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Preparing Snacks for Treating Acute Diarrhea and Malnutrition for Kindergarten and Experiments on Male Mice

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Abstract

Different formulae snacks were extracted using corn grits, parboiled brown rice, defatted soybean, and carrot powder to give four formulae and compared with control snacks made from corn grits to treat diarrhea in mice. The results observed that the Extrusion processing causes an increase in all color values when increasing parboiled brown rice and carrot may be caused by elevated phenolic and flavonoids. In control, the water absorption index (WAI) and water-soluble index (WSI) improved their extruded formulae. Effects of different formulae snacks on diarrhea mice feeding 20% substituted different snacks from the basal diet during fifteen days experiment period. The results showed a significantly increased protein efficiency ratio, and biological value may show important advantages because this effect reflects the palatability and increase in gain body weight when given to rats with diarrhea. Therefore, increased intake of dietary fiber, protein, and antioxidants may enhance recovery and improve stool consistency in diarrhea mice. In addition, it could be noticed that diarrhea mice fed on different formulae snacks showed the highest decreases in kidney functions. Moreover, an increase in the concentration of albumin, globulin and total protein in the group fed different formulae snacks compared to the control negative may be caused by the mixture of flours had contained antioxidants and rich nutritional value. From the obvious outcomes, it is recommended that the snack-fed mice diarrhea groups gave the best results through two weeks; due to these formulae, snacks had nutritional compounds to the anti-diarrhea for mice.

Keywords: Corn grits, Defatted soybean, Parboiled rice, Diarrhea rats

Introduction

Diarrheal diseases are a major health concern in children all over the world. According to the World Health Organization, they are the second greatest cause of mortality in children under five, accounting for over 750,000 deaths yearly (WHO, 2013).

Diarrhea is a gastrointestinal illness marked by alterations in gut motions, frequency, and the consistency of the stools, which become watery or pasty due to increased water content. Diarrhea can be either acute or persistent (Smieja & Goldfarb, 2016).

This disease has various causes, including polluted food and drink, the utilization of pharmaceuticals, endocrine problems, and microorganisms like Vibrio cholera. Diarrhea is classified as acute (1-13 days), persistent (14 days or more), or chronic (more than 30 days), as well as physiological processes, with most etiologies including one or more of these mechanisms (Bhutta & Syed, 2016).

Diarrhea is a sign of disruption of the proteins, channels, and physical and chemical barriers in the gut, leading to problems transporting water and electrolytes in the digestive system (Chu *et al.*, 2020). Diarrhea can also be a symptom of a range of disorders. Pathological bile acid absorption, bacterial and viral infections, and chronic inflammatory illnesses can induce diarrhea. Although the death rate linked with diarrhea has decreased dramatically over time, it remains one prevalent cause of pediatrics, particularly in Asian and African nations (Camilleri *et al.*, 2017).

Flour samples made from rice, soybean, and orange sweet potato fractions were utilized to improve suitable formulations pointing to achieving fairly high nutrient contributions in blends in terms of protein, vitamin A, iron, and zinc without sacrificing nutrients and sensory properties. The final mixture is utilized to make cake, biscuits, and snack meals for children aged 2 to 6. To help enhance micronutrient consumption among young children, the manufacture and consumption of such goods are essential (Hannah & Wisdom, 2017).

Soybean is a legume with proteins closely related to animal protein and high protein content, accounting for roughly 40–45 percent of total solids. They are one of the best plant-based protein sources, with protein concentrations ranging from 36 to 56 percent of dry weight. Nutrition value compounds are all abundant in soybeans. The soybean is the most commercially significant bean on the planet, which contains rich amounts of protein for millions of people; in addition, their high nutrition value constituents are used for chemical products (Edima-Nyah *et al.*, 2020).

Maize grains comprise rich amounts of fibers, a high oil, and are rich in carbohydrates. Maize grains contain about 11% protein, 4% fat, 3% fiber, and 65% starch in their whole form (Edima-Nyah *et al.*, 2019).

Brown rice proteins are also good carriers for phenolic and volatile chemical transfer (Kelemen *et al.*, 2022). The quality of each rice cultivar aids the user in selecting the appropriate rice for their

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needs, which is determined by the end-use of rice (Charoenthaikij *et al.*, 2022). Grain-based foods provide a significant portion of the world's energy and nutritional needs, accounting for 25–50% of energy in Western diets and more than half of the world's calories. Furthermore, they are rich in carbs and dietary fiber (Jones *et al.*, 2015).

Carrots (*Daucus carota* L.) are a vitamin food high in carotene and ascorbic acid, as well as moisture, protein, fat, carbohydrates, sugars, and fiber. Carrots are one-of-a-kind roots high in carotenoids and have a distinct flavor due to terpenoids and polyacetylenes. Monoterpenoids and sesquiterpenoids are the most common terpenes, whereas polyacetylenes are made up of falcarinol molecules. The pigments found in various colored roots offer a wide range of medicinal effects for human health; for example, lutein in yellow carrots aids in producing macular pigments, which are necessary for good eye function (Raees-ul & Prasad, 2015).

