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Formülasyonda Kullanılan Bileşen Oranlarının Balık Yemi Kalitesi Üzerindeki Etkisi

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Öz

Balık yetiştiriciliği, Nijerya su ürünleri yetiştiriciliği sektörünün önemli bir parçasıdır. Ancak kaliteli ve uygun fiyatlı yemlerin temin edilememesi, sektörün karşı karşıya olduğu başlıca sorunlardan biridir. Bu çalışma, bağırsak unu, sarı mısır kepeği, benni (susam) tohumu küspesi, buğday kepeği, yer fıstığı küspesi, palmiye çekirdeği küspesi, pirinç kepeği, kan unu, moringa yaprağı, kireç taşı ve kemik unu kullanılarak katma değerli bir balık yemi geliştirmek ve karakterize etmek amacıyla yürütülmüştür. Çalışmada, kısıtlı D-optimal karışım deneysel tasarımı kullanılmıştır. Formüle edilen yem örnekleri kalite özellikleri açısından değerlendirilmiştir. İncelenen kalite özellikleri; protein içeriği, nem içeriği, karbonhidrat, lipit içeriği, ham lif, kül içeriği, azotsuz öz madde (NFE), enerji değeri, suda çözünürlük indeksi, yığın yoğunluğu, çinko, magnezyum, kalsiyum, demir ve fosfor içerikleridir. Yem bileşenlerinin oranlarının bu kalite özellikleri üzerindeki etkileri araştırılmıştır. Deneysel verilerin istatistiksel analizi, formüle edilen yemin yalnızca lipit içeriği, ham lif içeriği ve enerji değerinin kullanılan bileşenlerden anlamlı şekilde etkilendiğini göstermiştir (L/Karışım p-değeri < 0,05). Yem bileşenleri arasındaki etkileşimlerin kalite özellikleri üzerindeki etkileri de belirlenmiştir. Bileşenler, ikili etkileşim düzeyinde, ham lif, çinko ve kalsiyum içeriklerini anlamlı şekilde etkilemiştir. Çinko ve kalsiyum modelleri genel olarak anlamlı bulunmamasına rağmen (model p-değeri > 0,05), analizler bazı bileşen kombinasyonlarının çinko ve kalsiyum içeriklerini istatistiksel olarak etkilediğini göstermiştir (terim p-değeri < 0,05). Bağırsak unu/benni tohumu küspesi, bağırsak unu/buğday kepeği, benni tohumu küspesi/buğday kepeği ve benni tohumu küspesi/yer fıstığı küspesi etkileşimleri, formüle edilen yemin çinko içeriğini anlamlı şekilde etkilemiştir. Buğday kepeği ile kireç taşının etkileşimi ise yemin kalsiyum içeriğini anlamlı şekilde etkilemiştir (p-değeri = 0,0422). Fosfor içeriği açısından ise ortalama değer, formüle edilen yemdeki fosfor miktarını doğru şekilde temsil eden bir ölçüt olarak belirlenmiştir.

Anahtar kelimeler: Balık yemi, Yem formülasyonu, D-optimal deney tasarımı, Tasarım matrisi, Karışım deney tasarımı

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Quality of Fish Feed as influenced by Formulation Ingredients Proportions

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Abstract

Fish farming is a crucial part of Nigeria's aquaculture industry. However, non-availability of quality and affordable feeds is one of the major challenges confronting the industry. This study was conducted to develop and characterize value-added feed from intestine meal, yellow corn bran, beni seed meal, wheat bran, groundnut meal, palm kernel cake, rice bran, blood meal, moringa leaf, limestone, and bone meal; using a constrained D-optimal mixture experimental design. Formulated feed samples were evaluated for quality properties. The quality properties of the feed evaluated were protein content, moisture content, carbohydrate, lipid content, crude fibre, ash content, nitrogen free extract, energy value, water solubility index, bulk density, zinc, magnesium, calcium, iron, and phosphorus. The effects of the ingredients proportions on the quality properties were investigated. Statistical analysis of the experimental data revealed that only the lipid, crude fibre, and energy value of the formulated feed were influenced by the formulation ingredients (L/Mixture p-value <0.05). Interaction effects of the ingredients on the quality properties of the feed were established. The ingredients interacted, at two levels, to significantly influence the crude fibre, zinc, and calcium contents of the formulated feed. Although the zinc and calcium models were non-significant (model p-value > 0.05), analysis revealed that specific ingredient combinations statistically influenced the zinc and calcium contents (terms p-value < 0.05)."Intestine meal/beni seed meal, intestine meal/wheat bran, beni seed meal/wheat bran, and beni seed meal/groundnut meal; interacted to significantly influence the zinc content of the formulated feed. Wheat bran interacted with lime stone to significantly influence the calcium content of the formulated feed (p-value = 0.0422). The mean value of phosphorus proved to be the true representation of phosphorus in the formulated feed.

Keywords: Fish feeds, Formulation, D-Optimal, Design matrix, Mixture design

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1. Introduction

Fish rearing, including freshwater fish like tilapia, carp, and catfish as well as marine fish, like salmon, trout, and shrimp; is referred to as aquaculture. Since feed typically constitutes the largest single expense in fish farming operations, developing value-added feeds is essential for the success of the industry [1]. Utilizing byproducts from agriculture, food processing and other industries to formulate feed presents a sustainable and cost-effective solution. The main goal of fish feed formulation is to develop feed that meets dietary needs of the fishes. The growth and well-being of farmed fish are directly impacted by the nutritional value of feeds [2]. Depending on the particular dietary needs of each species and life stage, a fish's diet should normally include protein, fats, carbohydrates, vitamins, and minerals. Balanced diet is essential to aquaculture operations' sustainability, promoting sustainable fish production and enhancing global food security [3]. Fingerlings and juveniles have significant protein requirements, whereas growers and mature fish demand progressively less protein. Similarly, fingerlings require vital fatty acids for rapid growth, while growers and mature fish require lower fat intake. Juveniles need a significant energy source, carbohydrates remain a significant source of energy for grower, while the adults use carbohydrates more effectively for energy. Vitamins are essential for immunological response, metabolism, and general health. For fingerlings vitamins A and D are necessary for bone development and metabolism, and vitamin C promotes collagen synthesis and immunological function. Poor growth and increased vulnerability to disease can result from deficiencies of vitamin A, D, E, and C levels in growers feed. Vitamins A, D, E, and C support the maintenance of both general bodily health and reproductive health in adults. The growth of the skeleton, metabolic control, and general health of fishes depend on minerals in fish feeds. Fiber promotes effective feed passage through the gut and helps in digestion [4–14]. The goal of this study was to develop and characterize value-added fish feed from intestine meal, yellow corn bran, beni seed meal, wheat bran, groundnut meal, palm kernel cake, rice bran, blood meal, moringa leaf, limestone, and bone meal; using mixture design experimental methodology.

