Antinutrient Profile of Three Mushroom Varieties Consumed in Amaifeke, Orlu, Imo State

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Abstract

Background: The importance of eliminating or minimizing antinutrients from foods human consume cannot be overemphasized. This study evaluated the antinutrient profile of three varieties of mushroom consumed in Amaifeke, Orlu, Imo State were determined. The mushroom species include white button mushroom (*Agaricus bisporus*), oyster mushroom (*Pleurotus ostreatus*), Crimini mushroom (*Agaricus bisporus*).

Methods: The mushroom varieties were harvested and dried, after which, they were taken to the laboratory for chemical analysis. Standard assay methods were used to analyze for antinutrient composition.

Result: Six anti-nutrients: hydrogen cyanide, saponin, phytate, oxalate, trypsin inhibitor and haemogglutinin were analysed and their values ranged from 0.198 - 0.236 mg/g, 0.6656 - 1.001 mg/g, 0.7794 - 1.558 mg/100g, 0.236 - 0.510%, 1.857 - 3.476 TIµ/mg, 1.350 - 2.899 Hµ/mg respectively.

Conclusion: Based on the result of the study, it is important that these mushrooms be properly processed so that the nutrients in them will not be rendered inaccessible by the body due to the presence of these antinutrients. **Keywords:** *Antinutrient, Mushroom, Consumed, Amaifeke*

Introduction

Antinutrients are natural or synthetic compounds that interfere with the absorption of nutrients (1). Plant foods may contain significant amounts of toxic or antinutritional substances, legumes are particularly rich source of natural toxicants including protease inhibitors, amylase inhibitors, metal chelates, flatus factors, hemagglutinins, saponins, cyanogens, lathyrogens, tannins, allergens, acetylenic furan and isoflavonoidphytoalexins (2). Mushrooms are referred to as the fruiting bodies of macrofungi. They include both edible/medicinal and poisonous species. Edible mushrooms once called the "food of the gods" and still treated as a garnish or delicacy can be taken regularly as part of the human diet or be treated as healthy food or as functional food (3). The extractable products from medicinal mushrooms, designed to supplement the human diet not as regular food, but as the enhancement of health and fitness, can be classified into the category of dietary supplements/mushroom nutriceuticals (4). Dietary supplements are ingredients extracted from foods, herbs, mushrooms and other plants that are taken without further modification for their presumed health-enhancing benefits. Mushroom is not just attributed to have many health benefits but studies has also shown that they are rich source of micronutrients (3 - 7).

This study is focusing on evaluating the antinutritional factors present this these three species of mushroom. Hence this will try to detect how safe it is for consumption and which of them needs more attention in processing in order to access its rich micronutrient content.

Methodology

Materials

Three varieties of mushroom that were commonly available were freshly harvested from the farm in Amaifeke, Orlu Local Government Area of Imo State. The samples were white button mushroom, crimini and oyster mushroom with common names "ero osisi", "ero otukwuru" and "ero ntioke". Their botanical names being *Agaricus bisporus, Pleurotus ostreatus* and *Agaricus bisporus* respectively.

Source of Material

The samples used for this study were collected from logs of wood in the fields and also purchased from the old Orlu Main market.

Preparation of Materials

Non-edible portions and unwanted wastes or particles like soil, portion of compost where they were grown, spoiled portion of mushroom etc were separated and removed from the samples. The mushrooms were then sundried by constant exposure to sunlight for 3 days while turning the mushrooms to avoid fungal growth.

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Determination of Anti-Nutrients

They involve the phytate, hemagglutinin, oxalate, saponin, trypsin inhibitor and cyanide content of the mushroom samples.

Phytate

This was determined using the method of Matyka *et al* (8). About 5g of each sample was extracted with 2.0m HCl, 0.1m NaOH and 0.7m. NaCl were added and passed through a resin (200 - 400 mesh) to elude inorganic phosphorous and other interfering compound. Modified wade reagents 0.03% Fecl₃ 6H₂O and reading taken at 500mm.

Saponins

This was determined using the method of Price *et al* (9). About 0.1mg of the sample was boiled with 50ml distilled water for 15 minutes and filtered with Whatman No. 1, 5ml of the filtrate was pippetted into a test tube and 2ml of olive oil was added. The solution was shaken vigorously for 30 seconds and read 620m against a blank.

