

# THE EFFECTS OF WET BREWER'S GRAIN WHOLE PLANT MAIZE MIXTURE SILAGES ON FERMENTATION CHARACTERISTICS AND NUTRIENT DIGESTIBILITY IN LAMBS

Fisun Koc, C. Polat, M. Levent Ozduven

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## SUMMARY

*This study was carried out to examine some quality characteristics and nutrient digestibility of wet brewers grain with whole plant maize mixture silages. Treatments were wet brewers grain, 25% + 75% whole plant maize (mixture 1) and %50 wet brewers grain + %50 whole plant maize (mixture 2). Relating to silage fermentation analysis of pH, ammonia nitrogen, water soluble carbohydrate, organic acids (lactic, acetic and butyric acid) and microbiological analyses were carried out. Digestion rate of crude nutritive matters of silages was determined by classical digestive experiments. Dry matter, crude protein, NH<sub>3</sub>-N, lactic acid content and pH value of the silages were found respectively as 23.3, 26.4, 25.4, 24.7%; 22.3, 7.4, 10.6, 14.3%; 1.9, 0.5, 0.9, 1.0 g/kg DM; %1.0, 2.5, 2.1, 1.8; 4.1, 3.8, 3.9, 3.8 for the group of wet brewers grain, whole plant maize, mixture 1 and mixture 2, (P<0.01). Dry matter, crude protein digestibility were determined as 65.0, 70.5, 70.0, 67.10%; 71.5, 55.8, 58.7, 62.3%, respectively. The results indicated that WBG is a suitable by-product for ensiling and, when ensiled with WPM as a mixture, it improved fermentation quality and stability against aerobic deterioration.*

**Key-words:** wet brewer's grains, whole plant maize, fermentation, aerobic stability, digestibility

## INTRODUCTION

Wet brewer's grains (WBG) are by-products of the brewing industry. These products are derived mainly from barley fermented to produce beer. They contain 230 to 290 g/kg CP of DM basis and are rich in digestible fiber (Pereira et al., 1998; Dhiman et al., 2003). Due to their fibrous nature and low energy content, WBG are suitable for ruminants, particularly in dairy cows, to balance intake of large amounts of rich starch diets (Dhiman et al., 2003).

Many by-products from the food and beverage industries have been used as animal feeds, and ensiling is sometimes used to preserve moist by-products for subsequent feeding. However, acceptable fermentation is often difficult to achieve when by-products are

ensiled alone due to high-moisture contents and lack of sugar substrates. Moreover, acetic and butyric acid production may increase especially after prolonged storage (Imai, 2001) and spoilage might occur when by-products are exposed to air after silo opening (Schneider et al., 1999; Nishino et al., 2003). Maize (*Zea mays*) is most popular cereal crop conserved as silage in many parts of world<sup>8</sup> due to relatively low buffering capacity and high water soluble carbohydrate for fermentation to lactic acid is responsible for the reduction of pH level (Meeske et al., 2000) although it has a low protein content. Ensiling of WBG and whole plant maize (WPM) forage with a crude protein and water soluble

*DSc Fisun Koc (fkoc@nku.edu.tr), DSc Cemal Polat, DSc Mehmet Levent Ozduven - Namik Kemal University, Agricultural Faculty, Department of Animal Science, Tekirdag, Turkey*

carbohydrate content can be used to ensure reasonable ensiling of WBG. The other objective of ensiling WBG with WPM is to complement positive characteristics of two materials and thus produce silage which being more complete feed.

However there is a little information available on the nutritive value of WBG and WPM silage mixtures when ensiled together in Turkey. The present study was, therefore, carried out to determine the chemical composition and digestibility. Technique *in vivo* was widely used to evaluate the nutritive value forage.

The objectives of this study were to ensile the different mixture of WBG and WPM to minimize ensiling risk in addition to supplement deficient nutrient by mixing them and to evaluate silage quality and digestibility of silage.

## MATERIAL AND METHODS

### Silage samples

Silages were prepared from different rate of WBG and WPM (Table 1).

**Table 1. Silages combination (fresh wet basis)**

Tablica 1. Mješavina silaža (svježa vlažna osnova)

Silages	Raw material	
	WBG (%)	WPM (%)
WBG	100	0
WPM	0	100
M1	25	75
M2	50	50

WBG: Wet brewer's grain; WPM: Whole plant maize, M1: Mixture 1; M2: Mixture 2

This experiment was conducted using WBG obtained from a private beer processing company at Luleburgaz of Turkey. WPM harvested during formation of milk in the vegetation period was the main material for this study.

