC = Feed moisture content; Lys = Lysine; Val = Valine; Meth - Methionine; Phe = Phenylalanine; His = Histidine; Try = Tryptophan; Leu = Leucine, Thre = Threonine; Isol = Isoleucine;

Table 2: Estimated regression coefficients of the fitted second order polynomial representing the relationship between the response and the process variable

Response coefficient	Lysine		Valine		Methionine		Phyalanine		Histidine		Tryptophan		Threonine		Isoleucine		Leucine	
	Coefficient	P-value	Coefficient	P-value	Coefficient	P-value	Coefficient	P-value	Coefficient	P-value	Coefficient	P-value	Coefficient	P-value	Coefficient	P-value	Coefficient	P-value
Linear																		
β_0	25.816		14.45		-6.09		7.52		6.98		4.85		0.47		7.46		6.93	
β_1	0.53*	0.0118	-0.029	0.0820	0.04	0.9589	-7.28E3*	0.0007	-6.90E3	0.2777	-0.01*	0.0009	-8.15E3*	0.0008	-0.06	0.6667	-0.01*	0.0021
β_2	-0.96	0.0178	-0.54*	0.0040	0.22	0.6772	-0.15*	0.0001	0.16*	0.0064	-0.11	0.4156	0.28*	0.0001	0.03*	0.3000	0.14*	0.0462
β_3	-0.15*	0.0011	-0.02*	0.0001	0.36	0.1062	-1.04E3*	0.0353	-0.02*	0.0001	-0.01*	0.0420	0.01*	0.0004	-6.09E3*	0.0001	-0.1	0.1353
Interaction																		
β_{12}	5.87E4	0.0717	5.34E4*	0.0004	-3.71E4*	0.0112	3.41E5*	0.0052	5.23E4*	0.0001	6.72E4*	0.0002	-7.58E5	0.1892	2.65E5	0.8320	-8.33E4*	0.0001
β_{13}	-3.39E4*	0.0002	-7.80E5*	0.0037	6.83E5*	0.0172	2.95E5*	0.0001	-7.58E5*	0.0005	1.09E4*	0.0008	3.03E6	0.7838	5.07E5	0.0637	8.50E5*	0.0001
β_{23}	1.03E4*	0.0825	1.10E3*	0.0002	-3.33E4*	0.1593	3.82E4*	0.0001	1.53E4*	0.2874	9.82E4*	0.0009	-8.61E4*	0.0001	1.60E4	0.4907	-1.00E3*	0.0001
Quadratic																		
β_{11}	-7.10E5*	0.0134	1.01E4*	0.0001	-1.33E4*	0.0001	6.28E6*	0.0001	2.11E5*	0.0058	-8.20E5*	0.0001	3.68E5*	0.0001	1.95E4*	0.0001	6.93E5*	0.0001
β_{22}	0.02*	0.0001	5.65E5*	0.0001	-2.40*	0.0150	1.61E3*	0.0001	1.48E3*	0.0155	-2.71E3*	0.0062	-3.03E3*	0.0001	-1.01E3	0.2507	2.70E3*	0.0001
β_{33}	5.5E4*	0.0001	-9.73E6	0.7382	-1.20*	0.0044	-4.76E5*	0.0001	9.98E5*	0.0006	-7.90E5*	0.0308	1.58E5	0.3083	4.61E6	0.8924	8.20E3*	0.0001
R^2	0.9534		0.9700		0.9562		0.9956		0.9467		0.9503		0.9709		0.9781		0.9889	
Adj. R^2	0.9114		0.9430		0.9168		0.9916		0.8987		0.9055		0.9447		0.9584		0.9789	

*Significant at 5% level

β_1 = Barrel temperature (BT); β_2 = Feed moisture content (FMC); β_3 = Screw speed (SS); β_{12} = BT* FMC; β_{13} = BT*SS; β_{11} = BT²; β_{22} = FMC²; β_{33} = SS²

Table 3: Analysis of variance for the fitted second order Polynomial model as per CCRD

Sum of squares										
	Df	Lysine	Valine	Methionine	Phenylalanine	Histidine	Tryptophan	Threonine	Isoleucine	Leucine
Regression										
First order terms	3	2.79 ^a	0515 ^b	0.041323 ^b	0.035327 ^a	0.302314 ^b	0.32132 ^b	0.217 ^a	0..537543 ^b	0.051398 ^a
Second order terms	6	12.34 ^b	61.44311 ^b	2.989 ^b	0.137212 ^b	0.59505 ^b	2.0039 ^a	0.578102 ^b	5.076522 ^b	1.868 ^a
Total	9	15.13	1.95811	3.030323	0.172539	0.897364	2.32432	0.795102	5.614065	1.99
Residual										
Lack of fit	5	0.00000534 ^b	0.0000002.04 ^b	0.000280 ^b	0.0000009522 ^b	0.003181 ^b	4.86590 ^a	0.000002572 ^b	0.0002611 ^b	0.003343 ^b
Pure error	5	0.74	0.094	0.12	0.0008000	0.045	0.12	0.025	0.13	0.017
Total error	10	0.74	0.094	0.12	0.0008010	0.048	0.12	0.025	0.13	0.020
Grand total	19	15.87	2.05211	3.150323	0.17334	0.945354	2.44432	0.820102	5.7440653	1.9393

Significant at P < 0.05

No Significant at P < 0.05

Tryptophan content of Noodles Analogue

Tryptophan content of the noodles analogue ranged from 0.83 mg/100 g (at barrel temperature of 237.5 °C, feed moisture content of 24 %, and screw speed of 150 rpm) to 1.96 mg/100 g (at barrel temperature of 90 °C, feed moisture content of 18 % and screw speed of 120 rpm). The result indicated decrease in tryptophan contents of noodles analogue with increase in barrel temperature, feed moisture content and screw speed respectively. Ikegwuet *al.* (2022) reported a range of 0.82 to 0.89mg/100g on germinated lablab bean seed, which was lower than the value in this study. Similarly, FAO/WHO (1991) reference pattern of 1.1mg/100g was also lower than the values in this work. Tryptophan is thought to be important in infants nutrition and is a precursor of serotonin and melatonin. Serotonin is a neurotransmitter that regulates appetites, sleep, mood and pain. Melatonin also regulates sleep. Therefore tryptophan appears to be sedative, and could be an ingredient in some sleep aids formulation. In human nutrition, tryptophan deficiency can cause a condition called pellagra, which can lead to dementia, skin rashes and digestive issues.