Thus, this study aimed to produce snacks made from yellow maize, defatted soybean, parboiled brown rice and carrot at different levels for treating diarrhea and malnutrition in kindergarten and experimenting with them on male mice.

Materials and Methods

Materials

Defatted soybean, brown rice and corn grits and carrots were acquired from the local market in Saudi Arabia.

Parboiled brown rice was prepared according to the method of Biswas and Juliano (1998), and it was dried in an oven at 120 °C for 5 – 6 hours.

Yellow carrots were peeled and sliced into slices after being cleaned and washed with tap water. According to Park (1987), the slices were immersed in boiling water for 5 minutes before being dried in an oven at 40 -50 °C for 24 hours.

The dried parboiled brown rice and yellow carrot were milled in a Willy Mill to a fine powder.

Male albino mice (42 mice), whose weight ranged between 80-90 g and were obtained from Pharmacy College at King Saud University, were used in this experiment. The basal diet, which contained starch (70%), casein (10%), salt mixture (4%), vitamin mixture (1%), and cellulose (5%), was prepared according to AOAC (2000).

Kits for all hematological and antioxidants were purchased from Sigma-Aldrich Corp., MO, for use in the analysis. Also

Methods

Preparation of Snack Formulae

The corn grits of each defatted soybean, parboiled brown rice and carrot powder were replaced, as shown in **Table 1**.

Nutritional Value of Raw Materials

Chemical compositions of raw materials were estimated according to AOAC (2010) methods. Fractionation of total dietary fibers was decided in raw materials with the method Prosky *et al.* (1988).

Qawasmeh *et al.* (2012) assessed total phenolic content using Folin-Ciocalteu reagent (TPC), and it was determined as mg of gallic acid Equiv./ 100g of dry weight. Whereas, Eghdami and Sadeghi (2010) determined the flavonoids content as mg of Quercetin Equi/100g dry weight sample

Table 1. Recipe of extrusion of different formulae snacks from raw materials

Formulae	Corn grits	Parboiled brown rice	Defatted soybean	Carrot powder
Control snack	100	---	---	---
Formula (1)	70.0	10.0	10.0	10.0
Formula (2)	60.0	20.0	10.0	10.0
Formula (3)	50.0	30.0	10.0	10.0
Formula (4)	40.0	40.0	10.0	10.0

Snacks Preparation for Extrusion

The resulting formulations were extruded in a Brabender laboratory Twins -screw extruder (Duisburg DCE 330, New Jersey, USA) connected to a 2 mm die nozzle at specified constant extrusion conditions: screw speed of 140 rpm and barrel temperature of 140ºC, according to Nwabueze's calculations (2008). The extradited blends were wrapped in a tagged high-density polyethylene bag and maintained at room temperature (28ºC) until analysis was required.

Sensory Evaluation of Snack Formulae

A sensory panel of twenty experienced panelists from the Family Education Department, Faculty of Education, Umm Al-Qura, Saudi Arabia, evaluated the quality attributes of the snacks made from corn grits (control) and the different formulae of parboiled brown rice, defatted soybean, and carrot powder. The panelists evaluated five sensory qualities for each snack formula (taste, color, crispness, flavor, and overall acceptability) using a 9-point Hedonic scale (Lim, 2011).

Nutritional Value of Extruded Snacks

Chemical composition, the fraction of dietary fiber and quantitative antioxidant as total phenolic and flavonoids content were determined like the above methods

Color Properties of Extruded Snacks

A Hunter Colorimeter with an optical sensor was used to create a color scheme based on the *L*, a*, and b** color schemes. (Hunter Lab. Colorimeter model D 25) was used to measure the color of different snacks, according to Francis (1998).

Physical Characteristics of Extruded Snacks

According to Anderson *et al.* (1969), the water absorption index (WAI) and water solubility index (WSI) of snack samples were measured using several developed formulae.

Nutritional Experimental

All animal experiments should comply with the ARRIVE guidelines and should be carried out in accordance with the UK Animals (Scientific Procedures) Act, 1986 and associated guidelines, EU Directive 2010/63/EU for animal experiments, or the National Research Council's Guide for the Care and Use of Laboratory Male albino mice. It indicated in the manuscript that such guidelines had been followed.

