

RESEARCH ARTICLE

Applications of hurdles in the processing of snail (*Achachatina marginata*) meat

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ABSTRACT

Inadequate protein consumption is one of the major challenges in most developing countries especially in Africa both to urban and rural dwellers. Snail meat was processed into dried snails using salt, sugar and citric acid at different concentrations of 0.1% (B), 0.2% (C), 0.3% (D), and 0.5% (E). The control sample (Sample A) was without treatment. Analyses of minerals, vitamins, proximate and anti-nutritional properties were carried out using standard methods. All results showed statistical difference (P<0.05%). The minerals sodium, iron, potassium, calcium, phosphorus, zinc and magnesium ranged from 71.350 to 99.420 mg/100g, 12.335 to 15.715 mg/100 g, 853.235 to 901.040 mg/100 g, 86.560 to 97.570 mg/100 g, 498.560 to 701.215 mg/100 g, and 52.450 to 76.250 mg/100 g and 163.230 to 128.00 mg/100 g, 0.165 to 0.300 mg/100 g, 0.054 to 0.073 mg/100 g and 0.955 to 0.715 mg/100 g respectively. The vitamin A, B1, B2, B3, B6 and E ranged from 6.595 to 9.685 μ g/100 g, 0.160 to 0.280 mg/100 g, 0.330 to 0.390 mg/100 g, 0.165 to 0.300 mg/100 g, 0.054 to 0.073 mg/100 g and 0.955 to 0.715 mg/100 g respectively. The proximate composition of snail meat: the crude protein, crude fat, ash, moisture and carbohydrate content of the samples ranged from 56.450 to 63.515%, 1.550 to 2.110%, 7.610 to 8.850%, 9.000 to 7.200% and 25.280 to 18.325% respectively. Crude fibre was not detected in the samples. The pH value of the snail meat ranged from 6.25 to 4.60. Anti-nutrients such as oxalate, tannin and cyanide were detected, oxalate ranged from 0.00-0.01%, tannin 0.00 - 0.01% and cyanide 0.00 - 0.01%. Phytate was not detected. This study shows that snails' meat can be treated with salt, sugar and citric acid and the samples treated with 0.3 and 0.5% are considered to be richer in nutrients.

Keywords: Storage fungi, Aspergillus flavus, essential oils, growth inhibition

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INTRODUCTION

Inadequate protein consumption is one of the major challenges in most developing countries especially in Africa both to urban and rural dwellers. And as such, human beings have been in search for alternative sources of protein in the wild to meet up with the body requirements (Yusuf and Oseni, 2004). This has made many to go into the consumption of other forms of animal protein from mini-livestock such as snail meat, rodents and other tiny livestock in the wild so as to improve their living conditions by acting as a valuable source of protein to supplement diets as well as generate income (Anon, 1992; Ezeama et al., 2007). Over the years, Snails have been used as human food with high nutritional value (Tremlova, 2001). They have been recorded to be high in protein, low in fat and a source of iron, magnesium, calcium and zinc (Ademolu et al., 2004; Adeola et al., 2010; Babalola and Akinsoyinu, 2009; Cobbinah et al., 2008). Snails also have been acknowledged to be richer in mineral than in other meat samples like beef, broiler, goat meat, mutton and pork. As compared to products from plant origins, snails have been found to contain anti-nutrients at levels not harmful to the human body (Tettey et al., 1997).

Snail just like other meat is susceptible to spoilage which sets in after 48 h of harvest (collection of snails). Shrinking speeds up the spoilage rate in snails as they cannot be kept in acceptable conditions after approximately 12 h. Hurdle technology is based on the combination of several factors/methods (hurdles) to achieve microbial stability and food safety. Application of hurdle technology illustrates the fact that complex interactions of temperature, water activity, pH, redox potential and preservatives are significant for microbial stability of foods (Leistner, 2000: 2002; Leistner and Gorris, 1995). The application of hurdle (also known as combined methods, combined processes, combination preservation, combination techniques or barrier technology helps to secure the microbial stability and safety as well as the sensory, nutritive and economic properties of foods.

Meat industries have evolved different processing methods and techniques which have the primary aim of improving the keeping quality of snails' meat as well as increasing palatability and acceptability of the meat product. In view of this, research has been in the area of using techniques that can address the problem of nutrient retention and increasing palatability and keeping quality of snail meat especially its protein. This present study looked at the application of different hurdles (salt, sugar and citric acid) on the processing of snails meat and assessing the effect of this hurdles on the proximate, mineral, vitamin and anti-nutritional content of snails' meat.

