NOVEL MULTINUTRIENT CATTLE BISCUIT AS AN ALTERNATIVE TO TRADITIONAL UREA SUPPLEMENTS FOR DAIRY COW

A Thesis By

Md. Nahid Sultan

Examination Roll No. 0213/05

Registration No. 164 (2013-2014)

Semester: July-December 2014

A thesis submitted in partial fulfillment of the requirements for the degree of Master of Science in Animal and Poultry Nutrition

Department of Animal Science and Nutrition

Faculty of Veterinary Medicine

Chittagong Veterinary and Animal Sciences University

December 2014

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This is to certify that we have examined the above Master's thesis and have found that the thesis is complete and satisfactory in all respects and that all revisions required by the thesis examination committee have been made

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Abstract

The study was carried out in a selected commercial dairy farm of Patiya under Chittagong Division, Bangladesh for a period of 60 days from September to October 2014 to innovate novel cattle biscuit as an alternative to traditional urea supplements for dairy cow. Twenty Local × Holstein crossbred milking cows were selected according to age, live weight, BCS and daily milk yield from the experimental farm. Animals were randomly distributed into five dietary treatment groups designated as T_0, T_1, T_2, T_3 and T_4 having four replicates per treatment. All animals were stall fed. Ration was prepared and supplied to the animal as per recommendation. Multi-nutrient Cattle Biscuit (MCB) was fed twice daily. All animals had free access to clean, cool drinking water. Intake of basal diet was recorded daily. All animals were kept in a single row stanchion barn. Body weight was measured, milk yield was recorded, milk and blood parameters were tested in the laboratory.

The daily milk yield of the cows in the experimental groups supplemented with varied levels of MCB significantly ($p<0.05$) increased for the last four weeks. The highest average milk yield (8.3 kg/d) was recorded in T_3 group and the lowest milk yield (6.3 kg/d) was recorded in T_0 group. Milk composition of the cows varied in an irregular fashion during the experimental period. Fat percent of milk significantly ($p<0.05$) increased during 1^{st}, 2^{nd}, 3^{rd}, 7^{th} and 8^{th} week in the treatment groups compared to control group. Besides fat, protein percent of milk increased significantly in the 1^{st} ($p<0.001$); 2^{nd}, 5^{th}, 8^{th} ($p<0.05$) and 7^{th} ($p<0.01$) week. The Solids not fat (SNF) percent differed significantly in the 1^{st}, 2^{nd}, 5^{th} ($p<0.01$); 3^{rd} and 7^{th} ($p<0.001$) week. Unlike SNF, the total solids (TS) percent differed significantly in the 1^{st}, 7^{th} ($p<0.01$); 3^{rd} ($p<0.001$) and 5^{th} ($p<0.05$) week. On average (1-8 weeks), milk fat, milk protein, SNF and TS percent were higher in the T_2 (25% urea supplemented MCB) and lower in T_0 (without MCB) group respectively.

Unlike milk components, there was no significant difference ($p>0.05$) in serum cholesterol, serum glutamic pyruvic transaminase (SGPT), bilirubin, urea and total protein level throughout the whole experimental period. However, serum glutamic oxaloacetic transaminase (SGOT) differed significantly ($p<0.05$) only in the 5^{th} week. Creatinine differed significantly in the 2^{nd} ($p<0.01$) and 3^{rd} ($p<0.05$) week. Glucose level differed significantly in the 1^{st} ($p<0.01$), 5^{th} ($p<0.001$) and 8^{th} ($p<0.01$) week. In the light of above observations, it might be concluded that, MCB supplementation substantially improved milk yield and milk composition and did not interfere blood parameters of the experimental cows. Therefore, 25% urea supplemented MCB in addition to basal diet may be suggested as a novel alternative to traditional urea supplements for dairy cow.

Keywords: Serum parameter, Dairy cow, Milk composition, Milk yield, Multi-nutrient cattle biscuit.

Chapter IV Results

List of Abbreviation

ANOVA	-	Analysis of variance
ARC	-	Agricultural Research Council
BBS	-	Bangladesh Bureau of Statistics
BCS	-	Body condition score
BER	-	Bangladesh economic review
BMD	-	Bangladesh Meteorological Department
BUN	-	Blood urea nitrogen
DCP	-	Digestible crude protein
FAO	-	Food and agriculture organization
GDP	-	Gross domestic product
HDL	-	Low density lipoprotein
IAEA	-	International atomic energy agency
LDL	-	High density lipoprotein
LW	-	Live weight
KG	-	Kilogram
MCB	-	Multi-nutrient cattle biscuit
ML	-	Milliliter
N	-	Nitrogen
NEFA	-	Non-esterified fatty acid
SGOT	-	Serum glutamic oxaloacetic transaminase
SGPT	-	Serum glutamate-pyruvate transaminase
SNF	-	Solid not fat
TDN	-	Total digestible nutrient
TMR	-	Total mixed ration
TS	-	Total solid
UMB	-	Urea molasses block
UMMB	-	Urea molasses multi-nutrient block
UTRS	-	Urea treated rice straw
VFA	-	Volatile fatty acids
VLDL	-	Very low density lipoprotein

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Chapter-1: Introduction

Bangladesh is a densely populated country with 149.8 million people (BBS, 2012). The majority of this population directly or indirectly depends on agriculture and the percentage is about 53.7% as proportion to the total population (FAO, 2005). The contribution of livestock sub-sector to GDP at constant prices was 2.6% during 2010-11 fiscal year. The estimated contribution to GDP during 2011-12 fiscal year from this sub-sector was 2.5%. The availability of milk in our country is only 33.0 ml per head per day against requirement of 250 ml per head per day (DLS, 2001). Though the share of the livestock sub-sector in GDP is small, it has immense contribution towards meeting the daily protein requirements through milk, meat and egg. During 2005-2006 fiscal year milk production were 22.7 lakh tones which increased up to 34.6 lakh tones in 2011-12 fiscal year (BER, 2012). Around, 10.4 million households rear cattle which is the 36.2% of the total households. Cattle population of Bangladesh is about 26.8 million of which 3.7 millions are milking cow. Household having crossbreed cattle is about 0.6 million and crossbred cows are about 0.2 million. The total number of improved or crossbreed milking cows are about 0.2 million (BBS, 2009).

In Bangladesh, a major constraint to ruminant livestock production is the severe scarcity of feeds and fodders both in quality and quantity. Due to high pressure on land for crop production farmers cannot spare it for fodder production. As a result, cattle and buffalo subsist mainly on straw based diet with limited supplementation of green fodder and little or no concentrate. Alam (2002) mentioned that 23.58 million tonnes of green fodder is available against the requirement of 70.42 million tonnes.

Rice straw is an important crop residue contributing more that 90% of total dry matter available to the dairy cattle. However, straw is severely deficient in protein and mineral content (Karim, 1988) and its cellulose and hemicellulose are poorly digested (Jackson, 1977). Now-a-days, the nutritive value of rice straw is improved by the appreciating efforts of many animal nutritionists (Itoh *et al.,* 1979; Liu *et al.,* 1988; Hock., 1988; Saadullah, 1991). From the results of those findings, the nutritional limitations could be overcome by physical and chemical treatments or by providing specific nutrients to improve an optimum

ruminal condition for ruminal microflora. In case of utilization of the roughage nitrogen is the main limiting factor and protein supplement undoubtedly could increase its voluntary intake and digestibility (Church and Suntos, 1981; Guthire and Wagner, 1988).

Straw has a poor nutritive value (4.0% crude protein and 5.0 MJ ME/kg DM). Its digestible crude protein (DCP) is near zero and total digestible nutrients (TDN) content is only 48.0% which is not satisfactory (Akbar and Khaleduzzaman, 2009). The possible alternative for much better utilization of straw is to improve its digestibility by treating with appropriate chemical or biological agents or by physical means so that its lingo-cellulose bond is broken or at least loosen to free major portion of cellulose to be digested by the ruminants. Between physical and chemical treatments, chemical treatment achieves most attention by the scientists, particularly treatment with urea and molasses (Akbar, 1992; Akber and Tareque, 1990; Saadullah *et al.*, 1982). Though urea treatment increases the digestibility of straw, it was not well accepted by the farmers because of the method is tedious and time consuming and dangerous particularly for rural farmer (Akbar, 1992).

The Urea Molasses Multinutrient Block (UMMB) supplementation is mainly recommended with the animals that are fed with poor quality roughages like rice straw or mature grass because they generally contain less nutrients, more energy and protein (Alam *et al.*, 2006). Urea in the block supply readily available nitrogen to the microbes in the rumen and this nitrogen is used by them to produce protein for growth and production (Tiwari *et al.*, 1990). Rumen microbes use molasses as a source of energy and sulphur. The fibrous substances present in rice straw and natural grass are degraded with increasing rate by the help of nitrogen, energy and sulphur present in the block (Alam *et al.*, 2006).

The supply of nitrogen, energy and sulphur from block increases the rate of degradation of fibrous substances present in rice straw and natural grass, which are ultimately utilized by the animal for higher performances (Alam *et al.*, 2006). Supplementation of basal diet with urea molasses block (UMB) is common practice in Bangladesh which has shown to have beneficial effect on growth performance, milk yield and milk composition. However, this is a tedious process of preparing and presenting block to the animal. It can never be stored for a

long time and cannot spread to the rural farmer as they don't have the precise manufacturing knowledge and skill.

Multinutrient Cattle Biscuit (MCB) on the other hand is a new concept in Bangladesh which can be prepared commercially in industrial level and it can be supplied, stored and easily portable to rural farmers. Therefore, before disseminating this cost effective technology the objectives of this study were to investigate the effect of supplementing MCB on milk yield, milk composition and blood parameters of crossbred dairy cows.

1.1 General objective

Develop cost effective high energy high protein commercial cattle biscuit to improve milk production and health of crossbred dairy cows.

1.2 Specific objectives

1. To measure daily milk yield and milk composition of the dairy cow.
2. To find out appropriate level of multi-nutrient cattle biscuit (MCB) supplementation for dairy cow.
3. To analyze blood profile of cows fed different level of multi-nutrient cattle biscuit (MCB) supplementation.

Chapter-2: Review of Literature

2.1 Livestock production scenario

Bangladesh is a densely populated country with a huge population of 149.8 million (BBS, 2012). The majority of this population directly or indirectly depends on agriculture and the percentage is 53.7% as proportion to the total population (FAO, 2005a). The contribution of the livestock sub-sector to GDP at constant prices was 2.5 percent during 2012-13. The estimated contribution to GDP during 2011-12 from this sub-sector was 2.5 percent. Though the share of the livestock sub-sector in GDP is small, it has immense contribution towards meeting the daily protein requirements through milk, meat and egg. During 2005-2006 fiscal year the milk production was 22.7 lakh tones which increased up to 50.7 lakh tones in 2012-13 fiscal year (BER, 2014).

The livestock consisting 25.8 million bovines, 17.3 million caprines and ovines and 135.1 million of poultry (BBS, 2012) contributes about US$ 2309.0 million as animal farming GDP sharing 18.6, 56.3, 19.8, 2.9 and 2.6%, respectively by dairy, meat, egg, hides, skin and others (BBS, 2011). It supported per capita intake of 14.3 kg milk, 8.9 kg meat and 115 eggs in 2011 (BBS, 2012). The supply of milk and meat is only 15 to 20% of their annual requirement and they are far below the average of the developing countries (55.0 kg and 32.0 kg respectively) (Thornton, 2010). The per capita annual egg consumption of the country in 2011 was 115 (BBS, 2012) and the number is close to the average consumption of the developing countries (120). About 73.8, 82.5 and 82.7% of the total bovines, caprines and ovines and poultry, respectively are kept by the landless and small farmers (Agri. Census, 2008) and their annual population growth was 3.9%, 3.1% and 54.8%, respectively during the period of the Agricultural and livestock Census 1983/84 to Agriculture Census 2008. The average share of the same animals by the medium and large farmers, on the other hand, was 26.2, 17.5 and 17.3%, respectively (Agri. Census, 2008).

2.2 Dairy cattle in Bangladesh

According to Bangladesh Bureau of Statistics 10.4 million household rear cattle which are the 36.2% of total household and household having milking cattle is about 3.7 million which is 12.93% of the total household (BBS, 2009). Cattle population of Bangladesh is about 26.8 million among these 3.7 million cattle are milking cow. Household having crossbreed cattle is about 0.6 million and household having milking cross breed cow is about 0.2 million. The total number of improved or crossbreed milking cow is about 0.2 million. The total number of milking cattle is about 3.8 million. The existing cattle breeding programme as adopted from 1982 was (i) female breed in urban, semi urban and milk pocket areas with 50% Friesian and 50% Shahiwal/indigenous bulls and (ii) breed females in rural areas with 50% Friesian and 50% indigenous bulls (Bhuiyan, 1997).

2.3 Milk yield in different cows

Sarker (1995) demonstrated that the milk production from crossbreds and indigenous dairy cows were 6.7 and 1.6 litter per day, respectively. In another study, Nahar *et al*. (1992) reported that the average daily milk yield of Holstein x indigenous, Sahiwal x indigenous, Sindhi x indigenous and Jersey x indigenous crossbreds were 5.5, 2.9, 3.0, 3.8 kg, respectively. Halim (1992) reported that lactation period for indigenous and crossbred dairy cows were 228 and 259 days, respectively. Hasan (1995) reported the average lactation period of Jersey, Holstein, Sahiwal and Sindhi crosses were 286, 272, 262 and 255 days, respectively. Khan (1990) reported that the average lactation period of Pabna, Sindhi cross and Sahiwal cross were 200, 251 and 282 days respectively. Kabir and Islam (2009) reported that the average milk yield of Holestein cross, Sindhi cross, Sahiwal cross and local cows were 12.0, 7, 5.1 and 2.1 litter/ day respectively.

Paul *et al*. (2013) reported that, the average milk yield of Desi, Shahiwal × Desi, Friesian × Desi and Jersey × Local was 2.3, 4.9, 6.0 and 5.7 (liters/day), respectively. It was observed that in Bangladesh, crossbreeding had a significant effect ($p<0.01$) on milk yield. Among different cows, highest milk production was recorded in case of Friesian × Desi cross (6.0

liters/day) and lowest milk yield was recorded in case of Desi cows (2.3 liters/day). These results are in agreement with findings of Islam *et al*. (1999) who found that the average milk yield of the Desi, Shahiwal × Desi, Friesian × Desi cows was 2.1, 4.7 and 6.2 liters/day, respectively. Shamsuddin *et al*. (2006) found that, the average milk yield per cow per day was 7.2 liters in Sirajgonj-Pabna region of Bangladesh, while it was 3.5 liters, 4.8 liters and 5.1 liters per cow/day in Mymensingh, Khulna, Satkhira and Chittagong, respectively. Talukder *et al*. (2001) reported that, Holstein-Friesian crossred cows yielded 2.5 kg more milk daily than that of Desi cows (7.2 vs. 4.7 kg per day).

2.4 Non-protein nitrogen (NPN) as feed ingredients

The history of the discovery of nitrogen, protein, amino acids, urea and other information that led to the development and use of non-protein nitrogen (NPN) compounds in ruminant nutrition was reviewed by Stangel (1967). During World War I, Germany began manufacturing NPN compounds as substitutes for plant and animal protein in ruminant diets. NPN products were widely used in Europe before research began on these in the United States. In 1935, on the other hand, urea began to be produced in the US and it became available to the feed manufacturer. Hart *et al*. (1939) after intensive research using NPN products, concluded that ruminants could synthesize protein from simple nitrogen compounds through the action of the rumen microorganisms and that the muscle tissue of steers fed a diet containing urea contained ordinary protein. Work and Henke (1940) found growing and finishing cattle receiving urea had regular livers and kidneys. Harris and Mitchell (1941a) determined the biological value of urea for maintenance and growth. They showed that NPN could be utilized effectively in diets defiant in protein but when an adequate amount of natural protein was present, urea was utilized poorly.

