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Short Research Article

Protein supplementation on onset of post-partum ovarian cyclicity of dairy cows: an isotopic immunoassay study†

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Abstract: Protein is the main nutritional constraint on livestock development in Bangladesh. With this idea in mind, a study was conducted to evaluate the effect of (pulse) concentrate supplementation of protein mixture on the post-partum onset of ovarian cyclicity using an isotopic immunoassay technique. Other relevant parameters were also studied. It was concluded that the protein concentrate supplementation could have produced better weight gain, milk yield and reproductive performance of cows. Copyright © 2007 John Wiley & Sons, Ltd.

Keywords: isotope; body weight; reproduction; milk yield

Introduction

Among the various factors involved on the onset of post-partum ovarian cyclicity, dietary protein supplementation is one of the most important for the resumption of post-partum ovarian function. In this research work, low-quality protein mixture (pulse) that is not considered for human consumption was used as a protein-rich concentrate for the daily ration of the experimental cows. The research was undertaken with the following objectives:

(i) To find out the voluntary intake of DM and ME of crossbred cows.
(ii) To find out the suitable level of dietary concentrate during early lactation in cows.
(iii) To determine the relationship between feed consumption and the onset of first post-partum estrous by using an isotopic approach in dairy cows.

Results and discussion

The experiment was conducted at the Bangladesh Agricultural University Dairy Farm and relevant laboratories during the period from July 2002 to December 2003. Ten post-partum cows were selected. The cows were divided into two groups with five cows in each; group A (Control) and group B (Supplemented). All cows calved normally and had no history of peripherturient diseases or disorders. The cows were regarded as apparently healthy and free from detectable abnormalities of the genital tract.

All cows were kept in intensive conditions. Cows were placed hygienically in individual stalls in a well ventilated face-out stanchion barn house, but twice allowed limited pasture grazing at the nearby University Dairy Farm. Attempts were made to keep all animals under the same management condition. All cows were milked twice daily by hand, keeping their calves at their feet during milking. Ten milliliters of available milk were collected directly from the udder of the individual cow during milking intervals of 10 days beginning from 10 days after calving up to 120 days post-partum. Sodium azide tablets (Merck, Germany) were used as a preservative at 8 mg/10 ml milk. The milk samples were then kept in the deep freeze at −20°C until analysis. The individual body weights of the cows were measured from the first day of calving. Morning and evening milk yields were recorded to get the daily milk yield of the cows. The milk yields of the individual cows were recorded at 10-day intervals by the Dairy Farm registrar.

Food and Agriculture Organization of the United Nations/ International Atomic Energy Agency (FAO/ IAEA) Progesterone (P4) Radioimmunoassay (RIA) Kits

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were used for P4 measurement by using a ^{125}I as tracer according to Plaizier.[1] The P4 concentration in the skimmed milk was determined by using a solid phase RIA technique. The RIA kits for milk progesterone were supplied by the laboratory of the IAEA, Vienna, Austria.

Briefly, milk samples were thawed at room temperature and individual vials well vortexed to ensure mixing of the milk samples. All standard milk samples supplied by the IAEA were reconstituted with 1 ml distilled water, vortexed and used for assessing the presence of P4 in the unknown milk samples. The P4 concentration was determined in duplicate samples. P4 antibody-coated tubes were loaded individually with 100 µl of defatted milk and 1.0 ml of the radioactive iodine (^{125}I) ligand solution. After that it was incubated for 3 h in an air-conditioned room. The contents of the tubes, except those of total count (TC), were decanted in a wash basin (where 10% sodium hydroxide was poured beforehand to reduce the risk of $^{125}I_2$ formation) and after decanting all tubes were struck sharply onto absorbent paper to shake off any residual droplets. The radioactivity of each tube was counted for 1 min using a portable Gamma Counter (Mini-instrument, Burnham on Crouch, England) and subsequent calculations were carried out by dividing average count per minute (CPM) of assay tube by the average CPM of the zero standard tubes (B_0) which was multiplied by 100. The percentage binding values for all standards, internal quality control (IQC) and unknown samples were determined by dividing each CPM of these tubes with that of zero standard tubes and this was multiplied by 100. Logit log graph papers (supplied by the IAEA) were used to plot the standard points by plotting percent bound (B/B_0) on the vertical axis (Y) against P4 concentrations of the standards on the horizontal axis (X). The assay standard curve, a straight line, was drawn through the standard points. The progesterone concentrations of unknown samples and IQC were determined by reading their per cent bound values and extrapolating to the axis of the standard curve.

The sample of feeds were collected and chemically analyzed to ascertain the nutrient composition according to AOAC[2] (Table 1).

