Journal of Animal and Veterinary Advances 6 (9): 1079-1082, 2007
ISSN: 1680-5593

Effects of Ensiling and Drying of White Grape Pomace on Chemical Composition, Degradability and Digestibility for Ruminants

[1]Rasoul Pirmohammadi, [2]Ahad Golgasemgarebagh and [3]Ali Mohsenpur Azari
[1]Department of Animal Science, Faculty of Agriculture,
Urmia University, 165 Urmia, Iran,
[2]Department of Animal Science, Islamic Azad University,
Maragheh Branch, Maragheh, Iran
[3]W. Azerbaijan Agricultural and Natural Resources Research Center, Urmia, Iran

Abstract: Grape Pomace (GP) is an agro-industrial by-product that can be utilized as a feed resource in ruminant diets. This study was done to determine the effects of ensiling and drying on degradability and digestibility of GP. Crude protein content in EGP and DGP was 144 and 132 g kg^{-1} DM, respectively. DM and NDF concentration of DGP was 497 g kg^{-1} and 504 g kg^{-1} and for EGP were 225 and 69.3 g kg^{-1}, respectively. Total Tannins (TT) and Total Phenols (TP) concentration of DGP (186 and 236 g kg^{-1}) was higher than EGP (92 and 132 g kg^{-1}). The soluble components of EGP (*a* fraction) were 16.6% while this value for DGP was 24.3%. Rapidly degraded fraction of EGP was significantly ($p<0.05$) lower than DGP. The insoluble but fermentable component (*b* fraction) in DGP (26.3%) was significantly ($p<0.05$) higher than that of EGP (14.2%). Also dry matter Effective Degradability (ED) of EGP was significantly ($p<0.01$) lower than that of DGP. ED value for DGP and EGP were 35.9 and 22.9%, respectively. Rate of degradation(*c* fraction) was not significant among DGP and EGP. DMD and OMD values obtained for DGP and EGP was 345, 285, 247 and 197 (g kg^{-1} DM), respectively.

Key words: Grape pomace, drying, ensiling, degradability, digestibility

INTRODUCTION

Supply of livestock feeds is a chief and basic problem. With ever increasing of population, attentive to natural resources limitation and forage production for livestock's. Therefore, recognition and better utilization of nonconventional feed resources may be essential. One of the agro-industrial co products is Grape Pomace (GP) which could form important components of ruminant diets. In Iran, GP production exceeds 50,000t/year (Alipour and Rouzbehan, 2006). Tannins are known to negatively affect digestibility, especially of crude protein, caused by the binding capacity to proteins and formation of tannin-protein complexes (Baumgartel *et al*., 2007).

Ensiling and drying can be inexpensive, simple and effective ways in reduction of antinutritional elements and improvement of nutritional value (Bensalem *et al*., 2005; Pirmohammadi *et al*., 2006).

Several investigations conducted on grape pomace, however little information is available with effects of ensiling and drying on digestibility and degradability values of GP. The aim of present study was to investigate effects of ensiling and drying white grape pomace on dry matter and crude protein degradability and digestibility in Azeri buffaloes.

MATERIALS AND METHODS

Silage preparing: Fresh GP provided from TATAO juice factory in Urmia city. Some of GP ensiled for 45 days in plastic vessels (Pirmohammadi *et al*., 2006) then ensiled samples analyzed approximately according to AOAC (1990).

Drying of samples: Some of GP samples were oven dried at 70°C for 48 h, then analyzed approximately according to AOAC (1990).

Chemical composition: The chemical composition of DGP and EGP was determined using methods recommended by AOAC (1990) (Table 1). Dry matter was determined by drying the whole sample in an oven at 65°C until a

Corresponding Author: Rasoul Pirmohammadi, Department of Animal Science, Faculty of Agriculture, Urmia University, 65 Urmia, Iran

Table 1: Mean chemical composition of DGP and EGP (g kg^{-1} DM or as stated)

	Feed stuffs	
Item	DGP	EGP
DM	497	225
OM	928	944
N	21	23
CP	132	144
NDF	504	693
TT	186	92
TP	236	132

DGP, Dried Grape Pomace; EGP, Ensiled Grape Pomace; DM, Dry Matter; OM, Organic Matter, CP Crude Protein; NDF, Neutral Detergent Fiber; TT, Total Tannins; TP, Total Phenols

constant weight was achieved (AOAC, 1990). Neutral Detergent Fiber (NDF) was determined using the methods of Van Soest *et al.* (1991). Determinations of N were conducted using the Kjeldahl method in an automated Kjelfoss apparatus (Foss Electric, Copenhagen Denmark). Total Phenolics (TP) were measured using the Folin Ciocalteau method (Makkar, 2000). Total Tannin (TT) was determined after adding insoluble Polyvinylpoly-Pyrrolidone (PVPP) and reacting with Folin Ciocalteau reagent (Makkar, 2000).