For eight days, experimental rats were fed a baseline diet. Seven groups consisted in this experimental biological (six rats for each). The negative control group was the first group fed on a basal diet, while the other six groups were given 3% castor oil plus 7 % corn oil as an alternative to 10 percent corn oil in the basal diet. As a result, castor oil produces diarrhea in mice 1–2 hours after treatment of 1.0–3.0 ml, according to various studies (Rouf *et al.*, 2003). The experimental rat diarrhea was allocated into six groups at random. The first six rats in the main group were fed a basal diet for two weeks and classified as control-positive animals. The second group was fed on 20% control corn snacks which were substituted from a basal diet, and the third, fourth, fifth, and sixth major groups were fed on a basal diet which was substituted with 20% snacks formulae one, two, three, and four, respectively. For two weeks, the body weight gain, protein efficiency ratio, and biological value were measured every three days. After the trial, blood samples were drawn from each rat's orbital plexus, centrifuged at 3000 ppm to obtain serum, and stored in the freezer until analysis.
Kidney functions such as uric acid, urea, and creatinine in serum were determined according to Khozeimeh *et al.* (2017).
Doumas *et al.* (1971) determined total protein, albumin and globulin.

Statistical Analysis

ANOVA was used to evaluate the data collected, and a significant difference (p 0.05) was found in all variables; the data means test was used to assess the difference between the samples in all analyses. According to SAS (2008), the results were evaluated using the software SAS System for Windows.

Results and Discussion

Nutritional Value of Raw Materials

The chemical properties of corn grits, defatted soybean, parboiled brown rice, and carrot powder were estimated. The results in **Table 2** indicated that the highest total protein in defatted soybean was 55.60%, followed by parboiled brown rice was 12.51%. At the same time, corn grits and carrot powder were nearly equal in the amounts of protein by 9.50 and 9.355%, respectively. The soybean is the cheapest plant protein source, potentially improving the nutrition of millions of people, particularly in developing countries. Furthermore, high-protein soy products prevent malnutrition and encourage farmers to cultivate more soybeans due to rising market demand (Neha & Ramesh, 2012).

Ash content and crude fiber the highest in carrot powder was 15.28 and 22.17%, defatted soybean was 6.93 and 9.71%, and the parboiled brown rice was 3.21 and 4.21% and also, and the lowest corn grits was 1.65 and 2.71%, respectively. In addition to their nutritional value, carrots may be a good source of antioxidants. Carrot cultivars had protein, crude fiber, lipid, and carbohydrate levels ranging from 6.46 to 10.73 percent, 7.18 to 8.87 percent, 1.97 to 4.31 percent, and 6.25 to 8.39 percent, respectively. Carrots had the most fiber, protein, and carbohydrate content, as well as the lowest moisture content, making them the favored vegetable (Boadi *et al.*, 2021).

Total lipids were nearly equal for raw materials and ranged from 3.01 to4.40%. Moreover, the corn grits were the highest in total carbohydrates (82.20%), followed by parboiled brown rice was 77.06%; in addition, carrot powder and defatted soybeans were 49.80 and 24.54%, respectively. These results indicated that decreasing total carbohydrates in defatted soybean and carrot powder might be due to elevated protein, ash and crude fiber.

According to increased ash content and crude fiber in carrot powder and soybean, the highest total dietary fiber in carrot powder and soybean was 42.23, and 20.45%, followed by parboiled brown rice and corn grits were 11.31 and 8.27%, respectively. Furthermore, soluble and insoluble dietary fiber outcomes were similar to total dietary fiber results. Carrot powder also has the highest fiber level, and this high crude fiber content benefits human digestion and prevents constipation. As a result, meals high in fiber have been linked to a lower risk of colon cancer. Moreover, soybean supplied a good amount of total dietary fiber (16.60-19.50%) (Gazalli *et al.*, 2013).

Parboiled brown rice contained the greatest amount of phenolic and flavonoids, having 174.50 mg 100/g GAE and 96.0 mg/100 QE on the dry weight. Epidemiological studies have revealed that the antioxidant component content of rice may be linked to a decrease in the occurrence of some chronic diseases in rice-consuming parts (Goufo & Trindade, 2014).

Total phenolic and flavonoid content in carrot powder defatted soybean has nearly amounted to 126.8 and 120.0 mg/100g GAE for phenolic content and 91.25and 81.40 mg/100g QE for flavonoid content, respectively. Antioxidants such as alpha- and beta-carotene, lycopene, and lutein are abundant in carrots. The carrots get their orange, red, and yellow colors from them. Fruits and vegetables with anthocyanins have a purple tint (Stahl, 2016). Phenols are a vast class of secondary plant metabolites with a wide range of structures, ranging from relatively basic structures, such as phenolic acids, to phenols with multiple groups, such as flavonoids, to polymeric compounds based on these various classes. Some phenolic compounds are widely distributed, such as isoflavones in soybeans, while others are restricted to or plentiful in specific plants (Cheynier, 2012).

Meanwhile, the corn grits with the lowest phenolic and flavonoid content were 86.52 mg/100g GAE and 58.85 mg/100g QE compared with other raw materials. Many compounds, like carotenoids and phenolic chemicals, are found in the whole kernel of colorful maize. The most common types are phenolic acids and flavonoids, which come in a variety of soluble, free, conjugated, and insoluble bound configurations (Žilić *et al.*, 2012).

