Development of Nutritious Biscuit by Substitution of Wheat Flour using Composite Flour of Pumpkin (*Cucurbita maxima*), Corn (*Zea mays*) and Soybean (*Glycine max*) and Quality Evaluation

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ABSTRACT

Nutrients are vital for the growth and development of the human body. Intake of inadequate nutrients substantially affects the growth and mental development of young children. The aim of this research was to formulate and develop a nutritious baby biscuit using composite flour of pumpkin (Cucurbita maxima), soybean (Glycine max) and corn (Zea mays) for children, under 5 years old. Composite flour was prepared by blending wheat flour with pumpkin flour, soybean flour and corn flour with the respective ratios of 100:0:0:0 (T1), 85:5:5:5 (T2), 70:10:10:10 (T3), 55:15:15:15 (T4). The suitability of the composite flour was evaluated by the functional properties, microbial qualityrelated parameters, sensory attributes and composition proximate analysis.

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Functional properties namely water absorption absorption capacity, oil capacity and foam capacity were significantly higher (p < 0.05) in composite flour than in wheat flour. The bestpreferred baby biscuit was developed using treatment 3 (T3: 70:10:10:10), representing 30% of composite flour. The colour, flavour, texture and overall acceptability of the baby biscuit were significantly higher (p <0.05). composite flour (T3) possessed 3.96% moisture (wb), 2.66% ash, 14.18% fat, 2.23% fibre, 3.43% protein, 74.75% carbohydrate, 46.00 mg/g calcium, 3.18 mg /g iron and 427.90 mg/ g potassium. Total plate count (540 CFU) and yeast and mould count (360 CFU) were below the standard microbial limits for biscuits. In conclusion, 30% substitution of wheat flour using composite flour (pumpkin, soybean and corn) was successful in the production of nutritious biscuits for young children.

Keywords: Biscuit, Composite flour, Sensory evaluation, Nutritious, Pumpkin.

INTRODUCTION

Wheat flour has a high nutritional value and processed into biscuits, breads, pasta, pizza, noodles, cakes and many foods worldwide (Cianferoni, 2016). Complementary feeding is a nutrientdense feeding practice for vulnerable malnourish groups such as young children. Under-nutrition and micronutrient deficiency are common children young in among developing countries. Malnourished children are suffering from growth and mental development (Likhar et al., 2022). Supply of energy and nutrient by diverse supplementary food significantly satisfying the daily needs. Diversification of formulation nutritious rich biscuits is one of the potential alternatives for the children in developing countries (Dewi et al., 2020).

Wheat is not grown in Sri Lanka due to inappropriate climatic conditions. As a result, wheat is imported to satisfy the local food and indusial uses. Furthermore, wheat is well recognized as a source of food that triggers for immune-mediated food allergies, both immunoglobulin E (IgE) and non-IgE mediated (Branchi et al., 2015) allergies. Ingestion (food allergy) or inhalation (respiratory allergy) are well-known reasons for IgE mediated reactions caused by wheat (Cianferoni, 2016). Therefore, some developing countries tropical in the region

encourage the use of composite flour, which replaces certain quantity of flour wheat in food product formulation (Adefegha and Oboh, 2013; Akubor and Obiegbuna, 2014). Formulation and development of baby biscuits substituting wheat flour is an alternative complementary food for young children.

The economical and nutritional benefits can be achieved by combining wheat flour with other composite flour in bakery and confectionery products. Composite flours are developed by blending several non-wheat flours in varied proportions, either with or without wheat flour (Chandra *et al.*, 2015). The addition of affordable staples like grains, pulses and other food items to wheat flour helps to increase the nutritious value of wheat based products (Noorfarahzilah *et al.*, 2014).

Pumpkin is one of the most significant vegetable crops in Sri Lanka, which has both medicinal nutritional benefits. The flesh pumpkin is rich in nutrients, that contains polysaccharides, pigments, active proteins, amino acids, minerals as well as antioxidants (Batool et al., 2022). Corn contains a number of important B vitamins, folic acid, vitamin C, and provitamin A (Gwirtz and Garcia-Casal, 2014). Soybeans have high protein content, fibre, antioxidants, and omega-3 fatty acids (Islam *et al.*, 2007).

