

RUMEN DEGRADATION CHARACTERISTICS OF GLUCOSE-TREATED CANOLA MEAL AND CANOLA SEED

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Summary. The study was conducted to determine the effects of glucose treatment of canola meal (CM) and canola seed (CS) on rumen degradability characteristics of dry matter (DM), organic matter (OM) and crude protein (CP) in four ruminally cannulated mature (2 years old) Merino rams. Canola meal and canola seed were treated first with water then heat, treated first with water then heat plus 2% glucose and treated first with water then heat plus 3% glucose. Crude protein degradability value of CM was reduced ($p<0.001$) by 3% glucose treatment at all rumen incubation times. Effective DM and OM degradabilities of CM were decreased ($p<0.001$) by 2 and 3% glucose treatments. Effective crude protein degradability of CM treated with 3% glucose was lower ($p<0.001$) than untreated CM, CM treated with water+heat and 2% glucose. While effective DM ($p<0.05$) and OM ($p<0.001$) degradabilities were reduced only in CS treated with 3% glucose, this effect was not determined for effective CP degradability of CS in any treatment. In conclusion, there was an effect in reducing of effective CP degradability of CM treated with 3% glucose although glucose treatment was ineffective on reducing of effective CP degradability value of CS.

Keywords: glucose treatment, canola meal, canola seed, rumen degradability.

GLIUKOZĖS ĮTAKA RAPSŲ IR JŲ SĖKLŲ VIRŠKINAMUMUI AVIŲ DIDŽIAJAME PRIESKRANDYJE

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Santrauka. Atliktas bandymas nustatyti gliukozės įtaką rapsų (R) ir jų sėklų (RS) virškinamumui avių didžiajame prieskrandyje. Tirta gliukozės įtaka R ir RS sausosios medžiagos (SM), organinės medžiagos (OM) ir proteinų (P) virškinamumui. Bandymas atliktas su keturiais suaugusiais (2 metų) Merinosų veislės aviniais, kuriems į didžiuosius prieskrandžius chirurginiu būdu implantuotos kaniulės. Rapsai ir jų sėklos iš pradžių buvo plaunami vandeniu ir kaitinami (1 variantas). Po savaitės avinai buvo šeriami pašaru, paruoštu papildomai plaunant bei kaitinant (2 variantas), o dar po savaitės šeriami R ir RS, kurie buvo plaunami bei kaitinami su 3 proc. gliukoze (3 variantas). Didžiojo prieskrandžio turinio mėginiai tyrimui buvo imami iš kaniulių avių didžiuosiuose prieskrandžiuose.

Nustatyta, kad, R papildžius 3 proc. gliukoze (3 variantas), proteinų virškinamumas didžiajame prieskrandyje ženkliai suprastėja ($p<0,001$). Taip pat ištirta, kad R ir RS sėklose esančių SM ir OM virškinamumas, papildžius 2 proc. ir 3 proc. gliukoze, statistiškai ženkliai suprastėja ($p<0,001$). Mūsų tyrimai parodė, kad R esančių proteinų virškinamumas, papildžius 3 proc. gliukoze (3 variantas), buvo statistiškai ženkliai prastesnis ($p<0,001$) palyginti su R ir RS, paveiktų vandeniu, karščiu ir 2 proc. gliukoze (2 variantas). Statistiškai ženklus SM ($p<0,05$) ir OM ($p<0,001$) suprastėjęs virškinamumas nustatytas tik tada, kai gyvuliai buvo šeriami RS su 3 proc. gliukoze. Panašaus poveikio nerasta tiriant RS esančių P virškinamumą pašare, paruoštame pagal visus tris variantus. Tyrimai parodė, kad 3 proc. gliukozė ženkliai sumažina R esančio proteino virškinamumą, tačiau neturi jokio poveikio RS esančio proteino virškinamumui.

Raktažodžiai: gliukozė, rapsai, rapsų sėklos, didysis prieskrandis, virškinamumas, avys.

