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SCIENCE DIRECT®
Animal Feed Science and Technology
119 (2005) 171–178
ANIMAL FEED SCIENCE AND TECHNOLOGY
www.elsevier.com/locate/anifeedsci

Short communication

Chemical composition, in vivo digestibility, N degradability and enzymatic intestinal digestibility of five protein supplements

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Received 8 April 2004; received in revised form 25 November 2004; accepted 9 December 2004

Abstract

The nutritive value of solvent extract palm kernel meal (SPKM), expeller palm kernel meal (EPKM), copra meal (CM), corn distillers dried grains (CDG) and corn gluten feed (CGF) were evaluated. Apparent in vivo digestibility was measured in rams using increasing levels of supplement studied. Dry matter (DM) and N degradability of feeds after 0, 12 and 16 h rumen incubation and enzymatic digestibility of rumen undegradable protein (UDP) were also determined. It was confirmed that supplements differ significantly in terms of digestibility (energetic value), N degradability and intestinal digestibility of UDP, the figures obtained being consistent with those found in the literature. The low energetic value of SPKM could limit its inclusion in high productive ruminant diets. Although CDG may be considered a good source of UDP, the intestinal digestibility of this fraction was low.

Abbreviations: ADF, acid-detergent fibre; ADL, acid-detergent lignin; CDG, corn distillers dried grains; CGF, corn gluten feed; CM, copra meal; CP, crude protein; DE, digestible energy; DM, dry matter; EE, ether extract; EPKM, expeller palm kernel meal; GE, gross energy; NDF, neutral-detergent fibre; OM, organic matter; SPKM, solvent extract palm kernel meal; UDP, rumen undegradable protein

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0377-8401/$ – see front matter
doi:10.1016/j.anifeedsci.2004.12.006

Additionally, the relatively high unsaturated fat content of CDG might restrict its incorporation level in ruminant diets.

Keywords: Enzymatic intestinal digestibility; In vivo digestibility; N degradability; Protein supplements

1. Introduction

Protein sources are a problematic component of animal's diets in Portugal and in the European Union. The high dependence on imports together with the fluctuation in prices, which show a tendency to increase rapidly, as well as the limited protein sources are some of the major problems. Although soybean meals are the largest source of protein for livestock animals in Portugal (78% of vegetable meals in 2002; IACA, 2003), there is a wide range of vegetable by-products (mainly from the malting, brewing and distilling industries and oilseed residues) with moderate protein content available in the market for feeding productive ruminants. Since knowledge of their nutritive value is essential for their proper use, a greater effort has been made to characterise and to predict the nutritive value of several by-products (e.g., Moss and Givens, 1994; O'Mara et al., 1999; Woods et al., 2003a).

No measurements of in vivo digestibility have been made in Portugal with the moderate protein sources commonly available for manufacturing compound feeds for ruminants. This study determines apparent in vivo digestibility of solvent extract palm kernel meal (SPKM), expeller palm kernel meal (EPKM), copra meal (CM), corn distillers dried grains (CDG) and corn gluten feed (CGF), using increasing levels of each supplement. As the evaluation of the protein value of feeds for ruminants requires its division into quickly and slowly degradable crude protein (CP) and undegradable CP (UDP; e.g., AFRC, 1993), these fractions were determined by the nylon bag technique. Additionally, the intestinal digestibility of UDP was estimated by an enzymatic method.

2. Materials and methods

2.1. Experimental feeds

The samples of SPKM, EPKM, CM, CDG and CGF studied were obtained from feed compound manufacturers of North and Central Portugal, over the years 2001 and 2002.

2.2. Apparent in vivo digestibility

Twelve Ile de France rams (77 ± 10.5 kg body weight) were used to measure the apparent in vivo digestibility of feeds at Campus Agrário de Vairão (University of Porto). The animals were kept in metabolism crates in a well-ventilated experimental barn. Before the studies, all animals were sheared, dewormed and injected subcutaneously with 1,000,000 IU of vitamin A, 150,000 IU of vitamin D and 100 IU of vitamin E. For each feed, the in vivo

digestibility was measured in four rams fed at the maintenance level (AFRC, 1993) with a diet comprising dehydrated alfalfa and increasing levels of the feed supplements (0, 150, 300 and 450 g kg^{-1} dry matter; DM), according to a 4 × 4 Latin square design. Due to the limited number of metabolism crates, the supplements were not all evaluated simultaneously. The diet was offered twice a day at 09:30 and 17:30 h. All animals received daily ca. 20 g of a commercial mineral and vitamin mixture and had free access to water. Each experimental period lasted for 21 days, comprising 14 days for adaptation to the diet and 7 days for total faeces collection. Samples of feeds offered and faeces were taken daily and bulked over the trial period.

