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Full Length Research Paper

Snails as meat source: Epidemiological and nutritional perspectives

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The microbiological (epidemiological), proximate and mineral element composition of the different species of snails (*Achatina fulica*, *Limcolaria sp*. and *Helix pomatia*) obtained from three different market in Uyo, Akwa Ibom state were investigated. Total bacterial count ranged from 1.00 - 1.50 x 10^8 cfu/g, Coliform count ranged from 1.68 - 2.20 x 10^7 cfu/g, *Salmonella/Shigella* count ranged from 5.2-8.2 x10^7 cfu/g, lactic acid bacteria count ranged from 1.03 - 1.30 x 10^8 cfu/g and fungi count ranged from 7.3 x 10^7 to 1.00 x 10^8 cfu/g. The organisms isolated were *Bacillus subtilis, Staphylococcus aureus, Lactobacillus spp., Escherichia coli, Micrococcus luteus and Bacillus cereus* while the fungal isolates were *Aspergillus terrus, Aspergillus fumigatus, Absidia* sp., *Fusarium oxysporum, Eurotium* sp. and *Aspergillus flavus.* The results showed that *Helix pomata* have the highest microbial load of 2.20 x 10^8 cfu/g. The proximate analysis showed that African giant snail (*Achatina fulica)* was nutritionally richer than the other snails. Mineral determination also showed that African giant snail had the highest amount of minerals. It was generally observed that snails though nutritionally rich are reservoirs of pathogenic microorganisms which are of public health importance.

Key words: Microbiological, proximate, mineral composition, coliform, *Helix pomata*

INTRODUCTION

Snail meat is a high quality food that is rich in protein (low in fats,) and a good source of iron, that is, 3.5 mg/100 g (USDA, 2006) . The comparative nutritive value of snail meat to some animal protein sources have been studied by some researchers. In an instance, the protein contents of 88.37, 82.42 and 92.75% were discovered in snail, pork and beef respectively (Imevbore and Ademosun, 1988).

With a fat content of only 1.3% and iron content of 12.2 mg/100 g in edible carcass, the nutritive value of snails is reported to be comparable to that of domestic livestock. It is estimated that snail is 15% protein, 2.4% fat and about 80% water (Saldanha et al., 2001). This makes snail healthy alternative food for people with high protein low fat diet requirements.

Besides, snail is high in health benefiting essential fatty acids such as linoleic acids and linolenic acids. A study on a snail species in Brazil estimated that 75% of the fat in snail is unsaturated fatty acids. That is 57% polyunsaturated fatty acids, 15.5% of monounsaturated fatty acids and 23.25% of saturated fatty acids (Su et al., 2004). However, it can be easily contaminated by pathogens and serve as vehicle of transferring infectious agents to consumers. Kirkan et al. (2006) reported the presence of *L. monocytogenes* in fresh snail sample which notably could have been contaminants from soil.

So, despite rich nutritional values of snail, the involvement of the mollusks in general in the transmission of infection mostly as secondary host for pathogens makes it necessary to study the microbiology of the resident snail without leaving their nutritive values behind. This project therefore aimed at isolation and identification of

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Table 1. Cultural and biochemical characteristics of bacteria isolated from snail.

Isolate	Gram reaction	Shape	Colour	Edge	Elevation	Catalase	Coagulase	Indole	M.R	V.p	Citrate	Spore	Glu.	Gal.	Fru	Mal	Suc	Raff	Prob. organism
1	+	Rod	Milky	Irregular	Raised	-	-	-	+	+	+	+	+	+	+	+	+	+	*Lactobacillus* sp.
2	+	Cocci	Yellow	Irregular	Raised	+	-	-	+	-	-	-	-		-	-	-	-	*M. luteus*
3	+	Cocci	Milky	Irregular	Raised	+	+	-	+	+	+	+	+	+	+	+	+	-	*S. aureus*
4	-	Rod	Yellow	Irregular	Raised	+	-	+	+	+	+	+	+	+		+	+	-	*E. coli*
5	+	Rod	Creamy	circular	Raised	+	-	-	-	+	+	+	+	+	+	-	+	+	*B. cereus*
6	+	Cocci	Milky	circular	Raised	-	-	-	-	+	+	-	+	+		+	+	+	*S. mutan*
7	+	Rod	Creamy	Irregular	Entire	+	-	+	+	+	+	+	+	+	+	+	+	-	*B. subtilis*

pathogenic microorganisms associated with snails and to study the proximate and mineral levels in the snail samples.