The body weight gain of the supplemented group (16.4 ± 2.7 kg) was significantly ($P<0.01$) higher than that of the control group (8.4 ± 3.4 kg) (Table 2). Hence, concentrate supplementation showed an increase in body weight.

The average milk yield of the supplemented group (4.48 ± 0.94 l/day) was not significantly higher than that of the control group (3.27 ± 0.96 l/d, Table 3) but biologically its importance may be due to the presence of amino acid (protein) in the ration which ultimately stimulate milk production.

The delayed onset of post-partum ovarian cyclicity prolongs the calving interval and low milk yield which ultimately cause great economic loss to farmers. The date of standing oestrus after parturition of the control and supplemented groups of cows were 141.68 ± 10.36 days and 101.00 ± 9.62 days ($P > 0.05$, Tables 4 and 5). This was higher than that of first P4 rise (first ovulation) meaning that farmers were unable to detect heat of their cows. However, it is evident from the above results (which are highly statistically significant, $P<0.05$) that the supplemented group of cows took less time (about 40 days) to express their standing oestrus.

Table 1 Proximate analyses of feed ingredients used in formulating experimental ration

Sample	DM (g/kg)	CP (g/kg)	EE (g/kg)	Ash (g/kg)
Napier grass	268	18.9	23	160
Para grass	278	101	29	112
Rice straw	890.2	29.8	19	120.2
Oil cake	899.8	386.2	116	103
Wheat bran	856.1	135.8	26.8	64.5
Common salt (NaCl)	955.7	—	—	911.5
Pulse mixture	860	314.7	38.0	47.7

Table 2 Summary of the results of ten days interval body weight changes of cows

Group	Mean body weight at different days (± SD)											
	Initial body wt.	10	20	30	40	50	60	70	80	90	Net wt. gain	Daily wt. gain (kg/day)
A($n = 5$) (Control)	238 ± 26.5	237 ± 26.3	238 ± 26.1	239 ± 26.2	240 ± 25.8	241 ± 26.1	243 ± 27.5	245 ± 27.5	246 ± 27.5	247 ± 27.9	8.4 ± 3.4	0.093 ± 0.037
B ($n = 5$) (Supplemented)	241 ± 30.4	237 ± 29.5	239 ± 29.4	241 ± 29.8	244 ± 29.7	246 ± 30.0	249 ± 30.2	252 ± 30.9	255 ± 31.0	257 ± 31.4	16.4 ± 2.7	0.22 ± 0.013

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Table 3 Average milk yield of cows

Group	Mean body weight at different days (± SD)									
	10th day	20th day	30th day	40th day	50th day	60th day	70th day	80th day	90th day	Average yield (kg/day)
A($n = 5$) (Control)	3.00 ± 0.00	3.60 ± 0.89	3.80 ± 1.09	3.80 ± 1.09	3.60 ± 0.89	3.40 ± 1.47	3.30 ± 1.48	2.60 ± 0.96	2.30 ± 1.20	3.27 ± 0.96
B ($n = 5$) (Supplemented)	2.80 ± 1.03	3.30 ± 1.03	3.80 ± 1.03	4.44 ± 1.12	4.70 ± 1.09	4.90 ± 1.34	5.00 ± 1.41	5.00 ± 1.41	5.00 ± 1.41	4.48 ± 0.94

Table 4 First expression of standing oestrus in post-partum cows (Group A)

Cows tag no.	Calving to standing oestrus (in days)	Mean ± SD	Level of significant
379	150		
5661	143		
190	130	141.68 ± 10.36	$^{**}P > 0.05$
5722	132		
18	153		

**=Non significance.

Table 5 First expression of standing oestrus in post-partum cow (Group B)

Cows tag no.	Calving to standing oestrus (in days)	Mean ± SD	Level of significant
196	105		
52	95		
611	90	101.00 ± 9.62	$^{*}P > 0.05$
5730	100		
38	115		

*=Non significance.

The above results are inconsistent with those of Nolan *et al.*[3] who suggested that inadequate nutrition causes delayed post-partum ovarian function by preventing gonadotropin release from the pituitary. The influence of nutrient intake causes weight change and hence the body condition at calving and during post-partum period and this could be related to the interval between calving to first post-partum oestrus.[4]

Conclusions

Inadequate feeding practice, low milk production, and delayed onset of post-partum cyclicity and consequently prolonged calving intervals are the major constraints in dairy cattle production in Bangladesh. This experiment suggests that the protein concentrate supplementation could improve weight gain, milk yield and reproductive performance during post-partum period of cows.

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