Pumpkin, Corn and soybean were grown in tropical climates. Post-harvest loss of pumpkin is high due to improper handling and storage during the season. The cheapest healthy composite flour ingredients such as pumpkin, corn, and soybean is a potential alternative to overcome the nutritional deficiency in vulnerable groups.

The objectives of the research were to formulate and develop a nutritious composite flour pumpkin (Cucurbita maxima), soybean (Glycine max) and corn (Zea mays) flour as substitute for wheat flour in biscuit production for children under 5 years. Furthermore, analysis of functional properties of composite flour, nutritional and physical properties of developed biscuits and sensory evaluation were conducted.

MATERIALS AND METHODS

Location

The experiment was conducted in the food processing pilot plant and food microbiology laboratory of the Department of Food Science and Technology, Faculty of Agriculture, University of Peradeniya, Sri Lanka and the Food Research Unit of the Department of Agriculture, Gannoruwa, Sri Lanka.

Materials

Pumpkin flour (variety Suprima) and soybean flour were prepared in the laboratory. Wheat flour, corn flour, sugar, skim milk powder, egg and margarine were purchased from a local super market, Peradeniya, Sri Lanka.

Preparation of Pumpkin Flour

A well mature pumpkin was washed, peeled, and fibrous inner soft segments and seeds were removed. The pumpkin flesh was cut into 0.5 cm thickness slices and hot water blanched at 90 \pm 2 $^{\circ}$ C for 2 minutes followed by dehydration using an air convection tray drier (DHG-9146A, China) at 65 \pm 2 $^{\circ}$ C for 8 hours. The dried pumpkin slices were

milled using a laboratory mill and sieved (300 micrometre sieve). Pumpkin flour was stored at 4 °C in sealed High Density Polythene (HDPE.300 gauge) pouches.

Preparation of Soybean Flour

Soybean was soaked in water for 2 hours and boiled in water at 95 ± 2 °C for 30 minutes. Boiled soybeans were dehulled manually, washed and allowed to drain excess water. The wet soybeans were dehydrated in the tray dryer at 65 ± 2 °C for 9 hours.

The dried soybeans were milled, sieved (300 μ m) and stored at 4 0 C in HDPE pouches (300 gauge) to avoid spoilage and rancidity. Nutritious baby biscuits were developed using wheat flour and composite flour blends (wheat flour: pumpkin flour: soybean flour: corn flour) at 100:0:0:0: 85:5:55, 70:10:10:10 and 55:15:15:15, respectively (Table 1).

Sugar, margarine, and egg were mixed using a beater until sugar dissolved. Ammonium bicarbonate, sodium bicarbonate, skim milk powder, wheat flour, composite flour and vanilla powder were added to the creamy mixture and kneaded lightly to

obtain a homogenous consistency (Table 2).

The dough was rolled to a uniform sheet and cut into circular shape using a biscuit cutter (5cm diameter) and baked at 170 ± 2 °C for 30 minutes in a preheated bakery oven. Baked biscuits were allowed to cool and packed hermetically in sealed containers for further analysis and sensory evaluation.

The functional properties of flour blends, T-1, T-2, T-3 and T-4 were analysed for water absorption capacity (WAC %), oil absorption capacity (OAC %), swelling capacity (ml), foam capacity (%), foam stability (%) and bulk density (g/ cm³). WAC was determined by the method described by Sosulski *et al.*, (1976). A sample of flour (1.0 g) w kept at ambient temperature (26 ± 1 °C) for 30 minutes and centrifuged for 10 minutes at 3000 rpm. WAC was expressed as percentage of water of the sample.

OAC was determined by mixing a sample of flour (1.0 g) with 10 ml refined soybean oil, kept at ambient temperature for 30 minutes and centrifuged for 10 min at 3000 rpm.

	Table 1.	Formulations	of flour blend	ds for the bis	cuit dough.
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		Composite flour		
Treatment	Wheat flour %	Pumpkin %	Soybean %	Corn %
T1	100	0	0	0
T2	85	5	5	5
T3	70	10	10	10
T4	55	15	15	15

T1: wheat flour (100%) or control, T2: wheat flour (85%) + pumpkin flour (5%) + soybean flour (5%) + corn flour (5%), T3: wheat flour (70%) + pumpkin flour (10%) + soybean flour (10%) + corn flour (10%), T4: wheat flour (55%) + pumpkin flour (15%) + soybean flour (15%) + corn flour (15%).

