



DETERMINATION OF TOTAL SOLUBLE PROTEIN IN DIFFERENT FOOD SAMPLES

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ABSTRACT

Soluble protein is that part of protein which is easily soluble in water or blood plasma and is readily available to cells. Excessive soluble protein causes protein loss through body waste such as sweat or urine. This study was designed to find total soluble proteins from a given food sample. A bio-RAD method was used and its absorption was measured in spectrophotometer. The readings were taken and compare with the known concentrations. A Standard curve has been drawn for protein content.

KEYWORDS: Soluble protein is that part of protein content.

INTRODUCTION

To ensure a constant supply of soluble protein, the pH level of blood needs to be ideal in 7.35-7.45. The total soluble protein, albumin, globulin and pseudo globulin those are soluble in NH₂So₄ and pure water (Jones, 1941). A soluble rice protein is from rice also very popular for being a dietary staple. It provides protein for growth of body and strengthening of cells (Pelosi *et al.*, 2006). The membrane protein helps in formation of membrane and strengthening of membrane. The globulin protein is only soluble protein which feeds the cells and used in synthesis of enzyme (Prosky *et al.*, 1987).

Bio-RAD method is very sensitive method in detecting protein in a given sample. A small quantity of protein in a total of 1ml is treated with 3ml Bio-RAD solution and contents can be mixed and left at room temperature for 15 minutes (Spector, 1978). The protein reacts with dye and blue colored complex is produced. The amount of protein is calculated from standard curve (Sedmak *et al.*, 1977).

METHODOLOGY

The sample of Germinating wheat seed, Onion, green chili and potato were collected. The samples were weighed and 25mg was taken in mortar and crushed by adding about 5ml of distilled water. The material was homogenized and filtered. Each sample was grind separately to extract the protein by bursting the cell wall and cell membrane with mechanical methods. The extract was filtered and preserved at 4⁰C for two days. 0.5ml of sample was taken and added in 0.5ml of distilled water with the help of micropipette. 3ml of Bio-RAD solution was added in the filtrate and vortex for 5

minutes. The mixture was left for 15 minutes at room temperature. Sample was taken into cuvettes and its absorption was recorded at 595nm in spectrophotometer.

Preparation of Bio-RAD solution

Bio-RAD solution was prepared by dissolving 500mg of brilliant blue G (dye) in 5ml of ethyl alcohol to 100ml of orthophosphoric acid and volume was diluted with 1 liter distilled water. The prepared solution was filtered and stored in amber color bottle in refrigerator at 4⁰C for 15 days (Congdon *et al.*, 1993).

Sample	Weight and added volume of distilled water	Volume of filtrate	Sample 0.5ml + 0.5ml of distilled water + 3ml of Bio-RAD	Absorption A (O.D)
Germinating wheat seed (leaflet)	0.034g + 5ml water	2.6ml	4ml	0.253
Onion	0.03g + 10ml water	8ml	4ml	0.184
Green chili	0.028g + 5ml water	2.6ml	4ml	0.282
Potato	0.080g + 10ml water	5.5ml	4ml	0.174

RESULTS

The more the concentration of soluble protein more is the absorption and hence more is the optical density. The highest value of soluble protein was estimated in green chili which is 0.39% and lowest was determined in potato which is 0.15%. The percentage of germination wheat seed and onion estimated 0.27% and 0.22%.