MATERIALS AND METHOD

Sample Collection

Snail samples (*Achatina fulica*l, *Limicolaria* sp. and land *Helix pomatia*) were purchased from Uyo main market, Itam market, Akpan Andem market in Uyo, Akwa Ibom State, Nigeria. The snail samples were collected in sterile plastic containers and taken to Microbiological laboratory for analysis.

Preparation and microbiological analysis of the samples

The snails were washed with running tap water to remove surface dirt. They were then washed with sterile distilled water and scrubbed with ethanol to remove external microorganism, the meat was aseptically extracted and homogenised.

One gram of the homogenate was used in carrying out ten-fold serial dilution up to the 10^{-6}. 1 ml of the dilluent was plated on Nutrient agar for total bacterial count, MacConkey agar for total Coliform count, *Salmonella/Shigella* agar for *Salmonella/Shigella* count, DeMan Rogosa and sharp agar was used for lactic acid bacterial count and Sabourand Dextrose agar for fungal count. The bacterial plates were incubated at 37^{O}C for 24 - 48 h, while fungal plates were incubated at room temperature (28 ± 2^{O}C) for 3 - 5 days. Colonies were selected randomly and were characterized using morphological and biochemical test such as gram stain, spore stain, motility, catalase, oxidize, coagulase, indole, MR-VP and Urease and sugar fermentation tests. Bacterial isolates were identified with reference to Cowan and Steel's Manual for the identification of Medical Bacteria (Cowan, 1985) and Beryey's Manual of Determinative Bacteriology (Hott et al., 1994). Fungal isolates were identified based on their morphological and cultural characteristics as recommended by Sampson et al. (1984).

Proximate and mineral analysis

Proximate composition was determined according to the method of A.O.A.C. (1998). This includes determination of Ash content, crude protein, Dry matter, moisture content and crude fiber. The mineral contents were also determined using Jenway Digital flame photometer (PFP 7 Model).

RESULTS

The microbial isolates associated with the snail samples were: *Bacillus subtilis, Staphylococcus aureus, Lactobacillus* spp., *Escherichia coli, Micrococcus luteus, Bacillus cereus* while the fungal isolates were *Aspergillus terrus, Aspergillus fumigates, Absidia* sp., *Fusarium oxysporum Eurotium* sp*. Aspergillus niger* and *Aspergillus flavus.*

A detailed result of the cultural and biochemical characteristics of the bacterial isolates is depicted in Table 1 while that of fungi and fungal frequency of occurrence are depicted on Tables 3 and 4.

Obviously, *Aspergillus niger* had highest number of occurrence (that is, with 6 times out of total of 15 times of fungal occurrence) . Its occurrence is equivalent to 40% frequency. *Absidia* sp. had 3 times (20%) frequency while *Aspergillus flavus* occurred twice (13.4%).

The microbial count of the snail samples are shown on Table 2. The microbial count for giant snail (*Achatina fulica)* ranges from 6.0 - 2.00 x 10^{7} cfu/g. Land snail (*Limicolaria sp)* ranged from 7.3 x 10^{4} to 1.68 x 10^{8} cfu/g while *Helix pomatia,* which had the highest microbial load, ranges from 5.2 x 10^{7} to 2.20 x 10^{8} cfu/g.

The proximate analysis and mineral composition of the snails are shown in Tables 5 and 6. The snail samples contained appreciable mineral compositions per 100 g.

They are rich in Na, K, Fe, Ca and P. Sample A (*A. fulica*) had highest mineral composition when compared to Sample B (*Helix pomatia*) and Sample C (*Limicolaria sp*) respectively (Table 5).

Table 2. Total bacterial count for snail samples.

