

Reproduction-Nutrition Relationship in Dairy Buffaloes. I. Effect of Intake of Protein, Energy and Blood Metabolites Levels

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ABSTRACT : Fifty one Nili-Ravi dairy buffaloes in their last two months of gestation were selected. After parturition, rectal examination of reproductive organs was carried out until the occurrence of the first oestrus (PEI). Milk samples were analyzed for milk progesterone levels (MPL). Ovulation (POI) was confirmed by rectal palpation and MPL. Feed and blood samples were collected fortnightly and analyzed. Body condition score (BCS) was recorded on a scale of 0 to 5. Crude protein (CP) intake varied among different seasons and correlated positively with serum urea levels, POI ($p < 0.01$) and PEI ($p < 0.05$). Excess CPI was lower in the group showing oestrus as compared to those remaining as anoestrus ($p < 0.05$). The dietary ratio of crude protein - metabolizable energy (CP:ME) in the oestrus animals was narrow and constant, while the anoestrus animals had a widely fluctuating one. In normal breeding season (NBS) calvers, mean serum urea level (SUL) was lower than the low breeding season (LBS) calvers. SUL was positively correlated with PEI and POI ($p < 0.01$). Up to six months postpartum, SUL were constantly higher in anoestrus than oestrus buffaloes. Mean metabolizable energy (ME) intake was lower in the NBS calvers than the LBS calvers ($p < 0.01$). BCS and postpartum ovulation interval were correlated with ME intake ($p < 0.01$). Prepartum ME intake was higher in oestrous as compared to anoestrous animals ($p < 0.05$). Higher and lower ME intakes were associated with anoestrus, while a moderate energy intake was associated with a PEI of less than 75 days. Buffaloes with poor BCS belonged to the LBS calving group and most of the NBS calving buffaloes had good BCS. BCS was negatively correlated with PEI ($p < 0.01$) and was higher in oestrous buffaloes than anoestrus. It was concluded that excess intake of crude protein, associated with higher serum urea levels and low energy intake, associated with poor body condition, are the key factors for low reproductive efficiency. It may be corrected by adopting a proper feeding strategy. (*Asian-Aust. J. Anim. Sci.* 2002. Vol 15, No. 3 : 330-339)

Key Words : Buffalo, Nutrition, Reproduction, Protein, Metabolizable Energy, Urea

INTRODUCTION

In the North-West Frontier Province (NWFP) of Pakistan and elsewhere in the Indo-Pakistan sub-continent, buffalo farming is practiced on non-scientific lines and there is no tradition of consulting animal health, reproduction or nutritional experts in identifying or addressing the relevant problems. The socio-economic status of the peri-urban dairy farmers is usually low, getting negligible inputs from livestock, financial or marketing institutions or experts. The major causes associated with the under-developed buffalo farms have been identified as: i) calf losses, irregular breeding, imbalanced feeding; ii) ungainly loans and; iii) a hostile marketing system. The three causes at commercial buffalo herds throughout Pakistan, lead to annual losses to the tune of Rs.1043.67 billions (US\$ 1=Rs.60) (Qureshi, 2000).

The normal breeding season in the NWFP begins in August and coincides with the feeding of non-leguminous fodders, such as sorghum and maize. Food proteins are

hydrolyzed to peptides and amino acids by rumen microorganisms but some amino acids are degraded to organic acids, ammonia and carbon dioxide (McDonald et al., 1984). Rumen degradable proteins are either converted by ruminal microflora, to microbial protein and digested in the small intestine; or they are hydrolyzed to ammonia and excreted as urea. Previously, no attempt has been made to work out availability of nutrients in comparison to their production and maintenance requirement or their effect on reproductive performance.

Low-protein diets are associated with reduced intake of energy and protein in beef cattle (Wiltbank et al., 1965). Sasser et al. (1988) found that a deficiency of crude protein reduces pre- and postpartum weight gains, decreased the percentage of heifers showing oestrus by 110 days after calving (89% vs 63%), decreased the first service conception rate (71% vs 25%), and increased the postpartum oestrus interval (75 vs 86 days), in beef heifers.

Under the conventional farming system in the NWFP, diet is not formulated according to the requirements of individual animals, resulting in decreased production and poor health and reproduction (Qureshi, 1995). The lactating buffaloes are fed green fodders plus concentrate feeds but dry and pregnant buffaloes are considered uneconomical and are mostly fed only low quality green fodders. Consequently, animals which receive adequate nutrients

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have higher body condition scores, which enable them to produce higher quantities of milk and they also are bred earlier.

In the absence of any ration formulation practice in the province, excessive or deficient intake of some nutrients may decrease reproductive performance. The present project was, therefore, designed to study the association of intake of protein and energy and the resulting serum urea levels and body condition, score with reproductive performance in Nili-Ravi buffaloes, under field conditions in the NWFP.

