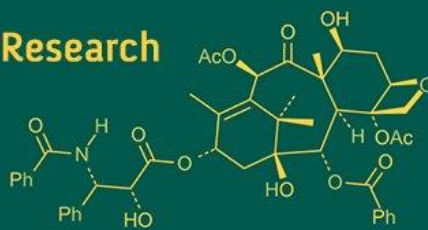


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Optimization of nutrient-enriched edible bowl formulation using response surface methodology

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Abstract

Single-use plastic tableware contributes substantially to non-biodegradable waste and microplastic pollution, highlighting the need for sustainable edible alternatives that can also help address micronutrient deficiencies. In this study, a nutrient-enriched edible bowl was developed using varying proportions of wheat, sorghum, ragi (finger millet) and water chestnut flours, supplemented with skimmed milk powder, sugar, salt, guar gum, butter, and beetroot juice. A four-factor, five-level Central Composite Rotatable Design (CCRD) comprising 30 experimental runs was employed using Response Surface Methodology (RSM) to evaluate the effects of wheat flour (2.5-12.5 g/100 g), sorghum flour (5-15 g/100 g), ragi flour (5-15 g/100 g) and water chestnut flour (5-15 g/100 g) on the physicochemical and sensory characteristics of the bowl. Numerical optimization targeted minimum moisture content and water absorption capacity (WAC), along with maximum true protein, iron (Fe), calcium (Ca) and overall acceptability. The optimized formulation contained 5.82 g/100 g wheat flour, 11.52 g/100 g sorghum flour, 7.50 g/100 g ragi flour and 12.50 g/100 g water chestnut flour, achieving a desirability of 0.76. The predicted values were 9.25% moisture, 20.03% WAC, 6.36% true protein, 32.33 mg/kg Fe, 1162.36 mg/kg Ca and 7.57 overall acceptability. The optimized bowl completely biodegraded in soil within 6-7 days, demonstrating its potential as a nutritious, functional and environmentally sustainable alternative to conventional plastic tableware.

Keywords: Edible bowl, response surface methodology, millets, water chestnut flour, biodegradable packaging

1. Introduction

Malnutrition and micronutrient deficiencies, particularly anaemia arising from inadequate intake of iron, calcium and other essential nutrients, remain widespread public health concerns, underscoring the need for accessible, nutrient-dense food products ^[1]. In parallel, conventional single-use plastic tableware has emerged as a critical environmental hazard: plastics are non-biodegradable, persist in the environment for thousands of years, contaminate soil when discarded and release toxic pollutants when incinerated, while microplastics generated from marine plastic debris enter the human food chain through seafood consumption ^[2, 3]. Projections indicate that, if current production and disposal trends continue, approximately 12,000 metric tons of plastic waste will accumulate in landfills and the environment by 2050, with associated risks of carcinogenic and toxic substances entering the food supply ^[4].

In response to these concerns, regulatory measures such as European Union Directive 2019/904, which bans a wide range of single-use plastic items, and national initiatives such as India's 'Plastic Challenge-Hackathon 2021' have sought to promote sustainable alternatives to disposable plastic products ^[5, 6]. Because conventional plastic cutlery and tableware are petroleum-derived and may expose consumers to carcinogenic and toxic residues, there has been growing interest in edible, biodegradable tableware developed from natural, nutrient-dense raw materials as a sustainable substitute ^[7, 8]. Edible tableware including plates, bowls, spoons and dessert cups offers the dual advantage of being either consumed with the meal or safely biodegraded in the environment without specialized composting conditions ^[9, 10].

Cereal and millet flours provide an attractive raw-material base for such products. Wheat flour contributes gluten-forming proteins (gliadin and glutenin) that impart viscoelastic dough properties essential for structural integrity ^[11].