Table 2. Formulation of ingredients.

T 1'	TA7 * 1
Ingredient	Weight (g)
Flour blend (wheat flour	100
and composite flour)	
Margarine	30
Sugar	35
Skim milk powder	25
Egg	20
Ammonium bicarbonate	1
Sodium bicarbonate	0.5
Vanila powder	0.13

OAC was expressed as percentage oil bound per gram of the sample (Sosulski and Youngs, 1979). The swelling capacity was determined by filling a flour sample up to 10 ml mark in 100 ml graduated cylinder and filled with water up to 50 ml. The top of the graduated cylinder was tightly covered

and mixed by inverting the cylinder. The suspension was inverted again after 2 min and allowed to stand for further 30 min. The volume occupied by the sample was taken after 30 minutes (Okaka and Potter, 1977).

Foam capacity and foam stability values were determined using the methods reported by (Narayana and Narasinga, 1982) with minor adjustments. A flour sample (1.0 g) was added into 50 ml distilled water at 28±2 °C in a graduated cylinder. The suspension was mixed and shaken for 5 minutes. The volume of foam after whipping for 30 sec was expressed as foaming capacity.

$$Foam \ capacity = \frac{AW - BW}{BW} \times 100\%$$

where, AW: after whipping, BW: before whipping.

Form stability is the volume of foam recorded after 1 hour whipping and presented as the percentage of the initial foam volume. Bulk density was determined using the method reported by Okaka and Potter (1977). A 100ml graduated cylinder containing the sample (50 g) was tapped 20-30 times to obtain a constant volume.

Chemical Analysis

Moisture, ash, crude protein, crude fibre and fat were determined in baby biscuits using the methods described in AOAC (2000). All experiments were conducted in triplicates. The carbohydrate content was calculated by subtraction method (Baljeet *et al.*, 2010). Minerals Fe and Ca were determined by Atomic Absorption Spectrophotometer (3300 Perkin-Elmer) as described in AOAC (2012). K was determined by Flame Emission Spectroscopy.

The diameter (cm) of baby biscuits was measured using vernier calliper with minimum value 0.05 mm. Three measurements were taken from the edge of the biscuits and calculated average value. The thickness was

measured in mm by Vernier calliper minimum value 0.05 Thickness was measured by stacking three biscuits on top of each other and taking average thickness. The spread ratio was determined by dividing the average diameter by the average thickness of the nutritious baby biscuits. The spread ratio of the supplemented biscuits was divided by the spread ratio of the control biscuits, and the result was multiplied by 100 to get the spread factor (Manohar and Rao, 1997).

Sensory Evaluation

Nutritious baby biscuit samples were evaluated for colour, flavour, aroma, texture/mouth feel, and overall acceptability with Hedonic scale (9: extremely like, 1: dislike extremely) using 30 semi-trained panellists.

Statistical Analysis

Parametric data was analysed using analysis of variance (ANOVA) and tested by One-way ANOVA. Mean comparison was done by Turkey test and 95% probability level. Friedman test was used to analyse nonparametric data. Statistical analysis was done using Minitab software package (version 21,

Minitab Pennsylvania, USA).

Microbiological Tests

Microbiological tests, total plate count and yeast and mould count were determined in baby biscuits using the methods described in Sri Lanka Standards for biscuits (SLS 251: 2010).

RESULTS AND DISCUSSION

The functional properties of composite flour are given in the Table 3. The water absorption capacity (WAC) of T3 (143%) and T4 (180%) was significantly (p <0.05) different from the control while no significant deference was observed between T1 and T2. The highest WAC in T4 was attributed to the presence of greater number of hydrophilic constituents like water-soluble fibre and proteins due to presence of 45% pumpkin, corn and soybean flour.

There was a significant difference (p <0.05) in oil absorption capacity (OAC) of the treatments vs the control. OAC is helpful in understanding the structural interaction in foods, particularly in flavour retention, and improves the mouth feel and shelf-life extension, especially in baking where

fat absorption is required. The results showed that all flour blends except the control performed equally the above characteristics.

