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Original Research Article

Comparative Studies of the Protein Content of Probiotic and Nonprobiotic Treated Chicken Meat

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ABSTRACT

Proteins are the major components for building body. The present study was to test the concentration of proteins of two meat samples. Sample one from the chicken meat was treated with probiotics and enzyme and the second sample was treated as control and treated with normal basal diet. Nowadays chicken consumption had increased because of good protein storage. In my present study, the chicken breast sample from probiotic treated chicken had protein concentration of 2mg/ml(protein concentration0.4mg/g) which is higher than the control sample which contained the 2 mg/ml(protein concentration 0.8mg/g). The protein sample was estimated the Biuret method. The Kindjal method was used for estimation of total nitrogen previously .but the actual protein content was estimated by Kindjal. So, replaced the Kindjal by Biuret method. This method was simple and accurate. The reason behind why the protein content high in the chicken meat sample was due to consumption of total feed high, and low litter quantity. When compared with control group.

Keywords: protein concentration, probiotics, Chicken meat,

INTRODUCTION

Proteins are polymers of amino acids. Proteins differ from each other according to the type, number, and sequence of amino acids that make up to polypeptide backbone. As a result, they have different molecular structure, nutritional attributes, and physiochemical properties. Proteins are important constituents of foods for several different reasons. They are a major source of energy, as well as contains essential amino acids, such as lysine, tryptophan, methionine, leucine, isoleucine and valine which essential to human health, but which the body not synthesized. Proteins are also the major structural components of many natural foods, Many food proteins are enzymes which are capable of enhancing the rate of certain biochemical reactions. These reactions either favorable or detrimental effect on the overall properties of foods.

Particularly, proteins were considered as the most important components of meat from a nutritional and processed viewpoint. Indeed, (Smyth *et al.*, 1999) meat proteins contains all the amino acids essential to the human body, thus making them highly nutritious (Friedman, 1996). Moreover, meat proteins greatly contribute to processed abilities by imparting specific functionalities. The overall properties of meat and meat products, included the appearance, texture, and mouthfeel are dependent on protein functionality (Xiong, 2004). It was well known that myofibrillar proteins (i.e., myosin and actin) are mainly responsible for the Whole human components and textural properties of meat and meat products, whereas sarcoplasmic proteins (i.e., muscle enzymes) played a minor role (Smith, 2010; Sun and Holley, 2011; Petrucci *et al.*,

2013c). The solubility of myofibrillar and sarcoplasmic proteins is highly correlates with water retention (Li-Chan *et., al*1987; Warner *et., al* 1997). Protein solubility also has a major role in the physical properties of the meat because lower protein solubility imparts poor functionality, as in the case of pale, soft, and exudative (PSE)-like meat (Van Laack *et. al* 2000; Bowker and Zhuang, 2013).

Brewer developed a Kjeldahl method for protein estimation in 1883called Johann Kjeldahl. The Kjeldahl method does not given a measure of the true protein since all nitrogen in foods are not in the form of protein. Different proteins need different correction factors because they have a different amino acid sequence. The use of concentrated sulfuric acid at high temperatures possessed a considerable hazard and the used some of the possible catalysts. The technique is time-consuming to carry-out.

One of the simplest and most common was the Biuret Protein Assay. (The name of this assay is somewhat confused because assayed for proteins used this method was not actually used biuret). Biuret protein assay method is caused by a binding reaction of proteins at basic PHamide and carboxyl group formed the peptide bond was found by Biuret. Protein samples combines with Biuret Reagent of copper ions will produce a blue color. The concentration of blue is directly proportional to the protein concentration and in samples. It is measured by Spectrophotometer.

MATERIALS AND METHODS

2.1 Biuret method &Samples preparation: In addition to the standard curve, we assayed one of seven different protein samples of unknowns from the instructor's bench. Weighed out 0.3-0.4g of the sample into a 50ml blue cap conical tube. Add around 15 glass beads and 0.5% of SDS in Quantity of 10ml to the sample the.SDS was a detergent that was helped to disrupt cellular membranes and release proteins into solution. Vortex the mixtures in a swinging bucket centrifuge. Removed the tubes from the centrifuge and carried them to your table, take care not to shake the sample.

2.2 Reagents:

1.0.1NNaOH: 1gm of NaOH was dissolved in 250ml of water

2.Alkaline sodium carbonate:2.0g of sodium carbonate was dissolved in 0.1Na OH and makeup to 100ml volume with 0.1N NaOH

3.1% Sodium potassium tartrate:100mg in 10ml water

4.Copper sulphate:0.5% Cuso4.% H20 was prepared by using 1Na k tartrate (50mg in 10ml NaK)

5.Alkaline copper Sulphate reagent:1ml of CusO4 regent was added to 50ml of alkaline

6.Sodium carbonate: The mixture was unstable and should be prepared freshly.

7. Biuret reagent

8.Stock:100mg of BSA was dissolved in 100ml of water

9.Working Stock solution:1ml of stock solution was added in 5ml of water.

