Protein Quality of Extruded Egg-Based Whole Wheat Snack

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ABSTRACT

This paper aims to evaluate the protein quality and the effect of extrusion on the extrudates from whole-wheat flour and chicken egg and also, the effect of storage on amino acid profile and protein solubility of the extrudates. The whole-wheat flour was mixed separately either with the raw whole egg, egg yolk or egg albumen, a twin-screw extruder was used for the extrusion of the samples. After production, one portion of the extrudates was used for initial quality evaluation (protein solubility, protein digestibility and amino acid profile). The second portion was stored at ambient room condition and the third portion was stored at 37 ^{o}C in a thermostatically controlled oven. The extrudates were stored for twelve weeks. Ambient room relative humidity and temperature were monitored throughout the period of storage. Amino acid profile of the extrudates were determined at the beginning and at the end of the storage period of the extrudates stored at ambient temperature conditions. Protein solubility of the extrudates were determined at the beginning and subsequently two weeks interval for the samples stored at ambient room condition and the samples stored at 37 ^{o}C for ten weeks. Protein digestibility of the extrudates were determined at the beginning for the samples stored at ambient room condition. Data analysis was carried out using one-way analysis of variance. Results show that extrusion cooking decreased the protein digestibility of the samples. Storage had no effect on the protein solubility of the samples which were in the same range (69.50 - 88.50%) in all of the samples. However, temperature of storage had a slight effect, samples stored at 37 ^{o}C reduced more in solubility compared to those stored at ambient room temperatures. Storage had no effect on the amino acid contents of the samples.

Keywords: Protein, Storage, Temperature, Amino acid,

INTRODUCTION

A snack, usually a light meal and consumed between meals may be defined as a food or drink (Miskelly, 2017). Adegunwa *et al*. (2017) defined snack in comparison with a regular meal as a smaller portion of food usually eaten between meals. Snacks are highly consumed all over the globe but sadly, most snacks are not nutritionally balanced because they do not contain proteins (Adegunwa *et al*., 2017). The world leading consumer of snack foods is the United States, followed by England, then Germany and finally, France (Saldivar, 2016). Tumuluru (2016) stated that snacks without nutritional benefit is regarded as junk and eating of such snack is unhealthy as it can lead to health related issues such as excess body fat, heart diseases, abnormally high blood pressure. The nutritive value needs to meet certain nutritional requirements. The global food technology landscape has evolved with consumers more conscious of their health status through what they consume (Contato and Conte-Junior, 2025) especially snacks which is consumed more often. Health facilities such as hospitals are not left out in nudging consumers to go for healthier mindful snack choices and hence better dietary decisions (Tzikas *et al*., 2026).

Nutritionally adequate diets are now produced all over the globe by researchers so as to combat protein malnutrition. Individuals in need of a light and/or fast food may eat snacks such as children as a result of their high activity which makes them easily hungry. Pies, chin chin, popcorn and biscuits are some examples of snacks (Anyika *et al*., 2009). Ervina *et al*. (2025) developed tortilla chips enriched with cricket flour with the aim of increasing the protein content of the snack. Mnayer and Joubrane (2025) produced a novel protein-rich

potato snack enriched with legume protein sources such as peanut, pea protein, lentil soybean flour, pea, chickpea and whey proteins.

Egg possess health-promoting functions (Lesnierowski and Stangierski, 2018). Food products which are egg-based may be regarded as a functional food owing to the high-quality protein content **(**Patrignani *et al*., 2013). Egg is the most affordable source of animal protein available to the general populace in developing countries. Egg is said to contain complete (first class) proteins (Khan *et al*., 2017; Guha *et al*., 2019). British researchers pronounced egg a "super food" due to its positive impact on health and helps in fighting obesity.

Miskelly (2017) reported that wholegrain snacks are commonly produced now for health benefits. Wheat is a commonly consumed cereal all over the globe (Gammoh *et al*., 2018) and has been classified as the world's most consumed cereal grain. Snacks such as pies, pizza, cookies and biscuits are produced mainly from wheat flour. Whole grain consumption prevents oxidative stress due to the content of bioactive compounds (Esfandi *et al*., 2019) therefore, may be regarded as an excellent health-building food with health promoting benefits. Wheat has a low glycaemic index, and a leading source of cereal protein compared with maize and rice (Bhat *et al*., 2016). Chicken egg and whole wheat have numerous benefits, therefore, producing a product from both will make available a very healthy product with high protein content.

Extrusion cooking involves a high temperature within a short time, however, the desired product determines the extrusion temperature, hot for snacks and cold for pasta production. Other examples of extruded products include breakfast cereal, beverages, pet treats and instant powders (Robin *et al*., 2014). Cereal possess viscoelastic property which enables expansion during extrusion cooking. Most extruded products are cereal-based and most consumers generally accept cereal including children and adults also the elderly. Addition of protein sources such as egg to ingredients for extrusion will result in a more balanced product. Egg has been used in one form or the other by some researchers in some extruded food products. Singh *et al*. (2007) used egg proteins, Stojceska *et al*. (2008) used egg whites, Valverde *et al*. (2016) used egg yolk. Zardetto and Rosa (2009) used fresh egg. Alamprese (2017) reviewed the use of egg and egg products in foods.