There significant was no difference (p >0.05) in swelling capacity value among the three flour blends compared to the control. Foam Capacity was ranged from 12.42 to 17.02% and it was significantly different among the treatments (p >0.05). Foam stability was lowest in T4 (2.92%) while there was no significant deference (p >0.05) in T3 and T4 flour blends. The bulk density of flour treatments ranged from 0.58 to 0.66 g/ cm³. Pure wheat flour (T1), possessed the highest bulk density (0.66 g/ cm³), while 45% composite flour added flour blend (T4) possessed the lowest bulk density (0.58 g/ cm³) due to presence of 15% pumpkin and 15% soybean flour with the lowest percentage of wheat flour (55%).

There was no significant difference (p >0.05) in bulk density of the flour blends, T3 (0.62 \pm 0.01 g/ cm³) and T4 (0.58 \pm 0.02 g/ cm³). Flour with lower bulk density is desirable for manufacturing complementing foods such as baby biscuits (Akpata and Akubor, 1999).

Table 3. The functional properties of composite flour.

Property	Composite flour			
	T1	T2	Т3	T4
WAC %	123.21 ± 5.07°	$132.82 \pm 3.60^{\circ}$	145.43 ± 6.25 ^b	180.03 ± 2.27^{a}
OAC %	93.91 ± 0.75^{b}	99.26 ± 1.67^{a}	101.33 ± 2.36^{a}	101.51 ± 2.63^{a}
SC (ml)	16.67 ± 0.58^{a}	15.48 ± 0.50^{a}	16.20 ± 0.35^{a}	17.00 ± 1.32^{a}
FC %	12.42 ± 0.15^{d}	$13.63 \pm 0.34^{\circ}$	15.13 ± 0.37^{b}	17.09 ± 0.18^{a}
FS %	9.01 ± 0.28^{a}	6.12 ± 0.53^{b}	5.03 ± 0.18^{b}	$2.92 \pm 0.67^{\circ}$
$BD (g/cm^3)$	0.66 ± 0.01^{a}	0.65 ± 0.02^{a}	0.62 ± 0.01^{ab}	0.58 ± 0.02^{b}

Significance level (p <0.05), Value = Mean \pm SD. WAC = Water absorption capacity, OAC = Oil absorption capacity, SC = Swelling capacity, FC = Foam capacity, FS=Foam stability, BD = Bulk density.

Nutritional Composition of Baby Biscuits

Nutritional composition of baby biscuits is given in Table 4. Moisture content of the baby biscuits of four treatments was ranged from 3.90% (T2) to 3.99% (T4) whereas 4.05 % in the control. Lower moisture content of biscuits ensures long shelf life and lower tendency for product deterioration due to lower water activity. Ash content was ranged from 2.64% (T2) to 2.70%, (T4). There was a significant difference (p <0.05) in ash content of the control sample vs composite flour samples, T2, T3, and

T4. The reason is presence of higher mineral content in pumpkin, soybean and corn flour compared to wheat flour.

Fat content in baby biscuits was ranged from 7.72% to 19.91%, and substantially higher (p <0.05) than the control biscuit sample (6.79%). Fat content of soybean, corn and pumpkin flour was higher than fat content of wheat flour, especially soybean contain high (22.3%) fat content (Farzana *et al.*, 2017).

The fibre content of baby biscuits in composite flour was substantially higher (p <0.05) than the control sample (0.72%). The reason is the consisting of higher fibre content in soybean, corn and pumpkin flour compared to wheat flour. Fibre content of biscuits produced from 45% composite flour (T4) was 2.98%.

The protein content of nutritious baby biscuits developed from composite flour ranged from 3.15% (T2) to 5.25% (T4), which is substantially higher (p < 0.05) than the protein content of the biscuit developed from wheat flour (2.10%). Similar trend was observed for protein content in developed baby biscuits. Soybean and pumpkin flour are rich in protein than wheat flour, especially soybean contain high (34%) protein content (Farzana and Mohajan, 2015). There was a significant difference (p < 0.05) in carbohydrate content among four treatments. The lowest, 67.54% was recorded from biscuits with 45 % of composite flour blend due to lower (55%) wheat flour while the highest, 85.21% was recoded from the control with 100 % wheat flour. Addition of 45% composite flour with soybean flour, pumpkin flour, and corn flour (T4) has been resulted in lower

carbohydrate content in the baby biscuit sample.