Millet, though nutritionally comparable to major cereals, remain underexploited despite being rich sources of protein, energy, B-complex vitamins and dietary fibre [12]. Ragi (finger millet) is particularly notable for its exceptionally high calcium and iron content, along with iodine, protein, phosphorus, fibre and vitamins, while sorghum contributes B-vitamins, resistant starch, magnesium and antioxidant phenolic compounds with demonstrated anti-inflammatory and anti-diabetic properties [1, 13-15]. Water chestnut flour is a gluten-free source of omega-3 fatty acids, minerals, B-complex and other vitamins and bioactive phenolics, with additional therapeutic relevance in traditional medicine [16, 17]. Beetroot, used here as a natural colourant and functional ingredient, is rich in betalains, antioxidants and micronutrients with recognized anti-inflammatory, antihypertensive and anti-anaemic properties [18-20]. The present study aimed to develop a nutrient enriched, biodegradable edible bowl formulated from wheat, sorghum, ragi and water chestnut flours, targeting simultaneous improvement of nutritional quality (protein, iron, calcium) and functional performance (moisture, water absorption capacity) together with high consumer acceptability, using Response Surface Methodology to identify and validate the optimum formulation. Specifically, the objectives were (i) to standardize process parameters for preparation of the nutrient enriched edible bowl and (ii) to evaluate the physicochemical and sensory properties of the developed bowl.

2. Materials and Methods

2.1 Preparation of the Edible Bowl

Rice flour served as the base ingredient, to which varying levels of wheat flour, sorghum flour, ragi flour and water chestnut flour were added along with SMP (10 g), powdered sugar (10 g), salt (1 g), guar gum (2 g), butter (1 g) and beetroot juice (55-60 mL) in fixed proportion. Dry ingredients were weighed and blended to a uniform mixture; beetroot juice was then incorporated and the mass kneaded into a homogeneous dough, which was rested for 30 min under a wet cloth. The dough was sheeted, moulded into

bowl shape and baked in a microwave oven (conventional mode) at 180 °C for 20-25 min. Baked bowls were cooled under ambient condition.

A four-factor, five-level Central Composite Rotatable Design (CCRD) was employed, comprising 16 factorial points, 8 axial points ($\alpha = 2$) and 6 centre-point replications, giving a total of 30 treatment combinations. The independent variables, expressed relative to 100 g rice flour base, and their coded/uncoded levels are presented in Table 1. Dependent (response) variables comprised physicochemical parameters such as moisture content, true protein, water absorption capacity, and mineral content (Ca and Fe) and sensory parameters (colour, taste, aroma and overall acceptability). Experimental design, regression modelling and ANOVA were performed using Design-Expert software (Version 13, Stat-Ease Inc.).

2.4 Analytical Methods

Moisture content (% w.b.) was determined by oven-drying at 105 °C to constant weight [21]. True protein content was determined by the Folin-Lowry method (1951), with absorbance measured at 660 nm against a bovine serum albumin (BSA) standard curve. Water absorption capacity (WAC) was measured by immersing 2 g of sample in water for 30 min and recording the change in weight. Mineral content (Ca and Fe) was determined following diacid (HNO₃:HClO₄, 2:1) digestion and analysis by Atomic Absorption Spectrophotometry. Sensory evaluation was carried out by a 10-member semi-trained panel using the 9-point hedonic scale, with an overall score ≥ 5 considered acceptable [22].

2.5 Optimization and Biodegradability

Numerical optimization of the formulation was performed using Design-Expert software with the goals of minimizing moisture content and WAC and maximizing true protein, Fe, Ca and overall acceptability. Biodegradability of the optimized bowl was assessed by the soil burial method, with samples buried 3 cm below the soil surface and degradation monitored daily.

Table 1: Treatment combinations for development of nutrient enriched edible bowl with coded and uncoded values

Treatment No.	Factor: Wheat flour (g/100 g)		Factor: Sorghum flour (g/100 g)		Factor: Ragi flour (g/100 g)		Factor: Water chestnut flour (g/100 g)	
	Coded	Uncoded	Coded	Uncoded	Coded	Uncoded	Coded	Uncoded
1	-1	5	-1	7.5	-1	7.5	-1	7.5
2	1	10	-1	7.5	-1	7.5	-1	7.5
3	-1	5	1	12.5	-1	7.5	-1	7.5
4	1	10	1	12.5	-1	7.5	-1	7.5
5	-1	5	-1	7.5	1	12.5	-1	7.5
6	1	10	-1	7.5	1	12.5	-1	7.5
7	-1	5	1	12.5	1	12.5	-1	7.5
8	1	10	1	12.5	1	12.5	-1	7.5
9	-1	5	-1	7.5	-1	7.5	1	12.5
10	1	10	-1	7.5	-1	7.5	1	12.5
11	-1	5	1	12.5	-1	7.5	1	12.5
12	1	10	1	12.5	-1	7.5	1	12.5
13	-1	5	-1	7.5	1	12.5	1	12.5
14	1	10	-1	7.5	1	12.5	1	12.5
15	-1	5	1	12.5	1	12.5	1	12.5
16	1	10	1	12.5	1	12.5	1	12.5
17	-2	2.5	0	10	0	10	0	10
18	2	12.5	0	10	0	10	0	10
19	0	7.5	-2	5	0	10	0	10
20	0	7.5	2	15	0	10	0	10