Introduction. Proteins are degraded peptides, amino acids and finally ammonia during rumen fermentation (Chalupa, 1981). As some feed proteins degrade rapidly in rumen, rumen microorganisms are not be able to fit this

degradation and major part of nitrogen coming out after feed degradation leaves from rumen as ammonia (Sinclair et al., 1995). Nitrogen and nitrogen compounds which arise from feed proteins not having much degradation

characteristics are used efficiently by rumen microorganisms. The undegraded dietary protein may be supplied by decreasing the ruminal degradation and hence increasing the quantity of protein digested post ruminally (Mustafa et al. 2000). There are different chemical treatments of feedstuffs such as glucose, formaldehyde and xylose in reducing crude protein (CP) degradation in the rumen. Experiments demonstrated the loss of protein depending on ruminal fermentation may reduce when some of important plant protein sources as meals or lupine seeds treats with heat (Thomas et al., 1979; Nakamura et al., 1994; Benchaar and Moncoulon, 1993), formaldehyde (Nishimuta et al., 1974; Thomas et al., 1979; Mir et al., 1984) acetic acid (Robinson et al., 1994), tannic acid (Driedger and Hatfield, 1972) lignosulfonate (McAllister et al., 1993), xylose (Windschitl and Stern

1988; McAllister et al., 1993; Can and Yilmaz, 2002; Tuncer and Sacakli, 2003) and glucose (Sacakli et al., 2009). Glucose reacts with amino groups in proteins of feeds which are rich in protein and it constitutes bonds resistant to pH in rumen.

The aim of this study was to determine the effect of glucose treatment of canola meal and canola seed on dry matter (DM), organic matter (OM) and crude protein rumen degradability characteristics.

Materials and Methods. Four ruminally cannulated Merino rams (average weight 60 kg) were kept in the individual cages and fed a ration containing of 200 g concentrate (50% barley, 25% sunflower meal, 22% wheat bran, 1% salt, 1% dicalcium phosphate and 1% vitamin + mineral premix,) and 1kg alfalfa hay (Table 1), twice daily at 09.00 h and 16.00 h.

Table 1. **Chemical composition of the experimental feeds** (% DM basis)

Feeds	Organic matter	Crude protein	Ether extract	Crude ash	Crude fiber
Alfalfa hay	91.1	13.7	1.7	8.9	27.9
Concentrate mixture	96.4	17.1	2.6	3.6	9.8
Canola meal	93.8	41.3	3.0	6.2	10.0
Canola seed	94.6	24.0	40.1	5.4	6.7

After grinding the feed as dry product to cross 3 mm screen size, canola meal and canola seed were treated with water + heat (this treatment was applied to determine the effects of water and heat at 100 °C for 2 h without glucose) or with first water then heat plus 2% glucose and then heat plus 3% glucose. The DM of meals was determined by drying at 105 °C for 24 h, and thereafter sufficient water or mixtures of water and glucose (2 and 3%) were added to elevate the moisture content of samples to 25 % (McAllister et al. 1993). These samples were thoroughly mixed with each solution and heated for 2 h at 100 °C in a convection air oven.

Nylon bag technique was used to measure disappearance of DM, OM and CP of untreated and treated CM and CS in the rumen. Nylon bags (45 µm pore size; 9x14 cm bag size) containing 5 g of test samples were incubated in the rumen of each ram. Two bags of each type of samples were removed after 2, 4, 8, 16, 24 and 48 h of incubation in the rumen. Then bags were washed in running tap water until getting clear rinse water. Bags were dried at 60 °C for 48 h and weighed. The following equation describing by Orskov and McDonald (1979) was used for calculating of digestion kinetics and effective rumen degradability of DM, OM and CP.

$$p = a + b(1 - e^{-ct})$$

In the equation, p is the amount degraded at a time, a rapidly soluble fraction (%), b potentially degradable fraction (%), c constant rate of b disappearance (% h⁻¹), t incubation time (h).

Effective rumen degradability of DM, OM and CP were estimated using the equation of Orskov and McDonald (1979):

$$Pe = a + bc/k + c$$

Where Pe is the effective degradation (%), k the

fractional ruminal outflow rate, and a , b and c are defined above. Effective degradability was calculated with an estimated solid outflow rate from the rumen (k) of 0.05 h⁻¹

Chemical composition of experimental feeds and DM, OM and CP content of their washed residues after rumen incubation were determined according to the methods of AOAC (1984).

Rams are not statistically different for physiological and other traits such as health, ages, live weights etc. during experimental periods. Therefore, ordinary least square procedure was used to determine differences among the treatment groups for the rumen degradation characteristics of dry matter, organic matter and crude protein of untreated and treated CM and CS (Searle et al. 1992).

