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In situ ruminal degradability and intestinal digestibility of dry matter and crude protein of low-protein fraction from sunflower meal

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Abstract

The objective of this study was to evaluate rumen degradation kinetics and intestinal digestibility of low-protein fraction from sunflower meal (LPSFM). 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. Five samples of LPSFM from different batches produced in 20-d intervals were collected and incubated in the rumen of the cows for 0, 2, 4, 8, 16, 24 and 48 h in 6 replicates. The average CP content of LPSFM was 175 (SD = 6.98) g/kg. Data were fitted to nonlinear regression model and analyzed statistically using SPSS. The soluble or rapidly degradable fraction *a* of LPSFM dry matter (DM) ranged from 105 to 137 (SD = 11.3) g/kg. The effective degradability of DM at assumed rumen outflow rate of 0.06/h ranged from 293 to 338 (SD= 16.3) g/kg. Fraction *a* of CP ranged from 124 to 189 (SD = 23.9) g/kg. Effective degradability of LPSFM CP at rumen outflow rate of 0.06/h ranged from 515 to 560 g/kg (SD = 16.2). Intestinal digestibility of LPSFM (after 16-h rumen incubation) DM and CP measured by the mobile bag technique varied from 318 to 349 g/kg (SD = 10.5) and from 766 to 820 g/kg (SD = 17.7), respectively. The average estimated Protein Digestible in the small Intestine (PDI) and Rumen Protein Balance (RPB) of the LPSFM samples analyzed in the current study, following equations in the Bulgarian feed evaluation system (at 0.06/h outflow rate), were 113 (SD = 1.67) g/kg and 125 (SD = 14.9) g/kg, respectively. The protein degradability and digestibility values obtained in this experiment can be effectively used in formulating rations for ruminant animals.

Keywords*:* rumen degradability; intestinal digestibility; low-protein fraction sunflower meal

Abbreviations: PDI, protein digestible in (small) intestine; RPB, rumen protein balance; CP, crude protein; DM, dry matter; EE, ether extract; ED, effective degradability; LPSFM, low protein fraction from sunflower meal; RUDM, ruminally-undegraded dry matter; RUCP, ruminally-undegraded crude protein; SD, standard deviation; CV, coefficient of variation;

Introduction

In most of the Eastern European countries sunflower meal is very important and cheap high-protein source for ruminants. Sunflower meal is the by-product of the extraction of oil from sunflower seeds and consists of the residue of the sunflower kernels and seed hull. In terms of production, it is the 4th most important oil meal after soybean meal, rapeseed meal and cottonseed meal (Oil World, 2011). There is a huge variety of products available on the market,

from low-quality fractions to high-protein meals containing 46 – 50% crude protein. The quality of sunflower meal depends on sunflower variety (seed composition, hulls/kernel ratio, dehulling potential), agronomic and storage conditions, and processing technology (Golob et al., 2002; Nedelkov et al., 2019).

Sunflower seeds from oil types contain about 25-35% hulls, which can be separated before, or after oil extraction process. Separation of sunflower hull from the kernel by means of mechanical fractionation on the centrifugal separators is hindered due to presence of firm conglomerates adhering to the hull (Sredanovic et al., 2011). To separate adherent particles, conglomerates need to be disintegrated, but at the same time, excessive break-up of the hull is to be avoided (Sredanovic, 2007; Levic, 2009), or otherwise, the quality of the sunflower meal will be compromised (reduced crude protein content and increased crude fiber content). As one of the most important sources of valuable proteins in our country, sunflower meal processing technology has been recently upgraded by its separation into two fractions using repeatedly rolling, sifting, and blowing (Draganov, 2015). The high-protein fraction from sunflower meal (HPSFM) contains only 5 – 8% crude fiber and 46 – 50% crude protein, while the low-protein fraction (LPSFM – about 30% of processed SFM) contains 36 – 55% crude fiber and 17 – 23% protein. Previous research from our team to estimate the ruminal degradability and intestinal digestibility of HPSFM revealed that the degradation kinetics and protein feeding values were almost the same as in the original/typical SFM (Nedelkov et al., 2021). However, there are no data available about the exact protein nutritional value for ruminants of the high-cellulose and low-protein fraction of sunflower meal (LPSFM). Hence, the objective of this study was to evaluate the rumen degradability, intestinal digestibility, and protein nutritional value of LPSFM samples. We hypothesized that the high-cellulose content of LPSFM would adversely affect its nutritional value, which might be a significant consideration for its inclusion in ruminants' diets.

