

## The effects of some agricultural By-products on blood metabolites, chewing behavior and physical characteristics of dairy cow diets

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**Abstract:** This study was carried out to evaluate the effects of feeding ensiled mixed tomato and apple pomace (EMTAP) on blood metabolites and chewing behavior of dairy cow. Six multiparous Holstein dairy cows in mid lactation were used in 3×3 Latin square design and fed alfalfa hay plus concentrate mixture with three levels replacement with EMTAP (0, 15, 30%) during 63 days. Results showed that, differences between treatments were significant. Feeding EMTAP resulted in higher glucose, cholesterol, BHBA, triglyceride, and total protein (P<0.01) concentrations than control diet. Data showed that, total eating time (hours per day) was not significantly (P > 0.05) affected by treatments, but time spent eating, ruminating and total chewing activity per daily intake (kg) of DM and NDF decreased significantly with increasing EMTAP in diet. It was concluded that, EMTAP can efficiently replace up to 30% alfalfa hay. The nutritive value of tomato and apple pomace could be improved when they are used together (50:50) in dairy cows diet.

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**Key words:** blood metabolites; chewing behavior, agricultural By-products.

### 1. Introduction

Ruminants which are able in converting waste items in to useful products such as meat, milk and skin, offers a feasible solution of using by-products and preventing pollution (Oni et al., 2008). Tomato and apple pomace are two alternative by-products that obtained from tomato paste and apple juice industrial production, respectively. These by-products are produced in huge amount annually. The chemical composition of final pomace is linked to the morphology of the original feed stock and the extraction technique used. Although tomato and apple pomace are varying from nutrient density, effective processing can improve their nutritive value. According to NRC, (2001) apple pomace (AP) is very low in protein (6.4% protein on DM basis), it also serves as a useful energy source because of high content of soluble carbohydrate for ruminants. Researches conducted on AP (Rumsey, 1978; Fontenot et al., 1977), showed that AP supplemented with natural proteins was comparable to protein enriched corn silage. In contrast, Elloitt et al. (1981) demonstrated that, tomato pomace (TP) have the potential to be a good source of protein, however its energy source may be limited due to the high fiber content. Previous researches reported different results from feeding TP and AP. The complementary composition of AP (low protein concentration) (Alibes et al., 1984; NRC, 2001; Pirmohammadi et al., 2006) and TP (high protein content) (Fondevila et al., 1994; Del Valle et al., 2006; Weiss et al., 1997) suggest to use those by-products together. Our

previous observations (unpublished data) showed that processed TP with AP (ratio of 50:50) had more palatability and digestibility than processing with urea, wheat straw, NaCl and NaOH for sheep. The aim of the present study is to evaluate the effect of ensiled mixed tomato and apple pomace (EMTAP) on blood metabolites and chewing behavior of dairy cow.

### 2. Material and methods

Fresh experimental samples of tomato pomace (TP) and apple pomace (AP) were collected from several factories in Urmia city (Iran). TP and AP were mixed together (50:50 on DM basis) and ensiled without any additive in a trench silo on a concrete floor. The mixed TP and AP silage (EMTAP) was sealed for 55 days, next fed as TMR diets in three levels replacement of alfalfa hay. Chemical composition of TP, AP and EMTAP was determined using the method suggested by AOAC (2000). Neutral detergent fiber (NDF) and acid detergent fiber (ADF) were determined using method of Van Soest et al. (1991). Six multiparous dairy Holstein cows were used in a 3×3 Latin square experimental design with three 3-wk periods. They were kept in individual concrete tie-stalls and had free access to drinking water at all times. The daily TMR diets were given in two equal feeds at 08:00 and 20:00 h to provide approximately 10% feed refusal each day (as-fed basis). Feed refusal were removed and weighed before feed offered at 08:00 h. Body weight was recorded prior to morning feeding

on 2 consecutive days at the beginning and at finish of each period. The experimental periods lasted 21 d, including 14 d of adaptation and 7<sup>th</sup> d of sampling and data collection. During the last 7<sup>th</sup> d of the experimental period collection and sampling of TMR diets, feed refusal, rumen fluid, blood, feces and urine were performed. Normal herd management practices were followed during the experiment.

On the morning of last day in each sampling period, blood samples were taken from the jugular vein of each cow 3 h after feeding and placed into vacuum tubes. The blood samples gently kept in ice, and then were centrifuged at 1500 g for 15 min to

separate the serum. The serums were transferred into storage pipe and labeled with data and animal identification and stored at 20°C until analysis. Concentration of plasma glucose, cholesterol, triglyceride, BHBA, albumin, urea, ammonia-N, total protein, calcium, phosphorus, were measured by using an auto analyzer Spectrophotometer, mark Unico, model S 2100 SUV, serial number 2165168, Japan. Sodium and potassium were measured by Flame photometer, model PFP7, Serial number 12377, Genewey factory, England.

