INFLUENCE OF LEVEL AND DEGRADABILITY OF DIETARY PROTEIN ON EARLY LACTATION IN CROSSBRED COWS

By

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DECLARATION

I here by declare that this thesis entitled " **INFLUENCE OF LEVEL AND DEGRADABILITY OF DIETARY PROTEIN ON EARLY LACTATION IN CROSSBRED COWS"** is a bonafide record of research work done by me during the course of research and that the thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title, or any other University or Society.

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Introduction

1. INTRODUCTION

Ruminants have the unique ability to subsist and produce without a source of dietary protein due to the synthesis of microbial protein within the rumen. But as a result of tremendous improvement in biotechnology and breeding techniques, the production capacity of the dairy cows has increased to high levels thereby increasing the requirement of dietary protein in appreciable quantity and quality. This has lead to a situation wherein protein formed the most significant constituent of ruminant ration and therefore it is important that this constituent should be used efficiently for economic benefits.

The protein requirement of a dairy cow can be defined as the minimum amount of protein that will support maximum production and can be expressed in terms of crude protein, digestible crude protein, rumen degradable protein, rumen undegradable protein or metabolisable protein. The requirement for amino acids in ruminants is met from the microbial protein and the dietary protein which is not degraded in the rumen but digested in the intestine. Rumen micro organisms are responsible for meeting a major part of the energy requirements of the host animal by transforming dietary carbohydrate into the volatile fatty acids such as acetate. propionate and butyrate. Thus satisfying the demands of the rumen micro organisms for available nitrogen is a major function of the ruminant diet for which a certain portion of the dietary nitrogen fraction must be degraded in the rumen. For high producing animals, the requirement of protein is considerably higher than that obtained from the products of rumen fermentation. Therefore it is also important that a source of undegradable protein should form a part of the total protein of the ration to support high milk production or growth. An excess of either the rumen degradable or undegradable protein can reduce the efficiency of feed utilisation and milk production. An excess of ruminally degradable protein will result in accumulation of ammonia in the rumen which will later on be excreted as urea leading to a wastage of nitrogen, whereas an excess of undegradable protein will result in a shortage of ammonia or nitrogen supply to rumen micro organisms, thereby reducing microbial protein production. Adequacy of levels of undegradable protein and rumen degradable protein in the diets of lactating cows should be considered independently and it is illogical to increase undegradable protein at the expense of degradable protein unless the latter is in excess.

There is considerable controversy regarding the amounts of dietary crude protein needed by dairy cattle and that the recommended level varies from 14 to 20 per cent. There are reports stating that for low producers, microbial protein synthesised in the rumen alone is sufficient to meet their amino acid demands. At the same time there are reports in which incorporation of undegradable protein was beneficial in increasing milk production especially when animals were energy deficient. The beneficial effects of undegradable protein is dependent upon their ability to supply the essential amino acids. The average peak daily milk production of the dairy cow in Kerala ranges between seven to eight kilogram. There are only scanty literature available on the requirement, if any, of the undegradable protein in the diets of such low to medium producing animals. Hence, the present work was undertaken to optimise the level and degradability of dietary protein in concentrate mixtures having sufficient energy to support maximum milk production under the managemental practices existing in the state.

Review of Literature

2. REVIEW OF LITERATURE

The most critical period in the life of the dairy cow is from the time of calving until peak milk production. Energy and protein are the two nutritional factors most likely to limit milk production. The type. source and the ratio between these two nutrients will influence the efficiency of utilisation of the feed as well as the amount of milk produced. As dairy cows in early lactation fail to consume sufficient dry matter. they are prone to be in negative energy balance at this stage (NRC. 1981). Increasing the proportion of concentrates to these cows above 60 per cent of total dry matter of the ration to provide a ration higher in energy density can result in situations such as off-feed and other metabolic disorders (Clark and Davis. 1980).

2.1 ENERGY FOR MILK PRODUCTION.

High producing cows in early lactation will be in negative energy balance as the maximum dry matter intake (DMI) is reached slowly compared to a rapid increase in milk production to reach the maximum by days 35 to 50 (NRC. 1978). Most of the rumen micro organisms grow on carbohydrate or the secondary products of carbohydrate fermentation and hence microbial yield is closely related to the carbohydrate level in the diet rather than the total organic matter content (Harsted and Vik-Mo. 1985; Nocek and Russell. 1988).

Moe and Tyrrell (1975) opined that high intake of feed and above average milk production in the case of cows producing around 25 kg milk per day can be achieved when ration contained 50 to 60 per cent concentrate and 40 to 50 per cent roughage. Scientists had assessed the effect of varying concentrate and roughage ratios as well as different roughage sources in the ration (De Peters and Kelser. 1980; Tyrrell. 1980; Broster *et al.*. 1981; Patel and Sharma. 1983). Sutton (1981) opined that no single proportion of concentrate and roughage could be recommended in the ration to optimise production. Broster and Thomas (1981) reported that the response in milk output to higher inputs of concentrate declined as the lactation advances. Broster *et al.* (1981) observed an increase in milk yield both in the multiparous and primiparous cows when the concentrate roughage ratio was 90:10. Mahal *et al.* (1997) observed significantly higher 4 per cent fat corrected milk (FCM) production but there was no change in the milk fat, solid not fat (SNF) or total solid and protein content of milk from cows receiving roughage and concentrate in 60:40 ratio compared to those receiving 50:50 or 70:30 ratio. Kennelly *et al.* (1999) reported that feeding concentrate to forage in the ratio 75:25 did not affect the feed intake but elevated the propionate levels significantly when compared to those fed at 50:50 ratio. They also reported that addition of buffer to the ration with concentrate roughage ratio of 75:25, prevented the milk fat depression.

McGregor *et al.* (1983) got significant increase in the milk production and DMI when high starch (32.9 per cent) diets were fed to cows compared to low starch (24.9 per cent) diets. Sutton (1985) reported that increasing the corn based concentrate mixture from 60 to 90 per cent of DMI reduced milk yield whereas increasing barley based concentrate mixture to the same extent increased milk yield. He also observed that regardless of the source, increasing the concentrate mixture in the diet had a deleterious effect on the milk fat per cent. Orskov (1986) reported that more than 90 per cent of the starch from the small grains are ruminally fermentable, but upto 40 per cent of the corn starch is rumen undegradable. Nocek and Russell (1988) opined that propionate stimulated milk production more effectively than glucose indicating that ruminal carbohydrate digestibility has a profound effect on milk production.

Punia and Sharma (1985) opined that the production of VFA was more with substrates containing barley and molasses than straw. Mc Carthy *et al.* (1987) reported that there was improvement in the milk yield by partially replacing barley with maize in the concentrate though there was no change in milk fat per cent. The yields of milk and milk fat were more when oats or maize was used as energy source compared to barley or wheat (Broster *et al.*, 1979; Moran, 1986).

Thomas *et al.* (1984) and Sutton *et al.* (1985) opined that the milk production of cows producing 25 to 30 kg milk per day was not affected by the type of concentrate in the ration and milk fat content and fat yield but were influenced by the fibrous concentrates rather than the starchy concentrates. Anis *et al.* (1991) reported that supplementation of molasses in the ration containing poor quality roughage such as rice straw increased milk and fat yields in comparison to maize stover diet in dairy cattle. Huhtanen *et al.* (1995) and Petit and Trembley (1995) reported similar increase in the milk production by supplementation of the ration with soluble sugars.

Jordan and Swanson (1979) opined that the ratio of protein to energy is an important factor in obtaining maximum efficiency of nitrogen and energy utilisation and for optimum animal production. Taminga (1980) reported that a shortage of nitrogen in the rumen decreased the activity of slowly growing cellulolytic bacteria and resulted in a decreased fibre digestibility. Nitrogen utilisation was reduced when there was a deficiency in energy. The optimum ratio between nitrogen and energy depended on the stage of lactation and Sohanae *et al.* (1997) listed out the optimum level for nitrogen and energy (g nitrogen per MJ of ME) for lactation which varied from 2.2 in the first trimester to 2.4 in the next two months of lactation, then decreased gradually as lactation proceeded.

Sievert and Shaver (1993) observed that when rations with 18.5 per cent CP and 21 per cent neutral detergent fibre (NDF) at two levels (35 and 42 per cent) of non fibre carbohydrate (NFC) were given to cows, the total tract digestibility of fibre, rumen acetate level and milk fat were increased. Batajoo and Shaver (1994) opined that diets for cows producing 40 kg milk per day should contain more than 30 per cent of NFC. Similar observations were made by Valadares *et al.* (2000) who got better yields when 37 to 38 per cent NFC was incorporated in the ration.

Broderick (1995) and Vagoni and Broderick (1997) reported that alfalfa silage having 44 per cent NDF and 26 per cent acid detergent fibre (ADF) stimulated feed intake and resulted in higher milk production compared to ryegrass silage with 50 per cent NDF and 10 per cent ADF. Ekinci and Broderick (1997) fed four rations viz. high moisture ear corn (HMEC). HMEC+ expeller soybean meal (ESBM). ground HMEC and HMEC+ESBM and observed that the yield of milk and milk components were highest for HMEC+ESBM. while highest nutrient digestibility and lowest rumen ammonia concentration were observed with ground HMEC ration. Leiva *et al.* (2000) reported that modifying the neutral detergent soluble carbohydrate (NDSC) proportion in the ration could alter milk production. milk components. pattern of rumen fermentation and nitrogen utilisation in cows.

Casper and Schingothae (1989) opined that different sources of readily soluble and fermentable carbohydrates had no significant effect on milk production. Khorasani *et al.* (1994) observed an interaction between protein and energy sources and reported that while protein source significantly increased milk yield. starch source influenced the milk composition.

McCormick *et al.* (2001) supplemented corn based grain rations with ESBM along with sucrose or lactose. They observed that sucrose at five per cent level lowered rumen ammonia nitrogen (NH_3-N). They also supplemented the grain ration with solvent SBM. which resulted in better utilisation of dietary energy for milk production than ESBM or sucrose or lactose supplementation. Broderick *et al.* (2002) observed that when three carbohydrate sources. namely high moisture ear corn (HMEC). cracked shell corn (CSC) and HMEC plus dried citrus pulp (DCP) were fed to cows, the DMI, milk yield and levels of milk components such as fat, protein, SNF and lactose, were more for HMEC and CSC diets than DCP diets.

2.2 PROTEIN FOR MILK PRODUCTION

The protein requirement of a dairy cow in early lactation can be expressed in terms such as crude protein (CP), digestible crude protein (DCP), rumen degradable protein (RDP), rumen undegradable protein (UDP) and metabolisable protein (MP) levels in the ration. Clark *et al.* (1977) opined that the requirement of protein expressed as quantity consumed per day was better than as per centage of CP in the diet. Clark and Davis (1980) and Bricano *et al.* (1988) found that the production of milk by cows in early lactation was directly proportional to the quantity of protein consumed.

Huber (1975) observed that high producing dairy cows benefited from the addition of non protein nitrogen (NPN) to diets containing CP above 12 to 13 per cent of DM while Satter and Roffler (1975) opined that use of NPN substances to replace preformed protein in rations with 12 to 13 per cent CP lowered milk production. Wohlt and Clark (1978) reported that when urea was used to increase the CP content of the ration to 12 per cent, it improved the production. Ruminants fed purified diets containing urea as the only source of nitrogen grew, reproduced and produced milk (Edwards *et al.*, 1980; Ferril *et al.*, 2001). However replacing preformed protein with NPN in the diets of dairy cows often resulted in reduced performance, indicating that all sources of nitrogen are not of equal value (Clark and Davis, 1980). Plant proteins were found to be better than urea for microbial protein production in the rumen (Bhatia and Pradhan, 1980). Clark and Davis (1980) and Sohanae *et al.* (1997) suggested that NPN substances can be included in the diets of dairy cows only if the production is below 20 kg per day. Similar observations were made by Jaquette *et al.* (1986). Santos *et al.*

that in the case of cows producing more than 40 kg milk per day, ration containing true protein as protein supplement increased milk yield than those having urea. Sannes *et al.* (2002) observed no difference in the DMI, milk yield or microbial protein yield (MPY) between rations containing urea and soybean meal (SBM) as protein supplements.

Satter and Roffler (1975) reported that higher rumen NH_3-N than 5mg per 100 ml of the rumen fluid had no beneficial effect on microbial protein synthesis (MPS). Bhatia and Pradhan (1980) observed that the NH_3-N concentration of 5 mg/100ml of rumen liquor was reached at about 13 per cent dietary crude protein. Ha and Kennelly (1984) reported that the microbial nitrogen concentration was highest in cows receiving 15 per cent CP ration compared to those containing 13, 17 and 19 per cent CP and that the ammonia nitrogen concentration in cows fed 15 per cent CP ration was 6mg/100 ml of rumen liquor. However, Devant *et al.* (2000) reported that in cows fed high concentrate diets, NH_3-N concentration was not the limiting factor for microbial growth.

A ruminant animal was found to receive 40 to 80 per cent of its daily amino acid requirement from the microbial protein flowing to the small intestine which in turn is influenced by a number of associated processes (Sniffen and Robbinson, 1987). Synthesis of microbial protein was at the rate of 20 to 22 g per 100 g of total organic matter digested and was increased as the feed intake increased (Robbinson *et al.*, 1985; Merchen *et al.*, 1986), while no correlation between the microbial yield and intake of a fixed diet was reported by several other scientists (Tamninga *et al.*, 1979; Vander-Aar *et al.*, 1984).

2.2.1 Effect of Crude Protein Level on Milk Production

The requirement of crude protein by a dairy cow is defined as the minimum amount of protein that will support maximum production (Clark and

Davis, 1980). Although the dietary protein requirements of dairy cows both for maintenance and for milk production are well established, reports on the influence of varying levels of dietary protein on milk production in early lactation are rather conflicting. There are numerous research reports indicating positive response on milk production to increasing dietary CP levels (Grieve *et al.*, 1974; Van Horn *et al.*, 1979; Edwards *et al.*, 1980; Macleod *et al.*, 1984; Glenn *et al.*, 1986). At protein inputs below requirements, additional increments of protein resulted in increased production and when the requirements were met, no further response on milk production to additional protein was observed (Chalupa, 1980; Hanigan *et al.*, 1998).

Various research workers had assessed the influence of varying levels of dietary protein on milk production. As per NRC (1971) a crude protein level of 13 to 14 per cent of dry matter of a complete ration is sufficient to meet the requirement of lactating cows unless the production is too high. An increase in milk production, DMI and nutrient digestibility with rations having 14 per cent CP compared to that with 11 per cent CP was reported by Kwan *et al.* (1977), Cowan *et al.* (1981) and Kung and Huber (1983). Clark and Davis (1980) reported that the response to increased dietary CP levels on milk production was maximum when the CP content of the ration was raised from 9 to 10 per cent to 13 to 14 per cent. Tolkamp *et al.* (1998) reported higher dry matter intake by increasing ration CP from 13.1 to 18.5 per cent.

Edwards *et al.* (1980) used rations containing three protein levels of 13, 15 and 17 per cent and reported that the 13 per cent CP ration was insufficient to meet the demands of cows producing more than 29 kg milk per day and that there was no significant difference in production between the 15 and 17 per cent CP rations. Roffler and Thacker (1983a) observed a reduction in milk yield of multiparous cows, when ration CP was decreased from 17 to 13.5 per cent after one month of calving. They also reported that reducing protein per cent from 17 to 13.5 after second month of parturition had no deleterious effect on milk yield. Claypool *et al.* (1980) using rations with 12.7, 16.3 and 19.3 per cent CP and Baxter *et al.* (1983) using 12.7, 12.9, 15 and 17.2 per cent CP rations did not get any significant difference in production of cows producing on an average 30 kg milk per day. Similar observations were made by Ha and Kennelly (1984), Holter *et al.* (1985), Lundquist *et al.* (1986), Zimmerman *et al.* (1992) and Robert *et al.* (2000). Christensen *et al.* (1993a) also did not get any significant difference in production between cows producing more than 30 kg milk per day when they were given rations containing 16.4 and 19.4 per cent CP.

Production performance and income above cost of feed for first trimester of lactation of cows producing around 30 kg milk per day were greatest when they received a 16 per cent CP ration (Satter and Roffler, 1975; Claypool *et al.*, 1980; Christensen *et al.*, 1993a). A ration CP of 15 to17 per cent is reported to be optimum for milk production by Roffler and Thacker (1983a) and Claypool *et al.* (1980). Lundquist *et al.* (1983) reported that the milk production by dairy cows receiving a 17.5 per cent CP ration was higher than those receiving 13 and 15 per cent CP rations. Chalupa (1984) reported that increasing CP content of the ration above 16 per cent resulted in diminishing returns because at this stage bacterial destruction of protein was more than the quantity of protein synthesised by microbes. Clark *et al.* (1987) also reported that CP content in excess of 14 to 16 per cent of dietary organic matter did not significantly increase dry matter intake or milk production. Leonard and Block (1988) reported an increase in milk production of cows producing more than 25 to 30 kg milk when the CP of ration was increased from 16 to 21 per cent. Klusmeyer *et al.* (1990) reported an increase in milk production of cows producing more than 30 kg milk per day when they were fed with 14.5 per cent compared to 11 per cent CP ration. But Zimmerman *et al.* (1991) did not get any change in milk production in cows producing around 30 kg per day when the CP level of the diet raised from 18 to 22 per cent. Cunningham *et al.* (1996) reported that the flow of all amino acids to the duodenum was increased when the level of CP in the ration was increased from 16.5 to 18.5 per cent. Pipenbrink *et al.* (1996) observed a similar increase in flow of amino acids to duodenum when 18 per cent CP rations were given compared to 14 per cent CP rations.

Botts *et al.* (1979) reported that high producing cows in early lactation do not consume sufficient dry matter to support maximum production of milk and the relatively small amount of labile protein reserves in the body are rapidly depleted during the period of negative nitrogen balance. The amount of nutrients reaching the site of absorption can be increased either by increasing the quantity and quality of the end products of rumen fermentation or by incorporating nutrients that bypass the rumen (Clark and Davis, 1980). According to Leng and Nolan (1984) the quantity and quality of protein flowing from the rumen is a major factor limiting the productivity of ruminal livestock production. National Research Council (NRC, 1989) recommended 19 per cent CP in the diet of high yielding cows during early lactation when the DMI lags behind the milk production. During early lactation when DMI is low, the microbial protein synthesis was found to be insufficient to meet the protein needs of the animal and inclusion of protein that escape rumen degradation was required for maximising the production (Christensen *et al.*, 1993a).

2.2.2 Effect of Rumen Degradable (RDP) and Undegradable Protein (UDP) Levels on Milk Production

Inclusion of rumen undegradable protein in the diet of lactating cows improved milk production or reduced the amount of conventional protein source required (Satter and Roffler, 1975; Holter *et al.*, 1985). Macleod *et al.* (1984) opined that the loss of ammonia in the rumen will be more in the case of degradable protein leading to lower efficiency of utilisation of these proteins compared to undegradable proteins.

There are large variations among and within feed stuffs in the extent to which they are degraded in the rumen. The protein degradability levels of the feed ingredients at different rumen out flow rates were reported by workers (Gupta and Gupta, 1984; Negi *et al.*, 1989; Garg and Gupta, 1994). The RDP and UDP values of various feed ingredients as such or after various chemical treatments were also assessed (Sampath *et al.*, 1989; Reddy *et al.*, 1992; Kalbande, 1995; Martillotti. *et al.*, 1995).

Of the total protein supplied to animals through diet a part of it should be fermentable in the rumen to meet the nitrogen requirements of the microflora of the rumen. Feeding of bypass protein supplemented with fermentable nitrogen was reported to be superior than bypass protein alone in terms of weight gain and production by animals (Giri and Das, 1993; Senger *et al.*, 1994).

The rumen micro organisms can be divided into two groups, the structural carbohydrate (SC) fermenting and the non-structural carbohydrate (NSC) fermenting group (Russell *et al.*, 1992). Most of the SC fermenting bacteria require ammonia as nitrogen source (Bhatia and Pradhan, 1980) and the nitrogen requirement of the NSC fermenting microbes which are the predominant population can be met by ammonia, peptides and amino acids (Cotta and Russell, 1982; Russell *et al.*, 1983; Jones *et al.*, 1998; Fu *et al.*, 1999; Meng *et al.*, 2000). But Dewhurst *et al.* (2000) opined that in actual feeding the effect of the supplementation of substances such as ammonia, peptides or amino acids might be masked by other factors present in the diet. They also suggested that the microbial protein synthesis (MPS) would be maximised by synchronising the availability of fermentable energy and degradable nitrogen in the rumen.

The effect of protein sources on MPS is more complicated than a synchrony effect or a limitation imposed by effective rumen degradable protein supply. In some situations the pursuit of diets that were high in nondegradable protein led to a deficiency of fermentable carbohydrates or end products of fermentation for rumen microbes and consequently reduced MPS (Siddons *et al.*, 1985; Cecava *et al.*, 1991; Cecava and Parker 1993; Christensen *et al.*, 1993 b). This negative effect of high levels of UDP supplementation on MPS was not there where the fermentable organic matter was not a limiting factor (Elizalde *et al.*, 1998) and the synthesis of microbial protein depended on the source or type of energy supplement available in the diet (Dewhurst *et al.*, 1999). Microbial protein synthesis was increased by supplementing silage based diets with moderate levels of readily fermented carbohydrate (Thomas *et al.*, 1980; Rooke *et al.*, 1985). Feeding of urea treated or ammoniated poor quality roughage such as straw resulted in higher microbial growth (Mohini and Gupta, 1991; Garg and Gupta, 1992).

Feeding of protein supplements of low rumen degradability such as corn gluten meal, dried distillers grains, extruded whole soybean and extruded or roasted cotton seeds to cows increased the amounts of protein passing to the small intestine (Stern *et al.*, 1985; Pena *et al.*, 1986; Cozzi and Polan, 1994; Polan *et al.*, 1997; Chiou *et al.*, 1999). Orskov and Reid (1985) reported that high producing cows require protein in excess of that which is available from the rumen and hence supplementing the diet with protein that escape rumen degradation improved milk production. It has been reported that cotton seed when incorporated at a level of 20 to 25 per cent of the ration, improved milk yield and fat percentage of milk (De Peters *et al.*, 1985; Coppock *et al.*, 1987; Belibasakis and Tsirgogianni, 1996; Sawal and Kurar, 1997). Inclusion of heated whole soybean also improved milk production (Ruegsegger and Schultz, 1985; Atwal *et al.*, 1995). Supplemental UDP in the diet of lactating cows increased milk production (Hibberd *et al.*, 1988; Rusche *et al.*, 1993) or decreased production (Forcherio *et al.*, 1995) while Wiley *et al.* (1991) found no effect on milk production. Reynal and Broderick (2003) opined that when 19 per cent CP rations containing either expeller SBM. solvent SBM. blood meal or corn gluten meal (CGM) were given to dairy cows. expeller SBM produced more milk and milk fat than those of the other treatments.

Compared to soybean meal, fish meal did not increase protein flow to the small intestine (Zerbini *et al.*. 1988; McCarthy *et al.*. 1989) while Waltz *et al.* (1989) observed an increase in the intestinal supply and absorption of amino acids by feeding a combination of blood meal and feather meal compared to those fed SBM. Feeds of animal origin such as fish meal. blood meal and meat cum bone meal were also been used successfully as protein supplements to improve production in cows (Atwal and Erfle. 1992; Carroll *et al.*. 1993; Grummer *et al.*. 1994; Akayezu *et al.*. 1997; Bernard. 1997). Kalbande (1995) reported that coconut cake is also a good source of UDP having an effective degradability of 25 to 30 per cent and cotton seed cake, which is a common ingredient of concentrate mixture is having an effective rumen degradability rate of 35 per cent.

Feeding of bypass protein increased the milk production by cows in early lactation especially when they were in energy deficiency (Clark *et al.*. 1973; Deering *et al.*. 1974; Cressman *et al.*. 1977; Orskov *et al.*. 1981; Roffler *et al.*. 1978). Burroughs *et al.* (1975) and Waldo and Glenn (1984) opined that rumen available protein at levels of 45 to 75 per cent of the ration CPgave maximum production while Majdoub *et al.* (1978) reported that a RDP level of 15 to 30 per cent is most beneficial for dairy cows. In developing countries where animals were mainly fed with crop residue based diets. feeding of by pass protein improved milk production in medium and high producers (Chiou *et al.*. 1995; Ramachandra and Sampath.1995; Misra and Rai 1996; Kalbande and Thomas. 1999). Kalbande and Thomas (1999) reported that increasing the level of UDP of the concentrate mixture from 30 to 63 per cent stimulated feed consumption resulting in better availability of nutrients for higher milk production in crossbred cows maintained on low quality roughage and producing around 10 kg milk per day.

Orskov *et al.* (1981) reported that when the metabolisable energy (ME) intake exceeded 160 MJ per day, changes in protein degradability did not cause any change in production but when the ME intake was below 135 MJ per day, increasing UDP level in the diet led to increased FCM production, milk protein content and a loss in body weight of cows in early lactation.

Multiparous cows responded positively to higher levels of UDP compared to primiparous cows (Roffler *et al.*, 1978; Cressman *et al.*, 1980; Roffler and Thacker, 1983b; Khorasani *et al.*, 1994; Khorasani *et al.*, 1996). On the contrary, Jaquette *et al.* (1986), Jaquette *et al.* (1987), Zimmerman *et al.* (1992) and Bernard (1997) reported a higher level of production for primiparous cows supplemented with UDP while no change in the production was observed in multiparous cows fed the same ration.