Table 2. Nutritional compounds of raw materials on a dry weight basis (g/100g)

Nutritional analysis	Corn grits	Defatted soybean	Parboiled brown rice	Carrot powder
Protein	9.50^{b}±0.83	55.60^{a}±3.48	12.51^{c}±0.97	9.35^{b}±0.81
Ash	1.65^{d}±0.01	6.93^{b}±0.17	3.21^{c}±0.03	15.28^{a}±1.05
Lipid	3.94^{ab}±0.02	3.22^{ab}±0.02	3.01^{b}±0.01	4.40^{a}±0.05
Crude fiber	2.71^{d}±0.01	9.71^{b}±0.76	4.21^{c}±0.03	22.17^{a}±1.64
Total carbohydrate	82.20^{a}±5.23	24.54^{d}±1.29	77.06^{b}±4.28	49.80^{c}±2.58
Total dietary fiber	8.27^{d}±0.46	20.45^{b}±1.38	11.31^{c}±0.94	42.23^{a}±2.49
Soluble dietary fiber	1.65^{d}±0.01	3.92^{b}±0.02	2.25^{c}±0.03	10. 76^{a}±0.97
Insoluble dietary fiber	6.62^{d}±0.18	16.53^{b}±0.98	9.16^{c}±0.79	31.47^{a}±2.13
Total phenolic mg/100g GAE	86.52^{c}±7.23	120.0^{b}±6.17	174.50^{a}±8.45	126.8^{ab}±7.61
Total flavonoids mg/100g QE	58.85^{c}±2.52	81.40^{b}±4.39	96.00^{a}±5.69	91.25^{a}±5.83

According to Duncan's New Multiple Range t-Test at the 5% level, means in a row followed by the same letter differ significantly.

Sensory Evaluation of Different Formulae Snacks

Since consumer approval is frequently encouraged in the marketing process of any food product, sensory evaluation is one of the most important aspects. Wójtowicz *et al.* (2013) employed a nine-point hedonic scale, while snacks deemed acceptable were those scoring above 5. **Table 3** showed that taste, color, and crispiness scores ranged from 5.00 to 8 for each, while flavor scores ranged from 6.00 to 8.00. Similarly, overall acceptability scores changed from 5.25 to 8.00. These results showed may be that the extrusion variables influence the sensory properties of the formula (4), made the same amounts of 40% from 40% corn grits and parboiled brown rice, as well as 10% from both defatted soybean and carrot powder, was the lowest score and maybe not be acceptability due to the highest-fiber. The fact that the taste, color, and flavor of the various snack formulae were acceptable could be attributable to the addition of carrot powder, which increased the taste, color, and flavor by 10%, making the formula snacks more delectable. The sensory study indicated that the enhanced 40 percent addition of parboiled brown rice observed in formula snacks (4) was unsatisfactory.

Furthermore, the gelatinization of the starch in yellow corn grits increased the texture and palatability of various snack formulations (El-Sohaimy *et al.*, 2019). Fortifying different formulae snacks with corn grits, parboiled brown rice, defatted soybean, and carrot powder might be an effect of some sensory properties like color and taste caused by the presence of defatted soybean. Because of the alleged health benefits of soy eating, soya products and goods with added soy are in high demand. Isoflavones and proteins in soybeans have been proven to increase lipid metabolism via activating or low-density lipoprotein receptors. Mohammed *et al.* (2012) found that the rich color of snacks could be due to the Maillard reaction, while the increased lysine level in extruded snacks could be owing to the Maillard reaction. Fortification of snacks up to 50% corn grits, 30% parboiled brown rice, and 10% from defatted soybean and carrot powder influenced the taste, color, and crispiness that judges in sensory evaluation did not accept.

Table 3. Sensory evaluation of different formulae snacks

Formulae	Taste	Color	Crispiness	Flavor	Overall acceptability
Control	9.00^{a}±0.94	9.00^{a} 0.89	9.00^{a}±0.86	9.00^{a} ±0.92	9.00^{a}±0.92
Formula 1	8.00^{b}±0.76	8.00^{b}±0.68	8.00^{b}±0.71	8.00^{b}±0.65	8.00^{b}±0.78
Formula 2	7.50^{bc}±0.54	7.50^{bc}±0.51	7.00^{c}±0.58	7.50^{bc}±0.49	7.38^{c}±0.38
Formula 3	7.00^{c}±0.53	7.00^{c}±0.56	6.00^{d}±0.47	7.00^{c}±0.43	6.75^{d}±0.31
Formula 4	5.00^{d}±0.37	5.00^{d}±0.42	5.00^{e}±0.31	6.00^{d}±0.38	5.25^{e}±0.24

Means in a row followed by the same letter differ, according to Duncan's New Multiple Range t-Test at the 5% level.

