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The effect of oil content of various feed protein sources on *in situ* and *in vitro* ruminal and post-ruminal protein disappearance

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Introduction *In situ* rumen incubation and post-rumen mobile nylon bag technique, and *in vitro* enzymatic procedure are currently used for estimating ruminal and post-ruminal disappearance of feed protein (Danesh Mesgaran and Stern, 2005). The objectives of the present study was to determine the ruminal and post-ruminal protein disappearances of various feed protein sources containing different oil concentrations, using *in situ* rumen incubation and post-rumen mobile nylon bag technique, and an *in vitro* enzymatic procedure.

Materials and methods Feed protein sources containing different oil concentrations, produced in various Iranian oil industries, were cottonseed meal (5, 40 and 80 g oil/kg dry matter (DM); CS5, CS40 and CS80, respectively), rapeseed meal (30 and 70 g oil/kg DM; RS30 and RS70, respectively) and soyabean meal (6 and 50 g oil/kg DM; SB6 and SB50, respectively). Ruminal and post-ruminal protein disappearance of the samples were evaluated *in vivo* using the *in situ* disappearance and mobile nylon bag techniques and *in vitro* using an enzymatic procedure (Danesh Mesgaran and Stern, 2005). Approximately 1.2 g of sample DM was placed in a polyester bag (3 cm × 6 cm; pore size of 52 µm; 16 bags/feed) and incubated in the rumen of four Holstein steers (395 ± 13 kg), fitted with rumen and T-shaped intestinal cannulae, for 12 h (assumed out flow rate of 0.08/h). After removal from the rumen, half of the bags were inserted into the small intestine via the intestinal cannulae, then removed from the voided faeces, washed and dried (64 °C, 48 h). For the *in vitro* enzymatic procedure, 1 g of each sample was weighed into polyester bags, pore size of 22 µm (n= 6), placed into borate-phosphate buffer and incubated for 1 h at 39 °C in a shaking Incubator. Then, 400 ml of protease (Sigma, P-5147) solution (1980 units protease in 400 ml of borate-phosphate buffer) were added and after 4 h incubation (39 °C), the bags were removed and rinsed. Three bags per sample were dried (64 °C, 48 h) and the remaining bags were placed in a pepsin solution [1.6 g of pepsin (Sigma P-7000) in 800 ml of a 0.1 N HCl]. Bags were incubated for 1 h at 39 °C. Then, 40 ml of 1 N NaOH and 1 litre of the pancreatin solution [6 g of pancreatin (Sigma, P-7545) and 68 g of KH₂PO₄ in 1 litre of distilled water] was added, then incubated for 24 h at 39 °C. After incubation, bags were removed and washed and dried (64 °C, 48 h). Nitrogen concentration of each pre-incubated and incubated sample was determined using the Kjeldhal method. Data were analysed using the general linear models procedure of SAS (1999) with the statistical model of Y= overall mean+ block effect of procedure+ feed effect within block effect+ residual error.

Results The ruminal and post-ruminal protein disappearance values of the various feed protein sources are shown in Table 1. The procedures had a significant effect ($P < 0.05$) on the rumen and post-rumen protein disappearance of the feeds evaluated in the present study. Results showed a significant ($P < 0.05$) effect of feed protein sources with different oil concentration on ruminal and post-ruminal protein disappearance.

Table 1 Rumen and post-rumen protein disappearance values of the various protein sources studied containing different oil concentrations using the *in situ* and enzymatic *in vitro* procedures

Protein sources	Rumen CP disappearance						Post-rumen disappearance of rumen-undegraded CP					
	Procedure		Feed effect*		Procedure effect*		Procedure		Feed effect		Procedure effect	
	<i>In situ</i>	<i>In vitro</i>	s.e.m.	P	s.e.m.	P	<i>In situ</i>	<i>In vitro</i>	s.e.m.	P	s.e.m.	P
CS5	0.74	0.81	0.04	<0.05	0.03	<0.05	0.91	0.81	0.04	<0.05	0.3	<0.05
CS40	0.46	0.71					0.83	0.73				
CS80	0.54	0.70					0.71	0.54				
RS30	0.89	0.85					0.70	0.88				
RS70	0.84	0.80					0.64	0.80				
SB6	0.84	0.91					0.85	0.96				
SB50	0.61	0.88					0.84	0.94				

* When the difference between means is greater than two times the s.e.m., it is considered as significant ($P < 0.05$).

Conclusions There was a considerable variation in rumen and post-rumen protein disappearance of the feed protein sources evaluated by *in situ* and *in vitro* methods. Samples of cottonseed meal with low oil concentration had higher rumen and post-rumen protein disappearance values compared with CS40 and CS80. Therefore, modifications of the *in situ* and *in vitro* methods for estimating the digestibility of high oil containing feed protein sources need to be developed.

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