Intestine meal from livestock is rich in protein (40-50%) and essential amino acids. It offers comparable quality to fishmeal when properly processed. Its high palatability and digestibility make it suitable for fish diets, promoting good feed intake and growth [15]. The cost-effectiveness of intestine meal, being a by-product of the meat industry, can significantly reduce feed costs, enhancing the profitability of fish farming. Research indicates that intestine meal can replace up to 50% of fishmeal in Titus diets without negative impacts on growth or feed conversion efficiency [16, 17]. Yellow corn bran, a by-product of maize milling, is widely available and rich in carbohydrates and fibre. It is often used in fish feed formulations to provide energy and improve feed bulk. It is a good source of energy, it contains about 10-12% protein and is a good source of dietary fiber, essential for maintaining gut health and improving digestion in fish. It can also provides essential vitamins and minerals, including B vitamins and magnesium, supporting overall fish health and growth [15–18]. Beni seed (Sesame) meal, derived from the residuals after oil extraction from sesame seeds, is an emerging alternative protein source for fish feed. It is rich in protein, containing approximately 35-45% protein, and essential amino acids like methionine and cysteine, which are vital for fish growth and health. Additionally, beni seed meal provides a good balance of essential fatty acids, contributing to the overall energy density of the diet and supporting optimal growth performance in Titus. Furthermore, beni seed meal contains beneficial minerals such as calcium, phosphorus, and magnesium, enhancing bone health and metabolic functions in fish. [15 – 17]. Wheat bran, a by-product of wheat milling, is rich in fibre and provides essential nutrients such as protein, vitamins, and minerals, making it a valuable component in aquaculture diets. Wheat bran typically contains around 14-18% protein, along with significant amounts of B vitamins, magnesium, phosphorus, and zinc, which are crucial for the growth and health of fish [18, 19]. Wheat bran also acts as an excellent binder in pelleted feeds, improving the physical quality and water stability of the pellets, which is essential for maintaining feed integrity in aquatic environments [17]. Studies indicate that wheat bran can replace up to 30% of the conventional feed ingredients in Titus diets, contributing to reduced feed costs while maintaining nutritional adequacy [15]. Groundnut meal, a by-product of groundnut (peanut) oil extraction, is a valuable protein source for fish. Rich in protein (approximately 45-50%) and essential amino acids, groundnut meal provides an excellent alternative to conventional protein sources like fishmeal. Its high nutritional value and relatively low cost make it an attractive option for formulating cost-effective and sustainable fish diets. Research indicates that groundnut meal can replace up to 30% of fishmeal in Titus diets without compromising growth performance or feed efficiency. It is also rich in energy, primarily from lipids and carbohydrates, which contribute to the overall energy density of fish diets [15, 17]. Palm kernel cake, a by-product of palm oil extraction, is an economically viable and nutritionally valuable ingredient for fish feed. With a protein content of 14-20% and a high energy level from lipids, palm kernel cake supports the growth and development of fishes. The high fiber content in palm kernel cake can benefit gut health but must be managed to prevent negative impacts on nutrient digestibility [15, 18]. Rice bran is a byproduct of the milling process that transforms brown rice into white rice. It contains various antioxidants that can benefit human health. A significant fraction of rice bran consists of 12%–13% oil and 4.3% highly unsaponifiable components. This fraction is rich in tocotrienols (a form of vitamin E), gamma-oryzanol, and beta-sitosterol, which may help lower plasma levels of various lipid profile parameters. Additionally, rice bran is high in dietary fibres, including beta-glucan, pectin, and gum, and contains ferulic acid, a component of non-lignified cell wall structures. Blood meal, a by-product of slaughterhouse operations, is a highly nutritious and cost-effective protein source for fish feed in Nigeria. It contains approximately 80-85% crude protein, making it one of the richest protein sources available for fish feed formulations. Blood meal is also rich in essential amino acids, particularly lysine, which supports optimal growth and health in fish [18, 19]. Moringa leaves are high in protein (20-30%), essential amino acids, vitamins (A, B, and C), and minerals (calcium, potassium, and iron), making them an excellent supplement in fish diets. Studies have shown that moringa leaf can replace a significant portion of conventional feed ingredients without negatively impacting fish growth or feed efficiency [16, 18-20]. Lime stone is the most widely used source of calcium in livestock diets, consisting primarily of calcium carbonate. Calcitic limestone contains 36-38% calcium and can be safely offered free choice (ad lib) when mixed with salt [21]. Bone meal, a by-product of the meat processing industry, is an important mineral supplement. It is rich in calcium and phosphorus, which are essential for bone development and metabolic processes in fish [16]. Including bone meal in fish diets helps meet dietary mineral requirements, promoting growth and preventing deficiencies. Bone meal also contains trace elements such as magnesium, zinc, and iron, which are crucial for immune function and enzyme activity [15, 18, 19].

2. Materials And Methods

Materials

Intestine meal, yellow corn bran, beni seed meal, wheat bran, groundnut meal, palm kernel cake, rice bran, blood meal, moringa leaf meal, limestone, and bone meal were the resources used for the feed formulation. They were sourced locally from Minna.

Experimental design for the fish feed formulation

An eleven-components constrained D-optimal mixture experimental design, with 76 randomized experimental runs, was employed. The formulation constraints and the design matrix are presented in Tables 1 and 2.

