

Subproduct of the Wine Industry to Replace Forage for Feeding Confined Ovine

Coproducto da Indústria de Vitivinificação como Substituto ao Volumoso na Alimentação de Ovinos Confinados

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ABSTRACT

The aim of the present study was to evaluate the ingestive behavior of sheep receiving alternative food with grape pomace to replace roughage, as well as its impacts on the rumen environment. It was observed that BU had higher TOC, TRAM and pH of the ruminal liquid, while there was less TRU ($P < 0,05$). As for the other parameters, no significant differences were found. In this study, just as the feeding time was not influenced due to the similarity of the concentrate: roughage ratio in the diet, the protozoan count did not change. This finding reinforces the possibility of using grape marc as a tool for formulating feed, reducing the risk of ruminal disturbances. It was observed that the group control had lower TRAM, indicating that the diet of this group promoted greater microbial activity and, consequently, greater digestibility, corroborating the already observed TOC. This result was the opposite of what was expected, since the grape pomace has probiotic properties, precisely because the fruits have more fungi and bacteria in their microbiota. This also confirms that the co-products of vitiviniculture can act as promoters of the intestinal health of ruminants, justifying further studies in order to improve this use. Therefore, the use of wine by-products is an alternative to reduce production costs, as its use in diets for confined sheep can improve the performance of the animals, increasing the total feed consumption and improving microbial activity.

Keywords: behavior; grape pomace; rumination; sustainability.

RESUMO

O objetivo do presente estudo foi avaliar o comportamento ingestivo de ovinos recebendo ração alternativa com bagaço de uva em substituição ao volumoso, bem como seus impactos no ambiente ruminal. Observou-se que a BU apresentou maiores COT, TRAM e pH do líquido ruminal, enquanto houve menor TRU ($P < 0,05$). Quanto aos demais parâmetros, não foram encontradas diferenças significativas. Neste estudo, assim como o tempo de alimentação não foi influenciado pela similaridade da relação concentrado: volumoso na dieta, a contagem de protozoários não se alterou. Esse achado reforça a possibilidade do uso do bagaço de uva como ferramenta na formulação de rações, reduzindo o risco de distúrbios ruminais. Observou-se que o grupo controle apresentou menor TRAM, indicando que a dieta desse grupo promoveu maior atividade microbiana e, conseqüentemente, maior digestibilidade, corroborando o COT já observado. Esse resultado foi o oposto do esperado, já que o bagaço de uva tem propriedades probióticas, justamente porque os frutos possuem mais fungos e bactérias em sua microbiota. Isso também confirma que os coprodutos da vitivinicultura podem atuar como promotores da saúde intestinal de ruminantes, justificando novos estudos no sentido de aprimorar esse uso. Portanto, a utilização de subprodutos do vinho é uma alternativa para reduzir os custos de produção, pois seu uso em dietas para ovinos confinados pode melhorar o desempenho dos animais, aumentando o consumo total de ração e melhorando a atividade microbiana.

Palavras-chave: comportamento; bagaço de uva; ruminação; sustentabilidade.

INTRODUCTION

Brazil is the largest producer of milk, beef and agro-industrial waste in Latin America; responsible for more than 150 tons of residue (ANUNCIACÃO, 2017). Buildup of this residue negatively impacts the environment becoming a major hurdle for the sector. Furthermore, transporting this residue to landfills or storing it close to the point of origin without a defined plan to manage it long term are important challenges for this sector.

The residue or subproduct is generated by the food and textile industry. Its utilization in the diet has been researched mainly in ruminants. This research attempts to reduce the cost of food and environmental pollution, preserving natural resources and increasing

productivity and environmental efficiency. Therefore, using this residue to feed livestock in regions close to these industries can benefit both the industry and the farmers (AMARAL, 2016).

