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Amino acids composition of pre-treated tree ear mushroom (*Auricularia auricula-Judae*)

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Abstract
This work is anchored on amino acids composition of pre-treated tree ear mushroom. Mushroom fruiting bodies of tree ear weighting 1000.32gm were brought from farmers. The sample were collected and appropriate methodology were followed to obtained the results indicated that Auricularia auricula-judae (AAJ) was shown to be a good source of almost all amino acids (34.7%) as compared to plant proteins. The pretreatment (blanching) and drying of (AAJ) has significant effect on both the essential and non-essential amino acids. The results from the twenty (20) sample runs with the blanching time of 2, 10, 15, 20, and 28 seconds; drying temperature of 40°C, 55°C, 65°C, 75°C, and 90°C as well as drying time of 30mins, 70mins, 90mins, 110mins and 140mins respectively, show Leucine (an essential amino acid) and Glutamic acid (a non-essential amino acid) to be the highest in concentration with values ranging from 4.61g/100g protein to 6.20g/100g protein and 5.00g/100g protein to 11.96g/100g protein respectively. On the other hand, Methionine (an essential amino acid) and Cystine (a non-essential amino acid) were lowest in concentration with values ranging from 0.32g/100g protein to 1.40g/100g protein and 0.48g/100g protein to 1.93g/100g protein respectively. These results strongly suggest that the fungus (Tree Ear Mushroom) is a rich and healthy food which could be explored for beneficial purposes. And it is recommended that incentive should be given to farmers to cultivate much of Tree Ear Mushroom to meet nutritional demands.

Keywords: amino acids, *auricularia auricula-judae*, fungus, pretreatment, blanching

Introduction
are several edible fungi that are found growing on the trees, and among them is the Tree Ear Mushroom which is universally known as *Auricularia auricula-judae* (AAJ). The Tree Ear Mushroom has been proven to be a very good source of almost all essential amino acids (34.7 percent) as compared to plant proteins. It is also asserted that the chemical content of this (AAJ) fungus proves that it is a valuable raw material to produce low-calorie dietary food as well as a good source of biologically active polysaccharides (Kadnikova *et al*., 2015). It is therefore fit to upheld that Tree Ear Mushroom is an edible fungi grown on plant residues containing 35 percent of protein, all essential amino acids and other substances.

Despites the health derivable benefits of the Tree Ear Mushroom (AAJ), there has been an issue of unawareness on the part of the people on what to do to get cheap food sources that are rich nutritionally. Therefore, there is urgent need for alternative sources of protein to meet the nutritional demand of people. Animal protein has been found to contribute to some health challenges such as heart disease, arteries blockage, and other health issues due to its high cholesterol content, hence alternative protein rich food is important (Ocansey, 2010). Mushrooms are considered as functional foods which can provide health benefits beyond the traditional nutrients, they are seen and used as vegetarian meat supplement.

Besides, Tree Ear Mushroom is in high demand in some part of Nigeria due to its numerous nutritional, economical, and medicinal properties, hence the need to optimize pre-treatment (blanching) and drying to preserve their nutritional quality. Apart from this, the issue of imbalance food nutrients consumed by the people especially in developing countries, where animal protein is grossly scarce, and relatively expensive, requires a shift to Tree Ear Mushroom. Mushrooms are commonly evaluated for their nutritional value. They are rich in proteins, chitin (dietary fibres), vitamins and minerals, low in fat but a high unsaturated fatty acids and have no cholesterols, in general, they contain many useful substances (He, *et al*, 2016).

The Tree Ear Mushroom (*A. auridula-judae*) is a good source of almost all essential amino acids (34.7 percent of total) as compared to plant proteins. The chemical content of this fungus proved that it is a valuable raw material to produce low-calorie dietary food as well as a good source of biologically active polysaccharides. It has been shown to be a good source of almost all the essential amino acid (Kadnikova, *et al*., 2015). Blanching is used to inactivate enzymes like catalase, peroxidase, lipoxygenase, polyphenoloxidase, the enzymes are attributed to the loss of flavours, colour, texture and nutritional qualities during product storage. Another desirable effect of blanching include the expelling of air and gases in the product, and a reduction in the product volume. Due to heat treatment, intercellular gases are removed which result in color retention.