2.3. In sacco degradability

Two dry Holstein cows (480 and 575 kg BW) fitted with rumen cannulae (10 cm diameter; Bar Diamond Inc., Parma, Idaho, USA) were used to measure rumen degradability of feeds. The cows were fed on a diet comprising (DM basis), 450 g kg^{-1} corn silage, 50 g kg^{-1} ryegrass hay, and 500 g kg^{-1} protein-rich commercial concentrate at 1.2× maintenance (AFRC, 1993). The cows were kept in individual tie-stalls with individual feed bins in an animal house which was well-ventilated, and had continuous access to water. Diets were given as total mixed ration with fresh feed offered twice each day (09:30 and 17:30 h). The nylon bag technique (Ørskov et al., 1980) was used to measure the DM and N degradation of feeds in the rumen. Nylon bags (10 cm × 20 cm; Bar Diamond Inc., Parma, Idaho, USA) containing 4 g of feed ground through a 4 mm screen were incubated in the rumen of each cow for 12 and 16 h, immediately after the morning feed, on two non-consecutive days. In total there were eight replicates for each feed sample and for each incubation time (2 cows × 2 days × 2 bags). Immediately after removal from the rumen, the bags were washed in cold water and frozen at −15 °C. At the end of the collections, they were unfrozen and washed together with the zero time bags (not incubated in the rumen) in a washing machine for 40 min at 40 °C and then dried at 65 °C for 24 h. The residues were accumulated by feed, cow and incubation time and submitted for analysis.

2.4. In vitro intestinal digestibility

Protein intestinal digestibility of nylon bag residues after 12 and 16 h rumen incubation was estimated according to the method described by Kopečný et al. (1998). Each feed residue was incubated in duplicate in three series, performed on different days.

2.5. Chemical analysis

Samples of feeds and faeces were dried in a forced-air oven at 65 °C for, respectively, 24 and 48 h and the DM content calculated. Ground samples (1 mm) were analysed for ash (AOAC, 1990, ID 942.05) and Kjeldahl N (AOAC, 1990, ID 954.01). Crude protein was calculated as Kjeldahl N × 6.25. Neutral-detergent fibre (NDF), acid-detergent fibre (ADF) and acid-detergent lignin (ADL) were determined by the detergent procedures of Van Soest et al. (1991) and Robertson and Van Soest (1981), with alpha amylase being

added, except for dehydrate alfalfa, during NDF extraction. Sodium sulfite was not added. NDF was expressed without residual ash. Ether extract (EE) was determined by extracting the sample with petroleum ether using a Gerhardt Soxtherm 2000 Automatic (AOAC, 1990, ID 920.39). Phosphorus was determined by official Portuguese standard method (Norma Portuguesa 873, 1997). Starch was analysed on samples after grinding to pass a 0.5-mm screen by the method described by Salomonsson et al. (1984). Gross energy (GE) of feeds and faeces was determined by complete oxidation in an adiabatic bomb calorimeter (Parr, Model A50M, series 1286).

2.6. Statistical analysis and calculations

Data were analysed using the statistical package Systat (Wilkinson et al., 1992). Apparent in vivo digestibility and digestible energy (DE) content of total diets data for each supplement were subjected to analysis of variance according to 4×4 Latin square design (Steel and Torrie, 1980). The nature of the response to the inclusion level of supplement was studied by partition of the diet sum of squares by use of orthogonal polynomials (Steel and Torrie, 1980). The apparent in vivo digestibility and DE content of each supplement were estimated by linear regression. When these total diet parameters increased quadraticaly with the inclusion level of supplement, digestibility coefficients and DE of supplement were estimated by quadratic regression. The apparent digestibility and DE content of each supplement were estimated for the inclusion level of supplement corresponding to the maximal total diet digestibility or DE content (i.e., when the first derivative of the quadratic equation equals zero). Data from in sacco degradability were subjected to least squares analyses of variance (Steel and Torrie, 1980). The model for 12 and 16 h degradability included supplement, cow and residual error and the model for 0 h degradability included supplement and residual error. In vitro intestinal digestibility of UDP data were subjected to least squares analyses of variance (Steel and Torrie, 1980), including cow, supplement, rumen incubation time, series of in vitro incubation, supplement $\times$ rumen incubation time and residual error in the model. As the interaction supplement $\times$ rumen incubation time was not significant, it was removed from the model.

