

Determination of rumen degradability, intestinal digestibility and protein nutritional value of sunflower cake produced in Bulgaria

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SUMMARY

The present study objective was to determine the rumen degradation kinetics, intestinal digestibility and protein nutritional value for ruminant animals of sunflower cake (SFC) produced in Bulgaria. Three non-lactating Jersey cows with an average body weight of 436 ± 18 kg fitted with a rumen and T-type duodenal cannulas were used in the experiment. Six samples of SFC were collected from six different sunflower processing companies located in Bulgaria (SFC1 to SFC6). Feed samples were incubated in the rumen of the cows for 0, 2, 4, 8, 16, 24 and 48 h in 6 replications. The rapidly degradable fraction *a* of DM was significantly lowest at SFC2 (15.8%) and highest at SFC6 (25.8%) ($P < 0.01$). The effective degradability of DM of SFCs at outflow rate 0.06/h ranged greatly from 47% to 63%. The soluble fraction of CP ranged from 24.9% for SFC3 to 34.1% for SFC4 ($P < 0.01$). Effective degradability of CP at different outflow rates ($k_p = 0.045, 0.06, 0.08$) for SFC6 were higher ($P < 0.01$) compared with the other five samples of SFC. The intestinal digestibility of the DM measured by mobile bag technique varied from 36.5% for SFC6 to 46.9% for SFC3. The values for intestinal digestibility of CP for SFC3 (94%) were significantly higher than the other samples ($P < 0.01$). The average value for protein digestible in the small intestine (PDI) according to the Bulgarian protein system, at a rumen outflow rate 0.06/h, was 154 g/kg DM, and the balance of protein in the rumen (BPR) was 101 g/kg DM. Although nutrient composition of SFCs from different processing plants is highly variable, the protein degradability and digestibility values obtained in this experiment can be used in formulating rations for ruminant animals. The observed differences could be mainly attributed to the different degree of squeezing the oil from sunflower seeds and variation of the quality of raw materials.

Keywords: rumen degradability, intestinal digestibility, protein value, sunflower cake

INTRODUCTION

Sunflower cake and meal are the main concentrate protein sources for ruminants in Eastern Europe, some Mediterranean countries and Argentina. Sunflower cake is a by-product from oilseed industry obtained by extracting the oil from sunflower seeds using continuous screw-presses (expellers). Subsequent processing of the cake with a solvent will result in production of sunflower meal. In the European Union, regulations forbid the use of solvents for the production of feed ingredients used in organic farming, which makes mechanically-extracted sunflower cakes suitable for organic animal production.

The sunflower meal as well the sunflower cake has high rumen degradability (Veresehyazy and Fekete, 1990), but it would be possible to improve its value by efficient heat processing or by using it with diets rich in structural carbohydrates that contain tannins (Alcaide et al., 2003). The improvement of quality of sunflower by-products should have a positive effect when inclusion levels of those feeds are increased (Griffiths, 2004). It is also well known that the protein nutritive value of sunflower by-products depends on the oil extraction process, variety of sunflower and proportion of the hulls removed during processing (Schinghoethe et al., 1977). However, there is scarce of information available about the amount of rumen degradable protein and intestinal digestibility of sunflower cakes, largely used as a protein sources for ruminants in Bulgaria. Therefore, the present study was taken up to estimate the DM and CP ruminal degradation kinetics and intestinal digestibility of SFC's produced in our country by the use of *in situ* and *mobile bag* techniques.

MATERIAL AND METHODS

Animals and samples

The animal experiment was carried out with consistency of Bulgarian legislation in field of the animal welfare and with the respect of the Bulgarian Food Safety Agency (*License № 126 registered in BFSAs*).

Three non-lactating Jersey cows with an average body weight of 436 ± 18 kg, each fitted with a rumen and T-duodenal cannulas, were used for evaluation of the rumen degradability and intestinal digestibility of DM and CP in SFC samples from different producers. During the adaptation and experimental periods cows were fed maintenance level ration of 80% roughages (63.6% alfalfa hay and 16.4% barley straw) and 20% concentrate (30.5% corn, 26.5% barley, 23.0% wheat bran, 17.0% SFC and 3% mineral and vitamin supplements). Cows received their assigned ration at approximately 8.00 h and 16.00 h. Six different samples of SFC were

collected from the biggest sunflower processing companies, members of the Oilseed Oil Producers Association in Bulgaria (*numbers indicate the individual factory*): „Gaytek“ Ltd., Nova Zagora (SFC1), „Dekadens“ Ltd., Bqla (SFC2), „Rosa“ Ltd., Popovo (SFC3), Olimex” Ltd., Vidin (SFC4), „Zlaten luch” Ltd., Dolni Dubnik (SFC5), „Faustina group”, Ltd., Montana (SFC6).

