

# Effects of fibre and non-fibre carbohydrate and level of intake on microbial protein yield in Sarda sheep

F. Boe<sup>1</sup>, V. Giovanetti<sup>2</sup>, E. Zerbini<sup>3</sup>, G. Bomboi<sup>4</sup>, B. Floris<sup>4</sup>

<sup>1</sup> Dipartimento di Scienze Zootecniche. Università di Sassari, Italy

<sup>2</sup> Istituto Zootecnico e Caseario per la Sardegna. Olmedo, Italy

<sup>3</sup> Cargill Animal Nutrition. Spessa, Italy

<sup>4</sup> Dipartimento di Biologia Animale. Università di Sassari, Italy

*Corresponding author:* Filippo Boe. Dipartimento di Scienze Zootecniche. Facoltà di Agraria, Università di Sassari. Via E. De Nicola 9, 07100 Sassari, Italy – Tel. +39 079 229308 - Fax: +39 079 229302 – Email: [fboe@uniss.it](mailto:fboe@uniss.it)

**ABSTRACT:** Three studies using Sarda dairy sheep in dry, mid-lactation and late-lactation were carried out. Forty ewes for each physiological stage were fed 8 complete pelleted diets, which differed from each other in NDF and NFC content and source. Based on their main ingredient, diets were denominated: corn meal (CM), wheat middlings (WM), corn flakes (CF), barley meal (BM), corn cobs (CC), beet pulp (BP), alfalfa (AA), and soybean hulls (SH). In each study, rumen microbial protein (MCP) synthesis was estimated measuring urinary purine derivatives. In dry sheep, MCP synthesis was not affected by diet, while in mid- and late-lactation sheep dietary effects were observed. In mid-lactation, the highest MCP production was found for BM and BP (171 and 166 g/d, respectively), while the lowest was observed with AA (63 g/d). In late-lactation, the highest MCP yield (146 g/d) was observed in BP, while the lowest were for SH and CM. MCP synthesis, for each diet, was higher in mid-lactation than in late-lactation, which in turn were higher than in the dry period. Dry matter intake (DMI) was positively associated to MCP. The MCP synthesis was best predicted by dietary energy (NEL) or digestible organic matter intake (dOMI).

**Key words:** Microbial protein, Purine derivatives, Sheep, Urea.

**INTRODUCTION** – Dietary NDF and NFC concentration and source can have large effects on microbial yield and efficiency, which can vary at different levels of intake (NRC, 2001). No sufficient information is available about these aspects for dairy sheep. Thus, we studied the effects of different dietary structural and non structural carbohydrates concentrations and sources on microbial protein (MCP) synthesis in Sarda sheep fed at various levels of intake during different physiological stages.

**MATERIAL AND METHODS** – Three trials on Sarda dairy ewes dry (E1), in mid (E2, 112 ± 7 days in milk, DIM) and late lactation (E3, 200 ± 10 DIM) were carried out in order to test diets at different levels of intake. In each study, forty ewes were put in individual metabolic cages, in which they were fed the experimental diets during 14 days of adaptation and 9 days of experimental measurements. All diets contained dehydrated alfalfa as a common basis and were added of various ingredients in order to obtain diets differing for NDF and NFC concentration and source (Table 1).

Based on their main ingredient, diets were denominated: corn meal (CM), wheat middlings (WM), corn flakes (CF), barley meal (BM), corn cobs (CC), beet pulp (BP), alfalfa (AA), and soybean hulls (SH).

In each experiment, the forty sheep were divided in 8 groups of 5 animals. Each group received one of the 8 diets twice a day, for a total supply of 600 g/d in E1, while in E2 and E3 sheep were fed *ad libitum*. In E3, WM was not used and only the other seven diets were tested. Total feces and urine collection was performed on each study to estimate dietary digestibility and total purine derivatives excretion. Plasma samples were analyzed for urea concentrations. The separation and quantification of allantoin and oxypurines in urine samples was carried out following the method proposed by Balcelles *et al.* (1992). Absorbed purine (mmol/day) and MCP were estimated based on the quantitative relationship defined by Chen *et al.* (1990). Energy concentration was estimated based on the

actual digestibility of the diet. Data were analysed using a general linear model procedure of SAS (1996) according to the following model:  $Y = \mu + \alpha_i + \varepsilon_{ij}$ , where:  $\mu$  = overall mean;  $\alpha_i$  = fixed effect of diets;  $\varepsilon_{ij}$  = random error.