2.5 Non-protein nitrogen utilization in ruminant

It is essential to indicate that NPN compounds are usual constituents in the biological fluids of ruminants, even when NPN is absent from the diet. Also, natural feedstuffs that are fed to ruminants contain a variable amount of NPN. Thus, the ruminant continually uses NPN as a

normal dietary and metabolic constituent. Ammonia is the common denominator in the utilization of NPN by ruminants (Hungate, 1966). If the rumen microorganisms cannot degrade the compound in question to yield free ammonia, it is of no use as a nitrogen source to the microorganisms. Allison (1969) reviewed the biosynthesis of amino acids by rumen bacteria. In general, amination and transamination reactions appear to be responsible for the major part of ammonia assimilation by the microflora.

Glutamic dehydrogenase (Hoshino *et al*., 1966) plays a key role in the initial fixation of ammonia to a carbon skeleton and glutamate-oxaloacetate and glutamate-pyruvic transaminases are important in the transfer of ammonia to other carbon skeletons, which are present in rumen fluid. Other dehydrogenase and transaminase enzyme systems also play a part in ammonia assimilation by rumen bacteria (Chalupa, 1971). Rumen microflora can use NPN for protein synthesis if the necessary carbon skeletons are present or if these can be synthesized fast enough from dietary carbohydrate or alternate carbon sources. The most important single fermentation characteristic is the amount of fermentable energy available in the diet for microbial growth and protein synthesis above that needed for maintaining equilibrium in the rumen between the feed protein degraded and the microbial protein resynthesized.

2.6 Urea in ruminant diet

In 1940, the Association of American Feed Control Officials (AAFCO, 1955) approved the use of urea and ammonium bicarbonate, the only acceptable sources of NPN at that time, by adopting a decision recommending that not more than one-third of the total protein in the diet be from NPN products. Oltjen (1969) showed that beef cattle can grow up and reproduce when fed diets in which urea supplied all the dietary nitrogen. Cattle have remained on such protein-devoid diets for over 4 years with no evidence of ill effects. Virtanen (1966) reported a moderate production of milk from dairy cows fed diets containing urea and ammonium salts as the exclusive sources of dietary nitrogen.

It has been a century over while Weiske *et al*. (1879) reported that ruminants could convert NPN to protein. During the following 60 years, this issue was intensively researched by German nutritionists. Later on, Krebs (1937) reviewed their research and summarized the status of the field at the time. Studies on the subject matter in the United States began in Wisconsin. Hart *et al*. (1939) reported that, either urea or ammonium carbonate might be used by growing dairy heifers. They also reported that, dietary soluble carbohydrate may increase NPN utilization in ruminants. Later on, a series of experiments were carried to study the metabolic aspects of NPN utilization by ruminants.

Another landmark in NPN research was conducted by Loosli *et al*. (1949) who demonstrated that urea could serve as the sole dietary nitrogen source for the lambs. Using the purified diet approach, they found that 10 amino acids that are dietary essentials for the laboratory rat were synthesized within the rumen. Lambs fed these diets grew and remained in positive nitrogen balance during the trial period. Results of parallel studies yielded information on the mechanism of NPN utilization and provided the facts for establishing the guidelines for the use of NPN in realistic ruminant rations. Finally, urea was approved in the United States as a feed ingredient in ruminant's diets in 1940 by AAFCO.

2.7 Performance of cattle feeding urea

Bos indicus and associated cross-breeds have higher urea production and recycling capacity than *Bos taurus*. Norton *et al*. (1979) reported that, cross-breeds from Brahman cattle produced 30% more urea-N and transferred 60% more urea- N into the gut compared with the Shorthorn breed. Higher renal re-absorption of urea-N seemed to account for this higher gut entry in Brahman cattle (Norton *et al*., 1979). *Bos indicus* crossbred cattle are often utilized in beef production in semi-arid environments due to their capability to adjust to high environmental temperature and low quality feed. In grazing studies in semi-desert rangeland, Brahman cows maintained higher body condition scores had greater serum concentrations of NEFA and urea-N in early lactation than *Bos taurus* cows (Obeidat *et al*., 2002). The authors suggested that different mechanisms exist between these breeds for tissue mobilization as

energy sources for maintenance and production. Different breeds of cattle for dairy and beef production also show quantitative variation in urea metabolism.

During growing and fattening stages, Japanese Black and Japanese Brown cattle had greater plasma concentration of urea-N than Holstein cattle under a similar feeding situation (Matsuzaki *et al*., 1997). In early-weaned calves of different breeds reared at the same body weight gain, Japanese Black calves have higher plasma urea concentration and better rate of urea production and recycling compared with Holstein calves (Shingu *et al*., 2007). Although the reasons for these differences between Japanese Black and Holstein calves are not clear, differences in body composition and endocrine status (Matsuzaki *et al*., 1997) possibly affect the variation of body protein yield in these cattle during growth.

2.8 Development of urea-molasses block

In South Africa the first trial of providing urea through feed supplementation blocks were done by Altona *et al*. (1960). The block included common salt and urea and provided acceptable results. Later on, other experiments using molasses, urea and salt confirmed these outcomes (Beames, 1963; Beames and Morris, 1965). Feed manufacturing companies also developed urea-molasses blocks, but the blocks made by industrial process were relatively costly and not affordable to these who needed this product the most, the small scale farmers in the developing countries.

In the early 1980s, the work of Professor Leng from Armidale University in Australia, in cooperation with the joint FAO/IAEA Division (Vienna) and the National Diary Development Board (NDDB) (India) renewed curiosity in this technology particularly for developing countries (Leng, 1984; Kunju, 1986). It appeared that, the technology could be tremendously being useful for Sahelian countries with sugar industries suffering from severe droughts, such as Senegal.

Unfortunately, the manufacture of urea-molasses blocks as studied in Australia used a “hot process” which required the pre-heating of the molasses. However, heavy and expensive

equipments (such as double jacket broiler) and foreign exchange to cover energy needs, usually imported as fossil fuel, was needed for this method. This was a serious impediment for African counties. It was for these reasons that the FAO Feed Resources Group (Sansoucy, 1986) tried to modify the technology to make it much simpler. The first trials were made at facilities provided by the Senegalese Agriculture Research Institute, in Dakar-Hann. The idea was to develop a "cold process" that incorporated the molasses into the mixture without any heating and to test various binding agents and ingredients.

The original formula was based on the work of an FAO projects in Egypt. It consisted of molasses 50%; wheat bran 25%; urea 10%, quick lime 10%; and common salt 5%. More than 70 different formulae were tested for final block quality. Several using locally available ingredients were found satisfactory and selected for the field trials. The new technology was applied by mixing the ingredients manually or with concrete or horizontal feed mixers depending on the scale. This improvement was a real breakthrough since it allowed the application of the technology at low cost and at small scale at village level by the farmers themselves.

Different formulae with or without molasses have been developed and tested according to the local availability, quality and price of ingredients. This demonstrates the adaptability of the technology. Eventhough designed mainly for dairy and beef cattle, the model has been used for buffaloes (Nguyen Van Thu, 2000), small ruminants (Houmani and Tisserand, 1999; Osuna *et al.*, 1996; Salman, 1997) and even rabbits (Binh *et al.*, 1991; Filippi *et al.*, 1992; Perez, 1990). Outstanding results have been obtained with different types of production, growth, meat, milk, work or wool (Sansoucy, 1995), although, one of the greatest effects seems to be obtained on reproductive performance of animals (Duc Vu *et al*., 1999; Ghosh *et al.*, 1993; Hendratno, *et al*., 1991; Vargas and Rivera, 1994).

At present, the technology of the cold process has been well mastered by many peoples in developing countries. Blocks are currently commercially produced on a large level in many countries (India, Mexico, Niger, Pakistan, Sudan, Venezuela, etc.) using a variety of equipments from a simple shovel to sophisticated industrial equipment. In Australia, the

achievement of the blocks is tremendous and growing from year to year. The possibility of using blocks as carriers of anthelmintic medicines was investigated at an early stage (McBeath *et al.*, 1979). However, in Asia, it has been fruitfully investigated more in recent times, in particular by the Australian Centre for International Agricultural Research (ACIAR). Other research has been conducted in Venezuela (Araque and Rosos, 1993), India (Sanyal *et al*., 1995), Ethiopia (Anindo *et al*., 1997) and Bangladesh (Saadullah *et al.*, 1991). The technology appears attractive, but the manufacture of such medicated blocks is only applicable at an industrial scale, not at village level.

2.9 Urea molasses block for dairy cows

German workers (Ehrenberg *et al*., 1891; Zuntz, 1891) determined that urea could be used to substitute a fraction of protein in ruminant rations. Reid (1953) concluded that:

- Conversion of urea to protein is mediated by the microorganisms of the rumen and reticulum which subsequently benefit the host animal.
- A low level of protein and high level of starch in the ration favor urea utilization.
- Bacteria may prefer highly soluble and readily hydrolysable protein rather than urea in the ration.
- Sugars and cellulose are inferior to starch as sources of energy for ruminal microorganisms.
- Application of *in vitro* to *in vivo* experiments may be misleading because the characteristics and kinds of microorganisms may differ at short periods.
- Urea N may provide up to 27% of required N from the standpoint of milk yield or reproductive behavior/general health.
- Urea may provide up to 3% of the concentrate ration or up to 1% of the total ration for milking cows from a practical standpoint.
- Small quantities of undiluted urea introduced suddenly into the rumen resulted rapid onset of toxicosis, whereas 180 to 272 g urea was consumed daily by beef calves/cows without toxicosis when fed along with hay or corn silage.
- Feeding urea at optimum level does not reduce palatability of basal diet.
- Molasses may improve palatability of urea-containing ration.

2.10 Degradation of urea in rumen

Urea was degraded in the bovine rumen ranged from 25 to 53% with higher percentages in response to lower N intakes (Bunting *et al*., 1989a; Huntington, 1989) or higher intake of readily fermented carbohydrates (Huntington, 1989). In goats, increasing dietary N slightly increased the percentage degraded in the rumen from 43 to 46% (Obara and Shimbayashi, 1980). Urea is rapidly hydrolyzed by bacteria adhering to ruminal epithelium and the resultant ammonia enters the ruminal ammonia pool (Bunting *et al*., 1989b). Amounts ranging from none to over 80% of ammonia from urea degradation are incorporated into bacterial N (Bunting *et al*., 1989a; Salter *et al*., 1979) and availability of energy is the major determinant of that percentage. In reality, the positive effects of organic matter digestibility and ruminal ammonia concentration on urea transfer are functions of the ruminal microbial capacity to assimilate products of fermentation.

Table 1. Normal range of urea intake

No. of cow	Milk yield (kg/day)	Urea intake (g/day)	Reference
42	23	200	Polan *et al*. (1968)
24	27	170	Huber *et al*. (1968)
12	26	191	Knott *et al*. (1972)
20	29	186	Huber and Thomas (1971)
45	30	180	Huber *et al*. (1973)

2.11 Urea toxicity in cattle

Huge amounts of dietary urea when consumed over a short period of time are lethal to ruminants (Clark *et al.,* 1951; Coombe *et al.,* 1960; Coombe and Tribe, 1958; Davis and Roberts, 1959; Dinning *et al*., 1948; Gallup, 1956; Repp *et al*., 1955). Elevated ruminal fluid ammonia levels and subsequent high levels of blood ammonia are chief characteristics of urea toxicity. Clark *et al*. (1951) found that, dietary urea toxicity was greater in sheep if they were fed diets of poor quality hay. Urea administered to cattle at a level of 0.44 gm/kg live weight was toxic (Davis and Roberts, 1959); conversely, if acetic acid was administered at a level to reduce the effect of ammonia released prior to the beginning of tetany the cattle survived.

Excessive absorption of ammonia into the blood can overwhelm the capability of the liver to detoxify it back to urea and ammonia toxicity results. The toxic effects of too much consumption of urea have been well documented (Antonelli *et al*., 2004; Bartley *et al*., 1981; Bartley *et al*., 1976; Davidovich *et al*., 1977). The symptoms of urea toxicity in order of appearance later than exposure include: fasciculation, apathy, hyperaesthesia, tremors, rumen stasis, incoordination, recumbancy, convulsions and death (Antonelli *et al*., 2004). The required amount of urea to cause toxicity varies widely, though urea fed at as low as 0.35g/kg BW resulted in death in some dairy cattle (Ryley and Gartner, 1968). However, ammonia toxicity from feed urea is somewhat situation dependant. There are wide reports that higher levels of urea are allowable in the diet when it is fed as part of a total mixed ration (TMR) instead of indiscrete meals (Kertz, 2010). Animals fed a TMR would be exposed to minor concentrations of urea, with more time for ammonia detoxification across the day.

However, Bartley *et al*. (1976) indicated that ammonia toxicity was poorly correlated to rumen ammonia concentration. Instead they showed that toxicity related more closely to rumen pH. When ruminal urea degradation results a fast accumulation of ammonia in the rumen, then the pH of the rumen may increase sharply, that time ionization of ammonia molecules removes free hydrogen ions from solution (Kertz *et al*., 1983). Increased ruminal pH facilitates a rapid transport of ammonia across the rumen epithelium, resulting in a quick increase in blood ammonia and the consequent ammonia toxicity (Abdoun *et al*., 2006). The ammonium chloride treatment resulted in increased rumen ammonia concentrations, but no pH elevation and subsequently no toxicity.

Table 2. Lethal dose of urea

Urea	Animal	Dosage (g/kgLW)	Given by	Result	Reference
Urea	Cattle	0.31	drench	Death	Davis and
Urea	Cattle	0.49	capsule	Death	Roberts (1959)
Urea	Cattle	0.45	feed	death	

2.12 Recycling of urea in ruminants

Ruminants as well as other mammals synthesize urea which helps put a stop to excess N from becoming lethal. However, other tissues have the enzyme activity compulsory to urea production (Emmanuel, 1980). Once released into blood, urea is excreted in urine or reenters the digestive tract by diffusion into saliva or directly across the gut wall. Urea production, excretion and recycling to the gut are linked to diet composition, intake and productive priorities of the animal. Depending on those factors, 19 to 96% of endogenous urea production may be recycled to the gut, 15 to 94% of the recycling may transfer in saliva and 25 to 90% of urea degraded in the gut may be degraded in the postruminal digestive tract. Urea excreted in the urine represents from 25 to 60% of endogenous urea production in goats (Obara and Shimbayashi, 1980), sheep (Sarraseca *et al*., 1998), beef heifers (Bunting *et al*., 1989a) and beef steers (Huntington, 1989).

Ureagenesis in the liver is closely linked to degradability of dietary N and subsequent absorption of ammonia. Ruminants, especially those consuming living or harvested legumes or immature grasses depend on liver to detoxify portal blood that contains ammonia absorbed from the gut. Basically, N recycling provides a continuous source of ammonia to maintain microbial fermentation in the rumen as well as other regions of the digestive tract. Kennedy and Milligan (1980) listed ruminal ammonia concentration, organic matter digestibility and plasma concentration of urea as the most important factors affecting rate of endogenous urea transfer from blood to the lumen of the gastrointestinal tract.