In situ procedures

Samples of each batch of SFC (approximately 2.5 g) were placed in a polyester dacron bags which were 5 cm ×10 and were prepared by double sewing of a nylon fabric with pore size 45 µm (SEFAR® PET 1500, 9410 Heiden, Switzerland) using a polyester thread.. Tested SFC was incubated in the rumen for 0, 2, 4, 8, 16, 24 and 48 in 6 replications. Immediately after removal from the rumen, the bags were carefully washed by hand under running tap water until the water remained clear. Then, the bags were dried at 65°C for 48 h and the DM content of the residue was determined at 105°C for 2 h.

Mobile bag technique

The polyester bags (4 x 8 cm with pore size 16 µm, SEFAR® PET 1500, 9410 Heiden, Switzerland) were filled with *ca.* 1 g of SFC previously incubated for 16 h in the rumen as described above. Samples were soaked for 1 h in HCl solution (pH 2.4) and thereafter incubated in HCl-pepsin solution (pH 2.4) for 2 h at 40 °C. The bags were inserted into the proximal duodenum by a T-shaped duodenal cannula almost 2 hours after morning feeding (14 bags per cow per day with 30 min. interval between the individual insertions). The bags were recovered in feces by rinsing with cold water through a large sieve. Finally, the bags were washed, dried (65 °C for 48 h and 105°C for 2 h) and weighed for the DM determination in residues.

Chemical analysis

Each SFC sample was milled to pass through a 2-mm screen. The chemical composition of the SFC was determined according to the AOAC (2007) (Table 1). The nitrogen content of the residues incubated in the rumen and passed through the intestine were assessed using an automatic N analyzer (Kjeltec™ 8400 Analyzer Unit, FOSS, DK-3400 Hillerod, Denmark). The CP content of the DM was determined by multiplying the percentage of nitrogen with a factor of 6.25.

Table 1. Chemical composition of sunflower meals, (% of DM)

Parameters	SFC1	SFC2	SFC3	SFC4	SFC5	SFC6	Mean (SD)
Dry matter, %	92.9	94.3	95.9	95.4	95.5	94.9	94.8 (1.023)
Crude protein (CP)	29.2	31.6	33.9	29.7	29.3	31.5	30.9 (1.853)
Crude fiber (CF)	15.4	20.2	16.2	18.6	22.6	18.1	18.5 (2.653)
Ether extract (EE)	12.4	9.23	8.55	11.4	10.6	9.67	10.3 (1.435)
Nitrogen free extract (NFE)	37.0	32.4	33.9	33.2	30.9	33.5	33.6 (1.999)
Crude ash	6.01	6.58	7.5	7.19	6.55	7.28	6.85 (0.560)
Ca	0.48	0.65	0.52	0.58	0.63	0.53	0.57 (0.067)
P	0.94	1.08	1.34	1.24	0.92	1.08	1.10 (0.165)

Calculations and Statistical analysis

The degradation kinetics of DM and CP for each sample of SFC were fitted to the equation described by Orskov & McDonald (1979), using the Marquardt algorithm for non-linear regression procedure (SPSS ver. 23, Chicago, USA).

$$d = a + b(1 - \text{Exp}(-ct))$$

Where d is the degradability at time, a is the water-soluble and rapidly degradable fraction, b is the potentially degradable fraction, c is the rate of degradation of potentially degradable fraction and t is the time of incubation (h).

The Effective degradability (ED) of tested feeds was calculated by the equation:

$$\text{ED} = a + (b \times c)/(c + k)$$

Where k is the particle passage rate, which was assumed to be 0.045, 0.06 and 0.08 h⁻¹.

The values for Protein truly digestible in small intestine (PDI) and Protein balance in the rumen (PBR) were calculated according to the Bulgarian protein system (Todorov et al. 2007) by the following equations:

$$\text{PDI} = 1.11 \times \text{CP}(1 - \text{ED}) \times \text{IDRUP} + 0.093 \times \text{FOM} + 4$$

$$\text{PBR} = \text{CP} \times (\text{ED} - 0.1) - 0.145 \times \text{FOM}$$

Where IDRUP is the intestinal digestibility of rumen undegraded protein (as part of 1), FOM is the fermented organic matter (as g/kg SFC) and ED is effective degradability (as part of 1). FOM is accepted to be equal for all SFC batches (517 g/kg, according to Todorov et al. 2007, for well dehulled SFC)

The data were analyzed for the fixed effect of protein source using the PROC GLIMMIX of SAS (2002-2012; SAS Institute Inc., Cary, NC). The significance was declared at $P < 0.01$.