MATERIALS AND METHODS

Animal selection and body condition scoring

Fifty one Nili-Ravi dairy buffaloes, in their last two months of gestation, were selected and tagged at seven peri-urban commercial farms, located in the Central Valley of the NWFP at 31 to 37°N and 65 to 74°E. The farms were visited twice a week. Reproductive history of the selected animals was recorded. Body condition score (BCS) was recorded at weekly intervals, using the method described for dairy and beef cows (scale 0 to 5, Peters and Ball, 1987). In this method, the thickness of fat over the lumber region and tail head area was estimated and assigned a score from 0 (emaciated) to 5 (very fat). Study on the animals prevailed over a period of 18 months (March, 1994 to August, 1995).

Clinical monitoring

After parturition, rectal examination of reproductive organs was carried out on days 14 and 21, and then fortnightly, until the occurrence of the first oestrus as described by Usmani et al. (1985). The position of the reproductive organs and the approximate size of the cervix, uterus and ovaries and the ovarian structures, were recorded. Oestrus detection was undertaken twice daily, from 15 days postpartum until resumption of oestrus. In addition to visual signs, an intact bull was used for heat detection at each farm. Ovulation was confirmed by palpation of an ovulation depression, a very soft corpus luteum or luteal tissue embedded in the ovary and milk progesterone levels (MPL).

Milk sampling and progesterone assay

Milk samples were collected weekly from each buffalo. The fat layer was removed and 100 microliters of 0.1% sodium azide solution was added to a 5 ml of milk sample as a preservative. Samples were stored at -20°C until analysis for milk progesterone levels (MPL) using radio-immunoassay (RIA) (FAO/IAEA, 1993). Intra- and inter-assay coefficients of variation were 3.54% and 9.21% respectively. Sensitivity (detection limit) of the assay was 0.09 ng/ml.

Feed and blood analysis and animal feed requirements

Feed ingredients were sampled at fortnight intervals. A portion of each samples was used for dry matter determination and the remaining portion was air dried at 60°C for 48 h, ground through a 1 mm sieve in a Wiley mill and stored for further analysis. The samples were analyzed for ash, crude protein, crude fiber and ether extract, according to AOAC (1980). Intra-ruminal protein degradability of feed samples was determined using the *in sacco* technique similar to that described by Ørskov et al. (1980). The digestibilities of fodders were calculated using the digestibility coefficients reported by Ishaque and Malik (1972) for Nili-Ravi buffaloes. Total digestible nutrients values of feed were estimated using the equations of Choo (1982). TDN, digestible energy (DE) and metabolizable energy (ME) of green fodders were calculated by the method of Crampton et al. (1957), Swift (1957) and Moe and Tyrell (1976).

The digestible protein requirements for maintenance were calculated through the equation of Kearn (1982) ($2.54 \text{ g/kg B.Wt}^{0.75}$) and the requirement for milk production was assumed to be 126 g/100 g of protein secreted in milk (Ranjhan, 1980). The protein content of Nili-Ravi buffalo milk was assumed to be 4.93% (Makhdam, 1983).

For calculating energy requirement of buffalo, the following values were used (1 cal=4.185 J):

- a) For maintenance, the value of 0.523 MJ/kg B.Wt^{0.75}, used by Sen et al. (1978) and followed by Kearn (1982), was adopted.
- b) For milk production, a value of 4.972 MJ per kg 4% fat corrected milk (FCM), as recommended by Ranjhan and Pattak (1979), was used.

Jugular blood samples were collected and analyzed for urea concentrations using Clonital kits (Sede amministrativa e produttiva 24030-Carvico (BG), Italy).

Statistical analysis

The data obtained were statistically analyzed using general linear model procedures and correlation analysis (Steel and Torrie, 1980). The following model was used:

1. For comparing the effect of calving period and seasons on intake of protein and serum urea levels (dependant variables)

$$Y_{ijk} = u + \alpha_i + \beta_j + e_{ijk}$$

Where Y_{ijk} =kth observation of the dependent variable during ith period of calving and jth season

u =Population constant common to all records

α_i =The effect of ith period of calving; i =normal breeding season (NBS) and low breeding season (LBS)

β_j =The effect of jth season; j =autumn, winter, spring and summer

e_{ijk} =The random residual term associated with each y_{ijk}

2. For comparing the difference in reproductive states

due to intake of protein and serum urea levels (dependant variables)

$$Y_{ijkl} = u + \hat{\alpha}_i + \hat{\beta}_j + \hat{\alpha}k + e_{ijkl}$$

Where Y_{ijkl} = lth observation of the dependent variable during ith postpartum ovulation interval, jth postpartum estrus interval and kth pregnancy status

u = Population constant common to all records

$\hat{\alpha}_i$ = The effect of ith postpartum ovulation interval; $i = <45, 45-74$ and $75-150$ days and anoestrus

$\hat{\beta}_j$ = The effect of jth postpartum oestrous interval; $j = <45, 45-74, 75-150$ days and anoestrus