21	0	7.5	0	10	-2	5	0	10
22	0	7.5	0	10	2	15	0	10
23	0	7.5	0	10	0	10	-2	5
24	0	7.5	0	10	0	10	2	15
25	0	7.5	0	10	0	10	0	10
26	0	7.5	0	10	0	10	0	10
27	0	7.5	0	10	0	10	0	10
28	0	7.5	0	10	0	10	0	10
29	0	7.5	0	10	0	10	0	10
30	0	7.5	0	10	0	10	0	10

3. Results

3.1 Model Fitting and Range of Responses

Across the 30 CCRD runs, moisture content ranged from 4.31 to 11.29%, WAC ranged from 18.55 to 40.89%, true protein ranged from 5.44 to 7.73%, Fe ranged from 16.58 to

74.35 mg/kg, Ca ranged from 936 to 1664 mg/kg and overall acceptability ranged from 6.67 to 7.89. Second-order polynomial models fitted to each response were statistically significant and the model summary statistics for the responses are presented in Table 2.

Table 2: ANOVA summary for the RSM-optimized quality responses of the edible bowl

Response	Model F-value	p-value	Significant terms	R ²	Adj. R ²	Adeq. Precision
Moisture content (%)	2.56	0.0407*	AD	0.70	0.43	6.54
Water absorption capacity (%)	3.01	0.0210*	D, B ²	0.74	0.49	7.25
True protein (%)	2.63	0.0369*	B, D ²	0.71	0.44	6.23
Iron, Fe (mg/kg)	3.13	0.0179*	B, D, BC, CD, C ²	0.74	0.51	7.97
Calcium, Ca (mg/kg)	8.71	<0.0001***	C, A ² , B ² , C ² , D ²	0.89	0.79	9.07
Overall acceptability	3.69	0.0085**	AD, CD, A ² , D ²	0.78	0.57	8.90

Level of significance: * = $p < 0.05$, ** = $p < 0.01$, *** = $p < 0.001$

3.2 Effect of Formulation Variables on Responses

3.2.1 Moisture content

The interaction between wheat flour and water chestnut flour (AD) was the only significant term affecting moisture content, while the linear and quadratic effects of the individual flours were non-significant, shown in Table 2. Moisture content increased with the simultaneous increase in wheat flour and water chestnut flour levels, indicating a positive interactive effect on moisture retention (Fig. 1). This behavior may be attributed to the high water-binding capacity of wheat proteins and the starch-rich composition of water chestnut flour, which enhanced water retention within the product matrix, consistent with Iqbal *et al.* [13]. In contrast, moisture content showed a slight decline with the simultaneous increase in sorghum flour and ragi flour levels, suggesting a comparatively lower water-holding capacity of these millet flours. Similar trends have been reported for ragi incorporated sample prepared by Solanki [23].

3.2.2 Water absorption capacity (WAC)

Water absorption capacity (WAC) was significantly influenced by the linear effect of water chestnut flour (D) and the quadratic effect of sorghum flour (B²), shown in Table 2. WAC exhibited a quadratic response with increasing sorghum flour and ragi flour levels, decreasing to a minimum at intermediate concentrations before increasing again at higher levels, indicating a significant interactive effect between the two millet flours. This curvilinear behaviour may be attributed to variations in the fibre and protein composition of the flours, which influence their hydration characteristics. In contrast, WAC decreased progressively with increasing water chestnut flour,

particularly in combination with moderate to higher levels of wheat flour, suggesting that water chestnut flour had a more pronounced effect than its interaction with wheat flour. The reduction in WAC at higher water chestnut flour levels may be associated with changes in the starch-protein matrix that limit water binding. Similar observations were reported by Manisha and Punia [24], who observed a significant decline in WAC with increasing substitution of wheat flour by water chestnut flour in composite blends.

