

***Effect of Chemically Treated of Sesame seed
Cake on Dry Matter and Crude Protein
Degradation***

Submitted by:

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DEDICATION

To my dear father, to my dear mother.

To my brothers and sisters.

To all my family.

To all my friends.

Colleges.

With love and respect.

AKNOLWLEDGEMENT

Iam Indebted to many friends and colleagues for help and advice.

I would introduce my special grateful to Dr. Ahmed Joffon Mahala, who read through various sections of the manuscript and offered constructive criticism, and then I thank Prof. Shadia Abdalati in University of Sudan, who gave un-stinting assistance during this study

I would like to express for my sincere thank to members of the Animal Production College, University of Khartoum, who were always willing and helpful.

Finally I would like to express gratitude to my parents, sister, brothers, and friends for them in loving and dedication.

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ABSTRACT

Nitrogen disappearance of Sesame cake (SSC) treated with 1) ethanol 50%, 2) ethanol 70% at 78°C and 3) tallow 10% was evaluated. Rumen degradability was carried in 3 fistulated bulls fed sorghum bicolor as basal diet with 2kgs supplements and salt lick stone, three bags (a bag in bull at a time) for each treatment were incubated for 0, 3, 6, 12, 24, 36, or 48 hours.

In situ crude protein and dry matter, potential degradability (PD), soluble fraction (a) and effective rumen degradability (ED) for treated sesame cake were significantly ($P < 0.05$) lower when compared with untreated sesame cake, which might be due to reduction in nitrogen solubility.

The maximum reduction of CP degradability as an average was reported in ethanol 50% followed by ethanol 70% and tallow respectively. Ethanol treatment protected (SSC) protein from being degraded by micro flora, and would be expected to improve nitrogen utilization.

المستخلص

تم تقييم النيتروجين الظاهري لامباز السمسم (1 غير المعامل 2) والمعامل بايثانول تركيزه 50 % (3) تركيزه 70 % في درجة حرارة 78°م و4) المعامل بالشحم بنسبة 10 % .

أجريت تجربة التكرارية للبروتين الخام والمادة الجافة في ثلاثة ثيران ذات ناسور كرشى غذيت على قصب الذرة كعليقة اساسية مع امدادها ب2 كجم عليقة مركزة و حجر اللحوس . ثلاثة اكياس (كيس/ ثور / فترة زمنية) في كل المعاملات ثم تم تحضيرها في الكرش للفترة الزمنية التالية صفر ,3, 6, 12 , 24 , 36, و48 ساعة

في اكياس النايلون البروتين الخام والمادة الجافة قورن جهد القابلية التكرارية (a) والجزء الذائب من الغذاء (b) وفاعلية الكرش التكرارية () لعينات السمسم المعاملة كانت معنويا () اقل عندما قورنت مع العينات الغير معاملة ويعزى ذلك لنقص زوبانية النيتروجين.

أقصى أختزال في تكسرية البروتين الخام كمتوسط , في المعاملة بالايثانول 50 % يليه المعامل بالايثانول 70 % والمعامل ب10 % شحوم.

معاملة أمباز السمسم بالايثانول تحمي البروتين الخام من التكرارية بواسطة الميكروفلورا في الكرش. ومن المتوقع أن يحسن في أستهلاك النيتروجين.

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INTRODUCTION

Diets of high producing ruminants are based on inexpensive and abundant supplies of cereal grains and oilseed meals (Zinn *et al*, 1981) Sesame cake protein serves as source of metabolizable protein to the ruminant by providing both ruminal undegradable protein for microbial growth and some ruminal undegradable protein for intestinal digestion. Crude protein content of sesame cake very well satisfies the animal requirements in many production systems.

The high solubility mainly due to non protein nitrogen and rate of rumen degradation of sesame cake result in excessive amount of ammonia production in the rumen, some of the ammonia produced in the rumen is utilized by the rumen bacteria for microbial protein syntheses. The remainder of the ammonia is either recycles and or excreted as waste. High producing animals can meet their protein requirements with microbial protein synthesized in the rumen, remainder of the required protein must be supplied by dietary protein that escapes ruminal degradation.

Several chemical and physical treatment of feedstuff have been describe in the literature to reduce the rate of ruminal protein degradation and increasing the proportion of protein that escape degradation in the rumen.

Exposing feedstuff to high temperature may cause over heating which is result in decreased digestibility of feed stuff.

Chemical treatment for the feed may be decrease the degradation of ruminal protein. The objective of this study was to estimate the effect of chemical treatment of sesame cake protein on nitrogen disappearance and rate of degradability.

CHAPTER TWO

LITERATURE REVIEW

2.1. Sesame seeds (*Sesamum indicum*):

Sesame seedcake is one of the chief sources of vegetable oil and protein, even though many varieties are known to commercial varieties white and black are cultivated in India on a large scale. Sesame seed contain about 62% oil also contain about 20% -22% vegetable protein mainly globulin, which has properties similar to vegetable casein and may be applied as a plastic and adhesive. White and black sesame have been analyzed for the total carbohydrates composition. Sesame cake seeds contained D-glucose 3.36% , D-glactose 0.40 % D-fructose 3.43% ,sucrose 0.17% and sesamose 0.14 % (William. *et al* 2003)

There are some reports on the nature of free sugars in accruing sesame but the poly sacharides have not been investigated. (William. H., *et al* 2003)

In Sudan the production of oil seed cake (1383tons) as follow 738t sunflower seedcake (SFSC), 313t sesame seed cake (SSC), 307t groundnut cake (GNC) and 25t cotton seed cake (CSC). (Maragan, 1996)

The extraction rate was 60, 50, 50 and 78% for SFSC, SSC, GNC and CSC, respectively, they are considered rich in protein (200-500g/Kg) and thus for a valuable food for animals as 950g /Kg of the N in oil seed meals is present as true protein (McDonald *et al* .,1996)

Little information is available on the quality of sesame cake in Sudan also little information about the outcome of feeding these material to ruminants.

2.2. Importance of protein:

Thousands types of protein present in the animal body they perform function too numerous to list. These functions include serving as carriers of vitamins, oxygen and carbon dioxide pulps structural kinetic, catalytic and signaling roles. However, it is not surprising that dire consequence can raise from mutation either that encode protein or in regions of protein (Robert, 1999).

Protein is critical nutrient for growing animals, it is more expensive than energy feeds, optimal diets from plant source. Due to the escalating price of plant protein non conventional protein sources needed to be investigated (pond *et al*, 1995)

2.2.1 The nutritive value of sesame cake:

Sesame meal or cake is a high protein concentrate containing about 46% crude protein, rich in arginine and lucien but low in lysine and methionine and may be used in feeding for animals in the same way as groundnut meal (Mc Donald *et al*, 1981).