The linear quadratic effect of barrel temperature and screw speed were negatively and significantly ($p < 0.05$) related to tryptophan.

The interaction effect of all the independent variables were positively and significantly ($p < 0.05$) related to tryptophan (Table 2). The R^2 for the fit was 0.9503 (95.03%). This meant that 95.03% of the total variations in tryptophan in noodles analogue was explained by regression model. The response surface plots for tryptophan content are shown in Fig 6a-c. The model equation developed for predicting tryptophan was shown in equation 8.

$$\text{Tryptophan} = 4.8787 - 0.01111\text{BT} - 0.013385\text{SS} + 6.72067\text{E}4\text{BT}*\text{MC} + 1.08656\text{E}4\text{BT}*\text{SS} + 9.82123\text{E}4\text{MC}*\text{SS} - 8.19267\text{E}5\text{BT}^2 - 2.71020\text{E}3\text{MC}^2 - 7.89460\text{E}5\text{SS}^2 \quad \text{Equation (8)}$$

Leucine content of Noodles Analogue

The value of leucine content of the noodles analogue varied from 6.87mg/100g(at barrel temperature of 90°C, feed moisture content of 18% and screw speed of 120rpm) to 7.90mg/100g (at barrel temperature of 200°C, feed moisture content of 18% and screw speed of 180rpm). This showed that there was increase in leucine contents of the noodles analogue due to increase in barrel temperature, and screw speed. There was no clear effect of feed moisture content on leucine content of the noodles analogue. Fasuan *et al.* (2022) reported a value of 7.52mg/100g on ready to eat flakes from acha partially defatted sesame meal and modified corn starch, which was within the range of the present study. Ikegwuet *al.* (2022) reported a range of 7.58 to 8.10mg/100g on germinated lablab bean seed, which was higher than the value in this study. The good news is that FAO/WHO (1991) reference pattern of 6.6mg/100g was lower than the values in this work. Leucine helps regulate blood sugar levels and aids growth and repair of muscle and bone. It is also necessary for wound healing and the production of growth hormone.

The linear effect of barrel temperature negatively and significantly ($p < 0.05$) affected the leucine content; linear effect of feed moisture content positively and significantly ($p < 0.05$) affected the leucine content. The interaction effects of the independent variables negatively and significantly ($p < 0.05$) affected the leucine content except the barrel temperature and screw speed. The quadratic effect of all the independent variables positively and significant ($p < 0.05$) affected the leucine content (Table 2). The (first and second order) terms were significant at 5 % level. However, lack of fit was not significant at 5 % level (Table 3) The R^2 for the fit was 0.9889 (98.89%). This meant that 98.89% of the total variation in leucine in noodles analogue was explained by regression model. The response surface plots for leucine content are shown in Fig 7a-c. The model equation developed for predicting leucine was shown in equation 9.

$$\text{Leucine} = 6.93 - 0.01\text{BT} + 0.14\text{FMC} - 833\text{E} - 4\text{BT}*\text{FMC} + 8.50\text{E} - 5\text{BT}*\text{SS} - 1.00\text{E} - 3\text{FMC}*\text{SS} + 6.93\text{E} - 5\text{BT}^2 + 2.70\text{E} - 3\text{FMC}^2 + 8.20\text{E} - 3\text{SS}^2. \quad \text{Equation (9)}$$

Isoleucine content of Noodles Analogue

The value of isoleucine content of the noodles analogue varied from 2.91mg/100g(at barrel temperature of 145°C, feed moisture content of 24% and screw speed of 99.55rpm) to 4.91mg/100g (at barrel temperature of 237.5°C, feed moisture content of 24% and screw speed of 150rpm). This showed that there was an increase in isoleucine content of the noodles analogue due to increase in barrel temperature and screw speed. There was no clear effect of feed moisture content on the isoleucine content of the noodles analogue Ikegwu *et al.* 2022 reported a range of 4.32 to 4.76mg/100g on germinated lablab bean seed. Similarly, Fasuan *et al.* (2022) reported the value of 5.82 on ready to eat flakes from acha partially defatted sesame meal and modified corn, and both were higher than the value in some of the samples this study. Nevertheless; FAO/WHO (1991)

reference pattern of 2.8mg/100g was also lower than the values in this work. Isoleucine helps with wound healing, immunity, blood sugar regulation, and hormone production. It is primarily present in muscle tissue and regulates energy levels. Older adults may be more prone to isoleucine deficiency than younger people. This deficiency may cause muscle wasting and shaking.