Macronutrient Composition, Fiber Fractions, and Antioxidant Content of Extruded Snacks

Chemical properties are one of the best characteristics to determine the quality of foods. Moreover, the proximate composition of any food determines its nutritional value; therefore, it needs to be determined. The present investigation following chemical properties and proximate analysis after extrusion-cooking of different formulae snacks have been studied, and the results are narrated in **Table 4**.

Results found the protein, ash, and crude fiber were gradually increased when increasing parboiled brown rice in different extrusion snacks from 14.95, 3.44, and 8.10 % to 16.18, 3.92, and 13.92%, respectively, compared with control corn grits snack was 9.27, 1.76, and 2.95%, respectively. The increases gradually in these results from extrusion snacks may be due to the highest protein content found in defatted soybean and parboiled brown rice, as well as ash content and crude fiber were the greatest in defatted soybean, parboiled brown rice, and carrot **(Table 2)**. Meanwhile, the lipid and total carbohydrates decreased gradually in different extrusion snacks ranging from 3.94, 69.75% to 3.67 and 62.31%, respectively, and compared with control corn grits snack was 3.82 and 82.20%, respectively. These gradually decreased may be caused by parboiled brown rice being the lowest in total lipid than other ingredients, and the defatted soybean was the lowest in total carbohydrates **(Table 2)**.

When the carrot and soybean were the greatest in total dietary fiber, the extrusion of different snacks increased gradually from 13.15% in formula (1) to 13.98% in formula (4), compared with control snack corn grits was 8.27%. Similarly, soluble and insoluble dietary fiber results were similar to total dietary fiber results in the extrusion of several snacks. Gluten-free whole-grain maize flours made without removing the germ from yellow and red kernels were utilized as functional food ingredients. Kernels maize demonstrated high-quality characteristics regarding physical qualities and chemical composition variations, indicating that they are suitable for various industrial applications, especially food production (Niklić *et al.,* 2020).

The same Table shows total phenolic and flavonoids in the different formulae snacks were the highest, especially for formula (4) contained 40% of parboiled brown rice in formula (4) (140.28 mg GAE/100 g and 79.19 mg/100g QE dw). These results indicated that increasing total phenolic and flavonoids content gradually for the different formulae snacks when increasing parboiled brown rice may be caused to the carrot powder, defatted soybean, and brown rice had contained the greatest natural antioxidant content. According to Acosta-Estrada *et al.* (2014), high temperatures can cause chemical bonds to rupture and phenolic compounds to be released, contributing to their determined amount. Although their no recommended level of antioxidants, the average daily intake of 591 mg vitamin C, 450 mg GAE, and 103 mg catechin, respectively, of fruits and vegetables. Meanwhile, legumes have recently gotten much attention because they're high in bioactive chemicals and can be used as ingredients in functional foods and other uses.

Table 4. Nutritional compounds of extruded snacks on a dry weight basis (g/100g)

Nutritional analysis	**Control snack**	**Different formulae snacks**			
		1	**2**	**3**	**4**
Protein	$9.27^{d}\pm0.94$	$14.95^{c}\pm1.15$	$15.40^{b}\pm1.28$	$15.78^{ab}\pm1.27$	$16.18^{a}\pm1.48$
Ash	$1.76^{d}\pm0.01$	$3.44^{c}\pm0.03$	$3.60^{bc}\pm0.03$	$3.76^{b}\pm0.03$	$3.92^{a}\pm0.03$
Lipid	$3.82^{b}\pm0.01$	$3.94^{a}\pm0.02$	$3.86^{b}\pm0.03$	$3.78^{c}\pm0.02$	$3.67^{d}\pm0.02$
Crude fiber	$2.95^{d}\pm0.02$	$8.10^{c}\pm0.86$	$10.04^{b}\pm0.79$	$11.98^{ab}\pm1.34$	$13.92^{a}\pm1.48$
Total carbohydrates	$82.20^{a}\pm8.36$	$69.57^{bc}\pm3.25$	$67.10^{bc}\pm4.26$	$64.70^{bc}\pm3.68$	$62.31^{c}\pm3.61$
Total dietary fiber	$8.27^{c}\pm0.74$	$13.15^{b}\pm1.12$	$13.42^{ab}\pm1.03$	$13.70^{ab}\pm1.21$	$13.98^{a}\pm1.31$
Soluble dietary fiber	$1.65^{e}\pm0.00$	$3.15^{d}\pm0.01$	$3.22^{c}\pm0.02$	$3.34^{b}\pm0.03$	$3.53^{a}\pm0.03$
Insoluble dietary fiber	$6.62^{c}\pm0.04$	$10.00^{b}\pm0.98$	$10.20^{ab}\pm0.94$	$10.36^{ab}\pm1.36$	$10.45^{a}\pm1.08$
Total phenolic mg/100g GAE	$80.52^{e}\pm7.69$	$122.68^{d}\pm7.69$	$128.58^{c}\pm6.86$	$134.43^{b}\pm7.21$	$140.28^{a}\pm7.95$
Total flavonoids mg/100g QE	$55.85^{d}\pm3.47$	$68.03^{c}\pm3.54$	$71.84^{b}\pm3.85$	$76.65^{ab}\pm4.01$	$79.19^{a}\pm4.16$

According to Duncan's New Multiple Range t-Test at the 5% level, means in a column followed by the same letter differ.