Table 1. Design Constraint for feed formulation

Low Limit (%)		Constraint		High Limit (%)
5	≤	x_1 : Intestine Meal	≤	40
3	≤	x_2 : Yellow Corn Bran	≤	40
5	≤	x_3 : Beni seed Meal	≤	20
2	≤	x_4 : Wheat Bran	≤	30
2	≤	x_5 : Groundnut Meal	≤	20
1	≤	x_6 : Palm Kernel Cake	≤	10

Table 1. (Continue) Design Constraint for feed formulation

Low Limit (%)		Constraint		High Limit (%)
5	≤	x_7 : Rice Bran	≤	35
2	≤	x_8 : Blood Meal	≤	10
1	≤	x_9 : Moringa Leaf	≤	5
0	≤	x_{10} : Lime Stone	≤	5
1	≤	x_{11} : Bone Meal	≤	5
		$x_1+x_2+x_3+x_4+x_5+x_6+x_7+x_8+x_9+x_{10}+x_{11}$	=	**100**

Table 2. Design Matrix for feed formulation

Run	x_1	x_2	x_3	x_4	x_5	x_6	x_7	x_8	x_9	x_{10}	x_{11}
1	5	14	5	24	20	10	5	2	5	5	5
2	5	40	5	2	2	1	29	10	5	0	1
3	5	3	5	30	6	10	35	4	1	0	1
4	5	29	20	29	2	1	5	2	5	1	1
5	21	3	11	2	2	10	35	10	1	0	5
6	19	3	20	30	9	1	5	6	1	1	5
7	5	3	20	30	18	10	5	2	1	5	1
8	5	3	20	30	18	10	5	2	1	5	1
9	5	40	12	12	4	1	5	10	1	5	5
10	35	3	5	6	2	2	35	2	5	0	5
11	40	23	5	3	2	1	5	10	5	1	5
12	5	3	6	4	20	10	35	10	1	5	1
13	20	3	5	2	20	1	35	10	3	0	1
14	37	3	5	30	2	1	5	2	5	5	5
15	16	3	13	12	10	1	27	8	5	0	5
16	5	18	5	14	10	10	31	4	1	1	1
17	5	33	16	2	20	6	5	2	1	5	5
18	5	12	5	26	20	10	5	10	1	1	5
19	5	3	20	30	2	10	10	10	5	0	5
20	5	40	20	2	2	10	5	10	1	0	5
21	38	13	13	5	2	9	8	3	3	3	3
22	5	12	20	9	18	1	24	2	5	3	1
23	15	14	5	30	2	10	5	10	1	5	3
24	24	14	5	8	12	4	25	2	1	2	3
25	5	18	5	30	20	1	5	10	5	0	1
26	40	3	20	2	20	5	5	2	1	1	1
27	40	3	18	2	11	1	9	10	5	0	1
28	21	40	5	3	14	1	5	2	3	1	5
29	5	40	20	2	2	1	13	2	5	5	5
30	5	35	20	2	2	10	5	10	5	5	1
31	10	3	5	30	2	1	35	10	1	2	1
32	26	3	20	14	2	3	17	6	3	5	1

Table 2. (Continue)Design Matrix for feed formulation

Run	x_1	x_2	x_3	x_4	x_5	x_6	x_7	x_8	x_9	x_{10}	x_{11}
33	5	40	5	26	2	10	5	2	1	3	1
34	40	3	5	2	2	10	24	10	3	0	1
35	5	3	5	2	20	10	35	10	5	0	5
36	33	3	20	2	2	10	5	10	5	5	5
37	5	3	20	25	20	1	5	10	1	5	5
38	15	14	5	30	2	10	5	10	1	5	3
39	19	19	20	2	11	1	16	10	1	0	1
40	5	26	20	2	20	5	5	10	5	1	1
41	5	3	20	2	20	10	32	2	1	0	5
42	5	40	5	2	20	10	10	2	5	0	1
43	7	3	20	2	13	1	35	10	1	5	3
44	5	3	20	13	2	10	34	2	1	5	5
45	15	3	5	30	2	10	22	2	5	5	1
46	23	9	5	20	10	10	5	10	5	2	1
47	26	34	9	2	4	7	5	10	1	1	1
48	23	40	20	2	2	1	5	2	1	3	1
49	23	10	6	2	2	1	35	10	5	5	1
50	5	40	5	2	20	1	5	10	5	5	2
51	19	18	7	21	2	1	19	9	2	1	1
52	17	25	5	2	6	10	18	2	5	5	5
53	5	29	13	15	13	1	16	2	5	0	1
54	5	26	5	30	2	1	19	2	5	0	5
55	5	38	5	2	2	1	35	6	1	0	5
56	27	3	20	12	20	1	5	2	5	0	5
57	5	3	19	20	20	1	27	2	1	0	2
58	25	3	20	22	2	10	5	10	1	1	1
59	20	3	20	2	2	10	35	2	5	0	1
60	40	3	5	10	19	1	5	10	1	5	1
61	5	35	20	2	2	10	5	10	5	5	1
62	5	3	20	30	2	10	10	10	5	0	5
63	5	3	5	19	20	1	22	10	5	5	5
64	40	3	5	2	20	10	5	5	5	0	5
65	40	13	20	2	2	1	13	2	1	1	5
66	29	40	5	2	2	1	5	5	1	5	5
67	5	34	5	7	3	1	35	2	2	5	1
68	37	3	11	2	2	1	35	2	1	5	1
69	26	26	5	17	13	1	5	5	1	0	1
70	31	3	5	30	19	1	5	2	3	0	1
71	20	3	20	2	2	10	35	2	5	0	1
72	18	3	7	2	20	10	26	2	5	5	2
73	30	28	5	11	2	10	5	2	1	1	5
74	5	3	5	23	20	1	30	2	1	5	5
75	40	25	5	2	3	7	5	2	5	5	1
76	38	11	5	2	20	10	5	2	1	5	1

2. Experımental Data

Using standard procedures [22], the formulated fish feed samples were analyzed and evaluated for proximate properties, some physical & physico-chemical properties, and mineral contents. The data generated are presented in Tables 3 and 4.