Statistical analyses of data were performed by repeated two-way linear model of SAS (Snedecor and Cochran 1980). The significance of differences between treatment means was compared by DUNCAN test (Duncan 1955). Statistical analyses were done using the SAS program (SAS 2007). Statements of statistical significance are based on $p < 0.05$.

Results. Chemical compositions of experimental feeds were shown in Table 1. Ruminal disappearances for rumen incubation times and rumen degradation characteristics of dry matter, organic matter and crude protein of canola meal and canola seed were shown in Tables 2, 3, 4 and 5, respectively.

Dry matter degradability values of CM treated with 2% and 3% glucose were lower ($p < 0.001$) after 8, 16, 24 and 48 h rumen incubation times than untreated CM. Glucose treatments, especially at the level of 3% reduced significantly OM and CP degradability of CM after all rumen incubation times compared to untreated CM (Table 2).

Table 2. **Disappearance of dry matter, organic matter and crude protein for rumen incubation times of canola meal (%)**

	Incubation times (h)					
	2	4	8	16	24	48
Dry matter						
CM	37.7b	44.0bc	54.0a	66.8a	73.8a	80.9a
CM + WH	40.9a	45.8a	54.0a	65.3b	72.1b	80.0ab
CM + % 2 G	40.4a	45.0ab	52.6b	63.3c	69.8c	78.0b
CM + % 3 G	38.2b	43.1c	51.2c	62.5c	69.4c	77.8b
	***	***	***	***	***	*
Organic matter						
CM	37.3a	44.0a	54.4a	67.1a	73.5a	79.5a
CM + WH	37.6a	43.0ab	51.9b	64.1a	71.4a	80.0ab
CM + % 2 G	36.7a	41.1b	48.5c	59.4b	66.7b	77.0b
CM + % 3 G	33.9b	37.6c	44.2d	54.2c	61.1c	71.4c
	*	***	***	***	***	***
Crude protein						
CM	54.4a	60.1a	70.3a	79.0a	84.0b	88.1b
CM + WH	53.0a	59.1a	68.8ab	80.8a	87.1a	93.2a
CM + % 2 G	53.4a	58.1a	65.8b	76.7b	82.3c	89.2b
CM + % 3 G	47.8b	52.4b	59.9c	69.9c	75.8d	82.5c
	***	***	***	***	***	***

Notes: CM: Canola meal, **a**: the rapidly soluble fraction, **b**: the potentially degradable fraction, **c**: the constant rate of disappearance of **b**, **Pe**: the effective degradation WH: water + heat treatment, G: glucose treatment.

p>0.05: means of same letters in column are nonsignificant.

NS: No significance, p>0.05; *p<0.05; ***p<0.001

Table 3. **Rumen degradation characteristics (%) of dry matter, organic matter and crude protein of canola meal**

	Degradability characteristics (%)			
	a	b	c <i>h⁻¹</i>	Pe <i>0.05 h⁻¹</i>
Dry matter				
CM	30.4c	51.8a	0.076a	61.7a
CM + WH	35.2a	47.0b	0.064b	61.6a
CM + % 2 G	35.2a	45.5b	0.061b	60.0b
CM + % 3 G	32.7b	47.5b	0.062b	59.0b
	***	*	*	***
Organic matter				
CM	29.3	51.3	0.084a	61.4a
CM + WH	31.5	50.9	0.064b	60.1a
CM + % 2 G	31.9	50.2	0.051b	56.8b
CM + % 3 G	29.7	47.0	0.047b	52.2c
	NS	NS	***	***
Crude protein				
CM	47.5a	41.1b	0.091a	74.1b
CM + WH	45.7ab	48.7a	0.082ab	75.6a
CM + % 2 G	48.0a	43.2b	0.067b	72.5b
CM + % 3 G	42.5b	42.0b	0.068b	66.4c
	*	*	*	***

Notes: CM: Canola meal, **a**: the rapidly soluble fraction, **b**: the potentially degradable fraction, **c**: the constant rate of disappearance of **b**, **Pe**: the effective degradation WH: water + heat treatment, G: glucose treatment.

p>0.05: means of same letters in column are nonsignificant.