Color and Physical Characteristics of Extruded Snacks

When it comes to determining the quality of snacks and food products, color is crucial. Consumers choose snacks and food goods based on their looks, making decisions based on color brightness. **Table 2** shows how extrusion processing affects the color (*L*a*b**) of various equations (4). When raising parboiled brown rice, extrusion processing increases all color values, possibly due to the elevating fiber. Water content is also a factor; the more water added, the darker the samples get (Liu *et al.,* 2019).

Hunter color values of control and extruded mixes were found in **Table 5**. In extrusion snacks, color analysis was evaluated as (*L**) brightness, (*a**) (redness), and (*b**) (yellowness). Extrusion snack turned brown and red due to a high content of protein in defatted soybean and parboiled brown rice, as compared to an extruded snack (1) that contained 70% corn grits, 10% of both parboiled brown rice, defatted soybean, and carrot to give the best color followed by an extruded snack (1) that contained 70% corn grits, 10% of both parboiled brown rice, defatted soybean, and carrot to give the best color followed by an extruded snack (2 and 3). The Maillard reaction, which happens between sugars and amino acids during the extrusion process, may be responsible for the brown color of an extruded food (4) (Cauvain, 2016). In an extruded snack (4), the (*L**) and (*b**) values elevated, while the (*a**) value fell, which could be due to an increase in natural pigments. Furthermore, the deep hue of an extruded snack can be attributed to its high protein and crude fiber content (El-Sohaimy *et al.,* 2021).

Table 5 indicated the physical features of the control snack and its extruded different formulae. Results from water absorption index (WAI) in different extrusion formulae snacks and compared with control snack corn grits reported that extrusion formulae (1) nearly results (4.12 and 4.95 gel/g) may be due to the formula (1) contained the highest amount from corn grits a lower parboiled brown rice. Meanwhile, when substituted increases the parboiled brown rice from the corn grits, the starch increased and the WAI in the extrusion different formulae snack were gradually increased by 5.23, 5.82, and 6.11 gel/g, respectively. The WAI measures how much water starch is after swelling in water, whereas the WSI measures how much-unbound polysaccharides are present in a swelled sample (Bouasla *et al.,* 2017). The WAI of extrusion snacks was increased significantly when the amount of parboiled brown rice increased in different extrusion snacks. This is due to an increase in the overall starch content replaced by corn grits and an increase in the amount of water absorbed by the residual starch in the extrudate.

Furthermore, the rise in WAI is temperature-dependent. It can be linked to an increase in gelatinized starch content when starchy raw materials are replaced with vegetables, fruit, or high-fiber. According to Ondo *et al.* (2013), WAI values for enhanced extrudate porosity in the different equations ranged from 4.12 to 6.11 g/g.

Extrusion blends had significantly higher WAI than snack corn control. This could be partly due to the large quantity of gelatinized starch in extrusion formulations. The WSI of extruded products is linked to various modifications that occur during processing; these treatments at various parts of the extruder, which involve changing heating, pressure, and residence time, cause these changes (Ondo *et al.,* 2013).

WSI is associated with dextrinization and is related to the number of water-soluble molecules. In other words, the WSI can be used as a measure of starch degradation caused by extrusion cooking and as an indicator of molecular component degradation (Ding *et al.,* 2005). WSI in the extrudate formula control corn grits and formula (1) was the same, and in the highest amounts (5.79 and 4.25%), formulae 2, 3, and 4 were 3.76, 3.21, and 2.89%, respectively. The water solubility index of the extruded formulas decreased when defatted soybean and carrot powder were a combination with a ratio of 10% and gradually parboiled brown rice until 40%% substituted from corn grits. The decreased water solubility index in extrusion snacks may be caused by gradually increasing dietary fiber in the different formulae snacks. Water-insoluble complexes produced during extrusion-cooking between the macromolecules of amylose and components such as proteins and lipids may diminish the WSI. Extruded snacks have a larger water-accessible surface area, which allows starch and other water-soluble ingredients to interact. After extrusion-cooking, the WSI might evaluate the molecular breakdown of components released as soluble polysaccharides from starch (Wójtowicz *et al.,* 2018).