3.2.3 True protein content

Sorghum flour (B) exerted a significant negative linear effect on true protein content, while the quadratic effect of water chestnut flour (D²) was also significant (Table 2). The Fig. 3 showed true protein content was higher at increased wheat flour and lower sorghum flour levels, indicating the positive contribution of wheat flour and the protein dilution effect associated with increasing sorghum flour, supported by Elizabeth *et al.* [25]. The quadratic response observed for sorghum flour suggests that its influence on protein content varied across the experimental range. Conversely, true protein content decreased with increasing water chestnut flour, particularly at higher levels of ragi flour, indicating that water chestnut flour had a more pronounced effect than its interaction with ragi flour. Similar results were reported by Din *et al.* [26] and Lutfi and Kalim [27] who observed a significant reduction in protein content with increasing replacement of wheat flour by water chestnut flour in composite cookies. This decline was attributed to the dilution of wheat gluten proteins by the inherently low-protein water chestnut flour.

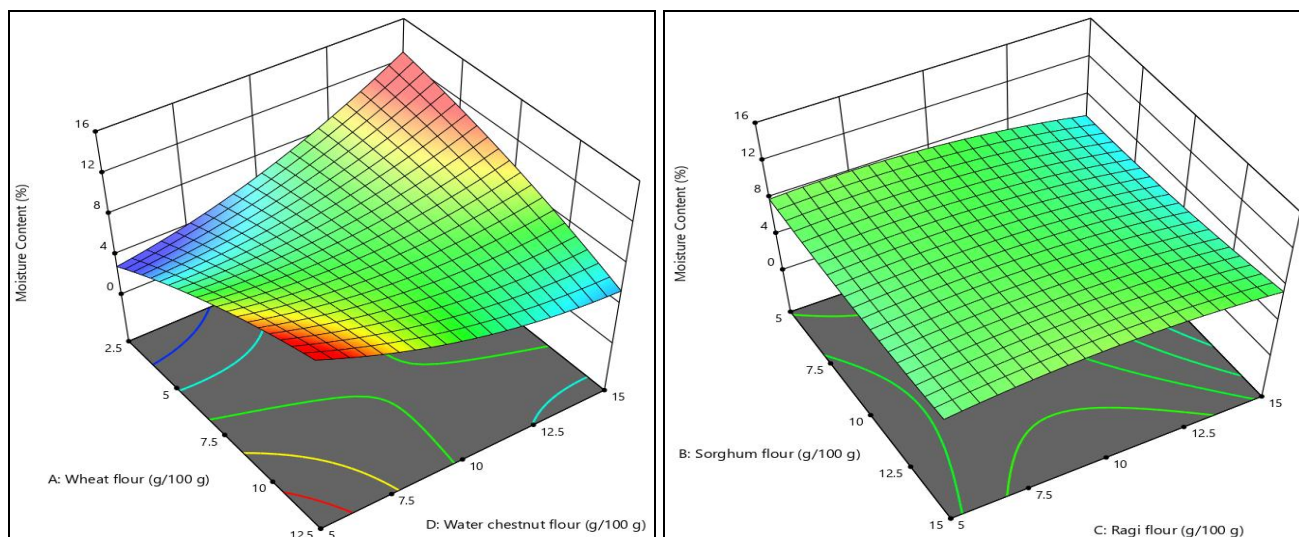


Fig 1: Effect of wheat flour, sorghum flour, ragi flour and water chestnut flour on moisture content of developed bowl