Egg is a very good option to complement extruded food products as a result of the high protein content. Nigeria is also a leading egg producer in Africa. Thus, egg is cheap and readily available in Nigeria. Many snacks are produced through extrusion cooking. Extrusion cooking involves high temperature which may alter the characteristic quality of the raw materials on extrusion. However, there is paucity of information on the quality characteristics of extruded egg-based wheat snacks. Therefore, there is the need to investigate the protein quality of egg-based wheat snack products.

MATERIALS AND METHODS

Materials

Four bags of 50 kg each of whole-wheat flour were purchased from Supreme Flour Mills, 6, President Burgers Street, Pretoria West 0183, Pretoria, South Africa. Thirty-two crates of thirty eggs each of freshly laid eggs were purchased from Northwest University farm, Mmabatho Unit 5, Mafikeng 2790, Mafikeng, South Africa.

Methods

The samples were prepared using the method of Nwadi *et al*. (2025). Egg whites were separated from the egg yolk for samples requiring either egg yolk or egg white using an egg separator. The whole-wheat flour and raw whole egg or raw egg yolk or raw egg white were mixed using a 50-litre paddle mixer constructed by Centre for Advanced Manufacturing (CFAM), Potchefstroom, South Africa, in different proportions (Table 1**)**. After mixing, extrusion of the samples was done in TX-32 Laboratory Scale (300 kg/h maximum) twin screw extruder made by CFAM. The extrusion was done in a batch size of 20 kg (using a Platform Scale, Model: Micro A12E in CFAM) per run for each of the samples and the extrusion parameters were constant for all the samples at a screw speed of 700 rpm, feed rate of 53.6 – 78.9 kg/h (50 – 60 %) driven by a 6 – 9.7 kW motor, 20.8 – 24.1 AMP, 50 – 55 % Torque, temperature of 139 – 153 ºC and 20 % feed moisture content. Each run lasted 14.22 – 15.02 minutes. A spaghetti die of 1.8 mm (2 rings) with a 40 – 60 % cutter was used in each run.

After production, the extrudates were divided into three portions. One portion was used for initial quality evaluation for protein solubility, protein digestibility and amino acid profile, using standard methods. The second portion was stored at ambient room condition and the third portion was stored at 37 ºC in a thermostatically controlled oven. The extrudates were stored for twelve weeks. Ambient room relative humidity and temperature were monitored throughout the period of storage.

Amino acid profile of the extrudates were determined at the beginning and at the end of the storage period of the extrudates stored at ambient temperature conditions. Protein solubility of the extrudates were determined at the beginning and subsequently two weeks interval for the samples stored at ambient room condition and the samples stored at 37 ºC. Protein digestibility of the extrudates were determined at the beginning for the samples stored at ambient room condition.

Table 1: Ingredient combinations for products

Sample	Ratio (whole wheat flour to chicken egg)	whole wheat flour (kg)	Chicken egg (kg)	Total quantity (kg)
RI (Whole wheat flour)	100:0	20	0 (no egg)	20
R2 (Whole wheat flour and raw whole egg)	85:15	17	3 (60 whole eggs)	20
R3 (Whole wheat flour and raw whole egg)	80:20	16	4 (80 whole eggs)	20
R4 (Whole wheat flour and raw egg yolk)	85:15	17	3 (187.5 egg yolks)	20
R5 (Whole wheat flour and raw egg yolk)	80:20	16	4 (250 egg yolks)	20
R6 (Whole wheat flour and raw egg white)	85:15	17	3 (81 egg whites)	20
R7 (Whole wheat flour and raw egg white)	80:20	16	4 (108 egg whites)	20

Calculation used above: One whole egg = 50g, one egg white = 37g, one egg yolk =16g

Source: (Nwadi *et al*., 2025)

Determination of protein solubility

The procedure reported by Annor *et al*. (2010) was used to determine the protein solubility of samples. One and a half grams (1.5 g) of the milled extrudate was weighed into a beaker and 75 ml of 0.2 % (0.36 N, pH 12.5) potassium hydroxide was added. The sample was then stirred for 20 min on a magnetic stirrer plate and centrifuged (Model: Hettich Zentrifugen D-7200 Type 2008) at 2,700 rpm for 15 min. The supernatant was then filtered through glass wool into a beaker, being careful to avoid transferring residue. The supernatant was centrifuged again and 15 ml supernatant was transferred into two Kjeldahl tubes for duplicate analysis (this gives 0.3 g aliquot of the original sample); 12.5 ml concentrated sulphuric acid and 2 ml hydrogen peroxide were added to each tube for nitrogen determination by the kjeldahl method. The total nitrogen of the original sample was also determined. Protein solubility was expressed as the soluble protein fraction (from supernatant) as a percentage of the total protein in the sample

$$\% \text{ protein solubility} = \frac{\text{protein in filtrate}}{\text{Total protein in sample}} \times 100$$

In vitro Protein digestibility of extruded snacks

This was determined according to the procedures reported by Onyango *et al*. (2004). Two hundred milligrams (200 mg) of the milled extrudate was transferred to a 100 ml Erlenmeyer flask containing 35 ml 0.1 M sodium

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citrate dehydrate (pH 2.0) with pepsin (1.5 g pepsin/litre, KEM LIGHT Laboratories Pvt. Ltd,. Mumbai, India). The mixture was incubated for 2 hours in a water bath at 37 ^{o}C, shaken every 20 min and then centrifuged (Model: Hettich Zentrifugen D-7200 Type 2008) at 6,000 rpm for 15 min. The residue was collected on a nitrogen free filter paper and washed with 10 ml phosphate buffer (pH 7.0). The filter papers were dried at 108 ^{o}C for 3 hours. The dried residue was analysed for nitrogen using Kjeldahl method (CP2).

$$\% \text{ in vitro protein digestibility} = \frac{CP1-CP2}{CP1}$$

Where:

CP1 = Total protein of extrudate (crude protein of sample)

CP2 = Total protein after digestion with pepsin

Determination of Amino Acid Profile

The Amino Acid profile in the sample was determined using methods described by Benitez (1989). The sample was dried to constant weight, defatted, hydrolyzed, evaporated in a rotary evaporator and loaded into the Applied Biosystems PTH Amino Acid Analyzer located in Jos, Plateau State, Nigeria.