Table 5. Physical characteristics of extruded snacks

Formulae	Color			WAI*	WSI*
	L*	***a****	***b****		
Control	$85.18^{a}\pm 5.35$	$0.04^{e} \pm0.001$	$20.62^{a} \pm1.48$	$4.12^{e} \pm0.09$	$5.79^{a} \pm0.21$
Formula 1	$77.42^{b}\pm4.19$	$0.1\ 1^{d} \pm0.001$	$18.81^{b} \pm1.38$	$4.95^{d} \pm0.11$	$4.25^{b} \pm0.18$
Formula 2	$73.25^{c} \pm4.36$	$0.23^{c} \pm0.01$	$17.76^{c} \pm1.17$	$5.23^{c} \pm0.12$	$3.76^{c} \pm0.16$
Formula 3	$69.43^{d}\pm4.27$	$0.45^{b}\pm0.02$	$16.69^{d} \pm1.04$	$5.82^{b} \pm0.24$	$3.21^{d} \pm0.12$
Formula 4	$64.23^{e}\pm3.48$	$0.63^{a}\pm0.03$	$14.32^{e} \pm0.98$	$6.11^{a} \pm0.23$	$2.89^{e} \pm0.19$

According to Duncan's New Multiple Range t-Test at the 5% level, means in a column followed by the same letter differ.
WAI: water absorption index, results expressed as weight of gel/gram of dry samples
WSI: water solubility index, result expressed as a percent of dry solids in the supernatant.

Biological Investigation

Effect of Feeding of Different Snacks on Weight Rats and Protein Efficiency Ratio and Biological Value for Diarrhea Mice

Table 6 showed that feeding diarrhea rats with 20% of different formulae snacks gives a significant increase in the gain body weight animals from 17.56 g in diarrhea mice fed on corn snacks to 35.94 g in diarrhea fed on formula snacks (4) compared to control negative diarrhea rats was 15.12g. This increase may be due to an increase in total food consumed and an indication that supplementation with different formulae snacks supports growth because different formulae snacks have a high amount of nutritional value, such as protein and dietary fiber.

Diarrhea mice fed a diet containing 20% from different formulae snacks showed a significant increase in protein efficiency ratio, and biological values were 1.36 and 64.22% in formula snacks (1) to 2.22 and 73.28% in formulae snacks (4) compared with negative control diarrhea mice was 1.01 and 60.54%, respectively. This means the significant increase in protein efficiency ratio and biological value may show important advantages because this effect reflects the palatability when given to mice with diarrhea. Increased dietary fiber intake may aid healing and improve stool consistency in diarrhea sufferers (Schiller, 2005). More children on the partly hydrolyzed guar gum (PHGG) diet (84%) than those on the control diet (62%) had persistent diarrhea resolve, while the duration of diarrhea was reduced; there was a trend toward a decrease in daily stool weight that became significant on days 4–7. Compared to a non-fiber control, PHGG considerably reduced the mean frequency of diarrhea in children, resulting in significantly lower days with diarrhea for all feeding days and significantly lesser diarrhea (Alam *et al.,* 2005).

According to various studies, tannins, saponins, steroids, terpenes, alkaloids, and flavonoids are responsible for the antidiarrheal activity of diverse plant extracts via various mechanisms. Tannins and flavonoids have been connected to antidiarrheal action by increasing colonic water and electrolyte reabsorption, while the others have been linked to intestinal motility slowing. Tannins are well known for decreasing intestinal irritation and lowering the peristaltic index (Daswani *et al.,* 2010).

Table 6. Body gain weight, protein efficiency ratio and biological value for diarrhea mice fed on different snack

Diets	Initial weight (g)	Final weight (g)	Body gain weight (g)	Protein efficiency ratio	Biological value
Control negative	85.24±5.68	120.38±7.26	35.14±2.21	2.93±0.03	80.75±4.35
Control positive	84.69±5.49	99.81±6.45	15.12±1.45	1.01±0.01	60.54±3.68
Control snack	87.38±6.12	104.63±7.19	17.56±1.68	1.25±0.01	63.06±4.12
Formula 1	88.53±5.28	108.88±8.11	20.35±1.37	1.36±0.02	64.22±3.57
Formula 2	86.16±5.34	111.65±7.82	25.49±1.89	1.65±0.02	67.27±3.86
Formula 3	83.28±4.39	114.03±9.15	30.75±2.15	1.94±0.02	70.32±4.16
Formula 4	87.92±5.12	123.86±8.36	35.94±2.38	2.22±0.02	73.28±4.34

According to Duncan's New Multiple Range t-Test at the 5% level, means in a column followed by the same letter differ.

Effect of Different Snacks on Kidney Function for Diarrhea Mice

The kidneys are an essential part of the human body. They regulate water, electrolytes, and acid/base balances, create some hormones, and contribute to the metabolism of others (Blandy & Kaisary, 2013).

The results in **Table 7** showed that there was an increase in serum urea, uric acid, and creatinine in positive control mice fed on a basal diet was 43.28, 6.82, 1.15 mg/dl, respectively, as compared with normal rats fed on a basal diet was 27.35, 3.57, and 0.83mg/dl, respectively. The most often utilized indicator of renal function is serum creatinine measurement. However, a large increase in creatinine and urea levels may observe kidney failure, followed by retention of urea and creatinine in the blood (Amin *et al.,* 2016). Urea is the primary carrier for eliminating harmful ammonia from the body and is the major end product of animal protein catabolism. When a higher urea concentration is in serum, it increases ammonium. Increased urea levels are also related to nephritis, renal ischemia, and urinary tract blockage in general, and urea determination is highly useful for monitoring kidney function (Taki *et al.*, 2005).

