Asian Journal of Agriculture & Life Sciences Vol. 5(1), January 2020: 4-6

Website: www.crsdindia.com/ajals.html e-ISSN: 2455-6149

ORIGINAL ARTICLE

Methodology of Protein Estimation from Helminth Parasites and its Host Tissue

Dhanraj Balbhim Bhure* and Sanjay Shamrao Nanware

Department of Zoology (UG & PG), Yeshwant Mahavidyalaya, Nanded-431602

*Email of corresponding author- drajbhure82@gmail.com

Received: 20th Nov. 2019, Revised: 30th Nov. 2019, Accepted: 8th Dec. 2019

ABSTRACT

The helminth parasites utilize the food from the gastrointestinal tract of the host so their metabolism depends on the nourishment available in the gut of the host. The metabolic and in vitro studies suggest that parasites need proteins from the predigested food from the host intestine for various metabolic activities. Ammonia and urea are obtained from intracellular compounds, so these are the true end products. The cestodes are used to live freely in a watery medium and they remove the end products so they are known as ammonotelic organisms. Proteins are fundamental units for all metabolic activities; they are most important agents for expression of the genetic material. The occurrence of proteins in the body of parasites. Biochemical indices have been employed in effectively monitoring the responses of organisms to stressors and thus its health status under such adverse conditions. Hence, the changes associated with Biochemical parameters due to various parasites establish a database, which could be used in disease diagnosis and in guiding the implementation of the treatment or preventive measures. Hence it is essential to know the methods for protein estimation from host and parasites. Therefore the present study undertaken on methodology of protein estimation from parasites and their host tissue.

Key words: *Helminth parasites, Methodology of Protein estimation*

INTRODUCTION

Proteins are very important molecules in our cells. They are involved in virtually all cell functions. Each protein within the body has a specific role. Some proteins are involved in structural support, while others are involved in bodily movement, or in defense against germs. Proteins vary in structure as well as function. They are constructed from a set of 20 amino acids and have distinct three-dimensional shapes.

In a universal phenomenon but the occurrence of polypeptides as stored food material is a less common feature compared to carbohydrates and fats. The main significance of the proteins is their role in the structural makeup of the body rather than in the yield of the energy. However, they took an important role in the production of energy by acting as a catalyst for varian metabolic processes. Further, the importance of proteins lies in their specificity in differentiating an enzyme with other or functionally as stored food material is very less yet they are important as they form the basic framework of the body. Parasitic helminthes are capable of efficient protein synthesis and incorporation of labeled amino acids into proteins has been demonstrated in several species.

The predigested food in hosts small intestine (especially ileum is the chief source of nourishment for tapeworm soluble nutrients like glucose amino glycogen and some lipid substances. The essential and non essential amino acids required proteins are also digested at the host parasites interface by the activity of photolytic enzyme and these are secreted by the cestode in tegument. Amino acid is absorbed by active transport but some amino acid tries inhibiting the uptake of others whereas some have no effect. In *Hymenolepis diminuta* there was interface by amino acid if the diet contained an incomplete protein or there was imbalance in dietary amino acids.

There are many different kinds of proteins each specialized for a different biological function moreover most of the genetic information is expressed by proteins. The term protein is derived from Greek word 'Proteios' meaning holding the first place berzeliu (Swedish Chemist) suggested the name proteins to the group of organic compounds that are almost important to life. Protein performs a great variety of specialized and essential functions in the living cells. These functions may be broadly grouped as static (structural) and dynamic.

The cestode parasites utilize the food from the gastrointestinal tract of the host so their metabolism depends on the nourishment available in the gut of the host. The occurrence of proteins is in the body of parasites. Hence, it is essential to know the methods for protein estimation from host and parasites. Therefore the present study is undertaken on methodology of protein estimation from parasites and their host tissue.

MATERIALS, METHODS AND RESULTS

For biochemical studies (estimation of Protein) of Helminth parasites, mature worms of the same size and length were chosen for sample preparation. Prepare samples from 1gm of parasite & host intestine tissue in 10ml distill water.

ESTIMATION OF PROTEIN BY LOWRY'S METHOD:

This is the best method for estimation of proteins of Parasites and its host tissue.
The following reagents are essential for protein estimation-

1. **Reagent (A):** Dissolve 100gm of Na2 Co3 in 1 liter (final Volume) of 0.5 N NaoH.
2. **Reagent (B):** Dissolve 1gm of Cuso4.5H2O in 100ml (final Volume) glass distilled water.
3. **Reagent (C):** Dissolve 2gm of Potassium tartrate in 100ml (final Volume) glass distilled water. The reagent prepared may be stored indefinitely.
4. **Folin's Phenol reagent:** (Folin ciocalteu reagent) take a 1500 ml flask. Placed 100 gm sodium tungstate, 25 gm sodium molybdate in 700ml of distilled water, 50 ml of 85% phosphoric acid, and 100 ml of Conc. HCl. reflux gently for 10 hours. Add 150 gm of lithium sulphate, 50 ml of distilled water and few drops of bromine. Mix well, cool and dilute to 1 liter and the filter. The reagent should have a greenish tint. This solution should be dilute with equal volume of distill water before used.

PROTOCOLS:

Following stepwise protocols used for this-

1. On a colorimeter to be used and allow it to warm up as directed by the manufacturer.
2. Take clean test-tubes and placed them in test-tube rack in each tube carefully pipette one of the following volumes of 0.3 mg /ml solution of bovine serum albumin: 0, 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9 and 1.0 ml.
3. Bring the total volume of liquid in each test-tube to 1 ml by adding an appropriate amount of glass distilled water.
4. Mixed thoroughly 15 ml reagent A, 0.75 ml reagent B and 0.75 ml reagent C. in a 50 ml flask.
5. Add 1 ml of solution made in step 4 to each of the tubes prepaid in step 2 vertex the tubes to mix them thoroughly.
6. Incubate the tubes for 15 min at room temperature.
7. At the conclusion of the incubation period, pipette 3 ml of Folin's phenol reagent to each tube vertex the solution immediately.
8. Incubate the sample at room temperature for 45 min.
9. Determine the optical densities at 660 m , setting to colorimeter to 0 density with blank.
10. Prepare sample from parasite & host intestine tissue (1gm in 10ml distill water) & proceed step no. 5 to 9 for protein estimation.

FORMULA:

Calculate the amount of protein in tissue using the following formula.

$$\% \text{ protein} = \frac{\text{O.D. of unknown X known protein}}{\text{O.D of standard}} \times 100 \text{ mg/gm Wet Weight.}$$

The Lowry Assay is the most widely used method for protein estimation in the world. Proteins are the most abundant organic molecules in cells constituting 50 percent or more of their dry body weight. Total protein contents in parasite tissues range between 20-80% of dry weight. Parasites

are a major concern to animal health all over the world, and of particular importance in India. Biochemical indices have been employed in effectively monitoring the responses of organisms to stressors and thus its health status under such adverse conditions. Hence, the changes associated with Biochemical parameters due to various parasites establish a database, which could be used in disease diagnosis and in guiding the implementation of the treatment or preventive measures. Hence Present study focused with methodology of protein estimation from parasites and their host tissue.

ACKNOWLEDGEMENT

Authors express sincere thanks to Principal, Yeshwant Mahavidyalaya Nanded for the facilities provided.

REFERENCES

1. Lowry O.H., Rosenburough N.J., Farr A.L. and Randall R.J. (1951): Protein measurement with folin phenol reagent. J. Biol. Chem., 193: 265-275.