Defatting Sample:

The sample was defatted using chloroform/methanol mixture of ratio 2:1. The sample (2.0 g) was put in extraction thimble and extracted for 15 hours in Soxhlet extraction apparatus (AOAC, 2010).

Nitrogen Determination:

A small amount (150 mg) of ground sample was weighed, wrapped in Whatman filter paper (No.1) and put in the Kjeldahl digestion flask. Concentrated sulphuric acid (10 ml) was added. Catalyst mixture (0.5 g) containing sodium sulphate (Na_2SO_4), copper sulphate ($CuSO_4$) and selenium oxide (SeO_2) in the ratio of 10:5:1 was added into the flask to facilitate digestion. Six pieces of anti-bumping granules were added.

The flask was then put in Kjeldahl digestion apparatus for 3 hours until the liquid turned light green. The digested sample was cooled and diluted with distilled water to 100 ml in standard volumetric flask. Aliquot (10 m1) of the diluted solution with 10 ml of 45 % sodium hydroxide was put into the Markham distillation apparatus and distilled into 10 ml of 2 % boric acid containing 4 drops of bromocresol green/methyl red indicator until 70 ml of distillate was collected.

The distillate was then titrated with standardize 0.01 N hydrochloric acid to grey coloured end point.

$$\text{Percentage Nitrogen} = \frac{(a-b) \times 0.01 \times 14 \times V \times 100}{W \times C}$$

Where:

a. = Titre value of the digested sample

b. = Titre value of blank sample

v. = Volume after dilution (100 ml)

W. = Weight of dried sample (mg)

C. = Aliquot of the sample used (5 ml)

14. = Nitrogen constant in mg.

Hydrolysis of the sample

The defatted sample (1.2740 g) was weighed into glass ampoule. 7 ml of 6N HCl was added and oxygen was expelled by passing nitrogen into the ampoule (this is to avoid possible oxidation of some amino acids during hydrolysis e.g methionine and cysteine). The glass ampoule was then sealed with Bunsen burner flame and put in an oven preset at 105 ^{o}C± 5 ^{o}C for 22 hours. The ampoule was allowed to cool before breaking open at the tip and the content was filtered to remove the humins.

The filtrate was then evaporated to dryness using rotary evaporator. The residue was dissolved with 5 ml to acetate buffer (pH 2.0) and stored in plastic specimen bottles, which were kept in the freezer.

Loading of the hydrolysate into analyzer

The amount loaded was 60 microlitre. This was dispensed into the cartridge of the analyzer. The analyzer is designed to separate and analyze free acidic, neutral and basic amino acids of the hydrolysate.

Method of Calculating Amino Acid Values

An integrator attached to the Analyzer calculates the peak area proportional to the concentration of each of the amino acids.

NOTE: g/100g Protein = g/16g N

Statistical analysis

One-way analysis of variance (ANOVA) in a completely randomized design (CRD) was used for data analysis; Duncan's New Multiple Range Test (DNMRT) was used for mean separation (Steel and Torrie, 1980). Significance was accepted at $p<0.05$.

RESULTS AND DISCUSSION

Amino acid profile

The amino acid content of a food determines its protein quality, and food nutritional value depends on the protein quality (Comai *et al*., 2011). The samples contain a good balance of the amino acids (Table 2). These amino acids are building blocks for certain compounds in the body (proteins, nucleic acids, nucleotides, peptide hormones and some neurotransmitters). Amino acids are also components of erythrocytes, lymphocytes and are useful in the catabolism of intestinal mucosa (Gutiérrez-Gamboa *et al*., 2019)

The essential amino acids include histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan and valine. Chicken egg contains these essential amino acids (Mori *et al*., 2020). Whole wheat flour also contains these essential amino acid (Cornell, 2012). Essential amino acids may be classified as dietary indispensable because the body cannot synthesize these amino acids (Lam and de Lumen, 2003; Courtney-Martin and Pencharz, 2016; Higgs and Boland, 2014). Since both the egg and the whole wheat flour contain these essential amino acids, the extrudates were rich in amino acids (Table 2). Proteins have a globular structure which is destroyed by extrusion cooking. The extrusion temperature did not affect the amino acids because they are thermally stable (Wang *et al*., 2012), therefore, they are able to withstand extrusion temperatures. Omwamba and Mahungu (2014) reported high percentage (88 to 95 %) of amino acids retention in extrudates.

The egg white and egg yolk contained higher quantity of the amino acids compared with the whole wheat flour except for glycine (3.99 mg/ 100 g), proline (8.45 mg/ 100 g) and glutamic acid (25.45 mg/ 100 g) (Table 2) which are not essential amino acids. At the beginning of storage, the samples (R2 and R3) containing whole egg had more of the amino acids, tryptophan, cysteine, methionine, histidine, tyrosine, lysine, threonine, isoleucine, alanine, glycine, phenylalanine, valine, arginine, aspartic acid, leucine, except for serine (R7 containing egg white had higher content than R2), proline (R6 containing egg white had higher content than R2) and glutamic acid (R6 containing egg white had higher content than R2).

