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Effect of Different Salt Concentrations on Protein Solubility of Mushroom Varieties Obtained in Akure, Nigeria

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Abstract The aim of this work is to determine the effect of some salts on the protein solubility of mushrooms found in Akure, Southwest of Nigeria. Standard methods were employed for the task and the results were subjected to statistical analysis. The results obtained compared favourably with other studies. From the results, it was observed that NaCl had the best protein solubility from 9.05 ± 2.00 to 56.31 ± 2.70% and significantly differed from one another at $p > 0.05$. It is recommended that in food formulations, different concentrations of NaCl should be harnessed.

Keywords: *food formulations, fungi, biotechnology, salt concentrations, functional properties*

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1. Introduction

Mushroom resources have been exploited in most developed economies because of their huge agro-industrial, medicinal and commercial benefits [1]. Nigerians utilized mushroom-forming fungi only for food and folk medicine for many decades. *Auricularia auricular* Judae (Bull.) Quél, *Lentinus squarrosulus* Mont., *Pleurotus tuberregium* (Fr.) Singer and *Volvariella volvacea* (Bull.) Singer was some of the common edible mushrooms that were successfully cultivated in Nigeria on small-scale basis. The mushroom resources in Nigeria are grossly under-studied and their attractive potentials under-exploited for addressing economic and industrial development. Resourceful biotechnological approach in the application of mushrooms in agriculture, medicine, industry and environment is inchoate and uncommon in the country.

Functionality of food protein has been defined as those physical and chemical properties which affect the behaviour of proteins in food systems during processing, storage, preparation and consumption [2]. Functional properties constitute the major criteria for the adoption and acceptability of proteins in food systems. Many factors influence the functional properties of proteins, including moisture, temperature, pH, enzymes concentrations, reaction time, chemical additives, mechanical processing, ionic strength and amount, sequence, rate and time of additives had been studied [3].

Protein functionality is dependent on hydrophobic, electrostatic, and steric parameters of the proteins, which are essential for defining the protein structure [4]. Functional properties may be classified according to the mechanism of action on three main groups. These groups are (1) properties related with hydration (absorption of water/oil, solubility, thickening, wettability); (2) properties related with the protein structure and rheological characteristics (viscosity, elasticity, adhesiveness, aggregation and gellying), and (3) properties related with the protein surface (emulsifying and foaming activities, formation of protein-lipid films, whippability) [5,6]. These properties vary with pH, temperature, protein concentration, protein fraction, prior treatment, ionic strength and dielectric constant of the medium as well as other treatments such as interactions with other macromolecules in the medium, processing treatments and modifications, physical, chemical and enzymatic methods [5].

The addition of salt may increase the total water content of the protein system at specific water activity value, although it may decrease the preferential binding of water to the protein. These effects are marked by dependence on anion and cation components [7,8,9].

Far more work must be carried out on those characteristics which will result in products which are more convenient to distribute, easier to process, and have the physical, chemical and organoleptic properties required by the target markets. For those foods such as mushrooms that do not have the native functional characteristics that are desired by the target market, an additional effort must be made to value-add or modify them so that they can compete internationally.

The aim of this research work is to determine the effect of NaCl, $NaSO_4$, $NaNO_3$, $NaNO_2$ and CH_3COONa on

protein solubility of seven mushroom samples found in southwest of Nigeria.

2. Materials and Methods

2.1. Source of Materials and Sample Pre-Treatment

The mushrooms; *Lentinus subnudus* Berk (M_1), *Chlorophyllum molybditis* (M_2), *Volvariella esculenta* (M_3), *Coprinus tramentarius* (M_4), *Pleurotus ostreatus Jacq* (M_5), *Termitomyces microcarpus* (M_6) and *Pleurotus pulmonarius* (M_7) were collected from Federal College of Agriculture campus, Akure, Ondo State, Southwest part of Nigeria. The bad or rotten samples were sorted out. The samples were oven dried at 65°C for 72 hours [ref?] and were then pound into powdered form using porcelain pestle and mortar. The milled samples were then sieved with a 2 mm mesh sized sieve [ref?] and stored in waterproof polyethylene bags at room temperature for further analysis.