2.3 Procedure:

Prepared the standard curve when sample spinning as follows. Label two sets of test tubes as 1,2,3,4,5,6. First water of the required amount was taken and Then pippeted the appropriate amount of BSA stock solution (10mg/ml). Do not add the biuret reagent until your unknown sample was ready.

Carefully pipetted1ml of the liquid portion of your known sample into clean, duplicate test tubes (DO NOT add the unknown to the tubes you made for the standard curve). Now added 2ml biuret reagent to every tube: the 12 tubes for the standard curve and 2 tubes for your unknown. Cover the tubes with parafilm and briefly vortexed to ensure that the sample and the biuret were thoroughly mixed. Allowed all of the tubes to stand at room temperature of 15minutes. You have waited for a turned on the Spectrophotometer and allowed it to warm up, this may take 1-2 minutes. Do not push any buttons until the

machine was ready. (When the ABS reading at 542 nm appears on the screen.) Adjust the wavelength to 550nm.

Each tube was wiped gently with tissue paper and read in the spectrophotometer, gently wiped the tube with a tissue paper to remove fingerprints and dust. After incubation of 15minutes, reading were taken. place tube 1 into the spectrophotometer and set the absorbance to zero. This tube was served as the blank. record the value in the table. Measured the absorbance of the other tube 1 as well. The absorbance should be very close to zero. DO NOT Zero/blank the instrument again. Standards absorbances values recorded continuously in the table. Finally, record the absorbance of your unknown. Determine the average ABS for all tubes. Dispose of your Biret solutions in the designed waste container at the instructor bench.

RESULTS & DISCUSSION

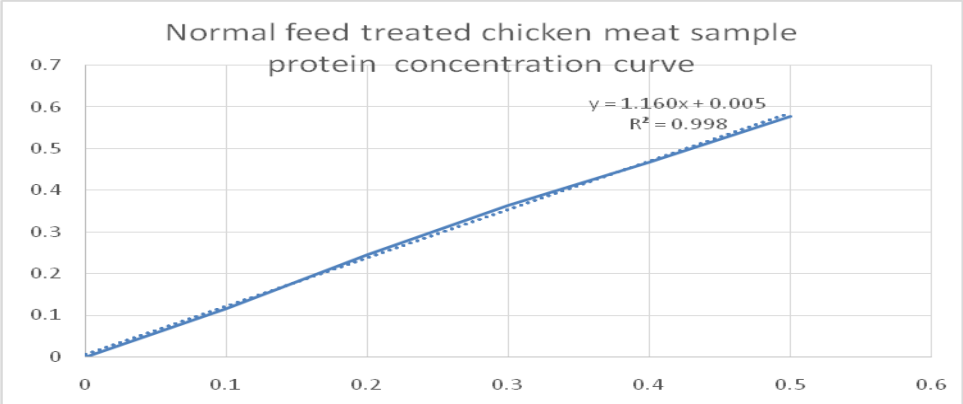
Table:1 Raw data for the calibration curve for the standard Bovine serum albumin. Bovine serum albumin concentration was increases as absorbance values also increases, At the first tube without BSA shows the absorbance value of 0.102.And average was 0.101.The final tube contained the BSA concentration of 5mg /ml and absorbance was 0.764 and average shows that 0.763.

Tube	BSA Conc.(mg/ml)	H2O(ml)	BSA stock(ml)	Biuret Reagent(ml)	ABS	ABS average
1	0	1.0	0	2	0	
2	1	0.9	0.1	2	0.102	0.101
3	2	0.8	0.2	2	0.247	0.246
4	3	0.7	0.3	2	0.387	0.385
5	4	0.6	0.4	2	0.675	0.673
6	5	0.5	0.5	2	0.764	0.763
7	6					
8	7					

Table-2 Raw data for the calibration curve (P-Value=0.227)for the normal feed treated chicken meat sample. The unknown sample concentration of two replicas shows the absorbance values like 0.554,0.549 & the concentration of the protein was shows 0.424,0.411.

Unknown	BSA Conc.(mg/ml)	H2O(ml)	BSA stock(ml)	Biuret Reagent(ml)	ABS	The concentration of proteins in the sample (mg/g)
1	0	1	0	2	0.554	0.424
1	0	1	0	2	0.549	0.411
0.9	0	1	0	2	0.531	0.404

Figure 1.calibration curve((P value=0.227)) using with BSA and Normal feedtreated chicken meat sample. X axis concentration of the protein and y axis absorbance values shows that absorbance of 0.005 and concentration of the protein was 0.46mg/ml.