NS: No significance, p>0.05; *p<0.05; ***p<0.001

Table 4. **Disappearance of dry matter, organic matter and crude protein for rumen incubation times of canola seed (%)**

	Incubation times (h)					
	2	4	8	16	24	48
Dry matter						
CS	58.7a	61.7b	66.4b	71.8a	74.4a	76.7a
CS + WH	60.2a	62.7ab	66.1b	70.3b	72.8b	74.8b
CS + % 2 G	59.3a	63.3a	68.1a	71.7a	72.5b	72.8c
CS + % 3 G	56.4b	62.0ab	68.0a	71.6a	72.3b	72.5c
	***	*	**	**	***	***
Organic matter						
CS	58.8a	61.5a	65.7a	70.6a	73.1a	75.3a
CS + WH	58.6a	61.1a	64.2bc	68.7b	71.0b	73.2b
CS + % 2 G	58.2a	60.9a	64.9ab	69.3b	71.1b	72.5b
CS + % 3 G	55.1b	58.6b	63.7c	69.0b	71.2b	72.7b
	***	***	***	***	***	***
Crude protein						
CS	63.3	65.2	68.4	73.0	76.0	80.2a
CS + WH	59.6	62.5	67.2	73.2	76.5	80.2a
CS + % 2 G	59.8	62.7	67.1	72.3	74.9	77.1b
CS + % 3 G	60.2	64.3	69.9	75.1	77.1	78.4ab
	NS	NS	NS	NS	NS	*

Notes: CS: Canola seed, a: the rapidly soluble fraction, b: the potentially degradable fraction, c: the constant rate of disappearance of b, Pe: the effective degradation WH: water + heat treatment, G: glucose treatment.

p>0.05: means of same letters in column are nonsignificant.

NS: No significance, p>0.05; *p<0.05; **p<0.01; ***p<0.001

Table 5. **Rumen degradation characteristics (%) of dry matter, organic matter and crude protein of canola seed**

	Degradability characteristics (%)			
	a	b	c h^{-1}	Pe $0.05 h^{-1}$
Dry matter				
CS	55.0b	22.0b	0.091c	69.2a
CS + WH	57.7a	17.5c	0.081c	68.5ab
CS + % 2 G	53.5b	19.3c	0.177b	68.5ab
CS + % 3 G	47.6c	24.9a	0.220a	67.8b
	***	***	***	*
Organic matter				
CS	55.6a	20.0ab	0.087bc	68.3a
CS + WH	56.2a	17.5c	0.078c	66.8b
CS + % 2 G	54.8a	17.8bc	0.105ab	66.8b
CS + % 3 G	50.8b	22.1a	0.110a	65.9b
	***	***	***	***
Crude protein				
CS	61.1a	20.8	0.053b	71.8
CS + WH	56.2ab	25.2	0.072b	70.9
CS + % 2 G	56.4ab	21.0	0.090ab	69.8
CS + % 3 G	54.8b	23.8	0.130a	71.7
	*	NS	***	NS

Notes: CS: Canola seed, a: the rapidly soluble fraction, b: the potentially degradable fraction, c: the constant rate of disappearance of b, Pe: the effective degradation WH: water + heat treatment, G: glucose treatment.

p>0.05: means of same letters in column are nonsignificant.

NS: No significance, p>0.05; *p<0.05; ***p<0.001

The effect of glucose treatment with 3% level was more pronounced on dry matter and organic matter degradability value of CS. Crude protein degradability value of CS was not affected by glucose treatment except 48 h incubation time (Table 4).

All treatments decreased dry matter *b* and *c* values of CM (Table 3) compared to those of untreated CM while increased *c* value. Organic matter *a* and *b* values of CM did not change with any treatments. Organic matter *c* value of CM was reduced ($p < 0.001$) by glucose treatments compared to that of untreated CM. Effective DM, OM and CP degradabilities of CM were decreased ($p < 0.001$) by 2 and 3% glucose treatments. Glucose treatments at both levels were reduced effective DM, OM degradabilities of CS. Effective CP degradability of CS was not changed by the treatments compared to those of untreated CS.

Discussion. The “Maillard Reaction” occurs between sugar aldehyde groups and free amino groups and it can provide to decrease protein degradability in the rumen (Chalupa 1974). McAllister et al. (1993) reported the xylose treatment of rapeseed commodities, in an aqueous calcium-magnesium lignosulfonate (LSO₃) solution, reduced degradability of DM and CP in the rumen. Tymchuk et al. (1997) mentioned in situ CP degradability of rapeseed decreased by formaldehyde treatment compared to that of heat treated and untreated canola seed. Wulf and Südekum (2005) showed crude protein was effectively protected in expeller rapeseed meal treated with xylose. Sacakli et al. (2009) reported that 3% glucose treatment was more effective on CP degradability of soybean meal (SBM) and soybean seed (SBS) after rumen incubation times than untreated, water + heat treated SBM and SBS. In the present study, crude protein degradability value of CM was reduced by 3% glucose treatment after all rumen incubation times while crude protein degradability value of CS was not affected by 3% glucose treatment.