Table 1. Ingredients and nutrient composition of experimental diets (DM basis)

Ingredients	Diets (EMTAP levels)		
	1	2	3
	0%EMTAP	15%EMTAP	30%EMTAP
Alfalfa hay	45.67	33.35	18.41
EMTAP‡	0	15	30
Soy bean meal	10.25	10.23	9.92
Barley	37.96	37.99	38.2
Fat (Oil plant)	0	0.57	0.99
Wheat bran	5.4	2.09	1.54
Caco3	0.22	0.27	0.44
Premix†	0.5	0.5	0.5
<b>Nutrient compositions</b>		<b>(% based DM)</b>	
DM	98	78.3	63.1
NEL (mcal/kg)	1.54	1.58	1.62
CP	15.4	15.5	15.5
NDF	35.4	35.2	35.1
ADF	21.4	23.1	24.3
Calcium	0.6	0.6	0.5
Phosphorus	0.4	0.4	0.4
Concentrate	54.33	51.65	51.59
Forage	45.67	48.35	48.41

‡EMTAP, ensiled mixed tomato and apple pomace; DM, dry matter; NEL, net energy for lactation; CP, crude protein; NDF, neutral detergent fiber; ADF, acid detergent fiber. †Premix supplied (on a concentrate DM basis): 400,000 IU of vitamin A/kg, 100,000 IU of vitamin D3/kg, 100 mg of vitamin E/kg, 219 mg/kg of Mn, 69 mg/kg of Zn, 116 mg/kg of Fe, 23 mg/kg of Cu, 1.8 mg/kg of I, 0.6 mg/kg of Co, and 0.46 mg/kg of Se.

The feces and urine pH were measured 3 h after morning feeding for two consecutive days on d 3 and 4 of each sampling period. Feces were collected and extracted through cheesecloth into a clean beaker; the urine samples were taken via vulva stimulation. Feces and urine pH was measured using a Schott Titrator Titroline easy pH-meter. Animal behaviors were monitored visually for 24 h period on day 5 of each sampling period. The assumption was that the particular chewing activity persisted for the

entire 5-min period between each visual observation (Beauchemin et al., 2003). Chewing activities were expressed as total hours on 24 hr period per unit of DM and NDF intake by dividing minutes of eating or ruminating by the mean daily nutrient intake.

Collected data were statistically analyzed using the GLM procedure (SAS, 1998, Inst. Inc., Cary, NC). Level of significance was  $\alpha = 0.05$ , and the Tukey test was used to test for all pairwise

comparisons among means. The model used for this analyze was:

$$Y_{ijk} = \mu + T_i + C_j + P_k + \epsilon_{ijk}$$

where Y is dependent variable,  $\mu$  is the overall mean, T is treatment effect (i = 1, 2, 3 EMTAP levels), C is cow effect (j= 1 to 6), P is period effect (k= 1, 2, 3) and  $\epsilon$  is random residual error term.

### 3. Results and discussion

Blood metabolite concentrations are shown in Table 2. Feeding EMTAP resulted in higher glucose, cholesterol, BHBA, triglyceride, and total protein (P<0.01) concentrations. In contrast, urea, albumin, calcium, phosphorous, sodium and potassium were not affected significantly (P > 0.05) by treatments. The results obtained are in good accordance with our expectations. Increasing the presence of soluble carbohydrates and digestible nutrients in AP (Rumsey, 1978) and high amount of protein (21.75) and fat (13.4) in TP resulted in higher concentration of blood metabolites. There was high correlation between rumen and blood ammonia-N, as

well the volume of rumen ammonia-N directly related to rumen degradability of diet protein. As above mentioned, TP has high content of not degradable proteins. In contrast with our results, Belibasakis (1995) reported that, replacing maize silage and soya bean meal with alone TP at 13% DM of dairy cows diet did not affect significantly same blood metabolites. While, in present study we saw significant differences. This inconsistency may be due to use of EMTAP in our study. Mean value of eating and ruminating times as well as particle size distributions for the diets are given in Table 3. Total chewing time (hours per day) varied from 13.58 to 12.09 with ruminating time varying from 7.30 to 6.13, for control and EMTAP diets, respectively. Data showed that, total eating time (hours per day) was not significantly (P > 0.05) affected by treatments, but time spent eating, ruminating and total chewing activity per daily intake (kg) of DM and NDF decreased significantly with increasing EMTAP in diet.