Calculation of total soluble protein

Leaflet of germinating wheat seed

Absorption (A) = 0.253

Fresh weight = 0.034g

Final volume = 2.6ml

0.5ml of leaflet extract contain = 18ug

1ml contains = 18/0.5

2.6ml contains = 18/0.5*2.6ml

= 93.6ug protein in 2.6ml

0.034g of sample contain protein = 93.6ug

1ml contains = 93.6/0.034

= 2752.9ug or 2.752mg or 0.00275g

If 1 g contains = 0.00275g

Or 0.00275/1*100

= **0.275 % total soluble protein in wheat seed.**

2- Onion

Absorption (A) = 0.184

Fresh weight = 0.03g

Final volume = 2.8ml

0.5ml of onion extract contain = 12ug of protein

1ml contains = 12/0.5

2.8ml contains = 12/0.5*2.8

= 67.2ug

If 0.03g contains protein = 67.2ug

1g contains = 67.2/0.03

= 2240ug or 0.00224g

1g contains = 0.00224g

= 0.00224/1*100

= **0.224% total soluble protein in onion.**

3- Green chili

Absorption (A) = 0.282

Fresh weight = 0.028g

Final volume = 2.6ml

0.5ml of green chili extract contain = 21ug protein

1ml contains = 21/0.5ml

2.6ml contains = 21/0.5ml*2.6ml

= 109.2ug of protein

0.0282g contains protein = 109.2ug

1g contains = 109.2/0.028

= 3900ug or 0.0039g

1g contains = 0.0039g

100 contains = 0.0039/1*100

= **0.39% total soluble protein in green chili.**

4- Potato

Absorption (A) = 0.174

Fresh weight = 0.080 g

Final volume = 5.5ml

0.5ml of potato extract contain = 11ug of protein

1ml contains = 11/0.5ml

5.5ml contains = 11/0.5ml*5.5

= 121ug

0.08g contains protein = 121ug

1g contains = 121/0.08

= 1512.5ug or 0.00151g

100g contain protein = 0.00151/1*100

= **0.15% total soluble protein in potato.**

DISCUSSION

Proteins are major components of living cells. IR spectra method is recently used for the estimation of soluble proteins. In terms of IR spectra no difference is found between soluble and membrane proteins (Goormaghtigh *et al.*, 1994). Many other improved sensitive techniques have been developed for the determination of soluble proteins. The interference of phenolic compounds and oxidases have been eliminated during extraction. It also includes the persistence of detected glycoprotein. The sensitivity for protein detection is increased 100-fold by using silver staining instead of Coomassie blue. The relative concentration of proteins have been changed on stained gels and now a days it can be determined by laser scanning densitometry (Hsu *et al.*, 1987).

REFERENCES

1. Goormaghtigh, E., Cabiaux, V., & Ruyschaert, J. M. Determination of soluble and membrane protein structure by Fourier transform infrared spectroscopy. In *Physicochemical methods in the study of biomembranes*, 1994; 405-450. Springer US.
2. Hsu, J. C., & Heatherbell, D. A. Isolation and characterization of soluble proteins in grapes, grape juice, and wine. *American Journal of Enology and Viticulture*, 1987; 38(1): 6-10.
3. Pelosi, P., Zhou, J. J., Ban, L. P., & Calvello, M. Soluble proteins in insect chemical communication. *Cellular and Molecular Life Sciences CMLS*, 2006; 63(14): 1658-1676.
4. Congdon, R. W., Muth, G. W., & Splittergerber, A. G. The binding interaction of Coomassie blue with

- proteins. *Analytical biochemistry*, 1993; 213(2): 407-413.
5. Sedmak, J. J., & Grossberg, S. E. A rapid, sensitive, and versatile assay for protein using Coomassie brilliant blue G250. *Analytical biochemistry*, 1977; 79(1-2): 544-552.
 6. Spector, T. Refinement of the Coomassie blue method of protein quantitation: A simple and linear spectrophotometric assay for ≤ 0.5 to 50 μg of protein. *Analytical biochemistry*, 1978; 86(1): 142-146.
 7. Jones, D. B. *Factors for converting percentages of nitrogen in foods and feeds into percentages of proteins* Washington, DC: US Department of Agriculture, 1941; 1-22.
 8. Prosky, L., Asp, N. G., Schweizer, T. F., DeVries, J. W., & Furda, I. Determination of insoluble, soluble, and total dietary fiber in foods and food products: interlaboratory study. *Journal-Association of Official Analytical Chemists*, 1987; 71(5): 1017-1023.