Study of Protein Profile Variation in Healthy and Diseased Black Bengal Goat

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Abstract

Goat is one of the main live stock animal it has much adaptive capabilities to disease resistance. Goat mainly used as protein source which combat the malnutrition as well as employment generation in the west Bengal. Some time few microorganism can infect the goat as well as protein is lost. The role of protein, as a lipostatic signal makes it one of the best physiological markers for body weight, food intake, energy expenditure and reproduction. Information regarding molecular characteristics of protein in different animal species is scarce. With the aim of characterizing the protein of Black Bengal goat of normal and healthy at molecular level, the present study was designed to isolate goat protein from its adiposites of both normal and diseased and molecular characterization of the same, subsequently. In the present study, chemical treatment and ultra-sonication technique was used for isolating protein from goat adipose tissue. The isolated protein are subjected to SDS-PAGE from both normal (healthy) and diseased goat (PPR Infected) respectively. There were protein profile variation in both normal (healthy) and diseased goat (PPR Infected).

Introduction

In this 21st century, the aquatic and terrestrial animal, viz. fish and goat are the important protein source in Indian perspective that may be exploited to combat malnutrition in human beings. Fish and goat are often exposed to several pollutants like pesticides, heavy metal toxicants, industrial effluent as well as pathogenic microorganisms. Which affecting as well as their growth and quality of meat content. Black Bengal goat is the traditional breed of West Bengal. Black Bengal goat is a dwarf breed and famous for high fertility, prolificacy, superior quality chevron, best quality skin, early sexual maturity, resistance against common diseases, short kidding interval and very good tropical adaptability, especially in the climatic conditions of West Bengal. However, Black Bengal goats are reported to be slower in growth, low producer of milk, higher kid mortality. Black Bengal goats are reared under subsistence farming where; environmental conditions (nutrition in particular) are not

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always conducive to express genetic potentiality of these animals.72% of the population of this state lives in the rural areas. Their livelihood is characterized by small holdings or landlessness, illiteracy, unemployment and malnutrition. Many of these people resort to goat production to assist in reaching self sufficiency Therefore, goats is considered to play important role in generating employment, income, capital, storage and improving household nutrition. However, this important protein has not yet been extensively studied and in the source of cheapest protein and Black Bengal goat (Capra hircus), an important Indian live stock species, till date. Bengal goat meat are widely used as daily meal and also used in various social ceremonies of Bengal.

Material and Methods:

- 1. Collection of goat sample
- A. Normal healthy sample: Goat adipose tissues were collected from Black Bengal goat (*Capra hircus*) from local market and Universities tissue bank. Clinical samples were collect from Postmortem unit of West Bengal University of Animal and Fishery sciences, Kolkata. The samples were kept on ice and transported to laboratory for processing. The samples were stored at -20°C for extraction of protein (Fig 1).
- B. Diseased sample: Clinical samples were collected from Post-mortem Section of West Bengal University of Animal and Fishery sciences, Kolkata. Adipose tissues of PPR infected goat (Fig 2) were used as clinical samples. The samples were kept on ice and transported to laboratory for processing. The samples were stored at -20°C for extraction of protein.
 - 2. Isolation of goat protein

Fig 1: Apparently healthy black Bengal goat (*Capra hircus*) along with its kids



Fig 2: PPR virus infected goat (*Capra hircus*) showing typical oral lesions



Adipose tissues were collected from Goat (Capra hircus) and kept in deep freezer (-20° C) for future use. Total adipose tissue was thawed in 0.4 ml of cold Urea/thiourea buffer (7M urea, 2M thiourea, 4% CHAPS, 45mM Tris, pH 7.4, 60mM DTT) and complete protease inhibitors (one tablet/20 ml, Roche, Barcelona, Spain) supplemented with 0.1 mM NaCl. Adiposites were mechanically disrupted and briefly sonicated (Hielscher Ultrasonics GmBH, Germany). Samples were adjusted to 900ìl with lysis buffer (20 mM Tris, pH 7.4; 100 mM Nacl; 1% Triton and protease inhibitors) and incubated for 15 min at 35°C. After cooling on ice (10 min), 100ìl of 0.1M Tris, pH 7, and 50 mM MgCl, were added to the homogenate, which were then incubated with DNase-1 (30 U Sigma) on ice (10 min). The homogenate was centrifuged (15 min, 10000 g, 4°C) and the aqueous phase between the upper lipid phase and the lower cellular debris phase was collected. Finally the extract was separated by chloroform/methanol precipitation and dissolved in phosphate buffer solution (pH-7.4) by Peinado et al, 2010. The total protein concentration was determined using Lowry's method (1951).

3. SDS-PAGE

Crude proteins were analyzed by sodium dodecyl sulphate polyacrylamide

electrophoresis (SDS-PAGE) as per Laemmli (1970) using 12.5% polyacrylamide gel in a vertical mini slab gel electrophoretic apparatus (Attao, Japan). The samples were mixed with sample buffer in a proportion of 1:1 and subsequently the solution was heated at 100°C for 2 min. The amount of protein applied was 50ìg per track. Proteins were run at 18 mA for 150 min. The bands were visualized by staining with monochromatic silver staining (Shevchenko *et al*, 1996). Standard molecular weight marker (PMW-M, Genei, India) was run parallel along with sample proteins to determine the relative molecular weights of the polypeptides.

Results

1. protein estimation

The concentration of the crude normal or healthy adipose tissue of Black Bengal goat protein from adipose tissue was estimated as-9.5 mg/ml and adipose tissues of PPR infected goat protein concentration was found to be 5.3 mg/ml respectively.

2. SDS-PAGE profile of normal and Disease goat

The concentration of the crude protein from normal or healthy adipose tissue of Black Bengal goat and adipose tissues of PPR infected goat were clinically estimated by as- 9.5 mg/ ml and 5.3 mg/ml respectively. When the crude protein obtained from adipocyte tissue from healthy and disease goat was subjected to SDS-PAGE analysis, 65 (sixty five) polypeptides in the molecular weight range of 3- to 100 kDa were obtained in healthy adipose tissue. 25 (twenty five) polypeptides in the molecular weight range of 3- to 100 kDa were obtained in adipose tissue of diseased goats. Presence of leptin protein in only crude adipose tissue (normal adipocyte) sample whereas less abundance in adipose tissue obtained from PPR infected goats (Fig 3).

Fig 3: Polypeptide profile of crude protein of adipose tissue obtained from normal and diseased Black Bengal goat as assessed by SDS-PAGE



^{1:} Crude protein profile of normal goat 2: Crude protein profile of diseased goat

Discussion

In case of Black Bengal goat normal and PPR infected goat adipocyte were used. When the crude protein obtained from adipocyte of normal and diseased goat were subjected to SDS-PAGE analysis, 65 (sixty five) polypeptides in the molecular weight range of 3- to 100 kDa were obtained in healthy adipose tissue. 25 (twenty five) polypeptides in the molecular weight range of 3- to 100 kDa were obtained in adipose tissue of disease goat. This study varied due to variation of species and tissue in diseased and control sample.

Livestock has been an important component of domestic animals has been well documented and mixed farming system practiced in India. The Black Bengal goats are dwarf goats and are known to be famous for its adaptability, fertility, fecundity, delicious meat superior skin, extreme disease resistance and wide range of acceptability under adverse agroclimatic condition.(FAO.2008). According to the current study, the protein concentration values of normal Black Bengal goat are

^{3:} Standard molecular weight marker

supported by the Kaneko et al, 1997.

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