After sufficient mixing, silage materials were ensiled in twelve plastic containers (120 liter, 3 replicates). The silages stored for 45 days in the experiment were used. At the end of the digestibility the silages were subjected to an aerobic aerobic stability test lasting for 7 days in which temperature changes were determined thermocouples (Model "875c" thermometer (Tegam, Geneva, OH, USA). Aerobic deterioration was considered to have started when the difference between the silage and surrounding air reached 2°C.

### Animal studies

Three mature Turkgeldi rams of 57 kg ( $\pm 2.5$ ) live weight were used to measure the digestibility organic

matter in the dry matter. The animals were allocated individually in metabolic cages with free-water accesses. The animals housed in were fed on a daily base at 08:00 in the morning and at 16:30 in the evening as two *ad libitum* meals. Ten days of adaptation were followed by 7 days of faeces collection in each period. Lambs were equipped with the bags for the faeces collection. For digestibility trial, each animal's faeces were weighed daily and 10% aliquot retained, composited and frozen. Composited samples were subsequently dried in a forced air oven at 60°C for 48 h. The fresh faeces were then completely mixed and a sample taken for chemical analysis.

### Analytical procedures

Chemical composition of forage, silages, and faces were determined following the procedures (AOAC, 1990). Neutral detergent fiber (NDF) and acid detergent fiber (ADF) were determined according to the method of Goering and Van Soest (1983). The pH value was measured using a portable digital pH meter (WTW multilab 540 Ionalyzer, pH/mV meter, Germany) and ammonia nitrogen (NH<sub>3</sub>-N) was analyzed as reported by Anonymous (1986).

Water-soluble carbohydrates (WSC) were determined by the anthrone method (Koehler, 1952). Lactic (LA), acetic acid (AA) were determined by the spectrophotometric method (Koc and Coskuntuna, 2003).

Microbiological evaluation included enumeration of lactic acid bacteria on pour-plate Rogosa agar (Oxoid CM627, Oxoid, Basingstoke, UK). Yeast and moulds were determined by pour plating in malt extract agar (Oxoid CM59) that had been acidified, after autoclaving, by the addition of 85% lactic acid at a concentration of 0.5% vol/vol. Plates were incubated aerobically at 32°C for 48 to 72 h (Seale et al., 1990).

Flight Point of the silages was described by Kilic (1986),

$$\text{Flight Points} = 220 + (2 \times \text{DM \%}) - (40 \times \text{pH})$$

Where Flight Points valued between 85 and 100 are of very good quality; 60 and 80, good quality; 55 and 60, moderate quality, 25 and 40, satisfying quality; <20 worthless.

Metabolizable energy (ME, MJ/kg) values were calculated using the following equations (Alderman, 1984):

$$\text{ME (MJ/kg DM)} = 0.0152 \times \text{DCP} + 0.0342 \times \text{DEE} + 0.0128 \times \text{DCF} + 0.0159 \times \text{NFE}$$

ME: Metabolic energy; DCP: Digestibility crude protein, g/kg DM; DEE: Digestibility ether extract, g/kg DM; DCF: Digestibility crude cellulose, g/kg DM; DNEF: Digestibility nitrogen-free extracts, g/kg DM.

### Statistical analysis

The statistical analysis of the results included one-way analysis of variance and Duncan's multiple range test, which were applied to the results using Statistical Analysis System (1988) and significance was declared at  $P < 0.05$ .

### RESULTS AND DISCUSSION

The chemical and microbiological composition of the fresh and ensiled WBG, WPM and mixture silages (M1, M2) are given in Tables 2 and 3. In vivo digest-

ibility of the WBG, WPM and mixture silages (M1, M2) are given in Table 4.

The DM content of the silage samples averaged 23.5%-26.8% at the beginning and 23.3%-26.4% at the end of the 45-d fermentation period. The reduction in the silage samples DM between day 0 and day 45, might be due to the fermentation process. The present result is not in agreement with previous findings by Ozduven and Ogun,<sup>19</sup> Wang and Nishino<sup>20</sup>. This variation at DM content can be explained by differences among methods and pressure applied during processing stage of brewer's grain production. Increasing WPM content WBG silages increased DM content of silages ( $P < 0.01$ ).