To examine the amino acid composition of pre-treated (blanching) and the nutritional quality of mushroom (*AAJ*)

with specific reference to determine the optimum parameters (time, for blanching), temperature and time for drying of mushroom (*AAJ*), optimize the processing conditions of mushroom (*AAJ*) which is the basis of this research work. It was hypothesized that there is no significant difference among the variables involves. To ascertain whether or not there was a significant difference, Analysis of Variance (ANOVA) was used to check for mean differences or variations among multiple variables.

Literature review

Tree Ear Mushroom

The tree ear or jelly ear mushroom was described scientifically in 1789 by Jean Baptiste Francois (Pierre) Bulliard, who named it *Tremella auricular- judae*. After several changes of genus this fungus was transferred into its present genus in 1897 by Austrian Botanist – Mycologist Richard Wettstein (1863-1934). However, in 1888, the species was given the name *Auricularia auricula-judae* by Joseph Schröter (Schröt, 1888). "Auricula" is a Latin word meaning "ear" and "Judae" means "Judas" – the Jew who is spoken of as the betrayer of Jesus (Bandoni, 1984). The fungus is associated with Judas Iscariot because of the belief that he hanged himself on an elder tree after his betrayal of Jesus Christ. Folklore suggests that the ears are Judas's returned spirit and are all that are left to remind us of his suicide (Kibby, 2003).

Food and Medicinal Value of Mushroom

Mushroom has a multifaceted medicinal and food value chain. It is nutritionally rich and used as food in Chinese cuisine and also regarded as vegetarian meat supplement. While the ground powder of the mushroom is used as a flavor and it's also used to remove excess liquid in soups as an alternative to flours because of its absorbent abilities. It is also used in soups such as: hot and sour soups, draw soups, and in all varieties of stews. The mushroom is used as a trail food which could be eaten raw from it source and is mostly used as a substitute for gum. It is worth noting that mushroom is a delicacy on it own and is commonly used as met in some rural communities of Cross River State.

Medicinally, it is regarded as fungi with high global medicinal value. Typically, in China, it is used in treating sore throat issues, sore eyes, jaundice, cold, fever, lungs infections, and serves as astringents due to its ability to absorb water. It is also used as a cleaning agent for intestine and the stomach. Mushroom is also used in treating hemoptysis (spitting of blood), angina (cardiac pain), diarrhea and warding against gastrointestinal upset. Sadhana G, (2011). Whereas, in Indonesia, Herbalist used mushroom as poultice to treat inflammation of the eye, as well as a palliative to treat sore throats, in the 16th centuries, it was boiled in milk or steeped in beer to produce the throat medicine (Taylor, 2015). Meanwhile, in Southern part of Chana, it was used as blood tonic (Irina, *et al*., 2015). Whereas, in Scotland and Ireland, mushroom is to treat sore throat and jaundice (Abby 2015).

Identification Guide

The tree ear mushrooms are easy to identify in their raw form. They grow on wood and they are distinctively shaped, typically reminiscent of a floppy ear, with fruit bodies covered with tiny hairs, wrinkle, and may have veins making it appear even more ear-like. The outer surface of the lobed fruit body is tan–brown with a purple tinge and covered in a fine grayish velvety down, while the inner surface is smooth. When the fruit bodies aged, they become darker in color while the inner surface is lighter grey purple in color, (Lama, 2019; Mohanan, 2011)

Source: Field data

Fig1: Mature Tree Ear Mushroom (*Auricularia Auricula-Judae*)

Methodology

To achieve the study objective, purposive sampling technique was adopted collect Mushroom fruiting bodies of tree ear (*A*AJ) weighing 1000.32gm from farmers who came from farming settlements in *Idomi* community, *Yakurr* Local Government Area of Cross River State, Nigeria, where there were evidence of visible logs and dead trees. The mushroom was collected during the early rainy season because during this period they are found in abundant, thus the choice of the season. The collected sampled mushroom was sorted out, washed, and dried in room temperature as shown in Table 4. The dried mushroom was properly identified based on their macro- and micro–morphological features.