The linear effect of feed moisture content positively and significantly ($p < 0.05$) affected the isoleucine content. The linear effect of screw speed, negatively and significantly ($p < 0.05$) affected the isoleucine content and the quadratic effect of the barrel temperature, positively and significantly ($p < 0.05$) affected the isoleucine. All other response coefficients were not significant at 5% level (Table 2). The (first and second order) terms and lack of fit were not significant at 5% level (Table 3). The R^2 for the fit was 0.9781. This meant that 97.81% of the total variations in isoleucine in noodles analogue was explained by regression model. The response surface plots for isoleucine content are shown in Fig 8a-c. The model equation developed for predicting isoleucine was shown in equation 10.

$$\text{Isoleucine} = 7.46 - 6.09E-3SS + 1.95E-4BT^2 \quad (10)$$

Threonine content of Noodles Analogue

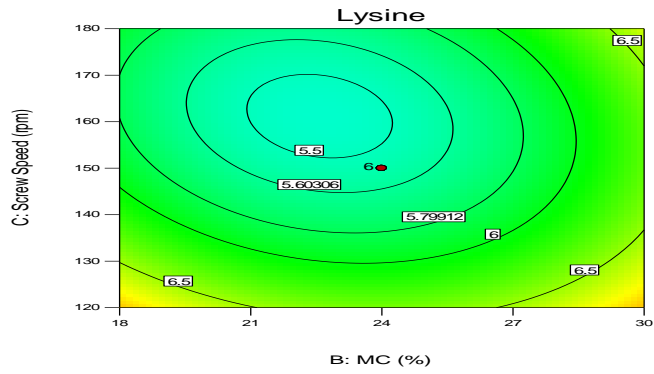
The value of threonine content of the noodles analogue varied from 3.55mg/100g (at barrel temperature of 145°C, feed moisture content of 34.09% and screw speed of 150rpm) to 4.44mg/100g (at barrel temperature of 237.5°C, feed moisture content of 24% and screw speed of 150rpm). This showed that there was a decrease in threonine content of the noodles analogue due to an increase in feed moisture content. There was no clear effect of screw speed on the threonine content of the noodles analogue. Ikegwue *et al.* (2022) reported a range of 5.10 to 5.31mg/100g on germinated lablab bean seed; Similarly, Fasua *et al.* (2022) reported the value of 4.96mg/100g on ready to eat flakes from acha partially defatted sesame meal and modified corn starch and both were higher than the values in this study. However, FAO/WHO (1991) reference pattern of 3.4mg/100g was also lower than the values in this work. Threonine is necessary for healthy skin and teeth, as it is a component in tooth enamel, collagen, and elastin. It helps aid fat metabolism and may be beneficial for people with indigestion, anxiety, and mild depression.

The linear effect of the independent variables, positively and significantly ($p < 0.05$) affected the threonine content except for barrel temperature which was negative. The interaction effects of feed moisture content and screw speed, negatively and significantly ($p < 0.05$) affected the threonine content. The quadratic effect of the barrel temperature, positively and significantly ($P < 0.05$) affected the threonine content except for moisture content which was negative (Table 2). The first order term was significant at 5% level. Whereas, the second order term and lack of fit were not significant at 5% level (Table 3). The R^2 for the fit was 0.9709. The response surface plots for threonine content are shown in Fig 9a-c. This meant that 97.09% of the total variations in threonine in the noodles analogue were explained by regression model for predicting threonine as shown in equation 11.

$$\text{Threonine} = 0.47 - 815E-3BT + 0.28FMC + 0.0155 - 7.58E-5BT * FMC + 3.03E-6BT * SS - 8.16E-4FMC * SS + 3.68E-5BT^2 - 3.30E-3FMC^2 + 1.58E-5SS^2 \dots \dots (11)$$

Design-Expert® Software
 Factor Coding: Actual
 Lysine
 ● Design Points
 7.54887
 4.55416

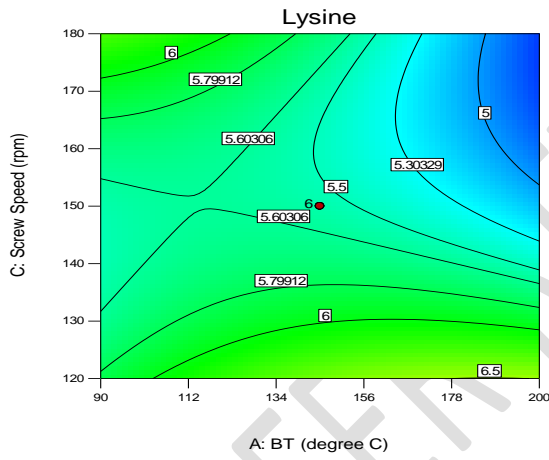
X1 = B: MC
 X2 = C: Screw Speed
 Actual Factor
 A: BT = 145



1a

Design-Expert® Software
 Factor Coding: Actual
 Lysine
 ● Design Points
 7.54887
 4.55416

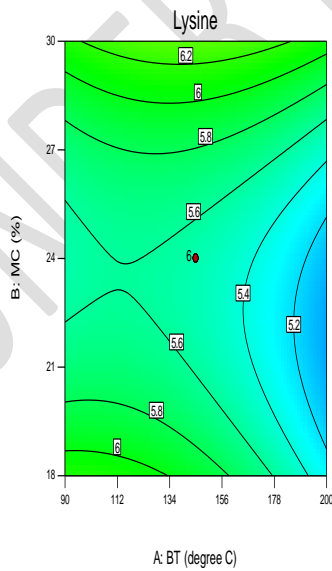
X1 = A: BT
 X2 = C: Screw Speed
 Actual Factor
 B: MC = 24



1b

Design-Expert® Software
 Factor Coding: Actual
 Lysine
 ● Design Points
 7.55
 4.55

X1 = A: BT
 X2 = B: MC
 Actual Factor
 C: Screw Speed = 150

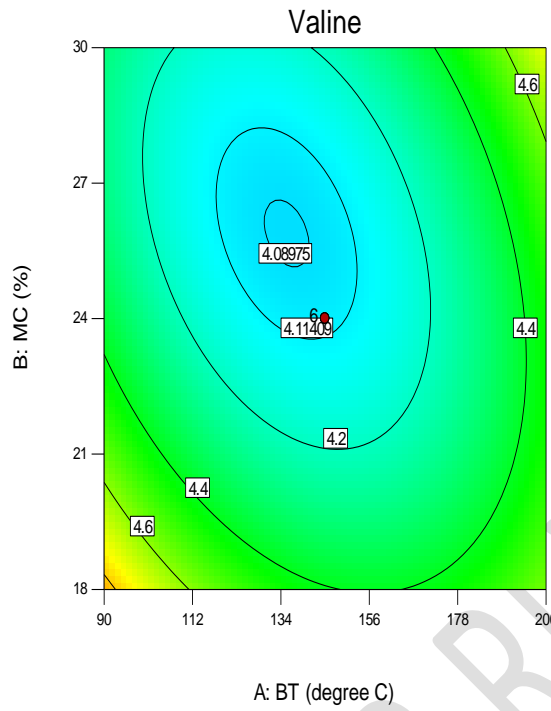


1c.