**Table 2. Chemical and microbiological analysis of the WBG, WPM and mixture silages (M1, M2) (FM)**

Tablica 2. Kemijska i mikrobiološka analiza WBG, WPM i mješavine silaža (M1, M2) (FM)

Item	WBG	WPM	M1	M2
Bc meq NaOH/kg DM	68.6	115.9	97.3	86.5
pH	4.35	5.74	5.33	4.80
DM, g/kg	23.5	26.8	25.8	24.8
WSC, g/kg DM	24.4	56.4	36.4	29.2
CP, g/kg DM	22.0	7.5	10.6	14.3
Cellulose, g/kg DM	15.8	20.6	22.4	20.2
LAB, log <sub>10</sub> cfu/g FM	1.2	4.5	3.3	3.8
Yeasts, log <sub>10</sub> cfu/g FM	4.5	3.1	2.9	3.0
Moulds, log <sub>10</sub> cfu/g FM	NF	NF	NF	NF

WBG: Wet brewer's grain; WPM: Whole plant maize; M1: (25% WBG–75% WPM); M2 (50% WBG–50% WPM), Bc: Buffering capacity; DM: Dry Matter; WSC: Water Soluble Carbohydrate; CP: Crude protein; LAB: Lactic acid bacteria; cfu: Colony forming unit; FM: Fresh Matter; NF: Not Found

The rate at which the pH drops is a function of the level of WSC and the epiphytic bacteria presence on the crop prior to ensiling (Meeske et al., 2000). If insufficient lactic acid bacteria are present on the crop and the readily available sugar concentration is low at ensiling this result in a slow drop in pH. In the present study the pH of WPM, M1 and M2 silage declined faster than the WBG. At the end of the fermentation, significant difference was shown between the pH values of WBG, WPM and mixture silages ( $P < 0.01$ ). A rapid drop in pH is desirable from preservation point of view, since this allows the fermentation to complete faster and preserves more nutrients. In the present study this was achieved through the provision of an energy source (maize) to the lactic acid bacteria which subsequently probably dominated the fermentation process (McDonald, 1991). The lactic acid bacteria require WSC for metabolism. The present result confirms previous findings on the benefit of maize addition when ensiling WBG because it stimulates maximum growth of lactic acid bacteria and consequently higher lactic acid concentrations in the silage (Cerci et al., 1992).

The  $\text{NH}_3\text{-N}$  concentration in silages shows the degree of protein degradation. The combined effects of plant and microbial enzymes result in extensive changes

to the nitrogenous fractions during ensiling. Relatively high CP and pH of WBG may have led to extensive proteolysis, and hence increased the  $\text{NH}_3\text{-N}$  concentration (McDonald, 1991). In this study compared with WBG, ammonia-N and CP contents were higher than ( $P < 0.01$ ) in WPM, M1 and M2 silages.

In the present study LA content was significantly ( $P < 0.01$ ) higher in WPM than WBG, M1 and M2 silages. LA level decrease as WPM level increase in the mixtures. No significant differences were observed among the WBG and WPM, mixture silages with regard to acetic acid and no butyric acid.

The LAB numbers were significantly differed among groups ( $P < 0.01$ ). The highest was obtained in WPM silage. No differences were detected among treatments for yeast numbers.

All of the silages were excellent in terms of physical quality criteria such as colour, smell and structure. The highest was obtained in WPM silage and there were significant differences among groups. WBG, WPM and mixtures were excellent quality based on Fleig point (Table 2,  $P < 0.01$ ). It has been reported that silages with low DM content might have low Fleig point and excellent silage can be obtained by increas-

**Table 3. Chemical and microbiological analysis of WBG, WPM and mixture silages (M1, M2) at 45 days (n=3)**

Tablica 3. Kemijska i mikrobiološka analiza WBG, WPM i mješavine silaža (M1, M2) (FM) tijekom 45 dana (n=3)

Item	WBG	WPM	M1	M2
DM, g/kg	233.3±0.3c	264.0±0.2a	254.0±0.3b	247.4±0.3c
OM, g/kg DM	963±0.1a	925±0.1c	951±0.1b	965±0.1a
CP, g/kg DM	223.0±0.1a	73.6±0.1d	106.2±0.2c	143.0±0.1b
Ash, g/kg DM	36.2±0.1c	56.0±0.1a	49.0±0.1b	36.0±0.1c
NDF, g/kg DM	553±0.3c	603±0.3a	559±0.2b	555±0.2b
ADF, g/kg DM	271±0.2c	308±0.2a	290±0.2b	287±0.2b
pH	4.12±0.1a	3.80±0.1c	3.90±0.1b	3.80±0.1c
WSC, g/kg DM	10.50±0.4b	15.03±0.2a	14.71±0.5a	11.50±0.5b
LA, g/kg DM	44.57±2.1d	94.05±2.7a	80.86±3.2 b	70.73±2.2c
AA, g/kg DM	30.47±3.4	25.02±0.8	26.03±1.2	31.53±2.2
NH <sub>3</sub> -N, g/kg DM	1.90±0.1a	0.52±0.1d	0.90±0.1c	1.01±0.1b
NH <sub>3</sub> -N /total N	53.2±0.1a	42.2±0.4b	53.0±0.3 <sup>a</sup>	28.0±0.2c
LAB, log <sub>10</sub> cfu/g FM	3.90±0.1c	6.60±0.3a	5.71±0.1b	4.40±0.2c
Yeasts, log <sub>10</sub> cfu/g FM	4.20±0.2	3.92±0.1	3.84±0.1	4.30±0.2
Moulds, log <sub>10</sub> cfu/g FM	NF	NF	NF	NF
Color	2	2	2	2
Total point	18	20	18	20
Quality classify	Excellent	Excellent	Excellent	Excellent
Fleigh points	87.2±0.1 <sup>d</sup>	106.9±0.1 <sup>a</sup>	99.7±0.1 <sup>c</sup>	102.5±0.1 <sup>b</sup>
Quality classify	Excellent	Excellent	Excellent	Excellent