2.2. Determination of Protein Solubility in Salt Solutions

0.2 g of each flour sample and 10 ml of a particular salt concentration were thoroughly mixed with a magnetic stirrer (1000 rpm) at room temperature (30°C). Insoluble materials were removed by centrifuging at 3500 rpm for 30 min. The supernatant was dissolved by the Biuret method. The salts used were sodium nitrite, sodium nitrate, sodium chloride, sodium acetate, sodium sulphate with their concentrations ranging between 2 and 12%). The nitrogen obtained was converted to crude protein by multiplying by 6.25 [ref?]. The results were recorded as means of triplicate determinations.

2.3. Statistical Analysis

Data obtained were generated in triplicates and analyzed using Mean, Standard deviation and one-way analysis of variance with Duncan Multiple Range test at 95% confidence or $p < 0.05$.

3. Results and Methods

3.1. Effect of NaCl on Protein Solubility (PS) of Mushroom Samples (%)

The results in Table 1 showed the effect of NaCl on the PS of the mushroom samples in percentage. The PS results varied from 9.05 ± 2.00 to 56.31 ± 2.70% and significantly differed from one another at $p > 0.05$. The PS of M_1, M_3, M_6 and M_7 decreased with increased salt concentrations while the PS of M_2, M_4 and M_5 increased with increased salts concentrations.

The results obtained in this work are in agreement with the works of Mepa *et al*., [10] on the maximum solubility (50.2%) using 1.0 M NaCl solution on coconut protein concentrates. *They also observed that at high NaCl concentration, solubility decreased drastically*. According to researchers, protein solubility has been recorded to be a useful indicator for the performance of protein isolates incorporated in food systems and to determine the extent of protein denaturation because of heat or chemical treatment at line with those reported by others [11].

Table 1. The Results of the Effect of NaCl on Protein Solubility (%) of Mushroom Samples

Salt Conc. (%)	M_1	M_2	M_3	M_4	M_5	M_6	M_7
2	33.15 ± 2.02^{c}	46.15 ± 2.02^{b}	56.31 ± 2.70^{e}	18.24 ± 2.04^{a}	7.31 ± 2.07^{a}	21.18 ± 2.03^{d}	14.28 ± 2.06^{b}
4	28.17 ± 2.02^{b}	45.03 ± 2.00^{ab}	17.28 ± 2.06^{d}	16.25 ± 2.05^{a}	7.31 ± 2.07^{a}	15.03 ± 2.00^{c}	12.26 ± 2.05^{ab}
6	24.01 ± 2.08^{a}	43.10 ± 2.00^{ab}	20.32 ± 2.08^{a}	17.28 ± 2.06^{a}	8.21 ± 2.04^{ab}	8.13 ± 2.01^{a}	$10.27.2.56^{a}$
8	21.02 ± 2.02^{a}	42.29 ± 2.06^{a}	26.02 ± 2.00^{b}	22.07 ± 2.00^{b}	12.06 ± 2.00^{c}	11.16 ± 2.02^{ab}	9.05 ± 2.00^{a}
10	21.11 ± 2.01^{a}	50.00 ± 2.00^{c}	32.28 ± 2.60^{c}	23.02 ± 2.00^{b}	11.22 ± 2.04^{bc}	14.04 ± 2.00^{bc}	9.20 ± 2.03^{a}
12	22.02 ± 2.00^{a}	54.01 ± 2.00^{d}	31.24 ± 2.04^{c}	23.24 ± 2.04^{b}	12.23 ± 2.04^{c}	13.26 ± 2.05^{bc}	10.09 ± 2.01^{a}