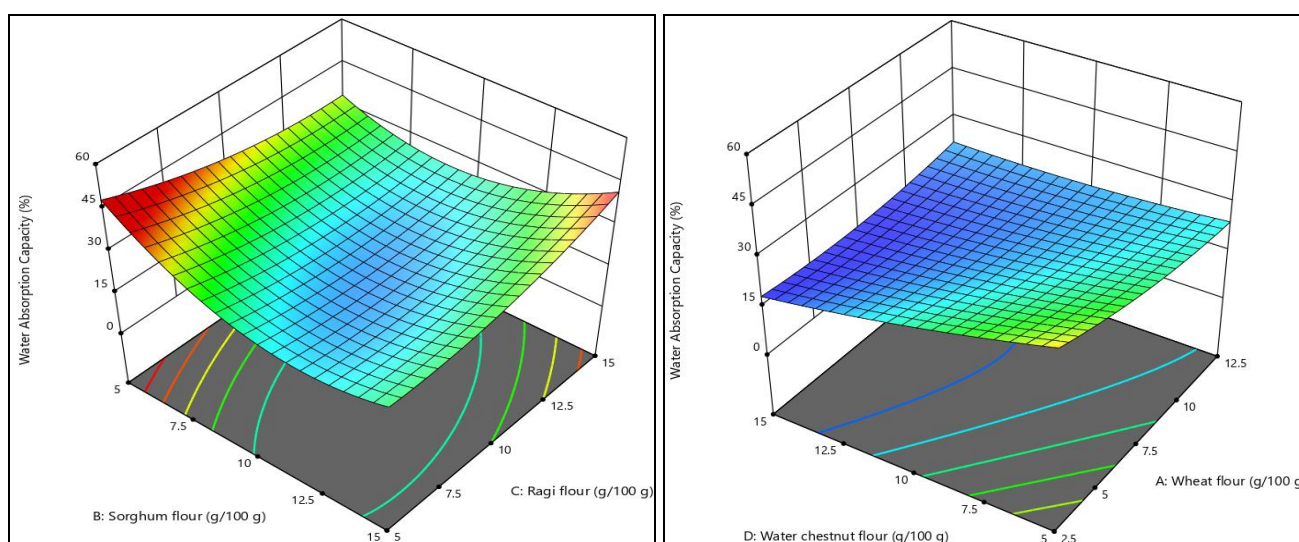


Fig 2: Effect of wheat flour, sorghum flour, ragi flour and water chestnut flour on water absorption capacity of developed bowl

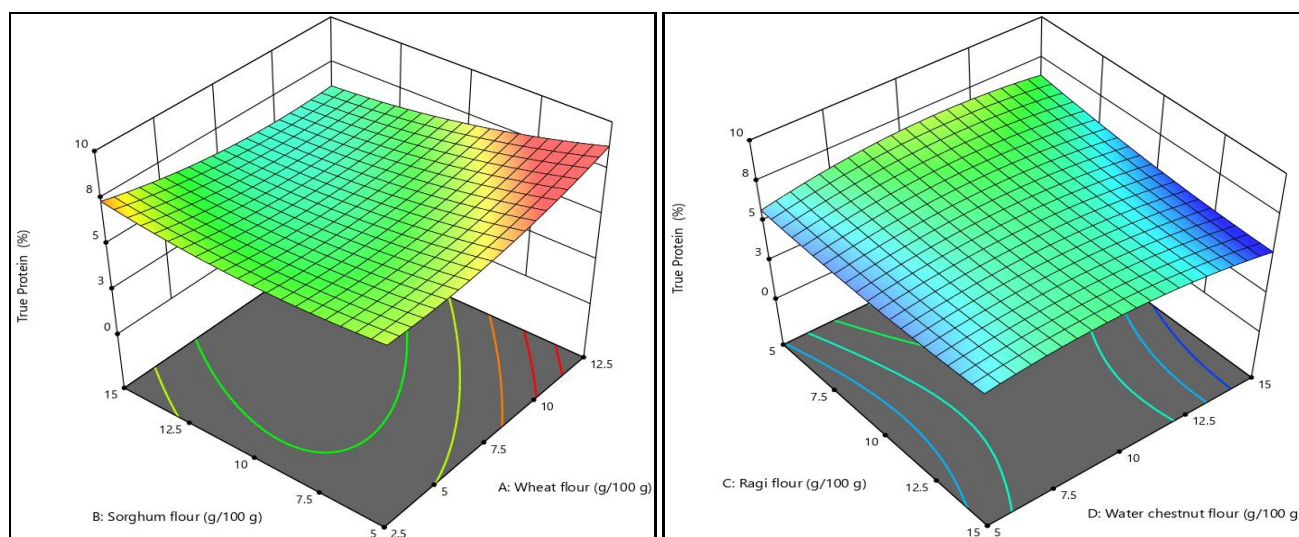


Fig 3: Effect of wheat flour, sorghum flour, ragi flour and water chestnut flour on true protein content of developed bowl