The mushroom (*A*AJ*)* was properly identified and the whole sample was soaked with 0.5% of sodium Chloride solution (or brine solution) of 2litres (2000 ml) for 3minutes. Ten (10) grams of sodium chloride (NaCl) was dissolved in two (2) liters of water that is brine solution. One thousand (1000) grams of the sample were soaked for three (3) minutes in the brine solution and then filtered.

In the blanching by using hot water at 100ºC, one thousand (1000) grams of the sample were divided into twenty (20) runs, that is, run 1 to 20, and each of the runs contained fifty (50) grams of the mushroom. Distilled water (500 cm3) was used for each of the runs for the boiling point of water at 100ºC and timed in seconds. The blanching time using hot water was 2, 10, 15, 20, and 28 seconds while the drying of sample using hot air (oven) was 40, 55, 65, 75 and 90ºC temperature, at the drying time of 30, 70, 90, 110 and 140 minutes. Each of the sample run was dried out according to the temperature and drying time duration. In order to evaluate the three (3) independent variables which include:

Blanching time (X_1 = 2, 10,15,20,28 Sec)

Drying temperature (X_2 = 40, 55, 65, 75, 90 ^{o} C)

Drying time (X_3 = 30, 70, 90, 110. 140 min) of the mushroom.

Further, in determining the Amino acid and profile, applied Biosystems PTH amino acid Analyzer was used for the determination process. Model 120A PTH amino acid analyzer (HPLC) automatically analyses phenyl

thiohydantoin (PTH) amino acids derived from Edman degradation of proteins and peptides. The PTH amino acids which were eluted for accurate integration included: Leucine, Lysine, Isoleucine, Phenylalanine, Nor-leucine, Tryptophan, Valine, Methionine, Proline, Arginine, Tyrosine, Histidine, Cystine, Alanine, Glutamic Acid, Glycine, Threonine, Serine and Aspartic acid. The amino acid profile in the known sample was determined using methods described by Benitez (1989). The known sample was dried to constant weight, defatted, hydrolyzed, evaporated in a rotary evaporator and loaded into the Applied Bio-systems PTH Amino Acid Analyzer. The samples were defatted by Bligh and Dyer (1959) method. The entire Procedure was carried out in approximately 10 minutes; it was efficient, reproducible, and free from deleterious manipulations. The wet tissue in miscible system was formed with water in the tissue. Dilution with chloroform and water separates the homogenate into two layers – the chloroform layer containing all the lipids and the methanolic layer containing all the non-lipids. A purified lipid extract was obtained by merely isolating the chloroform layer. A known weight (5.0) of ground mushroom was weighed into separating funnel. This was followed by the addition of 15 ml methanol, 30 ml distilled water and 15 ml chloroform. The separating funnel was shaken vigorously for 2 minutes and the liquid layer was decanted into 250 ml conical flask after 10 minutes. The fat free tissue was put into a clean petri dish and dried overnight at room temperature.

In order to calculate the amino acid value, an integrator attached to the Analyzer calculates the peak area proportional to the concentration of each of the amino acids. The calculation is presented thus:

Dry matter DM) content = 100 – M %

Dry matter of sample of weight Wg

$$= \frac{WX\ (100\text{-}M\ \%)\ g}{100}$$

$$= \frac{W \times DM}{100\ g}$$

$$\frac{W\ x\ (D.M)\ g}{100}\ \text{sample} = \frac{14\ x\ 0.1\ x\ T\ gN}{1000}$$

Add 100 g sample = 14 x 0.1 x T x 100 gN
1000 x W x DM
100

$$\%\ \text{Nitrogen} = \frac{14\ x\ T}{W\ X\ DM}$$

Using 0.1 N HCI

Again, the Nitrogen value was multiplied by 6.25 to obtain the weight of protein.