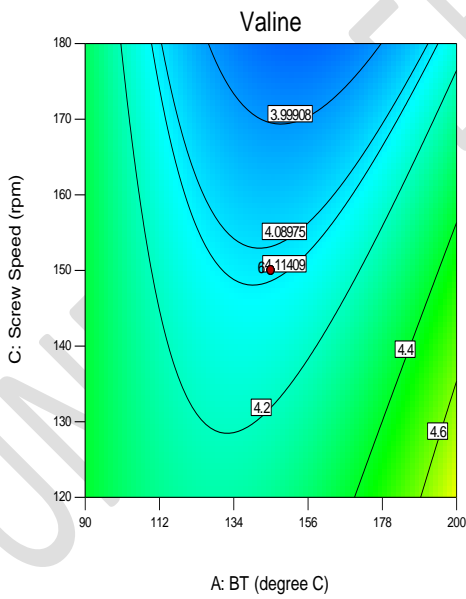
Fig 1a-c: contour plot showing the effect of barrel temperature, feed moisture content and screw speed on lysine content.

2a

Design-Expert® Software
 Factor Coding: Actual
 Valine
 ● Design Points
 5.06
 3.81
 X1 = A: BT
 X2 = B: MC
 Actual Factor
 C: Screw Speed = 150

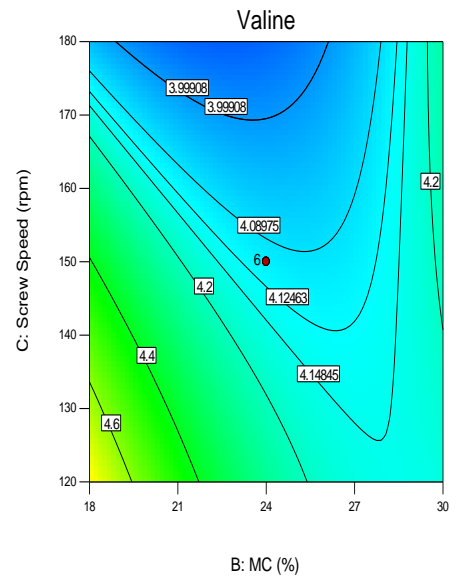


Design-Expert® Software
 Factor Coding: Actual
 Valine
 ● Design Points
 5.06405
 3.81342
 X1 = A: BT
 X2 = C: Screw Speed
 Actual Factor
 B: MC = 24



2b.

Design-Expert® Software
 Factor Coding: Actual
 Valine
 ● Design Points
 5.06405
 3.81342
 X1 = B: MC
 X2 = C: Screw Speed
 Actual Factor
 A: BT = 145

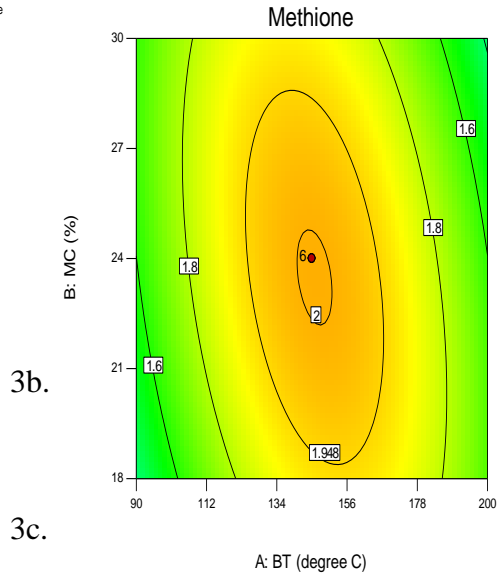


2c.

Fig 2a-c: contour plot showing the effect of barrel temperature, feed moisture content and screw speed on valine content.

Design-Expert® Software
 Factor Coding: Actual
 Methione
 ● Design Points
 2.24
 0.86

X1 = A: BT
 X2 = B: MC
 Actual Factor
 C: Screw Speed = 150

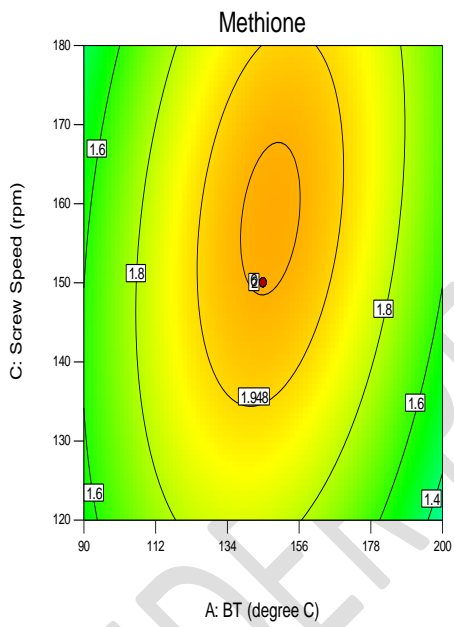


3b.

3c.