3.2.4 Iron (Fe) content

Sorghum flour (B), water chestnut flour (D), the BC and CD interactions and the quadratic effect of ragi flour (C^2) significantly influenced Fe content (Table 2). Iron content decreased progressively with increasing sorghum flour and

ragi flour levels, with the highest values obtained at lower concentrations of both flours, indicating a significant interactive effect between these ingredients, shown in Fig. 4. Ningtyias *et al.* [28] observed similar results for incorporation of sorghum flour. The reduction in Fe at higher ragi and

sorghum flour levels may be attributed to compositional interactions that affected mineral retention within the formulation. In contrast, Fe content increased with the simultaneous increase in wheat flour and water chestnut flour, reflecting the positive contribution of these

ingredients, particularly the naturally higher iron content of water chestnut flour. Similar observations on the iron-rich nature of water chestnut flour have been reported by Jamale *et al.* [29]

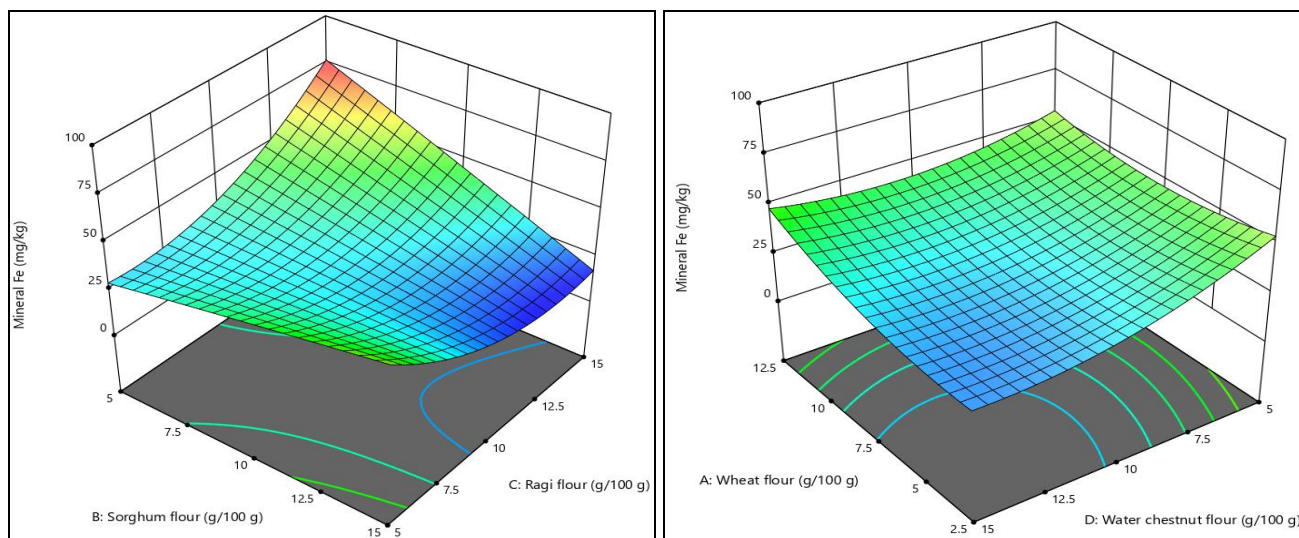


Fig 4: Effect of wheat flour, sorghum flour, ragi flour and water chestnut flour on iron content of developed bowl