The chemical composition of sesame seed cake varies according to the method of processing the sesame seeds (mechanical or solvent extraction)

The chemical composition dry matter (DM) was 83 to 96%, crud protein (CP) , ash , ether extract (EE) , nitrogen free extract (NFE) and crud

fiber (CF) are 23-46% ,7.5-17.5%,1.4 -27 % , 25 -31% and 5-12% for these nutrient , respectively (FAO, 1990).

Sesame press cake represents an important protein for human consumption. Some of the limiting factor was its high crude fiber content, and its bitter taste (Greed *et al*, 1987)

For ruminants the protein has a degradability of 0.65 - 0.75 it depending upon the rate of passage through the rumen (Mc Donald *et al* 2002)

The cake or meal if consumed in excessive amounts will increase milk fat and disagreeable flavor to milk may occur because the oil of meal or cake is highly unsaturated. The hulls of sesame seeds should be decorticated before process to avoid toxicity as it contains oxalates. Meals in good condition are palatable but have a laxative action. The diets of young ruminants should not contain more than 50 Kg /t, where as the maximum inclusion rate for adults is 100-150Kg/t, the meal should not be given to young pigs or poultry but may be included at upto50Kg/t for adults of these species (Mc Donald *et al*, 2002).

2.3 Rumen environment:

The ideal environment for maintaining stable microbial population on the adult ruminant animals is warm 39° - 42°c and anaerobic, chemically reducing environment. The microbial ecosystem in the rumen is vary within the animal with time after feeding, between days in the same animal and apparently in the animals of different countries fed similar diet for growth of bacteria, protozoa and possibly other microbes that produce cellulase enzymes (Hungate, 1975)

The pH of the rumen content is approximately 5.5-6.5 this is held relatively constant by the buffering action of the large amount of secreted saliva which is containing a high in sodium and potassium bicarbonate and urea, by absorption through the rumen wall in to the blood stream of volatile fatty Acid (VFA) and by ammonia (NH₃) produced during fermentation Type of microorganism that can exist in rumen are constrained by temperature, oxidation, reducing potential and PH those include bacteria, protozoa and fungi. Generally, proteins break down by the virtual enzymes. These enzymes secreted originally in the body these enzymes include pepsin, trypsin, carboxypeptidase, dipeptidases and amino peptidase, some origin secreted enzymes such as gastric mucosa, pancreas, and small intestinal. This enzyme converts proteins to polypeptides and amino acid to utilize it. Large amount of bacterial protein and vitamins are synthesized in the large intestine (Lenord *et al*, 1979).

Preston and Leng (1987) noted that ruminal environment appear to be controlled by type and quantity of food eaten, periodic mixing salvation rumination diffusion of secretion into the rumen absorption of nutrient from the rumen and passage of material down the digestive tract.

2.3.1 Rumen microorganism:-

The composition of rumen microbial cells is relatively constant .Atypical analysis are True protein 32-42%, Small nitrogenous molecules10%, Nucleic acids 8%, Lipids 11-15 %, Polysaccharide 17%, Ash 13 %. High lipid content have been noted in bacterial dry matter isolated from the particle associated phase of rumen content and from rumen where the microbial modifier rumen has been fed to animals (Leng, 1997)

According to Oigimoto and Iami, (1981). The rumen bacteria cell of rumen content and the majority of them are obligated. The classifications of the bacteria include the following types:

- cellulolytic bacteria
- Hemicellulolytic bacteria
- Amylolytic bacteria
- Sugar utilizing bacteria
- Proteolytic bacteria
- Methanogenic bacteria
- Lipolytic bacteria
- Vitamin synthesizing bacteria
- Acid utilizing bacteria

This classification depends on their utilization of carbohydrate, protein, lipids, methane production and ammonia producers.

Generally the total rumen microbial population was influenced by the type of physical form of diet, water intake and in frequency of feeding. More over than added that a diurnal variation is most obvious with rumen protozoa followed by bacteria, as protozoa population are highest immediately after feeding and decline for short time, mainly due to dilution of refeeding (Leedle *et al*, 1982).

Many factors affect microbial growth efficiency and the protein relative to volatile fatty acid and available for digestion and absorption

include a deficiency of any microbial growth factor in the feed or induced some time after feeding in rumen liquor because of rapid absorption of the nutrient and the relative amounts of carbohydrate and protein that are fermented, a high protein to carbohydrate ratio in the diet can lead to a relatively low microbial protein to volatile fatty acid ratio in the end products of fermentative digestion where the dietary protein is easily and rapidly fermented in the rumen (Leng 1993).

2.4 The protein:

Proteins are complex organic compounds of high molecular weight, they contain carbon, Hydrogen, oxygen, nitrogen and sulphur in addition some proteins contain phosphorus, iron, zinc, and copper. Proteins are found in all living cells and each species has its own proteins and a single organism has many different proteins (McDonald, 1981).

Proteins are made up of amino acids, which are produced when proteins are hydrolyzed by enzymes, acid or alkalis. Amino acids are characterized by having a basic nitrogenous group amino group – NH₂ and acidic carboxyl unit – COOH, the general formula of amino acid. Proteins are built up from amino acids by linkage between the α -carboxyl of amino acid and the α -amino group of another acid, this is known as the peptide linkage. Proteins are classified into three main groups according to their shape, solubility and chemical composition, the first one:-

➤ Fibrous protein: - Are insoluble proteins which are resistant to animal digestive enzymes. This group contains collagens (the main proteins of connective tissues).

- Globular protein: - This includes all enzymes, antigens and hormones, its divided into albumins which are water soluble and heat coagulable they are found in milk, eggs, blood and plants, globulin, lacto globulin, histones which are basic protein in cell nucleic they are also associated with DNA.
- Conjugated proteins: - such as glycoprotein, which are conjugated protein with one or more heterosaccharids, they are components of mucus secretions, which acts as lubricants in many parts of the body.

Also lipoprotein conjugated with lipids such as lecithin and cholesterol. Chromo proteins contain pigment like hemoglobin, haemocytinine, cytochrome and flavoprotein, Nucleoproteins are high molecular weight compound (Mc Donald *et al*, 2002).

2.4.1 Source of protein for dairy cows:

Protein sources are divided on the type of nitrogen, they provide:-

(a) Source ruminal degradable protein (RDP):- The sources of ruminal degraded proteins are oil seed meal such as soybean, in the Sudan the major source is groundnut cake. Oil seed meals are the only source of supplementary protein due to their high palatability.