In the determination of nitrogen, a small amount (150 mg) of ground sample was weighed, wrapped in What-man filter paper (No 1) and put in the Kjeldhal 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(jar) was then put in Kjeldhal 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 ml) of the diluted solution with 10 ml of 45% sodium hydroxide was put into 10 ml of 2% boric acid containing 4 drops of bromocresol green/methyl red indicator until about 70 ml of distillate was collected. The distillate was then titrated with standardize 0.01 N hydrochloric acid to grey colored end point. However, the nitrogen percentage was calculated which is this:

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

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 (10 ml)
14.	=	Nitrogen constant in mg.

In the hydrolysis of the sample, a known weight of the defatted sample was weighed into glass ampoule. 7 ml of 6 NHCL 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°c ± 5℃ for 22 hours. The ampoule was allowed to cool before broken open at the tip and the content was filtered to remove the humin. It should be noted that tryptophan was destroyed by 6 N HCL during hydrolysis. 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. However, in the loading of the hydrolysate into analyzer, the amount loaded was 60 microliters. 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.

Results

Amino Acid Analysis (Applied Biosystems PTH Amino Acid Analyzer)

The results of the analysis of the two groups of amino acids (essential and non-essential amino acids) using the Applied Biosystem PTH Amino Acid Analyzer.

Analysis of Variance (ANOVA) Among Amino Acids

Analysis of Variance (ANOVA) was used to check for mean differences or variations among multiple variables. The ANOVA was run with the aid of Statistical Science for Social Sciences (SPSS 23). Statistically, when the p-value (Sig.) is greater than 0.05, it indicates that there is no significant difference among the variables involves. On the other hand, if the significance value is less than the error margin (0.05) a significant difference exists among the variables under study (Ogee *et al*., 2015). The results of the ANOVA are presented in tables and figures as follows:

Table 1 shows the one-way analysis of variance (ANOVA) for differences (variations) among the various amino acids'

Table 1: shows the one-way analysis of variance (ANOVA) for

differences (variations) among the various amino acids' concentration per run

Essential Amino Acids	Concentration: g/100g Protein Range	Non – Essential Amino Acids	Concentration: g/100g Protein Range
Leucine	*4.61 – 6.20*	Proline	*1.83 – 6.70*
Lysine	*1.70 – 5.57*	Tyrosine	*3.44 – 4.82*
Isoleucine	*1.70 – 3.01*	Cystine	*0.48 – 1.93*
Phenylalanine	*2.66 – 4.10*	Alanine	*6.83 – 8.90*
Tryptophan	*0.73 – 1.60*	Glutamic acid	*5.00 – 11.96*
Valine	*3.39 – 6.20*	Glycine	*4.80 – 8.62*
Methionine	*0.32 – 1.40*	Serine	*8.86 – 11.90*
Arginine	*3.78 – 5.85*	Aspartic acid	*7.81 – 9.70*
Histidine	*2.04 – 3.17*		
Threonine	*4.16 – 6.66*		

***Source*:** Field Data

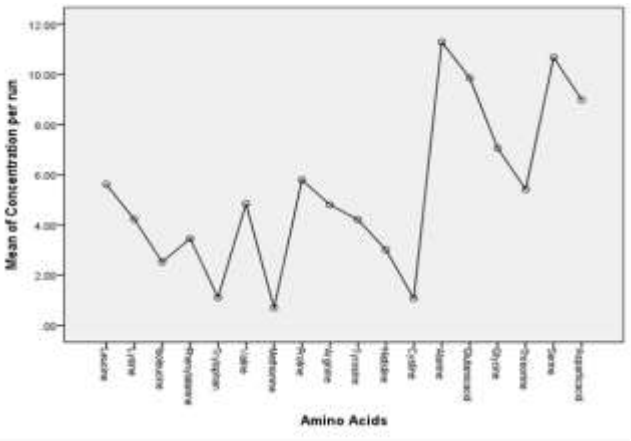
Italicized Data = Raw Data

Table 1

ANOVA					
Concentration per run					
	Sum of Squares	Df	Mean Square	F	Sig.
Between Groups	3560.620	17	209.448	20.119	.000
Within Groups	3560.414	342	10.411		
Total	7121.034	359			

Source: Field data

Figure 1 depicts the mean plot of the analysis of eighteen (18) amino acids revealing their maximum and minimum concentrations per run respectively.