Forster *et al.* (1983) observed that decreasing dietary degradability of 14 per cent CP ration from 54 to 43 per cent increased milk production linearly in cows producing around 30 kg milk per day, while Erdman and Vandersall (1983) did not observe any significant change in DMI, milk production, fat per cent and FCM yields when lactating cows were fed 14 per cent CP rations with 53 and 73 per cent degradability. An increase in milk yield by dairy cows in response to increase in UDP levels in the ration was reported by several scientists (Holter *et al.*, 1985; Glenn *et al.*, 1986; Schingoethe *et al.*, 1988; Grummer *et al.*, 1994; Cunningham *et al.*, 1996; Khorasani *et al.*, 1996; Gracia-Bajolil *et al.*, 1998; Volden, 1999; McCormick *et al.*, 1999; Todorov *et al.*, 2000). Higginbotham *et al.* (1989) observed no change in milk yield in cows producing more than 35 kg milk per day when 18 per cent CP rations with 60 and 40 per cent degradability and 15.4 per cent CP rations with 61 and 46 per cent degradabilities were fed.

Casper *et al.* (1990) observed that providing an alternative non structural carbohydrate source (barley Vs corn) did not increase the efficiency of utilisation of a more degradable CP source (urea Vs SBM) in a 16 per cent CP ration. Robbinson *et al.* (1991) reported that when three protein sources namely SBM, CGM or SBM plus whey protein were used in dairy cows, there was no effect on the milk yield by the source and degradability of the protein. Petit and Veira (1991) also observed a lack of response to source of protein or level of concentrate feeding on milk production when concentrate mixture containing SBM at the rate of 1.8 per cent of body weight or concentrate mixture containing SBM or fish meal at 1.3 per cent of body weight were given to cows producing more than 30 kg per day. Henson *et al.* (1997) using different sources of protein in rations with 17 per cent CP at 34 and 45 per cent RDP levels could not observe any influence on milk production by source or degradability of protein.

Christensen *et al.* (1993a) observed no change in milk yield, 4 per cent FCM yield, fat and SNF content of milk when 16.4 and 19.4 per cent CP rations with 55 or 70 per cent protein degradability were fed, while Armentano *et al.* (1993) observed an increase in milk production when UDP level of a 16 per cent CP ration was increased from 26 to 45 per cent levels. Aharoni *et al.* (1993) fed dairy cows producing more than 35 kg milk per day with a 17 per cent CP ration having 70 and 65 per cent degradabilities and observed that the 65 per cent degradable ration was more advantageous for yields of milk and milk components.

Cecava and Parker (1993) from their work using12.5 per cent CP rations containing either soy bean meal (SBM), corn gluten meal (CGM) or CGM plus blood meal (BM) as protein supplements concluded that the source of ruminally degradable protein influenced the efficiency of feeding ruminally degradable and undegradable CP in combination and that the source of supplemental CP had a greater influence on the quantity than the profile of absorbable amino acid supplied to the duodenum.

Triplet *et al.* (1995) used soybean meal (RDP source) and or fish meal (UDP source) to vary UDP levels (38, 56 and 75 per cent UDP) in three supplements to lactating Brahman cows. The milk production by cows receiving rations containing 56.3 per cent UDP was more than that of cows given 75.6 per cent UDP. Alderton *et al.* (2000) also got similar reduced production in crossbred beef cows when UDP sources were fed alone compared to a mixture of RDP and UDP. Santos *et al.* (1998a) opined that in dairy cows producing more than 40 kg per day, supplementation of true protein yielded more milk than urea supplementation.

Herera-Saldana and Huber (1989) and Gracia- Bojalil *et al.* (1998) observed that during early lactation when UDP was given to improve DMI and milk production, the response by the cows depended on the degradability of protein as well as on the availability of digestible carbohydrate. Tamlinson *et al.* (1993) observed 77 per cent increase in nitrogen excretion with no significant increase on milk nitrogen when CP concentration increased from120 to180 g/ kg DM. This emphazises the fact that merely increasing the protein concentration in the diet results in lower efficiency of protein utilization. Kalscheur *et al.* (1999) opined that in early lactation the milk yield increased as the UDP level of 15.2 and 17.4 per cent CP rations were increased from 35.5 to 46.5 per cent and in late lactation (wk 34 to 44) milk and 4 per cent FCM yields decreased linearly as UDP percentage increased. Wu and Satter (2000) opined that for cows producing around 10000 kg in 308 days, the protein level of the concentrate mixture. containing roasted soybean (SB) or expeller SBM can be reduced from 17.5 to 16 per cent at about 30 weeks of lactation.

Castilo *et al.* (2001) reported that feeding two rations with 21 and 29 per cent CP at three degradability levels. i.e. 64. 52 and 38 per cent did not affect the faecal and milk nitrogen output. They also reported that a low level of protein (150 g/kg DM) and medium to low rumen degradable protein (38 to 52 per cent) supplements significantly reduced nitrogen excretion without compromising lactation response (average production 25 kg/ day).

Based on the various reports of influence of protein degradability on milk production. Nocek and Russell (1988) concluded that the failure of undegradable protein to increase milk production as reported by many workers might be due to (1) high dietary CP levels in relation to the cows requirements (2) low productivity of cows (3) low availability of protein post ruminally and (4) low ruminal ammonia levels and decreased MPS.

2.3 EFFECT OF LEVEL AND DEGRADABILITY OF PROTEIN ON MILK COMPOSITION

Spires *et al.* (1975) and Orskov *et al.* (1981) observed an increase in the milk protein per cent and yield in cows infused with sodium caseinate post ruminally. Dietary protein levels had no effect on milk protein and fat levels (Roffler *et al.*, 1978; Foldger and Huber. 1979; Van Horn *et al.*. 1979; Claypool *et al.*. 1980; Edwards *et al.*. 1980). while Barney *et al.* (1981). Forster *et al.* (1983) and Henderson *et al.* (1985) reported a linear increase in both milk protein and fat levels in response to increasing dietary CP level from 16 to 22 per cent. Crooker *et al.* (1983) and Crawford and Hoover (1984) reported a reduction in the milk protein levels when cows were fed formaldehyde treated SBM incorporated ration compared to that containing untreated SBM. Cameron *et al.* (1991) reported increased milk protein levels when the diets were supplemented with urea. Faldet and Satter (1991) observed a reduction in milk protein level. but an increase in the milk and 3.5 per cent fat corrected milk (FCM) production when heat treated SBM was included in a concentrate alfalfa ration (50:50) in place of SBM.

Microbial protein is having an amino acid profile similar to milk protein and hence a shortage of MPS resulted in negative effect on milk protein synthesis (Santos *et al.*. 1998 b). They also stated that the amino acid profile of the escape protein and duodenal digesta influenced the milk protein synthesis and commonly used UDP sources had an amino acid profile inferior to microbial protein especially with regard to lysine and methionine contents. Wright *et al.* (1998) observed a linear increase in the milk protein level when the concentration of a UDP supplement. having amino acid composition similar to bovine casein. increased from 4.5 to 29.1 percentage.

There was no change in milk fat percentage by increasing the undegradable protein content of the ration (Kung *et al.*. 1983; Stern *et al.*. 1985: Zerbini *et al.*. 1988; Klusmeyer *et al.*. 1990; Christensen *et al.*. 1993b). while an increase in the milk fat percentage was reported in response to increased level of cotton seed in the ration by DePeters and Tayler (1985). Coppock *et al.* (1987). Blauwiekel *et al.* (1990) opined that when fish meal was used as UDP supplement. there was decrease in the milk fat and SNF contents compared to an isocaloric. isonitrogenous ration containing SBM.

Roffler and Thacker (1983a) observed that the milk composition in primiparous cows and fat content in the milk of multiparous cow were unaffected by the level of CP in the ration. Robbinson and Kennelly (1988) did not get any change in milk composition when two rations with 16 and 19 per cent CP were fed at 21 and 37 per cent RDP levels to dairy cows. Similarly. Armentano *et al.* (1993). Atwal *et al.* (1995) and Akayezu *et al.* (1997) also observed no effect of protein solubility level on milk composition. Nianogo *et al.* (1991) reported that the percentage composition and yield of milk. SNF. CP and total solids were higher in cows calving in fall than in cows calving in summer and that ration fat and escape protein had no effect on milk composition. Aharoni *et al.* (1993) observed that when a 17 per cent CP ration at 70 and 65 per cent degradabilities was given to cows there was no difference in milk protein content, but the milk fat percentage was higher for cows given diets of low degradability. O'Mara *et al.* (1998) reported that when fishmeal and SBM were included as protein supplements at 7 kg per day, the yield of milk constituents, such as protein and lactose were significantly higher than when they were fed at 3.5 kg per day.

A major component of the milk NPN is urea which accounts for 35 to 48 per cent of the total NPN and this level varies between individual cows, breed, stage of lactation, season and diet (Cebrulis and Forrel Jr, 1975). Blood is the major source of milk NPN and there is a close correlation between blood urea nitrogen (BUN) and milk urea nitrogen (MUN) content (Refsdal, 1983; Oltner and Wiktorson, 1983; Veen and Baker, 1988; Martinez *et al.*, 1991). The average NPN concentration of the milk is approximately 25 to 35 mg per 100 ml which forms five to six present of the total milk nitrogen (De Peters and Ferguson, 1992). Proportions of true protein and urea in the milk are influenced by the CP concentration, protein type and protein quality (Baker *et al.*, 1995).

Roseler *et al.* (1993) observed that while varying the RDP and UDP levels in the ration influenced the milk yield, it did not influence the CP and true protein content of milk. Oltner and Wiktorsson (1983) reported an increase in MUN levels from 8.7 to 29.4 mg /100ml when the dietary CP levels of isocaloric diets were increased from 11.6 to 17.6 per cent. Oltner (1985) reported that the MUN levels were negatively correlated with live weight changes and according to Jonker *et al.* (1998) a change of 50 kg and above in weight influences the MUN levels. Broderick *et al.* (1990) reported that supplementing alfalfa silage based diets with protein sources varying in protein degradability did not alter the rumen NH_3-N or BUN but altered MUN levels. Rodriguz *et al.* (1997) reported an increase in the MUN level by 8.8 per cent by increasing the UDP level from 29 to 41 per cent. Similar increase in MUN levels was reported by Moscardini *et al.* (1998) and Schepers and Meijer (1998). Schepers and Meijer (1998) also observed that parity and stage of lactation did not influence the MUN levels. Kanjanapruthipong and Butanong (2000) observed a significant increase in MUN levels by feeding untreated soybean meal based diets in comparison to an isonitrogenous, isocaloric diet containing formalin treated SBM as protein supplement.

2.4 EFFECT OF LEVEL AND DEGRADABILITY OF PROTEIN ON RUMEN AND BLOOD PARAMETERS

2.4.1 Effect on Rumen pH

Church (1976) suggested that ruminal pH was influenced by the amount of saliva entering the rumen and the amounts of organic acids particularly lactic acid and volatile fatty acids that are produced and accumulated in the rumen contents. Claypool *et al.* (1980) using three levels of CP in the ration (12.7,16.3 and 19.3 per cent) observed that there was no significant difference in the rumen pH (average 6.4) between the treatments. Lack of influence of dietary protein levels on rumen pH was also reported by other scientists (Cressman *et al.*, 1980; Forster *et al.*, 1983; Ha and Kenelly, 1984; Annexstad *et al.*, 1987; Warley *et al.*, 1994; Devant *et al.*, 2000). Crawford *et al.* (1978) reported that there was an increase in the rumen pH as the dilution rate was increased. Meng *et al.* (1999) and Meng *et al.* (2000) also reported similar observations. Klusmeyer *et al.* (1990) reported an increase in rumen pH by feeding a 11 per cent CP ration compared to 14.5 per cent CP ration. Broderick *et al.* (1993) observed an increase in rumen pH when urea was added as the supplemental protein source compared to true protein supplements. Christensen *et al.* (1993b) reported that the rumen pH was not affected by the CP content of the ration but was reduced when UDP was increased from 30 to 45 per cent without affecting the fibre digestibility. Devant *et al.*

(2001) using 14 per cent CP rations having 41, 49, 52 and 56 per cent RDP, reported that neither source nor degradability of the dietary protein affected the rumen pH. Similar observations were also made by Forster *et al.* (1983), Leonard and Block (1983), Krickpatrick and Kennelly (1989) and Kanjanapruthipong *et al.* (2002).

2.4.2 Effect on Total Volatile Fatty Acid Production (TVFA)

Mudgal and Puri (1977) reported that higher levels of VFA in rumen fluid were observed when groundnut cake was fed along with molasses than that of urea supplementation. Cressman *et al.* (1980) fed 12.4, 15.1 and 17.7 per cent CP rations results of which showed no change in the level of TVFA in the rumen between the treatments. Protein supplements such as soybean, soybean meal, sunflower seeds and cottonseed had no significant effect on the TVFA level in the rumen (Smith *et al.*, 1981; White *et al.*, 1987; Srivasthava and Mani, 1991). Kung.Jr. *et al.* (1983) and Khorasani *et al.* (1994) observed that the source of protein had no effect on the level of TVFA in the rumen. Ha and Kennelly (1984) observed a tendency towards an increase in acetic acid proportion even though there was no significant effect on TVFA production by increasing CP levels in the ration from 13 to 19 per cent. DeFarier and Huber (1984) reported that increasing CP levels from 8.1 to 11.3 and 13.3 per cent by adding urea increased TVFA level. Klusmeyer *et al.* (1990) fed 11 per cent and 14.5 per cent CP ration to lactating cows and found a reduced VFA production in the rumen of cows given the 11 per cent CP ration. They also reported that the source of supplemental protein did not alter the molar percentages of the VFAs but TVFA concentration was greater when SBM diets were fed compared to CGM diets. Krikpatrick and Kennelly (1990) and Calsaminglia *et al.* (1995) also reported a higher TVFA production when SBM containing rations were fed to cows.

Grubic (1991) reported an increase in the acetic acid and a decrease in the propionic acid levels when the dietary UDP levels were increased. Pires *et al.*

(1997) also reported an increase in acetate proportion by incorporation of roasted cottonseed in the ration. Christensen *et al.* (1993b) suggested that any factor which led to the excessive fermentation in the rumen resulted in higher levels of TVFA in the rumen fluid. Sawal and Kurar (1997) opined that the TVFA concentration in the rumen was governed more by the form and content of carbohydrate in the ration rather than the kind of protein supplement. Robbinson and Mcqueen (1994), O'Mara *et al.* (1998) and Meng *et al.* (2000) reported that the RDP level of the ration had no effect either on the production of TVFA or the concentration of individual VFA. Sawal and Kurar (1997) opined that the VFA concentration in the rumen was governed more by the form and content of carbohydrate in the ration rather than the kind of protein supplement.

Chan *et al.* (1997) reported an increase in the TVFA production with lowered ratio of acetate to propionate when animal protein supplements such as fish and blood meal were given. Mathis *et al.* (2000) reported an increase in the TVFA production in response to an increase in the UDP level and that the proportion of acetate reduced while that of propionate increased on UDP supplementation of animals fed medium to low quality forages.

2.4.3 Effect on Rumen Ammonia Nitrogen (NH_3-N) and Blood Urea Nitrogen (BUN)

Satter and Roffler (1975) reported an increase in the rumen NH_3-N concentration from 0.8 to 56.1 mg per 100 ml as the protein level of the ration was increased from 8 to 24 per cent. Rumen NH_3-N concentration increased linearly as the dietary CP levels were increased (Polan *et al.*, 1976; Kwan *et al.*, 1977; Ha and Kennelly, 1984; Janicki *et al.*, 1985; Warley *et al.*, 1992; Jones- Endsley *et al.*, 1997). There was a reduction in the ammonia production in the rumen as a result of reduced protein solubility of the diet (Howard *et al.*, 1987; Carrol *et al.*, 1988;; Scott *et al.*, 1991; Nicholson *et al.*, 1992; Carroll *et al.*, 1994; Shabi *et al.*, 1998 ; Casper *et al.*, 1999). De Farier and Huber (1984) reported that increasing the CP level of the ration from 8.1 to 13.3 per cent by adding urea resulted in elevated levels of rumen NH_3-N concentration. Santos *et al.* (1984) reported a 40 per cent increase in the NH_3-N concentration in the rumen when SBM containing rations were fed compared to isonitrogenous rations containing CGM. wet brewers grains or dried distillers grains. Janicki *et al.* (1985) reported that when RDP at 39.7 and 47.9 per cent were fed, nitrogen solubility of the diet had no effect on the rumen NH_3-N or BUN concentrations. Blauwiekel and Kincaid (1986) and Carroll *et al.* (1988) observed higher levels of rumen ammonia production and higher plasma nitrogen concentration when the ration CP level was increased from 13 to 20 per cent. Bowman *et al.* (1988) reported a decrease in the rumen NH_3-N by using sodium hydroxide treated SBM compared to untreated SBM. Zimmerman *et al.* (1992) reported that when the fibre levels were low (11 per cent ADF) rumen NH_3-N level was higher when 18.7 per cent CP ration containing untreated SBM was fed than those fed 14.5 per cent and 18.5 per cent CP rations containing UDP supplements. Devant *et al.* (2000) in their experiment with two rations having 17 and 14 per cent CP and 58 and 42 per cent RDP, observed that when low RDP low CP rations were fed rumen NH_3-N concentrations in the rumen fell below the critical level of 5 mg per 100 ml.

Kung *et al.* (1983) and Sahlu *et al.* (1984) observed no change in the rumen ammonia and BUN levels when fed rations containing heated SBM and untreated SBM. Jordan *et al.* (1983) reported an increase in the level of the BUN when CP of the ration was increased from 12 to 23 per cent. Similar elevated levels of BUN in response to increased CP levels in the ration were also observed by Cross *et al.* (1974). Botts *et al.* (1979), Cressman *et al.* (1980), Howard *et al.* (1987) and Higginbotham *et al.* (1989). Higginbotham *et al.* (1989) also observed no influence of protein degradability on the BUN levels when RDP at 60 and 40 per cent levels were given. Rodriguz *et al.* (1997) observed an increase in the plasma urea nitrogen (PUN) level when the UDP content of a 16 per cent CP ration was increased from 29 to 41 per cent. Similarly, Moscardini *et al.* (1998)

reported a significant increase in the PUN and milk urea nitrogen levels as concentration of UDP in the ration was increased from 4.5 to 29 per cent.

Ruegsegger and Schultz (1985) observed no change in the plasma glucose levels by replacing SBM with treated SBM in 18 per cent CP ration. Grubic (1991) also reported that increasing the UDP levels in the ration did not influence the blood urea and glucose levels.

2.5 EFFECT OF LEVEL AND DEGRADABILITY OF PROTEIN ON REPRODUCTION

High dietary protein intake was found to be detrimental to reproductive performance of dairy cows (Trecker *et al.*, 1976). Jordan and Swanson (1979) and Folman *et al.* (1981) reported an increase in the number of services per conception in dairy cows fed ration containing more than 16 per cent CP. The deleterious effects of high protein feeding was reported to be due to the elevated levels of plasma urea nitrogen (PUN) concentration (Jordan *et al.*, 1983; Duby *et al.*, 1984; Johnson *et al.*, 1986) which in turn was linked with ruminally degradable protein levels of dietary protein (Folman *et al.*, 1981; Canfield *et al.*, 1990; Ferguson *et al.*, 1993). Higher urea concentrations in the blood may be toxic to reproductive tissues (Jordan *et al.*, 1983; Ferguson *et al.*, 1989; Elrod and Butler, 1993) which may lead to reduced ova viability. Jordan *et al.* (1983) further reported that the decreased fertility might be by altering the ionic concentration of the utrine fluid during luteal phase in dairy cows. Kaim *et al.* (1983) reported that the pregnancy rates in cows fed 20 per cent CP rations were lower than those fed 15 per cent CP ration and the plasma urea nitrogen was 16.8 mg and 9.0 mg/dL when fed the 20 and 15 per cent CP rations, respectively. Carroll *et al.* (1988) on the other hand observed no difference in the reproductive efficiency between cows fed 13 and 20 per cent CP rations. Canfield *et al.* (1990) reported a lower conception rate in cows having a PUN level of 18.6 mg/dL than those having a PUN level of 15.7 mg/dL, where as Howard *et al.*

(1987) and Carroll *et al.* (1988) observed no deleterious effect of more than 24 mg/dL PUN on reproduction in cows fed high protein diets. The first pregnancy rates were lowered by 20 per cent in heifers fed 22 per cent CP ration with 83 per cent RDP in comparison with those received 15.5 per cent CP ration at 73 per cent RDP (Elrod and Butler, 1993). Ferguson *et al.* (1993) reported that BUN concentrations exceeding 20 mg per dL were associated with reduced conception rates in lactating cows. Carroll *et al.* (1994) reported that there was no effect on reproduction in cows fed 21 per cent CP rations at 40 (fishmeal) and 34 (SBM) per cent UDP level. A significantly higher incidence of cystic ovaries and delayed first observed heat were reported by Kanjanapruthipong and Butanong (2000) in cows given isonitrigenous, isocaloric total mixed rations with SBM as protein supplement compared to those fed rations containing formalin treated SBM.

Butler and Smith (1989), Ferguson and Chalupa (1989) and Swanson (1989) opined that cows in tropics are likely to be affected with reproductive problems caused by negative energy balance, higher rumen NH_3-N due to excess dietary RDP and deficiency of amino acids.

Materials and Methods

3. MATERIALS AND METHODS

The feeding experiments were conducted at University Livestock Farm. Mannuthy. using crossbred dairy cows in early lactation in two phases. In the first phase. rations varying in their crude protein (CP) content were given to cows in early lactation to optimize the level of CP in the ration. In the second phase the CP level that gave the best result in the first phase was used at two levels of rumen undegradable protein in another set of 12 early lactating cows.

3.1 PHASE 1

Twelve lactating crossbred cows belonging to University Livestock Farm (ULF& FRDS). Mannuthy were selected and grouped into two as uniformly as possible with respect to their age. stage of lactation. parity. body weight and previous milk yield. They were selected immediately after calving and the rations were randomly allotted to the two groups. The animals were fed with isocaloric experimental rations comprising of concentrate mixtures having 26 and 20 per cent CP on DM basis and paddy straw formed the sole roughage. The concentrate mixture and paddy straw were fed in the ratio 70:30 of the total DM intake so that the total protein in the rations were 19 and 15 per cent. respectively. Based on the observations during the first month. one more group with another six early lactating animals was introduced and they were fed with a concentrate mixture having 17 per cent CP (13 per cent CP in the total ration). The ingredient composition of the three experimental rations are given in Table 1. An adaptation period of seven days was given to each animal.

3.1.1 Feeding

The experimental animals were stall fed with the three rations. The total dry matter given was fixed at three per cent of the body weight in the beginning of the experiment, which later on was increased to four per cent of the body weight according to their intake. The quantity of concentrate mixture and roughage fed was in the ratio of 70:30 of the total DM for each animal. The calculated nutrient contents of the three rations are given in Table 2. The experimental animals had free access to clean, wholesome drinking water. The animals were fed with the concentrate twice daily, in the morning and in the afternoon before milking. Paddy straw was offered throughout the day. Individual data on the daily DM intake and milk produced were recorded through out the experiment. Animals were weighed at fortnightly interval and the quantity of feed given, was revised accordingly. The experiment was continued for a period of 100 days for all the groups.

3.1.2 Milk Production

Milk samples were collected fortnightly from each animal and analysed for total solids, fat (IS: 1224, 1977), protein (AOAC, 1990, N*6.38), and solids not fat (SNF).

3.1.3 Haematological Parameters

Blood from the experimental animals were collected on the first, seventh and 13th week of the feeding trial and were analysed for haemoglobin (Wong, 1928), blood glucose (Folin and Wu, 1920), plasma calcium (Clark and Collip, 1925), plasma phosphorus (modified Metol method using the kit supplied by Qualigens laboratory Pvt. Ltd.) and blood urea nitrogen (Coulombe and Favrreau, 1963).

3.1.4 Rumen Fermentation Parameters

Rumen liquor was collected from all the animals using a stomach tube at the beginning and towards the end of the feeding trial and was analysed for pH (pH meter), total volatile fatty acids (Barnett and Reid, 1957), rumen ammonia nitrogen (Beecher and Whitten, 1970) and lactic acid (Pennington and Sutherland, 1956).

3.1.5 Digestibility Trial

A digestibility trial was conducted during the last seven days of the experiment. Dung samples collected each day were weighed and samples at the rate of one per cent of the total weight were collected from each animal in double lined polythene bags after mixing thoroughly. The samples collected each day were kept frozen in a deep freezer. After the end of the trial, samples collected for each animal for the seven days were pooled, mixed together and sub samples were taken for analysis. The moisture was determined immediately and the crude protein was estimated using the fresh samples. The rest of the analysis was done using dried dung samples. During the collection period feed samples were collected daily from each animal and at the end of the trial, feed samples collected each day for each animal were pooled and sub samples were taken for analysis.

3.1.6 Analysis of Feed and Dung

Proximate analysis of the experimental concentrate mixture, dung and paddy straw was carried out as per standard procedure (AOAC, 1990). The acid detergent fibre (ADF) was estimated by the method suggested by Van Soest (1963) and neutral detergent fibre (NDF) by method suggested by Van Soest and Whine (1967). The calcium content in the feed and straw was found out by Atomic Absorption Spectrophptometry using hollow calcium cathode tubes. Phosphorus content in the feed and straw was found out by Vanado-Molybdate method (AOAC, 1990).

3.1.7 Statistical Analysis

The results were analysed statistically as per standard procedure (Snedecor and Cochran, 1985).

3.2 PHASE II

Another set of twelve crossbred cows in early lactation was selected and they were grouped into two as uniformly as possible with respect to their age, stage of lactation, parity, body weight and previous milk yield. Based on the results obtained, the concentrate mixture containing 17 per cent CP on DM was selected for the second trial. Two isocaloric concentrate mixtures containing 17 per cent CP having 26.8 and 42.9 per cent of their CP as undegradable protein were allotted to the two groups randomly. The ingredient composition of the two concentrate mixtures are given in Table 3.