Materials and Methods

Animals and samples

All procedures involving animals in the current study were consistent with Bulgarian animal welfare legislation and in compliance with the Bulgarian Food Safety Agency regulations (*Registration license № 126).*

Three non-lactating Jersey cows with an average body weight of 436 ± 18 kg, fitted with a rumen (made from polycarbonate material with internal diameter = 12 cm) and T-shape duodenal cannula (polycarbonate material with internal diameter = 4.4 cm) were used in the experiment. During the adaptation (10 d) and experimental (15 d) periods, cows were fed at maintenance level a ration containing 800 g/kg roughages (63.6 % alfalfa hay and 16.4% barley straw) and 200 g/kg concentrate (30.5% ground corn grain, 26.5% ground barley grain, 23.0% wheat bran, 17.0% SFM and 3% mineral and vitamin premix). Cows were housed in the large ruminant facility of Trakia University's Research Center, Faculty of Veterinary Medicine, and were fed at approximately 8.00 h and 16.00 h. Feeding was ad libitum targeting 5% refusals.

Low-protein sunflower meal samples were collected from the first company, which implemented the new technology for mechanical processing of sunflower meal – Bonmix Ltd., Lovech, Bulgaria (LPSFM1 to LPSFM5, numbers indicate the individual batch). The individual batches were collected in an interval of at least 20 days.

Briefly, samples (approximately 2.5 g) of each batch of LPSFM were placed in a polyester Dacron bags, made by double sewing of a nylon fabric with pore size 16 μm (4×8 cm, SEFAR®PET 1500), which were later incubated in the rumen of the cows for 0, 2, 4, 8, 16, 24 and 48 h in duplicates (i.e., a total of 6 bags per incubation timepoint). Then, the bags were dried at 65°C for 48 h and DM content of the residue was determined by drying at 105°C for 2 h in a mechanical convection oven. Intestinal digestibility of LPSFM DM and CP was analyzed by the mobile bag technique following the procedure of Woods et al. (2003b). A full description of the in situ rumen

incubation and estimation of the small intestinal digestibility of ruminal-undegraded fraction can be found in Nedelkov et al. (2019).

Chemical analysis

Low-protein sunflower meal samples were ground to pass through a 2-mm screen. Dried samples were analyzed by wet chemistry methods EE (method 2003.05; AOAC International, 2006), ash (method 942.05; AOAC International, 2000), and minerals (method 985.01; AOAC International, 2000) (Table 1). Sunflower meal samples and bag residues were analyzed for N (Kjeltec™ 8400 Analyzer Unit, FOSS, DK-3400 Hillerod, Denmark) and CP was found as N × 6.25.

Calculations and Statistical analysis

Ruminal DM and CP degradation data were fitted to the exponential equation of Orskov & McDonald (1979), using the Marquardt algorithm for non-linear regression procedure (SPSS ver. 23, Chicago, USA).

$d = a + b\,(1 - \exp(-^{c\,t}))$

Where *d* is degradability (%) at time *t*, *a* is the soluble and rapidly degradable fraction of DM or CP, *b* is the potentially degradable fraction, *c* is the rate of degradation of fraction *b*, and *t* is the incubation time (h).

Effective degradability (ED) of DM and CP were calculated using the following equation (AFRC, 1993):

$ED = a + (b \times c)/(c + kp)$

Where *a*, *b*, and *c* are as specified above and *kp* is the passage rate, assumed at 0.05, 0.06, and 0.08 h^{-1}.

The values for Protein Digestible in the small Intestine (PDI) and Rumen Protein Balance (RPB) were calculated according to the Bulgarian protein system (Todorov et al. 2007).

Data were analyzed for the fixed effect of protein source using the GLIMMIX procedure of SAS (2002-2012; SAS Institute Inc., Cary, NC). Significance was declared at $P < 0.05$. Means are expressed as least squares means.