MATERIAL AND METHODS

Procurement of raw materials

Snails, salt, sugar, ginger, pepper, seasoning cubes were purchased from the modern market and citric acid was purchased from Emole Nigeria LTD, all found in Makurdi, Benue state in Nigeria. Processed materials were taken to the Chemistry department, Benue State University laboratory for processing.

Preparations of snail samples

A total of 150 giant snails were sorted and purged, after purging; the snails were killed, washed and separated from the shell to give the foot (edible portion). It was then allowed to drain and weighed on a scale balance; the mass was recorded to be 693 g. The foot was then washed with clean water and separated into five samples with each sample containing 30 snails and each sample of 30 snails recorded a weight of 301 g. Each sample was then washed with 5% salt for 30 min to remove slime and blood, then rinsed with distilled water, drained and weighed; the weight was recorded to be 258 g. The water was changed several times until it was clean.

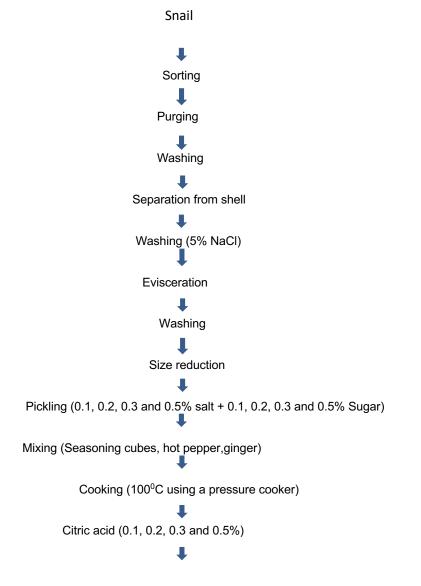
Method

The snail samples were eviscerated and soaked in pickling solution of 0.1%, 0.2%, 0.3%, 0.5% salt and 0.1%, 0.2%, 0.3%, 0.5% sugar respectively and allowed to equilibrate in the solution for 16 h at ambient conditions after which the solutions were

drained off. The snails were then mixed with spices (seasoning cube, hot pepper, and ginger) and cooked (using a pressure cooker at 100°C). The cooked snails were then treated in different concentrations of citric acid (0.1%, 0.2%, 0.3% and 0.5%) respectively (Table 1). Sample A which was the control sample was without salt, sugar and citric acid solution. The samples were all dried at a temperature of 60°C in an enclosed oven and then packaged.

Treatment/Sample	Salt (%)	Sugar (%)	Citric Acid (%)
Α	0	0	0
В	0.1	0.1	0.1
с	0.2	0.2	0.2
D	0.3	0.3	0.3
E	0.5	0.5	0.5

Table 1: composition of salt, sugar and citric acid blends



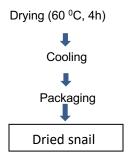


Fig 1: Flow chart of snail meat processing



Chemical analysis

The minerals content of the products were determined by the Otte (2012) method. Briefly, 2 g of the oven dried snail meat sample was ashed at 600°C in a muffle furnace and the resultant ash was transferred into 250 ml glass beaker and 120 ml conc. HN0₃ and 10 ml H₂O₂ added. The mixture was heated at 90°C for 1 h, cooled and filtered using glass wool. The filtrate was transferred into a 250 ml volumetric flask and made up to the mark with deionized water. After gentle shaking to mix, 2 ml was pipette into 250 ml flask and diluted to the mark with deionized water. Stock solution of 1000 mg/kg of Mg, Ca, K and Fe were prepared using deionized water. From stock solution working standard solution of 100 mg/kg was prepared by dilution with deionized water. Dilution comprising 0.4, 1.0, 1.5 and 2.0 mg/kg of each element was made with deionized water and together with the test sample was analysed using an atomic absorption spectrophotometer.