(b) Sources of ruminal undegradable protein (RUP)

This source of protein are slowly degraded in the rumen by microbial proteolyses enzymes

So large protein of RUP escapes ruminal degradation and becomes available for enzymatic digestion in the small intestine. The main sources of RUP are animal protein such as meat meal, bone meal, fish meal and feather

meal. In addition some heated plants sources like heated oil seeds. Sources of RUP have disadvantages because of their low palatability and they are expensive. Some of important protein definition that is commonly used in ruminant nutrition:-

- Crude protein (CP) :-

Crude protein measures total nitrogen required by the animal from the fodder nitrogen present as protein and the method used to measure CP is Kjeldahal

- Soluble protein (SCP):-

Is the CP which is soluble in buffer solutions, water or a rumen fluid, chemically measured as protein soluble in borate phosphate buffer (PH 0. 9)

SCP contains all the non-protein nitrogen and some true protein. It is degraded by rumen microbes rapidly. The source of SCP is young forage, silage, legume seed and oil seed.

- Non protein nitrogen (NPN):-

It is the nitrogenous compounds, which do not have the complex structure of a true protein. It includes ammonia, small peptides, free amino acids, amines and amid, NPN is rapidly converted to ammonia in the rumen.

- Ruminant undegradable protein (RUP):-

It the dietary protein which is not degraded by the rumen microbes or it the dietary protein that is resistant to microbial attaches in the rumen, in other terms it named bypass protein, it is estimated by in vivo, Invtro and

insitu methods, the insitu method is the common method used to estimates RUP.

- Ruminal microbial protein :-

It refers to the proportion of protein synthesized by rumen microbes, rumen microbes utilize ammonia amino acid and peptides to synthesize microbial protein, 80% of the microbial protein are digestible by the animal.

- Metabolizable protein (MP) :-

It is net quantity of true protein (microbial protein) or amino acids (feed protein) absorbed in the small intestine.

2.4.2 Protein digestion in ruminants:

Proteins are fermented to VFA, methane, carbon dioxide, and ammonia (end product), peptides and amino acids are intermediates and used by rumen microorganism to synthesized microbial cell. Ammonia is either absorbed dietary across the rumen wall or passes out of the rumen with the fluid phase of digesta or is incorporated into microbial protein. The dietary protein which is not totally degraded passes into the abomasums and duodenum and is digested by enzymes hydrolysis (Kempton *et al*, 2008) feed protein is usually hydrolyzed in the rumen rapidly, the breakdown of protein depend on a number of factors such as the nutritive value of protein. Chalmers and Synge (1954) investigated the factor of protein solubility, for example some (pure) soluble proteins are broken down more slowly than insoluble proteins, depending on the degree of secondary and tertiary structure also the nature of the diet has a major influence on the proteolysis activity of rumen content. Rumen microorganism which hydrolyzed, protein

named proteolytic microorganisms, and include ciliate protozoa. The acids produced by fermentation are theoretically capable of reducing the pH of the rumen liquor to 2.5-3.0 but under normal conditions the pH is maintained at 5.5 – 6.5. Phosphate and bicarbonate contained in the saliva act as buffers. In addition, the rapid absorption of the acids helps to stabilize the pH (McDonald *et al*, 2002). The rumen has a buffering action by the large amount of secreted saliva which is high in sodium and potassium bicarbonate (Ensiminger *et al*, 1990).

Degradability of protein refers to the rate and extent to which a feed protein may be broken down in the rumen. (Ensiminger *et al*, 1990). The rate of digestion of feed, particularly roughage feed and rate of passage of residues from the rumen, are important determinants of voluntary feed intake and productivity. Consequently, considerable emphasis is placed on maintaining optimal conditions in the rumen to maximize microbial growth and digestion (Leng, 1993).

Over-protected proteins are neither fermented in the rumen nor digested in the small intestine (Smith *et al.*, 1980). Microbial, dietary and endogenous proteins leaving the rumen are subjected to digestion and absorption in the small intestine. Any protein leaving the small intestine may be fermented by microorganisms in the caecum and colon or excreted in the feces but it is generally believed that the microbial protein produced in these organs is not available as amino acids to the animal.

The factors that influence the absorption and supply of amino acids to the tissues of ruminants are therefore complex (McDonald *et al.*...1979).

2.4.3 Ammonia metabolism:

The main product of amino acids deamination is ammonia which is used by structural carbohydrates bacteria as a nitrogen source for microbial protein synthesis in the rumen. Tran's protein the cell membrane is the first step in ammonia uptake. Glutamate is the first amino acid into which ammonia is metabolized. Synthesis of amino acids from available energy and carbon source occur, when nitrogen has been fixed into an appropriate compound such as glutamic acids. Ammonia in excess of microbial protein synthesis is converted to urea in the liver and recycled via saliva (McDonald *et al*, 1981)

2.4.4 Metabolism of urea:-

Urea is broken down in the rumen to ammonia by urease enzyme, this activity is come with microbial synthesis from ammonia enables ruminants to utilize urea entering the rumen either with food or in salivary secretion.

Recycling of blood urea to the rumen allows ruminant animals to survive on diet very low in nitrogen. The amount of blood urea recycled to the rumen depends on the ammonia concentration in the rumen and plasma urea concentration which enters the rumen with the saliva or by diffusion through ruminal wall. Microbes adhering to the ruminal epithelium have the ability to produce urease. The enzymes hydrolyze urea to ammonia and CO₂ –recycling urea is reduced by inhibiting urease activity when the ruminal ammonia level is high (McDonald *et al*, 2002).

2.4.5 Protein digestion in the small intestine:

Digestion of protein in the abomasum and the small intestine in ruminants is similar to that in monogastric animals, so the digestion of protein in the abomasums is carried out mainly by pepsin in a very acidic environment about PH. Optimal activity of trypsin, chemotropisin and carboxypeptide occur in middle jejunum, peak activity of exopeptidase and dipeptidases occur in midilium (McDonald *et al*, 2002)

2.5. Protein degradation:

Rumen degradation of proteins depends on the nature of feed. Food proteins are hydrolyzed by the rumen microorganism into peptides and amino acids or ammonia, volatile fatty acid, carbon dioxide and other materials (McDonald *et al*, 1996).

Degradation of protein in the rumen is important to determine the supply of nitrogen microbes and protein available for digestion in the small intestine (Miller. 1973).

Evaluation the quality presented to the small intestine for absorption is the sum of the microbial cell protein and feed protein that has escaped or bypasses ruminal digestion (Garray, 2005).