Table 1. Chemical composition (% of DM) and energy content (Mcal/kg DM) of the diets.

Diets	CM	WM	CF	BM	CC	BP	AA	SH
CP	18.0	17.8	18.7	18.1	19.8	17.9	19.7	17.5
NDF	23.9	26.2	28.5	33.2	45.8	44.9	45.6	51.9
NFC	51.2	47.9	47.3	41.0	26.1	31.8	25.1	23.6
Starch	35.6	34.1	28.8	25.2	12.4	8.0	7.1	3.2
NEL - E1	1.92	1.84	1.96	1.84	1.64	1.86	1.46	1.80
NEL - E2	1.80	1.72	1.80	1.74	1.52	1.78	1.30	1.50
NEL - E3	1.76		1.88	1.68	1.50	1.73	1.20	1.44

**RESULTS AND CONCLUSIONS** – A fairly high dietary CP concentration was used in all diets (Table 1) to avoid any possible shortage of nitrogen at a ruminal and intestinal level. When the diets were supplied *ad libitum* (E2 and E3), DMI was markedly affected by diet composition; the rank differed in the two stages of lactation (Table 2). In the study E1, MCP did not differ among the diets, probably because all diets were given in the same amount (Table 2). MCP, for each diet, was higher in E2 than in E3, which in turn had higher daily values than E1. It was clear that DMI was positively associated to MCP (Table 2).

Table 2. Dry matter intake, microbial crude protein yield (MCP), MCP per kg of digestible OM intake (dOMI), and blood urea concentration.

	Stage	Diet								P<
		CM	WM	CF	BM	CC	BP	AA	SH	
DMI, g/d	E1	489	511	517	528	514	520	525	509	NS
	E2	1718 <sup>c</sup>	1864 <sup>bc</sup>	1871 <sup>abc</sup>	1849 <sup>c</sup>	2602 <sup>ab</sup>	2055 <sup>abc</sup>	2196 <sup>abc</sup>	2634 <sup>a</sup>	0.01
	E3	890 <sup>c</sup>		1295 <sup>bc</sup>	1752 <sup>ab</sup>	2182 <sup>a</sup>	1992 <sup>a</sup>	1903 <sup>ab</sup>	1604 <sup>ab</sup>	0.01
MCP, g/d	E1	34	31	27.7	23.5	27	25.8	28	28	NS
	E2	112 <sup>c</sup>	131 <sup>abc</sup>	147 <sup>abc</sup>	171 <sup>a</sup>	129 <sup>bc</sup>	166 <sup>ab</sup>	63 <sup>d</sup>	126 <sup>bc</sup>	0.01
	E3	68 <sup>cd</sup>		94 <sup>bcd</sup>	108 <sup>b</sup>	92 <sup>bcd</sup>	146 <sup>a</sup>	96 <sup>bc</sup>	61 <sup>d</sup>	0.01
MCP, g/kg of dOMI	E1	92	82	70	62	85	70	94	77	NS
	E2	90 <sup>bc</sup>	100 <sup>bc</sup>	111 <sup>abc</sup>	135 <sup>a</sup>	87 <sup>cd</sup>	118 <sup>ab</sup>	54 <sup>d</sup>	82 <sup>cd</sup>	0.01
	E3	105		101	93	72	111	109	68	0.08
Blood urea, mg/dl	E1	31.1	33.6	32.2	34.4	32.2	32.5	40.0	34.4	NS
	E2	47.4 <sup>b</sup>	59.4 <sup>ab</sup>	51.2 <sup>ab</sup>	56.6 <sup>ab</sup>	56.6 <sup>ab</sup>	50.0 <sup>ab</sup>	76.5 <sup>a</sup>	58.3 <sup>ab</sup>	0.04
	E3	46.8		54.7	65.6	59.7	55.1	77.7	60.8	0.13

a,b,c,d = P<0.05.