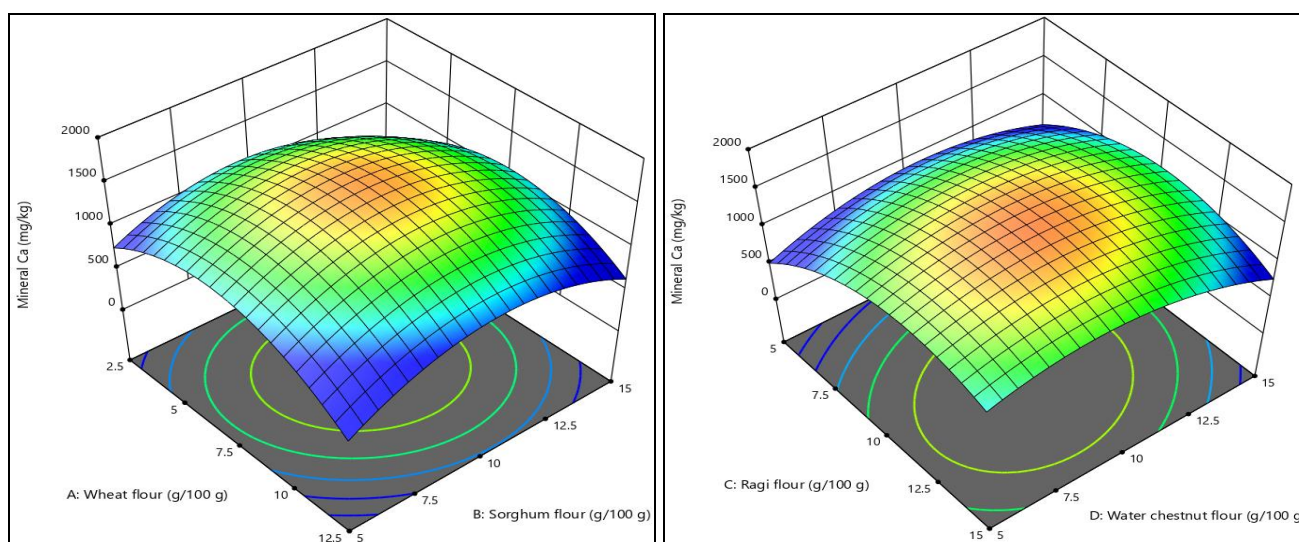


Fig 5: Effect of wheat flour, sorghum flour, ragi flour and water chestnut flour on calcium content of developed bowl

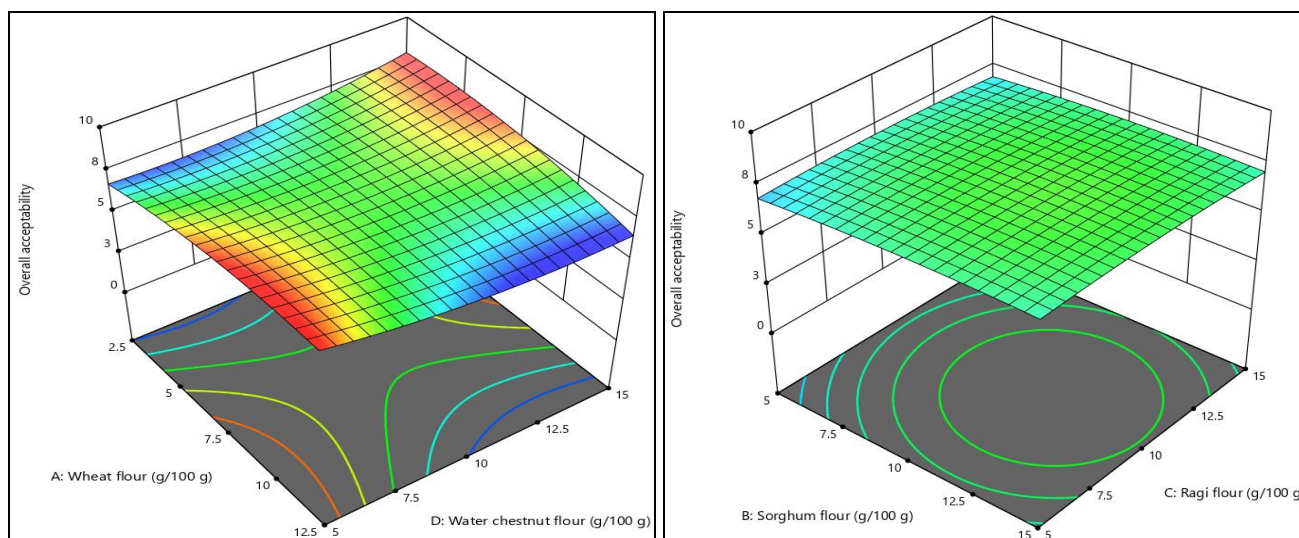


Fig 6: Effect of wheat flour, sorghum flour, ragi flour and water chestnut flour on overall acceptability of developed bowl

3.2.5 Calcium (Ca) content

The calcium model was highly significant, with the linear effect of ragi flour (C) and the quadratic effects of wheat flour (A^2), sorghum flour (B^2), ragi flour (C^2) and water chestnut flour (D^2) significantly influencing Ca content (Table 2). Calcium content increased with increasing wheat flour and sorghum flour up to intermediate levels before declining at higher concentrations, indicating a pronounced quadratic response (Fig. 5). A similar synergistic quadratic relationship was observed between ragi flour and water chestnut flour, with maximum calcium content achieved at intermediate levels of both ingredients, followed by a decline at higher concentrations. This behaviour reflects the mineral-rich nature of ragi and water chestnut flour, with water chestnut flour contributing more to calcium enhancement, consistent with Rana *et al.* [30] for ragi supplementation in sample.