3.2.1 Feeding

The animals were selected immediately after parturition and were fed with the two concentrate mixtures and paddy straw as the roughage in the ratio 70:30. The calculated nutrient content of the two rations are given in Table 4. The animals were fed at the rate of three per cent of their body weight at the beginning of the experiment and was revised fortnightly according to their body weight. The animals were maintained, fed and managed as in phase I. The total experimental period was for 100 days.

3.2.2 Collection of Materials

Milk samples collected every fortnight and were analysed for total solids, fat, protein and SNF as per the methods detailed in phase1. In addition, milk urea nitrogen was also estimated by the method suggested by Bector *et al.* (1998).

Blood samples collected were analysed for Hb. blood glucose. blood urea nitrogen, plasma calcium and plasma phosphorus and rumen liquor for pH, total volatile fatty acids, ammonia nitrogen and lactic acid in the same manner as in phase1.

3.2.3 Digestion Trial

A digestion trial was carried out during the last seven days of the feeding period. Total collection of dung was done and samples taken and analysed as described for phase-I

3.2.4 Statistical Analysis

The results obtained were analysed as per standard procedure suggested by Snedecor and Cochran, (1985).

Table 1. Per cent ingredient composition of the three experimental concentrate mixtures used in phase I.

Ingredient	Concentrate mixtures		
	I	II	III
Yellow Maize	46.0	46.0	46.0
Groundnut Cake	50.5	31.0	21.0
Rice Polish	0	20.0	30.0
Mineral Mixture *	1.5	0	0
Salt	1.0	1.0	1.0
Shell Grit	1.0	2.0	2.0

To 100 kg of the above concentrate mixture, 600 g sodium bicarbonate and 12 g of Indomix ** were also added

* Mineral mixture supplied by Cacils India Ltd. Bombay, containing Calcium (minimum) 24 per cent, Phosphorus (minimum) 12 per cent, Manganese (minimum) 0.12 per cent, Copper (minimum) 0.12 per cent, Zinc (minimum) 0.35 per cent, Magnesium (minimum) 6.5 per cent, Iron (minimum) 0.5 per cent, Cobalt (minimum) 0.03 per cent, Sulphur (minimum) 0.5 per cent, Acid insoluble ash (maximum) 2 per cent and Flourine (maximum) 0.45 per cent.

** Indomix supplied by Nicholas Prima India Ltd. Bombay, having 40,000 I.U. of vitamin A, 20.mg of vitamin B_2 and 5,000 I.U. of vitamin D_3 per gram.

Table 2. Calculated nutrient content of the three concentrate mixtures and rations used in phase 1.

Nutrient	Concentrate mixture (%)			Ration (%)		
	I	II	III	I	II	III
TDN	72.5	72.5	72.5	62.75	62.75	62.75
CP	26.07	20.74	17.00	19.4	15.72	13.10
ADF	2.7	3.5	4.0	17	17.6	18.0
Ca	0.82	0.79	0.80	0.65	0.63	0.63
P	0.63	0.65	0.77	0.46	0.48	0.56

Table 3. Per cent ingredient composition of the two experimental concentrate mixtures used in phase II.

Ingredient	Concentrate mixtures	
	I	II
Yellow maize	45.0	30.0
Ground nut cake	20.0	6.0
Wheat bran	26.4	15.4
Coconut cake	5.0	45.0
Salt	1.0	1.0
Shell grit	2.0	2.0
Sodium bicarbonate	0.6	0.6

To 100 kg of the above concentrate mixture 12 g of Indomix * was added

* Indomix supplied by Nicholas Prima India Ltd. Bombay, having 40,000 I.U. of vitamin A, 20 mg of vitamin B_2 and 5,000 I.U. of vitamin D_3 per gram.

Table 4. Calculated nutrient content of the two concentrate mixtures and rations used in phase II.

Nutrient	Concentrate mixture (%)		Ration (%)	
	I	II	I	II
TDN	71.11	72.02	61.78	62.41
CP	17.45	17.34	13.72	13.64
ADF	14.8	16.5	25.1	25.8
UDP	26.81	42.96	18.77	30.07
Ca	0. 83	0.85	0.70	0.72
P	0.65	0.63	0.45	0.44

Results

4. RESULTS

The results obtained in the present study are given under the following headings

4.1 CHEMICAL COMPOSITION

The data on the chemical composition of the concentrate mixtures and paddy straw used in phases one and two of the experiment are given in Table 5 and Table 17, respectively. The crude protein content (in per cent) of the three concentrate mixtures used in phase I were 26.5 ± 0.34, 20.01 ± 0.11 and 16.85 ± 0.10 while that of the straw used for feeding was 4.68 ± 0.46 on DM basis. The CP content of the concentrate mixtures used in the second phase was 17.24 ± 0.52 and 17.18 ± 0.18 while that of straw was 4.22 ± 0.22 per cent. The composition of the dung of the experimental animals of phase I and II are given in Tables 14 and 25, respectively.

4.2 BODY WEIGHT

The data on the fortnightly body weight of the experimental animals maintained on the various rations in phases I and II are given in Tables 6 and 18, respectively, and their covariance analysis in Tables 7 and 19, respectively. The summarised data on the body weight of the animals from both the phases are given in fig. 1 and 11, respectively. The initial body weight of the animals in the three groups I, II and III were 349.8 ± 21, 347.7 ± 12 and 337.5 ± 9.9 kg while the final weight was 323.3 ± 18.6, 356.0 ± 13.1 and 323.2 ± 15.3 kg, respectively.

4.3 DRY MATTER INTAKE (DMI)

The data on the daily average dry matter intake of the animals maintained on the experimental rations in both phases are given in Tables 8 and 20, respectively. The summarised data on the DMI of the animals of the three groups fed three levels of CP in phase I and that of animals fed rations with two levels of UDP in phase II are given in Fig. 2 and 12, respectively.

4.4 MILK PRODUCTION

The average daily milk production of the animals maintained on the experimental rations during phase I and II are given in Tables 9 and 20, respectively. The covariance analysis of these data is shown in Tables 10 and 21. The summarised data on the milk production by the animals in phase I and II are presented in Fig. 3 and 13, respectively.

4.5 MILK COMPOSITION

The composition of milk, viz. fat, total solids, solid not fat and protein from the animals of both the phases of the experiment collected at various intervals are given in Table 11 and 22, and the summarised data from both phases in Fig.4 and 14 (1) and (2), respectively.

4.6 RUMEN FERMENTATION PARAMETERS

The data on the rumen fermentation parameters namely, pH, ammonia nitrogen (NH_3-N), total volatile fatty acids (TVFA) and lactic acid levels of the rumen liquor from the animals of both the phases towards the end of the experiment are given in Tables 12 and 23, respectively. The summarised data on the rumen fermentation parameters from both the phases are given in Fig.5 and 15, respectively. The initial values of these parameters determined before the beginning of the experiment were 6.00 ± 0.12 for pH, 82.97 ± 6.84 meq/l for TVFA, 29.07 ± 0.36 mg/100ml for rumen NH_3-N and 0.32 ± 0.08 mg/ml for lactic acid levels in the first phase of the experiment while the corresponding levels were 7.38 ± 0.52, 47.86 ± 6.77 meq/l, 16.68 ± 0.45 mg/100 ml and 0.22 ± 0.04 mg/ ml, respectively in the second phase.

4.7 HAEMATOLOGICAL PARAMETERS

The data on the values of haemoglobin, blood urea nitrogen (BUN), blood glucose, plasma calcium and phosphorus content of the blood samples collected from the experimental animals of both the phases of the experiment are shown respectively in Tables 13 and 24. The summarised data of these parameters of phase I are presented in Fig. 6 and 7 and that of phase II in Fig. 16, respectively. Blood samples collected initially before the beginning of the first phase had 8.99 ± 0.68 g per cent of haemoglobin, 59.16 ± 4.98 mg per cent of blood glucose, 18.10 ± 0.06 mg per cent of BUN, 8.86 ± 0.81 mg per cent of plasma calcium and 6.65 ± 0.28 mg per cent of plasma phosphorus while the corresponding values of phase II were 11.56 ± 0.84 g per cent, 47.02 ± 2.81 mg per cent, 20.98 ± mg per cent, 10.86 ± 0.5 mg per cent and 9.83 ± 1.00 mg per cent, respectively.

4.8 DIGESTIBILITY COEFFICIENT

The digestibility coefficient of the nutrients of the rations, namely dry matter (DM), crude protein (CP), crude fibre (CF), ether extract (EE), nitrogen free extract (NFE), neutral detergent fibre (NDF) and acid detergent fibre (ADF) of phase I and II of the experiment are given in Tables 15 and 26, respectively. The summarised data on the digestibility coefficient of the nutrients in the two phases are given in Fig. 8 and 17, respectively.

4.9 DRY MATTER INTAKE AND COST OF PRODUCTION

The total dry matter intake (DMI), DMI per 100 kg body weight, DMI per kg metabolic live weight and cost per kg milk production of both the phases of the trial are given in Tables 16 and 27, respectively. The summarised data on DMI and cost of milk production in both phases are given in Fig. 10 and 19, respectively.

The summarised data on days to first postpartum insemination in both phases of the experiment are presented in Fig.9 and 18, respectively.

Table 5. Per cent chemical composition * of the three experimental concentrate mixtures and paddy straw used in phase 1 (on dry matter basis) .

Parameter	Concentrate mixtures			Paddy straw
	I	II	III	
Dry matter	95.88 ± 0.28	95.32 ± 0.22	95.34 ± 0.18	90.38 ± 0.09
Crude protein	26.55 ± 0.34	20.01 ± 0.11	16.85 ± 0.10	4.68 ± 0.46
Ether extract	5.31 ± 0.41	5.13 ± 018	5.45 ± 0.31	3.48 ± 0.22
Crude fibre	3.65 ± 0.33	5.42 ± 0.36	6.92 ± 0.22	32.70 ± 0.11
Total ash	6.52 ± 0.06	9.93 ± 0.51	9.76 ± 0.14	10.88 ± 0.36
Nitrogen free extract	57.97 ± 0.08	59.51 ± 0.22	61.02 ± 0.14	48.26 ± 0.18
Acid insoluble ash	2.01 ± 0.02	3.54 ± 0.48	3.62 ± 0.22	2.46 ± 0.08
Neutral detergent fibre	10.59 ± 0.06	12.85 ± 0.15	16.36 ± 0.07	77.71 ± 0.11
Acid detergent fibre	5.28 ± 0.09	8.88 ± 0.33	15.07 ± 0.08	44.99 ± 0.21
Acid detergent lignin	2.98 ± 0.12	4.17 ± 0.13	6.19 ± 0.11	5.34 ± 0.42
Calcium	1.08 ± 0.02	1.20 ± 0.08	1.20 ± 0.02	0.30 ± 0.05
Phosphorus	0.46 ± 0.05	0.55 ± 0.10	0.91 ± 0.21	0.13 ± 0.02

* Average of six values.

Table 6. Body weight of animals* maintained on the three experimental rations in phase I

Fortnight	Body weight (kg)		
	Group I	Group II	Group III
0	349.8 ± 21.0	347.7 ± 12.0	337.5 ± 9.9
1	335.0 ± 17.8	343.0 ± 13.1	330.0 ± 12.2
2	338.0 ± 18.3	354.0 ± 15.8	334.0 ± 12.7
3	331.0 ± 19.1	345.0 ± 13.9	325.0 ± 10.6
4	323.0 ± 17.8 ^{b}	345.0 ± 14.6 ^{a}	330.0 ± 10.2 ^{b}
5	321.0 ± 18.6	354.0 ± 14.6	336.0 ± 15.1
6	323.3 ± 18.6 ^{b}	356.0 ± 13.1 ^{a}	323.2 ± 15.3 ^{b}

*Average of six values.

a , b- means with different superscripts in the same row differ significantly ($P < 0.05$)

Table 7. Analysis of the covariance of the body weight of the animals maintained on the three experimental rations in phase I

Fortnight	Mean sum of the squares			P value
	Groups	Covariant	Error	
1	199.83	18203.9	67.26	0.08
2	530.3	18553.99	276.83	0.18
3	379.07	14830.61	381.16	-----
4	1568.88	14494.44	325.75	0.02
5	1866.17	15497.69	718.86	0.1
6	1984.548	177883.31	405.442	0.02

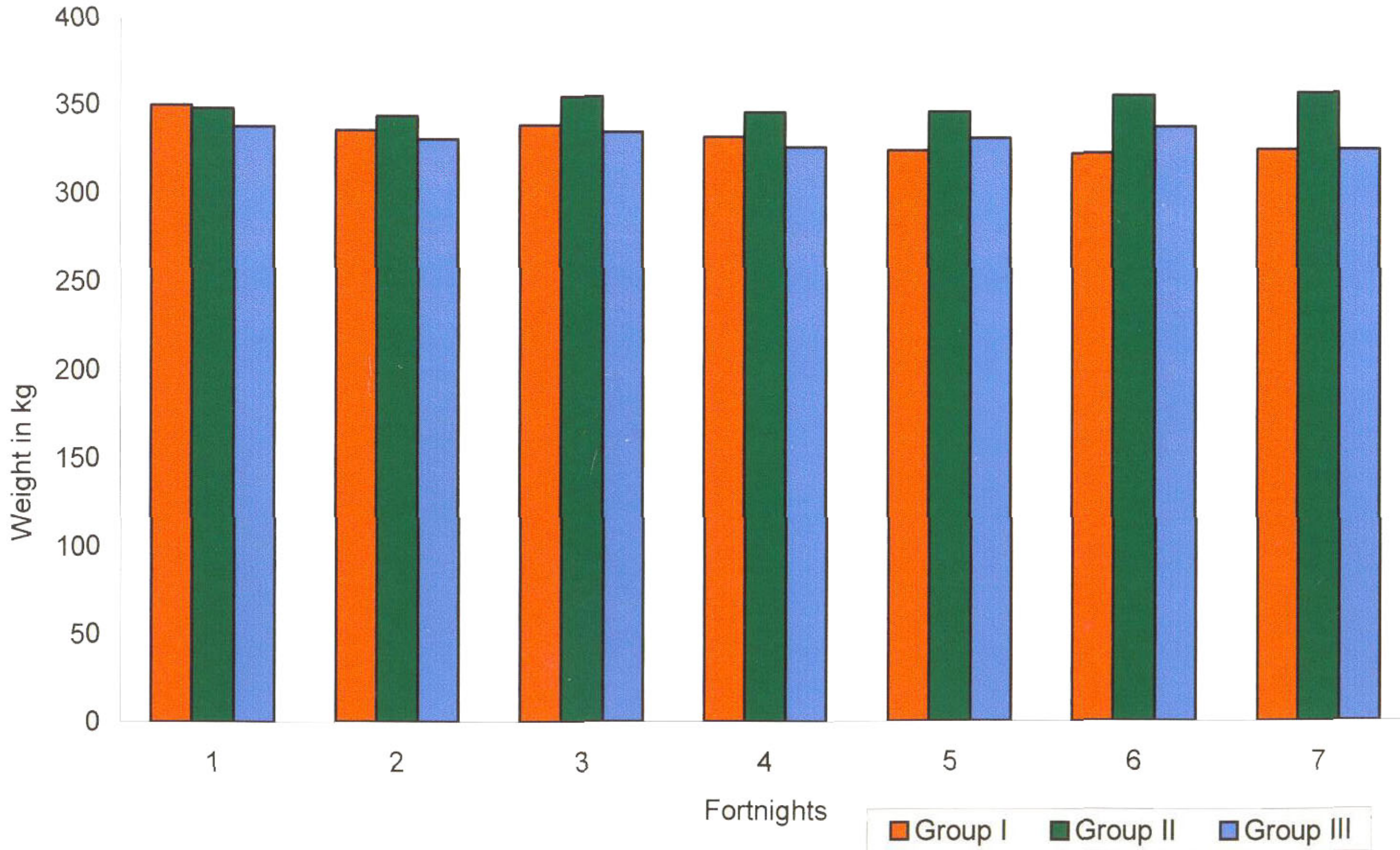

Fig. 1. Body weight of animals maintained on three experimental rations in phase I

Table 8. Average daily dry matter intake* of animals maintained on the three experimental rations in phase I.

Weeks	Dry matter intake (kg / animal)		
	Group I	Group II	Group III
1	9.67 ± 0.21 [A]	10.55 ± 0.28 [A]	12.5 ± 0.18 [B]
2	9.81 ± 0.18 [a]	11.23 ± 0.21 [b]	12.0 ± 0.12 [B]
3	9.62 ± 0.22 [A]	11.86 ± 0.22 [B]	11.36 ± 0.12 [B]
4	10.99 ± 0.42	13.23 ± 0.32	12.30 ± 0.14
5	11.07 ± 0.60	13.35 ± 0.02	12.73 ± 0.11
6	9.86 ± 0.8 [Aa]	13.45 ± 0.25 [B b]	12.74 ± 0.13 [b]
7	8.76 ± 0.24 [A]	13.48 ± 0.19 [B]	12.56 ± 0.21 [B]
8	10.35 ± 0.42 [A a]	13.6 ± 0.18 [B b]	12.46 ± 0.10 [b]
9	10.26 ± 0.43 [A a]	13.56 ± 0.16 [B b]	12.5 ± 0.11 [b]
10	10.08 ± 0.31 [A a]	13.5 ± 0.22 [B b]	12.65 ± 0.18 [b]
11	9.96 ± 0.35 [Aa]	13.3 ± 0.19 [B b]	12.51 ± 0.11 [b]
12	8.76 ± 0.38 [A]	12.66 ± 0.23 [B]	12.53 ± 0.09 [B]
13	9.32 ± 0.38 [A]	12.73 ± 0.19 [B]	12.67 ± 0.11 [B]
14	9.39 ± 0.32 [A]	13.30 ± 0.22 [B]	12.65 ± 0.11 [B]
Mean ± SE	9.85 ± 0.64 [A]	12.84 ± 0.79 [B]	12.44 ± 0.66 [B]

* Average of six values.

Means with different superscripts in the same row differ significantly

A, B-($P<0.01$) and a ,b-($P<0.05$)

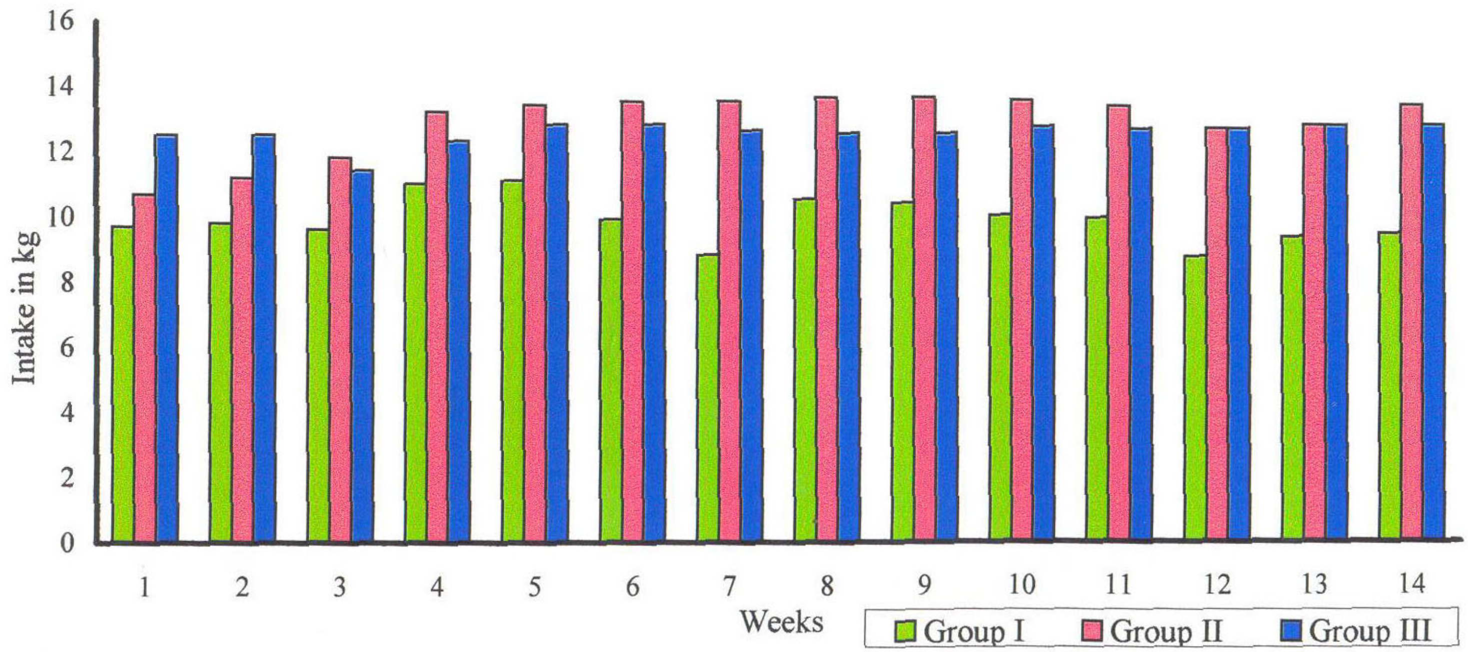

Fig. 2. Average daily dry matter intake of animals maintained on the three experimental rations in phase I

Table 9. Average daily milk production *of the animals maintained on the three experimental rations in phase I.

Weeks	Milk production (kg/ animal)		
	Group I	Group II	Group III
1	10.58 ± 0.96	9.81 ± 0.82	9.01 ± 1.12
2	11.02 ± 1.3	10.38 ± 0.41	9.61 ± 1.4
3	10.31 ± 1.03	10.51 ± 0.38	9.48 ±1.24
4	10.38 ± 0.96	10.98 ± 0.28	10.22 ± 1.2
5	10.30 ± 0.79	10.77 ± 0.61	10.21 ± 0.54
6	10.04 ± 1.01	10.52 ± 0.76	10.31 ± 0.52
7	8.87 ± 0.79	10.45 ± 0.67	10.04 ± 0.66
8	8.54 ± 0.89	10.30 ± 0.59	9.59 ± 0.57
9	8.26 ± 0.97	9.83 ± 0.96	9.16 ± 0.51
10	7.76 ± 0.82	9.41 ± 0.97	9.64 ± 0.54
11	7.12 ± 0.74	9.03 ± 1.01	9.34 ± 0.56
12	6.95 ± 0.93	9.12 ± 0.96	8.24 ± 0.34
13	6.45 ± 0.82	8.71 ±1.02	7.83 ± 0.39
14	6.70 ± 0.79	8.40 ± 1.06	7.71 ± 0.49
Mean± SE	8.81 ± 0.40	9.87 ± 0.22	9.31 ± 0.22

* Average of six values

Table 10. Analysis of covariance of milk produced by the animals maintained on the three experimental rations in phase I.