Source: Field data

Fig 1: Mean plot of Amino Acids concentration per run

Table 2: shows the ANOVA among Amino acids concentration with respect to blanching time

	Within Groups	2.575	15	.172		
	Total	2.844	19			
Lysine	Between Groups	4.367	4	1.092	.335	.850
	Within Groups	48.937	15	3.262		
	Total	53.304	19			
Isoleucine	Between Groups	.132	4	.033	.338	.848
	Within Groups	1.466	15	.098		
	Total	1.599	19			
Phenylalanine	Between Groups	.190	4	.048	.530	.715
	Within Groups	1.344	15	.090		
	Total	1.535	19			
Tryptophan	Between Groups	.030	4	.008	.247	.907
	Within Groups	.459	15	.031		
	Total	.489	19			
Valine	Between Groups	.460	4	.115	.239	.912
	Within Groups	7.213	15	.481		
	Total	7.673	19			
Methionine	Between Groups	.126	4	.031	.413	.796
	Within Groups	1.141	15	.076		
	Total	1.267	19			
Proline	Between Groups	4.119	4	1.030	.148	.961
	Within Groups	104.633	15	6.976		
	Total	108.753	19			
Arginine	Between Groups	.281	4	.070	.310	.867
	Within Groups	3.403	15	.227		
	Total	3.684	19			
Tyrosine	Between Groups	18.924	4	4.731	28.909	.000
	Within Groups	2.455	15	.164		
	Total	21.379	19			
Histidine	Between Groups	.285	4	.071	.444	.775
	Within Groups	2.410	15	.161		
	Total	2.695	19			
Cystine	Between Groups	.329	4	.082	.546	.705
	Within Groups	2.259	15	.151		
	Total	2.588	19			
Alanine	Between Groups	167.949	4	41.987	.212	.927

	Within Groups	2964.492	15	197.633		
	Total	3132.441	19			
Glutamic acid	Between Groups	9.694	4	2.424	.258	.900
	Within Groups	140.947	15	9.396		
	Total	150.641	19			
Glycine	Between Groups	3.982	4	.996	.329	.854
	Within Groups	45.405	15	3.027		
	Total	49.387	19			
Threonine	Between Groups	.623	4	.156	.513	.728
	Within Groups	4.559	15	.304		
	Total	5.182	19			
Serine	Between Groups	.244	4	.061	.153	.959
	Within Groups	5.981	15	.399		
	Total	6.226	19			
Aspartic acid	Between Groups	.731	4	.183	.343	.845
	Within Groups	7.997	15	.533		
	Total	8.727	19			

Source: Field data

Table 3: shows the ANOVA among Amino acids concentration with respect to drying time

Analysis of Variance (ANOVA) among Amino Acids by Drying Time						
		Sum of Squares	Df	Mean Square	F	Sig.
Leucine	Between Groups	.270	4	.067	.393	.811
	Within Groups	2.575	15	.172		
	Total	2.844	19			
Lysine	Between Groups	4.367	4	1.092	.335	.850
	Within Groups	48.937	15	3.262		
	Total	53.304	19			
Isoleucine	Between Groups	.132	4	.033	.338	.848
	Within Groups	1.466	15	.098		
	Total	1.599	19			
Phenylalanine	Between Groups	.190	4	.048	.530	.715
	Within Groups	1.344	15	.090		
	Total	1.535	19			
Tryptophan	Between Groups	.030	4	.008	.247	.907
	Within Groups	.459	15	.031		
	Total	.489	19			
Valine	Between Groups	.460	4	.115	.239	.912
	Within Groups	7.213	15	.481		
	Total	7.673	19			
Methionine	Between Groups	.126	4	.031	.413	.796
	Within Groups	1.141	15	.076		
	Total	1.267	19			
Proline	Between Groups	4.119	4	1.030	.148	.961
	Within Groups	104.633	15	6.976		
	Total	108.753	19			
Arginine	Between Groups	.281	4	.070	.310	.867
	Within Groups	3.403	15	.227		
	Total	3.684	19			
Tyrosine	Between Groups	18.924	4	4.731	28.909	.000
	Within Groups	2.455	15	.164		
	Total	21.379	19			
Histidine	Between Groups	.285	4	.071	.444	.775
	Within Groups	2.410	15	.161		
	Total	2.695	19			
Cystine	Between Groups	.329	4	.082	.546	.705
	Within Groups	2.259	15	.151		
	Total	2.588	19			
Alanine	Between Groups	167.949	4	41.987	.212	.927
	Within Groups	2964.492	15	197.633		
	Total	3132.441	19			
Glutamic acid	Between Groups	9.694	4	2.424	.258	.900
	Within Groups	140.947	15	9.396		
	Total	150.641	19			
Glycine	Between Groups	3.982	4	.996	.329	.854
	Within Groups	45.405	15	3.027		
	Total	49.387	19			