2.13 Prevention of urea poisoning

Feeding urea less than l% of total ration or not more than 3% of the concentrate mixture is not a reason of toxicity (Davis and Roberts, 1959). Acetic acid has been found to be an effective therapeutic measure (Davis and Roberts, 1959; Repp *et al*., 1955b). Eventhough Rummler *et al*. (1962) indicated that glutamic acid was effective in overcoming the toxic symptoms. Oltjen *et al*. (1964) found it was inferior to acetic acid on an identical carboxyl basis in its ability to neutralize rumen ammonia.

2.14 Urea feeding and milk yield

Golombeski *et al*. (2006) reported that, the addition of slow release urea had no effect on daily milk yield, which is in agreement with the results of Galo *et al*. (2003). A parity effect (p=0.02) was also observed by him for milk yield, where multiparous cows produced 6.8 kg/d more milk than primiparous cows (29.5 vs. 22.7 kg, respectively). Promma *et al*. (1984) reported that urea-treated straw could increase milk yield when fed to lactating cows and to lactating goats (Djibrillou *et al*., 1998). Leng (1997) found an increase in milk yield of 30% due to UMMB supplementation for lactating dairy cows in India. Whereas, In a Vietnamese studies, supplementation of crossbred dairy cows with UMMB resulted in an 11% increase in milk yield (Duc Vu *et al*., 1999).

2.15 Dietary urea and blood parameter

Blood metabolic profile (BMP) is a set of diagnostic procedures that are based on determining the various indicators in the blood of animals (Van Saun, 2000). Biochemical tests are used to evaluate the internal body condition of the function of different organs and the metabolic processes inside the body (Scamell, 2006). In case of cattle, the concentration of glucose is considered as a vital indicator of energy metabolism. The main indicators of protein metabolism are urea and total protein. Liver condition is represented in the activity of serum glutamic oxaloacetic transaminase (SGOT), Serum glutamate-pyruvate transaminase (SGPT) and gamma-glutamyl transferase and total bilirubin concentration, whereas creatinine is the basic parameter reflecting kidney function (Stojevic *et al*., 2005).

Chapter-3: Materials and Methods

3.1 Study area

The study was carried out in the Upazila of Patiya under Chittagong District in the Division of Chittagong, Bangladesh. It is located at 22.30° North 91.98° east. It is bounded by Kotwali, Chandgaon and Boalkhali on the north, Chandanaish and Anwara on the south, Rangunia and Chandanaish on the east, Bandar on the west. It has 70218 units of household and total area 316.47 km². Wahed Dairy Farm located in Patia Upazila was selected for the study. Milk and blood samples were collected from the farm during the study period.

Figure 1. Map of the study area

3.2 Study period

The study was conducted during September 2014 to October 2014. In September temperature was 31.6^{o} C-25.6^{o} C; average humidity was 83% and average rainfall was 259.3mm. In October temperature was 31.5^{o} C-23.9^{o} C; humidity was 81% and rainfall was 184.8 mm (BMD, 2014).

3.3 Experimental animals

Twenty Local × Holstein (F_2) milking cows were selected from the selected farm. Animals were selected based on age, live weight, body condition score (BCS), daily milk yield (5-8 litter/day), number of lactation and period of pregnancy. Individual histories like body weight (average 422kg); lactation length (1-4) was collected from the record sheet. All selected cows ranged within a BCS of 3-4 in a 5 scale.

3.4 Design of experiment

In order to minimize the experimental error between different groups (control and treatment) animals were grouped in Randomized completely Block Design (RCBD) where animals were blocked in five dietary treatments (T_0, T_1, T_2, T_3 and T_4) based on days in milk (DIM), body weight, body condition score and lactation having four replications in each group. T_1 contained MCB with 0% urea, T_2 contained MCB with 25% urea, T_3 contained MCB with 35% urea and T_4 contained MCB with 45% urea.

3.5 Management of animals

The animals were kept in single row face out system stanchion barn with well ventilated condition and sufficient space to keep them comfortable. All animals under the experiment were given a tag with identity number. All animals were given respective manger and other cares were taken for good husbandry condition. Animal to animal distance was maintained properly to ensure the proper feed intake. The regular cleaning of cow was done by a hose pipe with fresh water. A good sanitary condition was maintained throughout the experimental period. The milking was done twice in a day (6 am and 5 pm) regularly. During milking period, workers were maintained proper bio-security to guarantee best quality milk. Adlibitum fresh drinking water was supplied during that time. A complete balance ration was given as a basal ration. The concentrate and roughage ration was maintained properly.

2.6 Preparation of Multinutrient Cattle Biscuit

Multinutrient Cattle Biscuit was made with different compositions for different treatment groups. The percentages of ingredients of the MCB are given in Table 1.

Table 3. Composition and nutritive value of MCB

Ingredients (%)	Dietary treatments			
	T_1	T_2	T_3	T_4
Urea	0	25.0	35.0	45.0
Molasses	12.5	10.0	10.0	7.5
Sugar	5.0	5.0	5.0	5.0
Wheat flour	25.0	17.5	20.0	17.5
Rice polish	10.0	5.0	5.0	5.0
Maize	15.0	10.0	5.0	5.0
Soybean meal	15.0	15.0	10.0	7.5
Rice powder	10.0	5.0	5.0	0.0
Salt	2.5	2.5	0.0	2.5
Minerals	5.0	5.0	5.0	5.0
Total amount	100	100	100	100
ME (Kcal/kg)	2155.8	1569.6	1319.3	1043.6
CP (%)	13.9	80.5	105.9	131.6

T_0=Diet without MCB; T_1=Diet containing 0% urea supplemented MCB; T_2=Diet containing 25% urea supplemented MCB; T_3=Diet containing 35% urea supplemented MCB; T_4=Diet containing 45% urea supplemented MCB

Flow chart for preparation of MCB

Granulated urea ground to fine powder by a hammer mill

↓

Fine powder dissolved in water and mixed with molasses. Stirring for a homogenous dough. The dough stored in a jar for overnight

↓

Other ingredients mixed in the jar in next morning. The dough placed on a mixing machine following a definite rotation per minute

↓

The dough placed into the molding machine, forced into moulds where the negative shape of the dough pieces with patterns, name, type and docker holes

↓

Excess dough scraped off with knife bearing upon the mould and extracted into a web of cotton canvas/other fabrics

↓

Multinutrient Cattle Biscuit placed on a biscuit tray and dried for a specific time. Artificial air flow used to reduce the temperature

↓

Cooling MCB, packed in an airtight packet for preservation

Figure 2. Preparation of MCB

Figure 3. Heating tray

Figure 4. Packaging of MCB

Figure 5. Sampling of MCB

3.7 Feeding of animals

All animals were stall fed under single row face out system stanchion barn house. Ration was supplied to the animal based on its maintenance and milk production. Multinutrient Cattle Biscuit was fed to the experimental animals as per recommendation of Agricultural Research Council (ARC, 1980). All animals had free access to normal clean drinking water. MCB will was fed twice daily before milking in the morning and one hour before milking in the afternoon. Three MCBs was given to the animals during every feeding time. Intake of basal ration was recorded every day. Ration was provided to as per body weight and milk yield basis. Roughage and concentrate ratio was maintained properly. Green and dry roughage was provided to the animal as calculating the daily basal energy and protein requirement. There

was available green grass for the dairy cows near the farm. The cultivated high yielding German grass (*Echinochloa polystachya*) was supplied to the stanchion barn daily. Green grass, rice straw and concentrate were given to the animals as per requirement. Concentrate mixture was prepared with rice polish, broken rice, broken maize, wheat barn, molasses, soybean meal, mustard oil cake, pea barn, Di calcium phosphate (DCP), whereas the roughage feed ingredients were German grass and straw. The concentrate mixture was made by the following ingredients.

Table 4. Concentrate mixture for experimental animals

Ingredient	Amount (Kg)
Rice polish	40.37
Wheat barn	35.13
Broken maize	5.70
Soybean meal	5.70
Mustard oil cake	5.70
Mug powder	5.70
Di calcium phosphate	0.28
Growth Gold	0.57
Salt	0.85
Total	100

Figure 6. Feeding green forage

Figure 7. Offering MCB

Figure 8. Preparation of concentrate

Figure 9. Offering concentrate

3.8 Measuring body weight

Body weight was measured in all cows at the beginning of the study by using Shaffer's method with the help of a measuring tape which was used by Khan *et al*. (2004) and Moaeen-ud-Din *et al.* (2006) in small ruminant; Mcnitt (1983) in equine; Uddin *et al.* (2002) in buffalo; Alam *et al.* (2009) and Kamal *et al.* (2009) in deshi cows: [Body length (inch) x {Heart girth (inch)}^{2}] / 300 = Body weight (lb)

Body weight (lb) / 2.2 = Body weight (kg)

3.9 Measurement of milk yield

The milk yield was recorded carefully during the experimental period. A digital weight machine was used to record the milk yield. The weight of the empty milk bucket was taken at first. Then the milk containing bucket was measured by the digital weighing machine. By subtracting the weight of empty bucket from the milk containing bucket the amount of the milk was determined and recorded immediately in a milk register book. This procedure was maintained every time in morning and evening milking for milk collection during the experimental period.

3.10 Sampling of feed, blood and milk

The following sampling strategy was adopted for the collection of milk samples from the dairy farm. Approximately 100 gm of feed sample was taken from the farm and preserved in an air tight bag to carry away in the laboratory during the experimental period. Rice polish, wheat barn, soybean meal, broken maize, pea barn, molasses etc feed samples were collected directly from the farm and analyzed for dry matter (DM), crude fiber (CF), Crude protein (CP), ether extract and ash as per AOAC (2006). Blood samples were collected directly from jugular vein through syringe. Blood samples were collected in vacutainer tube. Samples were carried to the laboratory by using ice box and kept in a freezer at a temperature of -20°C. Four ml blood was collected as blood sample from the each experimental animal group and continued for 8 weeks. Every blood sample was given unique identification number. Milk samples were collected from individual group animal. 20 milk samples were collected every week. The milk sample collection was continued for 8 weeks. Approximately, 200 ml of milk sample was collected by individual bottle. Each milk sample was given unique identification number. Then the samples were transported to the laboratory by using ice box.

3.11 Analysis of milk sample

Without freezing milk sample, it was analyzed for fat, protein, lactose, total solids (TS), solid not fat (SNF) and mineral by using milk analyzer (Lactostar, Funke-Gerber, Berlin, Germany) on the day of milk sample collection. Lactostar adopted a combined thermo-optical procedure for determining milk components. This device measured both thermal and optical qualities of the milk constituents. Optical measuring procedure (turbidimetry) was based on the fact that all the colloidal and emulsified substances contributed to turbidity. By measuring turbidity, a sum of fat content and protein content was obtained. Thermo-analysis measured the fat content and SNF content of the sample through thermo physical effects and their arithmetical evaluation. Protein content was assessed by forming the difference between the results of optical measurement and the fat content thermodynamically via computational analysis. Before analyzing the milk sample Lactostar was calibrated well. It was recommended to carry out a zero calibration once per week. Multiple rinsing with distilled water was carried out until water in the disposal tube being clear. After the zero calibration milk sample was analyzed and the result was recorded carefully in every week.

Figure 10. Collection of milk sample

Figure 11. Recording lactometer reading

Figure 12. Calibration of milk analyzer

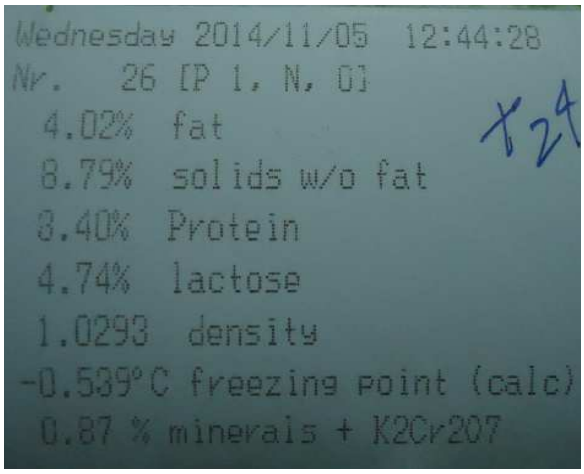

Figure 13. Report of milk test

3.12 Analysis of blood sample

Clotted blood in the vacutainer tube was centrifuged at 3000 rpm for 20 minutes and prepared serum was collected into the ependroff tube by micropipette. Sera were marked and stored in -20°C until being analyzed for glucose, total protein, urea, creatinine, albumin, serum glutamic oxaloacetic transaminase (SGOT), serum glutamate-pyruvate transaminase (SGPT) by HumaLyzer 3000 (Wisbaden,Germany). It was semi-automatic machine, microprocessor-controlled photometer with large graphic LCD screen. Randox® veterinary reagent kits were used for determination of the blood parameter of interest. Serum sample was mixed with the respective reagents with a specified time (as per manual) in an ependroff tube. Then the serum with reagent was aspired by the machine. By the spectrophotometric method which measured the target parameter and immediately the printed result was recorded in the blood parameter sheet.

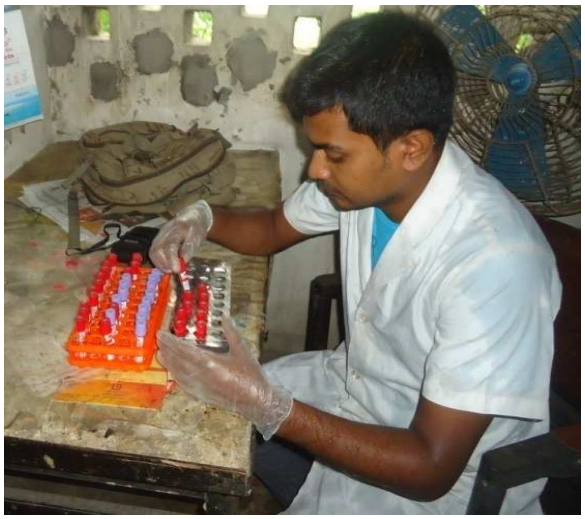

Figure 14. Collection of blood

Figure 15. Centrifuge of blood

Figure 16. Collection of serum

Figure 17. Mixing reagents with serum

Figure 18. Deep freezing of serum

Figure 19.Cheeking blood parameter

3.13 Statistical analysis

Data related to milk yield, milk composition and blood parameters were collected and compiled by using Microsoft Excel 2007 and analyzed for one way ANOVA (Winer *et al.*, 1991) by using Stata/IC-11.0 and SPSS 16.0. Means showing significant differences was compared by Dunnet Test (Duncan, 1955). Statistical significance was accepted as $p<0.05$

Chapter-4: Results

The present experiment was carried out to quantify the effect of MCB on the yield and composition of milk and blood parameters of Holstein-Friesian crossbred cows in a selected dairy farm. The results obtained from the study have been described in this chapter.

4.1 Milk yield

Milk yield of the experimental cows were recorded during 60 days of the experimental period (Table 5). Results indicated that, the daily milk yield of the cows in the experimental groups (T_1, T_2, T_3 and T_4) supplemented with varied levels of MCB had higher average milk yield (7.5, 7.8, 8.3 and 7.4 kg/d) than the control group (6.3 kg/d). The highest milk yield (8.9 kg/d) was recorded in 7^{th} and 8^{th} week in the T_3 group. Average daily milk yield did not differ significantly ($p>0.05$) among the all five dietary treatment groups irrespective of MCB supplementation for the first four weeks. However, the trend of milk yield appeared to increase from 1^{st} to 4^{th} week. As a consequence, milk yield differed significantly ($p<0.05$) from 5^{th} to 8^{th} week among all dietary treatment groups as the level of MCB supplementation increased from 0 to 45%. At the end of the experimental period, among all the treatment groups highest average milk yield (8.3kg/d) was observed in T_3 group and the lowest average milk yield (6.1 kg/d) was recorded in the T_0 group.