All values were expressed as averages of triplicate determinations ± the standard deviations and values bearing the same superscripts in the same row are significantly different ($p < 0.05$)

Table 2. The Results of The Effect of Na_2SO_4 on Protein Solubility (%) of Mushroom Samples

Salt Conc. (%)	M_1	M_2	M_3	M_4	M_5	M_6	M_7
2	29.13 ± 2.20^{b}	41.02 ± 2.00^{ab}	18.04 ± 2.00^{b}	27.21 ± 2.03^{c}	12.16 ± 2.01^{bc}	7.06 ± 2.00^{a}	13.05 ± 2.00^{a}
4	21.27 ± 2.05^{a}	43.25 ± 2.04^{c}	16.24 ± 2.04^{b}	28.10 ± 2.01^{c}	13.24 ± 2.04^{c}	6.27 ± 2.06^{a}	10.06 ± 2.00^{a}
6	24.01 ± 2.00^{a}	37.32 ± 2.07^{ab}	19.09 ± 2.01^{b}	17.21 ± 2.03^{a}	9.05 ± 2.00^{ab}	7.30 ± 2.07^{a}	10.20 ± 2.05^{a}
8	23.02 ± 2.00^{a}	39.32 ± 2.01^{ab}	10.13 ± 2.01^{a}	23.24 ± 2.04^{b}	11.22 ± 2.03^{abc}	8.33 ± 2.08^{a}	10.25 ± 2.00^{a}
10	22.27 ± 2.05^{a}	36.05 ± 2.00^{a}	36.05 ± 2.00^{c}	20.15 ± 2.02^{ab}	$9..05 \pm 2.00^{ab}$	7.22 ± 2.04^{a}	10.09 ± 2.01^{a}
12	22.19 ± 2.05^{a}	36.20 ± 2.03^{a}	36.20 ± 2.03^{c}	20.01 ± 2.00^{ab}	8.21 ± 2.03^{a}	9.03 ± 2.00^{a}	10.17 ± 2.02^{a}

All values were expressed as averages of triplicate determinations ± the standard deviations and values bearing the same superscripts in the same row are significantly different ($p < 0.05$)

3.2. Effect of Na_2SO_4 on Protein Solubility (%) of Mushroom Samples

The results in Table 2 showed the effect of Na_2SO_4 on the PS of the mushroom samples in percentage. The PS results varied from 6.27 ± 2.06 to 43.25 ± 2.04% and significantly differed from one another at $p > 0.05$. The PS of M_1, M_2, M_4, M_5 and M_7 decreased with increased salt concentrations while the PS of M_3 and M_6 increased with increased salts concentrations.

Our results are not comparable with results obtained by Masood and Rizwana [12] on legumes protein isolates. They recorded between 65 and 82%, which are far above our results. This higher solubility of legumes protein isolates as compared to mushroom samples may be due to the presence of low number of hydrophobic residues and the elevated charge. The solubility of a protein is usually affected by its hydrophilicity or hydrophobic balance, depending on surface active agents, can form and stabilize the amino acid composition, particularly at the protein surface [6].

3.3. Effect of $NaNO_3$ on Protein Solubility (%) of Mushroom Samples

The results in Table 3 showed the effect of $NaNO_3$ on the PS of the mushroom samples in percentage. The PS results varied from 6.27 ± 2.06 to 43.25 ± 2.04% and significantly differed from one another at $p > 0.05$.The PS of M_1, M_2, M_4, M_5 and M_7 decreased with increased salt concentrations while the PS of M_3 and M_6 increased with increased salts concentrations.

According to Mwasaru *et al*. [13], the calculated results obtained for protein solubility of pigeon pea and cowpeas were 53.4 and 61.8% respectively. The higher solubility of legume protein isolates as compared with our results on mushrooms may be due to the presence of low number of hydrophobic residues and the elevated charge. The identification of some functional properties of these mushroom varieties is essential in determining their potential uses in the formulation of foods.