RESULTS AND DISCUSSION

Chemical composition of SFC

The CP content of the sunflower cake's samples ranged from 29.2% at SFC1 to 33.9% at SFC3. The oil content of the sunflower cake in the form of ether extract listed in Table 1 is relatively high due to the sunflower seed's treatment technology (cold or hot pressing). In the present study, the

percentage of ether extracts ranged from 8.55% (SFC3) to 12.4% (SFC1). The results obtained at the present experiment are consistent with the levels of variability reported by Todorov et al., (2007).

Degradation parameters of DM

A huge variability was observed at the DM degradability parameters of sunflower cakes (Table 2). The rapidly degradable fraction *a* of DM was lowest at SFC2 (15.8%), and significantly higher was at SFC6 (25.8%) ($P < 0.01$). The potentially degradable fraction *b* ranged from 51.2% to 63.4%, and was significantly lower for SFC5 compared to all other samples ($P < 0.01$). Mohammadabadi et al. (2008) found that both DM fractions *a* and *b* of a high fat sunflower meal had the same values (39.0%), but neither the oil fat content of the raw material nor the processing technology were mentioned. We can speculate that it was a sunflower cake since it was reported as a high fat sunflower by-product and the values found at above noted study were inconsistent with our results. That difference as well the observed variability among all tested sunflower cakes could be explained both by the variation of the processing technologies and some methodological discrepancies such as bag pore size and substrate particle size (Nocek et al., 1979; Wadwa et al., 1998).

Table 2. Dry matter degradation parameters and effective degradability of dry matter of sunflower cakes at different rumen passage rates ($k = \% \cdot h^{-1}$)

Parameters	SFC1	SFC2	SFC3	SFC4	SFC5	SFC6	SEM	P value
a, %	16.3 ^{cd}	15.8 ^d	21.2 ^b	21.3 ^b	18.4 ^c	25.8 ^a	1.037	<0.01
b, %	56.2 ^b	54.2 ^b	63.4 ^a	54.8 ^{bc}	51.2 ^d	53.7 ^c	1.094	<0.01
c (h⁻¹)	0.082 ^b	0.088 ^b	0.041 ^c	0.148 ^a	0.095 ^b	0.135 ^a	0.0123	<0.01
Effective degradability of DM, %								
for $k=0.045$	52.6 ^{cd}	51.3 ^d	51.5 ^{cd}	63.4 ^b	53.2 ^c	66.0 ^a	0.049	<0.01
for $k=0.06$	48.8 ^{cd}	48.1 ^d	47.0 ^d	60.3 ^b	49.8 ^c	62.9 ^a	0.862	<0.01
for $k=0.08$	44.8 ^{cd}	44.2 ^d	42.8 ^d	56.9 ^b	46.2 ^c	59.5 ^a	0.875	<0.01

^{a-d} Means within a row lacking common superscript differ significantly at $P < 0.01$
SEM – standard error of the mean

The effective DM degradability of SFC at three different rumen passage rates was significantly higher for SFC6 and for SFC2 and SFC3 was lower compared to all other samples ($P < 0.01$). The values for effective DM degradability at the mean rumen outflow rate ($k = 0.06$) were lower compared to the results found by Mohammadabadi et al. (2008)

Degradation parameters of CP

The CP degradability of sunflower cakes estimated by *in situ* technique showed that the washable fraction *a* of CP was within the range of 23.7%

(SFC5) to 34.1% (SFC4) (Table 3). In contrast, the potentially degradable CP fraction b was the lowest at SFC4 - 61.0%, and highest at SFC5 - 71.0% ($P < 0.01$).

As a result of the observed differences among the relevant parameters it was found a large variation of the SFC's effective degradability of CP. At the lowest rumen passage rate, $k = 0.045$, the value for CP effective degradability of SFC6 (79.9%) was significantly higher compared to all other sunflower cakes samples ($P < 0.01$).

All the results for the ED of the rest of the samples obtained at different passage rates were also relatively high (Table 3). At a passage rate of $k=0.06$, Chrenkova et al., (2010) found values (69.0%) similar to our results for the CP effective degradability of sunflower cake. Maskalova et al., (2014) reported a significantly lower value (53.1%) for the ED of a sunflower cake with ether extract amounted to 7.94%.

Overall, the effective degradability of the CP of all tested samples of sunflower cake is over 60-65% and is approximately equal to the CP degradability of sunflower meal (Nedelkov et al., 2017).