Tryptophan: The egg yolk (1.84 mg/100 g) contained more tryptophan than the egg white (1.76 mg/100 g) and whole wheat flour (1.11 mg/100 g) (Table 2).

The samples containing whole egg (R2 and R3) had the highest quantity of tryptophan. This may be attributed to the additive effect of tryptophan contained in the egg white, egg yolk and whole wheat flour compared to other samples containing either egg yolk (R4 and R5) or egg white (R6 and R7) or only whole wheat flour (R1).

On extrusion, tryptophan contents of the resulting products were higher than that of the extruded whole wheat flour showing that the added egg products, through supplementation, contributed to upgrading the tryptophan in the products. On the other hand, the tryptophan contents of the extrudates were less than those of the raw egg white and yolk due to dilution effect from whole wheat flour. Although, the tryptophan contents of raw egg yolk was higher than that of raw egg white, extruded products containing egg yolk lost more tryptophan than samples containing egg white. This accounts for why products R2, R3, R6 and R7 contained more tryptophan than products R4 and R5. The reason for this may be connected to the fact that egg yolk contains more reactive substances (such as fatty acids and sugars) that can be involved in complexing reactions with tryptophan under heat and cause it to be diminished.

On storage, the tryptophan contents of the extruded products reduced further, except in products containing whole wheat flour and egg yolk. The reduction on storage could be attributed to deteriorative complexing reactions but the reasons for the increase in the case of yolk-containing products is not clear but could be attributed partly to the higher solids of egg yolk that reduced rate of reactions and partly to concentration due to moisture loss.

Friedman (2018) reported that tryptophan promotes normal growth and serves as precursor in the formation of many bioactive compounds such as serotonin and melatonin. Cereal proteins may be said to be deficient in tryptophan because it is available in very low quantities (Comai *et al*., 2011).

Cysteine: Cysteine is a Sulphur-containing non-essential amino acid but it is synthesized from methionine through transmethylation and transculturation reactions. Cysteine improves the antioxidant status of the body in health and disease by contributing to the decrease of oxidative stress (McPherson and Hardy, 2011; Clemente Plaza *et al*., 2018). The cysteine content of R1 and R6 at the beginning and end of storage were not significantly different ($p>0.05$) from each other, while the values were comparable in R2, R3 and R5 but significantly different in R4 and R7. The quantity of cysteine increased in R4 and R5, whereas it decreased in R1, R2, R3, R6 and R7 at the end of storage.

Table 2 shows that the cysteine contents of the whole wheat flour was 1.3 mg/100 g and that of raw egg yolk was 2.59 mg/100 g while that of raw egg white was 2.59 mg/100 g. The higher cysteine contents of the extruded products compared to whole wheat flour could be attributed to concentration effects and partly supplementation from the egg products. The extruded products also showed lower cysteine contents compared to egg yolk and egg white due to dilution from wheat flour. Those containing whole egg were higher in cysteine due to combined contributions from whole wheat flour, egg yolk and egg white. Those containing egg yolk only were the lowest due to complexing reactions of cysteine with other components of egg yolk during heat processing which diminished the quantity of cysteine in the products. On storage, all samples reduced in cysteine contents except R4 and R5. The reduction could be due to deteriorative complexing reactions. Those of R4 and R5 increased during storage due to concentration effects as well as slower reactions resulting from more solid contents. More antioxidant components of yolk could also contribute to preventing oxidation and loss of cysteine.

Methionine: Methionine has an important function of being a precursor of homocysteine (Garlick, 2006) and S-adenosylmethionine (Fagundes *et al*., 2020). The methionine content of R1 and R5 before and after storage period were not significantly different ($p>0.05$) from each other, R2 was comparable whereas R3, R4 and R7 were significantly different ($p<0.05$). The quantity of methionine increased in R1, R4 and R5, whereas it decreased in R2, R3, R6 and R7 at the end of storage.

Histidine: Histidine is required for protein synthesis and regulates secretion of hormones (Matsui *et al*., 2017). The histidine content of R1 before and after storage period were not significantly different ($p>0.05$) from each other, R2 and R5 were comparable whereas R3, R4, R6 and R7 were significantly different ($p<0.05$). The quantity of methionine increased in R1, R4 and R5, whereas it decreased in R2, R3, R6 and R7 at the end of storage.

Tyrosine: Tyrosine is a precursor for the synthesis of dihydroxy-L-phenylalanine and dopamine which are neurotransmitters in the body (Korner *et al*., 2019). The tyrosine content of R6 before and after storage period were not significantly different ($p>0.05$) from each other, R1 and R2 were comparable whereas R3, R4, R5 and R7 were significantly different ($p<0.05$). At the end of storage, the quantity of tyrosine in R1 remained the same, increased in R4 and R5, whereas it decreased in R2, R3, R6 and R7.