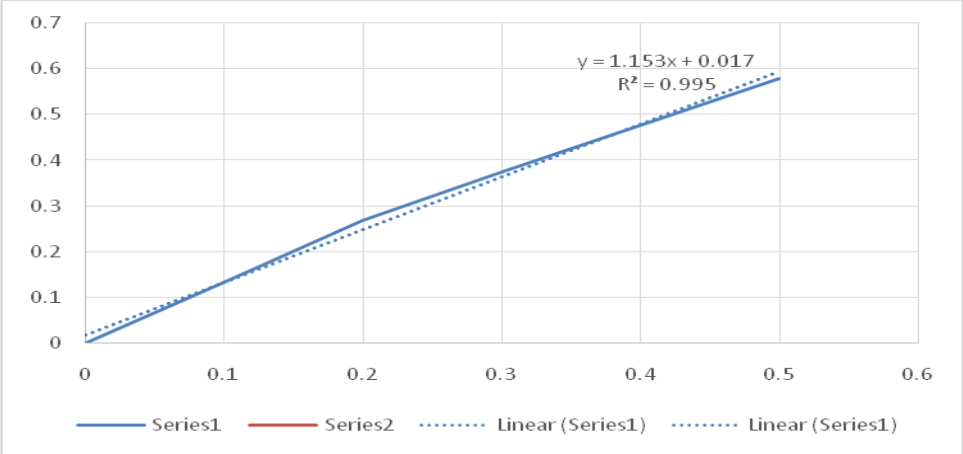


3.1 Calculations: y=1.1609x+0.005
X=y-0.005/1.1609
X=0.587-0.05/1.1609
=0.537/1.1609
=0.46mg/ml

Table-3 Raw data for the calibration curve P value=0.227)for the normal probiotic treated chicken meat sample. Un known sample 1 and Un known sample 2 shows the absorbance values of 0.954,0.967 and Concentration of the proteins 0.812mg/ml and 0.823mg/ml.

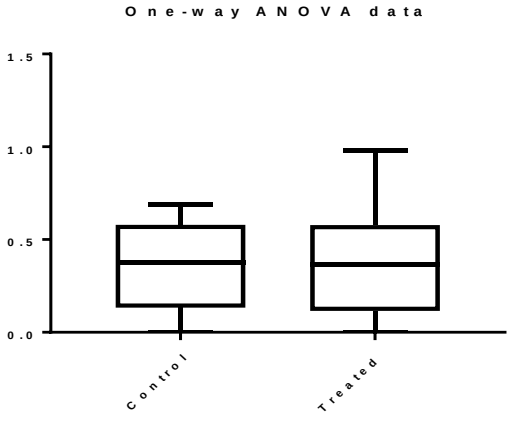
Unknown1	H2O(ml)	BSA stock(ml)	Biuret Reagent(ml)	ABS-Un known -1	ABS-Un known -2	The concentration of proteins in the sample (mg/g)
1	1	0	2	0.946	0.954	0.812
1	1	0	2	0.967	0.967	0.823
0.9	1	0	2	0.947	0.947	0.806

Figure 2.calibration curve((P value=0.227)) using with BSA and normal probiotic treated chicken meat sample. X axis shows the concentration and y axis the absorbance values. Final concentration of the protein at probiotic treated group was shows the 0.835mg/ml.



3.2 Calculations:
Y=1.153x+0.0172
X=y-0.0172/1.153
X=0.98-0.0172/1.153
=0.9628/1.153
= 0.835mg/ml

Figure:3 Calibration curve(P value=0.494) for a protein concentration of probiotic treated and normal feed treated chicken meat samples. Shows that highest protein value shows at probiotic treated group than the control group. The graph shows at 1 point and the control group was shows at between the 1-1.5 points.



A graph (Standard curve) was plotted to determine the concentration of the Unknown sample. To determine the actual concentration of protein in the unknown samples, it was necessary to graph the standard curve (Concentration on the X-axis and on the absorbance y-axis) and interpolate the absorbance values of the unknowns. A more accurate method was to perform a linear regression analysis on the standard curve which was l yield the equation for a straight line: $Y=m^{x+b}$**.** With this equation, calculated the concentration by entering the absorbance value of each unknown into the equation as the Y value and then solve for X.Table-1 showed that the concentration of BSA solution increased the absorbance value also increased the O.D values by using the spectrophotometer. Table-2 showed that concentration of control chicken meat samples of 1ml and 0.9ml of 3 samples the average of the 3 samples concentrations absorbance is 0.687

and concentration of protein was 0.80mg/ml. The other sample was probiotic treated chicken breast meat sample contained the average of absorbance of 3 samples 0.98 and concentration of meat samples 1.14mg/ml. Table -3 showed that comparison of protein concentration concentrations

Figure 1 was the calibration curve for the Bovine serum albumin as a standard Figure -2 was the calibration curve of the protein concentration of normal feed treated chicken meat sample Figure 3 was the comparison graph for both protein samples.

CONCLUSION

According to the above results, we concluded that the concentration of the meat sample from probiotic treated chicken had the highest protein content of 0.835mmg/ml. whereas the control meat sample contains 0.548mg/ml. The probiotic in feed was adhered to the intestine and allowed the food for complete digestion and absorption and caused the litter content reduction in probiotic-treated chickens. The complete absorption of feed had caused the muscles to build-up and make up the meat with high quality of proteins. This was the major reason for the increment of the proteins for the probiotic-treated chicken meat samples.

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