Weeks	Mean sum squares			P value
	Groups	Covariate	Error	
1	6.841	63.382	3.592	0.26
2	6.137	51.806	2.228	0.32
3	5.479	45.680	2.022	---
4	2.263	12.877	0.725	---
5	1.278	11.710	1.962	---
6	0.627	11.663	3.200	0.33
7	5.115	8.375	2.642	0.26
8	7.094	10.584	2.410	---
9	5.172	12.196	3.713	0.26
10	6.967	7.626	3.568	0.16
11	9.487	6.122	3.586	0.22
12	9.455	9.103	3.451	0.2
13	9.776	7.966	3.416	---
14	5.458	6.860	3.742	---

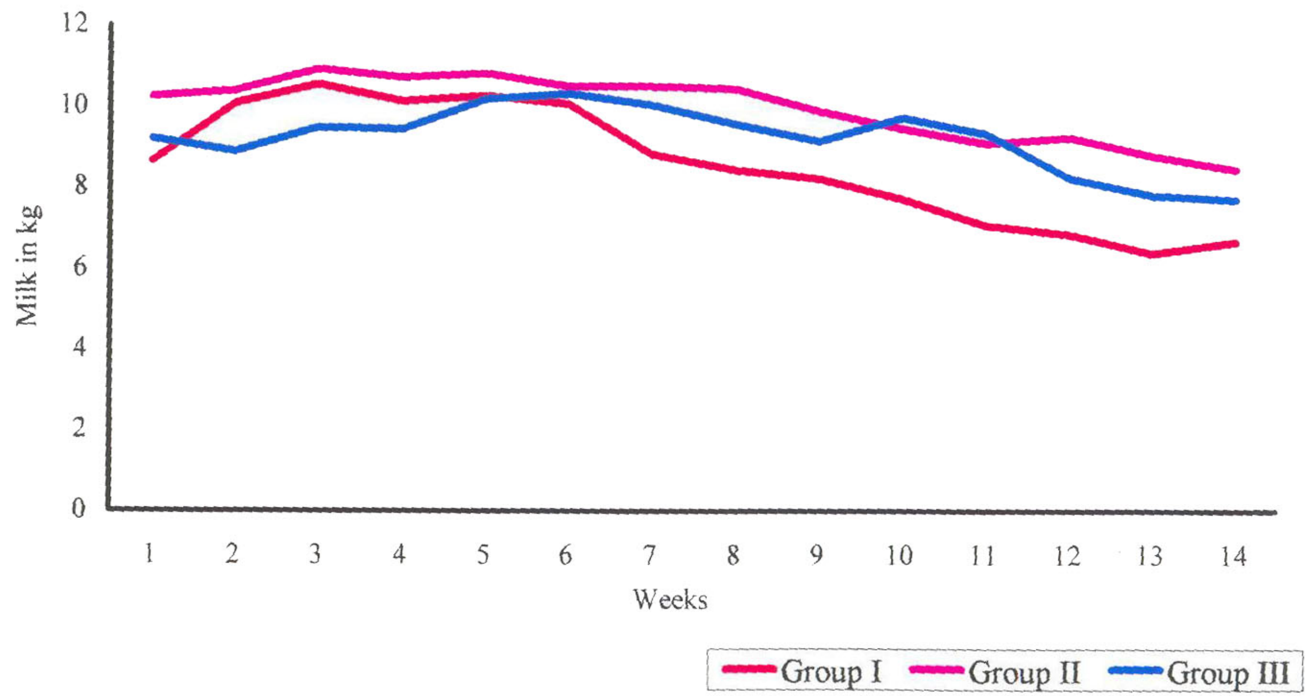

Fig. 3. Average daily milk production of animals maintained on three experimental rations in phase I

Table 11. Composition of milk*from animals maintained on the three experimental rations in phase I

Parameter	Group	Fortnights						Mean ± SE
		1	2	3	4	5	6	
Fat (g %)	I	3.62 ± 0.95	3.44 ± 0.13	3.38 ±.0.18	3.23 ± 0.19	3.78 [a] ± 0.16	3.43 ± 0.15	3.48 ± 0.08
	II	3.30 ± 0.28	3.37 ± 0.18	3.21 ± 0.04	3.80 ± 0.25	3.65 [a] ± 0.09	3.73 ± 0.16	3.51 ± 0.10
	III	3.40 ± 0.06	3.78 ± 0.28	3.50 ± 0.19	3.55 ± 0.20	3.33 [b] ± 0.10	3.33 ± 0.22	3.40 ± 0.07
Total solids (g %)	I	12.30 ± 0.93	11.50 ± 0.18	12.44 ± 0.51	11.90 ± 0.32	11.38 ± 0.44	11.31 ± 0.17	11.81 ± 0.20
	II	11.40 ± 0.40	11.21 ± 0.41	12.24 ± 0.25	12.50 ± 0.3	12.06 ± 0.26	11.80 ± 0.20	11.87 ± 0.20
	III	11.81 ± 0.30	12.31 ± 0.38	12.25 ± 0.34	11.71 ± 0.46	11.70 ± 0.39	12.30 ± 0.32	12.01 ± 0.12
SNF (g %)	I	8.68 ± 0.29	8.06 ± 0.06	9.06 ± 0.59	8.62 ± 0.23	7.60 ± 0.50	7.88 ± 0.21	8.33 ± 0.23
	II	8.10 ± 0.36	7.84 ± 0.36	9.03 ± 0.30	8.70 ± 0.21	8.41 ± 0.27	8.17 ± 0.2	8.36 ± 0.18
	III	8.41 ± 0.30	8.53 ± 0.31	8.75 ± 0.19	8.16 ± 0.41	8.37 ± 0.37	8.97 ± 0.44	8.53 ± 0.12
Protein (g %)	I	2.44 ± 0.14	2.91 ± 0.25	2.47 ± 0.27	2.43 ± 0.20	2.79 ± 0.21	2.47 ± 0.17	2.59 ± 0.09
	II	2.65 ± 0.30	2.30 ± 0.25	2.45 ± 0.18	2.21 ± 0.10	2.86 ± 0.18	2.74 ± 0.34	2.54 ± 0.10
	III	2.63 ± 0.14	2.58 ± 0.09	2.32 ± 0.18	2.50 ± 0.09	2.43 ± 0.17	2.65 ± 0.06	2.52 ± 0.05

*Average of six values.

a, b-Means within the same parameter with different superscript in the same column vary significantly ($P < 0.05$)

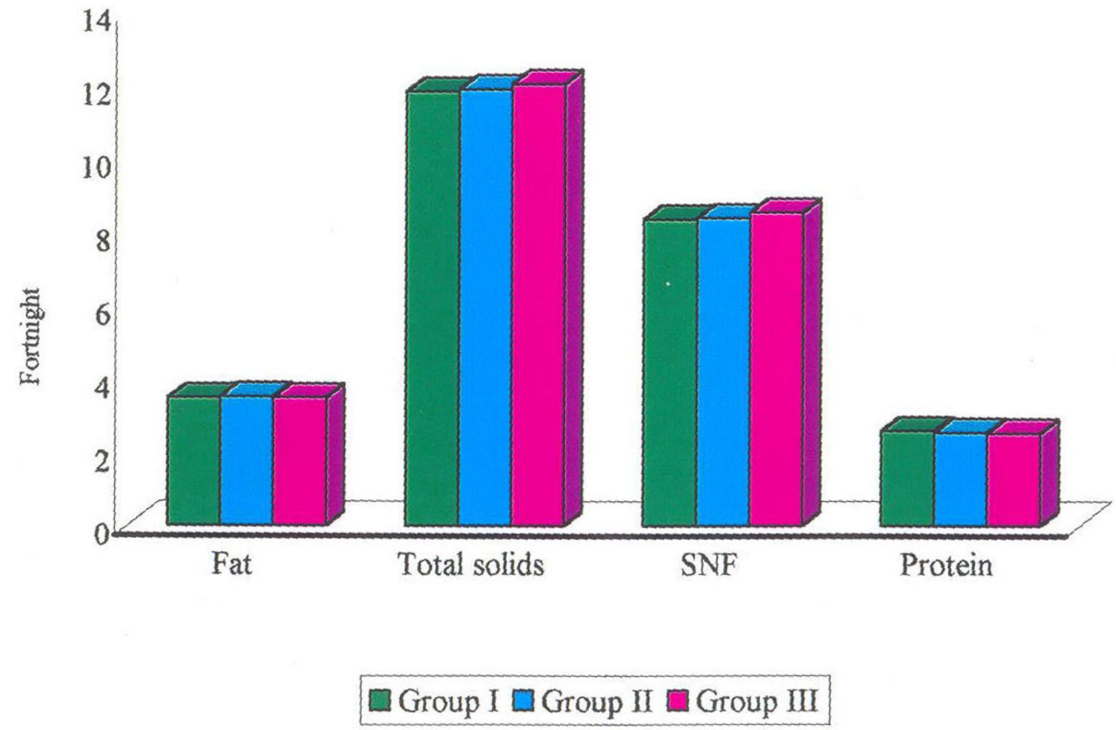

Fig. 4. Composition of milk from animals maintained on the three experimental rations in phase I

Table 12. Rumen fermentation parameters* of animals maintained on the three experimental rations in phase I

Groups	pH	Total volatile fatty acids (meq/l)	Ammonia nitrogen (mg/100ml)	Lactic acid (mg/ml)
I	7.71 ± 0.20^{a}	93.81 ± 11.6	39.0 ± 0.80^{a}	0.33 ± 0.02^{a}
II	7.01 ± 0.11^{b}	68.7! ±11.52	31.0 ± 0.21^{a}	0.21 ± 0.01^{b}
III	7.37 ± 0.21^{b}	86.50 ± 11.98	17.0 ± 0.22^{b}	0.22 ± 0.01^{b}

*Average of six values.

a,b.- Means in the same column bearing different superscripts vary significantly ($P< 0.05$)

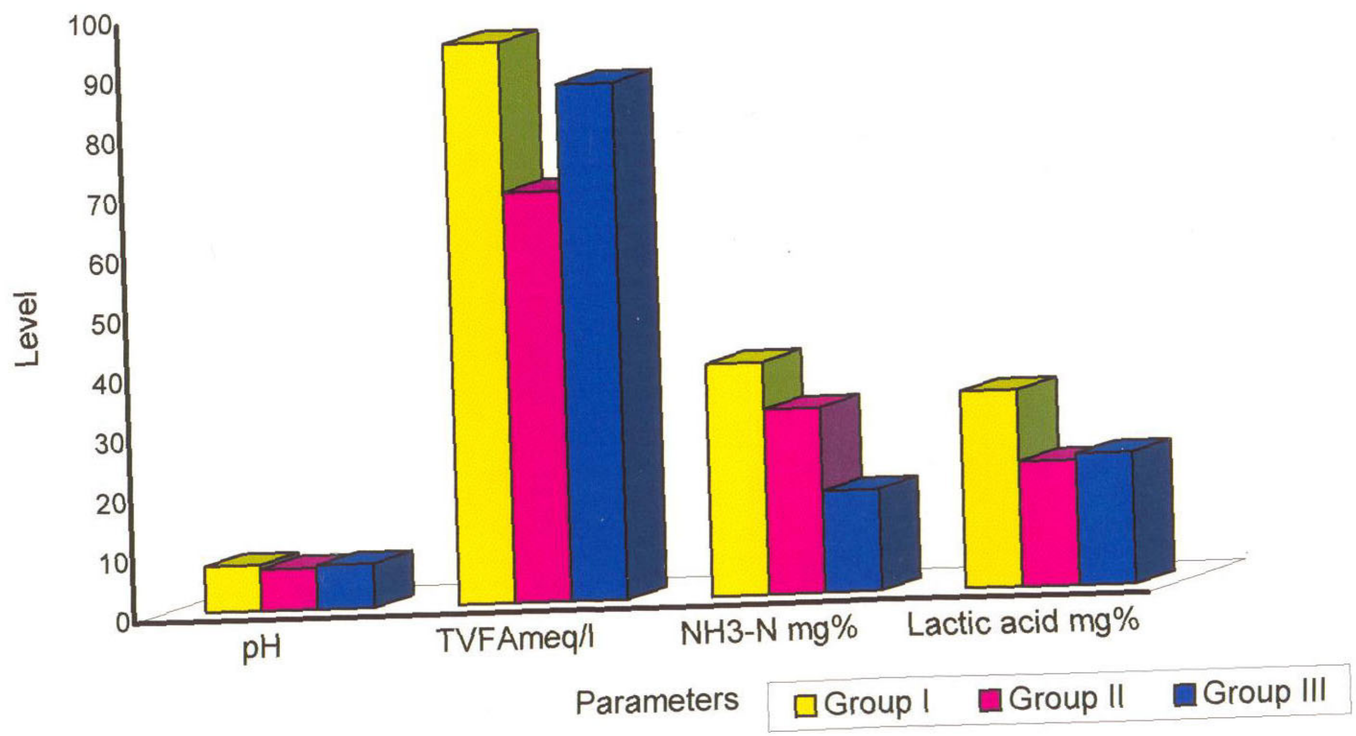

Fig. 5. Rumen fermentation parameters of animals maintained on three experimental rations in phase I

Table 13. Average haematological parameters* of the animals maintained on the three experimental rations in phase I

Group	Haemoglobin (g/100ml)		Blood glucose (mg/100ml)		Blood urea nitrogen (mg/100ml)		Plasma calcium (mg/100ml)		Plasma phosphorus (mg/100ml)	
	50th day	98th day	50th day	98th day	50th day	98th day	50th day	98th day	50th day	98th day
I	9.86 ± 1.3	10.31 ± 0.61	60.21 ± 2.59	51.71 ± 4.3	17.10 ± 2.02 AB	17.00 ± 1.01	9.50 ± 0.85	8.83 ± 0.60	7.12 ± 0.68	6.35 ± 0.72
II	10.4 3± 0 .65	10.70 ± 0.60	57.71 ± 7.13	53.70 ± 4.73	21.5 ± 1.65 A	18.00 ± 1.89	8.33 ± 0.88	9.17 ± 0.30	7.52 ± 0.56	8.13 ± 0.76
III	9.03 ± 0.41	8.35 ± 0.28	59.22 ± 3.74	62.21 ± 2.53	13.6 ± 1.31 B	18.01 ± 1.89	9.51 ± 0.72	9.67 ± 0.49	7.95 ± 0.99	6.58 ± 0.21

*Average of six values.
A, B- Means with different superscript in the same column for each parameter differ significantly (P<0.01).

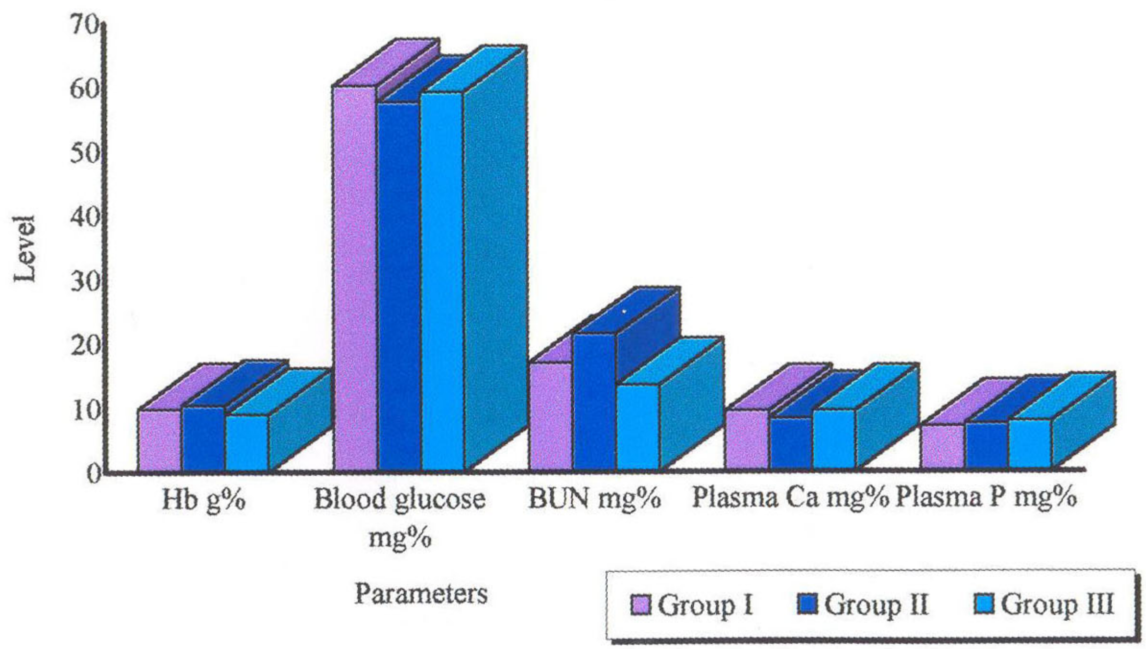

Fig. 6. Average haematological parameters of animals maintained on three experimental rations, collected on 50th day in phase I

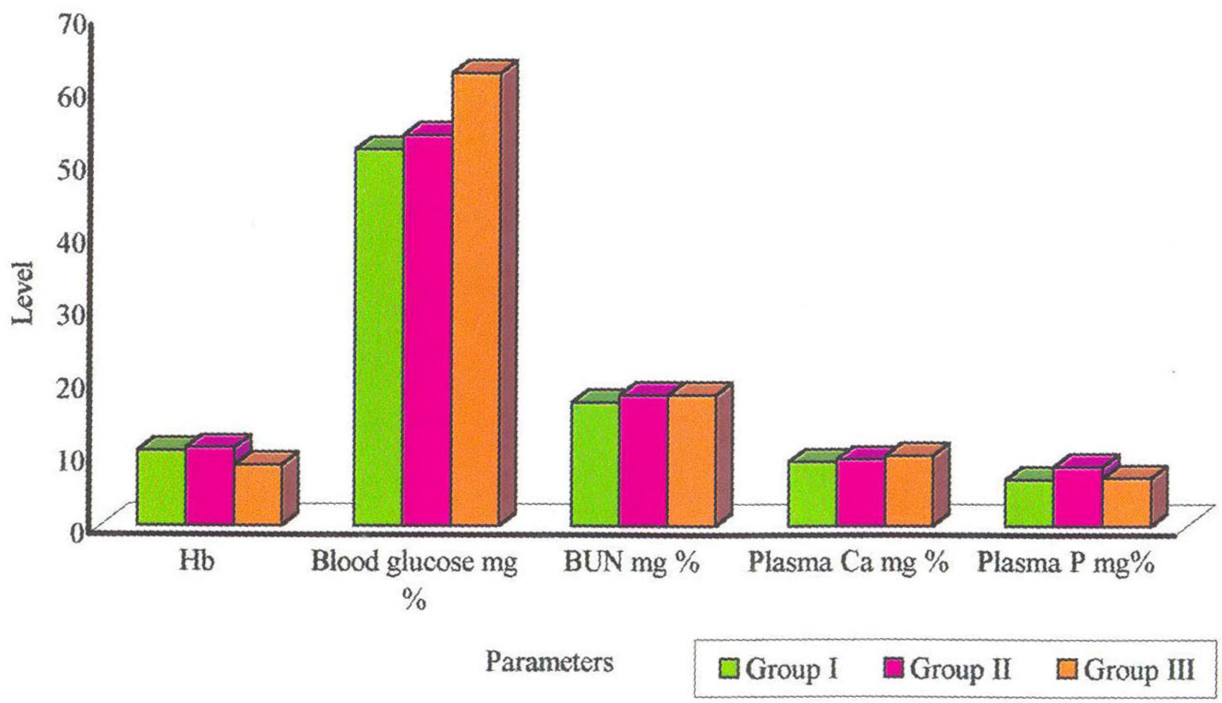

Fig. 7. Average haematological parameters of animals maintained on three experimental rations, collected on 98[th] day in phase I

Table 14. Per cent chemical composition* of the dung from experimental animals of phase I (on dry matter basis)

Nutrient	Group I	Group II	Group III
Dry matter	18.27 ± 0.52	20.11 ± 0.55	21.40 ± 0.54
Crude protein	14.23 ± 1.22	12.80 ± 0.96	11.88 ± 1.00
Ether extract	6.53 ± 0.74	6.93 ± 0.51	7.15 ± 0.25
Crude fibre	17.21 ± 1.51	19.27 ± 1.05	19.88 ± 0.96
Total ash	19.53 ± 0.51	18.28 ± 0.30	18.17 ± 0.29
Nitrogen free extract	42.50 ± 2.64	42.72 ± 4.06	42.92 ± 1.14
Acid insoluble ash	13.88 ± 0.73	12.20 ± 0.62	11.85 ± 0.37
Neutral detergent fibre	58.28 ± 0.66	59.62 ± 0.52	59.12 ± 2.18
Acid detergent fibre	40.96 ± 1.89	39.94 ± 0.51	42.01 ± 0.83

* Average of six values

Table 15. Digestibility coefficient* of nutrients of experimental rations in phase I (on dry matter basis) .

Nutrient	Group I	Group II	Group III
Dry matter	61.23 ± 3.69	62.15 ± 1.93	57.28 ± 1.44
Crude protein	69.23 ± 2.38	70.28 ± 2.34	62.10 ± 2.86
Ether extract	48.20 ± 1.08 ^{A}	42.93 ± 2.27 ^{B}	39.58 ± 1.70 ^{B}
Crude fibre	61.38 ± 4.10 ^{A}	45.80 ± 4.52 ^{B}	41.83 ± 3.81 ^{B}
Nitrogen free extract	68.32 ± 0.32	71.08 ± 2.38	68.04 ± 1.59
Neutral detergent fibre	46.34 ± 4.56 ^{A}	24.84 ± 1.32 ^{B}	26.81 ± 1.60 ^{B}
Acid detergent fibre	30.31 ± 4.04 ^{A}	18.95 ± 1.34 ^{B}	39.26 ± 3.15 ^{A}

* Average of six values

A. B-Means with different superscript in the same row vary significantly ($P < 0.01$)

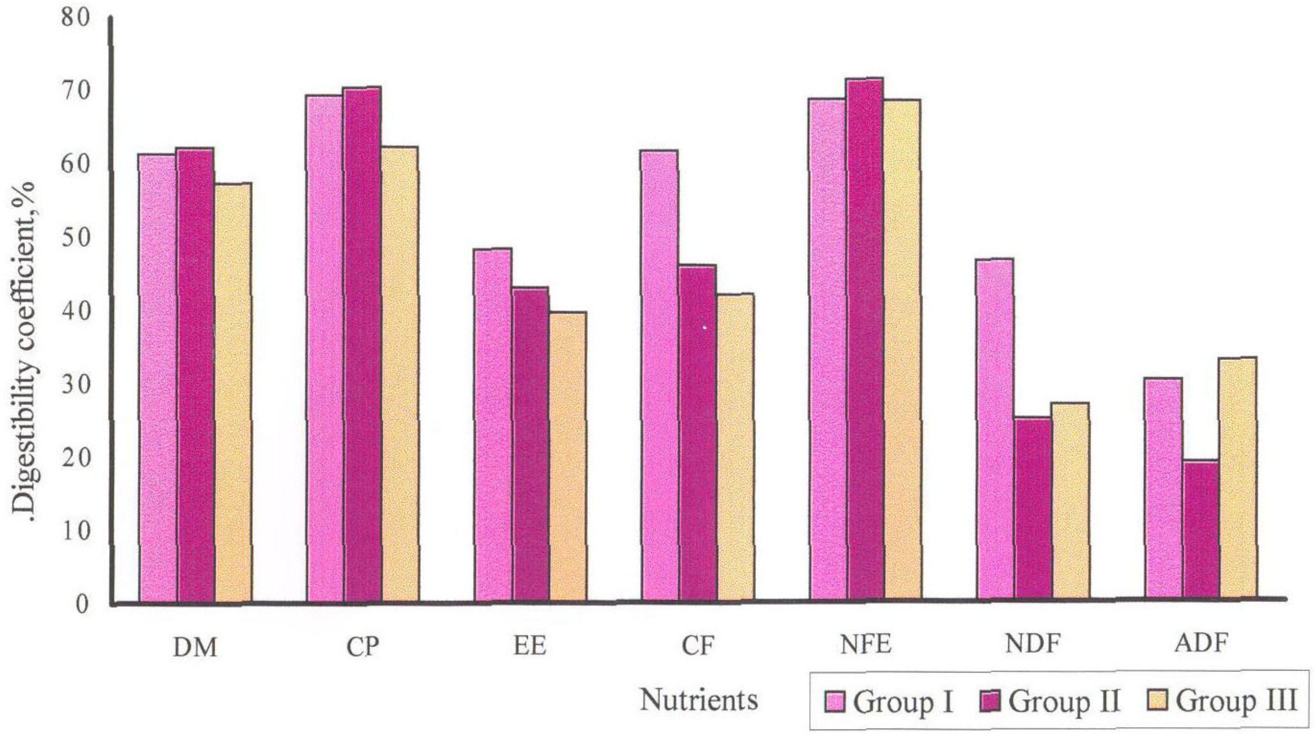

Fig. 8. Digestibility coefficient of nutrients of the three experimental rations in phase I

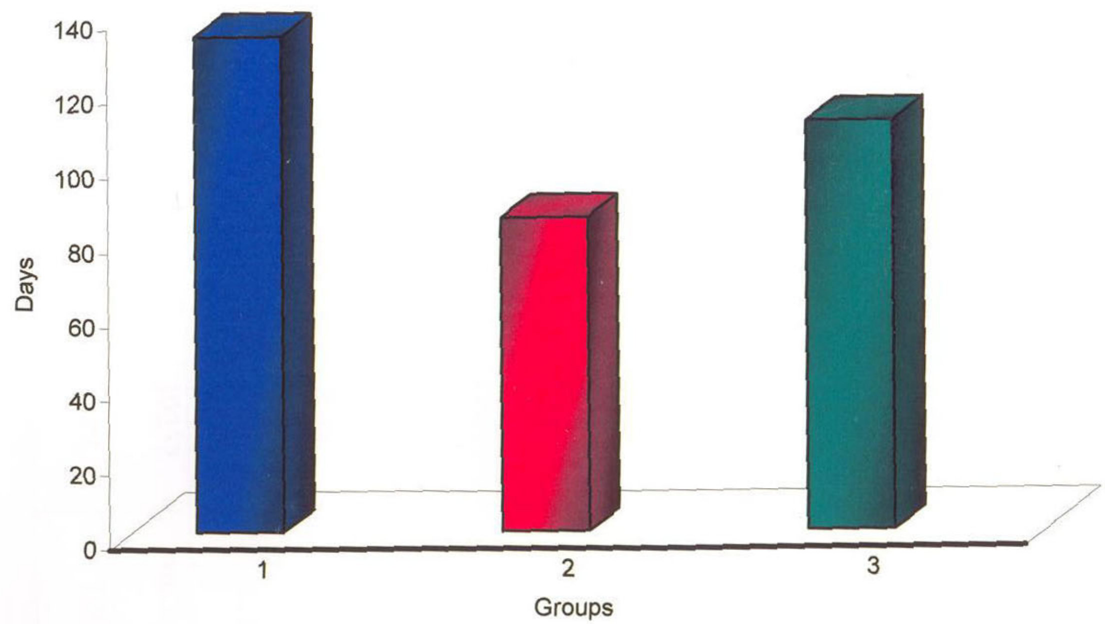

Fig. 9. Average number of days to first service in animals maintained on three experimental rations in phase I

Table 16. Total dry matter intake and cost per kg milk production of animals maintained on the three experimental rations in phase I

Parameter	Group I	Group II	Group III
Total concentrate consumed in 100 days (kg)	3690 ^{B}	5208 ^{A}	5132 ^{A}
Total straw consumed in 100 days (kg)	2104	2348	2226
Total feed consumed in 100 days (kg)	5794 ^{B}	7556 ^{A}	7358 ^{A}
Dry matter intake per 100 kg body weight (kg)	2.99 ^{B}	3.54 ^{A}	3.79 ^{A}
Dry matter intake per kg metabolic live weight (W kg $^{0.75}$)	12.67	15.37	16.08
Total milk produced in 100 days (kg)	5041	5855	5423
Feed intake per kg milk produced (Kg)	1.15	1.29	1.36
Cost of one kg concentrate * (Rs.)	9.44	7.95	7.24
Cost of one kg straw* (Rs.)	2.10	2.10	2.10
Total cost of feed (Rs.)	39252	46334.4	41830.28
Cost / kg milk produced	7.79	7.91	7.71

*Calculated using the rate contract values fixed for feed ingredients by College of Veterinary and Animal Sciences, Mannuthy.
A, B-Means with different superscripts in the same row differ significantly ($P<0.01$)