Lysine: Lysine is important for growth and muscle development (Lee *et al.*, 2020). Eggs, just like meat and milk are classified as complete protein because it contains all the essential amino acid in the proportion required by humans, on the other hand, most plant-based foods have a limiting amino acid. The limiting amino acids (methionine, lysine, histidine) are usually found in small amounts in foods (Yoder *et al*., 2020). The extrudates may be said to be very rich in protein because it contains egg. Omwamba and Mahungu (2014) reported lysine loss mostly through Maillard reaction, also, reported a low percentage (9.1 %) loss after extrusion in cereals**.** Lysine is the most limiting amino acid in cereals (Omwamba and Mahungu, 2014). R1, which is 100 % whole wheat flour contains the least quantity of lysine compared with other extrudates. There was no significant difference ($p>0.05$) between the quantity of lysine in R1, R2, R6 and R7 before and after storage, R3 was comparable whereas, R4 and R5 were significantly different. There was increase in R1 and R4 at the end of storage but there was decrease in R2, R3, R5, R6 and R7.

Threonine: Threonine maintains the mucosa of the intestine and is useful in the synthesis of mucin which serves as covering to the intestinal mucosa (Ahmad *et al*., 2020). The quantity of threonine before and after storage in the samples were either significantly different ($p<0.05$) (R4. R5, R7) or comparable (R1, R2, R3, R5). The quantity of threonine decreased in all the samples except in samples R4, R5, and R7.

Isoleucine: Isoleucine helps in regulation of some hormones in the body by stimulating its release such as insulin and somatotropin (Gutiérrez-Gamboa *et al*., 2019). The quantity of isoleucine before and after storage in the samples were either significantly different ($p<0.05$) (R2, R3, R4. R5, R7) or comparable (R1) or not significantly different (R6). The quantity of isoleucine at the end of storage period decreased in all the samples except in sample R4.

Alanine: The quantity of alanine before and after storage in the samples were comparable in R1 and R5, significantly different ($p<0.05$) in R2, R3, R4 and R6 but not significantly different in R7. The quantity of alanine at the end of storage period decreased in R3, R5, R6 and R7 but increased in R1, R2 and R4.

Glycine: The quantity of glycine before and after storage in the samples were comparable in R1 and R3 but significantly different ($p<0.05$) in R2, R4, R5, R6 and R7. The quantity of glycine at the end of storage period decreased in R1, R2, R3, R5, R6 and R7 but increased in R4.

Phenylalanine: The quantity of phenylalanine before and after storage in the samples were either significantly different ($p<0.05$) (R2, R3, R4, R6, R7) or not significantly different ($p>0.05$) (R1, R5). The quantity of phenylalanine at the end of storage period decreased in R2, R3, R4, R6 and R7 but increased in R1 and R5.

Serine: The quantity of serine before and after storage in the samples were either comparable (R1) or significantly different ($p<0.05$) (R2, R3, R4, R5, R6, R7). The quantity of serine at the end of storage period decreased in R1, R2, R3, R4, R6 and R7 but increased in R5.

Valine: The quantity of valine before and after storage in the samples were either comparable (R1 and R5) or significantly different ($p<0.05$) (R2, R3, R4, R6, R7). The quantity of valine at the end of storage period decreased in R2, R3, R5 and R7 but increased in R4 and R6.

Arginine: The quantity of arginine before and after storage in the samples were comparable in R5, significantly different ($p<0.05$) in R2, R3, R4, R6 and R7 but not significantly different in R1. The quantity of arginine at the end of storage period decreased in R1, R2, R3, R6 and R7 but increased in R4 and R5.

Table 2: Effect of extrusion and storage on amino acid profile of samples (g/100 g)