**Table 2.** Blood metabolites concentration of cows fed diets with different content of EMTAP.

Item	Diets (EMTAP Levels)			S.E.M	P value
	1 0% EMTAP	2 15% EMTAP	3 30% EMTAP		
Glucose, mg/dL	79.55 <sup>b</sup>	90.89 <sup>b</sup>	118.8 <sup>a</sup>	4.76	<0.01
Ammonia-N, mg/dL	6.87	6.88	6.92	0.02	0.12
Urea, mg/dL	30	33.67	34	1.72	0.33
Cholesterol, mg/dL	86.95 <sup>b</sup>	124.8 <sup>b</sup>	139.8 <sup>a</sup>	9.30	0.01
Triglycerides, mg/dL	9.69 <sup>c</sup>	11.43 <sup>b</sup>	13.2 <sup>a</sup>	0.47	<0.01
BHBA, mg/dL	6.15 <sup>b</sup>	8.25 <sup>b</sup>	8.68 <sup>a</sup>	0.31	<0.01
Calcium, mg/dL	8.44	8.68	8.96	0.53	0.48
Phosphorus, mg/dL	5.35	5.48	5.71	0.31	0.59
Sodium, mEq/L	136	136.33	136.5	1.57	0.98
Potassium, mEq/L	3.7	3.85	4.10	0.21	0.4
Total protein, g/dL	6.7 <sup>b</sup>	8.5 <sup>b</sup>	9.28 <sup>a</sup>	0.37	<0.01
Albumin, g/dL	4.05	4.33	4.64	0.24	0.26

Diets; 1= control or 0% EMTAP; 2= 15% EMTAP; 3= 30% EMTAP; S.E.M.= standard error of mean; a,b,cMeans in the rows with different superscripts are significantly different (P<0.05).

Dietary fiber plays a fundamental role in ruminant DM intake and stimulates chewing activity and rumen fermentation. The fiber of by-products has different physical and chemical properties from forage NDF (Zhu et al., 1997), in particular their particles have smaller dimensions and higher density (Firkins et al., 1991). Chewing activity is usually a good indication of rumen health because chewing stimulates saliva secretion. The influence of the diet treatments resulted in linearly decrease in proportion of particles on the sieves (PSPS) with increasing

amount of EMTAP in the diets (Table 3). Lower ruminating and total chewing time for the diets containing EMTAP compared to the control, reflected their low peNDF and particle size distribution. According to our results, Beauchemin et al. (2003) reported that dietary peNDF were moderately associated with ruminating time but not with eating time and increasing ruminating time rather than increasing eating time may be a more efficient means of improving ruminal pH status. The pef value for the EMTAP containing diets (59 to 65,

15 and 30% EMTAP, respectively) was significantly ( $P < 0.01$ ) smaller than that of the control diet (pef: 73), because a large amount of these diets included concentrate and EMTAP, which almost entirely passed through the 19 and 8-mm sieve. Generally, total chewing time decreases as forage NDF (Beauchemin, 1991) or particle size (Grant et al.,

1990) in the diets decreases. According to our results, Chumpawadee and Pimpa (2009) reported that, the use of non forage fiber sources (such as TP) caused a decrease of chewing time due to the smaller particle size and lower peNDF.

**Table 3.** Particle size distribution and physical effectiveness factors of diets differing in ratio of EMTAP.

Item	Diets (EMTAP Levels)			S.E.M	P value
	1 0% EMTAP	2 15% EMTAP	3 30% EMTAP		
<b>Eating activity</b>					
h/d	6.28	6	5.96	0.11	0.16
Min/kg DM	17.73 <sup>a</sup>	15.22 <sup>b</sup>	14.63 <sup>b</sup>	0.57	0.01
Min/kg NDF	50.11 <sup>a</sup>	43.22 <sup>b</sup>	41.68 <sup>b</sup>	0.80	$P < 0.01$
<b>Ruminating time</b>					
h/d	7.30 <sup>a</sup>	6.47 <sup>b</sup>	6.13 <sup>b</sup>	0.23	0.02
Min/kg DM	20.61 <sup>a</sup>	16.41 <sup>b</sup>	15.05 <sup>c</sup>	0.28	$P < 0.01$
Min/kg NDF	58.24 <sup>a</sup>	46.60 <sup>b</sup>	42.87 <sup>c</sup>	0.73	$P < 0.01$
<b>Total chewing time</b>					
h/d	13.58 <sup>a</sup>	12.47 <sup>b</sup>	12.09 <sup>b</sup>	0.19	$P < 0.01$
Min/kg DM	38.34 <sup>a</sup>	31.62 <sup>b</sup>	29.68 <sup>c</sup>	0.22	$P < 0.01$
Min/kg NDF	108.35 <sup>a</sup>	89.82 <sup>b</sup>	84.55 <sup>c</sup>	0.14	$P < 0.01$
<b>TMR offered, % DM retained on sieves.</b>					
19 mm	35 <sup>a</sup>	29 <sup>b</sup>	26 <sup>c</sup>	0.16	$P < 0.01$
8 mm	38 <sup>a</sup>	36 <sup>b</sup>	33 <sup>c</sup>	0.13	$P < 0.01$
Pan	27 <sup>a</sup>	35 <sup>b</sup>	41 <sup>c</sup>	0.07	$P < 0.01$
pef †	73 <sup>a</sup>	65 <sup>b</sup>	59 <sup>c</sup>	0.05	$P < 0.01$
PeNDF, % of DM‡	25.84 <sup>a</sup>	22.88 <sup>b</sup>	20.71 <sup>c</sup>	0.09	$P < 0.01$

† Physical effectiveness factor determined as the proportion of dry matter retained by both sieves of the Penn State Particle Separator

‡ peNDF measured as the NDF content of the TMR multiplied by the pef.

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