2.5.1. Methods of protein protection:-

Various methods have been used for protecting proteins from microbial degradation including the simple application of heat and chemical agents such as formaldehyde (Reis and Tunk, 1969), alcohol (Vande Aar *et al*, 1982), bentonites (Britton *et al*, 1978), zinc (Britton and Klopfenstein, 1986), tannins (Driedger and Hatfield, 1972), and sodium hydroxide (Mir *et*

al, 1984), all have been used successfully to treat protein as a mean reducing ruminal degradability.

All these methods of treatment, including heating, are through to act either by inhibiting proteolyses activity and or by modifying protein structure in such the number of proteins specific bonds that can be cleaved by microbial enzymes are decreased.

2.5.2. Heat Treatment:-

Heat treatment long has been recognized as effective way to reduced degradation of the protein in the rumen (Tagari *et al.*, 1962)

An increase in temperature at coagulation may increase the amount of protein that escapes ruminal degradation. Consequently, the amount of amino acids that would be available for absorption in the small intestine may be increased (Nishimuta *et al.*, 1974)

The most successful physical treatment has been heat. Heat facilitates the Millard or non enzymatic browning reaction between the sugar aldehyde groups and the free amino acid groups of protein to yield an amino-sugar complex (Grffin *et al*, 1993)

Heat treatment of feed stuffs can decrease proteolysis by blocking reactive sites for microbial proteolysis' enzymes. Heat has been used to decrease the supply of dietary protein the duodenum (Shezana *et al*, 2007).

Heat treatment of protein for the purpose of protecting it from degradation in the rumen has been approached through oven heating, roasting, extruding and autoclaving. Despite numerous attempts to heat – treat protein, little efforts has been made to relate systematically the extent

of protein degradation to temperature and length of heating (Tagari *et al*, 2002).

The heat treatment is the most economical process, and is the one with which the best results have been obtained in practice (Ali, 1983).

Heat treatment protects dietary protein for ruminants, but it is important that appropriate temperature and heating times are employed for particular feeds. However, the optimal conditions are often not known (FAO, 2003).

The temperature may affect the seeds not just chemically influenced (Reboller and Bals, 2001)

Oil seed protein sources are the most economical to treat with heat (Lyle, 1997).

The effect of heat –treated sesame meal was conducted by Tin Nagwe, Mar Mar kyi, Aung Aung and Ni Ni Maw (2002). in their experiment, the sesame meal were subject to heat treatment by placing them for two hours in a hot air oven maintained at a temperature of 150c and then cooling to room temperature, they expressed that the TDN%, DM were 59.6% and 61.6% for RSUS (rice straw+ untreated sesame meal) and RSHS (rice straw + heat-treated sesame meal) respectively and the value of RSHS was significantly higher ($p < 0.05$) than that of RSUS although there were no significant differences between them. In their experiment they also recommended that the total nitrogen intake for RSHS was 67.9/d and relatively higher than that of RSUS.

Sesame cake protein had a higher degradable protein when comparing to another source of protein, the fermentation characteristic of sesame cake protein (a), (b) and (c) values were 77.3, 26.8 and 0.07 respectively. (Mahala and Gomaa, 2007)

2.5.3. Formaldehyde Treatment:-

Formaldehyde reduce protein degradability by forming cross links between protein chains and has anti microbial properties that may alter the bacteria population and fermentation pattern (Woolford, 1975).

Formaldehyde (1g/100g crud protein) treatment reduced protein degradability of groundnut cake (GNC), gingelly (sesame) cake (GSC) and rubber seed cake (RS) by 69.48 and 35%, respectively, at an outflow rate of 0.04/h (Sampath and Sivaraman, 1987). Formaldehyde treatment of Lucerne partly protected plant proteins against microbial degradation during in vitro liquor digestion and so reduced ammonia nitrogen concentration in the supernatant (DC Broun and SC Valentine). Formaldehyde treated sesame meal as a main source of protected protein it was concluded that treated sesame meal decreased organic matter and protein degradability as well body weight losses and blood urea concentration. Moreover, there were slightly responses in milk production when treated sesame meal was added to the supplements (Apricio, Rafael, Maria, 1992).

2.5.4. Sodium hydroxide (NaOH) treatment:-

The use of NaOH to protect proteins from degradation has shown potential (Mir et al., 1984). Kategile and Federicen (1979) reported in vivo organic matter digestibility of 10% sodium hydroxide –treated maize cobs to be 32.4%. Nagole *et al* (1985) reported that treatment of maize cobs with

4.5% sodium hydroxide increased in vivo dry matter degradability from 44.7+1.6 to 54.2+2.0%. Sodium hydroxide treatment significantly ($p < 0.01$) improve the 48h DMD values of maize cobs. The highest level of treatment resulted in the highest DMD values. Feeding 2kg/day wheat treated with sodium hydroxide (NaOH) did not affect negatively the main parameters of rumen fermentation, i.e. pH, short chain fatty acid (SCFA) production, and microbial activity. (Schmidt *et al*, 2006). Alkaline hydrogen peroxide treatment has affected on physical and chemical properties of rice straw. The urea and Ca (OH) ₂ treated straws have the same lignin content as the untreated but showed significantly decreased NDF and enhanced organic matter (OM) degradability. This can be attributed mainly to the effect of Ca (OH) ₂ treatment in breaking specific lignin carbohydrates linkages (Mao Huaming and Feng Yanglian 1999).

2.5.5. HCL treatment:-

Treatment of soybean meal (SBM) by HCL protected the crude protein and amino acid (AA) from ruminal degradation, increasing rumen undegradable protein from 42% to 86%. This study showed that, based on in situ measures

Chemical treatment of soybean meal enhanced AA availability, and had a higher potential to enhance the AA supply to the small intestine of high-producing dairy cows. (Borucki Castro *et al* 2007).

However, tannic acid addition decreased the protein, of crude protein solubility at 0 times and increased the potential degradable fraction of CP (Santos *et al* 2000). However, including malic acid in the diet did not have an important effect on the degradation of dry matter and fiber fraction lucern

hay and the parameters of ruminal fermentation. (Mahfoud., *et al* 2006). Rioux *et al* (1995) who observed birds' foods trefoil cultivars with greater tannic concentration had lower soluble CP at 0 time and greater potentially degradable fraction of CP. A lower rapidly soluble fraction of CP and greater potentially degradable fraction of CP following tannic acid addition could be beneficial to the animal because it would decrease the excess of N in the rumen after feeding and maintain a more uniform availability of nitrogen.