$\hat{\alpha}k$ = The effect of kth pregnancy status; $k =$ pregnant and non-pregnant

e_{ijkl} = The random residual term associated with each y_{ijk}

3. For comparing the effect of calving period and seasons on intake of energy and body condition score (dependant variables)

$$Y_{ijk} = u + \hat{\alpha}_i + \hat{\alpha}j + e_{ijk}$$

Where Y_{ijk} = kth observation of the dependent variable during ith period of calving and jth season

u = Population constant common to all records

$\hat{\alpha}_i$ = The effect of ith period of calving; $i =$ NBS and LBS

$\hat{\alpha}j$ = The effect of jth season; $j =$ autumn, winter, spring and summer

e_{ijk} = The random residual term associated with each Y_{ijk}

4. For comparing the difference in reproductive states with intake of energy and body condition score (dependant variables)

$$Y_{ijkl} = u + \hat{\alpha}_i + \hat{\alpha}j + \hat{\alpha}k + e_{ijkl}$$

Where Y_{ijkl} = lth observation of the dependent variable during ith postpartum ovulation interval, jth postpartum

oestrus interval and kth pregnancy

u = Population constant common to all records

$\hat{\alpha}_i$ = The effect of ith postpartum ovulation interval; $i = <45, 45-74$ and $75-150$ days and anoestrus

$\hat{\alpha}j$ = The effect of jth postpartum oestrus interval; $j = <45, 45-74, 75-150$ days and anoestrus

$\hat{\alpha}k$ = The effect of kth pregnancy status; $\hat{\alpha} =$ pregnant and non-pregnant

e_{ijkl} = The random residual term associated with each Y_{ijk}

RESULTS

Feed composition

Feed components of the experimental animals comprised green fodders, concentrates and wheat straw with the composition reflected in table 1. Green fodder during the winter and spring comprised Egyptian clover with higher crude protein and moisture. During autumn and early winter, sorghum and maize were available, having higher levels of dry matter and relatively low crude protein. During late winter and spring wheat crop was fed while chopped sugar cane was fed during winter and spring. Wheat straw or dried maize stovers were fed through out the year with an increasing levels during winter and early summer. Concentrate feeds comprised wheat bran, cotton seed cake (undecorticated), mustard seed cake, maize oil cake, commercial concentrates, dried bread, wheat grain, beet pulp and molasses and were fed to lactating buffaloes at the same scale, irrespective of milk production, as per conventional practice.

Table 1. Nutrient composition of feeds used for experimental buffaloes

Feed	Months	Dry matter	AS % in dry matter		Metabolizable energy (Mcal/kg DM)
			Minerals	Crude protein	
Green fodders					
Berseem	Nov-May	14.43	12.94	21.03	2.67
Sorghum	Jul-Nov	34.35	8.82	6.09	3.06
Maize	Jun-Nov	30.00	8.91	7.51	2.95
Wheat	Jan-May	28.05	8.01	8.41	2.65
Dry roughage					
Maize stovers	Jan-Dec	95.09	6.04	3.72	1.73
Wheat straw	Jan-Dec	93.74	9.93	4.21	1.58
Concentrate supplements					
Wheat bran	Jan-Dec	90.44	3.87	17.13	3.68
Cotton seed cake	Jan-Dec	91.88	5.46	24.97	5.44
Mustard seed cake	Jan-Dec	92.01	6.79	31.61	2.93
Maize oil cake	Jan-Dec	95.38	1.94	20.50	2.72
Commercial conc.	Jan-Dec	92.60	6.68	15.66	2.98
Dried bread	Jan-Dec	82.66	2.49	20.39	4.57
Wheat grain	Jan-Dec	93.00	1.32	13.00	4.78
Beet pulp dried	Jan-Dec	95.06	4.49	11.96	2.31
Molasses	Jan-Dec	71.92	13.6	8.99	1.81

Protein intake

Crude protein intake (CPI) showed a mean value of 1.82 ± 0.47 kg/day, ranging from 0.95 to 2.64 kg/day (table 1). CPI varied between seasons ($p < 0.01$, table 3) and was positively correlated with serum urea levels ($r = 0.22$, $p < 0.01$, figure 1). Degradable protein intake (DPI) was 1.32 ± 0.01 kg/day and was significantly affected by season ($p < 0.01$) with summer > spring > autumn/winter.

CPI was positively correlated with the duration of placenta expulsion ($r = 0.21$, $p < 0.01$), postpartum estrus interval (PEI) ($r = 0.08$, $p < 0.05$) and postpartum ovulation interval (POI) ($r = 0.21$, $p < 0.01$, table 4). CPI excess to requirements was lower in animals which expressed oestrus than those which remained anoestrus ($p < 0.05$, figure 2). The difference was marked from one month prepartum to four months postpartum. Excess CPI was positively related to the duration of placental expulsion ($r = 0.37$, $p < 0.01$). The dietary ratio of crude protein-metabolizable energy (g CP/MJ ME) consumed by the buffaloes during the