Calculation:

Saponin =

 $\frac{\text{Reading from curve x dilution factor x 100}}{\text{Weight of sample x 10}^{\text{b}}}$

Haemagglutinin

This was determined by the method of Matyka *et al* (8). Weight of 29mg of each sample was put into a test tube, with 10 - 20ml H₂O, and shaken vigorously and filtered. The extracts were stored in the refregirator at 4^oc. In another four test tubes, 4ml of centrifuged fresh blood of mice were added together with 4ml of normal saline solution, centrifuged at 780g for 60 minute. The supernatant was decanted and kept for heamoglobin activity estimation. About 0.1ml of the blood sample was pipetted into the sample test tube, 4ml of saline solution was added and two drops of extract was also added and kept for 16 hours at a temperature of 4^oc. The turbidity formed was read at 600nm. The turbidity of the extract plus 4ml of saline solution were equally measured at the same wavelength as the control.

Oxalate

The oxalate content of the sample were determined by the method described by Alabi *et al.* (10). Weigh 0.05g of the sample into a test tube, add 10ml of they acetate and place in a water bath and boil for 3 minutes. Filter, shake 3ml of the filtrate with 0.1ml of dilute ammonia in a test tube. The presence of a yellow colouration in the lower layer indicates the presence of oxalate.

Cyanide

The cyanide content of the samples were determined enzymatically using the method of D'Mello (11). Five grams (5g) of sample was introduced into 300ml volumetric flask. 160ml of 0.1M phosphoric acid and homogenized for 15 minutes at low speed and made up to the mark. The solution was centrifuged at 10,000 rpm (revolutions per minute) for 30 minutes. The supernatant was transferred into a screw cap bottle and stored at 4° C. 5ml aliquot of the extract was transferred into quick fit stoppered test tube containing 0.4ml of 0.2M phosphate buffer pH 7.0. 10ml of diluted linamarase enzyme was added. The tube was incubated at 30° C for 15 minutes and the reaction was stopped by addition of 0.2M NaOH (0.6ml). The absorbance of the solution was measured using suitance spectrophotometer at 450nm against blank.

Cyanide is calculated as							
Cyanide =	absorbance x dilution factor x 100						
-	Extinction coefficient						
Extinction coefficient ($\Sigma 1/450$)		=	2250				
Value expressed in (mg/100g)							

Trypsin Inhibitor

Trypsin inhibitor was determined by the modified method of Hamerstrand *et al.* (12). The values were estimated by single dilution of each sample extract that inhibit at least 40% but not more than 60% of the trypsin. The values were calculated from differential absorbance readings and reported in absolute unit as milligram of trypsin inhibitor per gram sample.

Table 1: The Anti-nutrient Content in White Button, Oyster and Crimini Mushroom Varieties

Samples	HCN mg/g	Saponin mg/g	Phytate mg/100g	Oxalate %	Trypsin inhibitor μ/mg	Hemagglutinin µ/mg
А	0.216 <u>+</u> 0.03	1.001 <u>+</u> 0.08	1.558 <u>+</u> 0.25	0.510 <u>+</u> 0.06	3.476 <u>+</u> 0.22	2.899 <u>+</u> 0.74
В	0.198 <u>+</u> 0.06	0.6656 <u>+</u> 0.18	1.169 <u>+</u> 0.34	0.236 <u>+</u> 0.05	1.857 <u>+</u> 0.68	1.675 <u>+</u> 0.74
С	0.236 <u>+</u> 0.04	0.790 <u>+</u> 0.14	0.7794 <u>+</u> 0.19	0.342 <u>+</u> 0.08	3.352 <u>+</u> 0.04	1.675 <u>+</u> 0.74

Mean \pm SD of three replications. Key: A = White button mushroom B = Oyster mushroom C = Crimini mushroom HCN = Hydrogen cyanide mg = Milligrams g = grams % = Percentage μ = microgram