2.5.6. Alcohol treatment:-

Diets of high producing ruminants are based on inexpensive and abundant supplies of cereal grains and oilseed meals. Soybean meal (SBM) is widely used as a protein supplement in ruminant diets. Utilization is inefficient, 60 to 80% of the soybean protein is degraded in the rumen (Satter, *et al.*, 1977)

(Zinn, *et al.*, 1981) because young growing ruminants and lactating dairy cattle can only partially meet requirements with microbial protein synthesized in the rumen (Chalupa.1975). The remainder of the required protein must be dietary protein that escape ruminal degradation and is digested and absorbed in the small intestine. This can be accomplished by using feeds containing protein resistant to ruminal degradation, such as blood meal or dehydrated alfalfa (Loerch., *et al.*, 1983, and Stock, R., and Klopfenstein,1979) or by processing existing feedstuffs to reduce ruminal protein degradation.

Treatment of oilseed proteins with an aqueous alcohol solution produces a permanent change in three dimensional structure of the protein

molecule (Fukushima, 1969) When oilseed proteins are placed in an aqueous alcohol solution, water molecules disrupt the hydrophilic outer region of the protein molecule. This allows alcohol molecules to enter the interior of the molecule where they disrupt the bonding of hydrophobic amino acid chains.

Treatment of (SBM) with ethanol or propanol at room temperature reduced *in situ* N disappearance (Van der., *et al.*, 1982) and resulted in decreased ruminal ammonia (Lynch *et al.*, 1987) and increased N retention in lambs (Van der., *et al.*, 1982). The lipid coating of oilseed proteins (Peterson *et al.*, 1975 and Glenn *et al.*, 1977) reduced ruminal ammonia and increased N retention in lambs compared with those fed untreated oilseed meals. Chemical bonding between denatured vegetable proteins and lipids has been observed (Kamat., *et al.*, 1975). The resulting material improved stability in aqueous solution, which may render the protein more resistant to ruminal degradation.

Various lipids have been used to coat feed proteins (Preston *et al.*, 1975) and amino acids (Sibbald 1968) to protect them from ruminal degradation. Glenn *et al.*, (1977). An interaction between vegetable proteins and lipids was reported by Kamat *et al* (1978) when native soybean proteins were denatured with urea, the proteins were able to bind with lipids and form a lipoprotein- like complex the resulting product was more stable in aqueous solution, it was theorized that SBM proteins denatured number of ionic and hydrophobic residues to which lipids could bind.

Lynch *et al* (1987) found reductions in *in situ* N disappearance when comparing SBM treated with 40% ethanol at room temperature to untreated SBM because alcohol treatment at higher temperature reduced N

solubility (Lynch *et al* 1987), tallow reduced N disappearance at 12 h and reduced rate of N loss, no additive effect was seen when SBM previously treated with ethanol at 78°C was treated with either lipid.

Treatment of SBM with ethanol, especially at elevated temperature, appears to have potential to reduce ruminal degradation of soybean protein. Alcohols have the advantage of being readily incorporated into existing processing methods for soybean (Lynch *et al.*, 1987).

The values recorded for dry matter degradability, the degradation rate, soluble nitrogen, insoluble nitrogen and effective protein degradation of sesame cake are 0.20 ± 0.05 , 31.79 ± 11.34 , 66.06 ± 11.66 respectively concurred with Walli *et al.*, (2000), Haldar and Rai (2002) but differed from Walli *et al.*, (1999). The variable results were attributed to the animal factors like rumen environment, retention time and flow rate, reported by Vinil *et al.*, (2008).

CHAPTER THREE

MATERIALS AND METHODS

3.1. Animals and feeding:

Three castrated calves from a local breed (Kenana) aged 2 - 2.5 years were fitted with rumen cannulae as described by Brown *et al.* (1986). They were maintained with a well balanced ratio of concentrates and roughage. They were fed twice daily.

3.1.1. Sample collection:-

Sesame seed cake was obtained from local mill in the Khartoum state (Sudan).

3.2. Chemical treatment of sesame seed cake:-

Untreated sesame seed cake was oven dried (T1): 800 g sesame seed cake was heated to 78°C in heater prior to its addition to a vessel containing 2 liter of ethanol 50% (vol/vol) and named (T2) and/or ethanol 70% (vol/vol) and then named (T3) at 78°C. The treated sesame seed cake was mixed periodically and kept at 78°C for 1 h. after which it was strained through cheesecloth and dried oven at 50°C, and 100g of Tallow a mixture with 10 ml of Isopropanol as an organic solvent to making Tallow solution and then added to 1000g of sesame seed cake resulted in 10% tallow for the treatment and named (T4), mixed for 1 h and then air dried.

3.2.1. Chemical Analysis:-

Samples of sesame cake was analyzed for dry matter (DM), ash, ether extract (EE) and crude protein (CP), crude fiber (CF), and nitrogen free extract(NFE) according to equation, $NFE = DM - (CP\% + EE\% + ash\% + CF\%)$ using the standard procedures of the Association of Official Analytical Chemists (A.O.A.C., 1990). ME calculated according to equation of Qrskv and McDonald (1979).

$$ME (MJ/KgDM) = (0.018CP + 0.0315EE + 0.0163NFE + 0.0149CF)$$

3.2.2. Ruminal dry matter (DM) and crude protein (CP) Degradability:

According to the polyester bag technique of Mehrez and Qrskov (1977), the bags were prepared from nylon material of length 15.5cm, with 8.5cm and weighing 5g. They empty bags were individually weighed and their weights recorded. Five grams of treated or untreated cakes were put in a bag tied with a nylon ribbon, attached to a plastic tube, of 45.5cm length, 0.8cm diameter, and introduced inside the rumen. The bags (3bags/animals/period/treatment) were incubated for 3, 6, 12, 24, 36, or 48 hours each.

3.2.3. Calculation of ruminal degradability:-

Degraded dry matter percentage was calculated according to the formula:

$$\text{Dry matter loss} = \frac{(\text{Wt. of incubated sample} - \text{Wt. of residue after incubation}) \times 100}{\text{Wt. of incubated sample}}$$

Residual samples after incubation for each period were separately mixed, pooled and made ready for CP content determination (AOAC, 1980).

Degraded protein was calculated according to the formula:

$$\text{Dry matter loss} = \frac{(\text{Wt. of incubated sample} - \text{Wt. of residue after incubation}) \times 100}{\text{Wt. of incubated sample}}$$

The degradation kinetics of the incubated cake (treated or untreated) was described by curve –linear regression of DM or CP loss from the bags with time by the equation Oraskov and McDonald (1979).

$$P = a + b (1 - \exp^{-ct})$$

Where:-

P= potential degradability (percentage).

a = the soluble fraction (percentage).

b= the potentially degradable fraction (percentage).

c = the rate of degradation of b (percentage/hours).

t =time (hour).