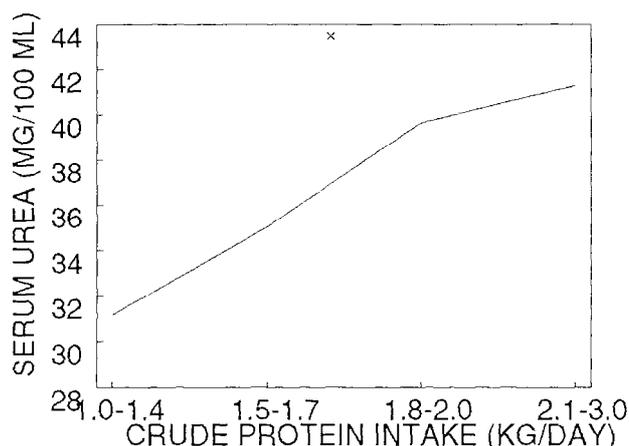


Figure 1. Relationship of crude protein intake (CPI) and serum urea levels (SUL) in buffaloes

Table 2. Number of observations (n), means and standard error (SE) for various parameters in buffaloes

Parameter	n	Mean±SE
Crude protein (CP) (kg/day)	1029	1.82±0.47
CP above requirements (g/day)	1029	92.5±17.1
Degradable protein (kg/day)	1027	1.32±0.01
Metabolizable energy (ME) (MJ/day)	1027	174.5±1.3
ME above requirements (MJ/day)	1027	50.6±1.6
CP/ME ratio (g CP/MJ ME)	1027	12.0±0.1
Dry matter (kg/day)	1029	16.6±0.1
Mean body condition score	1023	2.72±0.02
Body condition score (prepartum)	51	2.77±0.10
Body condition score loss 30 days prepartum	51	0.25±0.08

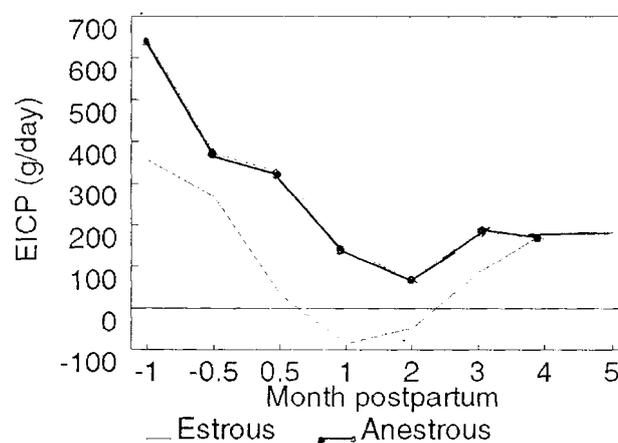


Figure 2. Excess intake of crude protein (EICP) in estrous and aneustrous buffaloes during various postpartum months (PMO)

Table 3. Effect of calving period and season on protein intake and serum urea concentration (least square means*±standard error)

Group	Crude protein (kg/day)	Degradable protein (kg/day)	CP/ME (g/MJ)	Serum urea conc. (mg/100 ml)
Calving periods				
NBS	1.81±0.02	1.32±0.02	12.2±0.07 ^a	31.7±1.43 ^b
LBS	1.83±0.02	1.34±0.02	11.8±0.10 ^b	39.4±21.6 ^a
Probability	NS	NS	$p < 0.01$	$p < 0.01$
Seasons				
Autumn	1.79±0.02 ^c	1.26±0.02 ^c	11.5±0.10 ^c	36.4±2.92 ^{ab}
Winter	1.68±0.02 ^d	1.22±0.02 ^c	12.0±0.09 ^b	29.9±1.96 ^b
Spring	1.89±0.03 ^b	1.42±0.02 ^b	12.2±0.09 ^b	38.6±2.95 ^a
Summer	2.08±0.05 ^a	1.55±0.04 ^a	12.7±0.19 ^a	40.1±2.53 ^a
Probability	$p < 0.01$	$p < 0.01$	$p < 0.01$	$p < 0.05$

* Means within a group in the same column with different letters differ significantly.

NBS: Normal breeding season, LBS: Low breeding season, NS: Non-significant.

Table 4. Correlation of various reproductive performance in buffaloes (Pearson's correlation coefficients¹).

Variate	Placenta expulsion duration	Lochia discharge duration	Postpartum interval	
			Ovulation	Oestrus
Nutrients intake				
Crude protein (CP)	0.21**	-0.14	0.21**	0.08*
Degradable protein	0.10*	0.22**	0.01	0.31**
CP above requirements	0.37**	0.06	-0.03	0.10**
Metabolizable energy (ME)	0.19*	0.28**	-0.03	0.03
ME above requirements	0.19*	0.24**	-0.27**	0.04
Other				
Serum urea	0.16	-0.18	0.30**	0.28**
Body condition score	-0.17	0.01	-0.06	-0.20**

¹The Pearson's product-movement correlation determined through correlation analysis.

* Significant at $p < 0.05$.