It could be noticed that diarrhea mice fed on different formulae snacks showed the highest decrease of urea, uric acid, and creatinine enzymes were 37.58, 5.54, and 0.89 mg/dL in formula (1) and the highest decrease was 28.26, 3.82, and 0.54 mg/dL in formula (4), respectively. These results mean different formulae snacks contained parboiled brown rice which contained high amounts of total carbohydrates, dietary fiber and antioxidants, defatted soybean was the greatest protein, and dietary fiber and natural antioxidant, carrot powder which contained dietary fiber, and corn grits rich in total carbohydrates. These components in the different formulae snacks protected kidneys in diarrhea mice.

Table 7. Kidney functions for diarrhea mice fed on different snack

Diets	Urea mg/dL	Uric acid mg/dL	Creatinine mg/dL
Control negative	27.35^{f} ±1.16	3.57^{f} ±0.02	0.53^{f} ±0.001
Control positive	43.28^{a} ±3.85	6.82^{a} ±0.06	1.15^{a} ±0.01
Control snack	$40,43^{b}$±3.25	6.18^{b} ±0.04	1.02^{b} ±0.01
Formula 1	37.58^{c}±2.48	5.54^{c}±0.03	0.89^{c} ±0.003
Formula 2	34.73^{d}±2.64	4.90^{d}±0.02	0.76^{d} ±0.002
Formula 3	31.65^{e}±2.16	4.36^{e}±0.02	0.63^{e} ±0.001
Formula 4	28.26^{f}±1.76	3.82^{f}±0.01	0.54^{f} ±0.001

According to Duncan's New Multiple Range t-Test at the 5% level, means in a column followed by the same letter differ.

Effect of Different Snacks on Protein Fractions for Diarrhea Mice

Serum protein fractions were estimated in the mice diarrhea fed on different formulae snacks for15 days. Results from **Table 8** noticed the total protein; albumin, and globulin were 4.94, 3.06, and 2.45g/dL in formula (1) and increased to 7.17, 4.52, and 3.25g/dL in formula (4), respectively in diarrhea mice fed on different formulae snacks. These results equal to or nearly healthy control rats fed on basal diet were 7.82, 4.92, and 3.45g/dL, respectively, and the lowest results from control positive diarrhea mice fed on basal diet were 3.52, 2.16, and 1.97g/dL, respectively, these means diarrhea in rats control negative was caused trouble in the kidney.

No significant differences or slight increases have happened between the concentration of albumin, globulin, and total protein in the group fed different formulae snacks compared to the control negative may be caused the mixture flours had contained antioxidants and good sources from the nutritional value. Moreover, the highest fiber content in the presently developed snacks may be because of the inclusion of soy flour, parboiled brown rice, corn grits, and carrot powder in the preparation of different snacks (Farzana & Mohajan, 2015).

Secondary metabolites of flavonoids are known to reach circulation, and mounting evidence shows that these secondary metabolites improve the total protein, globulin, and albumin; also, the original flavonoids found in food have biological effects on the body (Kroon *et al.,* 2004).

Table 8. Protein fractions for diarrhea mice fed on different snack

Diets	Total protein (g/dl)	Albumin (g/dl)	Globulin (g/dl)
Control negative	7.82^{a}±0.42	4.92^{a} ±0.02	3.45^{a} ±0.02
Control positive	3.52^{f}±0.06	2.16^{f}±0.01	1.97^{f}±0.06
Control snack	4.23^{e}±0.04	2.61^{e}±0.01	2.21^{e}±0.12
Formula 1	4.94^{d}±0.04	3.06^{d} ±0.03	2.45^{d} ±0.50
Formula 2	5.69^{c}±0.07	3.51^{c}±0.04	2.69^{c}±0.22
Formula 3	6.36^{b}±0.06	3.96^{b} ±0.02	2.93^{b}±0.07
Formula 4	7.17^{a} ±0.06	4.52^{a} ±0.03	3.25^{a} ±0.03

According to Duncan's New Multiple Range t-Test at the 5% level, means in a column followed by the same letter differ.

Conclusion

This study was designed to estimate defatted soybean, corn grits, parboiled brown rice, and carrot powder to produce different formulae snacks and improve the nutritional status, including biological experiments for diarrhea mice. The results observed that the extrusion formulae snacks contained the highest amounts of protein, dietary fiber, carbohydrates, and natural antioxidants. Moreover, the extrusion snack-fed mice diarrhea groups gave the best results through two weeks. Therefore, these formulae snacks had nutritional compounds such as anti-diarrhea in mice.

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