Effective degradability (Ed) of DM and CP was determined, at 0.02, 0.05, and 0.08 ruminal outflow rates, using the equation of Oraskov and McDonald (1979) stated above.

3.3. Statistical analysis: -

The data obtained were subject to one way analysis of variance to examine the effect of the treatment on DM and CP degradation kinetics. Significant differences among the treatment were determined using least significant differences (LSD) test according to Gomez and Gomez, (1984). The Statistix8 computer program was used for the analysis.

CHAPTER FOUR

RESULTS

The result of chemical composition of sesame cake of crude protein (CP), dry matter (DM), crude fiber (CF), ash (ash), ether extract (EE) and nitrogen free extract (NFE) were illustrated in Table (1)

4.1. Dry matter (DM) degradability:-

In situ dry matter degradability of treated and untreated S. S. C is shown in Table (2). The soluble fraction (a) of dry matter (DM) for untreated sesame cake meal was recorded significantly different ($P < 0.05$) higher level (15.5) than treatment with ethanol 50% and ethanol 70% which recorded 8.5 and 9.9 respectively, while Tallow 10% treatment closed value 5.2.

Potential degradable fraction (b) did not vary between treated and untreated sesame seed cake to never among to relative significant different ($P < 0.05$), but ethanol 70% was the highest value was observed in ethanol 70% treatment, ethanol 50%, Tallow 10% and untreated sesame cake meal recorded the lowest value.

PD was significantly ($P < 0.05$) different among the treatments, untreated cake was recorded 93.7 which considered as high level, ethanol 70% recorded 90.2, ethanol 50% and tallow was recorded a lower level 84.4 and 84.6 respectively.

Fraction (c) was significantly ($P < 0.05$) different untreated and tallow 10% recorded 0.07 each. Ethanol 50% and ethanol 70% were recorded 0.04 each.

Effective rumen degradability (Ed) at rumen out flow rate (0.02/h) was significantly ($P < 0.05$) difference. Untreated was recorded a high level which was 77.7, ethanol 70% and tallow 10% recorded 65.2 and 66.4 respectively and ethanol 50% recorded low level 59.9.

Ed at (0.05/h) was significantly ($P < 0.05$) different untreated was recorded 63.4, ethanol 70% and tallow 10% recorded 48.8 each, ethanol 50% recorded 42.5.

Ed at (0.08/h) was significantly ($P < 0.05$) difference untreated and ethanol 50% recorded 54.6, 33.6 respectively, ethanol 70% and tallow 10% recorded 40.5 and 41.6 each.

4.2. Crude protein (CP) degradability:

The in situ crude protein degradability of treated and untreated sesame cake meal was shown in Table (3).

The result showed that the soluble fraction of feed (a) was significantly ($P < 0.05$) different between untreated sesame cake meal and treated with ethanol 50%, ethanol 70% and tallow 10%. The data recorded that the untreated was 42.4, ethanol 50% 31.3 ethanol 70% 40.2, and 37.5 for tallow 10%. Despite the non significant difference between ethanol 70% and tallow 10%, the treatment were show significant differences with ethanol 50% (31.3)

For potential degradable fraction (b) the untreated sesame cake, sesame cake with ethanol 50% and tallow 10% did not show a significant ($P < 0.05$) differences. While the sesame cake which was treated with ethanol 70% show a significant with other treatments

c did no significantly ($P < 0.05$) difference with untreated sesame, sesame cake treated with ethanol 70% and tallow 10% which their values were 0.075, 0.075, 0.082 respectively, but ethanol 50% was differ ($P < 0.05$) significantly recorded 0.043.

Pd the potential degradability was showed a different result that all treated sesame cake ethanol 50%, ethanol 70% and tallow 10% did not show a significant ($P < 0.05$) differences in which their values 85.3, 87.4, 89.4, In which that the untreated sesame cake was differ significantly from the other treatments which was registered the highest value 92.5.

Ed 0.02 the effective rumen degradability at the rate 0.02 was registered a significant ($P < 0.05$) differences between all groups in which their values were 83.5 for untreated sesame cake meal and 68.1, 77.1, 79.2 for ethanol 50%, ethanol 70% and tallow 10% respectively.

Ed at flow rate 0.05 was significantly ($P < 0.05$) differences between the untreated sesame cake which was 75.4, and ethanol 50% which was 56.2, the other two treatments did not differ significantly in which their values were 68.2 for ethanol 70% and 69.6 for the tallow 10% these two treatments were significantly differ with the untreated sesame and the sesame cake treated with ethanol 50%.

Ed 0.08 show the same result, that the untreated sesame cake and the treated with ethanol 50% were differ ($P < 0.05$) significantly in which their

values 70.5, 50.1 while the treatment with ethanol 70% and tallow 10% did not register a significant ($P < 0.05$) differences their values were 62.7 and 63.7 respectively these two values were different significantly from the others two treatments.

Table, (1) Chemical composition of Sesame Seed Cake

DM%	EE%	CP%	CF%	Ash%	NFE%	ME(Mj/KgDM)
93.09	5.25	48.65	14.12	12.35	12.72	1.46

Table, (2) Insitu dry matter DM degradability for treated and untreated sesame cake.

Treatment	a	b	C	Pd	Ed0.02	Ed0.05	Ed0.08
T1	15.5 ^a	78.2	0.07 ^a	93.7 ^a	77.7 ^a	63.4 ^a	54.6 ^a
T2	8.5 ^b	75.9	0.04 ^b	84.4 ^b	59.9 ^c	42.5 ^c	33.6 ^a
T3	9.9 ^b	80.3	0.04 ^b	90.2 ^a	65.2 ^b	48.8 ^b	40.5 ^b
T4	5.2 ^c	79.3	0.07 ^a	84.6 ^b	66.4 ^b	48.8 ^b	41.6 ^b
SEM	1.36	5.39	0.55	2.40	0.89	1.42	1.6
L.S	*	NS	*	*	*	*	*

a: Soluble fraction of feed

b: Potential degradable fraction

C: Rate of degradation of fraction b (h-1)

Pd: Potential degradability

Ed: Effective rumen degradability calculated at out flow rate K= 0.02, 0.05, 0.08

SEM: Standard error of means

T1: Untreated SSC

T2: SSC treated with 50 % Ethanol

T3: SSC treated with 70 % Ethanol

T4: SSC treated with Tallow 10 %

NS: Not significant

L. S: Level of significance

* = P < 0.05

Table, (3) Insitu crude protein degradability of treated and untreated sesame cake