The NRC (2001) reported that numerous studies have found higher MCP synthesis when either the NFC level was increased or less degradable carbohydrates were replaced by more degradable ones. In our experiments, this pattern was true only in part. The BP had in E2 similar or higher MCP synthesis and efficiency (calculated as MCP/digestible OM intake) than the starchy diets (CM, WM, CF, BM). Moreover, BP had the highest MCP in E3. In this stage MCP efficiency did not differ among treatments, even though BP had the highest numerical value. The BP diet combined the highest NFC and digestible NDF concentration (not shown) among the fibrous diets with high NEL content in all stages. This might explain its high MCP. The BM diet had the highest MCP synthesis in E2, and the second highest in E3. This diet had the lowest starch concentration and the highest NDF concentration among the 4 starchy diets. Its starch probably had very high degradation rate and solubility, being made mostly by barley

starch. The high microbial yield of this diet can be the result of a good balance between starch and NDF concentration associated with the high degradability of its starch, confirming that synchronizing for fast degradable starch and protein sources stimulates greater synthesis or efficiency of MCP (NRC, 2001). In our experiments, SH produced different results in E2 and E3. MCP synthesis was two fold higher in E2 than in E3 and it was not different from that obtained using corn based diets (CM and CF, Table 2). In E3, intake as well as MCP synthesis and efficiency were decreased by feeding SH. Probably, because of the high environmental temperatures during E3, the sheep ate mostly in the cooler hours and a decline of rumen pH was induced. Many authors found that, despite their high NDF content, soyhulls increased the molar proportion of propionate in ruminal fluid when they replaced forage and did not affect rumen pH when they were fed in place of grains (Ipharraguerre *et al.*, 2002). Overall, MCP synthesis was best predicted by NEL or digestible OM intake ( $MCP = 35.8 \text{ NEL intake} - 5.9$ ;  $R^2 = 0.83$ ), confirming previous findings on this subject.

In E1 (dry sheep) no effect of diet on blood urea concentrations (BUC) was found (Table 2). In E2 (mid-lactating sheep) BUC was affected by the diet, being highest with AA and lowest with CM diet (Table 2). No effect of diet on BUC was found in E3 (late-lactating sheep); however sheep fed the AA diet tended to have a higher value ( $P < 0.13$ , Table 2). The AA had the highest value of soluble N concentration (not reported), and the lowest MCP (in E2) and NEL concentration. The sheep fed AA had also the lowest NEL intake. As a result, available nitrogen could not be efficiently used by microbes for the lack of energy substrates, as suggested by the fact that BUC was the highest when this diet was fed (Table 2).

In conclusion, the BP diet appeared to be the most efficient in terms of MCP, probably due to its fairly high NFC and NEL concentration combined with high digestible NDF concentration. The high MCP of BM could be the result of the good balance between starch and NDF concentration associated with the high degradability of its starch.

*The Authors want to thank Prof. Antonello Cannas and Dr. Giovanni Molle for their guidance.*

The research was supported by Cargill Animal Nutrition.

**REFERENCES** - Balcells, J., Guada, J.A., Peiró, J.M., Parker, D.S., 1992. Simultaneous determination of allantoin and oxypurines in biological fluids by high-performance liquid chromatography. *Journal of Chromatography*, 575:153-157. Chen, X.B., Hovell, F.D., Orskov, E.R., Brown, D.S., 1990. Excretion of purine derivatives by ruminants: effect of exogenous nucleic acid supply on purine derivative excretion by sheep. *British Journal of Nutrition*, 63:131-42. Ipharraguerre, I.R., Shabi, Z., Clark, J.H., Freeman, D.E., 2002. Ruminal fermentation and nutrient digestion by dairy cows fed varying amount of soyhulls as a replacement for corn grain. *Journal of Dairy Science*, 85:2890-2904. NRC, 2001. *Nutrient Requirements of Dairy Cattle*, 7<sup>th</sup> Revised Edition. National Academy Press, Washington, DC. SAS, 1996. *User's guide: statistics* - SAS Inst., Cary, NC, USA.