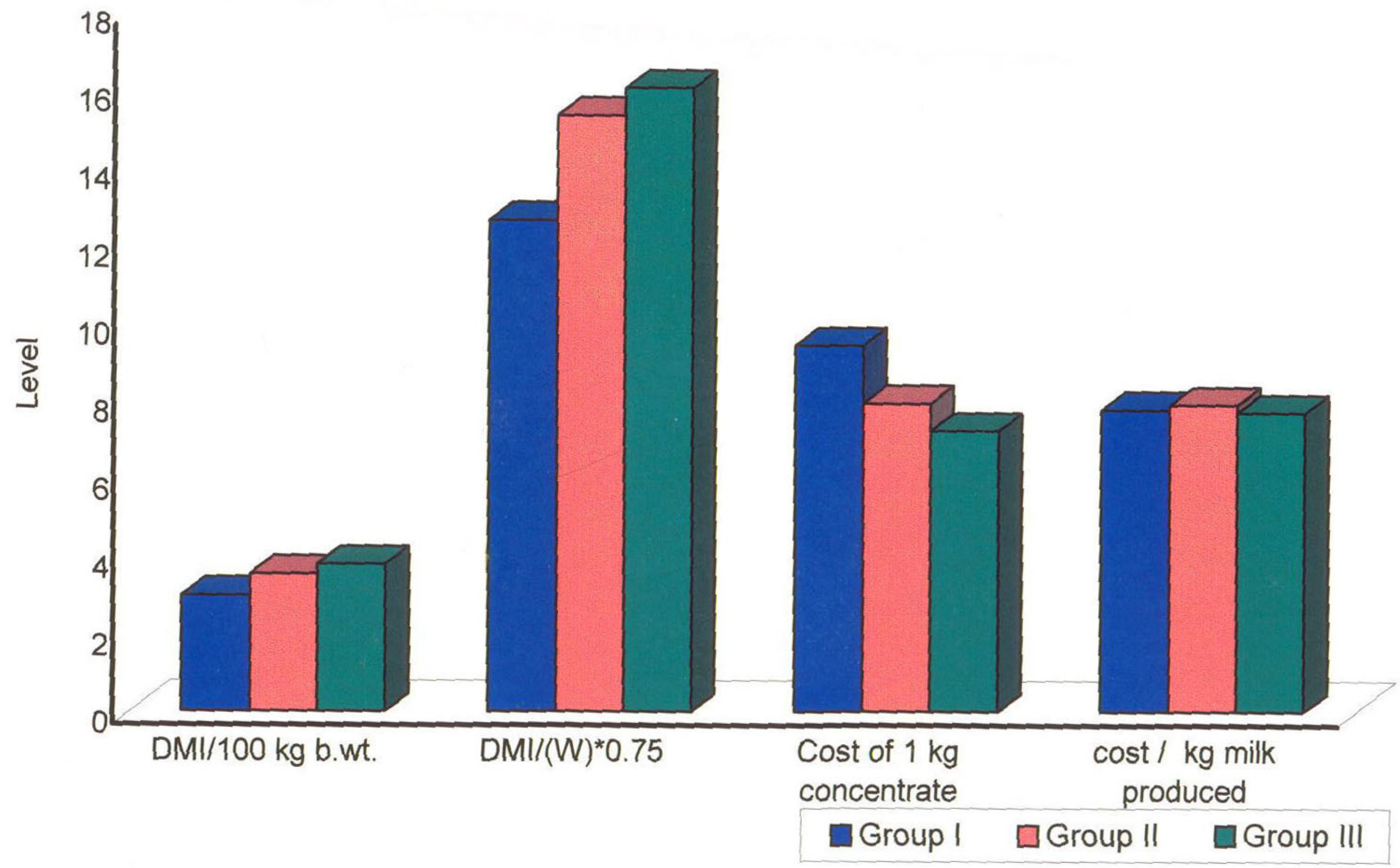

Fig. 10. Dry matter intake and cost of production of milk in animals maintained on three experimental rations in phase I

Table 17. Per cent chemical composition* of the two experimental concentrate mixtures and paddy straw of phase II (on dry matter basis)

Nutrient	Concentrate mixtures		Paddy straw
	Group I	Group II	
Dry matter	92.89 ± 0.24	93.25 ± 0.18	87.78 ± 0.11
Crude protein	17.24 ± 0.52	17.18 ± 0.18	4.22 ± 0.22
Ether extract	5.28 ± 0.32	5.55 ± 0.14	2.80 ± 0.19
Crude fibre	8.78 ± 0.48	8.24 ± 0.22	34.96 ± 0.16
Total ash	7.68 ± 0.28	8.18 ± 0.34	10.76 ± 0.11
Nitrogen free extract	61.02 ± 0.41	60.85 ± 0.18	47.26 ± 0.28
Acid insoluble ash	1.98 ± 0.28	3.08 ± 0.14	2.78 ± 0.08
Neutral detergent fibre	24.18 ± 0.09	23.08 ± 0.12	74.89 ± 0.12
Acid detergent fibre	15.28 ± 0.12	15.98 ± 0.18	54.23 ± 0.08
Acid detergent lignin	6.07 ± 0.02	6.28 ± 0.18	4.76 ± 0.04
Calcium	0.84 ± 0.06	0.88 ± 0.02	0.29 ± 0.02
Phosphorus	0.49 ± 0.14	0.58 ± 0.09	0.09 ± 0.07

* Average of six values

Table 18. Body weight*of the animals maintained on the two experimental rations in phase II.

Fortnights	Body weight (kg)	
	Group I	Group II
0	323.57 ± 15.11	309.43 ± 23.58
1	323.57 ± 15.11	313.29 ± 10.24
2	330.43 ± 15.68	315.00 ± 11.41
3	331.01 ± 19.16	314.43 ± 11.22
4	327.00 ± 19.53	312.31 ± 10.06
5	316.86 ± 16.49	312.30 ± 10.52
6	324.00 ± 15.66	319.29 ± 11.48
7	318.18 ±10.52	313.25 ± 11.28

* Average of six values

Table 19. Analysis of covariance of the body weight of the animals maintained on the two experimental rations in phase II.

Fortnights	Group	Covariate	Error	P value
1	0.051	12474.110	66.276	--
2	78.631	13621.128	117.186	---
3	84.445	16217.958	302.160	---
4	48.960	14400.908	429.684	---
5	102.309	12211.001	268.480	---
6	44.320	15891.652	70.360	---
7	99.429	12811.001	248.320	---

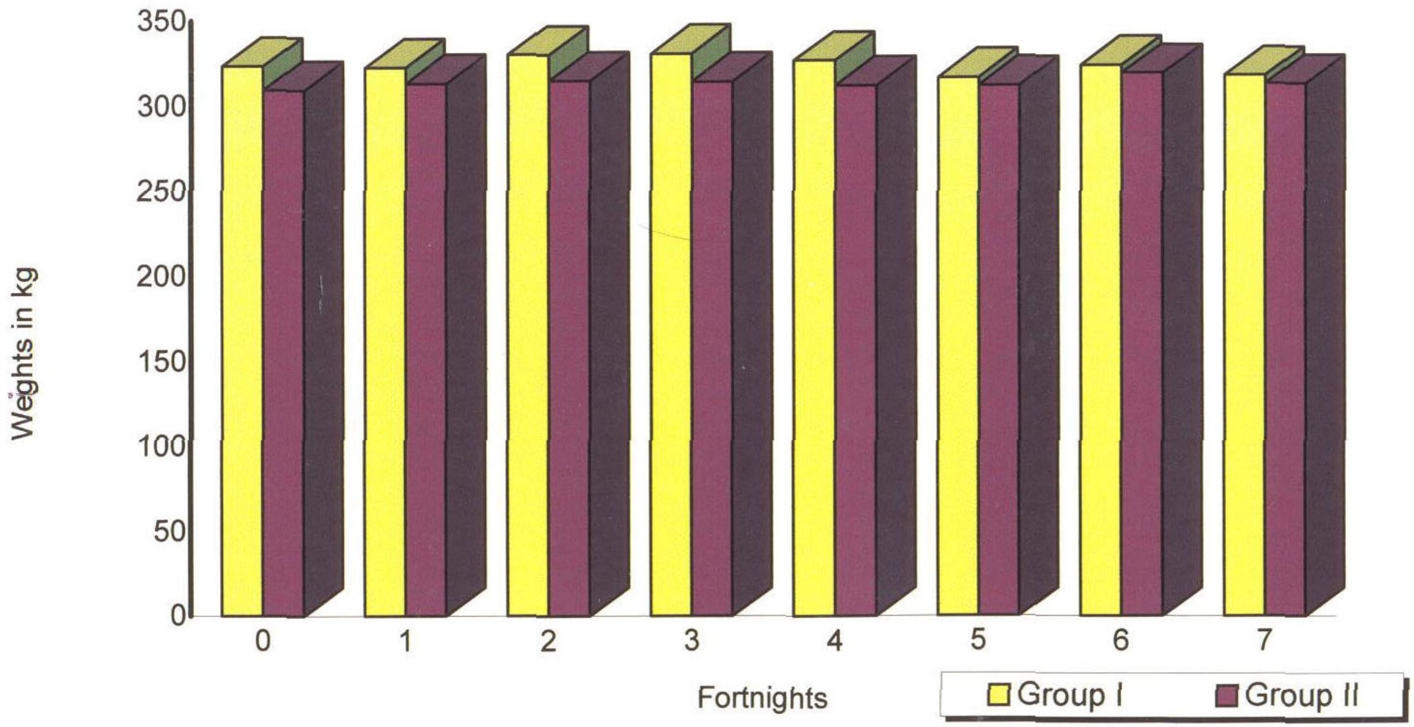

Fig. 11. Body weight of animals maintained on two experimental rations in phase II

Table 20. Average daily dry matter intake and milk production* of animals maintained on the two experimental rations in phase II

Weeks	Dry matter intake (kg/animal)		Milk production (kg/animal)	
	Group I	Group II	Group I	Group II
1	9.92 ± 0.15	9.55 ± 0.42	9.57 ± 0.48	9.31 ± 0.68
2	11.33 ± 0.51	10.54 ± 0.43	10.01 ± 0.71 ^{A}	8.86 ± 0.73 ^{B}
3	11.43 ± 0.48 ^{A}	9.56 ± 0.3 ^{B}	9.12 ± 0.46	8.84 ± 0.32
4	11.44 ± 0.48 ^{A}	9.52 ± 0.22 ^{B}	8.81 ± 0.46	8.81 ± 0.31
5	11.43 ± 0.48 ^{A}	9.74 ± 0.17 ^{B}	9.08 ± 0.62	8.51 ± 0.26
6	11.36 ± 0.52 ^{A}	9.52 ± 0.25 ^{B}	8.85 ± 0.51	8.06 ± 0.28
7	11.42 ±0.48 ^{A}	9.73 ± 0.13 ^{B}	9.13 ± 0.51 ^{a}	7.84 ± 0.19 ^{b}
8	11.46 ± 0.47 ^{A}	9.24 ± 0.23 ^{B}	9.02 ± 0.39 ^{a}	7.83 ± 0.19 ^{b}
9	11.38 ± 0.51 ^{A}	9.69 ± 0.24 ^{B}	9.12 ± 0.31 ^{a}	7.99 ± 0.28 ^{b}
10	11.39 ± 0.49 ^{A}	9.67 ± 0.29 ^{B}	9.10 ± 0.39	8.18 ± 0.35
11	11.36 ± 0.5 ^{A}	9.64 ± 0.26 ^{B}	9.08 ± 0.37 ^{a}	8.00 ± 0.34 ^{b}
12	11.38 ± 0.23 ^{A}	8.96 ± 0.37 ^{B}	8.92 ± 0.29 ^{a}	7.90 ± 0.39 ^{b}
13	11.05 ± 0.27 ^{A}	8.57 ± 0.65 ^{B}	8.86 ± 0.26 ^{a}	7.69 ± 0.39
14	11.47 ± 0.07 ^{A}	8.96 ± 0.12 ^{B}	8.51 ± 0.27 ^{A}	7.45 ± 0.33 ^{B}
Mean ±SE	11.27 ± 0.11	9.47 ± 0.12	9.08 ± 0.12	8.23 ± 0.12

* Average of six value

Means with different superscripts in the same row for each parameter vary Significantly- A, B- ($P<0.01$), a, b- ($P<0.05$).

Table 21. Analysis of covariance of the milk produced by animals maintained on the two experimental rations in phase II

Weeks	Group	Covariate	Error	P Value
1	0.143	13.400	1.341	-
2	2.900	6.828	0.292	0.01
3	0.250	5.402	0.284	-
4	0.000	11.547	6.255	-
5	0.725	5.219	0.431	0.22
6	1.356	5.681	0.545	0.14
7	3.664	1.799	0.510	0.025
8	3.079	1.757	0.438	0.026
9	2.734	1.647	0.614	0.06
10	1.809	2.430	0.632	0.124
11	2.532	2.974	0.528	0.056
12	2.240	3.299	0.496	0.06
13	2.970	3.492	0.366	0.02
14	2.422	4.038	0.239	0.01

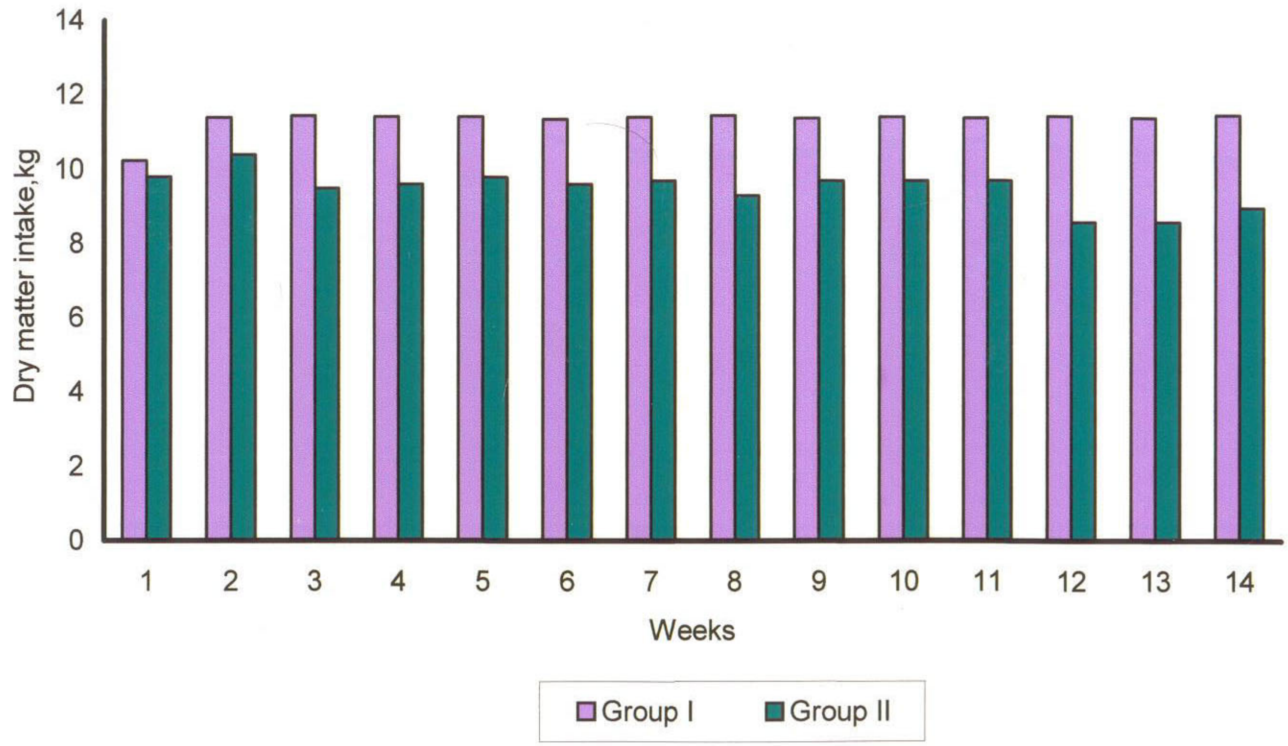

Fig. 12. Average daily dry matter intake in animals maintained on two experimental rations in phase II

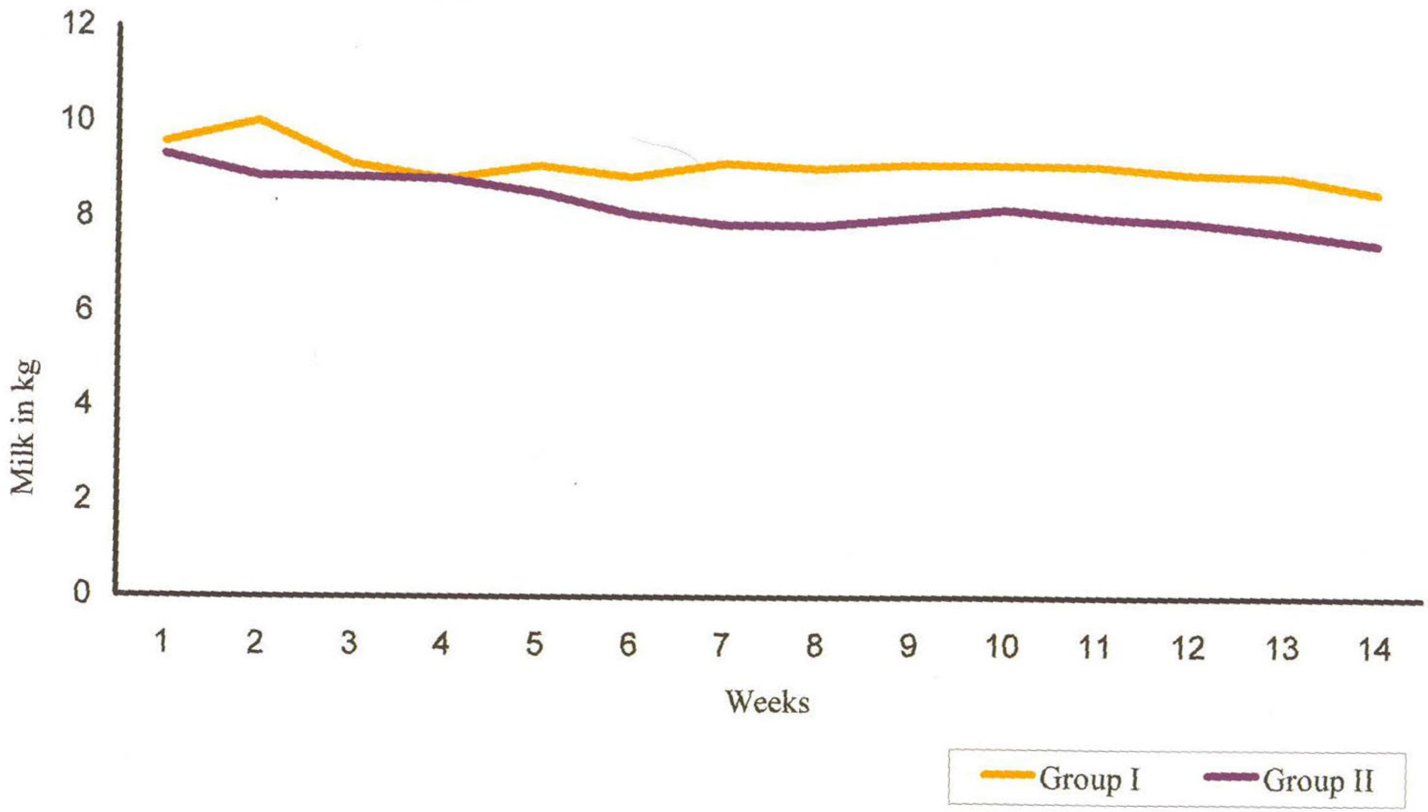

Fig. 13. Average daily milk production by animals maintained on two experimental rations in phase II

Table 22. Composition of milk* from the animals maintained on the two experimental rations in phase II .

Fortnight	Fat (g %)		Total solids (g%)		SNF (g%)		Protein (g%)		Urea nitrogen (mg %)	
	Group		Group		Group		Group		Group	
	I	II	I	II	I	II	I	II	I	II
1	3.36 ± 0.15	3.08 ± 0.03	12.24 ± 0.21	11.71 ± 0.28	8.88 ± 0.29	8.62 ± 0.29	2.86 ± 0.21	3.01 ± 0.15	34.43 ± 2.02	38.07 ± 1.93
2	3.49 ± 0.17	3.16 ± 0.08	11.56 ± 0.47	10.77 ± 0.46	8.08 ± 0.39	7.69 ± 0.39	2.76 ± 0.11	2.63 ± 0.09	54.11 ± 1.70	53.30 ± 2.95
3	3.67 ± 0.14	3.34 ± 0.13	12.11 ± 0.47	10.51 ± 0.51	8.44 ± 0.45	7.19 ± 0.46	2.86 ± 0.11	2.87 ± 0.21	50.57 ± 0.11	52.46 ± 1.52
4	3.86 ± 0.23 [a]	3.03 ± 0.22 [b]	12.97 ± 0.26 [A]	11.37 ± 0.41 [B]	8.97 ± 0.40	8.51 ± 0.44	2.96 ± 0.11	2.71 ± 0.09	51.26 ± 0.10	54.09 ± 1.89
5	3.40 ± 0.14	3.06 ± 0.24	12.42 ± 0.26	11.83 ± 0.37	9.02 ± 0.19	8.71 ± 0.15	3.05 ± 0.21	3.08 ± 0.11	52.46 ± 3.37	56.06 ± 2.04
6	3.83 ± 0.18	3.41 ± 0.21	12.53 ± 0.19	11.68 ± 0.35	8.96 ± 0.25	8.26 ± 0.34	2.79 ± 0.32	2.63 ± 0.09	43.20 ± 2.80	41.78 ± 2.54
Mean ± SE	3.60 ± 0.09 [A]	3.18 ± 0.06 [B]	12.31 ± 0.19 [A]	11.31 ± 0.22 [B]	8.70 ± 0.16 [A]	8.13 ± 0.16 [B]	2.88 ± 0.04	2.82 ± 0.08	47.67 ± 3.06	49.29 ± 3.04

* Average of six values

Means with different superscripts in the same row for each parameter vary significantly- A,B- (P< 0.01), a,b- (P< 0.05)

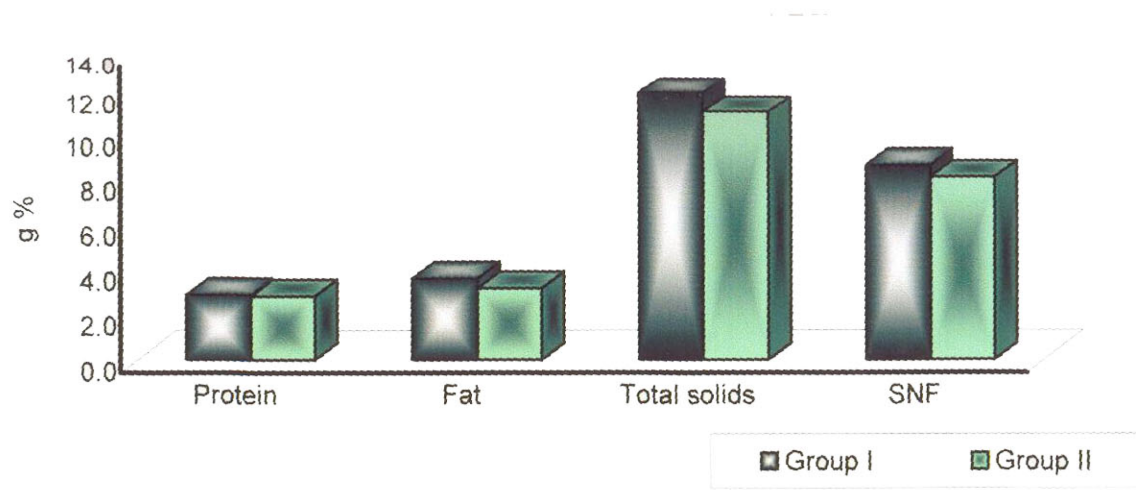

Fig. 14 (1). Composition of milk from animals maintained on two experimental rations in phase II

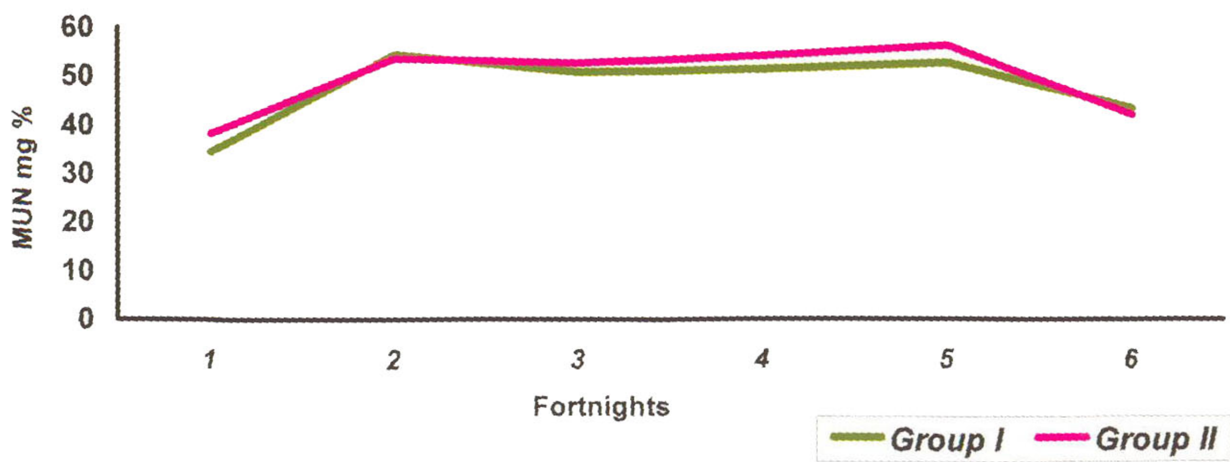

Fig. 14 (2). Milk urea nitrogen levels in animals maintained on two experimental rations in phase II

Table 23. Rumen fermentation parameters* of the animals maintained on the two experimental rations in phase II.