Treatments	a	b	C	Pd	Ed0.02	Ed0.05	Ed0.08
T1	54 ^a	42.4 ^c	0.075 ^a	96.5 ^a	83.5 ^a	75.4 ^a	70.5 ^a
T2	31.3 ^c	50.1 ^a	0.043 ^b	81.4 ^c	68.1 ^d	56.2 ^c	50.1 ^c
T3	40.2 ^b	47.0 ^b	0.075 ^a	87.4 ^b	77.1 ^c	68.2 ^b	62.7 ^b
T4	37.5 ^b	51.9 ^a	0.082 ^a	89.4 ^a	79.2 ^b	69.6 ^b	63.7 ^b
SEM	1.57	2.1	0.855	2.12	0.85	0.67	0.74
L. S	*	*	*	*	*	*	*

a: Soluble fraction of feed

b: Potential degradable fraction

c: Rate of degradation of fraction b (h^{-1})

Pd: Potential degradability

Ed: Effective rumen degradability calculated at out flow rate $K = 0.02, 0.05$ and 0.08

NS: Not significant

L. S: Level of significance

T1: Untreated SSC

T2: SSC treated with 50%Ethanol

T3: SSC treated with 70%Ethanol

T4: SSC treated with tallow 10%

SEM: Standard error of means

* = ($P < 0.05$)

CHAPTER FIVE

DISCUSSION

True dietary protein consists of amino acids joined together in various combinations. In the field of ruminant livestock nutrition, it is known that under same circumstances protecting dietary protein from extensive degradation in the rumen by microbial enzyme can lead to an increase in the out flow of amino acids from the rumen and /or a change in the balance of amino acids reaching the lower gut.

SSC protein was highly degradable (Mahala and Gomaa, 2007) there for chemical or physical treatment is required to protect it from microbial fermentation in the rumen. Finding the treatment of heat reduce the crude protein and DM degradability in the rumen this agreed with the results.

Chemical treatment of (SSC) by either ethanol or tallow reduced protein degradability by forming insoluble protein. This is inconsistent with Lynch *et al.*, (1987) who found the chemical bonding between denature vegetable protein and lipids, the resulting material improved stability in aqueous solutions which may render the protein more resistant to ruminal degradation, on the other hand this result matching with the Glenn, *et al.*, (1977) who used various lipids to coat feed protein and amino acids to protect them from ruminal degradation.

In this a study the treatment of sesame cake with Tallow protect the protein from ruminal degradation whereas compare with untreated sesame cake. The finding of Lynch *et al.*, (1978) found that an interaction between vegetable proteins and lipids were able to bind with lipids and form a

lipoprotein-like complex the resulting products was more stable in aqueous solution, denaturation of protein by alcohols provide an increased number of ionic and hydrophobic residues to which lipid could bind, these finding are in accordance with our result. Lipid treatment used in our experiment consisted of applying 10% tallow to untreated sesame cake meal which was similar of finding a Peterson *et al.*, (1975) and Glenn, *et al.*, (1977) who found that 10% lipid would be more practical in terms of feed storage and handling. Lynch *et al.*, (1986) found that tallow reduced N disappearance and rate of N loss, these are in line with our results. Fukushima, *et al.*, (1969) who reported treatment of oil seed protein with aqueous alcohol solution produces permanents change in the three dimensional structure of the protein molecule. Van der, *et al.*, (1983), suggested that alcohol treatment of SBM reduce degradation of soluble proteins as well, which was concurred with our results. Treated sesame cake meal with 50% and 70% ethanol at 78^oc resulted in lowest N solubility than untreated, these result are similar to Van der *et al.*, (1983), who found reduction in insitu N disappearance when comparing SBM treated with 40% ethanol at room temperature to untreated SBM, ethanol treatment at higher temperature reduce N solubility.

In the present study the values recorded for dry matter and crude protein degradability, the degradation rate, soluble nitrogen, insoluble nitrogen and effective protein degradation of sesame cake were similar to the results obtained for soybean meal (SBM), by Boruki, *et al.*, (2007), who reported that treatment of soybean meal with HCL protects the crude protein and amino acids from ruminal degradation, hence increasing rumen

undegradable protein and had a higher potential to enhance the amino acids supply.

Also formaldehyde treatment reduced protein degradability of groundnut cake (GNC), gengelly (sesame cake) and rubber seed cake (RSC), (Sampath and Savaram., 1987). Formaldehyde treated sesame cake as main source of protected protein it was concluded that sesame meal with Formaldehyde decreased organic matter (OM), crude protein degradability and blood urea concentration (Aparicio,1992).

Ethanol and tallow treatment of (SSC) protein revealed significantly ($P < 0.05$) lower degradability as compared to untreated sesame cake for all time of incubation this is inconsistent with (Aplang, 2008).

CONCLUSION

The results demonstrated that the treatment by ethanol or tallow of sesame cake increased the amount of dietary protein escaping degradation in the rumen and decreased the potential degradability and effective degradability of crude protein and dry matter.

RECOMMENDATIONS

- More continuous studies should be carried out using different chemical treatment on local produced feedstuff.
- Feeding trials should be performed to determine the effect of the treatment on feedlot performance.

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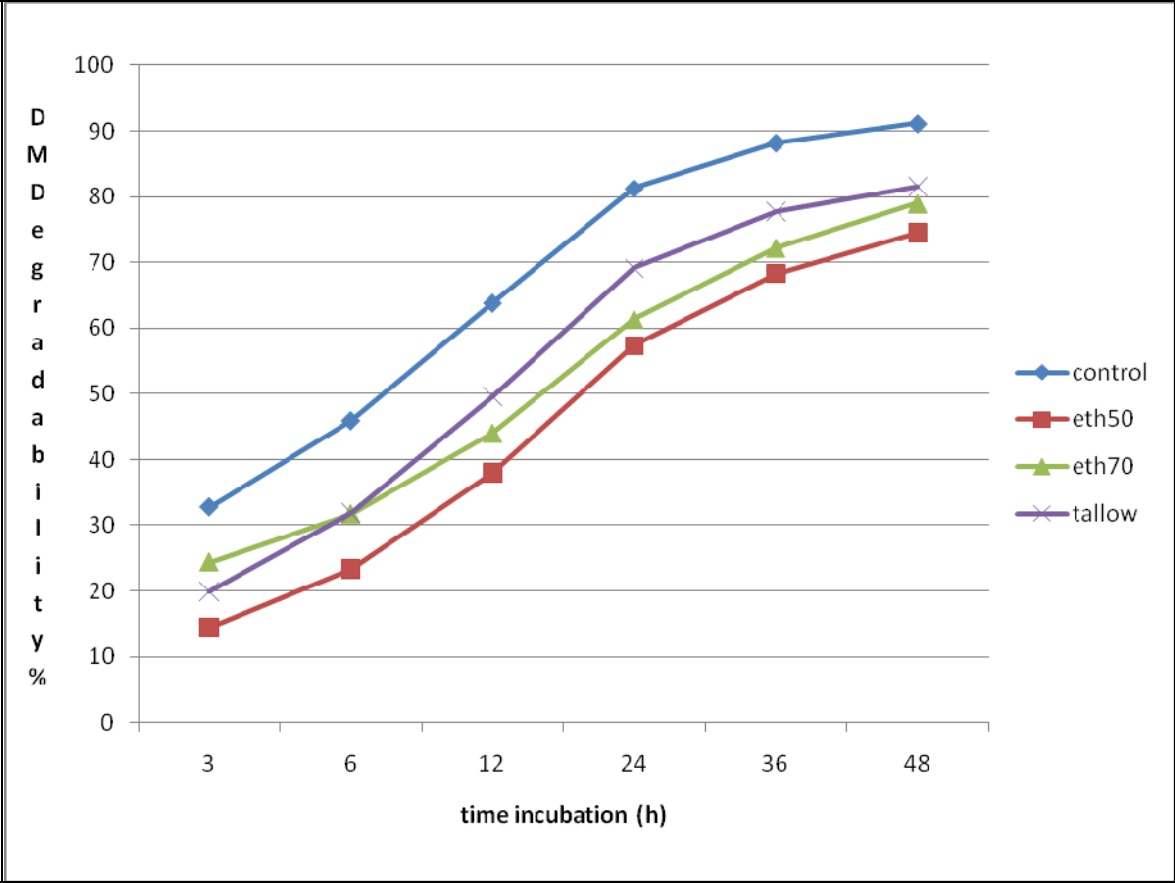