** Significant at $p < 0.01$.

prepartum and postpartum periods is given in figure 3. The comparison shows that the animals resuming to oestrus had a narrow and almost constant CP/ME ratio (11.9 to 12.2 g/MJ), while the anoestrous animals had a widely fluctuating ratio, ranging from 10.7 to 13.1 g/MJ. CP/ME was related positively with POI ($r=0.15$, $p < 0.01$, table 4).

Energy intake

The overall mean value of ME intake was 174.5 ± 1.1 MJ/day, ranging from 84.5 to 252.8 MJ/day (table 1). ME intake was lower in the NBS calvers than the LBS calvers ($p < 0.01$, table 6). ME intakes during winter and spring were similar and lower than those in summer while highest during autumn ($p < 0.01$). Increasing energy intake increased BCS ($r=0.16$) and duration of expulsion of placenta ($r=0.19$) and discharge of lochia ($r=0.24$) but decreased postpartum ovulation interval ($r=-0.27$, $p < 0.01$, table 4).

Prepartum ME intake was higher in animals observed in oestrous than those remaining anoestrous (177.2 vs 155.9 MJ/day, $p < 0.05$, already reported in Qureshi, 1998). Prepartum energy intake above requirement (MEAR) was also higher in animals returning to oestrus than the anoestrous ones ($p < 0.01$, figure 6). Higher MEAR during prepartum period were accompanied by a higher BCS in animals which came into oestrus (figure 7). The animals coming into estrus within 75 days postpartum showed a moderate intake of ME as compared to those coming into

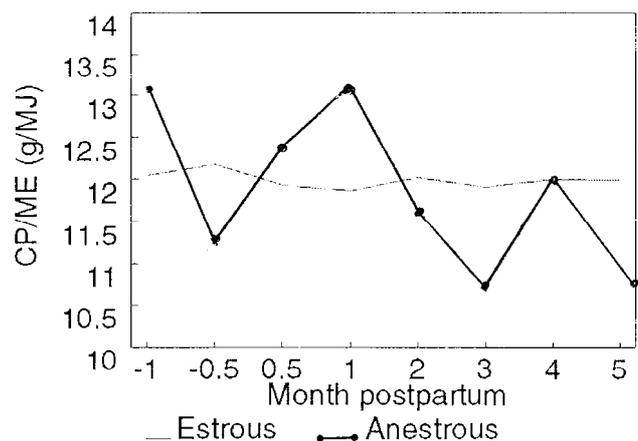


Figure 3. Ratio of crude protein/metabolizable energy intake (CP/ME) in estrous and anestrous buffaloes during various postpartum months (PMO)

estrus after 75 days postpartum, which showed either deficiency or excess of ME intake ($p < 0.01$, table 7).

Body condition score

The BCS of the 51 buffaloes varied from 1.0 to 4.0 during the late prepartum and early postpartum periods. The overall average BCS was 2.72 ± 0.02 (table 1). The buffaloes were grouped into three categories i.e. poor (BCS 1.0 to 2.0), moderate (BCS 2.5) and good (3.0-4.0). Figure 8 shows that non of the buffaloes in the NBS calving group had poor BCS (1.0 to 2.5). Conversely, none of the LBS calving buffaloes had good BCS (BCS > 3.0), which is consistent with the higher intake of metabolizable energy ($p < 0.01$) during summer and autumn (table 6).

BCS was significantly affected by the period of calving and season of the year ($p < 0.01$, table 6). Animals calving during the NBS, had significantly high BCS as compared to those calving during LBS (2.82 vs 2.60), which is the probable cause of low reproductive efficiency (animals coming into estrus within 45 days showed higher BCS than those coming after 45 days postpartum, table 7). BCS was highest (2.97) prepartum and lowest (2.65, $p < 0.01$) during the first two months postpartum. Placenta expulsion duration ($r=-0.17$, $p < 0.05$) and PEI ($r=-0.20$, $p < 0.01$, table 4) negatively correlated with BCS. Figure 7 shows that BCS in buffaloes resuming oestrus was constantly higher than those failing to resume oestrous activity.

Serum urea levels

In NBS calvers, serum urea levels were lower than those of the LBS calvers (31.69 vs 39.42 mg/100 ml, table 3).

Serum urea levels were positively correlated with PEI ($r=0.28$, $p < 0.01$) and POI ($r=0.30$, $p < 0.01$, table 4). Anoestrous animals had higher serum urea concentrations

Table 5. Protein intake and serum urea during various reproductive states (least square means*±SE)