Parameter	Group1	Group II
pH	6.42 ± 0.14	6.57 ± 0.19
Total volatile fatty acids (meq/l)	76.10 ± 4.21	77.60 ± 5.29
Rumen NH_3-N (mg/100 ml)	36.42 ± 0.53 [a]	27.27 ± 0.39 [b]
Lactic Acid. (mg/ml)	0.21 ± 0 04	0.10 ± 0.02

* Average of six values

a,b- Means with different superscripts in the same row vary significantly ($P < 0.05$)

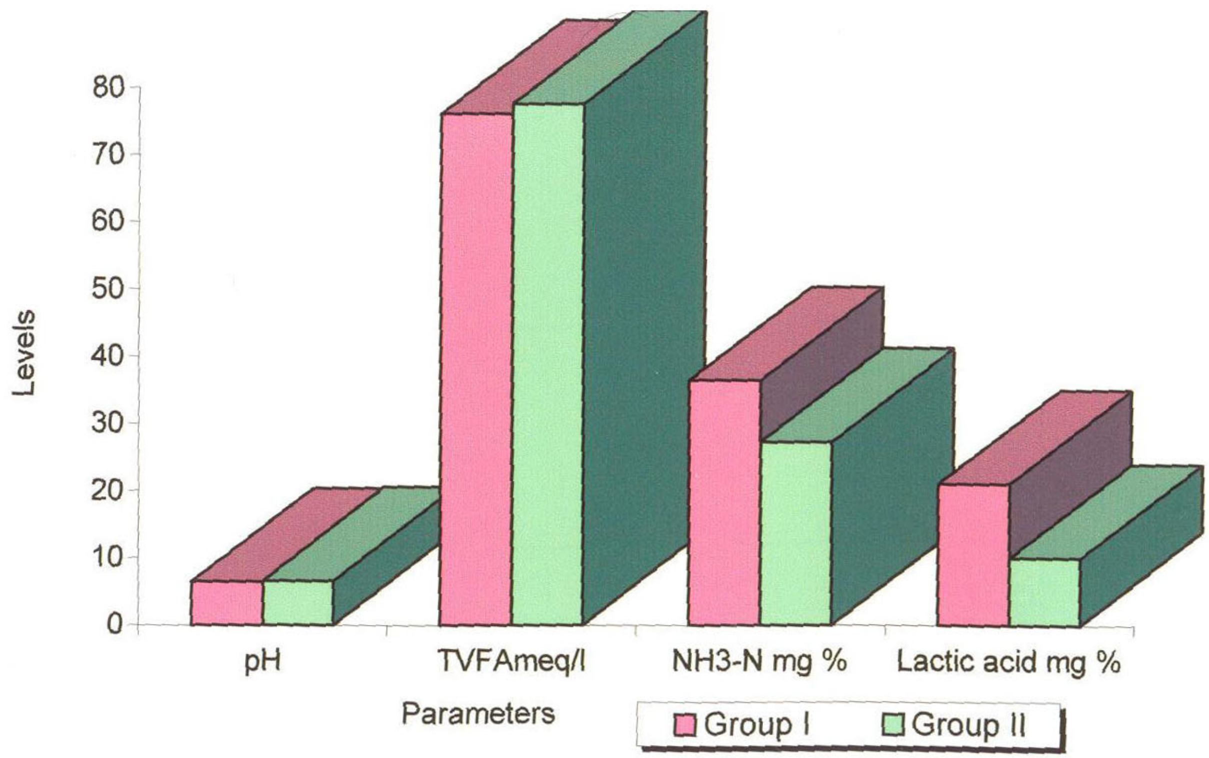

Fig. 15. Rumen fermentation parameters of animals maintained on two experimental rations in phase II

Table 24. Haematological parameters* of the animals maintained on the two experimental rations in phase II

Parameters	Days			
	50		98	
	Group I	Group I I	Group I	Group I I
Haemoglobin (g%)	10.27 ± 0.16	10.58 ± 0.54	9.69 ± 0.41	10.00 ± 0.73
Blood glucose (mg %)	43.57 ± 1.43 ^{A}	53.57 ± 3.46 ^{B}	62.14 ± 0.65	57.14 ± 2.14
Blood Urea Nitrogen (mg%)	44.40 ± 4.04	39.61 ± 3.03	43.01 ± 5.05	35.30 ± 2.03
Plasma calcium (mg %)	10.86 ± 0.51	9.8 ± 0.16	11.00 ± 0.44	11.0 ± 0.92
Plasma Phosphorus (mg%)	6.50 ± 0.31	6.24 ± 0.4	7.74 ± 0.29	6.7 ± 0.73

*Average of six values

A, B- Means with different superscripts in the same row differ significantly (P<0.01)

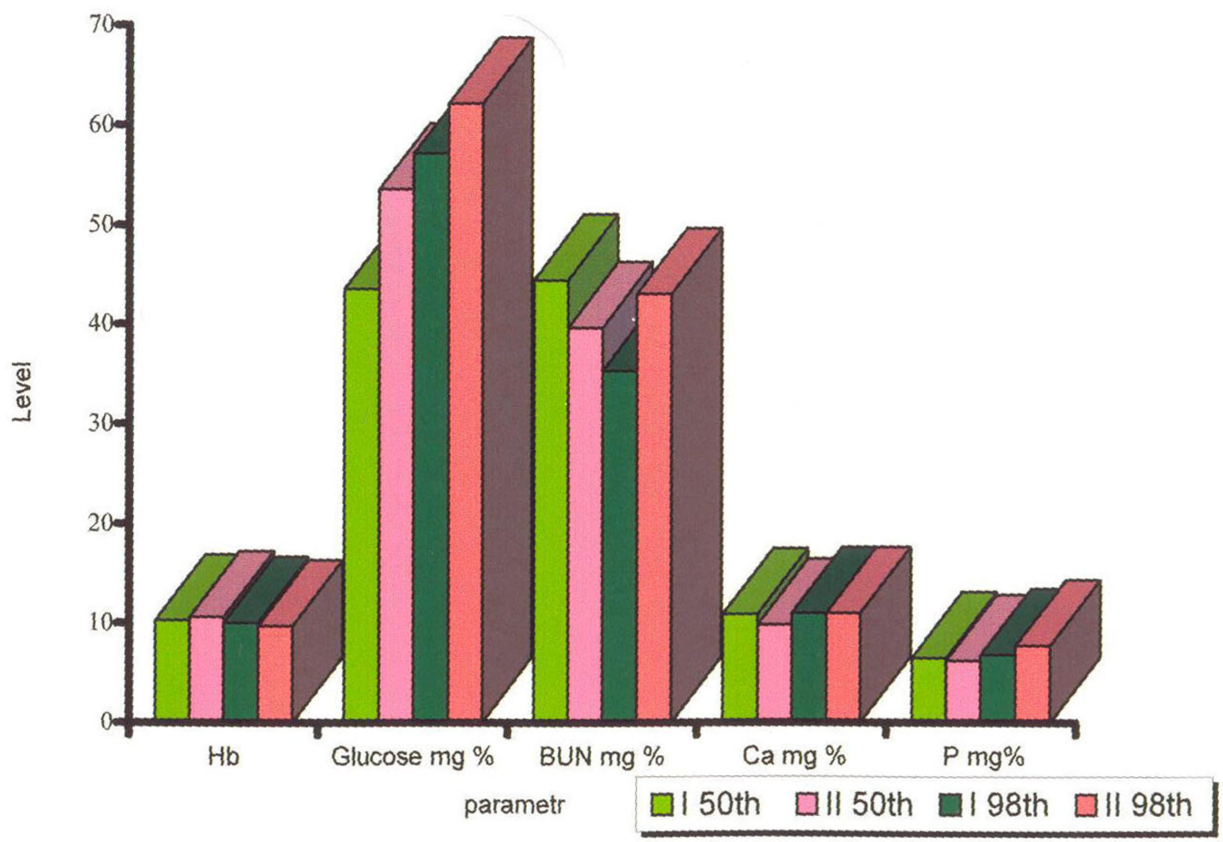

Fig. 16. Haematological parameters of the animals maintained on two experimental rations in phase II

Table 25. Percentage chemical composition* of the dung from the animals maintained on the two experimental rations in phase II

Nutrient	Group 1	Group II
Dry matter	19.57 ± 0.37	21.78 ± 0.75
Crude protein	9.45 ±1.01	11.05 ± 1.07
Ether extract	3.51 ± 0.39	2.43 ± 0.27
Crude fibre	21.06 ± 1.2	20.13 ± 0.71
Total ash	18.13 ± 0.30	15.63 ± 0.36
Nitrogen free extract	47.85 ± 1.71	50.76 ± 2.35
Neutral detergent fibre	59.55 ± 1.52	59.32 ± 1.07
Acid detergent fibre	36.04 ± 1.56	35.36 ± 1.05

* Average of six values

Table 26. Digestibility coefficient of nutrients* of the two experimental rations in phase II

Nutrient	Group 1	GroupII
Dry matter	62.34 ± 0.17	66.67 ±1.57
Crude protein	74.22 ± 2.41	73.69 ± 2.66
Ether Extract	71.33 ± 2.86 [A]	86.19 ±1.44 [B]
Crude fibre	52.07 ± 3.13	55.67 ± 1.76
Nitrogen free extract	68.11 ± 2.68	69.21 ± 2.45
Neutral detergent fibre	42.70 ± 2.94	45.02 ± 2.68
Acid detergent fibre	48.96 ± 4.11	54.02 ± 2.53

* Average of six values

A, B- Means with different superscript in the same row differ significantly ($P<0.01$)

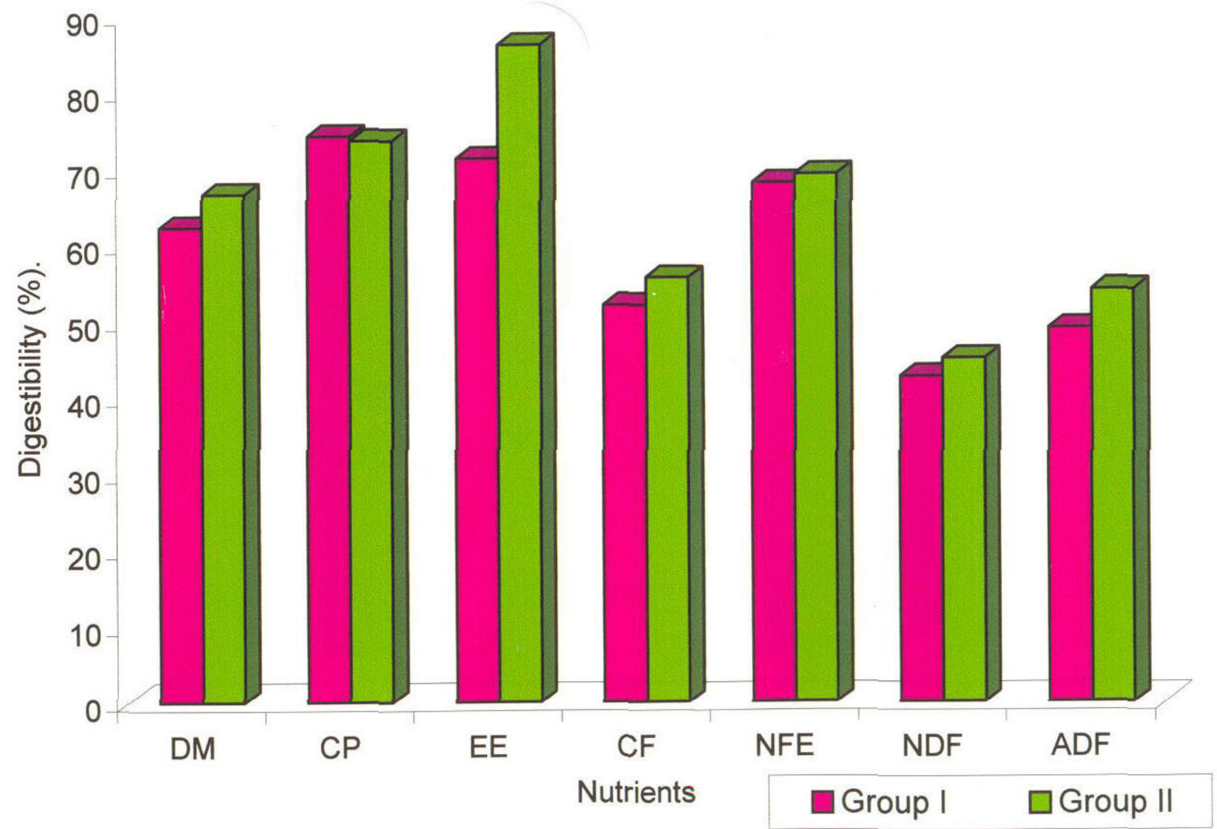

Fig. 17. Digestibility coefficient of nutrients of the two experimental rations in phase II

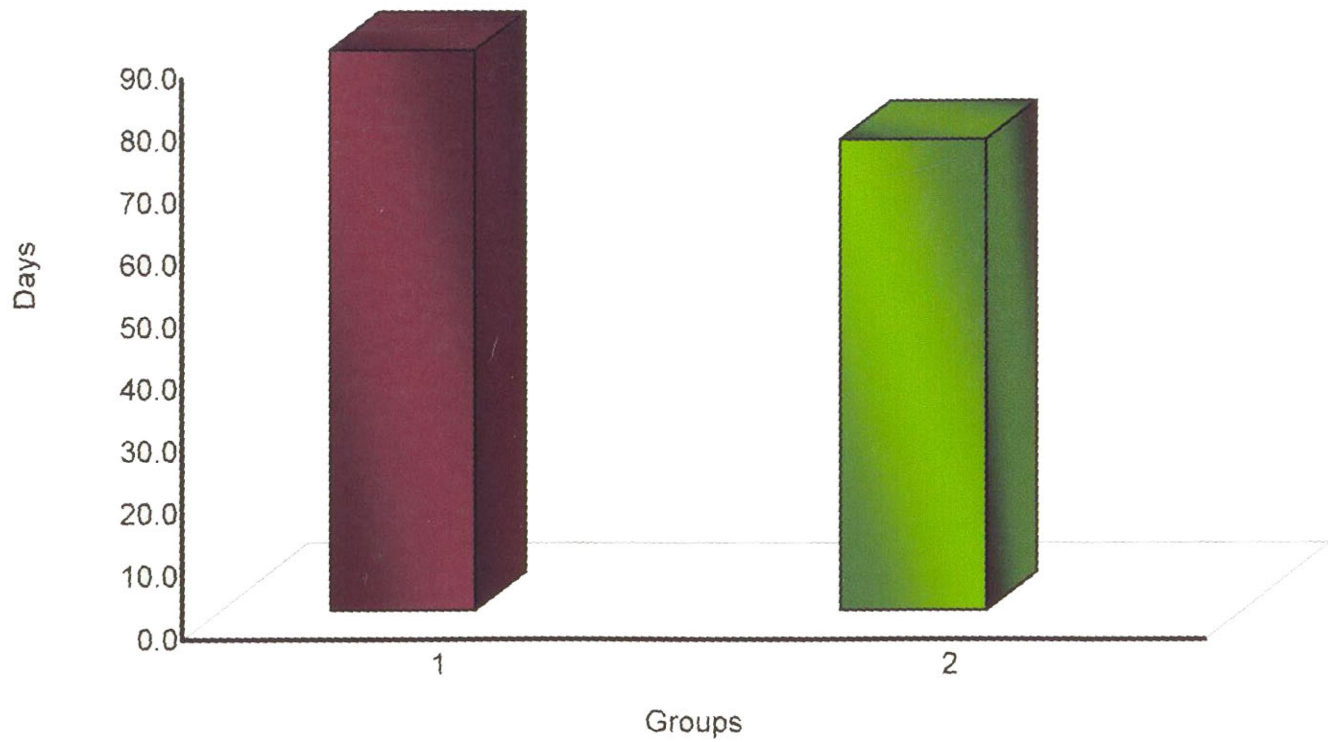

Fig. 18. Average days to first service in animals maintained on two experimental rations in phase II

Table 27. Total dry matter intake and cost per kg milk production of animals maintained on the two experimental rations in phase II

Parameter	Group I	Group II
Total concentrate consumed in 100 days (kg)	4620[A]	3889.2[B]
Total straw consumed in 100 days (kg)	2038[a]	1693[b]
Total feed consumed in 100 days (kg)	6658[a]	5582.2[b]
Dry matter intake per 100 kg body weight (kg)	3.49[a]	2.97[b]
Dry matter intake per kg metabolic live weight (W kg $^{0.75}$) (kg)	14.73[a]	12.5[b]
Total milk produced in 100 days (kg)	5549.9	4623
Feed intake per kg milk produced (kg)	1.2	1.21
Cost of one kg concentrate* (Rs.)	6.64	7.79
Cost of one kg straw * (Rs.)	2.19	2.19
Total cost of feed (Rs.)	35140.02	34004.54
Cost / kg milk produced (Rs.)	6.33	7.36

* Calculated using the rate contract values fixed for feed ingredients by College of Veterinary and Animal Sciences, Mannuthy.
Means with different superscripts in the same row for each parameter vary significantly- A,B- ($P< 0.01$), a,b- ($P< 0.05$)

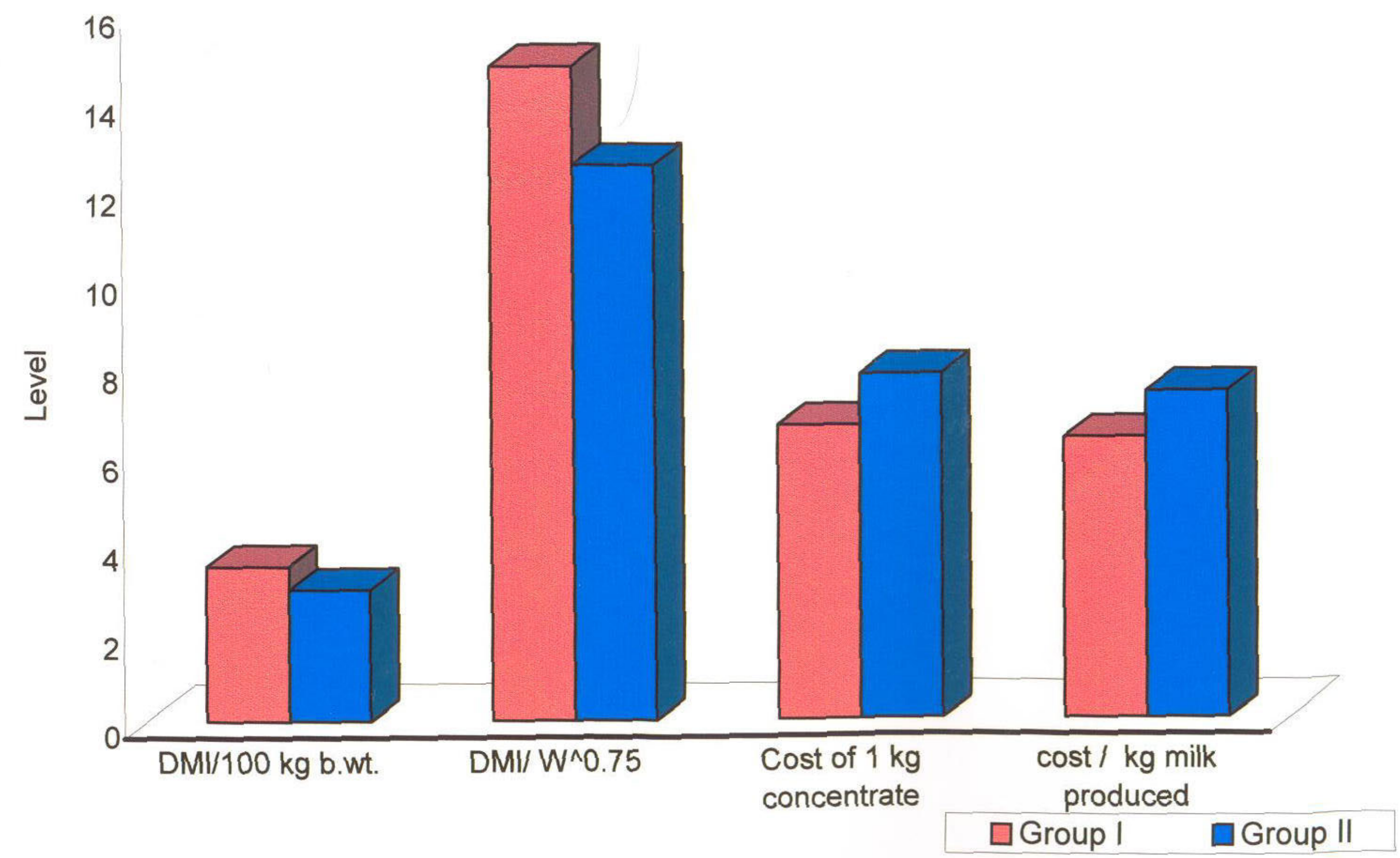

Fig. 19. Dry matter intake and cost of production of milk in animals maintained on two experimental rations in phase II

Discussion

5. DISCUSSION

5.1 PHASE I

5.1.1 **Chemical Composition**

The percentage of CP in the concentrate mixtures were 26.55, 20.01and 16.85, respectively and that of the paddy straw was 4.68 (Table 5). The CP content of the three rations formulated with concentrate and roughage in the ratio of 70:30 of the total dry matter intake was 20.0, 15.41 and 13.20 percentage. respectively. Clark and Davis (1980) suggested that a CP level of 13 to 14 per cent was sufficient to get maximum response in milk yield by dairy cow producing 20 kg milk per day. Chalupa (1984) and Clark *et al.* (1987) also opined that a CP level above 14 to16 per cent in the ration is not economical in the case of dairy cows in early lactation producing 20 to 25 kg per day. A protein content of 16 per cent was recommended in the ration of cows producing more than 30 kg milk per day by many scientists (Satter and Roffler, 1975; Claypool *et al.*, 1980: Christensen *et al.*, 1993 a). Increase in milk production by cows receiving 14 per cent CP ration compared to those fed 11 percent CP ration was reported by Kwan *et al.* (1977), Cowan *et al.* (1981) and Kung and Huber (1983). National Research Council (NRC, 1989) recommended a protein level of 19 per cent in the ration for high producing cows in early lactation. The Bureau of Indian Standards (IS: 2052. 1992) has recommended the level of CP as 22 per cent and 20 per cent. respectively in the grade I and II compounded cattle feed.

The calculated total digestible nutrient (TDN) content of the concentrate mixtures was 72.5 per cent in all the three mixtures, while the actual TDN content of the rations constituted by the concentrate mixture and straw in the ratio. 70:30 were 63.86, 61.28 and 60.74, per cent respectively. The acid detergent fibre (ADF) content of the three rations were 17.28, 19.7 and 23.5 per cent respectively. Acid detergent fibre level of 21 per cent in the ration was recommended by NRC

(1989), while Ensminger *et al.* (1990) opined that the ADF content in the ration of early lactating cows should not be below 17 to 18 per cent of ration. An ADF level of 17 to 18 per cent was followed also by Barney *et al.* (1981), Annexstad *et al.* (1987) and Christensen *et al.* (1993b).

5.1.2 Body Weight

A perusal of the data on the body weight of the animals at various periods of the feeding trial, fed with the three experimental rations, listed in Table 6 shows that there was no significant difference between the body weight of the animals of the three groups in the fortnights one, two, three and five, but the animals of the second group had significantly higher (P<0.05) body weight than those of first and third group in the fourth and sixth fortnight (Table 7). The animals in groupI lost weight from 349.8 kg to 323.3 kg, while the animals in group II gained weight from 347.7 to 356 kg while the animals in group III, more or less maintained their body weight, even though there was a decline in weight during the last fortnight.

There was a loss in the body weight of the animals in all the three groups, during the first three fortnights of feeding trial. A loss in body weight of animals during early stages of lactation when the dry matter intake (DMI) will not be sufficient to meet the requirements of production was also observed by Botts *et al.* (1979). The loss of body weight of the animals of first group correlates well with the low DMI of the animals of this group till the end of the experiment (100 days). Claypool *et al.* (1980), using 13, 16 and 19 per cent CP rations, reported that body weight loss was lowest in cows fed 16 percent CP ration compared to those of the other two groups and the loss in body weight was largest in cows fed 19 per cent CP ration.

5.1.3 Dry Matter Intake (DMI)

Dry matter intake of animals of group I was significantly lower in all weeks except week one, four and five ($P<0.01$) and during week two ($P<0.05$) than that of group II. The DMI of animals of group I was also significantly lower than that of group III during weeks one, two, three, seven, twelve, thirteen, fourteen ($P<0.01$) and in the rest of the period except weeks four and five ($P<0.05$). The DMI of animals of group II was not significantly different from that of group III during the entire experimental period except during the first week. Total intake of straw by the animals in all the three groups was more or less uniform (2104 kg, 2348 kg, and 2226 kg as given in Table 16). The concentrate intake by the animals of group I was lower than that of the animals in group II and III (3690, 5208 and 5132 kg, respectively as given in Table 16). The lower concentrate intake of animals fed the high protein diet may be due to the stress involved in catabolising the excess amino acids and its excretion as urea in the body. Since these animals consumed more or less the same quantity of roughage, it could be assumed that the animal did not develop any metabolic disorder. Lower palatability can also be a possible reason for lower intake in the high protein group since the concentrate mixture comprised exclusively of maize and GNC. However the average daily DM intake of animals of all groups were 9.85, 12.84 and 12.44 kg, respectively, which were higher especially for groups II and III than that recommended by NRC (1978) (10.94) and ARC (1980) (9.4) for cows weighing 600 kg and producing 10 kg milk.