Table 3. Formulated Feeds Quality Properties

Run	y_{prot}	y_{mc}	y_{cho}	y_{lip}	y_{cf}	y_{ash}	y_{nfe}	y_{ev}
1	25.50	6.25	26.79	10.71	18.0	12.75	26.79	3055.5
2	36.69	5.77	24.84	10.03	13.0	9.67	24.84	3363.9
3	49.94	5.79	11.39	12.46	10.3	10.17	11.39	3574.6
4	52.50	5.81	9.34	8.85	12.5	11.00	9.34	3270.1
5	54.69	6.37	6.72	12.22	12.5	7.50	6.72	3556.2
6	38.06	6.05	19.34	14.75	11.0	10.80	19.34	3623.5
7	33.75	6.25	29.32	11.18	9.5	10.00	29.32	3529.0
8	39.38	6.02	14.49	12.79	16.0	11.33	14.49	3305.5
9	55.06	5.80	5.92	12.34	11.0	9.88	5.92	3549.8
10	40.00	5.64	17.46	10.10	17.0	9.80	17.46	3207.4
11	39.56	6.49	28.13	5.52	10.0	10.30	28.13	3204.4
12	33.00	6.13	32.26	9.36	9.3	10.00	32.26	3452.8
13	43.94	5.92	12.96	11.18	16.0	10.00	12.96	3282.2
14	44.25	5.90	11.54	11.71	16.5	10.10	11.54	3285.5
15	56.13	5.97	9.50	12.66	7.0	8.75	9.50	3764.2
16	32.44	6.29	30.09	9.03	12.3	9.90	30.09	3313.9
17	42.63	6.21	18.69	9.15	12.0	11.33	18.69	3275.9
18	43.63	6.78	12.00	9.43	15.0	13.17	12.00	3073.5
19	43.63	5.96	18.91	11.18	8.5	11.83	18.91	3507.4
20	32.94	5.79	34.84	9.43	10.5	6.50	34.84	3559.9
21	33.19	6.00	35.36	5.45	9.0	11.00	35.36	3232.5
22	38.58	6.02	28.86	10.54	7.5	8.50	28.86	3646.2
23	44.69	5.76	20.52	11.78	7.0	10.25	20.52	3668.6
24	37.69	6.05	24.76	8.00	12.5	11.00	24.76	3218.0
25	43.63	5.74	24.14	8.12	7.5	10.88	24.14	3441.2
26	37.94	5.75	22.95	9.03	12.5	11.83	22.95	3248.3
27	49.25	6.01	13.41	14.33	5.0	12.00	13.41	3796.1
28	43.75	6.22	24.13	7.07	8.0	10.83	24.13	3351.5
29	22.06	5.72	45.96	6.38	9.0	10.88	45.96	3295.0
30	32.63	6.81	26.08	7.74	15.0	11.75	26.08	3044.6
31	33.63	5.99	27.53	15.03	8.0	9.83	27.53	3798.7
32	41.75	5.79	22.43	10.83	8.5	10.70	22.43	3541.9
33	49.44	5.61	19.02	9.43	7.5	9.00	19.02	3587.1
34	26.25	6.13	37.18	5.61	14.5	10.33	37.18	3042.1
35	36.50	5.63	20.74	11.96	15.5	9.67	20.74	3366.0
36	31.94	5.92	32.53	10.61	7.0	12.00	32.53	3533.7
37	40.25	5.01	9.09	25.65	11.0	9.00	9.09	4282.1
38	41.06	5.59	24.45	9.65	9.3	10.00	24.45	3488.9
39	45.56	5.57	15.58	12.91	10.8	9.63	15.58	3607.5

Table 3. (Continue) Formulated Feeds Quality Properties

Run	y_{prot}	y_{mc}	y_{cho}	y_{lip}	y_{cf}	y_{ash}	y_{nfe}	y_{ev}
40	45.63	5.86	16.38	10.47	12.5	9.17	16.38	3422.3
41	39.25	5.44	24.17	7.97	12.5	10.67	24.17	3254.1
42	45.19	5.90	19.61	9.30	10.0	10.00	19.61	3429.0
43	39.38	5.80	25.93	8.40	11.0	9.50	25.93	3368.0
44	25.50	6.25	26.79	10.71	18.0	12.75	26.79	3055.5
45	36.69	5.77	24.84	10.03	13.0	9.67	24.84	3363.9
46	49.94	5.79	11.39	12.46	10.3	10.17	11.39	3574.6
47	52.50	5.81	9.34	8.85	12.5	11.00	9.34	3270.1
48	54.69	6.37	6.72	12.22	12.5	7.50	6.72	3556.2
49	38.06	6.05	19.34	14.75	11.0	10.80	19.34	3623.5
50	33.75	6.25	29.32	11.18	9.5	10.00	29.32	3529.0
51	39.38	6.02	14.49	12.79	16.0	11.33	14.49	3305.5
52	55.06	5.80	5.92	12.34	11.0	9.88	5.92	3549.8
53	40.00	5.64	17.46	10.10	17.0	9.80	17.46	3207.4
54	39.56	6.49	28.13	5.52	10.0	10.30	28.13	3204.4
55	33.00	6.13	32.26	9.36	9.3	10.00	32.26	3452.8
56	43.94	5.92	12.96	11.18	16.0	10.00	12.96	3282.2
57	44.25	5.90	11.54	11.71	16.5	10.10	11.54	3285.5
58	56.13	5.97	9.50	12.66	7.0	8.75	9.50	3764.2
59	32.44	6.29	30.09	9.03	12.3	9.90	30.09	3313.9
60	42.63	6.21	18.69	9.15	12.0	11.33	18.69	3275.9
61	43.63	6.78	12.00	9.43	15.0	13.17	12.00	3073.5
62	43.63	5.96	18.91	11.18	8.5	11.83	18.91	3507.4
63	32.94	5.79	34.84	9.43	10.5	6.50	34.84	3559.9
64	33.19	6.00	35.36	5.45	9.0	11.00	35.36	3232.5
65	38.58	6.02	28.86	10.54	7.5	8.50	28.86	3646.2
66	44.69	5.76	20.52	11.78	7.0	10.25	20.52	3668.6
67	37.69	6.05	24.76	8.00	12.5	11.00	24.76	3218.0
68	43.63	5.74	24.14	8.12	7.5	10.88	24.14	3441.2
69	37.94	5.75	22.95	9.03	12.5	11.83	22.95	3248.3
70	49.25	6.01	13.41	14.33	5.0	12.00	13.41	3796.1
71	43.75	6.22	24.13	7.07	8.0	10.83	24.13	3351.5
72	22.06	5.72	45.96	6.38	9.0	10.88	45.96	3295.0
73	32.63	6.81	26.08	7.74	15.0	11.75	26.08	3044.6
74	33.63	5.99	27.53	15.03	8.0	9.83	27.53	3798.7
75	41.75	5.79	22.43	10.83	8.5	10.70	22.43	3541.9
76	49.44	5.61	19.02	9.43	7.5	9.00	19.02	3587.1

$y_{prot} = Protein(\%)$, $y_{mc} = Moisture\ Content(\%)$, $y_{cho} = Carbohydrate(\%)$,
$y_{lip} = Lipid\ Content(\%)$, $y_{cf} = Crude\ Fiber(\%)$, $y_{ash} = Ash\ Content(\%)$,
$y_{nfe} = Nitrogen\ Free\ Extract(\%)$, $y_{ev} = Energy\ Value(kcal)$,