Table 5. Milk yield (Kg/d/cow) of experimental cows fed diets supplemented with MCB from 1^{st} to 8^{th} week

Weeks	Dietary treatments					SE	Sig.
	T_0	T_1	T_2	T_3	T_4		
1^{st}	6.3	7.3	7.0	7.4	6.5	0.2	NS
2^{nd}	6.4	7.3	7.3	7.7	6.7	0.2	NS
3^{rd}	6.4	7.3	7.5	8.1	7.0	0.2	NS
4^{th}	6.3	7.5	7.8	8.2	7.3	0.2	NS
5^{th}	6.3	7.7	8.0	8.3	7.5	0.2	*
6^{th}	6.1	7.6	8.0	8.7	7.9	0.3	*
7^{th}	6.5	7.7	8.2	8.9	8.1	0.3	*
8^{th}	6.4	7.9	8.3	8.9	8.2	0.3	*
Overall	6.3	7.5	7.8	8.3	7.4	0.2	*

T_0=Diet without MCB; T_1=Diet containing 0% urea supplemented MCB; T_2=Diet containing 25% urea supplemented MCB; T_3=Diet containing 35% urea supplemented MCB; T_4=Diet containing 45% urea supplemented MCB; SE=Standard Error; NS=Non-Significant ($p>0.05$); *=Significant ($p<0.05$)

4.2 Milk fat

Milk fat percentage of the experimental cows varied in an irregular fashion during eight weeks of the experimental period (Table 6). It was found that fat percent in milk significantly ($p<0.05$) increased during 1st, 2nd, 3rd, 7th and 8th weeks in the dietary treatment groups. In contrast, it was statistically similar ($p>0.05$) during 4th, 5th and 6th week. The highest milk fat (5.4%) was recorded in the 2nd week in T_2 group. The lowest milk fat (2.7%) was recorded in 1st week in T_1 and T_2 groups jointly. The best average milk fat (3.9%) among the entire treatment group was T_2 group at the end of the experimental period (1st to 8th week).

Table 6. Fat percent in the milk of the experimental of cows fed diets supplemented with MCB from 1st to 8th week

Weeks	Dietary treatments					SE	Sig.
	T_0	T_1	T_2	T_3	T_4		
1st	3.2	2.7	2.7	3.3	3.4	0.1	*
2nd	4.7	5.3	5.4	4.6	4.4	0.2	*
3rd	3.2	3.3	3.4	3.5	3.5	0.0	*
4th	4.4	3.7	4.0	4.0	3.6	0.1	NS
5th	3.3	3.3	3.4	3.5	3.5	0.0	NS
6th	3.5	3.3	5.0	3.8	3.3	0.3	NS
7th	3.3	3.3	3.4	3.6	3.7	0.1	*
8th	3.0	3.3	3.8	3.4	3.3	0.1	*
Overall	3.6	3.5	3.9	3.7	3.6	0.1	*

T_0=Diet without MCB; T_1=Diet containing 0% urea supplemented MCB; T_2=Diet containing 25% urea supplemented MCB; T_3=Diet containing 35% urea supplemented MCB; T_4=Diet containing 45% urea supplemented MCB; SE=Standard Error; NS=Non-Significant ($p>0.05$); *=Significant ($p<0.05$)

4.3 Milk protein

The protein percent of milk increased significantly in the 1st ($p<0.001$); 2nd, 5th, 8th ($p<0.05$) and 7th ($p<0.01$) week (Table 7). However, the trend was non-significant in 3rd, 4th and 6th week. The highest protein percent (4.1%) was recorded in the 8th week in T_2 group and the lowest protein percent (2.2%) was recorded in the 1st week in T_1 group. The highest average protein percentage (3.5%) was observed in the T_1 and T_2 group equally and lowest (3.2%) in the T_0 group during 1st to 8th weeks of experimental period.

Table 7. Protein percent in the milk of the experimental of cows fed diets supplemented with MCB from 1st to 8th week

Weeks	Dietary treatments					SE	Sig.
	T_0	T_1	T_2	T_3	T_4		
1st	2.6	2.2	3.1	2.9	3.4	0.1	***
2nd	3.5	4.0	4.0	3.5	3.9	0.2	*
3rd	3.0	3.2	3.2	3.3	3.3	0.0	NS
4th	3.5	3.3	3.4	3.4	3.2	0.0	NS
5th	3.1	3.2	3.2	3.4	3.2	0.0	*
6th	3.6	3.6	3.8	3.6	3.5	0.0	NS
7th	2.9	3.4	3.3	3.5	3.7	0.1	**
8th	3.3	3.2	4.1	3.2	3.3	0.1	*
Overall	3.2	3.5	3.5	3.4	3.4	0.1	*

T_0=Diet without MCB; T_1=Diet containing 0% urea supplemented MCB; T_2=Diet containing 25% urea supplemented MCB; T_3=Diet containing 35% urea supplemented MCB; T_4=Diet containing 45% urea supplemented MCB; SE=Standard Error; NS=Non-Significant ($p>0.05$); *=Significant ($p<0.05$); **=Significant ($p<0.01$); ***=Significant ($p<0.001$)

4.4 Milk solids-not-fat

The SNF percent differed significantly in the 1st, 2nd and 5th ($p<0.01$); 3rd and 7th ($p<0.001$) week (Table 8). However, the difference was non-significant ($p>0.05$) in 4th, 6th and 8th week. The highest SNF percent (9.7%) was recorded in the T_1 group in 2nd week. The lowest SNF percent (8.3%) was recorded in the T_3 and T_4 group in 8th and 4th week respectively. The highest average value of SNF (9.0%) was observed in the T_2 group at the end of the 1st to 8th weeks of experiment.

Table 8. SNF percent in the milk of the experimental of cows fed diets supplemented with MCB from 1st to 8th week

Weeks	Dietary treatments					SE	Sig.
	T_0	T_1	T_2	T_3	T_4		
1st	8.9	8.8	8.9	9.0	9.1	0.0	**
2nd	9.3	9.7	9.5	9.1	9.2	0.1	**
3rd	8.8	8.9	8.9	9.0	9.0	0.0	***
4th	8.9	8.6	8.7	8.7	8.3	0.1	NS
5th	8.9	8.9	8.9	9.1	9.1	0.0	**
6th	9.0	9.1	9.5	9.0	8.9	0.1	NS
7th	8.9	8.9	8.9	9.0	9.2	0.0	***
8th	8.6	8.4	8.8	8.3	8.7	0.1	NS
overall	8.9	8.9	9.0	8.9	8.9	0.1	NS

T_0=Diet without MCB; T_1=Diet containing 0% urea supplemented MCB; T_2=Diet containing 25% urea supplemented MCB; T_3=Diet containing 35% urea supplemented MCB; T_4=Diet containing 45% urea supplemented MCB; SE=Standard Error; NS=Non-Significant ($p>0.05$); **=Significant ($p<0.01$); ***=Significant ($p<0.001$)

4.5 Milk total solids

TS percent of milk differed significantly in the 1st, 7th ($p<0.01$); 3rd ($p<0.001$) and 5th ($p<0.05$) week although it was similar ($p>0.05$) in 2nd, 4th, 6th and 8th week (Table 9). The highest TS percent (15.5%) was estimated in the T_1 group in 2nd week. The lowest TS percent (11.5%) was estimated in the T_1 group in 1st week. The highest average value of TS (12.9%) throughout the experimental period was found in T_2 group.

Table 9. TS percent in the milk of the experimental of cows fed diets supplemented with MCB from 1st to 8th week

Weeks	Dietary treatments					SE	Sig.
	T_0	T_1	T_2	T_3	T_4		
1st	12.0	11.5	11.6	12.4	12.5	0.1	**
2nd	13.9	15.5	14.8	13.7	13.9	0.3	NS
3rd	12.1	12.2	12.3	12.5	12.6	0.0	***
4th	13.3	12.3	12.7	12.7	11.9	0.1	NS
5th	12.1	12.2	12.3	12.5	12.6	0.1	*
6th	12.5	12.4	14.6	12.8	12.2	0.0	NS
7th	12.2	12.1	12.4	12.6	12.9	0.1	**
8th	11.6	11.6	12.6	11.6	12.0	0.1	NS
Overall	12.5	12.5	12.9	12.6	12.6	0.1	NS

T_0=Diet without MCB; T_1=Diet containing 0% urea supplemented MCB; T_2=Diet containing 25% urea supplemented MCB; T_3=Diet containing 35% urea supplemented MCB; T_4=Diet containing 45% urea supplemented MCB; SE=Standard Error; NS=Non-Significant ($p>0.05$); *=Significant ($p<0.05$); **=Significant ($p<0.01$); ***=Significant ($p<0.001$)

Table 10. Multiple correlation co-efficient matrix of milk components of the experimental cows fed diets supplemented with MCB from 1st to 8th week

Parameter	Fat %	Protein %	SNF %	TS %
Fat %	1.00			
Protein %	.76*	1.00		
SNF %	.802*	0.50	1.00	
TS %	.98**	.72*	.89**	1.00

SNF=Solids not fat; TS=Total solids; *=Significant ($p<0.05$); **=Significant ($p<0.01$)

4.6 Correlation co-efficient matrix

Table 10 showed the multiple correlation co-efficient matrix among the milk parameters estimated for all the experimental cows. Positive significant ($p<0.01$) correlations were

observed between fat to TS % and SNF to TS % of milk. Positive significant ($p<0.05$) correlations were also observed among fat, protein, SNF % and protein to TS %. However, the relationship between protein % and SNF % was statistically non-significant ($p>0.05$).

4.7 Serum cholesterol

There was no significant difference ($p>0.05$) in serum cholesterol. However, cholesterol level was moderately higher than the normal value both in the treatment and controls groups. The highest average value of serum cholesterol (301.8) was found in T_2 group whereas the lowest value (285.2) was found in the T_3 group during the experimental period (1^{st} to 8^{th} week).

Table 11. Cholesterol level (mg/dl) in the Blood serum of the experimental cows fed diets supplemented with MCB at 1^{st} week to 8^{th}

Weeks	Dietary treatments					SE	Sig.
	T_0	T_1	T_2	T_3	T_4		
1^{st}	261.5	315.4	253.8	341.8	355.3	23.0	NS
2^{nd}	272.3	321.2	306.5	290.9	311.6	9.6	NS
3^{rd}	327.7	270.7	264.1	250.0	277.1	14.8	NS
4^{th}	316.3	297.3	286.6	300.2	309.8	5.7	NS
5^{th}	230.2	235.5	340.8	279.0	293.6	22.7	NS
6^{th}	287.7	326.9	315.0	232.9	274.8	18.5	NS
7^{th}	354.8	317.1	263.6	291.0	301.1	16.9	NS
8^{th}	252.4	330.6	252.1	296.4	278.9	16.5	NS
overall	287.8	301.8	285.3	285.2	300.3	4.1	NS

T_0=Diet without MCB; T_1=Diet containing 0% urea supplemented MCB; T_2=Diet containing 25% urea supplemented MCB; T_3=Diet containing 35% urea supplemented MCB; T_4=Diet containing 45% urea supplemented MCB; SE=Standard Error; NS=Non-Significant ($p>0.05$)

4.8 Serum glutamic oxaloacetic transaminase

The SGOT (U/L) appeared statistically non-significant during the experimental period (Table 12). However in 5^{th} week, SGOT was significantly ($p<0.05$) low in the treatment groups compared to the control group. The level of SGOT was typical in the treatment group. At the end of the eight weeks experimental period, highest serum SGOT average value (105.8) was found in T_1 group whereas the average value (92.1) found in T_4 group but both of them lies in between the normal range.

Table 12. SGOT level (U/L) in the Blood serum of the experimental cows fed diets supplemented with MCB at 1st week to 8th

Weeks	Dietary treatments					SE	Sig.
	T_0	T_1	T_2	T_3	T_4		
1st	107.4	191.0	111.3	163.2	201.2	21.9	NS
2nd	142.9	152.9	101.9	77.6	74.5	18.2	NS
3rd	57.4	61.7	80.2	53.2	77.4	6.1	NS
4th	66.2	54.8	86.8	69.9	89.9	7.3	NS
5th	143.6	72.6	125.4	111.6	76.3	15.5	*
6th	111.5	76.3	73.4	108.3	65.4	10.7	NS
7th	90.3	153.3	100.4	136.8	74.0	16.5	NS
8th	88.8	84.4	102.8	75.3	78.3	5.4	NS
Overall	101.0	105.8	97.8	99.5	92.1	2.5	NS

T_0=Diet without MCB; T_1=Diet containing 0% urea supplemented MCB; T_2=Diet containing 25% urea supplemented MCB; T_3=Diet containing 35% urea supplemented MCB; T_4=Diet containing 45% urea supplemented MCB; SE=Standard Error; NS=Non-Significant ($p>0.05$); **=Significant ($p<0.01$)

4.9 Serum glutamate pyruvate transaminase

The SGPT level (U/L) remained non-significant during the experimental period (Table 13). The maximum average of SGPT level (34.4) was found in T_2 group; whereas the minimum level (30.3) was found in T_1 group and T_4 group jointly. The level of SGPT was typical in the treatment group.

Table 13. SGPT level (U/L) in the Blood serum of the experimental cows fed diets supplemented with MCB at 1st week to 8th

Weeks	Dietary treatments					SE	Sig.
	T_0	T_1	T_2	T_3	T_4		
1st	23.5	29.1	31.9	20.7	23.3	2.3	NS
2nd	29.3	35.9	36.8	23.5	26.6	2.9	NS
3rd	29.5	26.4	37.2	39.6	26.7	3.1	NS
4th	37.8	27.1	42.0	40.5	37.7	2.9	NS
5th	32.0	35.0	38.7	32.7	25.5	2.4	NS
6th	31.6	29.6	43.3	35.2	31.6	2.7	NS
7th	47.6	32.4	23.4	28.1	40.7	4.9	NS
8th	28.7	26.9	21.7	31.0	30.4	1.9	NS
Overall	32.5	30.3	34.4	31.4	30.3	0.9	NS

T_0=Diet without MCB; T_1=Diet containing 0% urea supplemented MCB; T_2=Diet containing 25% urea supplemented MCB; T_3=Diet containing 35% urea supplemented MCB; T_4=Diet containing 45% urea supplemented MCB; SE=Standard Error; NS=Non-Significant ($p>0.05$); **=Significant ($p<0.01$)

4.10 Serum bilirubin

The serum bilirubin appeared normal and did not differ significaty during the experimental period (1st to 8th week). The highest average value of serum bilirubin (0.17) was found in the T_4 group and lowest value (0.14) was found in the T_0 group.

