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Variability in rumen degradability and intestinal digestibility of sunflower meals protein

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Abstract

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The aim of this study was to evaluate variability in the nutritional value of sunflower meal (SFM) protein by determining both its ruminal degradability and intestinal digestibility. Three non-lactating Jersey cows fitted with a rumen and T-type duodenal cannulas were used to estimate rumen degradability and intestinal digestibility of SFM dry matter (DM) and crude protein (CP). Samples of SFM were collected from seven sunflower processing plants (SFM1 to SFM7). The SFM were incubated in the rumen of the cows for 0, 2, 4, 8, 16, 24 and 48 h in 6 replications. The soluble or rapidly degradable fraction *a* of SFM DM ranged from 208 to 264 (SD = 21.2) g/kg. The effective degradability of DM at assumed rumen outflow rate of 0.06/h ranged from 533 to 585 (SD= 22.1) g/kg. Fraction *a* of CP ranged from 208 to 279 (SD = 26.8) g/kg. Effective degradability of SFM CP at rumen outflow rate of 0.06/h was relatively high and ranged from 639 to 723 g/kg (SD = 29.4). Intestinal digestibility of SFM DM and CP measured by mobile bag technique varied from 387 to 473 g/kg (SD = 25.9) and from 863 to 930 (SD = 22.5), respectively. Estimated Protein Digestible in the small Intestine (PDI) was 159 to 201 (SD = 14.1) g/kg DM, and Protein Balance in the Rumen (PBR) varied from 105 to 146 (SD = 14.9) g/kg DM. This study showed that protein degradability and digestibility of commercial SFM samples varied considerably, suggesting that nutritive value of SFM protein will fluctuate depending on agronomic conditions, sunflower variety, and processing technology. This variability should be considered when formulating rations with SFM for ruminant animals.

Keywords: rumen degradability; intestinal digestibility; protein nutritive value; sunflower meal
Abbreviations: PBR – protein balance in the rumen; CP – crude protein; DM – dry matter; EE – ether extract; ED – effective degradability; SFM – sunflower meal; PDI – protein digestible in (small) intestine; RUDM – ruminally undegradable dry matter; RUCP – ruminally undegradable crude protein; SD – standard deviation; CV – coefficient of variation

Introduction

In parts of Europe, sunflower meal (SFM) is one of the most important and inexpensive protein sources for ruminants. Inclusion rate of SFM in ruminant rations is usually limited because of its high protein degradability in the rumen (Veresegyhazy and Fekete, 1990). Anderson et al. (2000) pointed out that SFM has the highest degradation rate of all basic protein meals used in ruminant nutrition. According to Kamalak et al. (2005) the rumen degradability of SFM CP, at an assumed passage rate of 0.06/h, was 470 g/kg while Woods et al. (2003b), Chrenkova et al. (2010), Marghazani

et al. (2013), and Gao et al. (2015) reported degradability values of 800 g/kg and higher. The large variability in SFM CP degradation rates among individual experiments could be explained with differences in type of SFM samples and differences in the in situ methodology, i.e. ratio of bag surface area to sample size, sample particle size, etc. (Wadwa et al., 1998; Habib et al., 2013). Additionally, processing plants may apply various modifications to the traditional manufacturing process based on experience, know-how, and availability of resources. For example, some processors do not separate the hulls before oil extraction, while others remove different parts of the hulls. Therefore, composition and nutritive value of SFM offered to livestock producers can vary significantly. In addition, there is limited data available regarding in vivo intestinal digestibility of SFM protein. Hence, the objective of this experiment was to evaluate the variability in rumen degradability, intestinal digestibility, and nutritional value of SFM samples collected from commercial sunflower processing plants. We hypothesized that variability in SFM protein degradability and digestibility, and consequently its estimated nutritive value, is large enough to be an important consideration in formulating rations for ruminant animals.

Materials and Methods

Animals and samples. All procedures involving animals in this experiment 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.

Sunflower meal samples were collected from the seven biggest sunflower processing plants in Bulgaria (SFM1 to SFM7, numbers indicate the individual plant/batch), all members of the Oilseed Oil Producers Association of Bulgaria.