Stanford et al. (1995) showed treating of CM with 7% lignosulfonate reduced rapidly soluble fraction of DM and CP in the rumen. McAllister et al. (1993) reported the level of 5% or 10% lignosulfonate treatment with heating at 100 °C for 1 h decreased solubility of CP for CM in the rumen. Similarly, xylose treatment was determined to reduce soluble CP of SBM (Harstad and Prestlokken 2000; Tuncer and Sacakli 2003; Sacakli and Tuncer 2006) and SBS rapidly (Wulf and Südekum 2005). Sacakli et al. (2009) reported that glucose treatment reduced the rapidly soluble fraction of CP in SBM and SBS. Rapidly soluble fraction (*a*) of CS reduced by treatment of 3% glucose compared to untreated CS. The major reducing effect of 3% glucose treatment was observed on the soluble fractions (*a* value) and effective crude protein degradability of CP in CM. The observed result of the study for CM may be attributed to formation of protein resistance to microbial fermentation. However, canola seed had lower CP than CM. These content of CS may cause in not reducing of soluble fractions (*a* value) and effective crude protein degradability of CS. Attempts to decrease the

degradability of proteins may be related industrial production procedures and the chemical composition of the samples as protein content in meals or seeds due to the fact that Maillard reaction occurring between sugar aldehyde groups and free amino acids.

Sacakli et al. (2009) reported that 2 or 3% glucose treatment did not affect on *b* value of CP in SBM and SBS. Tuncer and Sacakli (2003) indicated that *b* value of DM in SBM was not affected by 0.5, 1 and 2% xylose treatments. However, they have determined a decrease in *b* value of CP. The results obtained with xylose treatment (Sacakli and Tuncer 2006) and lignosulfonate treatment (Mir et al. 1984) reduced *b* value of CP in SBM. Stanford et al. (1995) found that lignosulfonate did not affect potentially degradable fraction of CP in SBM. In the present study *b* value of DM in CM was reduced by water + heat, 2% and 3% glucose treatments but *b* value of CP was not affected by 2% or 3% glucose treatments. These results for CP *b* value of CM treated with glucose did not conform to the results of other studies (Mir et al. 1984; Tuncer and Sacakli 2003; Sacakli and Tuncer 2006). Both CP *b* values of CM and CS treated with glucose were similar to the result of Stanford et al. (1995).

When glucose treatment was applied to CM the rates of disappearance (*c* value) of CP were reduced, but not to CS. Similarly, Sacakli et al. (2009) reported that CP *c* value of SBS was not affected by 2 or 3% glucose treatment. The results of many studies (Beauchemin et al. 1995; Stanford et al. 1995; Tuncer and Sacakli 2003) indicated that *c* value of CP of SBM was reduced by xylose or LSO₃ treatments.

The effective crude protein degradability of CM treated with 3% glucose was lower than untreated CM, CM treated with water+heat and 2% glucose but not to CS. Sacakli et al. (2009) mentioned effective CP degradability values of SBM and SBS treated with 2 or 3% glucose were lower than those of untreated, treated with water + heat. Effective CP degradability value of canola meal (McAllister et al. 1993) was reduced by 1% xylose treatment. These results of CM are in agreement with the other studies using CM (McAllister et al. 1993; Stanford et al. 1995).

By-pass protein production aims to provide more resistant protein sources directly pass to small intestines where they can be absorbed maximally especially for rapidly growing ruminants and high producing dairy cattle.

Conclusion. The present study indicates that chemical treatment of CS with glucose does not protect against ruminal crude protein degradation. However, glucose treatment is effective way to protect CM protein from rumen degradation. Also 3% glucose level is more effective than 2% glucose level. However, obtaining more reduction in effective CP degradability may obtain with different treatment conditions as higher glucose level, longer heat duration or temperature. It is concluded that glucose may be used in producing by-pass protein from protein rich supplements. Also it should be evaluated whether cost effective way of glucose treatment in further research.

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