Table 14. Bilirubin level (mg/dl) in the Blood serum of the experimental cows fed diets supplemented with MCB at 1st week to 8th

Weeks	Dietary treatments					SE	Sig.
	T_0	T_1	T_2	T_3	T_4		
1st	0.10	0.12	0.15	0.20	0.20	0.02	NS
2nd	0.12	0.15	0.15	0.15	0.15	0.00	NS
3rd	0.15	0.12	0.14	0.10	0.15	0.00	NS
4th	0.18	0.15	0.14	0.17	0.20	0.10	NS
5th	0.15	0.17	0.15	0.14	0.15	0.30	NS
6th	0.14	0.20	0.20	0.15	0.20	3.10	NS
7th	0.16	0.15	0.10	0.10	0.10	0.10	NS
8th	0.15	0.10	0.15	0.15	0.20	0.00	NS
Overall	0.14	0.15	0.15	0.15	0.17	0.45	NS

T_0=Diet without MCB; T_1=Diet containing 0% urea supplemented MCB; T_2=Diet containing 25% urea supplemented MCB; T_3=Diet containing 35% urea supplemented MCB; T_4=Diet containing 45% urea supplemented MCB; SE=Standard Error; NS=Non-Significant ($p>0.05$); **=Significant ($p<0.01$)

4.11 Serum urea

The serum urea level remained typical. The highest average value of serum urea (21.7) was found in the T_1 group in contrast the lowest average value of serum urea (19.3) was found in the T_0 group and T_2 group jointly.

Table 15. Urea level (mg/dl) in the Blood serum of the experimental cows fed diets supplemented with MCB at 1st week to 8th

Weeks	Dietary treatments					SE	Sig.
	T_0	T_1	T_2	T_3	T_4		
1st	28.0	25.9	28.6	26.8	28.3	2.1	NS
2nd	32.6	35.7	29.6	29.3	12.7	3.1	NS
3rd	23.4	12.8	12.2	19.8	16.2	0.0	NS
4th	17.2	12.4	13.8	19.1	17.5	1.4	NS
5th	17.7	25.1	19.5	15.5	14.4	2.2	NS
6th	12.4	20.6	17.2	10.8	30.5	3.1	NS
7th	12.3	20.7	14.8	19.9	18.5	1.2	NS
8th	11.2	20.4	19.0	21.1	19.1	1.7	NS
Overall	19.3	21.7	19.3	20.3	19.6	1.9	NS

T_0=Diet without MCB; T_1=Diet containing 0% urea supplemented MCB; T_2=Diet containing 25% urea supplemented MCB; T_3=Diet containing 35% urea supplemented MCB; T_4=Diet containing 45% urea supplemented MCB; SE=Standard Error; NS=Non-Significant ($p>0.05$); **=Significant ($p<0.01$)

4.12 Serum creatinine

Table 16 represented the serum creatinine level (mg/dl) of experimental dairy cows. In the 2nd week, blood parameters of the dairy cows remained same except creatinine. The creatinine level significantly ($p<0.01$) decreased among the dietary treatment groups than the control group. The mean value for creatinine was 3.4, 3.4, 2.2, 2.4 and 2.9 in T_0, T_1, T_2, T_3 and T_4 groups, respectively which was slightly higher than the normal value (1-2). Similar statistical significance trend was also observed in the 3rd week. The creatinine level significantly decreased ($p<0.05$) among the treatment groups compare to the control group. After the 8 week of observation the highest average (2.1) value of creatinine was observed in the T_0 group and lowest (1.8) in the T_2 and T_3 group equally.

Table 16. Creatinine level (mg/dl) in the Blood serum of the experimental cows fed diets supplemented with MCB at 1st week to 8th

Weeks	Dietary treatments					SE	Sig.
	T_0	T_1	T_2	T_3	T_4		
1st	3.6	3.5	2.8	1.4	2.4	0.3	NS
2nd	3.4	3.4	2.2	2.4	2.9	0.2	**
3rd	1.8	1.6	1.5	1.6	1.0	0.1	*
4th	1.1	1.3	1.1	1.3	1.5	0.1	NS
5th	1.5	1.8	1.8	1.4	1.4	0.3	NS
6th	1.3	1.7	2.0	1.6	1.9	0.1	NS
7th	2.1	1.6	2.1	2.9	2.4	0.2	NS
8th	2.4	1.1	1.2	1.7	1.9	0.2	NS
Overall	2.1	2.0	1.8	1.8	1.9	0.2	NS

T_0=Diet without MCB; T_1=Diet containing 0% urea supplemented MCB; T_2=Diet containing 25% urea supplemented MCB; T_3=Diet containing 35% urea supplemented MCB; T_4=Diet containing 45% urea supplemented MCB; SE=Standard Error; NS=Non-Significant ($p>0.05$); **=Significant ($p<0.01$)

4.13 Serum protein

The total protein (g/dl) was statistically non-significant (Table 17). The maximum average value of serum protein (10.6) was observed in T_4 group and the minimum average value (9.0) was observed in the T_0 group. During the experimental period the total protein level was slightly higher in the treatment group than the normal value.

Table 17. Total protein level (g/dl) in the Blood serum of the experimental cows fed diets supplemented with MCB at 1st week to 8th

Weeks	Dietary treatments					SE	Sig.
	T_0	T_1	T_2	T_3	T_4		
1st	10.8	10.9	8.5	9.5	10.9	0.5	NS
2nd	8.9	11.5	10.6	11.5	12.1	0.5	NS
3rd	8.3	9.9	8.9	8.1	6.9	0.5	NS
4th	8.6	8.3	8.7	9.8	10.5	0.6	NS
5th	8.3	9.5	10.4	10.4	11.5	0.6	NS
6th	8.9	9.4	9.1	9.4	12.8	0.8	NS
7th	8.7	9.8	9.5	10.4	7.8	0.8	NS
8th	10.0	13.5	13.4	13.1	12.6	0.8	NS
Overall	9.0	10.3	9.9	10.3	10.6	0.6	NS

T_0=Diet without MCB; T_1=Diet containing 0% urea supplemented MCB; T_2=Diet containing 25% urea supplemented MCB; T_3=Diet containing 35% urea supplemented MCB; T_4=Diet containing 45% urea supplemented MCB; SE=Standard Error; NS=Non-Significant ($p>0.05$); **=Significant ($p<0.01$)

4.14 Serum glucose

The serum glucose level (mg/dl) has been presented in the Table 18. Serum glucose level was highly significant ($p<0.01$) in the 1st week and moderately significant in 5th week ($p<0.05$). As a consequence, blood serum glucose level was very strongly significant in the 8th week among the treatment groups compare to the control group. At the end of the experiment the glucose was found statistically significant ($p<0.01$) in 1st to 8th week. The lowest average value of serum glucose (68.0) found in the T_3 group and the highest average value (78.0) was observed in the T_4 group.

Table 18. Glucose level (mg/dl) in the Blood serum of the experimental cows fed diets supplemented with MCB at 1st week to 8th

Weeks	Dietary treatments					SE	Sig.
	T_0	T_1	T_2	T_3	T_4		
1st	45.3	45.6	44.9	40.5	56.8	1.3	**
2nd	46.3	47.3	52.8	47.2	68.9	6.8	NS
3rd	75.4	95.0	90.6	83.9	85.8	10.0	NS
4th	77.3	85.6	84.6	62.0	75.0	8.3	NS
5th	80.6	88.7	70.5	87.0	85.0	6.2	*
6th	71.3	85.4	68.2	80.5	87.2	7.8	NS
7th	75.0	85.0	87.9	67.4	83.0	11.0	NS
8th	75.0	71.8	68.9	76.0	85.0	10.4	***
Overall	68.3	75.5	71.0	68.0	78.3	7.7	**

T_0=Diet without MCB; T_1=Diet containing 0% urea supplemented MCB; T_2=Diet containing 25% urea supplemented MCB; T_3=Diet containing 35% urea supplemented MCB; T_4=Diet containing 45% urea supplemented MCB; SE=Standard Error; NS=Non-Significant ($p>0.05$); *=Significant ($p<0.05$); **=Significant ($p<0.01$) ***=Significant ($p<0.001$)

4.15 Correlation co-efficient matrix

Table 19 showed the correlation co-efficient matrix among the serum parameters calculated for the all cows in the experiment. Very strong (-0.88) significant ($p<0.01$) negative correlation was observed between urea and glucose. Significant ($p<0.05$) negative correlation (-0.74) was also observed between creatinine and glucose. On the other hand, cholesterol, SGPT, SGOT, bilirubin and total protein were not significantly correlated with each other serum parameter and appeared to remain non-significant ($p>0.05$) throughout the whole experimental period.

Table 19. Multiple correlation co-efficient matrix of blood parameters of the experimental cows fed diets supplemented with MCB from 1st to 8th week

Parameter	Cholesterol	SGOT	SGPT	Bilirubin	Urea	Creatinine	T. protein	Glucose
Cholesterol	1.00							
SGOT	0.16	1.00						
SGPT	0.67	-0.36	1.00					
Bilirubin	0.20	-0.21	-0.09	1.00				
Urea	0.08	0.51	-0.07	-0.30	1.00			
Creatinine	0.01	0.31	-0.05	0.26	0.69	1.00		
Total protein	-0.07	0.34	-0.57	-0.21	0.14	-0.37	1.00	
Glucose	0.10	-0.42	0.05	0.41	-0.88**	-0.74*	0.06	1.00

SGOT=Serum glutamic oxaloacetic transaminase; SGPT=Serum glutamate-pyruvate transaminase *=Significant ($p<0.05$); **=Significant ($p<0.01$)

4.16 Serum glucose and milk parameter

There was a positive relationship among blood glucose and milk parameters of the experimental cows. For one unit increase in blood glucose level, milk fat, milk protein, milk SNF and milk TS was supposed to be increased by 0.002, 0.002, 0.003 and 0.002 unit and vice versa. However, regression coefficient (R^2) was extremely low (0.011-0.118) in all cases.

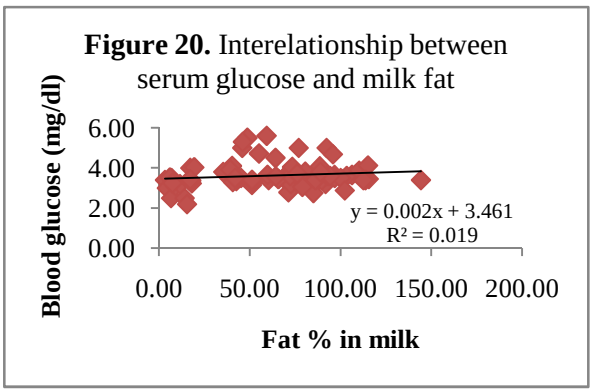

Figure 20. Interelationship between serum glucose and milk fat

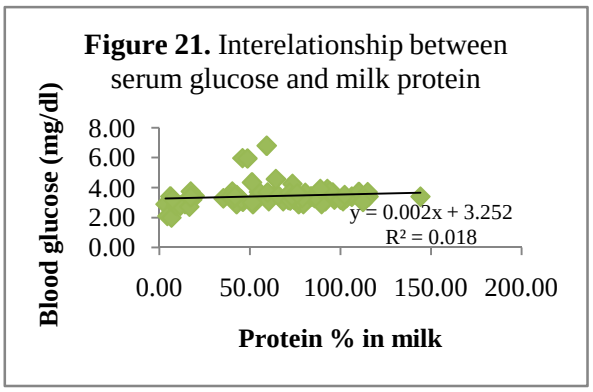

Figure 21. Interelationship between serum glucose and milk protein

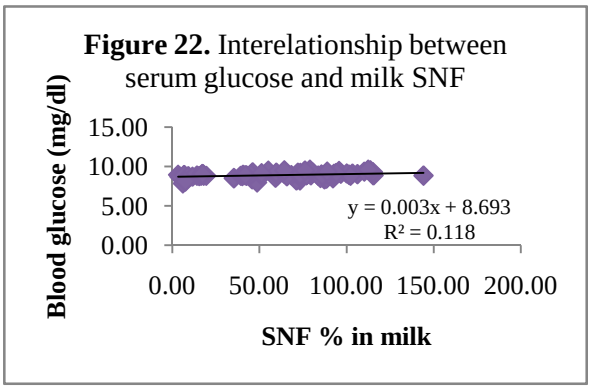

Figure 22. Interelationship between serum glucose and milk SNF

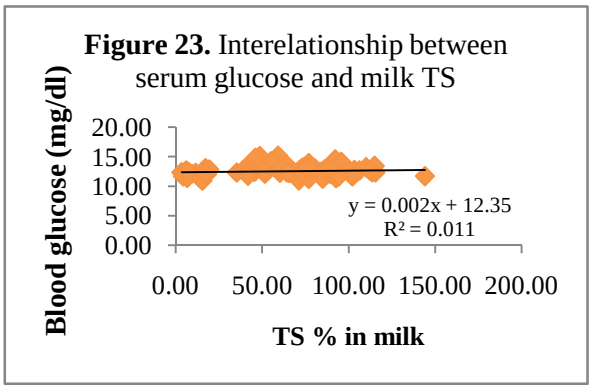

Figure 23. Interelationship between serum glucose and milk TS

Chapter-5: Discussion

Deficiency of nitrogen and minerals in cattle ration results due to feeding poor quality forages and mineral supplements. In present study, MCB was supplemented to the basal diet for incorporating additional nitrogen and minerals. The effect of MCB on milk yield, milk composition and blood parameters were investigated. It was evident that, milk yield differed significantly ($p<0.05$) in the treatment groups for the last four weeks. Similar result was reported by Mapato *et al.* (2010) who offered Holstein Friesian lactating cows urea treated straw. In another study, Duc Vu *et al.* (1999) obtained significantly ($p<0.05$) better daily milk yield in crossbred Holstein-Friesian cattle fed urea-treated rice straw. Additionally, increase in milk yield in crossbred cows were also in close agreement with other investigators (Alam *et al*., 2006; Chowdhury, 2004; Ferdous *et al*., 2007; Mazed, 1997; Miah *et al.*, 2000). The inherent reason for increased milk yield was described by Wanapat (1999) who reported that UTRS improved digestibility of nutrients, feed intake and fermentation endproducts which in terms resulted in increased milk production.

In contrast with previous finding, Wanapat *et al.* (2009) did not find any change ($p>0.05$) in milk yield by using treated rice straw with urea or urea and calcium hydroxide. Using conventional urea at different levels in dietary ration Erb *et al.* (1975) did not find any significant difference in milk yield. However, Erb *et al.* (1975) reported that, milk production was depressed by feeding conventional urea, eventhough, total feed intake was similar among control and treatment groups. In another study, Erb *et al.* (1975) showed that cows fed 180 g urea through concentrate mixture with corn silage as the sole source of forage produced less milk than those on conventional protein sources. Conversely, intake of urea in excess of 220 g per day did not depress milk yield when urea was added to corn silage during ensiling time (Polan *et al.* 1968).

Golombeski *et al.* (2006) reported that, the addition of slow release urea had no effect on daily milk yield in lactating dairy cow. Gonçalves *et al.* (2014) in another study, reported that 100% conventional urea in dairy cows significantly ($p<0.05$) reduced milk production. However, there was no remarkable changes ($p>0.05$) in milk production for treatments using 0, 44 and 88% coated urea. Souza *et al.* (2010) reported similar findings who offered coated urea to the lactating Holstein cows.

In current study, milk yield was similar ($p>0.05$) for the first four weeks because the cows might have used that time as adaptation period. However, significant ($p<0.05$) changes in milk yield at later stages were evident due to the effect of MCB. The best average milk yield was obtained from cows fed 35% urea supplemented MCB which could be due to the amount of blended cereal grains and molasses used in that group which released urea slowly and provided the ruminal microbes sufficient energy and minerals to utilize urea offered through MCB. It could also be inferred that, appropriate combination and composition of the nutrients specially energy, protein, fat and minerals in the MCB used in 35% urea supplemented MCB group might have triggered cows to exhibit best milk yield performance.