In situ rumen incubation. Samples (approximately 2.5 g) of each batch of SFM were placed in a polyester Dacron bags which were 5×10 cm and were prepared by double sewing of a fabric with pore size 45 μm (SEFAR® PET 1500, 9410 Heiden, Switzerland) using a polyester thread. Bags were 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 time-point). Bags were placed in the rumen sequentially and removed at the same time. Immediately after removal from the rumen, 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 DM content of the residue was determined by drying at 105°C for 2 h in a mechanical convection oven.

In situ small intestinal digestibility of ruminal undegraded fraction. Intestinal digestibility of SFM DM and CP was analyzed by the mobile bag technique following the procedure of Woods et al. (2003c). Polyester bags, made by double sewing of a nylon fabric with pore size 16 μm (4×8 cm, SEFAR® PET 1500), were filled with 1 g of SFM previously incubated for 16 h in the rumen as described above. Samples were soaked for 1 h in 1 N HCl solution (pH 2.4) and thereafter incubated in HCl/pepsin solution (pH 2.4) for 2 h at 40°C. Bags were then inserted into the proximal duodenum of the cows via the duodenal cannula, approximately 2 h after the morning feeding (14 bags per cow per day with 30 min interval between the individual bags' insertions). The bags were recovered by rinsing fecal matter with cold water through a large sieve (12 mm openings). Bags were washed, dried (at 65°C for 48 h and then at 105°C for 2 h) and weighed for DM and CP analysis of the residues.

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

Parameters	Average	Minimum	Maximum	SD	CV, %
Dry matter, g/kg	918	911	929	6.91	7.52
Crude protein	369	337	397	23.5	63.7
Ether extract	16.9	12.4	22.3	2.91	17.2
Ash	79.1	67.0	87.7	6.24	7.88
Ca	5.94	4.16	9.14	1.72	29.0
P	13.3	10.3	17.1	2.19	16.5

Chemical analysis. 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 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).

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 SFM. The average CP content of tested SFM was 369 (SD = 23.5) g/kg, ranging from 337 g/kg DM to 397 g/kg (Table 1). This indicates that all SFM were produced after an average degree of dehulling of the seeds (Todorov et al., 2007).

Rumen degradability of DM and CP. The rapidly-degradable fraction of DM varied from 208 to 264 (SD = 21.2) g/kg (Table 2). Potentially degradable fraction *b* ranged from 516 to 600 (SD = 27.7) g/kg. Values for soluble DM fraction *a* reported by Woods et al. (2003a), 284±47.2 g/kg and Marghazani et al. (2013), 271±13.5 g/kg are consistent with our results. Values for potentially degradable DM, fraction *b*, found in this study are higher than SFM values reported by others (Alcaide et al., 2003; Kamalak et al. 2005; Gao et al., 2015). According to Kamalak et al. (2005), variation in protein degradability of several protein sources, including SFM, was mainly associated with differences among laboratories in the methods used for sample preparation and processing, and in the bags used for *in situ* incubation. Madsen and Hvelpund (1994) previously also noted that in some cases differences in protein degradabilities between laboratories are too large. Another source of variation is the animal species (cattle vs. sheep) used in *in situ* experiments (Orskov et al., 1983).

Table 2. Ruminal degradation parameters and effective degradability of dry matter of commercial sunflower meal (SFM) samples (g/kg, or as specified)

Parameter	Average[a]	Minimum	Maximum	SD	CV
a[b]	248	208	264	21.2	8.51
b[b]	553	516	600	27.7	5.01
c[b]/h^{-1}	0.081	0.057	0.121	0.018	22.9
kp[c] = 0.05/h^{-1}	599	572	624	22.3	3.72
kp = 0.06/h^{-1}	562	533	585	22.1	3.93
kp = 0.08/h^{-1}	522	494	544	21.1	4.03

[a] $P < 0.01$ for the main effect of SFM 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 = 42

The average value (562 g/kg) for effective DM degradability at mean rumen outflow rate of 0.06/h is consistent with the results for SFM reported by others (Woods et al., 2003a; Mondal et al., 2008; Gao et al., 2015).

The washable fraction *a* of CP varied from 208 to 279 g/kg (Table 3). Similar results for SFM were reported by Gao et al. (2015; 274 g/kg) and Alcaide et al. (2003; 259 g/kg), whereas Woods et al. (2003b) and Mondal et al. (2008) reported values that reached 380 to 390 g/kg.