3. Results and discussion

3.1. Chemical composition

Chemical analysis (Table 1) yielded results similar to those reported in the literature (Masoero et al., 1994; Moss and Givens, 1994; Woods et al., 2003a.). The results reflect the influence of the type of supplement as well as of the technological process. The former is clearly shown by the higher lignified cell wall contents and lower CP contents of oilseed meals than corn by products. The influence of technological processing is clear with the PKM samples, EPKM having a higher EE content than SPKM in agreement with the results obtained by O'Mara et al. (1999). As expected, CGF presented lower CP and EE contents and higher starch content than CDG. Gross energy contents of the supplements used are in accordance with their EE contents.

Table 1
Chemical composition (g kg^{-1} DM) and gross energy (GE; MJ kg^{-1} DM) of the experimental feeds

Feeds	DM (g kg^{-1})	Ash	CP	NDF[a]	ADF	ADL	EE	Starch	*P*	GE
SPKM	930	48	175	731	460	146	25	ND	6.6	17.9
EPKM	945	50	172	665	357	120	74	ND	6.5	19.0
CM	910	67	222	639	306	150	47	ND	6.1	18.3
CDG	914	78	268	464	118	39	86	71	8.6	19.5
CGF	923	75	211	398	119	28	29	144	9.3	18.0
DL[b]	908	109	178	428	298	83	13	ND	2.8	17.1

SPKM: solvent extract palm kernel meal; EPKM: expeller palm kernel meal; CM: copra meal; CDG: corn distillers dried grains; CGF: corn gluten feed; DL:dehydrated alfalfa.

[a] Ash free. Assayed without sodium sulfite. Alpha amylase was not used for dehydrated alfalfa.

[b] Mean composition of three batches of dehydrated alfalfa.

3.2. Apparent in vivo digestibility and digestible energy

Table 2 presents the apparent in vivo digestibility and DE of total diets and of the supplements. For all protein sources, total diet digestibility of DM, organic matter (OM) and NDF increased significant and linearly ($P < 0.01$) with the inclusion level of supplement. The level of SPKM and CDG also had a significant quadratic effect ($P < 0.10$) on DM and OM digestibility of total diet. Although the high EE content of CDG might explain the quadratic response observed, through a decrease in rumen microbial activity (Van Soest, 1994), this explanation is not acceptable for SPKM. In this case, the response observed could be eventually attributed to its lower digestibility and the reduction in rumen retention time associated with a higher supplement/hay ratio.

The estimated SPKM digestibility was lower than that of EPKM. O'Mara et al. (1999) found precisely the opposite, but it must be noted that the mean NDF content of EPKM samples used by these authors was higher (810 g kg^{-1} DM versus 665 g kg^{-1} DM). However, OM digestibility of EPKM obtained agrees favourably with the values reported by Moss and Givens (1994). The digestibility coefficients for CM, CDG and CGF are consistent with the values previously reported (Moss and Givens, 1994; Woods et al., 2003a). The estimated DE content of the supplements reflects their OM digestibility and EE content, SPKM and CDG, respectively, having the lowest and the highest energy content.

3.3. In sacco degradability and enzymatic intestinal digestibility of protein

Table 3 presents the washing losses (0 h) and the in sacco degradability of feeds after 12 and 16 h rumen incubation. The incubation times were chosen to reflect the common rumen retention time of feeds in lactating dairy cows. The results shows that values for the washing losses (0 h) of N as well the N degradability after 12 and 16 h rumen incubation of SPKM were lower than those of EPKM, possibly as a consequence of the industrial production process modifying rumen protein degradability. Conversely, O'Mara et al. (1999) suggested that the heat generated by the expeller process might cause heat damage to the protein and thus reduce digestibility. Figures for N in sacco degradability of CM agree with those

Table 2
Apparent in vivo digestibility and digestible energy (MJ kg^{-1} DM) of total diets and estimated apparent in vivo digestibility and digestible energy of the supplements studied