Design-Expert® Software
 Factor Coding: Actual
 Methione
 ● Design Points
 2.24
 0.856204

X1 = A: BT
 X2 = C: Screw Speed
 Actual Factor
 B: MC = 24



Design-Expert® Software
 Factor Coding: Actual
 Methione
 ● Design Points
 2.24
 0.856204

X1 = B: MC
 X2 = C: Screw Speed
 Actual Factor
 A: BT = 145

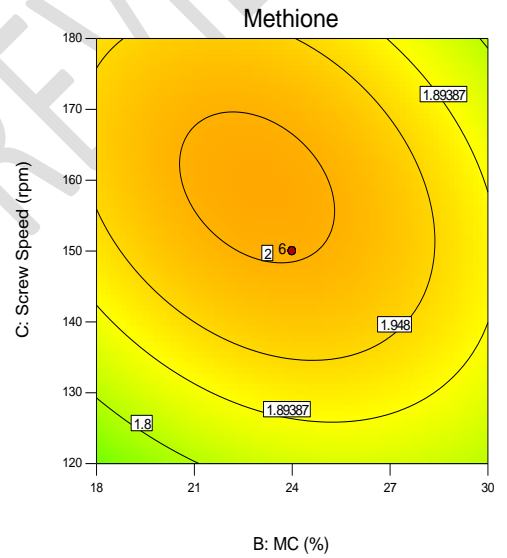
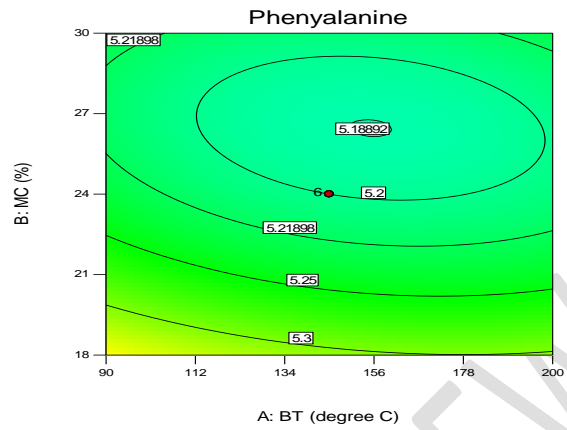


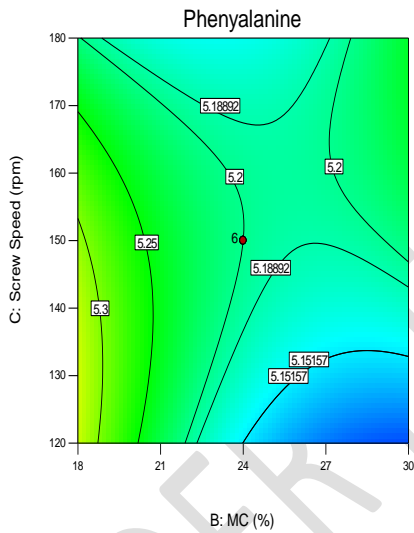
Fig3a-c: contour plot showing the effect of barrel temperature, feed moisture content and screw speed on methionine content.

4a

Design-Expert® Software
Factor Coding: Actual
Phenylalanine
● Design Points
5.45
5.06
X1 = A: BT
X2 = B: MC
Actual Factor
C: Screw Speed = 150



Design-Expert® Software
Factor Coding: Actual
Phenylalanine
● Design Points
5.44669
5.06089
X1 = B: MC
X2 = C: Screw Speed
Actual Factor
A: BT = 145



4b.

Design-Expert® Software
Factor Coding: Actual
Phenylalanine
● Design Points
5.44669
5.06089
X1 = A: BT
X2 = C: Screw Speed
Actual Factor
B: MC = 24

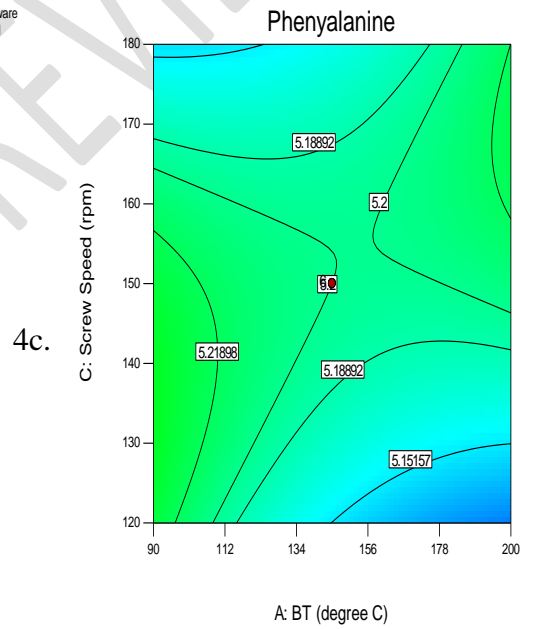
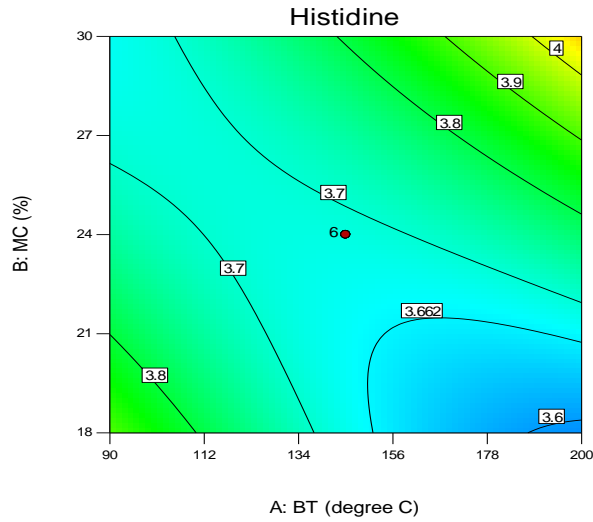


Fig 4a-c: contour plot showing the effect of barrel temperature, feed moisture content and screw speed on phenylalanine content.