As expected, estimated effective degradability of SFM CP decreased with increasing outflow rates, reaching a maximum of 762 g/kg at rumen passage rate of 0.05/h. Other

Table 3. Ruminal degradation parameters and effective degradability of crude protein of commercial sunflower meal (SFM) samples (g/kg, or as specified)

Parameters	Average[a]	Minimum	Maximum	SD	CV
a,[b]	263	208	279	26.8	10.2
b,[b]	724	652	772	36.8	5.08
c[b]/h^{-1}	0.083	0.070	0.129	0.019	22.9
kp[c] = 0.05/h^{-1}	726	693	762	26.1	3.59
kp = 0.06/h^{-1}	677	639	723	29.4	4.34
kp = 0.08/h^{-1}	626	604	681	32.3	5.16

[a] $P < 0.01$ for the main effect of SFM 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 = 42

authors reported considerably higher ED of SFM CP (750 to 900 g/kg; Alcaide et al., 2003; Chrenkova et al., 2010; Ganbari et al., 2015). According to NRC (2001), the proportion of rumen undegraded protein (RUP) of the total protein in SFM is 2 to 2.5 times lower than in soybean meal. Data from this study and other published reports indicate that there is a large variation in SFM CP degradability as a result of variation in SFM composition, preconditioning temperature, pressing, and removal of solvent after extraction of the oil.

Overall, differences in the manufacturing process, particularly excessive heat treatment (Ljokjel et al., 2000), are likely the main reason for the observed differences in SFM protein degradability. Heat or chemical treatment have been shown to decrease ruminal degradability of SFM (Mohammadabadi et al. 2009; Diaz-Royon et al., 2016) suggesting that feeding value of SFM protein could be substantially increased for ruminant animals if oil-extracting plant apply steam-heating or toasting. Another effective technology for decreasing SFM protein degradability in the rumen is gamma radiation (Ganbari et al. 2015). This latter treatment has also other advantages such as: decreased environmental pollution (Mani and Chandra, 2003), decontamination of the feed (Shawrang, 2006), and increased intestinal digestibility (Mani and Chandra, 2003; Song et al., 2009; Ganbari et al., 2015).

Intestinal digestibility of DM and CP. The intestinal digestibility of SFM DM varied ($P < 0.01$) from 387 to 473 (SD = 25.9) g/kg (Table 4). The range in intestinal digestibility of SFM CP was from 863 to 930 (SD = 22.5) g/kg. Average intestinal digestibility of SFM CP in the current study (899 g/kg) was comparable to values reported in the literature using an *in vitro* three-step incubation procedure (Weisbjerg et al., 1996; Woods et al., 2003c; Gao et al., 2015). All sunflower plants from which samples for this study were obtained applied similar processing technology of hot-pressing sunflower seeds followed by a solvent-extraction of the pressed, partially defatted cake. Deviation from this general protocol, however, likely resulted in the observed variation in SFM protein degradability and consequently, estimated feeding value. In addition to processing, seed variety and environmental factors (fertilization and climatic conditions) can also affect SFM degradability.

Table 4. Dry matter and crude protein intestinal digestibility (g/kg) of sunflower meal (SFM) samples following a16-h rumen incubation

Parameter	Average[a]	Minimum	Maximum	SD	CV
Intestinal digestibility of RUDM[b]	426	387	473	25.9	6.08
Intestinal digestibility of RUCP[c]	899	863	930	22.5	2.50

[a] $P < 0.01$ for the main effect of SFM sample
[b] RUDM – Ruminally-undegraded dry matter
[c] RUCP – Ruminally-undegraded crude protein

The average estimated PDI and PBR (at 0.06/h outflow rate) of the SFM samples analyzed in the current study were 181 (SD = 14.1) g/kg and 125 (SD = 14.9) g/kg, respectively. These values differ significantly from those reported in the Bulgarian feeding standards (Todorov et al., 2007; average of 148 and 184 g/kg for PDI and PBR, respectively), suggesting that a revision of the protein feeding value of SFM may be necessary.

Conclusions

This study showed that protein nutritional value of sunflower meals varies significantly among sunflower processing plants. This variation is a result of multiple factors, including sunflower variety, agronomic and environmental conditions, and processing technologies. It is recommended that PDI and PBR values for SFM protein in the current Bulgarian feeding standards are updated with average values determined in this experiment.

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