Among the utilized subproducts, grape pomace has the highest nutritional potential for ruminants due to its high concentration of fiber, protein, oils, condensed tannins and fenolic compounds. These can reduce the methane production in the rumen without negatively impacting productivity (MACIEL, 2012; GRAZZIOTIN 2017). The bromatological composition of grape pomace is 14 to 17% protein, 44 to 63% neutral detergent fiber (NDF), 65% total carbohydrates, 5 to 11% ether extract and 20 to 23% lignin (BARROSO, 2006; MENEZES, 2008). Additionally, this

residue has probiotic features due to its microbial profile (MONTEIRO, 2015). Nevertheless, eating behavior and rumen alterations must be evaluated in order to guarantee that alternate food sources can be used in the diet of ruminants.

Considering this, the present study aimed to evaluate the eating behavior of ovine receiving grape pomace as a replacement for forage and its impact on the ruminal environment.

MATERIALS AND METHODS

Study Location

The experiment was conducted at the Núcleo de Pesquisa, Ensino e Extensão em Pecuária (NUPEEC/UFPEl), located at the Universidade Federal de Pelotas, between the months of February and March of 2016.

Animals and diet

Ten *Texel* and *Corriedale* mix breed sheep were used. The animals weighed an average 45.7 Kg and were 13 months old. Sheep were identified individually for behavioral evaluation and grouped in stalls per treatment. The facility where the experiment was conducted had covered stalls with rice husk bedding. Sheep were randomly attributed to two groups: control (CON; n = 5) and grape pomace (GP; n = 5). The control group received a diet containing 39.7% forage (alfalfa hay), 44.3% commercial concentrate, and 17% rice straw. The grape pomace group received a diet containing 25% grape pomace, 44.3% commercial concentrate, and 30.63% forage (alfalfa hay) (Table 1).

Table 1. Bromatological composition of the diets.

Ingredients, g/Kg MS	Control Group	Grape Pomace Group
Alfafa hey	0,387	0,306
Rice Straw	0,170	-
Concentrate	0,443	0,444
Grape Pomace	-	0,250
Bromatological composition, %¹		
Ash	25,89	10,85
Ether extract	1,67	2,98
Acid detergent fiber	26,06	27,39
Neutral Detergent fiber	43,49	41,34
Lignin	5,00	12,12
Total digestible nutrients	55,62	55,88
Protein	16,43	16,55

The diets were given daily at 8:30 and 16:30 for a total of 35 days divided in 14 days adaptation and 21 days for sampling. The total amount given per day was calculates according to the amount eaten on the previous days, considering daily leftovers of 10%. The animals received water *ad libitum* during the entire experimental period, remaining healthy during this period.

Evaluation of Ruminant Parameters

Ruminal fluid was obtained using an orogastric probe on days -14 (basal), 1, 7, 14 and 21 after feeding in the morning. The following tests were carried out immediately after sampling: pH measurement using a benchtop pH meter, methylene blue reduction time (MBRT), and sedimentation (DIRKSEN, 1993). A 10 mL sample was obtained in a sterile tube for posterior

analysis of protozoa (DEHORITY, 1984).

Evaluation of the eating behavior

To evaluate the eating behavior, we used a visual approach (FISCHER, 1996), in which each individual animal was observed for 24 hours every 15 minutes and their activities classified as feeding time (FT), rumination time (RT) and resting time (RST). These evaluations were carried out on days 0, 3, 5, 7, 9, 11, 13, 15, 17, 19 e 21. Day 0 was the last day of feeding adaptation for the animals. Therefore, there was a total of 264 hours of sampling.

Statistical Analysis

The statistical analysis was carried out using SAS 9.1 (SAS[®] Institute Inc., Cary, NC, EUA, 2009). Normality of the data was verified by the Shapiro-Wilk test. The data for eating behavior and ruminal parameters were submitted to

the PROC MIXED procedure. This was to observe the effects of group and time (days), and their interactions, with statistical significance considered when $P < 0,05$. The Qui-square test was used for rectal temperature and respiratory rate, in order to observe the percent of animals within or out of physiologically accepted parameters throughout the experiment. Comparison between averages of the groups was done using the T test, with statistical significance considered when $P < 0,05$.