Concentrations of the elements in the test samples

$$Mineral\ content\ \left(\frac{mg}{kg}\right) = \frac{(At\ x\ Cs)}{As} \tag{1}$$

Where,

Cs = concentration of standard element

At = absorbance of test element

As = absorbance of the standard element

Vitamin A, E, B₁, B₃, B₆, B₁₂ were evaluated using HPLC (Model: BLC-10/11, buck scientific, USA) techniques as described by Otte (2012). For each snail meat, 3 g of ground sample was mixed with 5 ml of Hexane and 200 ml of HPLC distilled water. The mixture was homogenized (12000 r.p.m), centrifuged (3500 x g) for 30 minutes followed by sequential filtration through Whatman filter paper and 0.45 micrometre membrane. Then 15 µl of supernatant was injected into the HPLC equipped with a UV detector set at 254 nm. The peaks of the vitamins in the samples were calculated in relation to peaks of standard vitamins.

The dried samples were analysed for the presence of anti-nutrients using different method as follows; tannins was determined using burn method AOAC (2010). 5 g of the dried sample was treated with 50 mL methanol and kept for 24 hours before filtration. 5 mL of freshly prepared vanalin hydrochloric acid was added and the solution was allowed to stand for 20 minutes for colour development. The absorbance was measured at 550 nm using spectronic 20 and the machine value is used in calculating the tannin content. For oxalate determination, dye method described by (Krishnaiah et al., 2009) was used. 2.5 g of the sample was extracted with 20 mL of 25% H₂SO₄ and dissolved in hot water before titrating with 0.05 N KMnO₄ to determine the concentration of oxalate. Hydrogen cyanide was determined by using 5 g of the sample and soak in distilled water for 4 hours for the liberation of cyanide. The liberated cyanide is steam-distilled into 5 ml of 2.5% w/v 4 ml of 6N NH₄OH and 5% w/v KI is added to the distillate portion before titration with 0.02N AgNO₃ to a faint but permanent turbidity (ml of 0.02 N AgNO₃ = 1.08 mg HCN).

Proximate and anti-nutritional content were analysed using standard method described by (Gandy and Garrow, 2007).

Statistical analysis

All analyses were performed in triplicates and the data was subjected to one way analysis of variance (ANOVA) using Statistical Package (SPSS) 20 software version. Duncan Multiple Range Test was used to detect significant differences ($\alpha = 0.05$).

RESULTS AND DISCUSSION

Mineral Content of Snail meat

The results gotten from the mineral analysis is shown on Table 2. The values of the mineral content showed significant (p<0.05) difference. There was increase in sodium, potassium, calcium, zinc, iron and phosphorus except for magnesium that decreased from 163.23 to 128.00 mg/100g. The sample with 0.5% salt, sugar and citric acid was found to be a richer source of sodium, iron, calcium, potassium, phosphorus and zinc as compared to the other samples. The increase in sodium from 71.350 mg - 99.420 mg/100g could be as a result of the sodium chloride used during preparation of the samples and also due to the sodium chloride used in the treatment. Sodium chloride is needed for maintaining extra cellular fluid volume because of its role in osmotic action and is also important for the excitability of muscle and nerve cells (Garrow et al., 1999). The iron content was found to be slightly higher than that revealed by (Engmann et al., 2013; Uboh et al., 2014). Seafood such as snails contains both heme and non-heme iron which may have been made available after processing thus increasing its iron content. Hence snail meat acts as an excellent source of iron and helps in combating diseases such as anaemia resulting from iron deficiency. The calcium content 19.00 mg reported by Fagbuoro et al. (2006) was lower than what was revealed in this study. Calcium is mostly present in bones and teeth just one percent is present in soft tissues and body fluid. Snail meat on the other hand is mostly composed of muscles and lack bones and this could be the reason for its low calcium content. The zinc content of the samples increased from 52.45 to 76.25 mg/ 100g. The result was slightly higher than what was reported by Engmann et al., (2013) with 39.0 mg that studied on proximate and mineral composition of snail and (10.88 -12.00 mg) also reported by Uboh et al., (2014). Zinc deficiency is considered to be among the ten largest contributing factors to the problem

of morbidity in developing countries with high mortality (WHO, 2002). The high zinc content of snail may account for use of snail meat in nourishing lactating mothers and also pregnant women. The magnesium content of the samples decreased as

the concentration of the treatment increase. However, the magnesium content was found to be higher than what was reported by (Uboh et al., 2014).