Table 4: Formulated Feeds Quality Properties Continue

Run	y_{wsi}	y_{bd}	y_{zinc}	y_{mag}	y_{cal}	y_{fe}	y_{phos}
1	40	0.43	5.22	128.4	336.4	9.18	687.21
2	45	0.71	5.16	135.62	328.22	7.56	685.36
3	45	0.31	5.18	133.4	288.84	5.89	685.9
4	42	0.63	5.22	128.82	312.65	5.98	588.74
5	48	0.45	4.74	132.36	320.72	5.77	626.36
6	40	0.43	5.31	164.54	334.38	5.87	678.4
7	31	0.73	4.98	144.28	404.2	7.22	725.38
8	31	0.73	4.44	148.36	388.34	7.43	722.56
9	46	0.38	4.43	156.64	364.38	6.78	710.45
10	38	0.71	4.56	138.92	366.8	6.63	688.36
11	41	0.73	5.28	124.52	416.34	6.56	665.54
12	42	0.44	5.22	128.37	424.88	6.83	688.92
13	42	0.42	4.78	128.54	377.9	6.73	674.88
14	39	0.36	4.46	128.36	386.9	9.22	665.3
15	39	0.52	4.38	128.22	440.2	7.89	723.86
16	43	0.67	5.33	138.3	380.56	7.66	765.4
17	38	0.75	5.26	144.56	388.74	7.88	677.37
18	43	0.31	5.74	140.2	408.42	7.94	688.95
19	43	0.43	5.36	136.76	422.1	7.22	722.1
20	46	0.35	5.74	141.58	426.9	7.52	736.29
21	48	0.5	4.88	144.1	392.62	6.88	686.35
22	35	0.38	5.52	136.24	444.3	6.76	688.9
23	51	0.52	4.96	138.39	386.45	7.13	675.4
24	45	0.44	4.74	124.52	405.18	7.98	668.62
25	42	0.39	4.66	150.26	413.36	7.55	678.76
26	40	0.5	4.98	129.84	444.52	11.36	696.32
27	44	0.72	4.33	132.65	468.4	9.18	745.28
28	48	0.37	4.54	132.8	426.48	9.34	688.68
29	54	0.44	4.36	144.65	426.12	7.96	686.9
30	48	0.45	4.72	138.72	418.3	7.88	765.45
31	52	0.34	4.76	140.34	420.76	7.72	732.86
32	42	0.49	5.23	140.65	390.7	7.87	662.65
33	43	0.31	5.76	138.22	445.36	9.16	672.64
34	50	0.48	5.22	126.28	354.6	7.82	698.38
35	49	0.43	5.34	146.34	428.25	6.98	722.32
36	49	0.54	5.26	162.2	336.7	7.36	765.12
37	39	0.44	4.84	154.62	356.65	6.85	748.2
38	40	0.39	4.66	138.85	328.5	7.66	686.9
39	37	0.45	5.74	136.22	334.9	9.16	655.76
40	32	0.44	6.6	145.35	362.76	11.22	654.9
41	34	0.5	5.22	145.9	344.98	7.89	655.46
42	56	0.47	6.12	145.26	404.45	8.44	688.72
43	36	0.51	6.66	162	288.96	7.16	655.48
44	40	0.51	5.18	144.2	328.54	7.89	686.55

Table 4. (Continue) Formulated Feeds Quality Properties Continue

Run	y_{wsi}	y_{bd}	y_{zinc}	y_{mag}	y_{cal}	y_{fe}	y_{phos}
45	43	0.46	6.72	136.38	290.42	10.45	675.7
46	65	0.5	5.13	144.8	366.65	7.98	663.28
47	59	0.49	4.98	145.16	348.67	7.55	654.65
48	43	0.54	5.94	152.35	388.7	10.42	668.23
49	56	0.53	5.46	128.94	345.5	7.6	708.3
50	67	0.43	7.12	135.2	296.75	11.25	745.23
51	45	0.69	6.83	126.54	288.4	9.4	726.34
52	58	0.63	4.78	128.88	310.98	7.88	688.76
53	56	0.5	5.22	144.32	322.74	9.65	712.38
54	81	0.52	4.89	132.78	340.26	9.72	735.56
55	41	0.56	5.22	134.5	316.9	7.98	698.7
56	55	0.71	5.18	136.34	320.18	7.44	660.54
57	58	0.58	5.12	128.7	338.54	7.69	692.38
58	40	0.72	7.1	165.33	336.2	11.28	644.76
59	61	0.52	7.62	158.54	298.86	11.33	668.87
60	55	0.53	6.18	166.35	264.75	9.63	676.42
61	61	0.64	5.15	144.92	280.5	7.88	680.65
62	57	0.67	5.22	142.28	322.76	7.48	656.38
63	46	0.45	5.74	150.36	374.8	7.52	705.78
64	46	0.54	4.96	154.22	410.3	7.16	722.45
65	57	0.53	5.18	148.42	388.56	7.9	738.2
66	53	0.47	5.14	166.32	440.92	7.75	718.32
67	62	0.44	5.88	145.6	388.35	8.12	696.74
68	66	0.5	5.15	136.28	365.72	7.66	688.32
69	60.5	0.45	5.72	155.3	406.3	7.88	666.48
70	61	0.49	5.34	148.26	390.68	7.56	689.68
71	51	0.47	5.7	145.78	356.45	7.93	688.9
72	63	0.53	5.45	148.24	372.3	7.71	656.75
73	52	0.47	5.86	136.68	365.58	7.86	693.42
74	58.5	0.34	5.54	150.45	326.44	7.32	674.34
75	61	0.61	7.3	156.42	346.23	7.22	640.34
76	54	0.56	6.3	133.32	434.56	9.34	733.43

$y_{wsi} = Water\ \text{Solubility}\ index(\%)$, $y_{bd} = Bulk\ Density\left(g/cm^3\right)$, $y_{zinc} = Zinc(mg)$,

$y_{mag} = Magnesium(mg)$, $y_{cal} = Calcium(mg)$, $y_{fe} = Iron(mg)$, $y_{phos} = Phosphorus(mg)$.