Amino acid	Time	R1	R2	R3	R4	R5	R6	R7	Egg white	Egg yolk	Whole wheat flour
Tryptophan	Start	1.21^{a} ±0.01	1.32^{b} ±0.01	1.33^{a} ±0.02	1.18^{a} ±0.04	1.21^{a}± 0.01	1.26^{a} ±0.0 0	1.27^{b} ±0.0 5	1.76	1.84	1.11
	End	1.24^{a} ±0.04	0.94^{a} ±0.00	1.21^{a} ±0.00	1.40^{b} ±0.04	1.26^{a}± 0.00	1.21^{a} ±0.0 0	1.05^{a} ±0.0 7			
Cystine	Start	1.48^{b} ±0.04	2.05^{de} ±0.07	2.22^{cd} ±0.03	1.36^{b} ±0.06	1.54^{ab} ±0.04	1.62^{b} ±0.0 1	1.86^{d} ±0.0 6	2.59	2.59	1.23
	End	1.45^{b} ±0.00	1.76^{cd} ±0.09	1.94^{c} ±0.00	2.36^{e} ±0.08	2.00^{b} ±0.08	1.57^{b} ±0.0 0	1.69^{c} ±0.0 0			
Methionine	Start	1.76^{c} ±0.07	2.27^{e} ±0.04	2.29^{d} ±0.04	1.81^{c} ±0.01	1.80^{b} ±0.14	2.13^{c} ±0.0 4	2.22^{f} ±0.0 2	3.59	3.07	1.63
	End	1.79^{c} ±0.04	1.58^{bc} ±0.04	2.08^{bc} ±0.00	2.22^{d} ±0.04	2.02^{b} ±0.02	1.76^{b} ±0.0 0	2.00^{e} ±0.1 1			
Histidine	Start	2.30^{d} ±0.00	3.95^{ghi}±0.0 7	4.21^{g} ±0.13	2.27^{de} ±0.04	3.51^{c}± 0.01	3.54^{d} ±0.0 4	3.80^{h} ±0.0 0	2.36	2.47	2.22
	End	2.33^{d} ±0.04	3.61^{fg} ±0.04	4.00^{f} ±0.13	3.87^{h} ±0.05	3.97^{cd} ±0.09	2.30^{c} ±0.0 0	3.65^{g} ±0.0 9			
Tyrosine	Start	3.40^{e} ±0.00	3.91^{ghi}±0.1 6	4.29^{g} ±0.02	3.31^{f} ±0.01	3.57^{c}± 0.06	3.64^{d} ±0.0 4	3.90^{hi}±0.1 4	4.39	3.99	3.06
	End	3.40^{ef} ±0.00	3.78^{fg} ±0.00	3.96^{f} ±0.00	4.05^{ij} ±0.12	4.22^{de} ±0.12	3.44^{d} ±0.0 0	3.53^{g} ±0.1 2			
Lysine	Start	3.43^{ef} ±0.01	3.98^{ghi}±0.0 8	4.12^{fg} ±0.13	3.37^{f} ±0.06	3.51^{c}± 0.01	3.56^{d} ±0.0 6	3.81^{h} ±0.0 1	6.20	7.29	3.33
	End	3.44^{ef} ±0.02	3.82^{gh} ±0.00	3.98^{f} ±0.08	3.92^{hi} ±0.00	4.45^{ef} ±0.00	3.50^{d} ±0.0 0	3.79^{h} ±0.0 4			
Threonine	Start	3.61^{fg} ±0.08	4.56^{jk} ±0.08	4.77^{i} ±0.09	3.33^{f} ±0.04	3.96^{cd} ±0.07	4.20^{e} ±0.0 6	4.42^{j} ±0.0 3	4.35	4.80	3.11
	End	3.53^{efg} ±0.04	4.25^{ij} ±0.04	4.66^{hi} ±0.00	4.80^{o} ±0.04	4.53^{def} ±0.04	3.61^{d} ±0.0 8	4.86^{kl}±0.0 4			
Isoleucine	Start	3.64^{g} ±0.05	4.19^{hi} ±0.04	4.51^{h} ±0.01	3.55^{g} ±0.07	3.64^{c}± 0.04	3.72^{d} ±0.0 1	4.01^{i} ±0.0 1	5.26	5.50	3.50
	End	3.60^{fg}	3.44^{f}	4.13^{fg}	4.49^{m}	4.33^{de}	3.70^{d}	3.83^{h}			

		±0.00	±0.05	±0.09	±0.05	±0.09	±0.04	±0.04			
Alanine	Start	4.31^{h} ±0.01	4.66^{kl} ±0.06	5.06^{j} ±0.08	4.09^{jk} ±0.04	4.56^{ef} ±0.06	4.52^{fg}±0.02	4.56^{j} ±0.06	5.50	4.66	3.76
	End	4.33^{hi} ±0.11	3.94^{ghi}±0.00	4.7^{i}± 0.00	4.44^{m} ±0.05	4.51^{def} ±0.06	4.29^{ef}±0.05	4.44^{j} ±0.05			
Glycine	Start	4.54^{j} ±0.09	5.26^{n} ±0.08	5.22^{jk} ±0.12	4.22^{kl} ±0.06	4.56^{ef} ±0.07	4.85^{h} ±0.07	4.98^{lm}±0.05	3.33	3.27	3.99
	End	4.49^{ij} ±0.10	4.25^{ij} ±0.10	5.09^{j} ±0.13	5.37^{p} ±0.06	5.49^{ghi} ±0.04	4.49^{fg}±0.04	4.75^{k} ±0.07			
Phenylalanine	Start	4.52^{j} ±0.13	5.30^{n} ±0.00	5.48^{l} ±0.03	4.51^{m} ±0.01	4.63^{ef} ±0.03	4.87^{h} ±0.05	5.06^{m}±0.06	5.37	4.19	4.21
	End	4.57^{j} ±0.06	3.99^{ghi}±0.88	3.28^{e} ±0.13	4.26^{l} ±0.00	4.66^{ef} ±0.06	4.61^{gh}±0.00	4.79^{k} ±0.00			
Serine	Start	4.79^{kl} ±0.02	4.97^{lmn}±0.09	5.27^{k} ±0.04	4.53^{m} ±0.04	4.60^{ef} ±0.08	4.81^{h} ±0.07	4.98^{lm}±0.04	6.53	7.77	3.31
	End	4.65^{jk} ±0.08	4.78^{klm}±0.04	5.11^{j} ±0.04	5.30^{p} ±0.15	5.22^{gh} ±0.04	4.23^{e} ±0.67	4.73^{k} ±0.11			
Valine	Start	4.93^{lm} ±0.11	5.80^{o} ±0.00	6.07^{n} ±0.10	4.66^{n} ±0.06	5.36^{gh} ±0.06	5.41^{i} ±0.01	5.66^{o} ±0.06	6.34	5.91	4.42
	End	4.97^{m} ±0.08	5.09^{mn}±0.08	5.79^{m} ±0.00	5.32^{p} ±0.00	4.97^{fg} ±0.08	4.88^{h} ±0.04	5.21^{n} ±0.16			
Arginine	Start	5.45^{n} ±0.07	6.44^{q} ±0.11	6.69^{p} ±0.04	5.37^{p} ±0.06	5.63^{hi} ±0.10	5.81^{j} ±0.01	6.06^{p} ±0.06	5.13	6.45	5.02
	End	5.42^{n} ±0.12	6.28^{p} ±0.12	6.41^{o} ±0.06	5.77^{q} ±0.12	6.02^{i}± 0.00	5.50^{i} ±0.00	5.72^{o} ±0.06			
Aspartic acid	Start	6.29^{o} ±0.04	7.47^{t} ±0.04	7.73^{t} ±0.04	6.08^{r} ±0.04	6.67^{j}± 0.04	7.06^{l} ±0.06	7.29^{r} ±0.04	10.14	9.68	5.51
	End	6.48^{p} ±0.04	6.92^{rs} ±0.13	7.29^{r} ±0.04	6.98^{t} ±0.04	7.54^{k} ±0.05	6.61^{k} ±0.05	7.07^{q} ±0.08			
Leucine	Start	6.59^{p} ±0.02	7.16st ±0.06	7.53^{s} ±0.04	6.46^{s} ±0.08	6.71^{j}± 0.00	6.79^{k} ±0.02	7.00^{q} ±0.00	7.96	8.20	6.25
	End	6.60^{p} ±0.01	6.71^{qr} ±0.00	7.03^{q} ±0.04	7.97^{u} ±0.04	6.62^{j}± 1.46	6.62^{k} ±0.04	6.98^{q} ±0.04			
Proline	Start	8.94^{q} ±0.11	9.23^{u} ±0.12	9.48^{v} ±0.10	8.72^{w} ±0.02	9.28^{m} ±0.08	9.25^{n} ±0.07	8.98^{s} ±0.03	3.31	3.83	8.45
	End	8.93^{q}	9.09^{u}	9.04^{u}	8.58^{v}	8.23^{l}±	8.78	8.93^{s}			