Figure (1) Dry Matter degradability

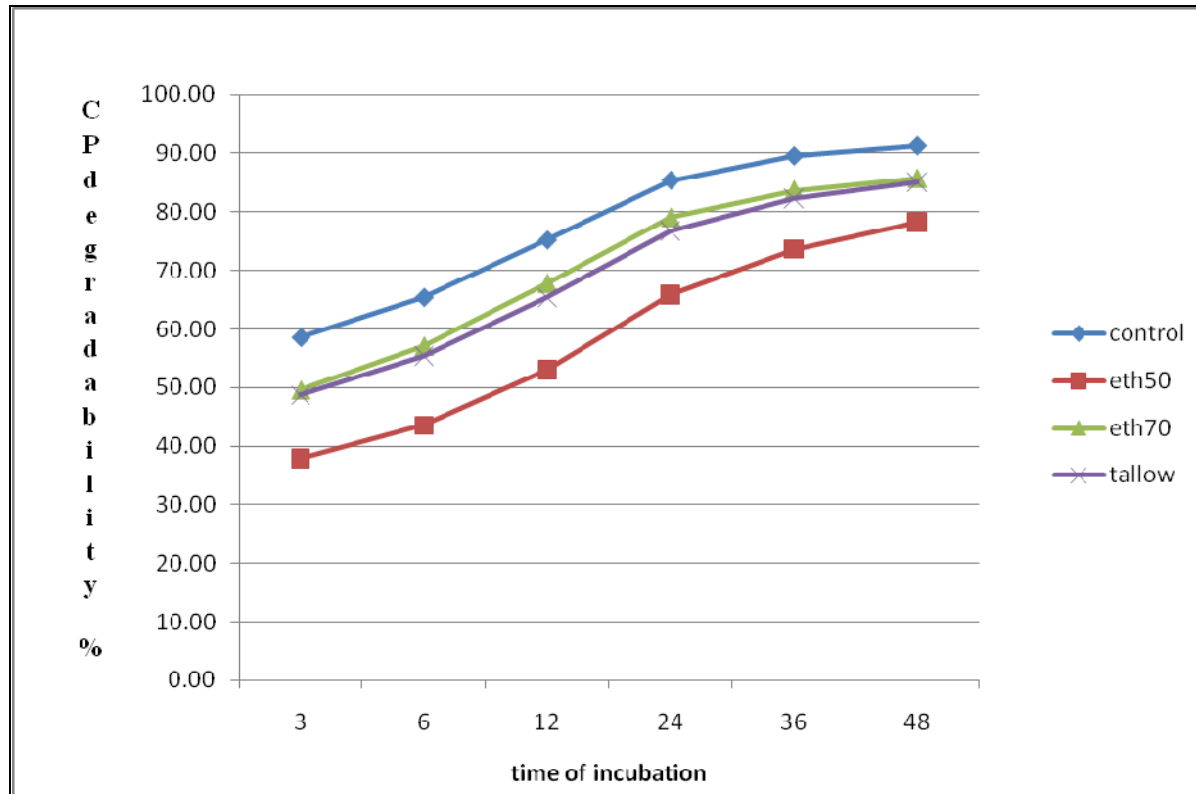


Figure (2) Crude protein degradability

تأثير المعاملة الكيميائية لامباز السمسم على تكسرية المادة الجافة

والبروتين الخام

مقدمة بواسطة :

مجاهد بابكر الامين محمد على

ماجستير العلوم فى التغذية

المستخلص

تم تقييم النيتروجين الظاهرى والمادة الجافة لامباز السمسم غير المعامل والمعامل بايثانول تركيزه 50% تركيزه 70% فى درجة حرارة 78°م والمعامل بالشحم بنسبة 10% .

أجريت تجربة التكسرية للبروتين الخام والمادة الجافة فى ثلاثة ثيران ذات ناسور كرشى غذيت على قصب الذرة كعليقة اساسية مع امدادها ب2 كجم/يوم عليقة مركزة و حجر اللحوس . ثلاثة اكياس (كيس/ ثور / فترة زمنية) فى كل المعاملات ثم تم تحصيلها فى الكرش للفترة الزمنية التالية صفر , 3, 6, 12 , 24 , 36 و 48 ساعة

فى اكياس النايلون البروتين الخام والمادة الجافة قورن جهد القابلية التكسرية والجزء الذائب من المادة الغذائية وفاعلية الكرش التكسرية لعينات السمسم المعاملة كانت معنويا () اقل عند مقارنتها مع العينات الغير معاملة ويعدى ذلك لنقص زوبانية النيتروجين.

أقصى أختزال فى تكسرية البروتين الخام كمتوسط وجد فى العينات المعاملة بالايثانول 50% يليه المعامل بالايثانول 70% والمعامل بالشحوم بنسبة 10% .

معاملة أمباز السمسم بالايثانول تحمى البروتين الخام من التكسرية بواسطة الميكروفلورا فى الكرش. ومن المتوقع أن يحسن فى أستهلاك النيتروجين.