Dry matter intake of animals of group I and III peaked during the fifth week of the feeding trial, while peak DMI was observed during the eighth week in the second group. The DMI of animals in the three groups were 2.99, 3.54 and 3.79 per cent of their body weight. Dry matter intake of 3 to 3.6 per cent of body weight have been reported by other scientists also (Edwards *et al.*, 1980; Robbinson *et al.*, 1991; Winsyrg *et al.*, 1991; Annexstad *et al.*, 1987).

An increase in the DMI in response to an increase in the ration CP had been made by other scientists also (Barney *et al*., 1981; Cowan *et al*., 1981; Glenn *et al*., 1986). Tolkamp *et al*. (1998) observed an increase in the DMI by dairy cows in early lactation when ration CP was increased from 13 to 18.5 per cent. Atwal *et al*. (1995) reported no significant difference in the DMI of cows given 15 and 17 per cent CP rations. Lack of influence by the ration CP level on the DMI or on body weight of early lactating dairy cows was reported by Claypool *et al*. (1980) with 12 to 19.3 per cent CP rations and Baxter *et al*. (1983) with 12 to 17 per cent CP rations and Howard *et al*. (1987) with 14.5 and 19.4 per cent CP rations.

5.1.4 Milk Production

The weekly average milk production by the animals of the three groups presented in Table 9 shows that the milk production by animals in the three groups increased during the first month of lactation. Milk production by the animals in group I peaked at 11.02 kg during second week of lactation, then declined to 6.7 kg in the 14th week of lactation. The milk production of the second group peaked at 10.98 kg during the fourth week, maintained the production during the fifth week and then declined to 8.4 kg during the 14th week. In the third group, the milk yield peaked at 10.31 kg during sixth week of lactation, then declined to 7.71 kg at the end of 14th week. The production curve was more or less similar for the second and third group while in the first group there was a very sharp decline in yield from seventh week onwards. Covariance analysis on the data on the milk production of the animals of the three groups listed in Table 10 reveals that there was no significant effect on the milk production by the three levels of CP in the concentrate mixture or ration, indicating that 13.2 per cent CP in the ration was sufficient to meet the protein requirement. The total milk production in 100 days by the animals of the second group was the highest among the three groups, which may be due to higher initial milk yield of the animals of the second group. This difference in the initial milk production of animals of three groups occurred due to the inclusion of first lactation cows in equal numbers in all the groups, whose production could not be predicted.

The average decline in milk production from the first week to the last week of the experiment was 2.01, 1.83 and 1.52 kg, respectively in the groups I, II, and III, showing that the third group maintained their milk production better than those of the groups I and II. National Research Council (NRC, 1985) compared the levels of CP recommended by nine systems and it varied from nine to thirteen per cent for cows producing 10 kg milk / day which agrees with the results obtained in the present study.

Edwards *et al.* (1980) using 15 and 17 per cent CP rations in cows producing on an average 29 kg per day, Claypool *et al.* (1980) with 13, 15 and 17 per cent CP rations and Christensen *et al.* (1993a) with 16.4 and 19.4 per cent CP rations in cows in early lactation producing around 30 kg per day also did not observe any significant increase in milk production of early lactating cows when CP content of the ration was increased. On the other hand, the findings of the present study are in disagreement with that of Lundquist *et al.* (1983) and Roffler and Thacker (1983a) who reported an increase in milk yield when ration CP was increased from 13 per cent to 17.5 per cent. An increase in milk production by raising the CP of the ration from 11 to 14.5 per cent also had been reported by Klusmeyer *et al.* (1990) in cows producing more than 30 kg per day. An increase in milk production in response to increased dietary CP levels was reported by other scientists also (Macleod *et al.*, 1984; Glenn *et al.*, 1986). The low production capacity of the animals used in the present study may be the reason for not getting an increase in milk production with increased CP levels as obtained by the above scientists.

5.1.5 Milk Composition

Data on the composition of the milk collected fortnightly from the animals maintained on the three treatments, (Table 11) shows that there was no significant difference between the three groups in any of the parameters studied throughout the experiment except in the fifth fortnight, fat percentage of milk from animals of group III was significantly lower ($P<0.05$) than those of the other two groups. However there was no significant difference in the average fat percent between the groups (3.48, 3.51 and 3.48, respectively).

The average total solids in the milk from animals of the groups I, II and III were 11.81, 11.87 and 12.01 g per cent, respectively, while that of SNF were 8.33. 8.36 and 8.53 g per cent, respectively. Edwards *et al.* (1980) reported a significantly higher SNF percentage in the milk of cows given 15 per cent CP ration compared to those of 13 per cent CP ration, but there was no difference in the SNF level between the 13 and 17 per cent CP and 15 and 17 per cent CP rations. Henderson *et al.* (1985) observed no significant difference in the SNF content (8.78, 8.80 and 8.74 g per cent) of milk produced by cows fed 22, 15 and 16 per cent CP rations. Christensen *et al.* (1993a) also did not observe any significant effect on the SNF level in the milk by the CP level in the ration (16 and 19 per cent).

The average protein content of the milk from the animals of the three groups were 2.59, 2.54 and 2.52 per cent, respectively, and the values ranged between 2.43 and 2.91 in the first group, 2.21 and 2.86 for the second group and 2.32 and 2.63 in the third group. Claypool *et al.* (1980) reported a fat per cent of 3.6, 3.2 and 3.4 in the milk of animals fed 12.7, 16.3 and 19.3 per cent CP rations while the protein per cent was 3.5 per cent in all the three groups. Barney *et al.* (1981) observed that the milk of cows fed 17 per cent CP ration contained 3.15 and 3.2 per cent fat and protein, respectively. Klusmeyer *et al.* (1990) using 11 and 14.5 per cent CP rations reported that milk contained 3.42 and 3.53 per cent fat and 3.31 and 3.27 per cent protein, respectively, while Christensen *et al.* (1993 b) reported a fat level of 3.0 per cent and protein content of 2.8 per cent when fed a ration with 16.4 per cent CP.

The values for all the milk parameters studied in the present work agree well with the reported values, except for protein. Lower protein percent in the milk observed in the present study may be due to the difference in the genetic make up of the animals used for the study since the milk fat and protein contents are determined by their genetic make up. However, Sathian'(2001) reported a fat level of 4.64 and 4.6 per cent, 13.23 and 12.95 g per cent total solid, 8.59, 8.34 g per cent SNF and protein content of 3.13 and 3.0 per cent in the animals of University Livestock Farm and field animals, respectively, values being higher than those obtained in the present study. In the fifth fortnight there was no significant difference in the fat level of milk of the first and second group animals but it was significantly higher ($P<0.05$) than that of the third group. Dietary CP level did not affect the milk composition in the present study which correlates well with the reports of other scientists (Roffler *et al.*, 1978; Van Horn *et al.*, 1979; Edwards *et al.*, 1980), while Forster *et al.* (1983) and Henderson *et al.* (1985) reported an increase in the fat and protein levels of milk in response to an increase in ration CP.

5.1.6 Rumen Fermentation Parameters

The initial values of pH, NH_3-N, TVFA and lactic acid levels in the rumen liquor collected before the beginning of the experiment were 6.00, 82.97 meq/l, 29.07 mg percent and 0.32 mg/ml respectively. The rumen fermentation parameters of the animals of the three groups I, II and III estimated from the rumen liquor collected on the 92nd day are given in Table 12. The pH of the rumen fluid collected towards the end of the feeding trial was 7.71, 7.0 and 7.37, respectively, for the three experimental groups, values for group I being significantly higher (P<0.05) than that of the other two. A rumen pH of 6.4 was reported by Claypool *et al.* (1980) and Forster *et al.* (1983), while Klusmeyer *et al.* (1990) and Weigel *et al.* (1997) reported rumen pH of below six. The pH in all the groups of the present studt were slightly above the normal range of 5.5-6.5 to 7. Crawford *et al.* (1978), Meng *et al.* (1999) and Meng *et al.* (2000) reported an increase in the rumen pH corresponding to an increase in the dilution rates. Lack of any significant influence of the ration on rumen pH were reported by many scientists (Cressman *et al.*, 1980; Ha and Kennelly, 1984; Annextad *et al.*, 1987; Warley *et al.*, 1994; Devant *et al.*, 2000), while Klusmeyer *et al.* (1990) and Broderick *et al.* (1993) observed an increase in rumen pH as the ration CP was increased.

Total volatile fatty acid levels were 93.81, 68.71 and 86.50 meq/l which were not significantly different and were within the normal range of 50-150 meqv /l (Mc Donald *et al.*, 1995). The higher numerical levels of TVFA in group I, though not significant, may probably be because of the significantly higher (P<0.05) level of lactic acid in rumen liquor of these animals. The observations made in the present study are in agreement with those of Cressman *et al.* (1980) and Ha and Kennelly (1984) who also did not get any significant effect on the TVFA production by the various levels of CP in the ration. Klusmeyer *et al.* (1990) reported an increase in the TVFA level from 98.6 to 112.7 mM when the ration CP was increased from 11 to 14.5 per cent. Sawal and Kurar (1997) opined that the TVFA levels were governed more by the form and content of carbohydrate in the ration rather than the protein level.

The average lactic acid levels in the rumen liquor from animals of three groups were 0.33, 0.21 and 0.22 mg/ml. The lactic acid level in the group I was significantly higher (P<0.01) than that of the other two groups. But in all the three groups, the lactic acid levels were within the normal range of 10 to 30 mg per cent as reported by Rossenberger (1979). Verma *et al.* (1975) reported 5 to 10 mg per cent of lactic acid in the rumen liquor of goats.

Ammonia nitrogen content of the rumen fluid collected from animals of the three groups were 39, 31 and 17 mg/ml, all of which were higher than the critical value of 5 mg/100ml of liquor (Bhatia and Pradhan, 1980) required for the optimum microbial protein synthesis (MPS). The significantly higher ($P<0.05$) rumen NH_3-N levels in the first (39.0 mg per cent) and second (31.0 mg per cent) group may be because of the higher CP (26.5) level in the first and second groups and higher DMI and CP digestibility in group II than that of group III. An increase in the rumen NH_3-N concentration in response to an increase in the ration CP levels were also reported by Polan *et al.* (1976), Kwan *et al.* (1977), Janicki *et al.* (1985), Warley *et al.* (1992) and Jones-Endsley *et al.* (1997). Satter and Roffler (1975) reported a rumen NH_3-N concentrations of 0.8 mg per cent in animals fed 8 per cent CP rations and 56.1 mg per cent in animals fed 24 per cent CP rations.

5.1.7 Haematological Parameters.

The initial values of haemoglobin, blood glucose, blood urea nitrogen (BUN), plasma calcium and plasma phosphorus were 8.99 g per cent, 59.16 mg percent, 18.10 mg per cent, 8.86 mg per cent and 6.65 mg per cent. The values of various haematological parameters such as haemoglobin, blood glucose, blood urea nitrogen, plasma calcium and plasma phosphorus estimated from the blood collected on the 50^{th} and 98^{th} day are listed in the Table 13. A perusal of these data indicates that all the values for the parameters studied ranges between the normal levels reported for the species. There was no significant difference between the three groups in any of the parameters studied, except that of BUN.

Blood urea nitrogen levels in the second group (21.5 mg per cent) was significantly higher ($P<0.01$) than that of third group (13.6 mg per cent) on the 50th day collection. There was no significant difference between the first and second group as well as the first and third group. Though the level of CP was highest in the first group (20 per cent CP in the ration), because of reduced DMI especially that of concentrate mixture, no significant higher BUN level (17.1 mg per cent) was observed in these animals. There was no significant difference in the BUN level (17, 18 and 18 mg per cent) between the three groups on the 98th day collection.

Gustafsson and Palmquist (1993) and Elrod and Butler (1993) reported that plasma urea nitrogen (PUN) concentrations fluctuate through out the day. Butler *et al.* (1996) reported a PUN level of 16 to 25 mg per cent in cows and that the maximum PUN level was observed in four to six hours after feeding. The observations made in the present study regarding the BUN levels are in agreement with those of Botts *et al.* (1979), Howard *et al.* (1987) and Higginbotham *et al.* (1989). Jordan *et al.* (1983) also reported elevated BUN levels in dairy cows as the level of CP of the ration was increased from 12 to 23 per cent.

5.1.8 Digestibility Coefficient of Nutrients

The digestibility coefficient of the nutrients such as DM, CP, CF, NDF, ADF, EE and NFE are given in Table 15. The digestibility coefficient of EE, CF, NDF and ADF were significantly higher ($P<0.01$) for ration I compared to the other two rations. The digestibility coefficients of nutrients of ration II and III were not significantly different. The low concentrate intake and the higher protein per cent in the first ration might be the reasons for the significantly higher fibre digestibility in this group. Cressman *et al.* (1980) used 12, 15 and 18 per cent CP rations and observed a linear increase in CP digestibility along with increase in the CP level in the ration, but there was no difference in the DM and ADF digestibility. Kwan *et al.* (1977) and Grieve *et al.* (1980) also reported increased CP digestibility with increased level of CP in the ration. Cowan *et al.* (1981) observed better digestibility of nutrients as the level of CP in the ration was increased. Ha and Kennelly (1984) and Stern *et al.* (1985) observed an increase in the digestibility of DM and OM as the CP level in the ration was increased. Christensen *et al.* (1993 b) reported better ADF digestibility with 16 per cent CP ration than with 19 per cent CP ration, while there was no difference in the OM or NDF digestibility.

5.1.9 Reproductive Parameters

The average number of days to first post partum insemination were 122, 79.4 and 80 days respectively, in the three groups and the difference between the groups was nonsignificant. All the animals in group II conceived during the experimental period, while only one each in the first and second group conceived during this period. Jordan and Swanson (1979) and Folman *et al.* (1981) reported an increase in the number of services for conception, when the CP level of the ration was above 16 per cent. But Howard *et al.* (1987) reported that the service period (80.4 and 79.9) and number of AI for successful insemination (1.39 and 1.40) were not affected by the CP level in the ration (14.5 and 19.4 per cent). Similarly Carroll *et al.* (1988) reported average service periods of 72 and 82 days and the average number of AI for conception as 1.5 and 1.8 in dairy cows in early lactation fed 13 and 20 per cent CP rations, respectively. They also reported that the level of CP in the ration was not having any significant effect on the reproduction.

The animals in the second group had a BUN level of 21.6 mg per cent on the 50th day, which was slightly above critical level of 20 mg per cent as suggested by Ferguson *et al.* (1993) for optimum reproduction, while the BUN level on the 98th day (18 mg per cent) was below the critical level. Animals of the three groups had a BUN level below 20 mg per cent on the 98th day and hence the nutritional status of the animals might have been the factor, which controlled the fertility. The animals in the second group gained weight, those in the third group more or less maintained their body weight, while the animals in the first group lost weight, indicating that these animals were in negative energy balance. The conception rate was highest in the second group and poor in the first group. Dunn and Moss (1992) opined that the negative energy balance due to restricted feeding can lead to weight loss in animals, which can result in cessation of oestrus cycle and development of quiescent ovaries. Positive correlations between body weight and conception rates were reported by Wishart *et al.* (1977), Day *et al.* (1986) and Houghton *et al.* (1990), where as the observations of Gardner *et al.* (1977) and Harrison and Randel (1986) disagree with this.

5.1.10 Cost of Production

From the data given in Table 16, it can be observed that the total DMI of the animals in the three groups were 5794, 7556 and 7358 kg, respectively. The lowered DMI in group I was due to the reduced concentrate intake by these animals (3690 kg in comparison to an intake of 5208 kg and 5132 kg in the second and third groups, respectively). Higher DMI by the animals in the second group led to an increase in the weight of the animals in this group (356 kg compared to 323.3 and 323.2 kg in the other groups on the 96th day).

The DMI per 100 kg body weight in the three groups were 2.99, 3.54 and 3.79 kg, respectively and the DMI per kg metabolic live weight were 12.67, 15.37 and 16.08 kg, respectively. It could be seen that both the DMI per 100 kg body weight and per kg metabolic live weight were significantly higher ($P<0.01$) in the second and the third group than the first group. The feed intake for every one kg milk production was 1.15, 1.29 and 1.36 kg, respectively for the three groups.

From the Table 16 it could also be seen that the total milk production in the three groups were 5041, 5855 and 5423 kg. The total milk yield in 100 days from the animals of the second group was numerically more than that of the other two groups. The cost for producing one kg milk of the three groups were 7.79, 7.91 and 7.71 rupees, respectively. Claypool *et al.* (1980) reported that cows fed 16 per cent CP rations had highest return over feed cost than those fed 19 per cent CP ration. Barney *et al.* (1981) reported a better return above feed cost in animals maintained on 17 per cent rations for longer periods (14 weeks) compared to those maintained on 17 per cent ration for four, nine or nineteen weeks after which they are maintained on 13.5 per cent CP ration.

From the overall evaluation of the results obtained in the first phase of the experiment it could be observed that the animals of third group, fed concentrate mixture containing 16.85 per cent CP maintained their body weight and milk production better than those of the other two groups throughout the experimental period. The cost for producing one kg milk was also lowest for the animals of third group. Thus it could be concluded that a concentrate mixture with 16.85 per cent CP or a ration with 13.2 per cent CP was sufficient to meet the requirements of crossbred dairy cows in early lactation with a peak of 10 kg milk per day. Hence the concentrate mixture with 16.85 per cent CP was selected for the second phase of the experiment.

5.2 PHASE II

5.2.1 Chemical Composition

The proximate composition of the two concentrate mixtures used in the second phase of the study given in Table17 reveals that both the concentrate mixtures were isonitrogenous and isocaloric, the CP levels being 17.24 and 17.18 per cent, respectively. The calculated TDN content of the two rations formed by concentrate and roughage in the ratio 70:30 were 64.33 and 66.98 per cent.

respectively. Undegradable protein content (UDP) content of the two concentrate mixtures, calculated by using the published UDP content of the ingredients were 26.8 and 42.9 per cent of the CP, respectively. Acid detergent fibre content of the rations were 26.7 and 27.34 per cent, respectively on DM basis. National Research Council (NRC, 1989) recommended 36.6 to 43.3 per cent UDP and 49.8 to 74 per cent RDP (of ration CP) for 400 kg cows producing 7 to 13 kg milk per day. Agricultural Feed Research Council (AFRC, 1993) recommended 261 g of metabolisable protein (MP) and 57 MJ of ME for dairy cows weighing 550 kg. Burroughs *et al.* (1978) and Waldo and Glenn (1984) reported that rumen available protein at levels of 45 to 75 per cent of the CP of ration gave maximum production, while Majdoub *et al.* (1978) recommended RDP level of 15 to 30 per cent. Kalbande and Thomas (1999) reported an increase in the milk production by increasing the UDP level of the concentrate mixture from 30 to 63 per cent. An acid detergent fibre level above 25 per cent in the ration was recommended by Baxter *et al.* (1983), Roffler and Thacker (1983a) and Krikpatrick and Kennelly (1989).

5.2.2 Body Weight

The average body weight of the animals maintained on the two isonitrogenous rations are given in Table 18 and covariance analysis of data in Table 19. It could be observed that there was no significant effect on the body weight by the UDP level in the concentrate mixture. The body weight of the animals in group I, increased from 323.57 kg to 331 kg in the third fortnight of the trial and then decreased to 318.18 kg. The animals fed concentrate mixture with 42.9 per cent UDP more or less maintained the weight around 315 kg throughout the experimental period. Atwal and Erfle (1992) reported that feeding of SBM along with fish meal resulted in lowest weight gain in animals compared to feeding SBM or fish meal alone. Crish *et al.* (1986) observed that the animals fed 15 per cent CP rations with lower level of UDP (43 per cent) lost weight compared to the higher UDP (67 per cent) ration. Casper *et al.* (1999) reported no change in the body weight of cows, when SBM was replaced by extruded soybean meal (ESBM) in the ration.

5.2.3 Dry Matter Intake

The dry matter intake of the animals maintained on the two isonitrogenous rations, is given in Table 20. It could be seen from the table that the animals of group I had a significantly higher (P<0.01) dry matter intake from the third week of feeding trial to the end of the trial. The DMI by the animals of both the groups peaked at second week of the trial and there after the intake was maintained at that level in group I, while it was decreased gradually in group II. The DMI of animals of the two groups were 3.49 and 2.97 per cent of their body weight (Table 27). Both the concentrate and straw intake of the animals of group II were lower (P<0.01 and P<0.05, respectively) than those of group I. A decrease in the DMI was reported in animals fed fish meal incorporated ration in comparison to those containing SBM (Atwal and Erfle, 1992; Calsaminglia *et al.*, 1995; Gracia-Bajolil *et al.*, 1998), while Bernard (1997) observed an increase in DMI of cows when meat meal or meat cum bone meal was included in the ration of multiparous cows replacing SBM or cotton seed. Atwal *et al.* (1995) observed no change in the DMI of animals given treated and untreated SBM. Erdman and Vandersall (1983) did not observe any change in the DMI of cows given 14 per cent CP rations at two levels (28 and 47 per cent) of UDP. Similarly, source or degradability of protein in the ration did not influence the DMI as reported by Crawford and Hoover (1984), Bowman *et al.* (1988), Blauwiekel *et al.* (1990) and Christensen *et al.* (1993a). Carroll *et al.* (1994) also failed to observe any change in DMI or body weights of animals given rations with 34 and 40 per cent UDP.

5.2.4 Milk Production

The daily average milk production by the animals of the two experimental groups are given in Table 20 and the covariance analysis of this data are presented in Table 21. The production in group I peaked at 10.01 kg at the second week of trial, then declined sharply to 9.12 kg during the third week. Then the production was maintained at this level, then declined to 8.51 kg on the 14th week of trial. In the case of animals of group II there was no distinct peak production, but the production of 8.84 kg during the third week of trial, gradually declined to 7.45 kg at the 14th week of trial. It could be seen from Table 21 that the difference in milk production was significant between the two groups during the second and fourteenth ($P<0.01$), seventh, eighth and thirteenth ($P<0.05$), ninth, eleventh and twelfth ($P=0.06$) weeks of trial. The difference in the initial milk yield of the animals of the two groups was taken care of by doing covariance analysis and using the adjusted means, variation in milk production was calculated which was lower for the animals of the second group, indicating that the production of the animals fed high UDP ration was maintained better.

A positive response in milk yield to UDP feeding in dairy cows maintained on crop residue based diets was observed by Ramachandra and Sampath (1995) and Misra and Rai (1996). Clark *et al.* (1973), Deering *et al.* (1974) and Roffler *et al.* (1978) also made similar observations. Kalbande and Thomas (1999) reported an increase in the milk yield of cows when UDP level was 63 per cent of the CP compared to those fed 30 per cent UDP ration maintained on poor quality roughage. This positive response in milk production may be because of low microbial protein production due to energy deficiency in these animals fed low quality roughage and providing UDP helped to provide the required amino acid in the small intestine for absorption.

Forster *et al.* (1983) observed an increase in production of cows producing around 30 kg milk per day when the UDP level of 14 per cent CP ration was increased from 43 to 54 per cent. The positive response in the milk production to increased UDP level made in the present study correlates well with the observations of other scientists (Holter *et al.*, 1985; Glenn *et al.*, 1986; Schingoethe *et al.*, 1988; Volden, 1999; Mc Cormick *et al.*, 2001; Todorov *et al.*, 2000). Armentano *et al.* (1993) using 16.4 per cent CP rations at 45 and 35 per cent UDP levels, Aharoni *et al.* (1993) with 17 per cent CP rations at 30 and 35 per cent UDP levels and Carroll *et al.* (1994) with 21 per cent CP rations at 34 and 40 per cent UDP levels, observed higher milk yields when the ration UDP levels were more. The source or the degradability of dietary protein did not influence milk production as reported by Erdman and Vandersall (1983), Higginbotham (1989), Casper *et al.* (1990), Robbinson *et al.* (1991), Petit and Veira (1991) and Henson *et al.* (1997).

5.2.5 Milk Composition

The fortnightly average composition of milk produced by the animals of the two groups is given in Table 22. An evaluation of the data indicates that all parameters estimated were well within the range specific for the species. There was no significant effect of UDP level of the diet on any of the milk parameters studied such as milk fat, protein, SNF, total solids or milk urea nitrogen. In both the groups the fat per cent increased from 3.36 to 3.83 in group I and from 3.08 to 3.41in group II. During the fourth fortnight, there was significantly higher milk fat content ($P<0.05$) and a correspondingly higher ($P<0.01$) total solids in the first group. De Peters *et al* (1985), Coppock *et al.* (1987) and Aharoni *et al.* (1993), observed an increase in the milk fat content in response to an increase in the UDP level of the ration. However, Blauwieckel *et al.* (1990) observed no change in milk fat content by the incorporation of UDP supplements such as fish meal in comparison to SBM. A depression in the milk fat per cent by replacing SBM with fish meal in the ration was reported by Zerbini *et al.* (1988) and Spain *et al.* (1990).

Total solids and SNF levels were lower in the second group than those of the first group. The average total solids content in group I (12.31) was significantly higher (P<0.01) than that of the second group (11.31 g per cent), while the average values of the total solids in the various fortnights ranged between 11.56 to 12.97 g per cent in the first group and between 10.51 to 11.83 g per cent in the second group, which were not significantly different. Kung *et. al.* (1983) reported that there was no significant effect on the total solid content of milk (11.00 and 11.21 g per cent) of cows receiving SBM or heated SBM in the ration as protein supplement. Similarly, Kim *et al.* (1991) and Casper *et al.* (1999) could not observe any significant difference in the total solids content by replacing ESBM in the ration with SBM.