In lactating cows, milk fat is usually affected by physiological and environmental factors (Doreau *et al.,* 1999; Grummer, 1991; Palmquist *et al.,* 1993; Sutton, 1989). Casper and Schingoethe (1986) reported an unexplained decrease in milk fat percentage for cows fed urea. In contrast, Gonçalves *et al.* (2014) reported reasonably fair fat percent of milk (4.0%) with the experimental diet using 100% conventional urea. Another investigators (Galo *et al.,* 2003; Van Horn *et al.,* 1967) reported that, the milk fat percentage was unchanged by addition of urea in the diet of lactating cow.

Susmel *et al.* (1995) and Wanapat *et al.* (2009) reported that milk protein percentage increased significantly in urea supplemented diets. Xin *et al.* (2010) reported that the polyurethane coated urea diet significantly ($p<0.04$) increased milk protein than Feed-grade urea diet. These observations are in close agreement with current finding. The SNF percent was unaffected ($p>0.05$) in 4^{th}, 6^{th} and 8^{th} week which is in well agreement with Wanapat *et al.* (2009). The TS percent was non-significant in 3^{rd}, 4^{th}, 6^{th} and 8^{th} week in the treatment groups compared to the control group. Similar finding was reported by Golombeski *et al.* (2006).

Santos *et al*. (2011) reported no difference in milk composition of cows fed diets with different levels of urea. Similarly, Inostroza *et al*. (2010) found that the yields of milk fat and milk protein were unaffected ($p> 0.10$) by treatment with urea containing feed. Van Horn and Mudd (1971) showed no differences in milk yields, milk fat content and feed intake in

cows fed dry or liquid urea supplements. Mba *et al*. (1975) showed that urea-treated straw increased milk fat and protein concentrations. Similar results were obtained by Wanapat *et al*. (2009). Jaquette *et al*. (1986) reported that, there was no significant difference in daily milk fat feeding high and low protein diet.

In present study, milk fat might be increased due to urea supplementation through MCB. Lock and Shingfield (2003) stated that starch was converted to acetyl coA through TCA cycle and joined fatty acid pool to form milk fat. Therefore, lactating cow fed MCB had better performance in terms of fat percent in milk. This is the reason why the lowest fat percent was found in control group. Milk protein significantly increased due to MCB supplementation which provided sufficient N for the microbial protein synthesis.

Efficient gluconeogenesis is the most important pathway in high-producing dairy cows for maintaining adequate glucose supply in the mammary gland (Reynolds *et al*., 1988). In ruminants carbohydrates are fermented to volatile fatty acids and energy is supplied almost entirely from these fatty acids. However, this does not mean that ruminants do not require glucose. Glucose is required for the maintenance of nerve tissue, retina, germinative epithelia, heart and even synthesis of lactose for milk (Bolukbasi, 1989). In present study, the mean value of serum glucose was lower than normal minimum range in 1st week but it significantly ($p<0.01$) increased as the level of MCB supplementation increased later on.

The reason could be that, some cows in early lactation might be in mild hypoglycemic condition and did not have enough glucose in the circulation. This argument is in well agreement with Cenesiz *et al*. (2006). Another reason is that, the N supplied by MCB was utilized by rumen microbes to synthesize available microbial protein which was broken down into amino acids in the gut. The portion of glucogenic amino acids along with other keto-acids worked as the precursor of serum glucose which was converted to blood glucose (McDonald *et al.*, 2011). However, this observation is in disagreement with Debasis and Shingh (2003) who found no changes in the serum glucose when fed UMMB in lactating cow.

The serum total protein did not differ significantly ($p>0.05$) in the experimental period. Similar finding was observed by other investigators (Cenesiz *et al.*, 2006; Hosamani *et al.*, 1988). The total protein was higher than the normal value. Serum protein tended to increase due to the effect of MCB. Hosamani *et al.* (2003) found higher serum total protein in the experimental group compared to control.

A key finding in renal disease is the elevation of serum creatinine. The majority of serum creatinine originates from the endogenous conversion of phosphocreatine in muscle. Creatinine is not reutilized in body. It is modified by conditioning and muscle disease and distributed throughout the compartment of total body water. Creatinine concentration is not affected significantly by diet, protein catabolism and urinary flow (Meintjes *et al.*, 2005). In present study, creatinine level significantly ($p<0.01$) reduced in the 2^{nd} and 3^{rd} ($p<0.05$) week indicating no renal disorders in experimental cows.

The level of serum urea appeared constant ($p>0.05$) during the study period and the value was normal till the end of the experiment which was in agreement with Radostits *et al.* (2006). Nozad *et al*. (2012) also reported similar results. Hosamani *et al*. (2003) found slightly higher urea level in treatment group compared to control. Serum bilirubin is derived from hemoglobin and is formed by macrophages and other leptomeningeal cells that degrade the hemoglobin from lysed red blood cells (Kaneko, 1997). In present study, no abnormal change in serum bilirubin was found indicating the proper hepatic function.

During the entire experimental period the total serum cholesterol was non-significant ($p>0.05$). Similar finding was observed by other investigators (Adedibu *et al*., 2013; Cenesiz *et al*., 2006). The amount of total serum cholesterol was higher during the experimental period which could be due to the basal diet which contained several grains and succulent green grasses.

Occurrence of all biochemical reactions and continuation of life is supported by enzymes. Therefore, changes in enzyme activities are considered to be an indicator of the health of an organism (Kuchmar and Moss, 1982). Liver is the main organ controlling metabolism in

entire body. SGPT and SGOT are the specific enzymes of the liver which increases in the plasma by the destruction of the cell membrane and cell necrosis in acute liver disease and due to accumulation of toxic substances (Dunman and Erden, 2004). Normal values of SGPT and SGOT do not appear to differ greatly between sexes, although reported values for cows were somewhat higher than values for bulls (Cornelius *et al.*, 1959; Roussel and Stallcup, 1966). In present study, serum SGPT was normal and did not differ significantly ($p>0.05$) in the treatment group feeding MCB which is in well agreement with Cenesiz *et al*. (2006). Clampitt and Hart (1978) found that the serum SGPT activity per gram of liver was at least four times greater than in other organs although considerable activity was found in both heart and skeletal muscle. But moderate increase in the serum SGPT level does not indicate any hepatic injury in the lactating cow (Kaneko, 1997). In current study, there was no significant ($p>0.05$) changes in the SGPT level indicating functional liver of the experimental cows.

Serum SGOT significantly ($p<0.05$) decreased at the 5^{th} week but remained in normal range among the treatment groups compared to control. It could be inferred that, the dietary urea supplementation by MCB might have provided available amino acids for maintaining tissue repair which helped maintaining normal serum SGOT in the treatment groups. Cenesiz *et al*. (2006) did not find any change in SGOT in the treatment groups compared to control.

Glucose is a universal fuel used in energy metabolism and synthesis pathways of all mammalian cells (Cankaya *et al.*, 2007; Cárdenas *et al.*, 1998). Among all the nutrient sources, glucose is the important predictor to explain the variability of milk production (Ingvartsen and Friggens, 2005). Glucose requirement and glucose status are critically dependent on lactation and the level of milk production and its components are closely interconnected with endogenous glucose production (Hammon *et al.*, 2010; Reynolds, 1995).

Serum glucose is the indicator of the functional liver (Bobe *et al*., 2004). Lower level of serum glucose, urea and total protein are the indicators of fat infiltration into the liver (West, 1990). In current research, a very strong ($r= -0.88$) negative correlation between glucose and urea was found. González *et al*. (2011) found a non-significant negative correlation ($r= -0.10$) in high yielding dairy cows. The increase in serum glucose could be due to the fortification of starch and glucogenic materials that was supplied through MCB. In another study, González *et al*. (2011) reported a significant ($p<0.01$) negative correlation ($r= -0.51$) between glucose and creatinine which was similar to current finding.

Chapter-6: Conclusion

The study investigates the effects of MCB on milk yield, milk composition and serum parameters in crossbred dairy cows reared in commercial dairy farms under traditional farming system. It was speculated that, milk production increased due supplementation of MCB without exhibiting harmful effects on blood parameters. The highest milk yield was recorded in the cows fed diet containing 25% urea supplemented MCB. Similar to milk yield, fat, protein, SNF and TS content of milk substantially improved after feeding MCB during study period in the experimental groups compared to control.

Most of the serum parameters appeared normal in the treatment group except cholesterol and protein. Wide range of MCB supplementation did not influence normal level of serum bilirubin, SGPT and SGOT which clearly indicated functional liver. Similarly, normal level of serum creatinine and urea reflected soundness of the functioning of kidney. Serum glucose was higher in the treatment groups. Since lactating cows require more serum glucose for milk synthesis, therefore, the additional glucose supplied through MCB obviously resulted beneficial effect to produce more milk for the experimental cows.

Most of the conventional methods for feeding urea used in Bangladesh or elsewhere are laborious and time consuming. None of them are suitable either for long term preservation or for marketing. MCB on the other hand can be produced in large scale industrial level. It is convenient for transportation, feeding and storage as well. It can quickly be supplemented with basal diet as an additional source of protein, starch and mineral. Therefore, 25% urea supplemented MCB may be suggested for the dairy farmer as a novel alternative to traditional urea supplements used for dairy cows.

Chapter-7: Recommendation

Due to financial constraints and technical limitations, some vital blood parameters like high density lipoprotein (HDL), low density lipoprotein (LDL), very low density lipoprotein (VLDL), calcium, phosphorus and other trace minerals both in meat and milk were not analyzed. These parameters could have vital impact on human health. These parameters could be analyzed as future study.

In this study, postmortem examinations were not carried out during and after study period. For future recommendation of MCB, microscopic as well as gross observation of liver, kidney, digestive tract of the dairy cows should be carried out. During the study period, hormonal profile of the experimental animals were not estimated which might be done in future.

The interaction between rumen environment and MCB should be investigated. The lethal dose of multi-nutrient cattle biscuit should be estimated. Finally, the long term effect of MCB on reproductive performance of lactating cows should be investigated in future.

Chapter-8: References

Association of American Feed Control Officials. 1955. Historical Sketch 1909-1955.

Abdoun K, Stumpff F and Martens H. 2006. Ammonia and urea transport across the rumen epithelium: a review. Animal Health Research Reviews. 7(1-2): 43-59.

Adedibu II, Opoola E and Jinadu LA. 2013. Associations between milk yield, parity, physiological status and certain serum biochemical properties of Friesian x Bunaji cows. International Journal Agriculture and Biosciences. 2(3): 127-131.

Akbar MA and Khaleduzzaman ABM. 2009. Feeds and Feeding: Survey of Feedstuff Availability. In Handbook of Dairy Nutrition - Bangladesh, Edited by Robinson PH, Krishnamoorthy U, Anand PEV, American Soybean Association, International Marketing, Creative Views, New Delhi, India. pp. 117-135.

Akbar MA and Tareque AMM. 1990. Proper utilization of byproducts and locally available feedstuffs in buffalo rations in Bangladesh. In: Domestic Buffalo Production in Asia. Published by the International Atomic Energy Agency, Viena (Austria). pp. 63-74.

Akbar MA. 1992. Methods of urea incorporation in straw and their effects on performance of buffalo heifers. Asian-Australian Journal of Animal Sciences. 5(3): 545-548.

Alam J. 2002. Dairy and poultry farming in Bangladesh. Current status and potentials. Paper presented at the workshop on: Policies and Institutions for Poultry and Dairy Development in Bangladesh, Farmgate, Dhaka, Bangladesh Agricultural Research Council.

Alam MGH, Ul-Azam MDS and Khan MDJ. 2006. Supplementation with urea and molasses and body weight, milk yield and onset of ovarian cyclicity in cows. Journal of Reproduction and Development. 52(4): 529-535.

Alam MGS, Rahman MA, Khatun M and Ahmed JU. 2009. Feed supplementation and weight change, milk yield and post-partum oestrus in Desi cows. The Bangladesh Veterinarian. 26(2): 39-47.

Allison MJ. 1969. Biosynthesis of amino acids by ruminal microorganisms. Journal of Animal Science. 29(5): 797-807.

Altona RE, Rose CJ and Tilley TJ. 1960. Urea as supplementary protein for bulk feeds. South African Journal of Agricultural Science. 3: 69-81.

References

Anindo D, Toe F, Tembely S, Mukasa-Mugerwa E, Lahlou-Kassi A and Sovani S. 1997. Effect of molasses-urea-block (MUB) on dry matter intake, growth, reproductive performance and control of gastrointestinal nematode infection of grazing Menz ram lambs. Small Ruminant Research. 27(1): 63-71.

Antonelli AC, Mori CS, Soares PC, Kitamura SS and Ortolani EL. 2004. Experimental ammonia poisoning in cattle fed extruded or prilled urea: clinicalfindings. Brazilian Journal of Veterinary Research and Animal Science. 41: 67-74.

Association of Official Analytical Chemists. 2006. Official Methods of Analyasis of AOAC International. 18th edition (Editor: William Horwitz, George W . Latimer), Gaithersburg, USA.

Araque CAH and Rosos LRO. 1993. Evaluacion de bloques multinutricionales con y sin antihelmintico en la alimentacion de mautas. Zootecnia Tropical. 11(1): 49-58.

Bangladesh Beuro of Statistics. 2009. Report of the Household-based Livestock and Poultry Survey 2009.

Bangladesh Bureau of Statistics. 2011. Statistical Year Book of Bangladesh, Statistics Division, Ministry of Planning, The Government of peoples Republic of Bangladesh.

Bangladesh Bureau of Statistics. 2012. Bangladesh population and housing census 2011.

Bangladesh Bureau of Statistics. 2012. Statistical Year Book of Bangladesh, Statistics Division, Ministry of Planning, The Government of peoples Republic of Bangladesh.

Bangladesh Economic Review. 2012. p.111. Ministry Of Finance, The Government of Bangladesh, Dhaka.

Bangladesh Economic Review. 2014. p.104. Ministry Of Finance, The Government of Bangladesh, Dhaka.

Bangladesh Meteorological Department. 2014. Ministry of Defense of the Government of Bangladesh.

Bartley EE, Avery TB, Nagaraja TG, Watt BR, Davidovich A, Galitzer S and Lassman B. 1981. Ammonia toxicity in cattle. V. Ammonia concentration of lymph and portal, carotid and jugular blood after the ingestion of urea. Journal of Animal Science. 53(2): 494-498.

References

Bartley EE, Davidovich AD, Barr GW, Griffel GW, Dayton AD, Deyoe CW and Bechtle RM. 1976. Ammonia toxicity in cattle. I. Rumen and blood changes associated with toxicity and treatment methods. Journal of Animal Science. 43(4): 835-841.

Beames RM and Morris JG. 1965. Effect of salt/urea blocks on body-weight, body composition and wool production of sheep fed low-protein native grass hay. Queensland Journal of Agriculture Science. 22: 369-379.

Beames RM. 1963. Provision of urea to cattle in a salt/urea/molasses block. Queensland Journal of Agriculture Science. 20: 213-230.

Bhuiyan AKFH. 1997. Cattle breeding and Improvement Strategy in Bangladesh-Past, Present and Future. Keynote paper presented at a national seminar organized by the Directorate of Livestock Services, Government of the Peoples Republic of Bangladesh Agricultural Research Council (BARC), Dhaka, 25 May, 1997. pp.1-16.

Binh BV, Bui Van Chin BV and Preston TR. 1991. Molasses urea blocks as supplements for rabbits. Livestock Research for Rural Development. 3(2): 13-18.