3.2.6 Overall acceptability

The interactions of wheat flour with water chestnut flour (AD) and sorghum flour with ragi flour (CD), together with the quadratic effects of wheat flour (A^2) and water chestnut flour (D^2), significantly influenced overall acceptability (Table 2). Overall acceptability exhibited a saddle-shaped response between wheat flour and water chestnut flour, with higher sensory scores obtained at opposite extreme combinations of the two ingredients, indicating a significant interaction rather than a simple linear trend (Fig. 6). In contrast, overall acceptability increased with increasing

sorghum flour and ragi flour up to intermediate concentrations before declining slightly at higher levels, demonstrating a quadratic relationship. The improved acceptability at moderate levels of sorghum and ragi flour may be attributed to their complementary effects on texture, colour, flavour and overall sensory quality, whereas excessive or insufficient incorporation of either flour slightly reduced consumer acceptability.

3.3 Numerical Optimization and Validation

Numerical optimization using Design-Expert software, applied across all measured responses with moisture content and WAC set to minimize and true protein, Fe, Ca and overall acceptability set to maximize, identified an optimum formulation of 5.82 g/100 g wheat flour, 11.52 g/100 g sorghum flour, 7.50 g/100 g ragi flour and 12.50 g/100 g water chestnut flour, with an overall desirability of 0.76 (Table 3).

The optimized formulation was subsequently prepared under the identified conditions and characterized experimentally to validate the regression models. Experimental values for all responses, including the moisture content (8.86%), WAC (20.63%), true protein (6.25%), Fe (31.29 mg/kg), Ca (1134.78 mg/kg) and overall acceptability (7.83), were in close agreement with predicted values, with percentage deviations ranging from 1.23 to 3.05% across all evaluated parameters, confirming the adequacy of the RSM models for predicting product quality.

Table 3: Optimized values of responses

Parameter	Optimum/Predicted value
Moisture content (%)	9.25
Water absorption capacity (%)	20.03
True protein (%)	6.36
Fe (mg/kg)	32.33
Ca (mg/kg)	1162.36
Overall acceptability	7.57
Overall desirability	0.76

3.4 Biodegradability of the Optimized Bowl

The soil burial test revealed rapid biodegradation of the optimized edible bowl. Partial disintegration was evident by day 3, and complete degradation of the buried fragments occurred within 6-7 days, facilitated by the natural, hygroscopic composition of the flour blend, increased surface-area exposure of the fragmented samples and soil moisture-driven microbial activity. This confirms the suitability of the developed product as an environmentally sustainable alternative to conventional disposable food containers.

Conclusion

A nutrient enriched, biodegradable edible bowl was successfully developed with wheat, sorghum, ragi and water chestnut flours, using a four-factor CCRD under Response Surface Methodology. Second-order regression models for moisture content, water absorption capacity, true protein, iron, calcium and overall acceptability were all statistically significant, with the calcium model showing the strongest fit. Numerical optimization identified 5.82 g/100 g wheat flour, 11.52 g/100 g sorghum flour, 7.50 g/100 g ragi flour and 12.50 g/100 g water chestnut flour as the optimum

formulation, yielding an overall desirability of 0.76 with predicted moisture content of 9.25%, WAC of 20.03%, true protein of 6.36%, Fe of 32.33 mg/kg, Ca of 1162.36 mg/kg and overall acceptability of 7.57. The optimized bowl exhibited adequate mechanical strength for functional use as tableware, complete soil biodegradation within 6-7 days. Collectively, these findings establish the developed edible bowl as a nutritionally beneficial and environmentally sustainable alternative to conventional single-use plastic tableware, with potential to simultaneously address plastic pollution and dietary micronutrient inadequacy.

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