Threonine	Between Groups	.623	4	.156	.513	.728
	Within Groups	4.559	15	.304		
	Total	5.182	19			
Serine	Between Groups	.244	4	.061	.153	.959
	Within Groups	5.981	15	.399		
	Total	6.226	19			
Aspartic acid	Between Groups	.731	4	.183	.343	.845
	Within Groups	7.997	15	.533		
	Total	8.727	19			

Source: Field data

Table 4: shows the ANOVA among Amino acids concentration with respect to drying temperature

Analysis of Variance (ANOVA) among Amino Acids by Drying Temperature						
		Sum of Squares	Df	Mean Square	F	Sig.
Leucine	Between Groups	1.261	4	.315	2.989	.053
	Within Groups	1.583	15	.106		
	Total	2.844	19			
Lysine	Between Groups	3.363	4	.841	.253	.904
	Within Groups	49.941	15	3.329		
	Total	53.304	19			
Isoleucine	Between Groups	.839	4	.210	4.143	.019
	Within Groups	.760	15	.051		
	Total	1.599	19			
Phenylalanine	Between Groups	.797	4	.199	4.050	.020
	Within Groups	.738	15	.049		
	Total	1.534	19			
Tryptophan	Between Groups	.175	4	.044	2.090	.133
	Within Groups	.314	15	.021		
	Total	.489	19			
Valine	Between Groups	2.594	4	.648	1.915	.160
	Within Groups	5.080	15	.339		
	Total	7.673	19			
Methionine	Between Groups	.235	4	.059	.856	.512
	Within Groups	1.031	15	.069		
	Total	1.267	19			
Proline	Between Groups	15.596	4	3.899	.628	.650
	Within Groups	93.157	15	6.210		
	Total	108.753	19			
Arginine	Between Groups	1.341	4	.335	2.147	.125
	Within Groups	2.343	15	.156		
	Total	3.684	19			
Tyrosine	Between Groups	1.794	4	.449	.344	.844
	Within Groups	19.584	15	1.306		
	Total	21.379	19			
Histidine	Between Groups	1.176	4	.294	2.901	.058
	Within Groups	1.520	15	.101		
	Total	2.695	19			
Cystine	Between Groups	.581	4	.145	1.085	.399
	Within Groups	2.008	15	.134		
	Total	2.588	19			
Alanine	Between Groups	3131.713	4	782.928	16141.409	.000
	Within Groups	.728	15	.049		
	Total	3132.441	19			
Glutamic acid	Between Groups	27.412	4	6.853	.834	.524
	Within Groups	123.229	15	8.215		
	Total	150.641	19			
Glycine	Between Groups	2.865	4	.716	.231	.917
	Within Groups	46.522	15	3.101		
	Total	49.387	19			
Threonine	Between Groups	2.030	4	.507	2.415	.095
	Within Groups	3.152	15	.210		
	Total	5.182	19			
Serine	Between Groups	3.768	4	.942	5.751	.005
	Within Groups	2.457	15	.164		
	Total	6.226	19			
Aspartic acid	Between Groups	2.177	4	.544	1.246	.334
	Within Groups	6.550	15	.437		