Sample Code	Na	Fe	Mg	к	Ca	Ρ	Zn
Α	71.35±0.01ª	12.34±0.01ª	163.23±0.01e	853.24±0.01ª	86.56±0.01ª	498.56±0.01ª	52.45±0.01ª
B (0.1%)	75.46±0.01 ^b	12.66±0.01 ^b	149.55±0.01 ^d	863.24±0.02 ^b	91.33±0.01 ^b	517.44±0.01 ^b	54.57±0.01 ^b
C (0.2%)	82.57±0.02°	13.66±0.01°	145.23±0.01°	880.06±0.01°	93.05±0.02°	644.33±0.01°	72.67±0.02 ^c
D (0.3%)	88.45±0.01 ^d	14.84±0.02 ^d	138.44±0.01 ^b	896.74±0.02 ^d	95.02±0.01 ^d	690.04±0.01 ^d	75.35±0.01 ^d
E (0.5%)	99.42±0.01°	15.72±0.01°	128.00±0.00ª	901.04±0.01°	97.57±0.01°	701.22±0.01°	76.25±0.00 ^e

Table 2: Effect of salt, sugar and citric acid on the mineral content (mg/100g) of snail meat

All data are means of triplicates expressed on dry weight. Different superscripts between columns depict significant difference (p≤0.05).

A= 0% treatment (control), B= 0.1% salt, sugar and citric acid, C= 0.2% salt, sugar and citric acid, D= 0.3% salt, sugar and citric acid, E= 0.5% salt, sugar and citric acid

Vitamin Content of Snail Meat

The result of the vitamin content is presented in Table 3 below. It was reported that the sample with 0.5% salt, sugar and citric acid concentration is an important source of Vitamin A, B₁, B₂, B₃ and B₆ as compare to other samples, but had the least amount of vitamin E. Statistically, there was a significant (P<0.05) difference in all the vitamins. Vitamin A content was recorded to increase from 6.60 to 9.69 ug/100g. The results obtained were slightly higher than the results reported by(Çağıltay, 2011). Preformed vitamin A is found almost exclusively in animal products, such as human milk, glandular meats, liver and egg yolk, whole milk etc. Snails just like other animal products are very rich in vitamin A. Vitamin A is an important vitamin for proper and clear vision thus snail meat could be consumed to improve on the vitamin A content of the body. This result obtain for thiamine (vitamin B₁)was in line with report by(Çağıltay, 2011) that worked on Amino acid, fatty acid, vitamin and mineral contents of the edible garden snail . It is reported that the human body requires 1.4 mg vitamin B₁(Belitz et al., 2001).

Thiamine plays a role as the coenzyme thiamine pyrophosphate (TPP) in the metabolism of carbohydrates and branchedchain amino acids. It was revealed from the result that the snail samples examined had an increase in riboflavin (vitamin B_2). However it was slightly higher than study reported by (Çağıltay, 2011) (0.065 mg), the slight difference could be attributed to the difference in snail species. Riboflavin is vital in protein metabolism in the body (WHO., 2003). The niacin and pyridoxine present in snail helps the body turn food into energy. Lack of vitamin B_6 leads to Dandruff-like eruptions, Pink eye and Epilepsy (UBI., 2016). There was a decrease in vitamin E from 0.96 to 0.72 mg/100g. However, it was in line with report by(Çağıltay, 2011). Vitamin E helps to enhance healing of wounds with much less scar tissues, assists in breaking down blood clots in the circulating system, and reduces the incidence of heart diseases or cancer.

Sample Code	A (µg/100g)	B1 (mg/100g)	B2 (mg/100g)	B3 (mg/100g)	B6 (mg/100g)	E (mg/100g)
Α	6.60±0.04ª	0.16±0.00 ^a	0.33±0.00ª	0.17±0.02ª	0.05±0.00 ^a	0.96±0.01 ^e
B (0.1%)	6.88±0.01 ^b	0.18±0.00 ^b	0.38±0.01 ^b	0.19±0.00 ^{ab}	0.06±0.00 ^b	0.88±0.01 ^d
C (0.2%)	7.65±0.01°	0.25±0.00 ^c	0.38±0.00 ^{bc}	0.21±0.00 ^b	0.06±0.00 ^b	0.81±0.01°
D (0.3%)	8.31±0.01 ^d	0.25±0.00 ^c	0.39±0.01 ^{bc}	0.28±0.01 ^c	0.07±0.00°	0.76±0.01 ^b
E (0.5%)	9.69±0.02 ^e	0.28±0.00 ^d	0.39±0.00°	0.30±0.00°	0.07 ± 0.00^{d}	0.72±0.01ª

Table 3: Effect of salt, sugar and citric acid on the vitamin content (mg/100g) of snail meat

All data are means of triplicates expressed on dry weight. Different superscripts between columns depict significant difference (p≤0.05).