WBG: Wet brewer's grain, WPM: Whole plant, M1: (25% WBG–75% WPM); M2: (50% WBG–50% WPM), DM: Dry Matter; OM: Organic matter; CP: Crude protein; NDF: Neutral detergent fiber; ADF: Acid detergent fiber; WSC: Water Soluble Carbohydrate; LA: Lactic acid; AA: acetic acid; NH<sub>3</sub>-N: ammonia-N; LAB: Lactic acid bacteria; cfu: Colony forming unit; NF: Not Found; <sup>a-d</sup> Means with different superscript within same row significantly differ (P<0.01)

ing carbohydrate content of silage material (Iptas and Avcioglu, 1992). In addition, Kilic (1986) reported a positive relationship between Fleigh point and silage quality.

Based on temperature changes, WBG silage was considered to have deteriorated after exposure to air (Figure 1). The silage temperature peaked after 5 days at 6°C above the ambient and cooled quickly thereafter. The WBG silage appeared to be resistant to aerobic deterioration. Results of the aerobic stability experiment showed that, when WBG was ensiled as a M1, the silage had considerable resistance to aerobic deterioration. According to evidence that silages of higher lactic acid contents as more remaining sugars are less stable exposure to air, (Nishino et al., 2003), it was anticipated that M1 silage would deteriorate faster than WBG silage. The opposite result observed in this study is difficult to explain but low number of yeast in M1 silage may be associated with the enhanced stability (Kung

et al., 1998). The difference in silage DM was probably uninvolved, because no significant relations were observed between DM content and aerobic stability of silage (O'Kiely and Muck, 1992). Reasons explaining the inhibited deterioration may, therefore, be concentrations of undissociated acids in WBG and M1 silages. It has been reported that there is positive correlation between fermentation quality and organic acid levels taken place in silages and especially, acetic, propionic and butyric acids inhibit aerobic yeast and mould growth in silages (Moon, 1983).

The in vivo digestibility of CP, and EE were higher WBG silages compared to WPC, M1 and M2 silages (P<0.01). However, in vivo DM, OM, CP, NDF and ADF were lower than WPM, M1 and M2 silages (P<0.01). In our study, DM and CP values determined in the WBG prepared with different mixtures are in accordance with those of the literatures. Thus the DM results are similar to those reported by Ozduven and Ogun (2006) while the

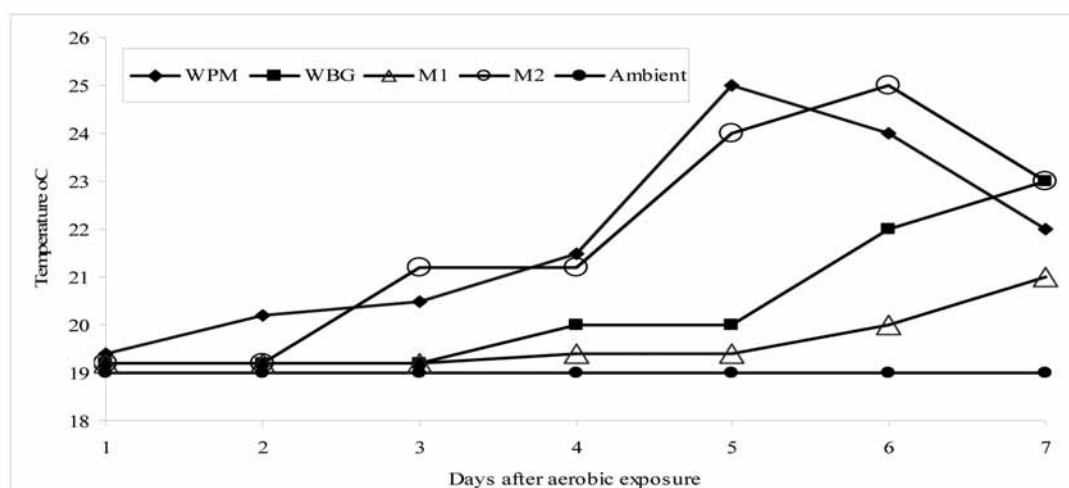


Figure 1. Changes in temperatures after aerobic stability exposure of WBG, WPM and mixture silages