Sample	Total bacteria count x 10^8 Cfu/g	Total coliform count x 10^7 cfu/g	Salmonella/ shigella count x 10^7 cfu/g	Fungal count x 10^7cfu/g	Lactic acid bacteria count x 10^8cfu/g
A	1.45	2.00	6.0	9.0	1.20
B	1.00	1.68	9.2	7.3	1.03
C	1.50	2.20	5.2	1.0	1.30

Key: A: *A. fulica* (African giant snail), B: *Limicolaria* sp (Land snail), C: *Helix pomatia* (Land snail).

Table 3. Cultural and morphological characteristics of the isolated fungi.

Isolate code	Colony colour	Type of soma	Nature of hyphae	Special vegetative structure	Asexual spore	Special reproductive structure	Conidial head	Vesicle shape	Probable organism
Lss 1	Gray green wrinkled spread colony	Filamentous	Septate	None	Ovate conidia	Ascoma	Radiating conidial head	subglobose	*Eurotium* sp
Lss2	Dense felt yellow green colony	Filamentous	Septate	Foot cell	Globose conidia	Philides borne directly on the vesicle scerotia	Radiate	subglobose	*Aspergillus flavus*
Lss3	Compact white or yellow basal dark colony	Filamentous	Septate	Foot cell	Globose conidia	Smooth balled erect conidiospore	Globose	subglobose	*Aspergillus niger*
Lss4	Brownish colony becoming darker with age	Filamentous	Septate	Foot cell	Globose conidia	Short conidiospore	Long columnoir	Hemispherical	*Aspergillus terrus*
Lss5	Floccose felty whitish colony with a purple tinge	Filamentous	Septate	None	Cell conidia, clamydospores	Sporodochia absent	None	None	*Fusarium oxysporum*
Lss6	Floccose light grayish, rapidly growing colony	Filamentous	Coenocytic	Stolons and rhizoids	Oblong sporangiospore	Zygospores sporangiospores	None	None	*Absidia sp*
AGS1	Compact white or yellow basal dark colony	Filamentous	Septate	Foot cell	Globose conidia	Smooth walled erect conidiosphore	Globose	Globose	*Aspergillus niger*
AGS2	Floccose light grayish, rapidly growing colony	Filamentous	Septate	Stolons and rhizoids	Oblong sporangiosphore	Zygospores sporangiospores	None	None	*Absidia* sp.
AGS3	Smoky or gray green colony	Filamentous	Septate	Foot cell	Globose conidia	Short conidiospore	Typically columnar	Dome shaped broadly clavate	*Aspergillus fumigatus*
Hp1	Compact white or yellow basal	Filamentous	Septate	Foot cell	Globose conidia	Smooth walled erect conidiosphore	Globose	Globose	*Aspergillus niger*

Table 3 contd.

Hp2	Dense felt yellow green colony	Filamentous	Septate	Foot cell	Globose conidia	Philides borne directly on the vesicle scerotia	Radiate	Subglobose	*Aspergillus flavus*
Hp3	Floccose light grayish, rapidly growing colony	Filamentous	Septate	Stolons and rhizoids	Oblong sporangiosphore	Zygospores sporangiospores	None	None	*Absidia* sp.

Key: AGS = *Achatina fulica* (African giant snail), Hp = *Helix pomatia* (land snail), Lss = *Limicolaria sp*. (land snail).

Table 4. Frequency of occurrence of fungi isolated from snails.

S/N	Fungi isolates	Frequency of occurrence (%)
1	*A. niger*	40.0
2	*A. terrus*	6.7
3	*F. oxysporum*	6.7
4	*Absidia* sp	20.0
5	*Eurotium* sp	6.7
6	*A. flavus*	13.4
7	*A. fumigates*	6.7
	Total	100

Table 5. Mineral composition of the snail samples.

Sample	Na_1 (mg/100 g)	Na_2 (mg/100 g)	K_1 (mg/ 100 g)	K_2 (mg/100 g)	Fe_1 (mg /100 g)	Fe_2 (mg/100 g)	Ca_1 (mg/100 g)	Ca_2 (mg/100 g)	P_1 (mg/100 g)	P_2 (mg/100 g)
A	28.97	28.94	64.15	64.15	1.51	1.49	186.35	186.32	62.73	62.76
B	24.78	24.83	56.72	56.76	1.36	1.36	179.65	179.64	58.34	58.32
C	13.63	13.65	31.59	31.57	1.17	1.14	13.51	132.48	31.17	31.14

Key: A: *A. fulica* (African giant snail), B: *Limicolaria* sp (Land snail), C: *Helix pomatia* (Land snail).