Results and Discussion

Chemical composition of LPSFM

The average CP content of tested LPSFM was 175 (SD = 6.98) g/kg, varied from 166 g/kg DM to 183 g/kg, while the crude fiber content was 411 (SD = 16.3) g/kg, ranging from 381 g/kg DM to 428 g/kg (Table 1). In our previous study, the average CP content of commercial sunflower meal samples collected from the seven biggest sunflower processing plants in Bulgaria produced after an average degree of dehulling of the seeds was 369 (SD = 23.5) g/kg, ranging from 337 g/kg DM to 397 g/kg (Nedelkov et al., 2019). This suggests that during the separation process, most of the sunflower hulls remained in the LPSFM.

Table 1. Dry matter content and chemical composition (g/kg dry matter or as indicated) of low-protein sunflower meal samples used in the study.

Parameters	Average	Minimum	Maximum	SD	CV, %
Dry matter, g/kg	932	926	938	4.62	0.49
Crude protein	175	166	183	6.98	3.99
Crude fiber	411	381	428	16.3	3.96
Ether extract	21.1	13.4	2.93	5.23	24.8
NFE	345	323	372	15.7	4.56
Ash	47.4	45.1	50.3	1.84	3.88
Ca	4.47	4.01	4.90	0.33	7.47
P	5.82	4.60	8.04	1.16	20.05

Rumen degradability of DM and CP

The soluble or rapidly degradable fraction *a* of LPSFM dry matter (DM) ranged from 105 to 137 (SD = 11.3) g/kg (Table 2). Higher variations were observed at the potentially degradable DM fraction *b*, wherein the results varied from 282 to 414 (SD = 48.1) g/kg among different samples. The effective degradability of DM at assumed rumen outflow rate of 0.06/h ranged from 293 to 338 (SD= 16.3) g/kg. After the separation process, a significant percentage of the hulls remained in the LPSFM (theoretically, about 85-90%). Thus, expectedly the fractions *a*, *b*, and overall DM degradability of LPSFM were much lower compared to the data for commercial SFM reported in some previous studies (Woods et al. 2003b; Nedelkov et al., 2019).

Fraction *a* of CP ranged from 124 to 189 (SD = 23.9) g/kg (Table 3). These values were lower compared to the results reported by others for washable CP fraction of common SFM (Habib et al., 2013; Gao et al., 2015). However, in terms of potentially soluble fraction *b* of the CP, the established results ranging from 749 to 801 (SD = 19.4) g/kg are in agreement with previously reported values for common SFM samples (Alcaide et al., 2003; Chrenkova et al., 2010; Nedelkov et al., 2019). Overall, the estimated effective degradability of LPSFM CP at a rumen outflow rate of 0.06/h was considerably low and ranged

Table 2. Ruminal degradation parameters and effective degradability of dry matter of low-protein fraction of sunflower meal (g/kg, or as specified).

Parameter	Average[a]	Minimum	Maximum	SD	CV, %
a,[b]	119	105	137	11.3	9.48
b,[b]	319	282	414	48.1	15.1
c[b]/h^{-1}	0.091	0.077	0.104	0.01	10.9
kp[c] = 0.045/h^{-1}	332	312	366	19.7	5.94
kp = 0.06/h^{-1}	310	293	338	16.3	5.26
kp = 0.08/h^{-1}	288	273	308	13.2	4.60

[a] *P < 0.01 for the main effect of LPSFM sample.*

[b] *a, b, and c are soluble, potentially degradable fraction and rate of degradation of fraction b, respectively;*

[c] *kp is the passage rate from the rumen;*

N = 30.

Table 3. Ruminal degradation parameters and effective degradability of crude protein of low-protein fraction of sunflower meal (g/kg, or as specified).