***In situ* study:** The nylon bag technique (AFRC, 1992) was used to determine the rate of degradability of DM and protein with 3 rumen-fistulated Azeri buffaloes. All of the dried samples ground and milled through a 2 mm sieve. Buffalos were fed with a maintenance extent diet. Then 5 g of each sample put in the nylon bags (21×10 cm with a pore size of 45 μm) and incubated in the rumen for 0, 3, 6, 12, 24, 48 and 96 h. In each buffalo one bag was used for each time interval. After withdrawing the bags in incubation times from the rumen, they were washed with cold water by a washing machine for 1 h. When all the bags had taken from the rumen, they were dried for 48 h at 70°C in oven.

The value of degradability at time 0 was obtained by washing two bags in a washing machine for 1 h using cold water. For each bag, the residue was analyzed for DM and protein. The degradability at each time interval was calculated by taking the mean value obtained from the three bags. The percentage of degradability (Y) of DM and protein at time (t) was obtained from an exponential curve of the type: $Y = a + b (1 - e^{(-ct)})$.

This was fitted to the exponential data by iterative regression analysis (Ørskov and McDonald, 1979). In the equation, 'e' is the base of natural logarithms, the constant 'a' represents soluble and very rapidly degradable component and 'b' represents the insoluble but potentially degradable component, which degrades at a constant fractional rate © per unit time. The effective degradability of DM and protein in each feedstuff was then estimated (Ørskov and McDonald, 1979) by the following equation:

$$\text{Effective degradability (\%)} = \frac{a + bc}{c + k}$$

In this equation, k refers to the fractional outflow rate of small particles from the rumen. A value of 0.05 fraction/h for k was used.

***In vitro* study:** Dry Matter and Organic Matter Digestibility (DMD and OMD) were determined using (Tilly and Terry, 1963) method. This study contained 6 steps:

- Preparing of samples and materials.
- Anaerobic digestion.
- Acidic pepsin digestion.
- Separating of no digest content.
- Determination of digestion and ash residue.
- Determination of DM, OM and OM in DM digestibility.

Preparing of samples and material requirement

Preparing of samples: Primarily, samples milled through a 2 mm sieve. Then, from each sample was put 2 g and spilled into 100 mL dishes.

Buffer supply: Buffer was contained 9.8 g Sodium bicarbonate, 9.35 g Mono hydrogen sodium phosphate, 0.57 g Potassium chloride, 0.47 g Sodium chloride and 0.12 g Magnesium sulfate, solve in distilled water. Then, before starting of anaerobic digestion, 1 mL Calcium chloride 4% solution, added to buffer (per 1 L of buffer) and solution reached to 1 L. Buffer pH set in 6.9-7 range with entering CO_2 gas for 10-15 min. Forty mililiter of solution used for each sample.

Rumen fluid supply: Rumen fluid prepared by rumen fistulated Azeri buffaloes that were fed with a maintenance diet. Rumen fluid kept in anaerobic conditions in 39°C water.

Anaerobic digestion: Filtered rumen fluid mixed with the buffer 4:1 ratio then CO_2 gas entered for getting anaerobic culture for 4-5 min in 39°C. Fifty mililiter from this mixture spilled on 100 mL flasks that contained GP sample. Then let the flasks to be incubated in 39°C for 48 h (flasks was shake each 8 h interval).

Acidic pepsin digestion: After anaerobic digestion, 6 mL of Hydrochloride 20% solution was added to all of the flasks in 3 steps, respectively (4, 1, 1 mL). Then 2 mL of

Pepsin 5% solution was added to the flasks then they incubated in 39°C for 46 h (flasks was shake each 8 h interval).

Separating of no digest content: Separating of no digest content was done using ash less Wattman strainer paper number 41.

Determination of residual digestion and ash: Residual material weight determined by drying of separated material using strainer paper in 72°C for 48 h. Furthermore, ash weight of residual digestion determined with 550 °C oven for 3.5 h.

Determination DM, OM and OM in DM digestion: Determination of digestibility was used with the equations suggested by Tilley and Terry (1963).