	Level of supplement (g kg^{-1} DM)				SEM	*P*	Contrasts		Estimated[a]
	0	150	300	450			L	Q	
DM digestibility									
SPKM	0.583	0.605	0.628	0.625	0.0039	***	***	*	0.688
EPKM	0.611	0.640	0.651	0.685	0.0096	**	**	NS	0.758
CM	0.632	0.642	0.676	0.697	0.0060	***	***	NS	0.775
CDG	0.593	0.647	0.673	0.677	0.0061	***	***	**	0.806
CGF	0.612	0.629	0.652	0.680	0.0037	***	***	NS	0.755
OM digestibility									
SPKM	0.616	0.635	0.658	0.656	0.0048	**	***	†	0.707
EPKM	0.645	0.676	0.688	0.719	0.0093	**	**	NS	0.790
CM	0.688	0.698	0.723	0.741	0.0063	**	***	NS	0.800
CDG	0.627	0.680	0.704	0.710	0.0063	***	***	*	0.823
CGF	0.650	0.664	0.685	0.712	0.0045	***	***	NS	0.777
NDF digestibility									
SPKM	0.475	0.532	0.572	0.591	0.0090	***	***	†	0.683
EPKM	0.472	0.556	0.599	0.665	0.0139	***	***	NS	0.791
CM	0.586	0.633	0.690	0.733	0.0076	***	***	NS	0.843
CDG	0.507	0.575	0.614	0.637	0.0129	**	***	NS	0.796
CGF	0.495	0.509	0.539	0.585	0.0123	**	**	NS	0.685
Digestible energy									
SPKM	9.8	10.2	10.8	10.8	0.07	***	***	*	11.8
EPKM	10.3	11.1	11.6	12.5	0.19	***	***	NS	14.8
CM	11.1	11.4	12.1	12.6	0.11	***	***	NS	14.3
CDG	9.9	11.3	12.1	12.5	0.11	***	***	**	15.1
CGF	10.3	10.7	11.2	11.9	0.08	***	***	NS	13.7

NS: no significant; SPKM: solvent extract palm kernel meal; EPKM: expeller palm kernel meal; CM: copra meal; CDG: corn distillers dried grains; CGF: corn gluten feed; L: linear; Q: quadratic.

[a] Estimated by linear or quadratic regression.

† $P < 0.10$.

* $P < 0.05$.

** $P < 0.01$.

*** $P < 0.001$.

reported by Masoero et al. (1994) and Moss and Givens (1994). As expected, the two corn by products studied differ in N degradability, the N soluble and degradable fractions of CGF being very much higher than those of CDG, reflecting some solubilisation of protein during the wet-milling process and the high temperatures applied and the presence of prolamins and glutelins (proteins of high molecular weight with disulfide bonds) in CDG (Clark et al., 1987).

Enzymatic intestinal digestibility of UDP (Table 3) was not significantly affected by the duration of rumen incubation, as observed by Hindle et al. (1995) using the mobile bag technique in palm kernel by products. The values confirm significant differences between supplements, CM and CDG being the supplements with lower intestinal digestibility. This

Table 3
In sacco degradability and enzymatic intestinal digestibility of rumen undegradable protein of experimental feeds

	Feeds					SEM	*P*	Rumen incubation time (h)		SEM	*P*
	SPKM	EPKM	CM	CDG	CGF			12	16		
DM degradability											
0 h	0.190	0.343	0.340	0.355	0.508	0.0043	***				
12 h	0.322	0.439	0.554	0.605	0.681	0.0197	**				
16 h	0.381	0.491	0.650	0.633	0.710	0.0231	**				
N degradability											
0 h	0.200	0.407	0.302	0.305	0.663	0.0041	***				
12 h	0.302	0.422	0.366	0.536	0.780	0.0422	**				
16 h	0.376	0.524	0.480	0.536	0.859	0.0314	**				
UDP digestibility	0.803	0.764	0.644	0.509	0.698	0.0250	***	0.679	0.688	0.0158	NS

SPKM: solvent extract palm kernel meal; EPKM: expeller palm kernel meal; CM: copra meal; CDG: corn distillers dried grains; CGF: corn gluten feed.
** $P < 0.01$.
*** $P < 0.001$.

can be attributed to the high fibre content of CM and to the extensive heat treatment of CDG. This study used only one source of each feed and consequently did not allow the evaluation of the effects of different sources of the same feed. However, the existent significant variation in intestinal digestibility of UDP between different sources of concentrate feedstuffs as well between the different sources of one concentrate feedstuff (Woods et al., 2003b) should be considered in formulating diets.

4. Conclusions

This study confirms that the supplements evaluated differ significantly in terms of digestibility (energetic value), N degradability and intestinal digestibility of UDP, the figures obtained being consistent with those found in the literature. The low energetic value of SPKM could limit its utilization in the diets of highly productive ruminants. Although CDG may be considered a good source of UDP, the intestinal digestibility of this fraction was low. Additionally, the relatively high unsaturated fat content of CDG might restrict its incorporation level in ruminant diets.

Acknowledgments

This work was partially financed by the Instituto Nacional de Investigação Agrária e Pescas (INIAP, Portugal), Projecto 342, Medida 8, Acção 8.1 do Programa Agro (European Union), which is gratefully acknowledged. The authors thank the help of the staff of the Nutrition laboratory of AGROS/University of Porto for the analysis of feed samples and the constructive and helpful suggestions of Dr. Arnaldo Dias-da-Silva.

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