Table 3. Crude protein (CP) degradation parameters and effective degradability of CP of sunflower cakes at different rumen passage rates ($k = \% \cdot h^{-1}$)

Parameters	SFC1	SFC2	SFC3	SFC4	SFC5	SFC6	SEM	P value
a, %	30.1 ^{bc}	27.1 ^{cd}	24.9 ^{de}	34.1 ^a	23.7 ^e	31.8 ^{ab}	1.149	<0.01
b, %	65.7 ^b	69.8 ^a	61.5 ^c	61.0 ^c	71.0 ^a	63.7 ^{bc}	1.030	<0.01
c (h⁻¹)	0.079 ^c	0.054 ^d	0.085 ^{bc}	0.098 ^b	0.083 ^{bc}	0.138 ^a	0.0117	<0.01
Effective degradability of CP, %								
for $k=0.045$	72.0 ^c	65.2 ^d	65.0 ^d	75.8 ^b	69.8 ^c	79.9 ^a	1.24	<0.01
for $k=0.06$	67.4 ^c	60.2 ^d	60.8 ^d	71.9 ^b	65.0 ^c	76.3 ^a	1.17	<0.01
for $k=0.08$	62.7 ^c	55.3 ^d	56.5 ^d	67.6 ^b	60.0 ^c	72.2 ^a	1.05	<0.01

^{a-e} Means within a row lacking common superscript differ significantly at $P < 0.01$
SEM – standard error of the mean

Intestinal digestibility of DM and CP

The values for the intestinal digestibility of the DM in an undegraded residue after 16 h of rumen incubation presented at Table 4 were significantly lowest at SFC6 - 36.5%, and the highest at SFC3 - 46.9% ($P < 0.01$).

The results found for intestinal digestibility of CP ranged from 82.4% (SFC6) to 94.0% (SFC3) being significantly higher for SFC3 compared to all other samples ($P < 0.01$). Maskalova et al. (2014) also reported a high CP intestinal digestibility of sunflower cake (90.2 ± 0.1), although their results were obtained by *in vitro* procedure.

Table 4. Dry matter and crude protein intestinal digestibility of the sunflower cake's residue after 16 h of rumen incubation

Parameters	SFC1	SFC2	SFC3	SFC4	SFC5	SFC6	SEM*	P value
Intestinal digestibility of IDM, %	37.9 ^{bc}	38.0 ^{bc}	46.9 ^a	39.8 ^b	39.7 ^b	36.5 ^c	0.457	<0.01
Intestinal digestibility of ICP, %	88.3 ^b	90.7 ^b	94.0 ^a	91.2 ^b	86.2 ^{bc}	82.4 ^c	0.363	<0.01

^{a-d} Means within a row lacking common superscript differ significantly at P<0.01

SEM – standard error of the mean

IDM – Intestinal dry matter

ICP – Intestinal crude protein

Calculation of protein digestible in intestine (PDI) and protein balance in the rumen (PBR) values

Hedqvist and Uden (2006) pointed out that the estimation of protein degradability in the rumen is one of the essential steps in feed evaluation systems to predict the nutritional requirements of ruminants. Considering that and using also the values for intestinal digestibility obtained at the present study, we were able to calculate the results for the main parameters of the protein nutritional value of SFC (PDI and PBR). The final values of both PDI and PBR, at different rumen outflow rates, are presented in Table 5.

There are significant differences among SFCs from different processing plants. The values for PDI were slightly higher compared to those reported by Todorov et al., (2007), but the calculated results for PBR differed significantly from the values indicated in the above mentioned Handbook of Animal Nutrition, which largely used in Bulgaria for formulating diets for ruminants.

The variations in the present study may be due to many factors, including agronomic conditions, processing technologies and the different varieties of sunflower seeds used for oil extraction.

Table 5. Protein digestible in intestine (PDI) and protein balance in the rumen (PBR) values of 1 kg dry matter of sunflower cakes calculated at different outflow rates (k)

Feed-stuffs	SFC1	SFC2	SFC3	SFC4	SFC5	SFC6	Average	Todorov et al. 2007
PDI								
k = 0.045	136 ^{bc}	161 ^{ab}	175 ^a	123 ^c	141 ^b	113 ^c	141	-
k = 0.06*	145 ^{bc}	179 ^{ab}	191 ^a	137 ^c	150 ^b	120 ^c	154	134
k = 0.08	159 ^{bc}	194 ^{ab}	206 ^a	149 ^c	164 ^b	132 ^c	168	-
PBR								
k = 0.045	106 ^b	99.6 ^b	112 ^b	120 ^{ab}	101 ^b	145 ^a	114	-
k = 0.06*	92.5 ^b	83.9 ^b	97.6 ^b	109 ^{ab}	86.5 ^b	134 ^a	101	166
k = 0.08	78.7 ^b	68.3 ^b	82.9 ^b	96.0 ^{ab}	71.7 ^b	121 ^a	86.4	-

^{a-c} Means within a row lacking common superscript differ significantly at P<0.01.

* PDI and PBR values were calculated using 517 g FOM for well dehulled SFC according to data published by Todorov et al., (2007).

CONCLUSIONS

Although nutrient composition of SFCs from different processing plants is highly variable, the protein degradability and digestibility values obtained at the present study can be used in formulating rations for ruminant animals. The observed differences could be mainly attributed to the different degree of squeezing the oil from sunflower seeds and variation of the quality of raw materials.

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