A= 0% treatment (control), B= 0.1% salt, sugar and citric acid, C= 0.2% salt, sugar and citric acid, D= 0.3% salt, sugar and citric acid, E= 0.5% salt, sugar and citric acid.

Proximate Composition of Snail meat

The result gotten from the proximate analysis of snail meat revealed that; they was a significant difference (P<0.05) in protein, moisture, fat, fibre, pH, carbohydrate and energy content of the samples (Table 4). An increase in protein, fat, fibre and pH was observed while the carbohydrate, moisture and energy content of the samples were found to decrease. The increase in protein can be attributed to the fact that most animal products contain more proteins than plant products and snail meat is basically made up of muscles; and also due to the fact that living cells contain proteins that are indispensable. Proteins are made available due to the removal of water present in the snail meat using citric acid. Snails being a good source of animal protein can be made into powder and used to prepare nutritious foods for babies to prevent diseases such as Kwashiorkor, marasmic-kwashiokor and stunted growth that are common in the rural areas. Eke et al. (2013) reported an increase in protein for meat treated with hurdles (salt, citric acid, spices). The value of the crude protein was in line with results published by(Ademolu et al., 2004; Uboh et al., 2014). The result of the fat content in this study was within the range reported by (Ademolu et al., 2004). The lower fat content recorded maybe as a result of the purging process where the snails used some fat for energy to keep themselves alive. With the low level of fat content found in snail meat as compared to other animals and fatty foods, eating of snail meat helps to prevent the incidence of health problems such as hypertension and other cardiovascular diseases resulting from eating excessive fatty foods and inadequate exercise (Okonkwo and Anyaene, 2009). From this research, it was reported that crude fibre was not detected in any of the samples. This may be due to the fact that snail meat on like plant products is made basically of muscle and doesn't contain any fibre. The findings from this study agrees with the reports of (Adegoke et al., 2010; Ademolu et al., 2004; Eruvbetine, 2012; Obande, 2013). The ash content was found to range from 7.61 to 8.85%

The high values of the ash content maybe as a result of the location and food eaten by the snail. Increase in ash content indicates an increase in minerals and samples with high percentage of ash are a good source of mineral. Snails consume different food sources including eating the soil, and this gives snail meat a relatively high ash value. The findings in this work was in line with the values reported by(Eneji et al., 2008) that worked on nutrition assessment of some Nigerian land and water snail species, and nutritional evaluation of giant African land snail fed with different diets respectively. The result indicated a decrease in moisture content and pH. Moisture content is an important factor in controlling food deterioration. The decrease in moisture content is as a result of the treatments used. This is because salt, sugar and citric acid help to draw out moisture

from the meat thereby reducing free or unbound water and acting as a food preservative. This finding agrees with the range of results published by (Krishnaiah et al., 2009). The lower pH is as a result of the citric acid that was used which contributed in lowering the pH of the treated samples and this helps to lower microbial activities of the snail meat by reducing moisture content and creating an acidic environment where some spoilage microorganisms cannot strive, thus enhancing the keeping and storage life of the product. The values for carbohydrates were in line with results reported by (Engmann et al., 2013; Soniran et al., 2013). Snails are very slow animals as such they don't use much energy for their activities and locomotion. Snail meat could be concluded to be a poor source of carbohydrate and thus poor energy source.

Sample Code	PROTEIN	FAT	FIBRE	ASH	MOISTURE	СНО	рН
Α	56.45±0.00 ^a	1.55±0.00 ^a	0.00±0.00	7.61±0.01ª	9.11±0.01 ^e	25.28±0.04°	6.25±0.00 ^e
B (0.1%)	57.17±0.00 ^b	1.60±0.00 ^b	0.00±0.00	7.72±0.01 ^b	9.00±0.00 ^d	24.51±0.02 ^d	5.00±0.00 ^d
C (0.2%)	57.83±0.01°	1.66±0.00 ^c	0.00±0.00	8.20±0.00 ^c	8.91±0.01°	23.39±0.04°	4.80±0.00°
D (0.3%)	60.75±0.00 ^d	1.85±0.00 ^d	0.00±0.00	8.45±0.00 ^d	8.00±0.00 ^b	20.95±0.03 ^b	4.71±0.01 ^b
E (0.5%)	63.52±0.01°	2.11±0.01 ^e	0.00±0.00	8.85±0.00 ^e	7.20±0.00ª	18.33±0.02ª	4.60±0.00ª

Table 4: Effect of salt, sugar and citric acid on the proximate composition (%) and pH of snail meat.