	Total	8.727	19			

Source: Field data

Discussion

The concentration of eighteen (18) amino acids (both essential and non-essential) in 100g of protein were understudied using the Applied Biosystems PTH Amino Acid Analyzer. The results as depicted in "Experimental sample runs: 1 – 20" show Leucine (an essential amino acid) and Glutamic acid (a non-essential amino acid) to be the highest in concentration with values ranging from 4.61g/100g protein to 6.20g/100g protein and from 5.00g/100g protein to 11.96g/100g protein respectively. On the other hand, Methionine (an essential amino acid) and Cystine (a non-essential amino acid) were lowest in concentration with values ranging from 0.32g/100g protein to 1.40g/100g protein and from 0.48g/100g protein to 1.93g/100g protein respectively. This result agrees with the research work of Dabbour and Takruri, (2002) that focused on the amino acid concentration of four (4) species of muchrooms, namely: *T.claveryi*, *P. Ostreatus*, *T. terreum* and *A. macroseorus*. The amino acid analysis showed that Glutamic acid had the highest concentration meanwhile Cysteine and Methionine were the least concentrated amino acid in 100gram dry weight of the mushrooms.

The one-way analysis of variance (ANOVA) for differences (variations) among the various amino acids' concentration per run as shown in Table 1 reveals that there is a significant difference in the concentration of all the amino acids understudied ($p > 0.05$) given the three independent variables Blanching time (X_1), Drying temperature (X_2) and Drying time (X_3). Figure 1, which depicts the mean plot of the analysis, makes transparent the information on Table 1 by revealing the amino acid with the highest and least concentration respectively. Alanine an essential amino acid was found to have the highest concentration while Methionine had the least concentration per run. However, further investigation on the effect of blanching time on the amino acid concentration in AAJ was undertaken as reflected in Table 2. The result therein reveals that there is no significant difference in concentration per blanching time for all the amino acids. This decision was taken because the significant value is greater than the error margin ($p > 0.05$) according to Ogee *et al*. (2015).

The ANOVA among Amino acids concentration with respect to drying time was examined as shown in Table 3. The results revealed that there is no significant difference in concentration per drying time for all the Amino acids except Tyrosine. Tyrosine had a Significant value less than error margin of 0.05 ($p = 0.000$; $F= 28.909$). Statistically, as noted by Ogee *et al*. (2015), when the p-value (Sig.) is less than the error margin (0.05) a significant difference exists among the variables under study. On the other hand, if the significant value is greater than 0.05, it indicates that there is no significant difference among the variables involved. Thus, while other amino acids had significant values greater than the error margin ($p > 0.05$), Tyrosine alone had a significant value less than the error margin ($p < 0.05$). Moreover, the drying temperature had no significant effect on the concentration of the amino acids in AAJ as reflected in Table 4. The p-values of these 18 amino acids were greater than the error margin ($p > 0.05$). The concentration of four of the amino acids understudied were significantly different with regards to the drying temperature per run. These include: Isoleucine ($p = 0.019 < 0.05$; $F=4.143$), Phenylalanine ($p = 0.020 < 0.05$; $F= 4.050$), Alanine ($p = 0.000 < 0.05$; $F= 16141.409$), and Serine ($p = 0.005 < 0.05$; $F= 5.751$).

Conclusion

This study affirms that the Tree Ear Mushroom (AAJ) contains a good number of amino acids both essential and non-essential. The amino acids understudied ($p > 0.05$) – given the three independent variables: blanching time (X_1), drying temperature (X_2), and drying time (X_3) – exhibited significant differences in their concentration. The mean plot of the analysis of these amino acids showed that Alanine had the highest concentration while Methionine had the least concentration per run. However, in 100g of protein sample analyzed, Leucine (an essential amino acid) and Glutamic acid (a non-essential amino acid) were the highest in concentration whereas Methionine (an essential amino acid) and Cystine (a non-essential amino acid) were the lowest in concentration.

Recommendation

Details from the study findings, analysis and result informed the following recommendations:

1. Due to the richness and health importance of Tree Ear Mushroom, it cultivation should be encouraged to the health benefits of the teeming populace.
2. The government through the Ministry of Agriculture, should provide incentives for farmers to cultivate basically Tree Ear Mushroom due to it nutritional value to meet commercial demands.

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