Statistical analysis: Data obtained from degradability was subjected to Neway 1992 software and mean values were statistically compared with T test ($p<0.05$). Mean digestibility values were also subjected to statistical T test ($p<0.05$).

RESULTS AND DISCUSSION

Crude protein content in EGP and DGP was 144 and 132 g kg^{-1} DM, respectively. This outcome is in agreement with (Alipour and Rouzbehan, 2006) who found that anaerobic storage of GP had no effect on crude protein concentration. DM and NDF concentration of DGP was 497 and 504 g kg^{-1} and for EGP were 225 and 69.3 g kg^{-1}, respectively. Chemical analysis of DGP and EGP are shown in Table 1. Higher concentration of NDF in EGP may be due to fermentation process, as ensiling cause increasing of cell walls (ADF, NDF and lignin) consequent from fermentation processing and decreasing of Water Soluble Carbohydrates (WSC) (Van Soest, 1994).

Decrease of WSC and pH in the silage is probably due to fermentation of WSC by lactic acid bacteria that produce organic acids leading to a decrease in pH (pH of DGP = 3.92 and pH of EGP = 3.11) (Mc Donald *et al*., 2002). Total Tannins (TT) and Total Phenols (TP) concentration of DGP (186 and 236 g kg^{-1}) was higher than EGP (92 and 132 g kg^{-1}). Lower concentration of TT and TP of EGP in comparison to DGP may attribute to ensiling of EGP, because the gradual reduction in level of TT and TP during ensilage could be due to the polymerization (Makkar and Singh, 1993) or oxidation of tannins in GP during ensiling (Ben Salem *et al*., 2005).

Characteristics of DM degradation of the feedstuffs are given in Table 2. Ensiling reduced degradation of DM. The soluble components of EGP (*a* fraction) were 16.6% while this value for DGP was 24.3%. Rapidly degraded fraction of EGP was significantly ($p<0.05$) lower than DGP. The insoluble but fermentable component (*b* fraction) in DGP (26.3%) was significantly ($p<0.05$) higher than that of EGP (14.2%). Also dry matter Effective Degradability (ED) of EGP was significantly ($p<0.01$) lower than that of DGP. ED value for DGP and EGP were 35.9 and 22.9%, respectively. Rate of degradation (c fraction) was not significant among DGP and EGP. It is obvious that degradability of a feedstuff may decrease by decreasing its soluble components and increasing its NDF (Van Soest, 1994). Lower a, b and c values of EGP compared to DGP in our study may partly be due to lower soluble components specially WSC in EGP due to soluble component losses during ensilage (McDonald *et al*., 1991).

Table 3 shows lower values of Dry and Organic Matter Digestibility (DMD and OMD) of EGP in comparison to DGP. DMD and OMD values obtained for DGP and EGP was 345, 285, 247 and 197 (g kg^{-1} DM), respectively. As mentioned above, WSC may decrease and cell wall (ADF, NDF and lignin) increase during ensiling and fermentation processing (McDonald *et al*., 1991). Thus, digestibility and degradability values could be decreased during ensiling in our study. Lower DMD, OMD and DOMD values seen for EGP may be reflection of ensiling. Similar results reported by Alipour and Rouzbehan (2006) who worked on ensiling of GP for ruminants. They found that reducing of gas production of GP in anaerobic storage period to cause subsidence of digestibility that, reducing of gas production may be due to the decrease in water soluble carbohydrates which is source of energy for microorganisms in the rumen. Furthermore, our results are in agreement with (Baumgartel *et al*., 2005) who worked on nutritive value of GP for sheep.

Table 2: Mean and S.E.M differences of degradation values (DM) for DGP and EGP (% or as stated)

	Item				
Treatment	a	b	a + b	c	ED
DGP	24.3±0.8^{a}	26.3±1.2^{a}	49.7±1.7^{a}	0.04±0.01^{a}	35.9±0.6^{a}
EGP	16.6±0.8^{b}	14.2±1.4^{b}	30.9±1.0^{b}	0.03±0.03^{a}	22.9±0.3^{b}
P	0.05	0.05	0.05	ns	0.01

DGP, Dried Grape Pomace; EGP, Ensiled Grape Pomace; DM, Dry Matter; CP, Crude Protein; a, rapidly degraded fraction(%); b, slowly degraded fraction(%); c, rate of degradation (fraction/h); are constants in the exponential equation [$p = a + b(1 - e^{-ct})$]; ED (%), effective degradability (out flow rate: 0.05h). Means in the same row with difference superscripts (a–b) letters are significantly difference