Parameters	Average[a]	Minimum	Maximum	SD	CV, %
a,[b]	161	124	189	23.9	14.8
b,[b]	778	749	801	19.4	2.49
c[b]/h^{-1}	0.056	0.054	0.058	0.02	3.29
kp[c] = 0.045/h^{-1}	591	573	615	14.7	2.47
kp = 0.06/h^{-1}	536	515	560	16.2	3.01
kp = 0.08/h^{-1}	480	459	506	17.1	3.57

[a] *P < 0.01 for the main effect of LPSFM sample.*

[b] *a, b, and c are soluble, potentially degradable fraction and rate of degradation of fraction b, respectively;*

[c] *kp is the passage rate from the rumen;*

N = 30

from 515 to 560 (SD = 16.2) g/kg. It is evident that the higher amount of hulls impregnated by lignin and silica and remaining in LPSFM after separation will result in lower degradability. However, in previous work, it was highlighted that it could be more efficient to remove the hulls partly from the LPSFM fraction and to increase CP content up to 25% (Nedelkov, 2023).

Intestinal digestibility of DM and CP

The intestinal digestibility of LPSFM DM (after 16-h rumen incubation) varied (*P < 0.01*) from 318 to 349 g/kg (SD = 10.5) g/kg (Table 4). The range in intestinal digestibility of LPSFM CP was from 766 to 820 (SD = 17.7) g/kg. The average intestinal digestibility of LPSFM RUCP (788 g/kg) was slightly lower compared to the previously reported data for CP intestinal digestibility of the typical SFM samples ranging from 838 to 899 g/kg (Woods et al., 2003b; Nedelkov et al., 2019). The remaining larger proportion of the hulls after the fractionation process is likely the main reason for the decreased digestibility of LPSFM previously incubated for 16 h in the rumen.

Calculation of Protein Digestible in the small Intestine (PDI) and Rumen Protein Balance (RPB)

The average estimated PDI and RPB (at 0.06/h outflow rate) of the LPSFM samples analyzed in the current study were 113 (SD = 1.67) g/kg and 16.6 (SD = 5.83) g/kg, respectively. These values are significantly lower compared to those reported in the Bulgarian feeding standards (Todorov et al., 2007; average of 148 and 184 g/kg for PDI and RPB, respectively), for common (not mechanically treated) SFM. As previously suggested, additional removal of the hulls to increase the CP content of LPSFM might be an excel-

Table 4. Dry matter and crude protein intestinal digestibility (g/kg) of low- protein fraction of sunflower meal (LPSFM) following a 16-h rumen incubation.

Parameter	Average[a]	Minimum	Maximum	SD	CV, %
Intestinal digestibility of RUDM[b]	329	318	349	10.5	3.18
Intestinal digestibility of RUCP[c]	788	766	820	17.7	2.25

[a] *P < 0.01 for the main effect of LPSFM sample.*

[b] *RUDM – Ruminally-undegraded dry matter*

[c] *RUCP – Ruminally-undegraded crude protein*

Table 5. Protein digestible in intestine (PDI) and rumen protein balance values (RPB) of 1 kg dry matter of low-protein fraction of sunflower meals (LPSFM) calculated at different outflow rates (g/kg).

Parameter	Average[a]	Minimum	Maximum	SD	CV, %
PDI					
kp[b] = 0.045/h^{-1}	105	103	107	1.53	1.46
kp = 0.06/h^{-1}	113	111	116	1.67	1.47
kp = 0.08/h^{-1}	122	118	124	1.81	1.49
RBP					
kp = 0.045/h^{-1}	26.4	18.8	34.5	5.93	22.5
kp = 0.06/h^{-1}	16.6	9.15	24.4	5.83	35.2
kp = 0.08/h^{-1}	6.76	-0.15	13.6	5.36	79.3

[a] *P < 0.01 for the main effect of LPSFM sample.*

[b] *kp is the passage rate from the rumen;*

N = 30.

lent alternative for its use in ruminant nutrition. Moreover, further inclusion of molasses, macro-, and microminerals, to such an extra fractionated LPSFM will convert it to an excellent compound feed for dry cows, or other categories of low-productive ruminants.

Conclusions

The protein nutritional values (PDI and RPB) obtained in this experiment can be effectively used in formulating rations for ruminant animals. The remaining larger proportion of the hulls after the fractionation process is likely the main reason for the decreased digestibility of LPSFM. There is some room for improvement of the protein nutritional value of LPSFM by additional partial removal of the hulls and subsequent inclusion of molasses, macro-, and microminerals.

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