Table 3. The Results of Effect of $NaNO_3$ on Protein Solubility (%) of Mushroom Samples

Salt Conc. (%)	M_1	M_2	M_3	M_4	M_5	M_6	M_7
2	28.09 ± 2.01^{b}	45.08 ± 3.51^{c}	33.33 ± 2.08^{c}	19.12 ± 2.01^{a}	9.21 ± 2.03^{a}	9.15 ± 2.01^{a}	13.12 ± 2.01^{b}
4	29.25 ± 2.05^{b}	43.25 ± 2.05^{bc}	34.09 ± 2.01^{c}	20.15 ± 2.02^{ab}	15.18 ± 2.02^{b}	22.01 ± 2.00^{c}	10.09 ± 2.01^{ab}
6	22.02 ± 2.00^{a}	39.24 ± 2.04^{ab}	28.11 ± 2.01^{b}	21.04 ± 2.00^{ab}	8.18 ± 2.02^{a}	$12.2 \pm .2.03^{ab}$	10.06 ± 2.00^{ab}
8	22.02 ± 2.00^{a}	45.18 ± 2.05^{c}	17.16 ± 2.01^{a}	23.31 ± 2.07^{bc}	17.22 ± 2.03^{b}	14.12 ± 2.01^{b}	10.17 ± 2.02^{ab}
10	20.25 ± 2.00^{a}	36.05 ± 2.00^{a}	27.25 ± 2.05^{b}	26.11 ± 2.05^{c}	22.11 ± 2.01^{c}	13.26 ± 2.05^{b}	9.08 ± 2.01^{a}
12	30.16 ± 2.01^{b}	41.17 ± 2.02^{bc}	26.21 ± 2.03^{b}	26.19 ± 2.03^{c}	17.09 ± 2.01^{b}	15.27 ± 2.05^{b}	10.32 ± 2.08^{ab}

All values were expressed as averages of triplicate determinations ± the standard deviations and values bearing the same superscripts in the same row are significantly different ($p < 0.05$)

3.4. Effect of CH_3COONa on Protein Solubility (%) of Mushroom Samples

The results in Table 4 showed the effect of CH_3COONa on the PS of the mushroom samples in percentage. The PS results varied from 6.00 ± 2.00 to 45.00 ± 2.00% and significantly differed from one another at $p > 0.05$. The PS of M_1, M_2, M_4, M_5 and M_7 decreased with increased salt concentrations while the PS of M_3 and M_6 increased with increased salts concentrations. The results obtained using $NaNO_3$ had a similar PS when compared to these results, but PS of NaCl was quite higher than these results. In this work, CH_3COONa may have decreased the preferential binding effect of water to the mushrooms' protein.

Table 4. The Results of the Effect of CH_3COONa on Protein Solubility (%) of Mushroom Samples

Salt Conc. (%)	M_1	M_2	M_3	M_4	M_5	M_6	M_7
2	29.13 ± 2.20^{b}	41.02 ± 2.00^{bc}	18.04 ± 2.00^{b}	27.21 ± 2.03^{c}	12.16 ± 2.02^{bc}	7.06 ± 2.00^{a}	13.05 ± 2.00^{a}
4	21.27 ± 2.05^{a}	45.00 ± 2.00^{c}	16.24 ± 2.04^{b}	28.10 ± 2.01^{c}	13.24 ± 2.04^{c}	6.00 ± 2.00^{a}	10.06 ± 2.00^{a}
6	24.01 ± 2.00^{a}	37.32 ± 2.07^{ab}	19.09 ± 2.01^{b}	17.21 ± 2.03^{a}	9.05 ± 2.00^{ab}	7.30 ± 2.07^{a}	10.21 ± 2.03^{a}
8	23.01 ± 2.00^{a}	39.10 ± 2.01^{ab}	10.13 ± 2.01^{a}	23.24 ± 2.04^{b}	11.22 ± 2.04^{abc}	8.33 ± 2.07^{a}	10.25 ± 2.04^{a}
10	22.27 ± 2.00^{a}	36.05 ± 2.00^{a}	36.05 ± 2.10^{c}	20.15 ± 2.02^{ab}	9.05 ± 2.00^{ab}	7.22 ± 2.04^{a}	10.09 ± 2.01^{a}
12	22.19 ± 2.03^{a}	36.20 ± 2.03^{a}	36.20 ± 2.03^{c}	20.01 ± 2.00^{ab}	8.21 ± 2.03^{a}	9.03 ± 2.00^{a}	10.17 ± 2.03^{a}