All data are means of triplicates expressed on dry weight. Different superscripts between columns depict significant difference (p≤0.05).

A= 0% treatment (control), B= 0.1% salt, sugar and citric acid, C= 0.2% salt, sugar and citric acid, D= 0.3% salt, sugar and citric acid, E= 0.5% salt, sugar and citric acid.

Anti-nutritional content of snail meat

Table 5 reveals the anti-nutrient composition of snail meat. Oxalate, tannin and cyanide where detected in negligible (0.01%) amount while phytate on the other hand was not detected in any of the samples. Sample E treated with 0.5% salt, sugar and citric acid was reported to not contain any of the anti-nutrients (Okon et al., 2016) Oxalate values ranged from 0.01 to 0.00%. The presence of oxalate may be as a result of the food consumed by these animals since snail feed on plant materials. Study had shown that oxalates in large amounts bind with calcium forming calcium oxalate, which is insoluble and not absorbed by the body but harmless when present in small amounts(Chai and Liebman, 2004). No phytate was detected in any of the samples that were analysed. The absence of phytate in the sample might have been as a result of enzymatic reactions during processing. Phytates contains complex zinc, iron, magnesium and calcium ions in the digestive tract; they can cause mineral ions deficiency in animals and human(Bora, 2014). The mean value for tannins was recorded to range from 0.01 to 0.00%, the result from this study was reported to be lower than what was reported by Udoh et al. (1995) (112 mg/100g). Report by Onwuka (2005) showed that the presence of tannins in high proportion may cause browning or other pigmentation problems in both fresh food and processed products and also it can provoke an astringent reaction in the mouth and make the food unpalatable. The levels of anti-nutritional factors present in these samples were not at levels that are toxic to humans as such can be consumed by humans.

Samples	Oxalate	Phytate	Tannin	Cyanide
Α	0.01±0.00 ^b	0.00±0.00	0.01±0.01 ^b	0.01±0.00 ^b
В	0.01±0.00 ^b	0.00±0.00	0.01±0.01 ^b	0.01±0.00 ^b
С	0.01 ± 0.00^{b}	0.00±0.00	0.01±0.01 ^b	0.01±0.00 ^b
D	0.01±0.00 ^b	0.00±0.00	0.01±0.00 ^b	0.01±0.00 ^b
Е	0.00±0.01ª	0.00±0.00	0.00±0.00ª	0.00±0.00ª

Table 5: Effect of salt, sugar and citric acid on anti-nutritional content (%) in snail meat

All data are means of triplicates expressed on dry weight. Different superscripts between columns depict significant difference (p≤0.05).

A= 0% treatment (control), B= 0.1% salt, sugar and citric acid, C= 0.2% salt, sugar and citric acid, D= 0.3% salt, sugar and citric acid, E= 0.5% salt, sugar, citric acid.

CONCLUSION

The research presented shows that snails meat can be treated with salt, sugar and citric acid which can help improve the nutritional content of snail meat and also provide additional benefits to consumers. The mineral and vitamin content as well as the proximate composition and anti-nutritional content of snail meat showed variable results. The hurdles applied in treating the snail meat helped to improve its mineral and vitamin content. It also increased the protein content, ash content, crude fat content and pH of the snail meat. Crude fibre was not detected in any of the samples. The moisture content and carbohydrate content decreased significantly (P<0.05) as the concentration of the treatment decreased, which is necessary for storage. The snail meat treated with hurdles had an increase in minerals (sodium, zinc, potassium, iron, calcium, phosphorus) and vitamins (A, B1, B2, B3, B6) except for magnesium and vitamin E that showed a decrease. Anti-nutrients such as oxalate, tannin and hydrogen cyanide that were examined were found to be present at levels not toxic to man while phytate was not detected at all. This shows that snails' meat can be treated with salt, sugar and citric acid and the samples treated with 0.3 and 0.5% are considered to be richer in nutrients.

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