3. Results of Statistical Analyses of Experimental Data and Discussion

The experimental data were subjected to statistical analyses and the statistical significance of the regression terms were tested using analysis of variance (ANOVA) for each response. Contour as well as the 3-D response surface plots were generated for the quality properties using the Desıgn Expert 13.0.0 statistical software package. Inferential statistics were derived based on the coefficient of determination, F-value, and p-values at a 5% level of significance." The summary statistics of the feeds' quality properties that were significantly influenced by the formulation ingredients, at p-value < 0.05, are presented in Table 5.

Table 5. Summary statistics of the feeds' quality properties significantly influenced at p-value < 0.05)

Response	Sources	**F-value**	**p-value**	**R^2**	**Adj R^2**	**Pre R^2**	**C.V. (%)**	**Adeq Precision**
y_{lip}	Model	3.43	0.0012	0.3454	0.2446	0.0974	23.03	8.1724
	L/Mixture	3.43	0.0012					
y_{cf}	Model	3.99	0.0108	0.9629	0.7216	-5.6444	16.10	10.0859
	L/Mixture	3.68	0.0259					
	x_1x_5	24.96	0.0005					
	x_1x_8	6.68	0.0272					
	x_1x_9	7.06	0.0240					
	x_2x_5	27.34	0.0004					
	x_2x_8	6.30	0.0309					
	x_2x_9	6.49	0.0290					
	x_3x_5	7.23	0.0228					
	x_3x_6	8.35	0.0161					
	x_3x_8	8.84	0.0140					
	x_3x_9	7.09	0.0238					
	x_4x_5	25.83	0.0005					
	x_4x_6	7.05	0.0241					
	x_4x_8	7.07	0.0240					
	x_4x_9	6.91	0.0252					
	x_5x_7	27.36	0.0004					
	x_5x_9	8.78	0.0142					
	x_6x_7	6.32	0.0307					
	x_6x_8	10.74	0.0083					
	x_6x_{10}	6.68	0.0272					
	x_7x_8	8.40	0.0159					
	x_7x_9	6.78	0.0263					
	x_8x_{10}	5.46	0.0415					
	x_9x_{11}	8.69	0.0146					
y_{ev}	Model	2.53	0.0123	0.2798	0.1690	0.0095	5.84	7.1855
	L/Mixture	2.53	0.0123					
y_{zinc}	Model	1.51	0.2458	0.9075	0.3060	13.4826	11.10	5.5016
	L/Mixture	1.46	0.2787					
	x_1x_3	5.34	0.0434					
	x_1x_4	6.40	0.0299					

Table 5. (Continue) Summary statistics of the feeds' quality properties significantly influenced at p-value < 0.05)

Response	Sources	**F-value**	**p-value**	**R^2**	**Adj R^2**	**Pre R^2**	**C.V. (%)**	**Adeq Precision**
	x_3x_4	5.48	0.0413					
	x_3x_5	10.22	0.0095					
y_{cal}	Model	1.13	0.4509	0.8798	0.0987	-4.8331	12.35	5.0279
	L/Mixture	0.8631	0.5897					
	x_4x_{10}	5.42	0.0422					

Note: The full summary statistics of all the feeds' quality properties and respective plots are provided as supplementary files.

$y_{lip} = Lipid\ Content(\%)$, $y_{cf} = Crude\ Fiber(\%)$, $y_{ev} = Energy\ Value(kcal)$,

$y_{zinc} = Zinc(mg)$, $y_{cal} = Calcium(mg)$,

In the summary statistics Table, p-values less than 0.05 indicate terms that were significant at 5% level of significance. The quality properties of the feed evaluated were protein content, moisture content, carbohydrate, lipid content, crude fibre, ash content, nitrogen free extract, energy value, water solubility index, bulk density, zinc, magnesium, calcium, iron, and phosphorus. However, as presented in the summary statistics Table, only the lipid, crude fibre, energy value, zinc, and calcium were influenced by the formulation ingredients (p-value <0.05). The lipid model was significant (model p-value = 0.0012) and all the ingredients had significant influences on the lipid content of the formulated feed (L/Mixture p-value = 0.0012). The crude fibre model was significant (model p-value = 0.0108) and all the ingredients had significant influences on the crude fibre content of the formulated feed (L/Mixture p-value = 0.0259). In addition, nearly all the ingredients interacted to influence the crude fibre significantly (p-values <0.05). The energy value model was significant (model p-value = 0.0123) and all the ingredients had significant influences on the crude fibre content of the formulated feed (L/Mixture p-value = 0.0123). The zinc model was not significant (model p-value = 0.2458 (greater than 0.05)) and all the ingredients were also found not to have significant influences on the zinc content of the formulated feed (L/Mixture p-value = 0.2787 (greater than 0.05)). However, intestine meal/ beni seed meal (x_1x_3), intestine meal/ wheat bran (x_1x_4), beni seed meal / wheat bran (x_3x_4), and beni seed meal / groundnut meal (x_3x_5); interacted to significantly influence the zinc content of the formulated feed. The calcium model was not significant (model p-value = 0.4509 (greater than 0.05)) and all the ingredients were also found not to have significant influences on the calcium content of the formulated feed (L/Mixture p-value = 0.5897 (greater than 0.05)). However, wheat bran interacted with lime stone (x_4x_{10}) to significantly influence the calcium content of the formulated feed (x_4x_{10} p-value = 0.0422). The quality properties contour plots for the formulated feed are presented in Figures 1.