Design-Expert® Software
 Factor Coding: Actual
 Histidine
 ● Design Points
 4.20
 3.49

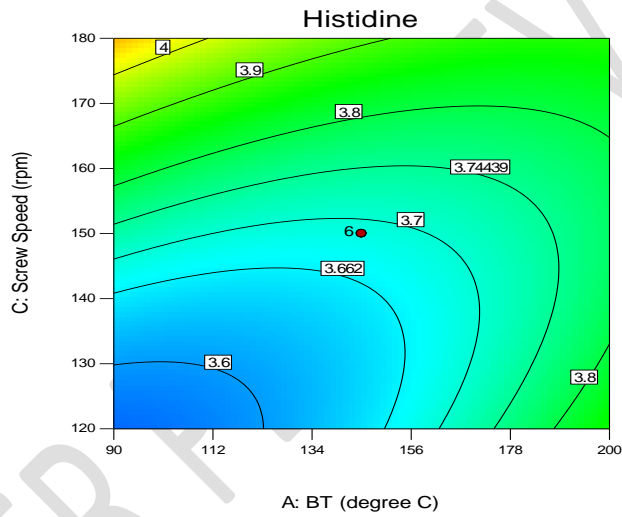
X1 = A: BT
 X2 = B: MC
 Actual Factor
 C: Screw Speed = 150



5a

Design-Expert® Software
 Factor Coding: Actual
 Histidine
 ● Design Points
 4.2
 3.49025

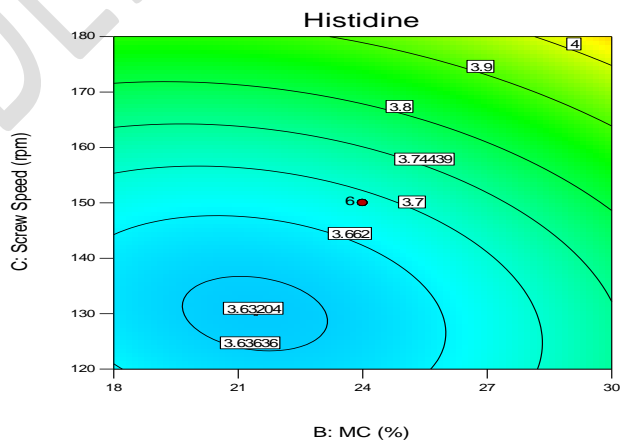
X1 = A: BT
 X2 = C: Screw Speed
 Actual Factor
 B: MC = 24



5b

Design-Expert® Software
 Factor Coding: Actual
 Histidine
 ● Design Points
 4.2
 3.49025

X1 = B: MC
 X2 = C: Screw Speed
 Actual Factor
 A: BT = 145

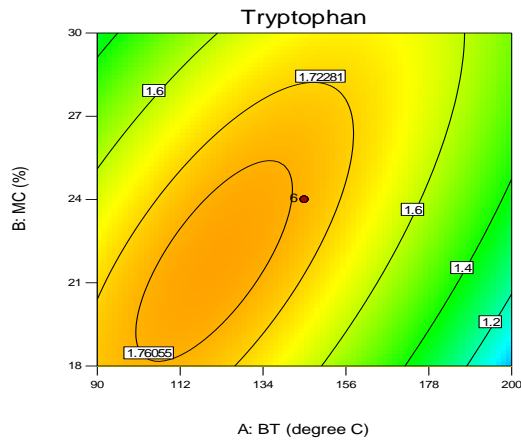


5c

Fig 5a-c: contour plot showing the effect of barrel temperature, feed moisture content and screw speed on histidine content.

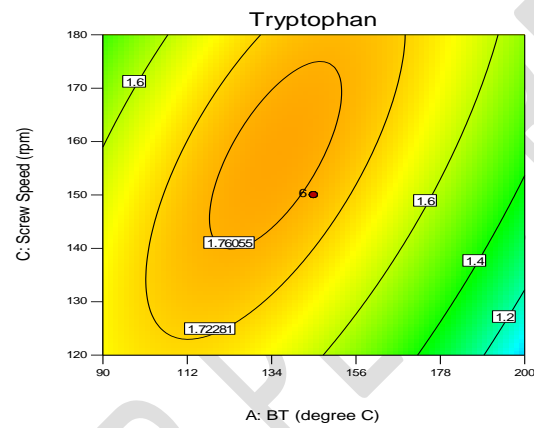
6a.

Design-Expert® Software
Factor Coding: Actual
Tryptophan
● Design Points
1.96
0.83
X1 = A: BT
X2 = B: MC
Actual Factor
C: Screw Speed = 150



6b

Design-Expert® Software
Factor Coding: Actual
Tryptophan
● Design Points
1.95666
0.826278
X1 = A: BT
X2 = C: Screw Speed
Actual Factor
B: MC = 24



6c

Design-Expert® Software
Factor Coding: Actual
Tryptophan
● Design Points
1.95666
0.826278
X1 = B: MC
X2 = C: Screw Speed
Actual Factor
A: BT = 145