		±0.00	±0.07	±0.15	±0.07	0.15	^{m}±0.07	±0.00			
Glutamic acid	Start	26.58^{s} ±0.11	26.84^{w}±0.16	27.48^{x}±0.11	26.58^{y}±0.11	26.98^{o} ±0.04	27.34^{p}±0.09	26.88^{u}±0.11	13.52	13.15	25.45
	End	26.12^{r} ±0.32	24.68^{v}±0.21	26.27^{w}±0.11	25.06^{x}±0.11	24.15^{n} ±0.11	25.97^{o}±0.11	26.19^{t}±0.00			

Results are means of three replicates. Means with the same superscript in the same row are significantly different ($p>0.05$). R1 - 100:0 (100 % Whole wheat flour), R2 - 85:15 (85 % Whole wheat flour and 15 % raw whole egg), R3 - 80:20 (80 % Whole wheat flour and 20 % raw whole egg), R4 - 85:15 (85 % Whole wheat flour and 15 % raw egg yolk), R5 - 80:20 (80 % Whole wheat flour and 20 % raw egg yolk), R6 - 85:15 (85 % Whole wheat flour and 15 % raw egg white), R7 - 80:20 (80 % Whole wheat flour and 20 % raw egg white).

Aspartic acid: The quantity of aspartic acid before and after storage in the samples were significantly different ($p<0.05$) in all the samples (R1, R2, R3, R4, R5, R6, R7). The quantity of aspartic acid at the end of storage period decreased in R2, R3, R6 and R7 but increased in R1, R4 and R5.

Leucine: The quantity of leucine before and after storage in the samples were either significantly different ($p<0.05$) (R2, R3, R4) or not significantly different (R1, R5, R6, R7). The quantity of leucine at the end of storage period decreased in R2, R3, R5, R6 and R7 but increased in R1 and R4.

Proline: The quantity of proline before and after storage in the samples were either significantly different ($p<0.05$) (R3, R4, R5, R6) or not significantly different (R1, R2, R7). The quantity of proline at the end of storage period decreased in all the samples (R1, R2, R3, R4, R5, R6 and R7).

Glutamic acid: The quantity of glutamic acid before and after storage in the samples were significantly different ($p<0.05$) in all the samples (R1, R2, R3, R4, R5, R6, R7). The quantity of glutamic acid at the end of storage period decreased in all the samples (R1, R2, R3, R4, R5, R6 and R7).

Protein digestibility

Table 3 shows that the protein digestibility of the whole wheat flour (75.9 ± 0.09 %) was higher than those of raw egg yolk (61.99 ± 2.84 %) and raw egg white (63.82 ± 3.95 %) Chinma *et al*. (2016) reported a high protein digestibility value (72.30 %) for 100 % wheat bread which is almost the same as that reported for the whole wheat flour. Also, Evenepoel *et al*. (1998) observed that the digestibility of raw egg is not high (about 50 %) which is close to the value reported in Table 3 but that the digestibility could reach up to 91 % on cooking. On extrusion, the digestibility of the whole wheat flour (R1) reduced to 69.82 % suggesting that extrusion cooking reduced the digestibility of foods. This is in agreement with Ajita and Jha (2017) who reported that non-extruded foods had higher digestibility value compared to their extruded counterparts. The reason for this reduction could be attributed to heat-induced complexing reactions and cross-linkages that could lead to formation of less digestible end products.