Group	Crude protein (kg/day)	Degradable protein (kg/d)	CP/ME ratio** (g/MJ)	Serum urea (mg/100 ml)
Postpartum ovulation interval (days)				
<45	1.66±0.03b	1.18±0.04	11.9±0.11b	26.3±1.68
45 to 74	1.72±0.03b	1.23±0.03	11.9±0.13b	36.5±2.42
75 to 150	1.84±0.02a	1.35±0.03	12.5±0.07a	39.8±2.79
Anestrus	1.90±0.02a	1.35±0.02	11.9±0.10b	38.7±2.81
Probability	p<0.05	NS	p<0.01	NS
Postpartum estrus interval (days)				
<45	1.90±0.02a	1.39±0.03a	12.2±0.06a	31.0±1.89b
45 to 74	1.69±0.03b	1.22±0.02b	11.9±0.12b	31.7±1.93b
75 to 150	1.87±0.03a	1.36±0.03a	11.6±0.14b	33.0±2.80b
Anestrus	1.86±0.02a	1.37±0.02a	12.3±0.10a	46.0±1.83a
Probability	p<0.01	p<0.01	p<0.01	p<0.01
Pregnancy status				
Pregnant	1.70±0.02b	1.22±0.02b	11.9±0.08	33.2±1.68
Non-pregnant	1.94±0.02a	1.43±0.02a	12.1±0.08	37.4±2.02
Probability	p<0.01	p<0.01	NS	NS

* Means within a group in the same column with different letters differ significantly.

** Crude protein/metabolizable energy ratio.

Table 6. Effect of calving period and season on body condition score and metabolizable energy (ME) intake (least square means*±standard error)

Group	Body condition score (scale=0-5)	ME intake (MJ/day)	Crude protein/ME ratio (g/MJ)
Calving periods			
NBS**	2.82±0.02a	172.3±1.3b	12.15±0.07a
LBS***	2.60±0.03b	177.5±1.7a	11.78±0.10b
Seasons			
Autumn	2.91±0.03a	195.7±1.7a	11.47±0.10c
Winter	2.86±0.02a	163.8±1.8c	11.95±0.09b
Spring	2.61±0.03b	162.6±1.9c	12.19±0.09b
Summer	2.22±0.05c	177.0±2.6b	12.66±0.19a

* Means within a group in the same column with different letters, differ (p<0.01).

** Normal breeding season.

*** Low breeding season.

(46.0 mg/100 ml) than those resuming oestrus (table 5). From one month prepartum to six months postpartum, serum urea levels were repeatedly higher in anoestrus than oestrus buffaloes (figure 5).

DISCUSSION

The protein contents of the concentrate feeds were highly degradable. Figure 4 shows relationship of the degradable protein intake: 4% fat-corrected milk production ratio with conception rates. Conception rates decreased

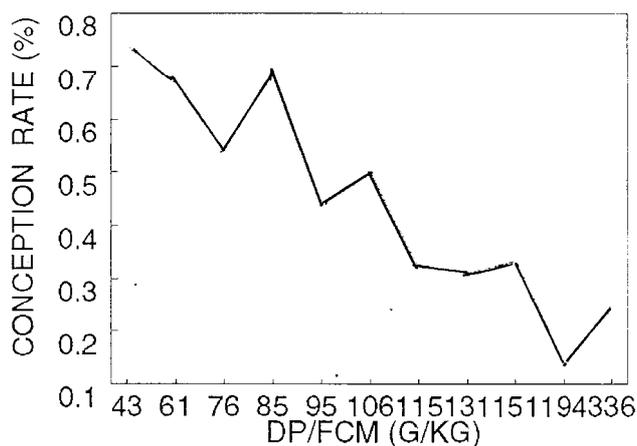


Figure 4. Relationship of degradable protein intake/4% fat-corrected milk production ratio (DP/FCM) with conception rate (CR%) in buffaloes

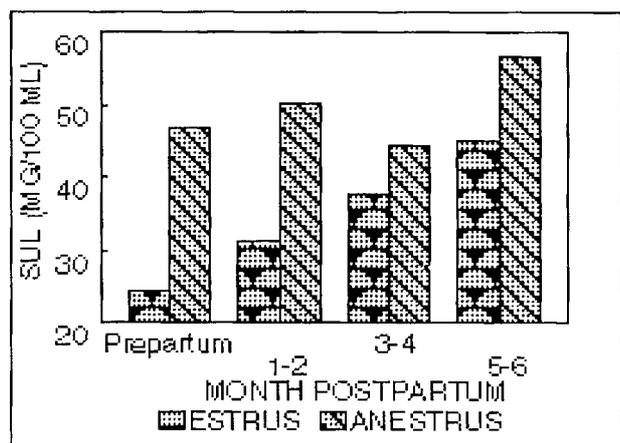
linearly as the ratio increased. Protein supplements were fed to lactating buffaloes irrespective of their milk production following the conventional practice on the farms in the study. Consequently, the low producers had excessive intake of dietary protein. The relationship in figure 4 demonstrates that such a practice of indiscriminate use of CP of higher degradability, adversely affects conception rates in low milk producing buffaloes. As milk production increases, more degradable protein is utilized reducing its detrimental effects, and resulting in enhanced conception rates.