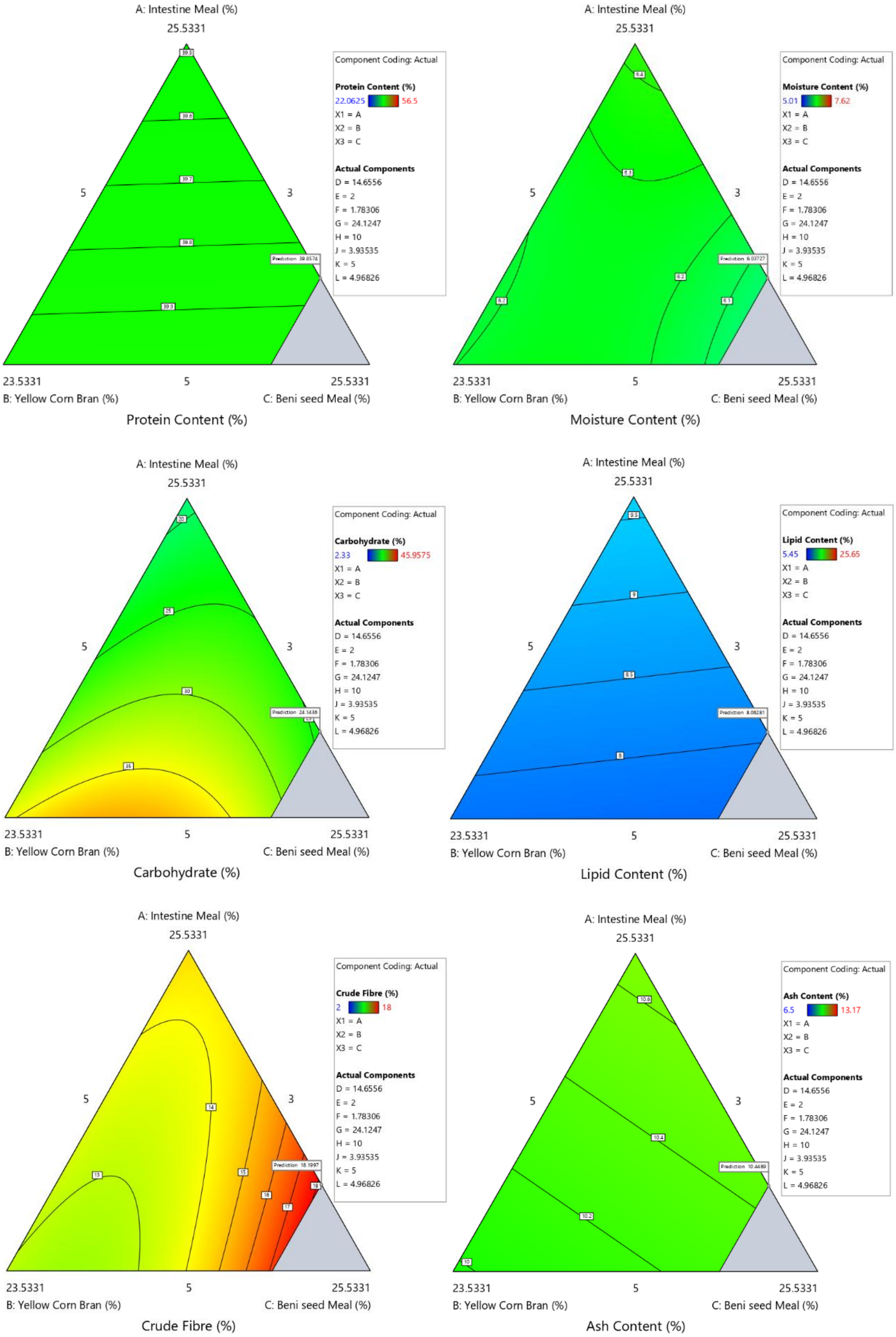

Figure 1. (Continue) Contour plots for the quality properties of the formulated feeds

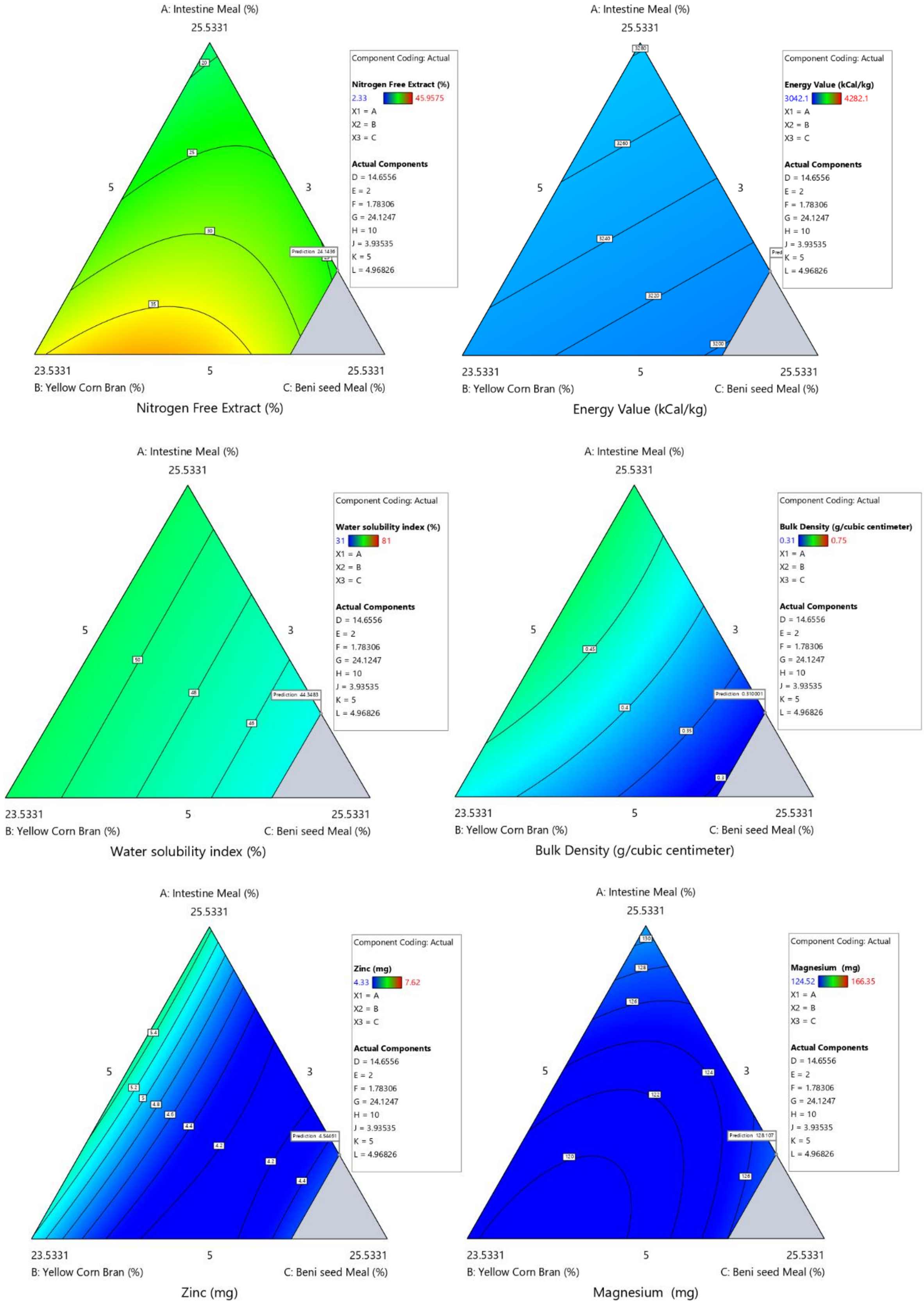

Figure 1. (Continue) Contour plots for the quality properties of the formulated feeds