RESULTS

In this study, the parameter FT of the eating behavior did not differ between groups ($P = 0,071$). However, RT decreased ($P = 0,05$) and RST increased ($P < 0,05$) for animals supplemented with grape pomace in comparison to the control (Table 2).

Table 2. Evaluation of the parameters related to the eating behavior such as feeding time (FT) rumination time (RT) and resting time (RST) of ovine supplemented with grape pomace (GP) or not (control)

Parameter	Groups			<i>P value</i>		
	GP	Control	T	D	T*D	
FT (hours/day)	2,51	2,78	0,071	<0,025	0,243	
RT (hours/day)	5,70 ^b	6,15 ^a	0,050	<0,001	0,537	
RST (hours/day)	15,51 ^b	14,92 ^a	0,040	<0,001	0,322	

Different letters in the same line indicate a statistical difference ($P < 0,05$) between groups.

T: comparison of averages between groups; D: comparison between days; T*D: group/day interaction

Ruminal Parameters

As observed in table 3, the ruminal fluid pH for animals in the GP group was higher than for animals in the control group ($P = 0,045$). The total protozoa

amount ($P = 0,18$) and time to sedimentation did not differ between groups ($P = 0,72$). Additionally, no group-day interaction was observed for the variables tested.

Table 3. Ruminal parameters for ovine supplemented with grape pomace (GP) or not (control)

Parameters	GP	Control	P-T	P-D	P-T*D
pH	7,01	6,92	0,045	<0,001	0,378
Total protozoa ¹	970920	841220	0,181	<0,001	0,153
Sedimentation (min)	6,80	6,61	0,720	<0,001	0,527

¹number of protozoans/mL.

T: comparison of averages between groups; D: comparison between days; T*D: group/day interaction

For the methylene blue reduction time (MBRT) and interaction was observed between treatment and day with

significant reduction in the control group in comparison to the treatment group on days 1 and 7 (Figure 1).

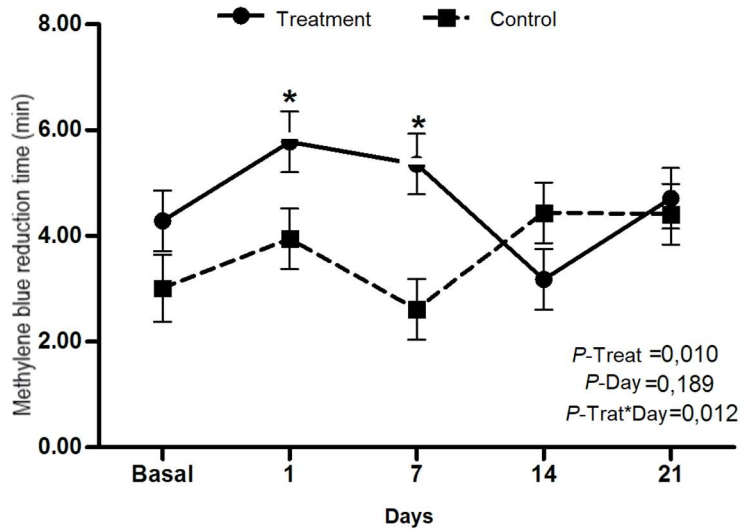


Figure 1. Bacterial reductive activity time

DISCUSSION

In the present study the eating behavior was similar to that observed in another study (AMARAL, 2019) in which no difference was observed in the amount of food consumed by lambs supplemented or not with grape pomace. On the other hand, a different study found that the food intake increased when alfalfa hay was substituted with grape pomace (KLINGER, 2013). The grape pomace has more lignin which can influence the food intake (JUNIOR, 2005). Nevertheless, these data show that grape pomace can substitute hay without negatively impacting the food intake (FERREIRA, 2007).