Dairy animals in the Indo-Pakistan are mostly owned by

Table 7. Body condition score and metabolizable energy (ME) intake during various peripartum periods under various reproductive states (least square means* \pm standard error)

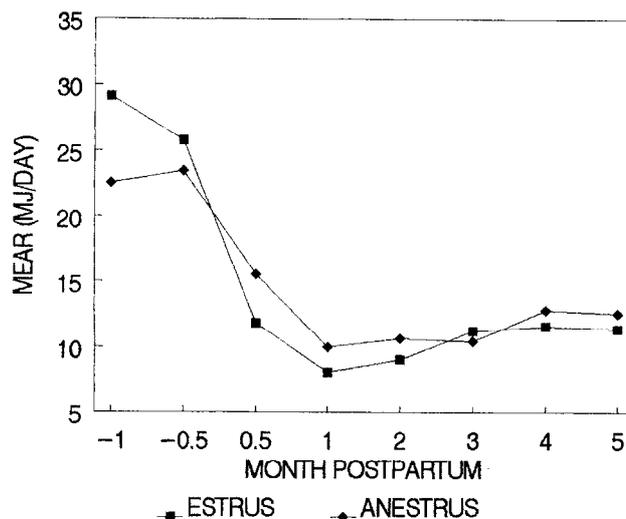
Group	Body condition score (scale=0-5)	ME intake (MJ/day)	Crude protein/ME ratio (g/MJ)
Postpartum ovulation interval (days)			
<45	2.91 \pm 0.03a	169.9 \pm 2.3bc	11.85 \pm 0.11b
45 to 74	2.70 \pm 0.05b	174.1 \pm 2.0ab	11.93 \pm 0.13b
75 to 150	2.58 \pm 0.02c	165.6 \pm 1.4c	12.51 \pm 0.07a
Anoestrus	2.74 \pm 0.02b	179.7 \pm 1.4a	11.85 \pm 0.10b
Probability	p<0.05	p<0.01	p<0.01
Postpartum oestrus interval (days)			
<45	2.91 \pm 0.02a	177.2 \pm 1.5b	12.24 \pm 0.06a
45 to 74	2.73 \pm 0.04b	166.8 \pm 2.5c	11.89 \pm 0.12b
75 to 150	2.58 \pm 0.04c	184.3 \pm 2.1a	11.60 \pm 0.14b
Anoestrus	2.66 \pm 0.03bc	171.4 \pm 1.6c	12.28 \pm 0.10a
Probability	p<0.01	p<0.01	p<0.01
Pregnancy status			
Pregnant	2.83 \pm 0.02a	167.3 \pm 1.5b	11.94 \pm 0.08
Non-pregnant	2.63 \pm 0.03b	180.8 \pm 1.5a	12.07 \pm 0.08
Probability	p<0.01	p<0.01	Non-significant

* Means within a group in the same column with different letters differ significantly.

**Figure 5.** Serum urea levels (SUL) in estrous and anestrus buffaloes during various postpartum months (PMO)

farmers of low socio-economic status and there is no system of ration formulation in relation to the animal's requirements for maintenance and milk production (Qureshi, 1995). The farmers try to promote higher milk production by feeding more concentrates. In such feeding systems, the low milk producers get more protein relative to their requirements, compared to the high producers. The higher intake of protein, mostly degradable, leads to formation of ammonia which is converted into urea in the liver which is an energy consuming process. It increases blood urea levels which adversely affects fertility, as already reported (Fergusson et al., 1991).

In agreement with the present study, Elrod and Butler (1993) reported detrimental effect of high CPI in cattle

**Figure 6.** Metabolizable energy intake above requirement (MEAR) in estrous and anestrus buffaloes

indicated by significantly lower first-service conception rates in Holstein heifers. In dairy cattle (Francos et al., 1992), feeding of diets higher in protein with low energy/protein ratio for lactation and maintenance, was a factor contributing to low fertility. Similarly, Jordan and Swanson (1979b) found that services per conception were increased in cows fed diets containing more than 16% crude protein. Detrimental effects of excess protein have been attributed to the ruminally degradable fraction of dietary protein (Folman et al., 1981; Canfield et al., 1990). Endocrine functions of the pituitary (LH secretion) and ovary (progesterone secretion) have been greatly elevated

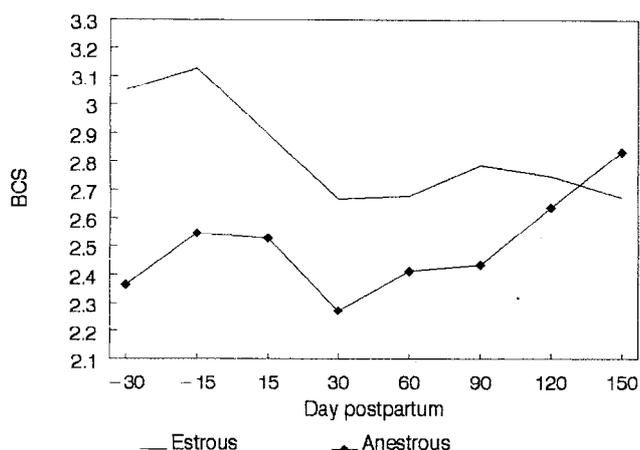


Figure 7. Postpartum body condition score (BCS) of estrous and anestrous buffaloes

and depressed, respectively, in cows consuming feed high in protein (Jordan and Swanson, 1979a).