Calcium content in baby biscuit was increased significantly (p <0.05) with increasing the percentage of substitution of composite flour of pumpkin, soybean and corn flour. The biscuits with 100% wheat flour contained 3.21 mg/ g while Ca 45% wheat flour substituted biscuits (T4) contained 58.95 mg/g. Iron content of baby biscuit was significantly different (p < 0.05) and ranged from 2.64 mg/g (T1) to 5.44 mg/g (T4). A similar trend has been shown for calcium and potassium as well due to gradual increase of composite flour. The potassium content of baby biscuits showed significant (p < 0.05) difference between T1 (288.13 mg/g) and T4 (711.68 mg/g) respectively. The reason could be due to the presence of extremely higher potassium content in pumpkin and soybean flour than in wheat flour.

The physical properties of biscuits are given in Table 5. The diameter of baby biscuits was 5.98 cm in T4, 6.25 cm in T2 while 6.13 cm in control sample. The thickness of biscuits ranged from 0.9 to 1.02 cm. Incorporation of pumpkin, corn and soybean flour has

Table 4. Nutritional composition of developed baby biscuits.

Constituent	T1	T2	T3	T4
Moisture %	4.05 ± 0.03^{a}	3.99 ± 0.04^{ab}	3.96 ± 0.03 bc	$3.90 \pm 0.025^{\circ}$
Ash %	2.50 ± 0.06^{b}	2.64 ± 0.02^{a}	2.66 ± 0.01^{a}	2.70 ± 0.03^{a}
Fat %	6.79 ± 0.04^{d}	$7.72 \pm 0.04^{\circ}$	14.18 ± 0.02^{b}	19.91 ± 0.02^{a}
Fibre %	$0.72 \pm 0.02^{\rm d}$	$1.69 \pm 0.03^{\circ}$	2.23 ± 0.01^{b}	2.98 ± 0.01^{a}
Protein %	2.10 ± 0.02^{d}	3.15 ± 0.02^{c}	$3.43 \pm 0.01^{\rm b}$	5.25 ± 0.01^{a}
Carbohydrate %	85.21ª	82.28 ^b	74.75°	67.54 ^d
Ca (mg/g)	13.21 ± 0.06^{d}	$31.09 \pm 0.13^{\circ}$	46.00 ± 0.39^{b}	58.95 ± 0.11^{a}
Fe (mg/g)	1.51 ± 0.08^{d}	2.64 ± 0.08^{c}	$3.18 \pm 0.21^{\rm b}$	5.44 ± 0.13^{a}
K (mg/g)	288.13 ± 6.70^{d}	$455.59 \pm 5.51^{\circ}$	527.88 ± 9.00^{b}	711.68 ± 14.65^{a}

Significance level (p < 0.05), n = 3, Value = Mean $\pm SD$.

resulted slight increase in the thickness of biscuits. However, there was no significant difference (p >0.05) in thickness values of biscuits prepared from T3 and T4 treatments. Increase in thickness may be due to the decrease in diameter. The changes in diameter and thickness were reflected by spread ratio and spread factor of biscuit. The spread ratio and spread factor of control biscuits were 6.62 and 100, respectively. The spread ratio and spread factor with decreased substitution composite flour comprised of pumpkin, corn and soybean flour.

The hardness of biscuits was significantly decreased with higher substitution of composite flour ratio. Thus biscuits possess softer texture and desirable mouths feel. The reduction of

hardness is mainly due to reduction of gluten content by substitution of wheat flour with others. Lightness value (70.14) of the biscuits with 100% wheat flour indicated a lighter surface colour compared to other three treatments due to presence of fibre and off white colour of the composite flour. Lightness value was significantly higher (p <0.05) in the control samples than the other treatments.

Table 5. Physical properties of developed baby biscuits.