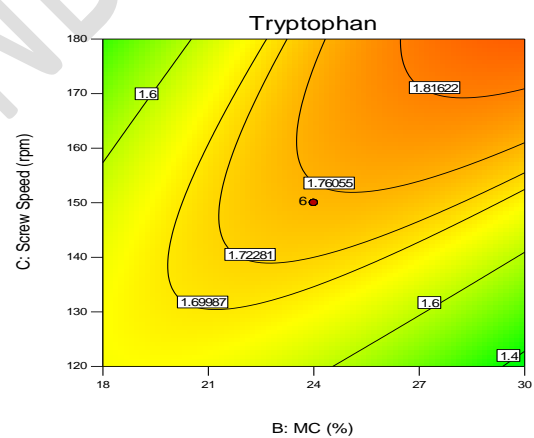
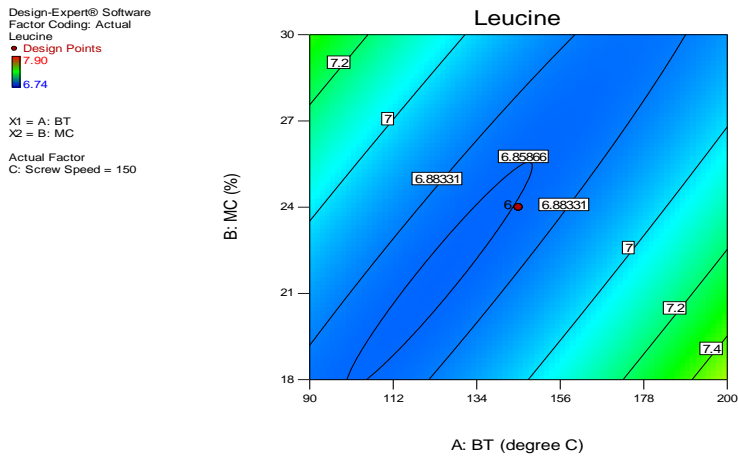
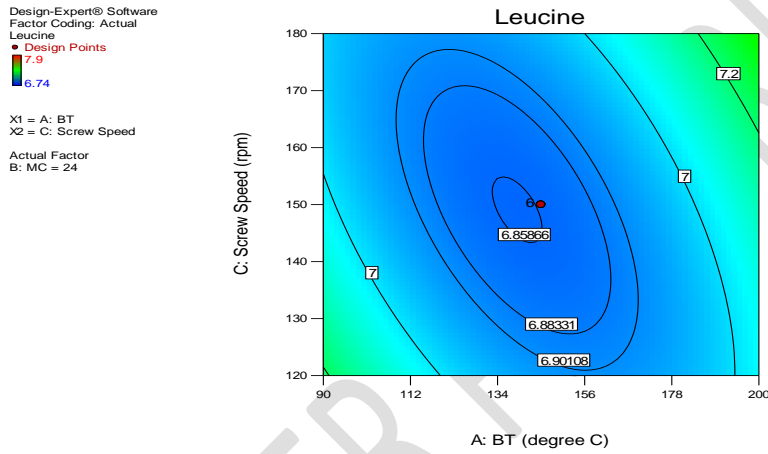


Fig 6a-c: contour plot showing the effect of barrel temperature, feed moisture content and screw speed on tryptophan content.

7a



7b.



7c.

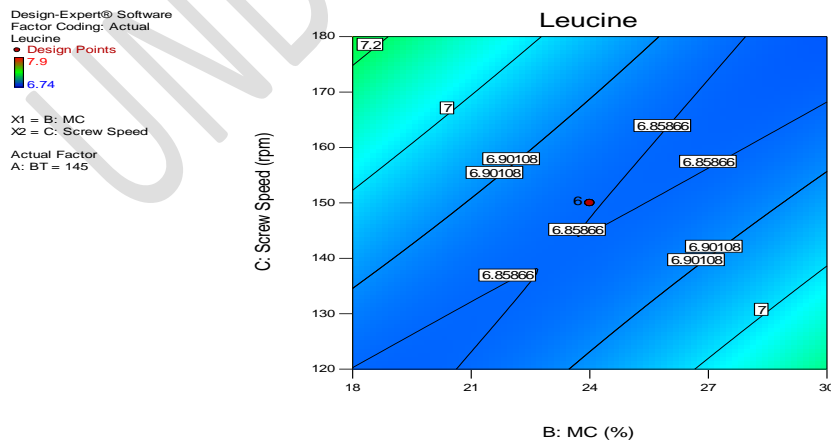
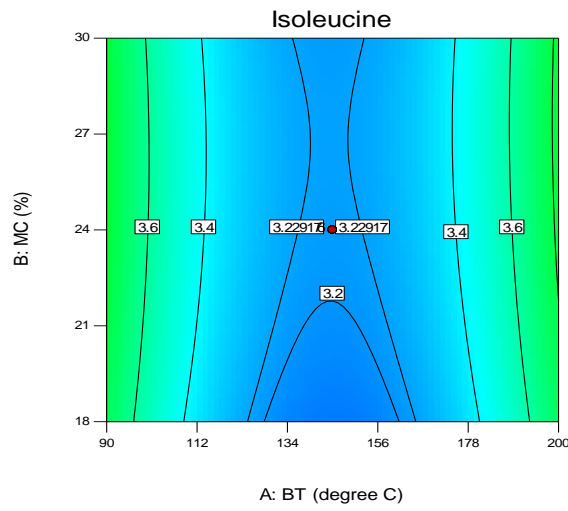


Fig 7a-c: contour plot showing the effect of barrel temperature, feed moisture content and screw speed on leucine content.

8a.

Design-Expert® Software
Factor Coding: Actual
Isoleucine
● Design Points
4.91229
2.91

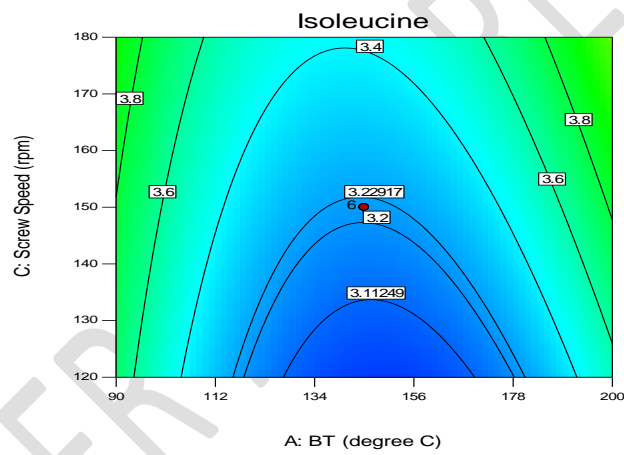
X1 = A: BT
X2 = B: MC
Actual Factor
C: Screw Speed = 150



8b

Design-Expert® Software
Factor Coding: Actual
Isoleucine
● Design Points
4.91229
2.90694

X1 = A: BT
X2 = C: Screw Speed
Actual Factor
B: MC = 24



8c.