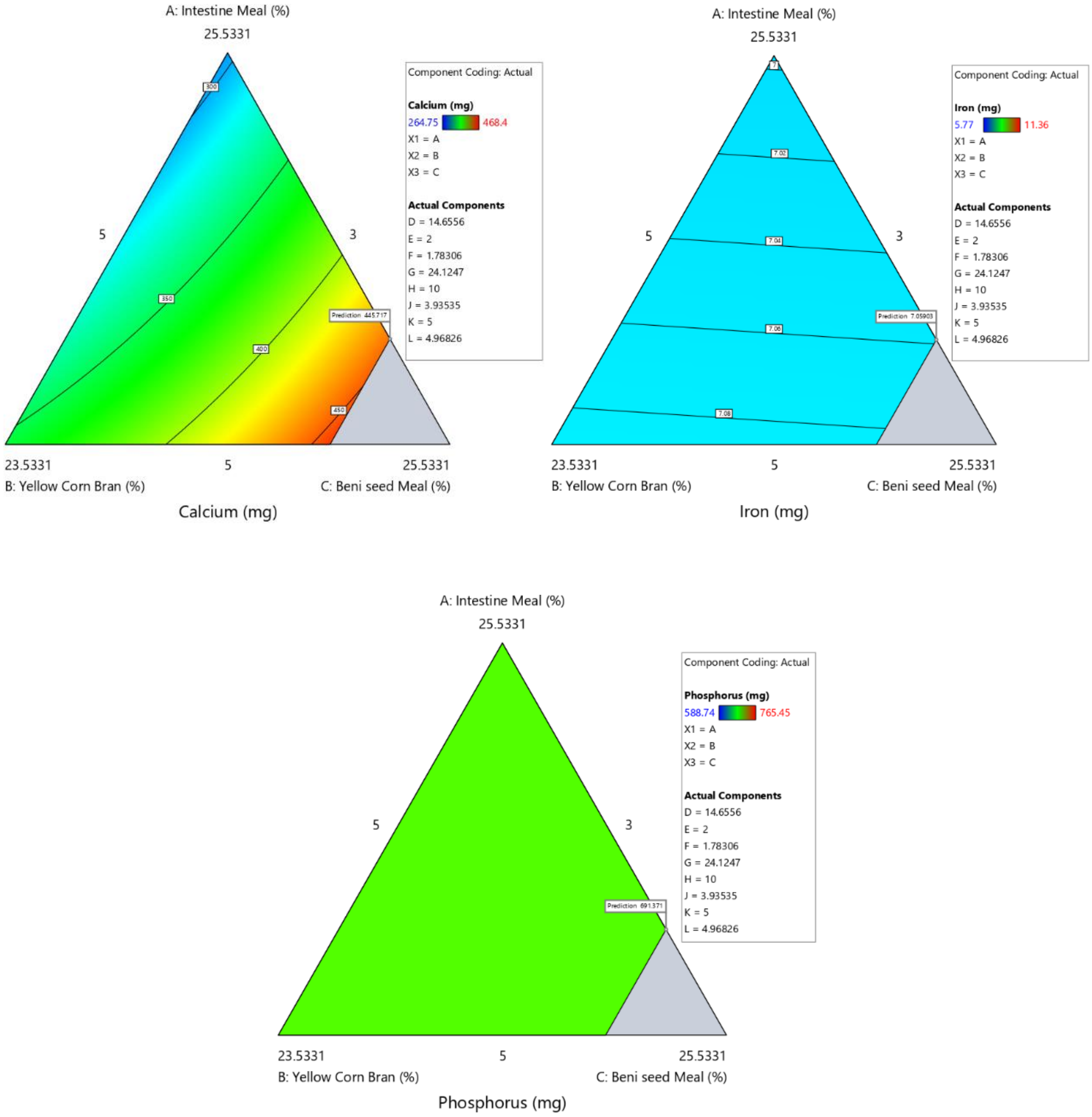

Figure 1. (Continue) Contour plots for the quality properties of the formulated feeds

4. Conclusion

Fish feed was developed from intestine meal, yellow corn bran, beni seed meal, wheat bran, groundnut meal, palm kernel cake, rice bran, blood meal, moringa leaf, limestone, and bone meal. The lipid, crude fibre, and energy value of the formulated feed were influenced by the formulation ingredients (L/Mixture p-value <0.05). Interaction effects of the ingredients on the quality properties of the feed were established. The ingredients interacted, at two levels, to significantly influence the crude fibre, zinc, and calcium contents of the formulated feed. Though zinc and calcium models were non-significant (model p-value > 0.05), analysis revealed that some of the ingredients combined together to statistically influence zinc and calcium (terms p-value < 0.05). Intestine meal/beni seed meal, intestine meal/wheat bran, beni seed meal/wheat bran, and beni seed meal/groundnut meal; interacted to significantly influence the zinc content of the formulated feed. Wheat bran interacted with lime stone to significantly influence the calcium content of the formulated feed (p-value = 0.0422). The overall mean value of phosphorus proved to be the true representation of phosphorus in the formulated feed.

5. Author Contribution Statement

Samuel Tunde Olorunsogo*:* Conceptualization, methodology, Investigation, performance of laboratory analysis, data collection, analysis and interpretation, visualization, software, validation, drafting the article, review and editing, critical revision, final approval of the version to be published

Akande Eunice Kemi: Review and editing, critical revision,

6. Ethics Committee Approval and Conflict of Interest

There is no need for an ethics committee approval and there is no conflict of interest with any person/institution in the prepared article. The authors alone are responsible for the content and writing of the manuscript.

7. Ethical Statement Regarding the Use of Artificial Intelligence

Declaration of Generative AI and AI-assisted technologies in the writing process. During the preparation of this study, the authors used AI-assisted technologies only for English language checking and improving academic style. All figures, data analysis graphs, experimental results, and scientific inferences in the article were entirely created and evaluated manually by the authors without the use of artificial intelligence. As authors, we accept full responsibility for the originality of the article and the scientific findings it contains. The entire content of the study was produced by the authors in accordance with scientific research methods and academic ethical principles.

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