Property		T1	T2	Т3	T4
Weight		11.98 ± 0.14 ^b	12.27 ± 0.04 ^b	13.14 ± 0.21 ^a	13.42 ± 0.20^{a}
Diameter		6.13 ± 0.03^{b}	6.25 ± 0.04^{a}	6.09 ± 0.05^{b}	$5.98 \pm 0.03^{\circ}$
Thickness		0.93 ± 0.01^{b}	1.00 ± 0.02^{a}	1.02 ± 0.00^{a}	1.02 ± 0.01^{a}
Spread ratio		6.62 ± 0.04^{a}	6.23 ± 0.18 ^b	5.97 ± 0.05 ^{bc}	$5.85 \pm 0.08^{\circ}$
Spread factor		100	94.15	90.07	88.37
Colour	L*	70.14 ± 0.29^{a}	67.95 ± 0.07 ^b	$65.85 \pm 0.43^{\circ}$	63.75 ± 0.31^{d}
	a*	$4.28 \pm 0.14^{\circ}$	4.56 ± 0.10^{bc}	4.92 ± 0.23^{b}	6.64 ± 0.17^{a}
	b*	$32.49 \pm 0.55^{\circ}$	$39.19 \pm 0.40^{\text{b}}$	41.80 ± 0.25^{a}	42.93 ± 0.73^{a}
Hardness		65.15 ± 0.08^{a}	57.94 ± 0.53^{b}	$52.65 \pm 0.86^{\circ}$	42.01 ± 0.47^{d}

Significance level (P<0.05), Value = Mean±SD.

Sensory Evaluation

The mean scores of sensory attributes are shown in Figure 1. Treatment, T3 was scored significantly (p <0.05) the highest mean score for colour (7.9, like moderately), followed by T2 (7.6, like moderately) and the control, T1 (6.0, like slightly), while the lowest was

received by T4 (nether like nor dislike). Thus, biscuits prepared form 100% wheat flour and T3 were scored the same mean score (like moderately) for aroma. There was no significant difference in aroma of the biscuits among the treatments and prefered as 'like slightly' to or 'like moderately'.

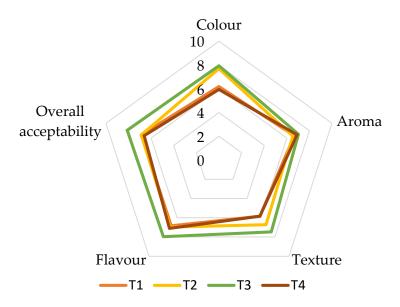


Figure 1. Mean scores of the sensory attributes of developed baby biscuits.

Biscuits samples of T3 scored significantly (p <0.05) the highest mean score for flavour (7.6), while other samples scored as like slightly (T4, T2 and T1). Biscuits sample, T3 scored significantly the highest (p <0.05) mean score for texture (7.1) followed by T2 (6.1) and T1 (5.8), while T4 was the lowest (5.5).

Substitution of composite flour has been improved the texture and mouth feel of the baby biscuits. In overall acceptability, T3 (7.7) scored significantly the highest mean score (p <0.05) followed by T2 (6.5) and T1 (6.3), while T4 (6.3) scored the lowest. Therefore, baby biscuits prepared using composite flour blend with 10% pumpkin, 10% soybean and 10% corn

flour (T3) was scored the highest mean scores for colour, flavour, texture, aroma and overall acceptability. The all sensory attributes of T3 samples were subjectively evaluated as "like moderately". Therefore treatment T3 was selected as the best composite flour formulation for baby biscuits production.

Microbiological Analysis

Both total plate count (540 CFU) and yeast and mould count (360 CFU) of T3 up to 30 days at ambient (28 ± 2 °C) storage were below the standard microbial limits for biscuits (Sri Lanka Standards, 2010).

CONCLUSION

Formulation and production of baby biscuits substituting wheat flour using the composite flour of pumpkin, soybean and corn flour was successful. The ratios of composite flour treatments (wheat flour: pumpkin flour: soybean flour: corn flour) were 100:0:0:0 (T1), 85:5:5:5 (T2), 70:10:10:10 (T3) and 55:15:15:15 (T4), respectively. The acceptability of the best flour blend was selected based on functional properties of the composite flour.

Fibre, fat, protein and minerals significantly improved were by substituting wheat flour by the composite flour (p < 0.05). The optimum baking conditions was 170 °C for 30 minutes. The highest mean scores for the sensory attributes were received by treatment, T3 with 70% wheat flour. The produced baby biscuits were safe for consumption up to one month stored in a hermetically sealed pouches or container. The microbial parameters tested at 30 days storage was below the product limits. Substitution of wheat flour using the formulated composite flour (pumpkin, soybean and corn flour) has been provided a higher potential for nutrient enrichment and biscuit manufacturing for young

children at lower cost. The developed composite flour formulation is an applicable alternative to combat the prevailing malnutrition condition among the children in Sri Lanka.

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