The increase observed in rumination time for the animals receiving the control diet (6,19 hours/day) in comparison to the treatment group (5,73 hours/day) can be associated with the higher amount of

NDF in the control diet in comparison to control. According to previous studies, increasing the amount of NDF increases the rumination time since more time is required to break the fiber through regurgitation and rumination (FIGUEIREDO, 2013; SANTOS, 2014). Alterations in the time spent on feeding activities have been frequently documented in studies with varied the fiber content in experimental diets (CARVALHO, 2006). Therefore, the NDF content in the control diet can explain the data in this study for resting time which was lower in the control group (approximately -59 min/d) in comparison to the treatment group. The resting time decreased because of the increase in rumination time (FIGUEIREDO, 2013). Furthermore, the data in this study are consistent with previous data suggesting a linear increase in average feeding time and

rumination according to the NDF content in the diet (BEUACHEMIN, 1991; DADO, 1995; BURGUER, 2001; PEREIRA, 2018).

The data for eating behavior in this study suggest that the higher the FT and RT the lower the RST (MINERVINO, 2014). The higher RST value in the treatment group can be related to the more easily digestible diet given to this group. Previous data has shown that animals receiving more protein in the diet have higher RST (FIGUEIREDO, 2015). This was observed in this study since the protein content was higher in the treatment group.

The pH in both groups remained within the physiologically acceptable range of 6,8 to 7,0 (RADOSTITS, 2007; TABELÃO, 2007), although it was lower in control animals (GUERRA-RIVAS, 2016). The food that reduces the pH is usually high starch grain, which are rapidly degraded, and some fruit subproducts due to their naturally low pH (GONZÁLEZ, 2003; OLIVEIRA, 2013). Grapes have a low pH of 3 after the crushing process, and this can cause reductions in the ruminal pH (RIZZON, 2002). Even though the rumination time was lowered, these results demonstrate the effective buffering activity of the animal's saliva, which may have helped to maintain the pH levels constant (GONZÁLEZ, 2003).

The average protozoan concentrations in the ruminal fluid remained within the physiologically accepted range. In general, the protozoa counts vary between 10^4 and 10^6 per mL of fluid (KAMRA, 2005). This is considered satisfactory since an alteration in the microorganisms can lead to a drop in the digestion rate and nutrient availability (HUNERBERG, 2015). This study

differs from other which found a reduction in total protozoa content in animals receiving wine industry residue. The same study reports that discrepancies in the ruminal microbiota are due to the type of diet, the animal and the sampling method (ABARGHUEI, 2010).

Furthermore, the concentrate to forage ratio is inversely proportional to the protozoa content (BURGUER, 2000). In this study, neither FT or protozoa count were altered by the similar amounts of concentrate and forage in the diet. This highlights the potential for using grape pomace in diets with a reduced risk of ruminal disorders.

The sedimentation time was similar between groups and remained within the physiologically accepted values of 4 to 8 minutes (RADOSTITS, 2007). This parameter enables a prompt examination of the ruminal microbiota through the esterification of the components in the ruminal fluid. Reduced values indicate ruminal acidosis (JONES, 2009).

The MBRT analysis indicates the microbial activity in the ruminal fluid (DIRKSEN, 1968). If the microbial activity increases the discoloration of the methylene blue occurs faster. Values between 3 and 6 minutes are considered normal; when values supersede 8 minutes it is considered simple indigestion; and values above 30 minutes are considered acute acidosis (RADOSTITS, 2007). Both groups in this experiment had MBRT values within the physiologically accepted range. However, the control group had lower MBRT values, indicating that the control diet promoted higher microbial activity and digestibility, corroborating the results obtained for RST.

This result was opposite to the expected considering that grape pomace has probiotic properties (MONTEIRO, 2015), since fruit has more fungus and bacteria in its microbiota (BEUCHAT, 1996; MONTEIRO, 2015). The result also confirms that the subproducts of the wine industry can promote intestinal health, justifying further research to optimize their utilization.

CONCLUSIONS

Considering the data, we conclude that grape pomace can potentially substitute conventional forage without negatively impacting the ruminal environment. Therefore, grape pomace is a viable alternative that would reduce the costs in livestock production. Furthermore, this study has an invaluable role in optimizing technology for the utilization of grape pomace. This research benefits the livestock industry while providing a sustainable and economic purpose for grape pomace.

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