Table 3: Protein digestibility of samples

Samples	Protein digestibility
Whole wheat flour	75.97^{h} ± 0.09
Egg white	63.82^{cd} ± 0.95
Egg yolk	61.99^{c} ± 0.84
R1	69.82^{fg} ± 0.40
R2	67.70^{ef} ± 0.82
R3	72.88^{gh} ± 0.11
R4	51.85^{a} ± 0.00
R5	66.14^{def} ± 0.87
R6	55.86^{b} ± 0.00

R7	$64.68^{cde} \pm 0.62$

Results are means of three replicates. Means with the same superscript in the column have no significant difference ($p>0.05$). R1 - 100:0 (100 % Whole wheat flour), R2 - 85:15 (85 % Whole wheat flour and 15 % raw whole egg), R3 - 80:20 (80 % Whole wheat flour and 20 % raw whole egg), R4 - 85:15 (85 % Whole wheat flour and 15 % raw egg yolk), R5 - 80:20 (80 % Whole wheat flour and 20 % raw egg yolk), R6 - 85:15 (85 % Whole wheat flour and 15 % raw egg white), R7 - 80:20 (80 % Whole wheat flour and 20 % raw egg white).

The lower values of the protein digestibility of the egg-based wheat snacks, compared to the extruded whole wheat snacks could be attributed partly to the protein complexing/cross linkage reactions and partly to dilution from the egg products. The egg on cooking (including extrusion) was expected to contribute to higher digestibility of the snacks while complexing/cross linkage reactions would lead to reduced digestibility of the products. Thus the digestibility of the products would depend on the relative rates of these two processes. It is obvious from the result in Table 3, that complexing/cross linkage reactions were occurring more. This would account for why the products containing egg yolk only (R4 and R5) had the lowest values because egg yolk contains more reactive substances (proteins and lipids) that could be involved in complexing and cross linkage reactions with the carbohydrates (which is abundant in wheat). The above notwithstanding, it is also observed from Table 3 that all the digestibility results were above 50 %. This may mean that the nutritional value of the extruded egg-based wheat snacks is still high and would be beneficial to consumers.

Protein solubility

The protein solubility of the egg-based wheat snack products is shown in Figure 1. The protein solubility of the products containing wheat only (R1) was 71 % and significantly ($P< 0.05$) lower than those containing egg (R2 - R7) which were higher (85.5 % to 88.5 %). The solubility of products containing egg could be attributed to the presence of egg which is known to be highly soluble. On the average, products containing whole egg were the most soluble. The order was whole egg (88 %) > egg white (87 %) > egg yolk (86.5 %). Due to mass action, those containing larger amount of egg (20 %) were slightly more soluble (87.17 %) compared to those containing 20 % (87 %). During storage (at ambient room temperature and 37 ^{o}C) the solubility of all products remained high (69.5 - 88.5 %), although, with slight reduction, as shown in Figure 1. This means that the nutritional value of the products would remain high during storage, at least for 10 weeks. The slight decrease in protein solubility has been attributed to protein cross linkage and complexing reactions, resulting from protein-protein, protein-carbohydrate and protein-lipid interactions (Obanu *et al*., 1975; Okonkwo *et al*., 1992). Those products containing egg white (R6 and R7), reduced more than others, suggesting that egg white contained more reactive proteins than egg yolk. Also, those stored at 37 ^{o}C reduced more in solubility compared to those stored at ambient room temperatures due to greater reaction rate at 37 ^{o}C. The slight increase in protein solubility from the 8th week of storage may suggest that cross linkage may have started to breakdown and this would lead to production of more brittle products. This later increase is more pronounced at 37 ^{o}C than at ambient room storage.

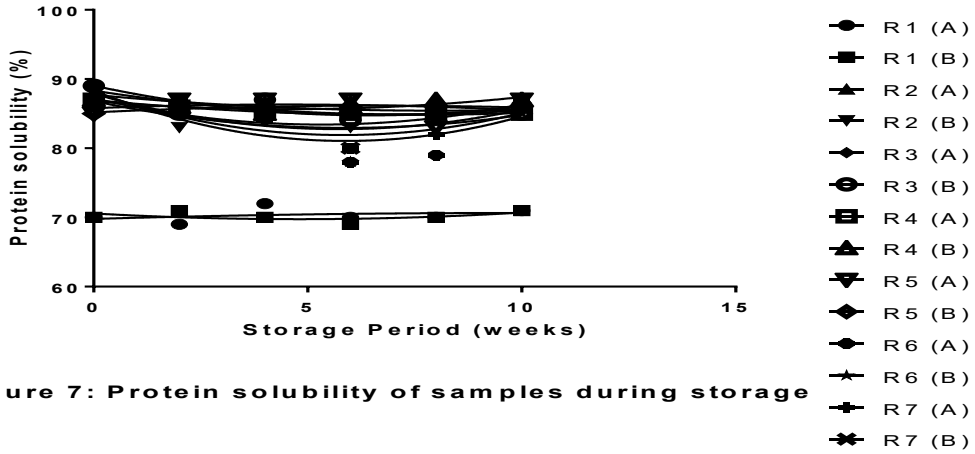

Figure 7: Protein solubility of samples during storage

Figure 1: Protein solubility of samples during storage

Source: (Nwadi and Okonkwo, 2021)

CONCLUSION

The use of chicken egg improved the protein content of the extruded snack which had acceptable protein solubility and digestibility values. The extruded snack proved to be nutritious based on the amino acid profile. The extruded snack can therefore be said to be of high quality and safe for consumption across all age ranges.

RECOMMENDATIONS

Further investigations on storage period using adequate packaging material so as to determine the shelf-life of the extruded snacks. Use of raw egg powder so as to consider a possibility of increasing quantity of egg in the extruded snacks for a product with a higher protein content so as to have a cheaper alternative of protein source in low-income communities.

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