Solids not fat content of the milk of the first group animals ranged between 8.08 to 9.02 g per cent while in the second group it was between 7.19 to 8.71 g per cent. Crawford and Hoover (1984) reported a decrease in the SNF level of milk by feeding ration containing treated SBM in comparison to those with untreated SBM, while Christensen *et al.* (1993 a) did not observe any change in SNF by the source or degradability of protein in the ration. Klusmeyer *et al.* (1990) reported that when SBM containing rations with two levels of protein (11 and 14.5 per cent) were fed to cows, there was no significant change in the SNF (8.95 and 8.75) levels of milk. Kalbande (1995) reported that feeding three levels of UDP (63, 48 and 30 percent) did not affect fat, total solids, SNF or protein of milk whose average values were 4.64, 13.5, 8.86 and 3.52 g per cent, respectively.

The protein level in milk of the animals of the first group was maintained at 2.86 g per cent except in the fourth and fifth fortnight, which was 2.96 and 3.05 g per cent, respectively. Milk protein content of the animals of the second group was

3.01 g per cent in the first fortnight which then decreased to 2.63 g per cent in the sixth fortnight. There was no significant effect of levels of UDP on the protein content of milk. The average protein content of milk from two groups were 2.88 and 2.82 g per cent. Milk protein content of 2.9 to 3 g per cent in animals fed 19 per cent CP ration at 28, 30 and 35 per cent UDP levels had been reported by Robbinson *et al.* (1991). They also reported milk protein contents of 2.8 to 2.9 g per cent with 16 per cent CP ration at 17, 29 and 41 per cent UDP levels. A reduction in milk protein content by inclusion of treated (heat or formaldehyde) SBM in the ration compared to untreated SBM was reported by Crooker *et al.* (1983), Crawford and Hoover (1984) and Faldet and Satter (1991). Wright *et al.* (1998) observed an increase in milk protein content by increasing the UDP supplement similar in composition to bovine casein in the ration, while Aharoni *et al.* (1993) reported no change in milk protein level by increasing the UDP from 25 to 35 per cent in the ration. Baker *et al.* (1995) also reported an increase in the milk protein content from 2.89 to 3.01 g per cent by increasing the UDP level in the ration.

The milk urea nitrogen (MUN) levels observed in the present study were 34.43 and 38.07 mg per cent in the two groups, respectively in the first fortnight of the trial. The corresponding levels were 43.20 and 41.78 mg per cent in the sixth fortnight in the two groups. Oltner and Wiktorson (1983) reported a MUN level between 38 to 76 mg per cent, while Oltner (1985) observed a MUN level of 54 and 71 mg per cent in cows fed 14 and 16 per cent CP rations. He also opined that the cows in first lactation had a lower MUN value. Gustafsson and Palmquist (1993) reported that compared to PUN levels there was only little variation in the MUN levels, while Oltner and Wiktorson (1983) and Moscardini *et al.* (1998) reported an increase in MUN levels when the dietary CP levels were increased. Baker *et al.* (1995) reported an increase in the MUN levels from 31 to 39 mg per cent when the level of urea was increased in the ration. Bector *et al.* (1998) reported that the MUN level in dairy cattle ranged between 22.8 to 92.4 mg per cent, the average being 53.36 mg per cent. Roderiguz*et al.* (1997), Schepers and Meijer (1998) and Kanjanapruthipong and Butanong (2000) observed an increase in the MUN levels by increasing the UDP level of the ration.

5.2.6 Rumen Fermentation Parameters

Table 23 presents the data on the rumen fermentation characteristics of the animals maintained on the two levels of UDP. The initial values of pH, NH_3-N, TVFA and lactic acid levels in the rumen liquor collected before the beginning of the experiment were 7.38, 47.86 meq/l, 16.88 mg per cent and 0.22 mg/ml, respectively. The pH of the rumen liquor from the animals of the two groups were 6.47 and 6.57 which are within the normal range. A rumen pH between 6.3 to 6.9 was reported in animals fed rations containing UDP levels between 25 and 45 per cent (Annexstad *et al.*, 1987; Robbinson *et al.*, 1991; Winsryg *et al.* 1991; Broderick *et al*, 1993). There was no significant effect on rumen pH by the UDP level in the ration. This observation is in agreement with that of Devant *et al.* (2001) who opined that neither source nor degradability of dietary protein affected rumen pH. Foster *et al.* (1983), Krickpatrick and Kennelly (1989) and Kanjanapruthipong *et al.* (2002) also did not observe any influence of UDP levels of the ration on rumen pH. But Christensen *et al.* (1993b) reported a reduction in rumen pH when UDP level of the ration was increased from 30 to 45 per cent.

Total volatile fatty acid and lactic acid contents in the rumen liquor were 76.1 meq/l and 0.21 mg per ml in group I and 77.6 meq/l and 0.1 mg per ml in group II, respectively. Annexstad *et al.* (1987) reported a TVFA level of 68 to 81 mM when 16 per cent CP rations were fed at two levels of UDP (31 and 42 per cent), while Vagoni and Broderick (1997) reported a TVFA level between 150 to 170 mM by feeding 17 to 18 per cent CP rations containing fish meal. The observations made in the present study agrees with the findings of Robbinson and Mcqueen (1994), O'Mara *et al.* (1998) and Meng *et al.* (2000), in which TVFA level of rumen liquor was not affected by the level of UDP. However, Chan *et al.* (1997) and Mathis *et al.* (2000) reported an increase in TVFA production as the UDP level of the ration increased.

Rossenberg (1979) reported a lactic acid level of 10 to 30 mg per cent in the rumen liquor of cattle. Pillai (1988) reported average lactic acid levels of 20 to 22 mg per cent in the rumen liquor of goats.

The rumen NH_3-N levels were 36 mg per cent in group I and 27 mg per cent in group II of the present study. Broderick *et al.* (1993) reported NH_3-N concentration of 33.79, 31.0 and 28.67 mg per 100 ml in the rumen liquor when animals are fed 19 per cent CP ration. They also reported NH_3-N levels of 22.9, 17.7 and 18.7 mg per 100 ml when 16 per cent CP rations were given at 29, 41 and 17 per cent UDP levels. Broderick *et al.* (1993) also reported rumen NH_3-N level of 20 mg per 100 ml when 16 per cent CP rations were given to dairy cows. The rumen NH_3-N level in group I was significantly higher ($P<0.05$) than that of the second groupof the present study. This may be because of the higher degradability of the concentrate mixture in the first group, which had a UDP level of 26.6 per cent of CP of concentrate mixture compared to a UDP level of 42.9 per cent in the second group. Increased rumen NH_3-N with increased degradability of protein was reported by Carroll *et al.* (1988), Scott *et al.* (1991), Shabi *et al.* (1998), Casper *et al.* (1999) and Devant *et al.* (2000). On the contrary, Sahlu *et al.* (1984) and Janicki *et al.* (1985) reported that the UDP level of the ration had no significant effect on the rumen NH_3-N levels.

Sampath (1985) reported that when 17 percent CP concentrate mixture was fed at 77, 65 and 45 per cent RDP, there was no significant difference in the rumen pH (6.6, 6.44, 6.48) and TVFA (73, 71, 83 meq/l), but the rumen NH_3-N was significantly higher in the animals fed 77 per cent RDP ration (16.07, 8.9 and 3.94 mg /100ml). Kalbande (1995) reported values which ranged between 6.43 to 6.8

for pH, 8 to 16.3 mg percent for NH_3-N and 84 to 101 meq\1000 ml of TVFA in the rumen liquor of Jersey crossbred cows fed 63, 48 and 30 per cent UDP rations.

5.2.7 Haematological Parameters.

The data on the haematological parameters presented in the Table 24 are within the range specific to the species. The initial values of haemoglobin, blood glucose, BUN, plasma calcium and plasma phosphorus were 11.56 g per cent, 47.02 mg per cent, 20.98 mg per cent, 10.86 mg per cent and 9.83 mg per cent, respectively. There was no significant effect of the UDP levels of the ration on the haematological parameters except for blood glucose levels measured on the 50th day collection which was higher ($P<0.01$) in animals of group II than that of group I. However there was no difference in glucose level on day 98 between the two groups. Grubic (1991) and Palmquist *et al.* (1993) reported that UDP level of the ration had no effect on the blood glucose levels.

The haemoglobin levels were 10 and 9.69 g per cent respectively for the 50th and 98th day collections in first group while the values were 10.58 and 10.00 g per cent in second group.

Blood urea nitrogen levels were 44 and 43 mg per cent for first group and 39.6 and 35 mg per cent for second group for the two collections and the values were higher than that of phase I experiment. The NH_3-N levels are affected by the collection time and according to Howard *et al.* (1987) the highest level of PUN could be observed within 2.5 to 3 hours after feeding. Even though there was a significant effect of UDP level on rumen NH_3-N in the present study, there was no significant difference on BUN levels of animals fed rations with two UDP levels. Higginbotham *et al.* (1989) and Grubic *et al.* (1991) also could not observe any effect on BUN levels by feeding rations with different UDP levels. Rodriguz *et al.*

(1997) and Moscardini *et al.* (1998) observed an increase in the PUN level corresponding to an increase in the UDP levels of the ration.

The plasma calcium levels were 10.86 and 9.80 mg /100 ml in the first group for the 50th and 98th day collections, respectively, and the corresponding levels in the second group were 9.8 and 11.08 mg per cent. The plasma phosphorus levels obtained in the present study were 6.5 and 7.74 mg per cent in the first group and 6.24 and 6.79 mg per cent in the second group, respectively, for the 50th and 98th day collections.

5.2.8 Digestibility Coefficient of Nutrients

The digestibility coefficient of nutrients such as DM, CP, CF, NDF, ADF, EE and NFE are given in Table 26. There was no significant effect of the UDP level of the concentrate mixture on the digestibility of nutrients, except for the digestibility of EE, which was significantly higher ($P<0.01$) in animals of group II. Crooker *et al.* (1983) reported a low CP digestibility in cows given formaldehyde treated SBM, while Atwal *et al.* (1995) observed a significantly low digestibility coefficients of CP and ADF in cows given treated SBM compared to those given untreated SBM rations. King *et al.* (1990) did not observe any effect on the nitrogen metabolism of cows by the source or degradability of protein in the ration. Christensen *et al.* (1993b) reported that the digestibilities of DM, ADF and NDF and nitrogen metabolism in cows were not influenced by the amount of UDP (30 and 45 per cent) in the ration. Similar observations were also made by Kung *et al.* (1983), Santos *et al.* (1984), Waltz *et al.* (1989), Klusmeyer *et al.* (1990) and Herrera-Saldana *et al.* (1989). Palmquist *et al.* (1993) did not observe any significant change in the CP digestibility by the two levels of UDP in the ration (31 and 51), while Devant *et al.* (2000) reported better nitrogen digestibility when the degradability of protein in the ration was increased, irrespective of CP level.

5.2.9 Reproductive Parameters

The average number days to first insemination were 90.7 and 74.8. respectively in groups I and II. Three animals in the first group and two animals in the second group conceived during the experimental period. There was no significant effect by the UDP levels of the ration on the days to first heat or rate of conception. The blood urea nitrogen levels of the animals of both groups ranged between 35 to 43 mg per cent which was above the critical level of 20 mg per cent reported by Ferguson *et al.* (1993). Reduced fertility in case of cows with BUN levels above 20 mg per cent were also observed by other scientists (Jordan *et al.*, 1983; Johnson *et al.*, 1986; Canfield *et al.*, 1990; Erlod and Butler, 1993), while Howard *et al.* (1987), Carroll *et al.* (1988) and Carroll *et al.* (1994) opined that a PUN level above 20 mg per cent and variation in the degradability of protein in the ration had no effect on the fertility.

5.2.10 Cost of Production

A perusal of the data given in Table 27 reveals that the total DMI of the animals of the two groups fed 17 per cent CP concentrate mixture at two levels of protein degradabilities (26.8 and 42.9 per cent) were 6658 and 5562 kg, respectively. The DMI of the animals of group I was significantly higher ($P<0.05$) than that of the second group. The intake of DM per 100 kg body weight in the two groups were 3.49 and 2.97 kg. The DMI per kg metabolic live weight (12.5 kg) also was higher ($P<0.05$) in the first groupcompared to the second group (14.73 kg). Even though the DMI in the first group was higher than the second group, the feed intake per kg milk production was similar (1.20 and 1.21 kg).

The total milk production in 100 days were 5549.9 and 4623 kg, respectively, for animals of groups I and II. Although the total milk production was more in the first group, the covariance analysis of the data on milk production revealed that the milk production by animals of the second group fed 42.9 per cent UDP concentrate was maintained better than that of the animals of the first group. But the cost of production of one kg milk was 1.15 rupees higher in the second group than that of the first group.

From the overall results obtained in the present study it could be concluded that a level of 13.2 per cent of CP in the total ration was sufficient for meeting the protein requirements of early lactating crossbred cows producing around 10 kg per day. Increasing the UDP level from 26.6 to 42.9 per cent of the CP maintained production better, but cost of milk production was higher for the high UDP group indicating that it is not economical to provide high UDP ration for cows when the average daily production is around 10 kg

Summary

6. SUMMARY

A study was conducted in two phases to assess the influence of level and degradability of dietary protein on early lactation in crossbred cows.

6.1 PHASE I

Twelve crossbred cows in early lactation (within one month of lactation) were selected and grouped into two as uniformly as possible with respect to age, stage of lactation, parity, body weight and previous milk yield. The animals were fed with the experimental rations comprising of concentrate mixture having either 26 or 20 per cent CP on DM basis and paddy straw, as the sole roughage, in the ratio 70:30 of total DM. The two rations contained 20 and 15.41 per cent CP and 63.86 and 61.28 per cent TDN, respectively. Based on the observations made during the first month, third group with six early lactating cows was introduced and they were fed with concentrate mixture having 17 per cent CP and 72.5 per cent TDN, and paddy straw, which gave 13.2 per cent CP and 62.7 per cent TDN in the total ration.

The experiment was carried out for a period of 100 days and the animals were maintained under similar managemnetal conditions. Individual records of daily intakes of concentrate and paddy straw, daily milk production, fortnightly body weight and milk composition (fat, total solid, SNF and protein) were maintained throughout the experiment. Blood samples were collected before the beginning of the feeding trial and subsequently on the 50^{th} and 98^{th} day for assessing the levels of haemoglobin, blood glucose, blood urea nitrogen (BUN), plasma calcium and plasma phosphorus. Rumen liquor was collected before the beginning and towards the end of the experiment and rumen fermentation parameters such as pH, rumen ammonia nitrogen (NH_3-N), total volatile fatty acids (TVFA) and lactic acid levels were assessed. A digestibility trial was conducted during the 15th week of the experiment with a collection period of seven days. Data on the reproductive performance of the animals during the experimental period were also collected. The data gathered during the course of the experiment were statistically analysed.

There was no significant difference in the body weight of animals recorded throughout the experimental period except during the fourth and sixth fortnight when the animals of group II registered significantly higher ($P<0.05$) body weights than those of the first and second group. The average daily DMI of animals in the three groups were 2.99. 3.54 and 3.79 per cent of the body weight and 12.67. 15.37 and 16.08 kg per kg metabolic live weight. respectively. Average daily dry matter intake was significantly lower ($P<0.01$) in the first group than that of the second and third group. There was no significant difference between the second and third group regarding DMI.

Crude protein levels (20. 15.41 and 13.2 per cent) of the ration did not affect the milk production or the levels of milk constituents. The average milk composition was 3.48. 3.51 and 3.48 per cent fat. 11.81. 11.87 and 12.01 per cent total solids. 8.33. 8.36 and 8.53 per cent SNF and 2.59. 2.54 and 2.52 per cent protein. respectively. for the animals of groups I. II and III.

The rumen parameters estimated on the 92nd day of the experiment were pH. 7.71. 7.01 and 7.37. TVFA. 93.81. 68.71 and 86.5 meq/l. NH_3-N. 39. 31 and 17 mg per cent and lactic acid 0.33. 0.21and 0.22 mg /ml. The rumen NH_3-N levels were significantly higher ($P<0.05$) in the first and the second groups than that of the third group. The lactic acid and pH levels in the first group were significantly higher ($P<0.05$) than those of second and third groups.

All the estimated blood parameters were within the normal range for the species. Blood parameters such as haemoglobin. blood glucose. plasma calcium and plasma phosphorus were not affected by the level of CP in the ration used in the present study. while the BUN level was significantly higher ($P<0.01$) in the second group than the third group on the 50^{th} day collection. But there was no significant effect on the BUN levels on the 98^{th} day collection.

The digestibility coefficients of the nutrients such as ether extract (EE), crude fibre (CF), neutral detergent fibre (NDF) and acid detergent fibre (ADF) were significantly higher ($P<0.01$) in the first group. All the animals in the second group and only one each in the first and third groups conceived during the experimental period . The average days to first AI in the animals were 122, 79.4 and 80, respectively in the three groups, and the difference between groups was nonsignificant. The cost of producing one kg milk for animals in the three groups fed 20, 15 and 13 per cent CP rations was Rs. 7.79, 7.91 and 7.71, respectively.

From the results obtained in the first phase of study, it was found that the animals in the third group maintained their body weight and production through out the experimental period in a better manner than those of other two groups. The cost of producing one kg milk was also lower for the third group indicating that 13.2 per cent CP in the ration was sufficient to meet the requirements of cows in early lactation, producing a peak of around 10 kg milk

6.2 PHASE II

In this phase 12 crossbred cows in early lactation (within one month of calving) were selected and grouped into two as in the first phase. The animals were fed 17 per cent CP concentrate mixture with either of the two levels of protein degradability (26.8 and 42.9 per cent of CP) and paddy straw as the roughage in 70:30 ratio. The crude protein and TDN content of the two rations were 13.66 and 13.68 and 64.33 and 66.98 per cent, respectively. The experiment was conducted for 100 days in the same manner as to that of phase I with regard to the feeding, management, observations made and materials collected for analysis. In addition to the parameters estimated in the first phase, milk urea nitrogen also was estimated. A digestion trial for seven days, as in first phase, was carried out towards the end of experimental period.

There was no significant difference in the body weight of animals between the two groups during the course of the study. Dry matter intake as per cent of body weight (3.49 and 2.97) as well as the DMI per kg metabolic live weight (14.73 and 12.5) were higher ($P<0.05$) in the first group than that of the second group. The average daily DMI was also significantly higher ($P<0.01$) in the first group from the third week of experimental period.

The milk production of the animals of the second group was better than that of the first group during the seventh, eighth and thirteenth week ($P<0.05$), ninth, eleventh and twelfth ($P=0.06$) and during second and fourteenth week ($P<0.01$). There was no significant difference in the milk constituents between the two groups except for the fat and total solids which were higher ($P<0.01$) for the first group. The milk protein levels in the two groups were 2.88 and 2.82 g per cent and the milk urea nitrogen levels were 47.67 and 49.29 mg per cent, respectively.

There was no significant difference between the groups with regard to the rumen pH (6.42 and 6.57), TVFA (76.10 and 77.60 meq/l) or lactic acid levels (0.21 and 0.10 mg per ml, respectively for group I and II), while the rumen NH_3-N level in the first group (36.42 mg per cent) was significantly higher than that of the second group (27.27 mg per cent).

Except for a significantly higher ($P<0.01$) blood glucose level in the first group (43.57 Vs 53.57 mg per cent) on the 50th day collection, there was no significant difference in any of the blood parameters studied between the two groups.

The digestibility coefficient of all nutrients except that of CP of the second ration was numerically higher than those of the first ration and the EE digestibility in the second group being significantly higher (P<0.01).

The reproductive performance of the animals was not affected by the UDP level of the ration, since three animals of the first group and two animals of the second group conceived during the experimental period. However, the average number of days from calving to first service was numerically lower for the animals of second group (90.7 and 74.8 days for animals of group I and II, respectively).

The cost of production of one kg milk in the two groups was Rs. 6.33 and 7.36, respectively. From the evaluation of the results, it can be concluded that, even though the milk production in the second group was better maintained during the experimental period, the cost of production was higher, indicating that a level of 42.9 per cent UDP in the concentrate mixture is not economical in dairy cows producing around 10 kg milk as peak production.

From the overall assessment of the results obtained in the present study, it could be concluded that a level of 13.2 per cent of CP in the total ration was sufficient for meeting the protein requirements of early lactating crossbred cows producing around 10 kg per day. Increasing the UDP level from 26.6 to 42.9 per cent of the CP maintained production better, but cost of milk production was higher indicating that it is not economical to provide high UDP ration for cows when the average daily production is around 10 kg . It is also indicated from the present study, that RDP sources particularly NPN substances like urea can be incorporated economically for these cows.

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INFLUENCE OF LEVEL AND DEGRADABILITY OF DIETARY PROTEIN ON EARLY LACTATION IN CROSSBRED COWS

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ABSTRACT OF A THESIS

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ABSTRACT

A study was conducted in two phases to assess the influence of level and degradability of dietary protein on early lactation in crossbred cows. In phase I. eighteeen crossbred cows in early lactation (within one month of lactation) were selected and grouped into three as uniformly as possible with regard to age. sex. parity. previous production and stage of lactation and were fed with the experimental rations comprising of concentrate mixtures having 26. 20 and 17 per cent CP on DM basis and paddy straw, which were fed in the ratio 70:30 of total DM. The three rations contained 20, 15.41 and 13.2 per cent CP and 63.86, 61.28 and 62.5 per cent TDN on DM, respectively. Individual records of daily intakes of concentrate and paddy straw. daily milk production. fortnightly data on body weight and milk composition were maintained throughout the experimental period of 100 days. Blood samples were collected for assessing levels of haemoglobin. blood glucose. blood urea nitrogen (BUN). plasma calcium and plasma phosphorus. Rumen liquor samples were collected for assessing pH. rumen ammonia nitrogen (NH_3-N). total volatile fatty acid (TVFA) and lactic acid contents. A digestibility trial was conducted towards the end of experiment and reproductive performance of the animals during the experimental period was also assessed.

The average dry matter intake (DMI) of animals in the first group (2.99 per cent of body weight) was lower ($P<0.05$) than those of group II and III (3.54 and 3.79 per cent of the body weight. respectively). The body weight of animals of the three groups were similar. except in the fourth and sixth fortnight. when the weight of animals in the second group was significantly higher ($P<0.01$) than those of first and third. There were no significant differences in the milk production or composition between the groups during the course of study. except during the fifth fortnight. milk fat content of group III was lower ($P<0.05$) than that of groups I and II. The pH and lactic acid contents of the rumen liquor

collected from animals of group I were higher ($P<0.05$) than those of groups II and III. while the rumen NH_3-N concentration was lower ($P<0.05$) for the third group than that of the first and second groups. There was no significant difference on the blood parameters such as haemoglobin. blood glucose. plasma calcium and plasma phosphorus. between the groups while the blood urea nitrogen (BUN) level in the 50th day collection was significantly higher ($P<0.01$) in the second group than that of the third group. The digestibility coefficient of the nutrients such as ether extract (EE). crude fibre (CF). neutral detergent fibre (NDF) and acid detergent fibre (ADF) were significantly higher ($P<0.01$) in the first group. All the animals in the second group and one each in the first and third group conceived during the experimental period. The average number of days from calving to first AI of the animals of the three groups were 122. 79.4 and 80. respectively. the difference being nonsignificant. The cost of producing one kg milk in the three groups fed 20. 15.41 and 13.2 per cent CP rations was Rs. 7.79. 7.91 and 7.71. respectively. Results of the study suggest that CP level of 13.2 per cent in the total ration was sufficient to meet the requirements of cows in early lactation. producing a peak of 10 kg milk.

During phase II. 12 crossbred cows in early lactation (within one month of lactation) were selected and grouped into two as uniformly as possible as done in phase I. They were fed 17 per cent CP concentrate mixture with two levels of undegradable protein (UDP) (26.8 and 42.9 per cent of CP) and paddy straw as the roughage in 70:30 ratio. The rations had 64.33 and 66.98 per cent TDN respectively on DM basis. The experiment was conducted similar to that of phase I for a period of 100 days. In addition to the parameters estimated in the first phase. milk urea nitrogen also was estimated.

There was no significant difference in the body weight of animals of the two groups during the experiment. The weekly average DMI was significantly higher ($P< 0.01$) in the first group from the third week of experimental period. The milk production of the animals of the two groups was significantly different during the second and fourteenth week ($P<0.01$). during the seventh. eighth and thirteenth week ($P<0.05$) and during the ninth. eleventh and twelfth week ($P=0.06$). There was no significant difference in the milk constituents between the two groups except for the fat and total solids which were higher ($P<0.01$) for the first group. There was no significant difference in the protein and urea nitrogen levels in the milk. or any of the rumen parameters studied except for the rumen NH_3-N level which was significantly higher ($P<0.01$) in group I. Except for a significantly higher ($P<0.01$) blood glucose level in the first group on the 50th day collection. there was no significant difference in the blood parameters studied between the groups. The digestibility coefficient of the nutrients of the two rations were not different. except EE. which was higher ($P<0.01$) for the second ration. The reproductive performance of the animals were not affected by the UDP level of the ration since three animals of the first group and two animals of first group conceived during the experimental period. However the average days from calving to first service was numerically lower. though not significant. for the animals of the second group (90.7 in the first group and 74.8 in the second). The cost of production of one kg milk in the two groups was Rs. 6.33 and 7.36. respectively. The ration containing higher UDP (42.9 per cent of CP of concentrate mixture) maintained the milk production better than the low UDP ration. but the cost of production was also higher for high UDP fed group.

From the overall results obtained in the present study it could be concluded that a level of 13.2 per cent of CP in the total ration was sufficient for meeting the protein requirements of early lactating crossbred cows producing around 10 kg per day. Increasing the UDP level from 26.6 to 42.9 per cent of the CP maintained production better. but cost of milk production was higher indicating that it is not economical to provide high UDP ration for cows when the average daily production is around 10 kg