The higher protein intake was associated with higher blood urea. The results also demonstrate a negative effect of high serum urea levels on reproductive performance in buffaloes. The adverse effects of higher serum urea levels were perhaps due to altered uterine pH and ovarian function. Results of the present study support the findings of Fergusson et al. (1991) who suggested that serum urea nitrogen concentration >20 mg/100 ml, would impair fertility in dairy cattle. Similarly, Kaim et al. (1983) reported that in cows with plasma urea nitrogen of 16.8 mg/100 ml, pregnancy rate was lower than in cows with plasma urea nitrogen of 9.0 mg/100 ml. However, according to Zaman (1984), the difference in urea levels of cyclic, non-cyclic and anoestrus Nili-Ravi buffaloes was not significant.

The high prepartum excess ME intake in the oestrus group probably resulted in the accumulation of more body reserves, which supported high postpartum milk production and early resumption of oestrus activity. In the last month prepartum, the ME intake was sufficient to support fetal growth (tissues and fluids). The present findings suggest that the animals receiving more ME above requirements during prepartum period were able to maintain a relatively good BCS despite mobilization of body reserves.

Increased energy intake were observed to enhance conception rates and the percentage of cows, cycling early in the breeding season (Wiltbank et al., 1964; Dunn et al., 1969), in agreement to our findings. While low energy intake led to ovarian inactivity and anoestrus in suckled beef cows. It was shown that increasing the dietary energy intake during gestation resulted in shortening of the anovulatory period in primiparous beef heifers (Echternkamp et al., 1982). Miettinen (1990) reported that

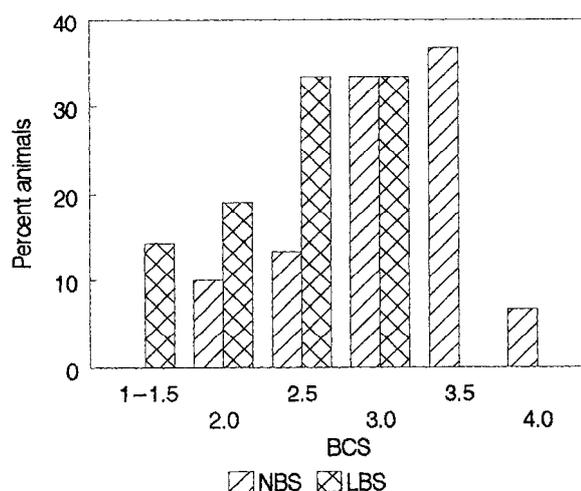


Figure 8. Distribution of buffaloes (%) on the basis of body condition score (BCS) during normal (NBS) and low breeding season (LBS)

in dairy cows, a low energy level in early puerperium delayed uterine involution and onset of ovarian activity and prolonged the PEI and service period. Marston et al. (1995) concluded that conception rates were significantly improved by feeding higher levels of supplemental energy prepartum, but not postpartum. Wongsrikeao and Taesakul (1984) reported that improved nutrition reduced the postpartum service period in Swamp buffalo cows and increased the growth rate of their calves.

The results of this study confirm the findings of Osoro and Wright (1992) and De Rouen et al. (1994), who concluded that BCS at calving significantly affected postpartum reproductive performance in cows. O'Rourke et al. (1991) reported that cows with BCS of ≥ 8 had a conception rate 33% higher than those with score ≤ 5 (scale 3-9). Buffaloes in poor BCS had inactive ovaries and postpartum anoestrus periods (Jainudeen and Wahab, 1987). In line with this study, Bhalaru et al. (1987) reported that conception rates were significantly higher (88.3%) for buffaloes with moderate BCS (2.5 to 3.5) than for females scoring 1 to 2 (65.8%) or 4 to 5 (70.8%). The reason for low reproductive performance in the animals with low BCS was perhaps the non-availability of nutrients for reproduction, being the third candidate in partitioning of nutrients, after health and milk production. In the fat animals the low reproductive performance was perhaps due to abnormal physiological functions during the estrous cycle.

The interval to first postpartum oestrus was significantly shorter in moderate and fat females (66.2 and 66.9 days, respectively) than in thin females (77.7 days). The service period of moderate BCS females was significantly shorter (128.3 days) than fat or thin females (144.1 and 164.5 days,

respectively). In the fat animals the delayed breeding was probably due to hinderance in transportation of gametes in the reproductive tract. Dunn and Kaltenbach (1980) found that in cows with moderate BCS, a prepartum gain in weight resulted in 25% better cyclicity than those losing weight. In cows with poor BCS, the cows gaining weight showed 45% higher cyclicity.

CONCLUSION

Excess intake of crude protein, associated with increasing serum urea levels, may lead to delayed postpartum ovarian activity in Nili-Ravi buffaloes under field conditions. Low energy intake associated with poor body condition of buffaloes (BCS<2.75), may be a key factor for low reproductive efficiency in the animals. Improving the pre- and postpartum nutrition of such animals is likely to overcome the problem.

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