Design-Expert® Software
Factor Coding: Actual
Isoleucine
● Design Points
4.91229
2.90694

X1 = B: MC
X2 = C: Screw Speed
Actual Factor
A: BT = 145

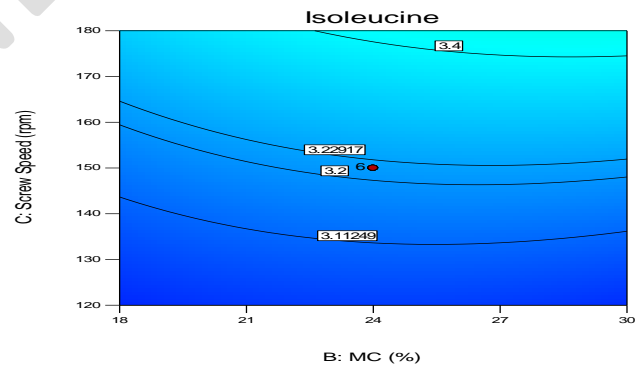
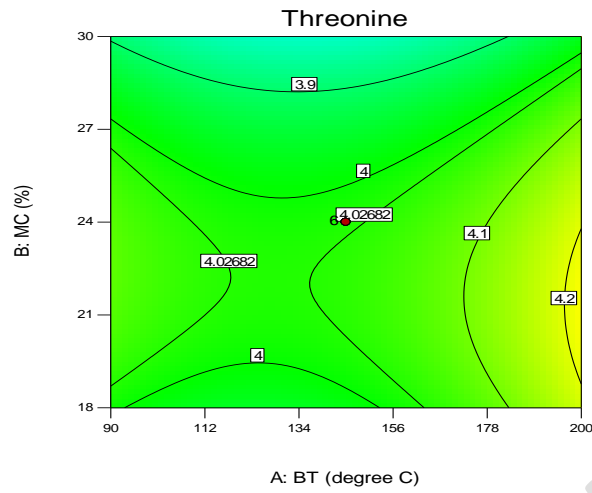


Fig 8a-c: contour plot showing the effect of barrel temperature, feed moisture content and screw speed on isoleucine content.

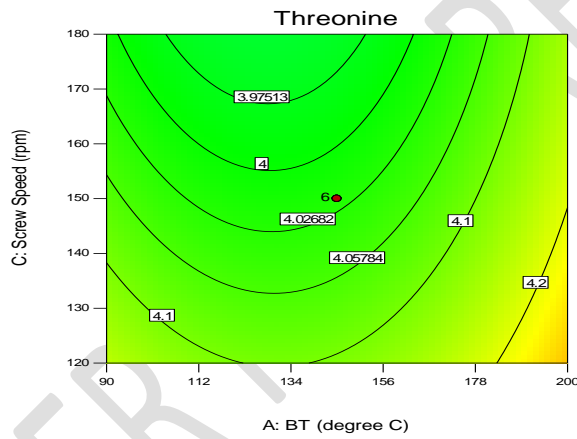
9a

Design-Expert® Software
 Factor Coding: Actual
 Threonine
 ● Design Points
 4.44
 3.55
 X1 = A: BT
 X2 = B: MC
 Actual Factor
 C: Screw Speed = 150



9b.

Design-Expert® Software
 Factor Coding: Actual
 Threonine
 ● Design Points
 4.44322
 3.54705
 X1 = A: BT
 X2 = C: Screw Speed
 Actual Factor
 B: MC = 24



9c

Design-Expert® Software
 Factor Coding: Actual
 Threonine
 ● Design Points
 4.44322
 3.54705
 X1 = B: MC
 X2 = C: Screw Speed
 Actual Factor
 A: BT = 145

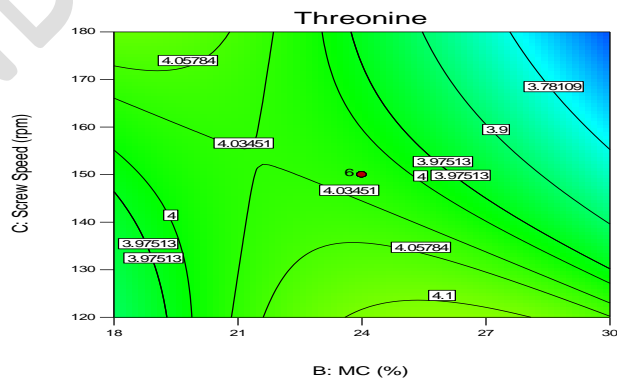


Fig 9a-c: contour plot showing the effect of barrel temperature, feed moisture content and screw speed on threonine content.

CONCLUSION

This study has shown that the essential amino acid profile of noodles analogue from aerial yam, rice, african yam bean flour blend are dependent on the extrusion process parameters (barrel temperature, screw speed and feed moisture content); because the all had significant effect ($p < 0.05$). The second order model was found to be sufficient in describing essential amino acids, the data obtained from the study could be used for control of product characteristics with more consistent quality. Traditionally, aerial yam and African Yam bean are popularly consumed in Nigeria as a local balanced meal. Expectedly, rice will contribute its sulfur containing amino acids deficient in African Yam bean. This work gives credence for supplementation and complementation in human nutrition as most of essential amino acid content of the noodles analogue were within the amount recommended for both adult and children consumers. In commercializing the product; rice, aerial yam, and african yam bean would gain global visibility as an alternative base material in noodle production.

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