Bobe G, Young JW and Beitz DC. 2004. Invited Review: Pathology, Etiology, Prevention, and Treatment of Fatty Liver in Dairy Cows. Journal of dairy science. 87(10): 3105-3124.

Bolukbasi FM. 1989. The Physiology of cattle. Fizoloji Dars Kitabu. ankara üniversitesi veteriner fakültesi yatay. pp. 413-417.

Bunting LD, Boling JA and MacKown CT. 1989a. Effect of dietary protein level on nitrogen metabolism in the growing bovine: I. Nitrogen recycling and intestinal protein supply in calves. Journal of Animal Science. 67: 810-819.

Bunting LD, Boling JA, MacKown CT and Davenport GM. 1989b. Effect of dietary protein level on nitrogen metabolism in the growing bovine: II. Diffusion into and utilization of endogenous urea nitrogen in the rumen. Journal of Animal Science. 67(3): 820-826.

Cankaya M, Hernandez AM, Ciftci M, Beydemir S, Ozdemir H, Budak H and Kufrevioglu OI. 2007. An analysis of expression patterns of genes encoding proteins with catalytic activities. BMC genomics. 8(1): 232.

References

Cárdenas ML, Cornish-Bowden A and Ureta T. 1998. Evolution and regulatory role of the hexokinases. Biochimica et Biophysica Acta (BBA)-Molecular Cell Research. 1401(3): 242-264.

Casper DP and Schingoethe DJ. 1986. Evaluation of urea and dried whey in diets of cows during early lactation. Journal of dairy science. 69(5): 1346-1354.

Cenesiz S, Atakisi O, Ozcan A, Yuceyurt R and Unal Y. 2006. Effect of feed supplemented with urea molasses mineral blocks on activity of serum AST, ALT and levels of total protein, glucose, triglyceride, total lipid, total protein, total cholesterol in lambs. Kafkas Universitesi Veteriner Fakultesidergisi. 12(2): 137-140.

Census of Agriculture. 2008. Structure of Agricultural Holdings and Livestock Population National Series Volume 1; Bangladesh Bureau of Statistics 2010, Statistics Division, Ministry of Planning, Government of The Peoples Republic of Bangladesh.

Chalupa W. 1971. Metabolic aspects of nonprotein nitrogen utilization in ruminant animals. In Federation Proceedings. 31(3): 1152-1164

Chowdhury MAR. 2004. Effects of urea molasses block on productive and productive performance of crossbred dairy cattle. (PhD Thesis).

Church DC and Santos A. 1981. Effect of graded levels of soybean meal and of a nonprotein nitrogen-molasses supplement on consumption and digestibility of wheat straw. Journal of Animal Science. 53: 1609-1615.

Clampitt RB and Hart RJ. 1978. The tissue activities of some diagnostic enzymes in ten mammalian species. Journal of comparative pathology. 88(4): 607-621.

Clark R, Oyaert W and Quin JI. 1951. Studies on the alimentary tract of the Merino sheep in South Africa. XXI. The toxicity of urea to sheep under different conditions. Onderstepoort Journal of Veterinary Research. 25: 73.

Coombe JB and Tribe DE. 1958. Toxicity of urea to sheep. Nature. 182: 116.

Coombe JB. Tribe DE and Morrison JWC. 1960. Some experimental observations on the toxicity of urea to sheep. Australian Journal of Agricultural Research. 11: 247.

Cornelius CE, Kaneko JJ and Benson DC. 1959. Erythrocyte survival studies in the mule deer, aoudad sheep, and springbok antelope, using glycine-2-C14. American journal of veterinary research. 20: 917-920.

References

Davidovich A, Bartley EE, Chapman TE, Bechtle RM, Dayton AD and Frey RA. 1977. Ammonia toxicity in cattle. II. Changes in carotid and jugular blood components associated with toxicity. Journal of Animal Science. 44(4): 702-709.

Davis GK and Roberts HF. 1959. Urea toxicity in cattle. University of Florida Agricultural Experiment Station. Bulletin. 611.

Debasis D and Singh GP. 2003. Effect of cold process monensin enriched urea molasses mineral blocks on performance of crossbred calves fed a wheat straw based diet. Animal feed science and technology. 103(1): 5 1-61.

Department of Livestock Services 2001. Department of Livestock Services Annual Report. Government of The People's Republic of Bangladesh.

Dinning JS, Briggs HM, Gallup WD, Orr HW and Butler R. 1948. Effect of orally administered urea on the ammonia and urea concentration in the blood of cattle and sheep, with observations on blood ammonia levels associated with symptoms of alkalosis. American Journal of Physiology. 153: 41.

Djibrillou OA, Pandey VS, Gouro SA and Verhulst A. 1998. Effect of urea-treated or untreated straw with cotton seed on performances of lactating Maradi (Red Sokoto) goats in Niger. Livestock production science. 55(2): 117-125.

Doreau M, Chilliard Y, Rulquin H and Demeyer DI. 1999. Manipulation of milk fat in dairy cows. Recent Advances in Animal Nutrition. 81-110.

Duc Vu D, Xuan Cuong L, Anh Dung C and Ho Hai P. 1999. Use of urea-molasses-multinutrient block and urea-treated rice straw for improving dairy cattle productivity in Vietnam. Preventive Veterinary Medicine. 38(2): 187-193.

Duman C and Erden F. 2004. Brief review of the biochemical laboratory data for primary health care (translated title). Sürekli Tıp Eğitimi Dergisi. 13(7): 256-262.

Duncan DB. 1955. Multiple range and multiple "F" test. Biometrics. 11:1-42

Ehrenberg P, Nitsche H and Muller J. 1891. Report on protein substitutions in feeding experiments at Bettlerm (translated title). Zentra Tierernahr Futtermittlek. 1: 33.

Emmanuel B. 1980. Urea cycle enzymes in tissues (liver, rumen epithelium, heart, kidney, lung and spleen) of sheep (Ovis aries). Comparative Biochemistry and Physiology Part B: Comparative Biochemistry. 65(4): 693-697.

References

Erb RE, CaDahan CJ and Garverick HA. 1975. Effect of dietary urea on re-production. Dairy Science Handbook. Agriservices Foundation, Qovis, California press. pp.189-194.

Ferdous Z, Khan MAS, Siddiki MSR and Habib MA. 2007: Response to Urea Molasses Block (UMB) feeding of lactating cows reared under village condition of Bangladesh. Journal of Bangladesh Society for Agricultural Science and Technology. 4: 161-164.

Filippi BG, Amici A and Machin D. 1992. Initial studies on the production and use of molasses blocks in the feeding of forage fed rabbits. Journal of Applied Rabbit Research 15: 1053-1057.

Food and Agriculture Organization 2005. Food and Agricultural Organization of United Nations. Rome.

Food and Agriculture Organization 2005a. The state of food insecurity in the world, Food and Agricultural Organization of United Nations, Rome.

Gallup WD. 1956. Review of utilization of non protein nitrogen in the ruminant. Journal of Agricultural and Food Chemistry. 4: 625.

Galo E, Emanuele SM, Sniffen CJ, White JH and Knapp JR. 2003. Effects of a polymer-coated urea product on nitrogen metabolism in lactating Holstein dairy cattle. Journal of Dairy Science. 86: 2154-2162.

Ghosh A, Alam MGS and Akbar MA. 1993. Effect of urea-molasses-mineral block supplementation on postpartum ovarian activity in zebu cows. Animal Reproduction Science. 31(1): 61-67.

Golombeski, GL, Kalscheur KF, Hippen AR and Schingoethe DJ. 2006. Slow-release urea and highly fermentable sugars in diets fed to lactating dairy cows. Journal of Dairy Science. 89: 4395-4403.

Gonçalves GDS, Pedreira MDS, Azevedo JAG, Del RAJ, Silva HGO and Silva FF. 2014. Replacement of soybean meal by conventional and coated urea in dairy cows: intake, digestibility, production and composition of milk. Acta Scientiarum. Animal Sciences. 36(1): 71-78.

González FD, Muiño R, Pereira V, Campos R and Benedito JL. 2011. Relationship among blood indicators of lipomobilization and hepatic function during early lactation in high-yielding dairy cows. Journal of veterinary science. 12(3): 251-255.

References

Grummer RR. 1991. Effect of feed on the composition of milk fat. Journal of Dairy Science. 74(9): 3244-3257.

Guthrie MJ and Wagner DG. 1988. Influence of protein or grain supplementation and increasing levels of soybean meal on intake, utilization and passage rate of prairie hay in beef steers and heifers. Journal of Animal Science. 66(6): 1529-1537.

Halim 1992. Comparative economics analysis of local and cross-bred dairy cows in a selected area of Dhaka district, Bangladesh, M.S. thesis, Dept. of Agricultural economics, Bangladesh Agricultural University, (BAU), Mymensingh.

Hammon HM, Metges CC, Schulz A, Junghans P, Steinhoff J, Schneider F and Kühn C. 2010. Differences in milk production, glucose metabolism, and carcass composition of 2 Charolais × Holstein F2 families derived from reciprocal paternal and maternal grandsire crosses. Journal of dairy science. 93(7): 3007-3018.

Harris LE and Mitchell HH. 1941. The Value of Urea in the Synthesis of Protein in the Paunch of the Ruminant I. In MaintenanceTwo Figures. The Journal of Nutrition. 22(2): 167-182.

Hart EB, Boitstedt G, Deabald HJ and Wegner MI. 1939. The utilization of simple nitrogenous compounds such as urea and ammonium bicarbonate by growing calves. Journal of Dairy Science. 22: 785-798.

Hasan MM. 1995. Distribution pattern and some economic dairy characters of locals and crossbred cows in Mymensingh Sadar, M.S. Thesis, Department of Dairy Science, Bangladesh Agricultural University, Mymensingh.

Hendratno C, Nolan JV and Leng RA. 1991. The importance of urea-molasses multinutrient blocks for ruminant production in Indonesia. In: Proceedings of an international symposium on nuclear related techniques for animal production and health. International Atomic Energy Agency, Vienna, Austria.

Hock RDV, Muttentuwengama GS and Sciere JB. 1988. Overcoming the nutritional limitations of rice straw for ruminants. 1. Urea ammonia treatment and supplementation with rice bran and gliciridia for lactating Surti buffaloes. Asian-Australasian Journal of Animal Sciences. 1: 201-208.

Hosamani SV, Mehra UR and Dass RS. 2003. Effect of different source of energy on urea molasses mineral block intake, nutrient utilization, rumen fermentation pattern and blood profile in Murrah buffaloes (Bubalus bubalis). Asian-Australasian Journal of Animal Sciences. 16(6): 818-822.

References

Hoshino S, Skatsuhara K and Morimoto K. 1966. Ammonia anabolism in ruminants. Journal of Dairy Science. 49: 1523-1528.

Houmani M and Tisserand JL. 1999. Complémentation d'une paille de blé avec des blocs multinutritionnels: effets sur la digestibilité de la paille et intérêt pour des brebis taries et des agneaux en croissance. Annales de Zootechnie. 48: 199-209.

Huber JT and Thomas JW. 1971 Urea-treated corn silage in low protein rations for lactating cows. Journal of Dairy Science. 54: 224.

Huber JT, Lichtenwalner RE and Thomas JW. 1973. Factors affecting response of lactating cows to ammonia-treated corn silages. Journal of Dairy Science. 56: 1283.

Huber JT, Thomas JW and Emery RS. 1968. Response of lactating cows fed urea-treated corn silage harvested at varying stages of maturity. Journal of Dairy Science. 51: 1806.

Hungate RE. 1966. The rumen and its microbes. Academic Press, New York . pp. 533.

Huntington GB. 1989. Hepatic urea synthesis and site and rate of urea removal from blood of beef steers fed alfalfa hay or a high concentrate diet. Canadian Journal of Animal Science. 69(1): 215-223.

Ingvartsen KL and Friggens NC. 2005. To what extent do variabilities in hormones, metabolites and energy intake explain variability in milk yield? Domestic animal endocrinology. 29(2): 294-304.

Inostroza JF, Shaver RD, Cabrera VE and Tricárico JM. 2010. Effect of diets containing a controlled-release urea product on milk yield, milk composition, and milk component yields in commercial Wisconsin dairy herds and economic implications. The Professional Animal Scientist. 26(2): 175-180.

Islam A, Wadud A, Rabbani MG and Hossain B. 1999. Rearing practices and milk production of dairy cattle in Thakurgaon district. Bangladesh Journal of Animal Science. 27: 172-176.

Itoh H, Terashima Y and Tohari N. 1979. Evaluation of ammonia treatment for improving the utilization of fibrous materials in low quality roughages. Japanese Journal of Zootechnical Science. 50: 54-61.

Jackson MG. 1977. Rice straw as livestock feed. World Animal Review. 23: 25-31.

References

Jaquette RD, Rakes AH and Croom WJ. 1986. Effects of dietary protein on milk, rumen and blood parameters in dairy cattle fed low fiber diets. Journal of Dairy Science. 69: 1026-1034.

Kabir F and Islam MR. 2009. Comparative study on productive and reproductive performance of local and different crossbreed dairy cows at Dawlatpur, Khulna in Bangladesh Bangladesh. Research Publications Journal. 3(2): 909-914.

Kachmar JF and Moss DW. 1982. Fundamentals of clinical chemistry., WB Saunders Company, Philadelphia. 562-698.

Kamal MM, Iqbal DMH and Khaleduzzaman ABM. 2009. Supplementation of maize-based concentrates and milk production in indigenous cows. Bangladesh Veterinarian. 26(2): 48-53.

Kaneko JJ, Harvey WJ, Bruss ML. 1997. Clinical biochemistry of domestic animals, Fifth edition, Academic Press, San Diego, California.

Karim F. 1988. Effect of supplimentation of groundnut straw on rice enlisted with poultry excreta and urea on its digestibility in vitro. M.S. Thesis, Bangladesh Agricultural University, Mymensingh, Bangladesh.

Kennedy PM and Milligan LP. 1980. The degradation and utilization of endogenous urea the gastrointestinal tract of ruminants: A review. Canadian Journal of Animal Science. 60: 205-221.

Kertz AF. 2010. Review: Urea feeding to dairy cattle: a historical perspective and review. The Professional Animal Scientist. 26(3): 257-272.

Kertz AF, Davidson LE, Cords BR and Puch HC. 1983. Ruminal Infusion of Ammonium Chloride in Lactating Cows to Determine Effect of pH on Ammonia Trapping. Journal of Dairy Science. 66: 2597-2601.

Khan AA. 1990. A comparative study on the reproductive efficiency of native and crossbred cows. M.S. Thesis, Department of Dairy Science. Bangladesh Agricultural University, Mymensingh.

Khan BB, Iqbal A, Riaz M, Yaqoob M, Younas M. 2004. Livestock Management Manual I, Department of Livestock Management, University of Agriculture, Faisalabad, Pakistan.

Knott FN, Polan CE and Huber JT. 1972. Further observations on utilization of urea by lactating cows. Journal of Dairy Science. 55: 466.

Krebs K. 1937. Value of the Amides in the Feeding of Cattle. Historical onsideration of the Development of the Amide Question, Critical Evaluation of the State of Our Present Knowledge (Translated title). Biedermanns Zentr Biologie Tierernähr. 9: 394-507.

Kunju PJG. 1986. Urea molasses block lick: a feed supplement for ruminants. pp. 261-274, In: Ibrahim MNM and Schiere JB. (eds). Rice straw and related feeds in ruminant rations. Proceedings of an International Workshop, Kandy, Sri Lanka.