Table 3: Mean and S.E.M differences of DMD, OMD and DOMD values for DGP and EGP (g $kg^{-1}M$)

	Feedstuffs		
Item	DGP	EGP	S.E.M
DMD	345[a]	285[b]	3.77
OMD	247[a]	195[b]	3.37
DOMD	230[a]	182[b]	3.25

DGP, Dried Grape Pomace; EGP, Ensiled Grape Pomace; DMD, Dry Matter Digestibility; OMD, Organic Matter Digestibility; DOMD, dry Organic Matter Digestibility. Means in the same row with different superscript (a-b) letters are significantly different ($p<0.05$)

CONCLUSION

It may conclude that although, digestibility and degradability of EGP was reduced by ensiling, but, it caused to decreasing of TT and TP concentrations, that finally, may improve nutritive value of ensiled grape pomace.

ACKNOWLEDGEMENT

The authors thank W. Azerbaijan Agricultural and Natural Resources Research Center for financial support, Mr. Gader, Manafi Azar for his associate to statistical analysis of the data and Dr. A. Safamehr for his scientific help.

REFRENCES

Agricultural and Food Research Council (AFRC), 1992. Nutrient Requirements of Ruminant Animals: Protein. Technical Committee on Responses to Nutrients, Report Nutrition Abstracts and Reviews, Series B., 62: 787-835.

Alipour, D. and Y. Rouzbehan, 2006. Effects of ensiling grape pomace and addition of polyethylene glycol on *in vitro* gas production and microbial biomass yield. Anim. Feed. Sci.Tech. Doi: 10.1016/j.anifeedsci.

Association of Official Analytical Chemists (AOAC), 1990. Official Methods of Analysis, (15th Edn.), Association of Official Analytical Chemists, Arlington, VA, USA, Vol. II.

Baumgartel, T., Kluth, H. Epperelein and K. Rodehutscord, 2007. A note on digestibility and energy value for sheep of different grape pomace. Small Rumin. Res., 67: 302-306.

Ben Salem, H., L. Saghrouni and A. Nefzaoui, 2005. Attempts to deactivate tannins in fodder shrubs with physical and chemical treatments. Anim. Feed Sci. Tech., 122: 109-121.

Makkar, H.P.S., 2000. Quantification of tannins in tree Foliage. A Laboratory Manual For the FAO/IAEA Co-ordinated Research Project on Use of Nuclear and Related techniques to Develop simple Tannin Assays for Predicting and Improving the safety and Efficiency of Feeding Ruminants on Tanninferous Tree Foliage. Joint FAO/IAEA, FAO/IAEA of Nuclear Techniques in Food and Agriculture. Animal Production and Health Sub-Programme, FAO/IAEA Working Document. IAEA, Vienna, Austria.

Makkar, H.P.S. and B. Singh, 1993. Effect of storage and urea addition on detanninification and in Sacco dry matter digestibility of mature oak (*Quercus incana*) leaves. Anim. Feed Sci. Tech., 41: 247-259.

McDonald, P., A.R. Henderson and S.J.E. Heron, 1991. The Biochemistry of Silage, (2nd Edn.), Chalcombe Publications, Marlow, UK.

McDonald, P., R.A. Edwards, J.F.D. Greenhalgh and C.A. Morgan, 2002. Animal Nutrition, (6th Edn.), Longman, London, UK, pp: 451-464.

Ørskov, E.R. and P. McDonald, 1979. The estimation of protein digestibility in the rumen from incubation measurements weighed according to rate of passage. J. Agric. Sci., Cambridge, 92: 499-503.

Pirmohammadi, R., Y. Rouzbehan, K. Rezayazdi and M. Zahedifar, 2006. Chemical composition, digestibility and in situ degradability of dried and ensiled apple pomace and maize silage. J. Anim. Small Rumin. Res., 66: 150-155.

Tilly, Y.M.A. and R.A.A. Terry, 1963. A two stage technique for the digestion forage crops. J. Br. Grass, L. Soc., 18: 104-111.

Van Soest, P.J., J.B. Robertson and B.A. Lewis, 1991. Methods of dietary fiber, neutral detergent fiber and non-starch polysaccharides in relation of animal nutrition. J. Dairy Sci., 74: 3583-3597.

Van Soest, P.J., 1994. Nutritional Ecology of the Ruminant. Cornell University Press, Ithaca, NY, USA.