The crude protein, fat, Ash and moisture contents are higher in A than B and higher in B than C (Table 6).

DISCUSSION

Microbiological assessment of snail coupled with pathogenicity test in this study showed that the snail samples harbour quite a number of highly pathogenic bacteria of potential public health hazard to the dependants as protein source. Such hazards are more appropriate in some regions where the demand for the snail meat is high and the vendors in an attempt to meet with the demand usually undercook. Food- borne illnesses due to consumption of snails may occur when the mollusks that contain pathogenic microorganisms are consumed raw or improperly cooked. The health implications of these pathogenic microorganisms with high count in this study can not be overemphasized. *Escherichia coli* for instance can

Table 6. Proximate composition of the snail samples.

Sample	% Crude protein$_a$	% Crude protein$_b$	%Crude fat$_a$	% Crude fat$_b$	% Crude fiber$_a$	%Crude fiber$_b$	%Ash$_1$	%Ash$_2$	%Dry matter$_1$	% Dry matter$_2$
A	72.64	72.86	1.48	1.52	0.00	0.00	4.78	4.88	90.29	90.27
B	58.35	58.49	1.39	1.43	0.00	0.00	3.88	3.91	89.97	89.98
C	15.44	15.35	1.18	1.15	0.00	0.00	1.26	1.28	20.43	20.41

Key: A: *Achatina fulica* (African giant snail), B: *Limicolaria* sp (Land snail), C: *Helix pomatia* (Land snail).

induce gastroenteritis (Olowe et al., 2008). *Aspergillus* sp causes aspergillosis while *Staphylococcus aureus* isolates have been implicated in a number of clinical cases (Lowry, 1998; Komolafe and Adegoke, 2008; Adegoke and Komolafe, 2009) . Like in this study, Serrano et al. (2004) reported the presence of mesophilic aerobic bacteria, *Enterobacteriacea*e, *S. aureus* and coliforms among ready to eat snails. The coagulation of plasma in both the tube test and slide test by *S. aureus* coupled with their ß-haemolysis confirmed their pathogenicity. This organism (though easily killed by boiling heat) produces enterotoxin that is stable to heat at 100°C for 30 min and this toxin is known for food poisoning (Brook et al., 2004)

Snail is of epidemiological importance not only because it harbours pathogenic bacteria but also because it can serve as an intermediate host of the liver fluke; *Fasciola* spp. (Legaspi and Jovellanos, 1990) and Schistosoma (Oyawoye, 2008). There is therefore the need to set a thermal standard and time of cooking that would be sufficient for complete inactivation of pathogens while the nutritive contents would remain unaffected.

Milinsk et al. (2003) reported that the nutritional composition of snail meat meets the required balanced diet for an average individual. In this study high contents of 72.75 and 1.50% for the crude protein and fat were observed, (Table 6) beside having 28.96 mg/100 g, 64.15 mg/100 g, 1.50 mg/100 g, 186.34 mg/100 g and 62.75 mg/100 g of Na, K, Fe, Ca and P respectively for one of the three species of snails in question (Table 5). These contents met the USDA National Nutrient Standard Reference, release 19 (2006).

Meanwhile, of the three specie of snails studied [(*Achatina fulica*), (*Limcolaria* sp.) and *Helix pomatia*), their rating in term of nutritional composition could be *A. fulica* > *Limicolaria* sp, > *Helix pomatia*. The nutritional compositions of the *A. fulica* (African giant snail) which are multiples of those of the other two make the species the dependable source of nutrient for the obessed or hypertensive individuals more so since snails have low cholesterol (Su et al., 2004; USDA, 2006).

While, the trend of poverty worldwide seems to be higher unabaited and hunger with malnutrition is endemic in the subsaharan African countries, African giant land snail can be a good source of nutrient especially because it is easy to rear by the rich and the poor. It is imperative in the same premise to ensure adequate care in its preparation so that the supposed source of nutrients would not be source of epidemic threat.

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