Leng RA. 1984. The potential of solidified molasses-based blocks for the correction of multinutritional deficiencies in buffaloes and other ruminants fed low-quality agro-industrial by-products. pp.135-150, In: The use of nuclear techniques to improve domestic buffalo production in Asia. Final research coordination meeting. FAO/ IAEA, Manila, Philippines.

Leng RA. 1997. Balancing nutrition to maximize forage utilization. In: Tree foliage in ruminant nutrition. FAO Corporate Document Repository.

Liu JX, Okubo M and Asahida Y. 1988. Effect of soyabean mea supplementation on fiber digestion in the rumen and voluntary intake of rice straw by sheep. Japanese Journal of Zootechnical Science. 75: 1034-1039.

Lock AL and Shingfield KJ. 2003. Optimising milk composition. In: Kebreab E, Mills J and Beever D. (eds.) UK Dairying: using science to meet consumers' needs. Nottingham University Press, Nottingham, UK. (in press).

Loosli JK, Williams HH, Thomas WE, Ferris FH and Maynard LA. 1949. Synthesis of amino acids in the rumen. Science. 110 (2849): 144-145.

Mapato C, Wanapat M and Cherdthong A. 2010. Effects of urea treatment of straw and dietary level of vegetable oil on lactating dairy cows. Tropical Animal Health and Production. 42(8): 1635-1642.

Matsuzaki M, Takizawa S and Ogawa M. 1997. Plasma insulin, metabolite concentration, and carcass characteristics of Japanese Black, Japanese Brown, and Holstein steers. Journal of Animal Science. 75: 3287-3293.

Mazed MA. 1997: Effect of urea molasses multinutrient block on the productive and reproductive performance of indigenous zebu cows under the village condition of Bangladesh using Radio-Immunoassay techniques. MS Thesis, Department of Dairy

Science, Faculty of Animal Husbandry, Bangladesh Agricultural University, Mymensingh, Bangladesh.

Mba AU, Boyo BS and Oyenuga VA. 1975. Studies on the milk composition of West African dwarf, Red Sokoto and Saanen goats at different stages of lactation. I. Total solids, butterfat, solid-non-fat, protein, lactose and energy contents of milk. Journal Dairy Research. 42: 217-226.

McBeath DG, Preston NK and Thompson F. 1979. Studies in sheep on the efficacy of fenbendazole administered via feed-block carrier. British Veterinary Journal. 135(3): 271-278.

McDonald P, Edwards RA, Greenhalgh JFD, Morgan CA, Sinclair LA and Wilkinson RG. 2011. Animal Nutrition. Seventh Edition. Harlow, Essex UK. Pearson Education Limited. pp: 203-229.

McNitt JI. 1983. Livestock Husbandry Techniques. Granada.

Meintjes RA, Botha CJ and Prozesky L. 2005. Toxicity, pathophysiology and pathology in sheep following dosing of the nephrotoxic plant Nolletia gariepina (DC) Mattf . The Onderstepoort Journal of Veterinary Research. 72: 39-53.

Miah AG, Salma U, Khan MAS and Ali ML. 2000. Effect of urea-molasses multinutrients-blocks on the productive performance of indigenous cows. Bangladesh journal of Animal Science. 29: 135-142.

Moaeen-ud-Din M, Ahmad N, Iqbal A and Abdullah M. 2006. Evaluation of different formulas for weight estimation in Beetal, Teddi and crossbred (Beetal x Teddi) goats. Journal of Animal and Plant Sciences.16: 3-4.

Nahar TN, Islam M and Hashath MA. 1992. A comparative study on the performance of F1 crossbred cows under rural conditions. Asian-Australian Journal of Animal Science. 5: 435-338.

Nguyen Van Thu. 2000. Urea-molasses based supplements for multipurpose buffaloes. Doctoral thesis. Swedish University of Agricultural Sciences, Uppsala, Sweden.

Norton B W, Moran JB and Nolan JV. 1979. Nitrogen metabolism in Brahman cross, buffalo, banteng and Shorthorn steers fed on low-quality roughage. Crop and Pasture Science. 30(2): 341-351.

References

Nozad S, Ramin AG, Moghadam G, Asri-Rezaei S, Babapour A and Ramin S. 2012. Relationship between blood urea, protein, creatinine, triglycerides and macro-mineral concentrations with the quality and quantity of milk in dairy Holstein cows. Veterinary Research Forum. 3(1): 55-59.

Obara Y and Shimbayashi K. 1980. The appearance of recycled urea in the digestive tract of goats during the final third of a once daily feeding of a low-protein diet. British Journal of Nutrition. 44: 295-305.

Obeidat BS, Thomas MG, Hallford DM, Keisler DH, Petersen MK, Bryant WD, Garcia MD, Narro L and Ropez R. 2002. Metabolic characteristics of multiparous Angus and Brahman cows grazing in the Chihuahuan Desert. Journal of Animal Science. 80: 2223-2233.

Oltjen RR, Robbins JD and Davis RE. 1964. Studies involving the use of glutamic acid in ruminant nutrition. Journal of Animal Science. 23: 767.

Oltjen RR. 1969. Effects of feeding ruminants non-protein nitrogen as the only nitrogen source. Journal of Animal Science. 28(5): 673-681.

Osuna, DB, Ventura MS and Casanova A. 1996. Alternativas de suplementacion para mejorar la utilizacion de los forrajes conservados. II. Efecto de diferentes concentrationes de dos fuentes de energia en bloques nutricionales sobre el consumo y ganancia de peso de ovinos en crecimiento. Revista de la Facultad Agronomia (LUZ). 13(2): 191-200.

Palmquist DL, Beaulieu AD and Barbano DM. 1993. Feed and animal factors influencing milk fat composition. Journal of Dairy Science. 76: 1753-1771.

Paul AK, Maruf AA, Jha PK and Alam MGS. 2013. Reproductive performance of crossbred and indigenous (desi) dairy cows under rural context at sirajgonj district of Bangladesh. Journal of Embryo Transfer. 28(4): 319-324.

Perez R. 1990. Manual de crianza: Conejos. Ministerio del Azucar, Havana, Cuba. p. 79.

Polan CE, Hube JT, Sandy RA, Hall JW and Miller CN. 1968. Urea-treated corn silage for lactating cows. Journal of Dairy Science. 51: 1445.

Promma S, Tuikumpee S, Ratnavanija A, Vidhyakorn N and Froement RW. 1984. The effects of urea treated rice straw on growth and milk production of crossbred Holstein Friesian dairy cattle. Proceedings of the 4th Annual Work Shop of the Australian-

Asian Fibrous Agricultural Residues. Research Network held in Khon Kaen, Thailand, 10-14 April. pp.88-93.

Radostits OM, Gay CC, Hinchcliff KW and Constable PD. 2006. Veterinary Medicine 10th edition: A textbook of diseases of cattle, sheep, goats, pigs and horses. Saunders Elsevier, Edinburgh. pp. 2048-2049.

Reid JT. 1953. Urea as a protein replacement for ruminants: A review. Journal of Veterinary Science. 36: 955.

Repp WW, Hale WH and Burroughs W. 1955a. The value of several non-protein nitrogen compounds as protein substitutes in lamb fattening rations. Journal of Animal Science. 14: 901-908.

Repp WW, Hale WH, Cheng EW and Burroughs W. 1955b. The influence of oral administration of nonprotein nitrogen feeding compounds upon blood ammonia and urea levels in lambs. Journal of Animal Science. 14: 118-131.

Reynolds CK, Engelhardt WV, Leonhard-Marek S, Breves G and Giesecke D. 1995. Quantitative aspects of liver metabolism in ruminants. In Ruminant physiology: digestion, metabolism, growth and reproduction. Proceedings 8th International Symposium on Ruminant Physiology. Delmar Publishers. pp. 351-371.

Reynolds CK, Huntington GB, Tyrrell HF and Reynolds PJ. 1988. Net portal-drained visceral and hepatic metabolism of glucose, L-lactate, and nitrogenous compounds in lactating Holstein cows. Journal of Dairy Science. 71(7): 1803-1812.

Roussel, JD and Stallcup OT. 1966. Influence of age and season on phosphatase and transaminase activities in blood serum of bulls. American Journal of Veterinary Research. 27(121): 1527.

Rummler H, Laue JW and Bersehneider F. 1962. Biochemical changes and therapeutic methods in urea poisoning in cattle. Monatsh Veterinaermed Journal. 17: 156.

Ryley JW and Gabtner R. 1968. Effects of sub-lethal doses of urea on pregnant cattle. In Proceedings of the Australian Society of Animal Production. 7: 385-390.

Saadullah M, Haque M and Dolberg F. 1982. Treated and untreated rice straw for growing cattle. Tropical Animal Production. 7: 20-25.

Saadullah M. 1991. The importance of urea molasses blocks and by pass protein in animal production, the situation in Bangladesh, Vienna, IAEA. pp: 147-156.

References

Salman AD. 1997. The role of multinutrient blocks for sheep production in integrated cereal-livestock farming system in Iraq. In 2nd FAP Electronic Conference, Livestock Feed Resources within Integrated Farming Systems. 9: 209-219.

Salter DN. Daneshvar K and Smith RH. 1979. The origin of nitrogen incorporated in to compounds in the rumen bacteria of steers given protein- and urea-containing diets. British Journal of Nutrition. 41: 197-209.

Sansoucy R. 1986. The Sahel: Manufacture of molasses-urea blocks. World Animal Review (FAO). 57: 40-48.

Sansoucy R. 1995. New developments in the manufacture and utilization of multinutrient blocks. Short communications. World Animal Review. 82(1): 78-83.

Santos JF, Dias Jr GS and Bitencourt LL. 2011. Resposta de vacas leiteiras a substituição o parcial de farelo de soja por ureia encapsulada. Arquivo Brasileiro de Medicina Veterinária e Zootecnia. 6(32): 423-432.

Sanyal PK, Srivastava SM, Panchal AG and Singh DK. 1995. Effect of feeding fenbendazole incorporated urea molasses block on nematode infection and milk yield of dairy cattle. Journal of Veterinary Parasitology. 9(1): 11-16.

Sarker MA. 1995. 'Economic analysis of dairy cattle enterpriseand its pattern of contribution to Farm income in a selectedarea of Bangladesh.' M.S. thesis, Department of Agricultural Economics, B.A.U. Mymensingh, Bangladesh.

Sarraseca A. Milne E. Metcalf MJ and Lobley GE. 1998. Urea recycling in sheep: effects of intake. British Journal of Nutrition. 79: 79-88.

Scamell JM. 2006. Healthy land for healthy cattle. Cattle Practice. 14: 143-152.

Shamsuddin M, Goodger WJ, Hossein MS, Azizunnesa Bennett T and Nardlund K. 2006. A survey to identify economic opportunities for smallholder dairy farms in Bangladesh. Tropical Animal Health and Production. 38: 31-140.

Shingu H, Hayashi H, Touno E, Oshibe A, Kushibiki S, Oda S, Katoh K and Obara Y. 2007. Characteristics of developmental changes in the kinetics of glucose and urea in Japanese Black calves: Comparison with Holstein calves. Journal of Animal Science. 85: 2910-2915.

References

Souza VLS, Almeida R, Silva DFFS, Piekarki PRB and Jesus CP. 2010. Substituição parcial de farelo de soja por ureia protegida na produção e composição de leite. Arquivo Brasileiro de Medicina Veterinária e Zootecnia. 62(6): 1415-1422.

Stangel HJ. 1967. History of the use of urea in ruminant feeds. In Briggs MH. (Ed.) Urea as a Protein Supplement. Pergamon Press. New York.

Stojević Z, Piršljin J, Milinković-Tur S, Zdelar-Tuk M and Ljubić BB. 2005. Activities of AST, ALT and GGT in clinically healthy dairy cows during lactation and in the dry period. Veterinary Archives. 75: 67-73.

Susmel P, Spanghero M, Mills CR and Stefanon B. 1995. Rumen fermentation characteristics and digestibility of cattle diets containing different whey: maize ratios. Animal feed science and technology. 53(1): 81-89.

Sutton JD. 1989. Altering milk composition by feeding. Journal of Dairy Science. 72: 2801-2814.

Talukder MSU, Hoque MN, Hossain MI, Aziz SA and Rahman MR. 2001. Effect of different genotypes of milk yield and reproductive performance of cows. Pakistan Journal of Biological Science. 4: 1421-1424.

Thornton PK. 2010. Livestock production: recent trends, future prospects. Philosophical Transactions of the Royal Society B: Biological Sciences. 365: 2853-2867.

Tiwari SP, Singh UB and Mehra UR. 1990. Urea, molasses mineral blocks as a feed supplement: Effect on growth and nutrient utilization in buffalo calves. Animal Feed Science and Technology. 29: 333-341.

Uddin MJ, Shahjalal M, Kabir F, Khan MH, Chowdhury SA and Chittagong B. 2002. Beneficiary Effect of Feeding Urea-Molasses Treated Straw on Buffalo Cows in Bangladesh. OnLine Journal of Biological Science. 2(6): 384-385.

Van Horn HH and Mudd JS. 1971. Comparison of a liquid supplement of nonprotein nitrogen with urea and soybean meal for lactating cows. Journal of Dairy Science. 54(1): 58-64.

Van Horn HH, Foreman CF and Rodriquez JE. 1967. Effect of high-urea supplementation on feed intake and milk production of dairy cows. Journal of Dairy Science. 50: 709-714.

Van Saun R. 2000. Blood profiles as indicators of nutritional status. In: Proc.18th Annu. Western Canadian Dairy Seminar. Red Deer, Alberta, Canada. pp.1-6.

References

Vargas JE and Rivera JG. 1994. Efecto del bloque multinutricional sobre el comportamiento productivo en ovejas africanas. Livestock Research for Rural Development. 6: 2.

Virtanen AI. 1966. Milk production of cows on a protein free feed. Science. 153: 1603.

Wadhwa M and Bakshi M. 2014. Nutritional evaluation of urea molasses multi-nutrient blocks containing agro-industrial wastes in buffaloes. The Indian Journal of Animal Sciences. 84(5): 71-75.

Wanapat M, Polyorach S, Boonnop K, Mapato C and Cherdthong A. 2009. Effects of treating rice straw with urea or urea and calcium hydroxide upon intake, digestibility, rumen fermentation and milk yield of dairy cows. Livestock Science. 125(2): 238-243.

Wanapat M. 1999. Feeding of ruminants in the tropics based on local feed resources. KhonKaen Publication. Company Limited, Khon Kaen, Thailand. p: 236.

Weiske H, Schrod, H and Dangel ST. 1879. Über die Bedeutung des Asparagins für die tierische Ernährung. Zeitschrift für Biologie. 15: 261-296.

West HJ. 1990. Effect on liver function of acetonaemia and the fat cow syndrome in cattle. Research in veterinary science. 48(2): 221-227.

Winer BJ, Brown R and Michels KM. 1991. Statistical Principle in Experimental Design. 3rd ed. New York; McGraw-Hill.

Work SH and Henke LA. 1939. The value of urea as a protein supplement replacement for dairy heifers. Journal of Animal Science. 1: 404-406.

Xin HS, Schaefer DM, Liu QP, Axe DE and Meng QX. 2010. Effects of polyurethane coated urea supplement on in vitro ruminal fermentation, ammonia release dynamics and lactating performance of Holstein dairy cows fed a steam-flaked corn-based diet. Asian-Australian Journal of Animal Science. 23: 491-500.

Zuntz N. 1891. Observations on the digestion and nutritive value of cellulose (translated title). Pflügers Archiv European Journal of Physiology. 49: 477.