Table 1 above showed that the content of hydrogen cyanide was highest in sample E (0.236 ± 0.04 mg/g) and lowest in sample B (0.198 ± 0.06 mg/g). Sample A had a value of 0.216 ± 0.03 mg/g. The saponin composition was highest in Sample A (1.001 ± 0.08 mg/g) followed by sample C (0.790 ± 0.14 mg/g) and sample B having the lowest content of 0.6656 ± 0.18 mg/g). The phytate content in the mushroom samples was seen in the descending order A>B>C with values 1.558 ± 0.25 mg/100g, 1.169 ± 0.34 mg/100g and 6.7794mg/100g respectively. Sample A had the highest content of $0.342 \pm 0.08\%$ and sample B had the lowest value of $0.236 \pm 0.05\%$. The content of trypsin inhibitor in the mushroom samples were seen in the descending order of A>C>B with values 3.476 ± 0.22 µ/mg, 3.352 ± 0.44 µ/mg and 1.857 ± 0.68 µ/mg respectively. Sample A had the highest content (1.675 ± 0.74 µ/mg) and sample B had the lowest value of 1.350 µ/mg.

Discussion

Cyanide is a normal constituent of the blood but it is usually at low concentrations of less than 12μ mol (13), at high concentration, it is a potent inhibitor of the respiratory chain. HCN is reported to be very toxic at low concentration to animals. HCN can precipitate dysfunction of the central nervous system, respiratory failure and cardiac arrest (14). In this study, hydrogen cyanide content of the mushrooms was found to be within the range of 0.198 - 0.236mg/g. These values are significantly higher thaan 5.8ppm (0.58mg/100g) reported by Chang (15) and also higher than 0.00 - 4.51mg/100g reported by Afiukwa et al., (16).

Phytates are inositol hexaphosphoric acids which form complexes with salts as calcium, zinc, magnesium, iron and render them unavailable for absorption and utilization in the body. Phytates can also affect digestibility by chelating with calcium or by binding with substances or proteolytic enzyme (17). The phytates content of the studied mushrooms are seen in the range of 0.7794 - 1.558 mg/100g. These values are over ten times lower than the safe limit (22.10mg/100g) (WHO, 2003). The results are also comparable to $2.43 \pm 0.09\%$ reported by Ogbe and Obeka (2013), and 0.5 - 0.12% reported by Afiukwe et al., (16).

Oxalate is an antinutritional factor mostly found in cocoyam, legumes and vegetables. Dietary oxalate has been known to complex with calcium, magnesium, and iron and inhibits their absorption by humans. Oxalates cause calcium deficiency both in man and in non-ruminants. At a high dose of 1g - 2g of body weight, it is toxic to the kidney and heart (20 - 21).

The oxalate content of the mushrooms was found to be within the range of 0.236 - 0.510%. These values are lower than the tolerable limit given by WHO (105.00mg/100g) and within the range of 0.412% reported by Harden (21). It is also within the range reported by Ogbe and Obeka (22).

Trypsin inhibitors are proteins that are found in some raw foods. They reduce the proteolytic activity of the trypsin enzyme. The presence of trypsin inhibitors in foods can lead to the formation of irreversible trypsin and trypsin inhibitor complexes. The trypsin inhibitor content of the mushrooms was found to be within the range of

 $1.857 - 3.476 \mu/mg$. This is within the range of $2.39\% \pm 0.11$ reported by Ogbe and Obaka (22).

Saponins possess a carbohydrate moiety to a steroid aglycone. They form a group of compounds, which on consumption cause deleterious effects such as haemolysis and permeabilization of the intestine . In this study, the saponin of the mushrooms was seen to be within the range of 0.6656 - 1.001 mg/g, comparable to 4.05% and 3.03% for the exotic and wild species of oyster mushroom by Kayode *et al.*, (23) and $1.26 \pm 0.06\%$ reported by Ogbe and Obeka (22). It is however higher than $0.11 \pm 0.56\%$ reported by Afiukwa *et al.*, (16) and lower than the maximum permissible limit of (48.05 mg/100g) set by WHO (24).

Hemagglutinins or lectins are widely distributed in the plant kingdom including legumes and pose high degree of specificity towards sugar component and thus have diagnostic importance. On ingestion, hemagglutinins exhibit unique property to bind carbohydrate-containing molecules and resist digestion. The hemagglutinin content of the mushrooms was found to be within the range of $1.350 - 2.899\mu/mg$

Conclusion

Based on the result of this study, in the consumption these mushrooms, proper processing should be carried out to reduce the values of these antinutrients in the mushroom to a tolerable level. However, these mushrooms could be subjected to further studies to evaluate other phytochemicals present in them and as well consider its beneficial use.

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