All values were expressed as averages of triplicate determinations ± the standard deviations and values bearing the same superscripts in the same row are significantly different ($p < 0.05$)

3.5. Effect of $NaNO_2$ on Protein Solubility (%) of Mushroom Samples

The results in Table 5 showed the effect of $NaNO_2$ on the PS of the mushroom samples in percentage. The PS results varied from 6.20 ± 2.00 to 54.28 ± 2.06% and significantly differed from one another at $p > 0.05$. The PS of M_1 and M_2 decreased with increased salt concentrations while the PS of M_3, M_4, M_5, M_6 and M_7 increased with increased salts concentrations. The results obtained here were in agreement with results of Mepa *et al*. [10]. The coconut protein in their work showed solubility of about 54.6% and 53.2% in FFC-PC and DFC-PC, respectively.

Table 5. The Results of The Effect of $NaNO_2$ on Protein Solubility (%) of Mushroom Samples

Salt Conc. (%)	M_1	M_2	M_3	M_4	M_5	M_6	M_7
2	31.16 ± 2.02^{b}	53.19 ± 2.03^{b}	15.30 ± 2.07^{a}	25.08 ± 2.01^{a}	16.08 ± 2.01^{bc}	6.20 ± 2.00^{a}	14.09 ± 2.01^{a}
4	26.09 ± 2.01^{a}	50.01 ± 2.00^{b}	22.31 ± 2.07^{b}	38.03 ± 2.01^{b}	17.17 ± 2.02^{bc}	9.19 ± 2.02^{ab}	12.29 ± 2.06^{a}
6	28.16 ± 2.02^{ab}	45.03 ± 2.00^{a}	32.28 ± 2.06^{c}	16.62 ± 1.47^{c}	9.05 ± 2.00^{a}	14.12 ± 2.01^{cd}	13.24 ± 2.04^{a}
8	25.09 ± 2.01^{a}	46.29 ± 2.06^{a}	39.68 ± 2.07^{d}	41.26 ± 2.05^{c}	18.26 ± 2.05^{bc}	$19.17.2.02^{e}$	13.38 ± 2.51^{a}
10	30.07 ± 2.00^{b}	44.07 ± 2.00^{a}	36.27 ± 2.06^{cd}	48.18 ± 2.02^{d}	19.10 ± 2.01^{c}	16.05 ± 2.00^{de}	14.21 ± 2.03^{a}
12	30.07 ± 2.11^{b}	51.11 ± 2.01^{b}	31.33 ± 2.08^{c}	54.28 ± 2.06^{e}	15.08 ± 2.00^{b}	11.32 ± 2.08^{bc}	15.14 ± 2.02^{a}

All values were expressed as averages of triplicate determinations ± the standard deviations and values bearing the same superscripts in the same row are significantly different ($p < 0.05$)

4. Conclusion

From the results obtained in this work, it is clear that salts have effects on the protein solubility of the different mushroom varieties. In comparing the different salts used it was observed that NaCl was better in comparisons with other salts concentrations. It had higher protein solubility at the least and higher concentrations.

Recommendation

It is recommended that in food formulations, different concentrations of NaCl should be harnessed.

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