Slika 1. Promjene temperature nakon dodavanja vlažnog pivskog tropa i cijele biljke kukuruza

Table 4. In vivo digestibility of nutrients (%) and metabolize energy (MJ/kg DM) of silages (n=3)

Tablica 4. In vivo probavljivost hranjivih tvari (%) i metabolička energija (MJ/kg ST) silaža (n=3)

Digestibility (%)	Silages			
	WBG	WPM	M1	M2
DM	65.1±0.2c	70.5±0.1a	70.0±1.6a	67.1±1.2b
OM	68.1±0.4b	73.7±0.1a	74.2±1.5a	73.3±1.0a
CP	71.5±0.5a	55.8±0.8b	58.7±3.1b	62.3±4.5ab
CF	54.0±1.1b	67.4±0.5a	70.9±3.0a	69.1±2.3a
EE	89.5±0.3a	63.1±0.7d	87.0±0.4b	84.9±0.3c
NDF	55.6±0.3c	60.6±0.3a	56.0±0.2b	55.8±0.2b
ADF	27.2±0.2c	30.9±0.2a	29.1±0.2b	28.7±0.2b
ME	11.2±0.1a	10.6±0.1b	11.4±0.1a	11.5±0.1a

WBG: Wet brewer's grain; WPM: Whole plant maize; M1: (25% WBG–75% WPM); M2 (50% WBG–50% WPM), DM: Dry matter; OM: Organic matter; CP: Crude protein; CF: Crude cellulose; EE: Ether extract; NDF: Neutral detergent fiber; ADF: Acid detergent fiber; ME: Metabolic energy; a-d. Means with different superscript within same row significantly differ ( $P < 0.01$ )

CP results found in our study are either similar or lower than the values indicated in these reports (Nishino et al., 2003).

Starch and cell wall contents of feedstuffs have a great impact on digestibility (Meeske et al., 2000). Increases in starch content and decrease in cell wall content of feedstuffs increase feed value (Hart, 1990). Level of NDF digestibility is reported to be an important criteria for feed quality (Bal et al., 1997). Valdez et al. (1988a, 1988b) stated that reduction of DM and OM digestibility was caused by an increase in ADF and NDF contents or decrease in ADF and NDF digestibility of silage.

## CONCLUSION

The benefit of mixing WBG with WPM is an increase of the DM content of the silage, decrease

of seepage and improvement of silage quality (less protein breakdown as shown lower  $\text{NH}_3\text{-N}$  as %TN, higher CP and lower AA content). The brewer's grain was well preserved when ensiled without adding of whole crop maize.

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## DJELOVANJE VLAŽNOGA PIVSKOGA TROPA I CIJELE BILJKE KUKURUZA U MJEŠAVINAMA SILAŽA NA FERMENTACIJSKE KARAKTERISTIKE I PROBAVLJIVOST HRANJIVE TVARI KOD JANJADI

### SAŽETAK

*Ovo je istraživanje imalo za cilj ispitati neke kvalitativne karakteristike i probavljivost hranjive tvari pivskoga tropa i cijele biljke kukuruza u mješavinama silaža. Mješavina 1 sadržavala je 25% vlažnoga pivskoga tropa + 75% cijele biljke kukuruza, a mješavina 2 sadržavala je 50% vlažnoga pivskoga tropa + 50% cijele biljke kukuruza. Tijekom istraživanja obavljale su se analize na pH tijekom fermentacije silaže, amonijski dušik, vodotopiv ugljikohidrat, organske kiseline (mliječnu, octenu i maslačnu), kao i mikrobiološke analize. Probavljivost sirovih hranjivih tvari u silažama utvrdila se klasičnim eksperimentima za probavljivost. Sadržaj suhe tvari, sirovih bjelančevina, NH<sub>3</sub>-N, mliječne kiseline, kao i pH vrijednosti silaža iznose redosljedom: 23,3; 26,4; 25,4; 24,7%; 22,3; 7,4; 10,6; 14,3%; 1,9; 0,5; 0,9; 1,0 g/kg ST; %1,0; 2,5; 2,1; 1,8; 4,1; 3,8; 3,9; 3,8 za grupu s vlažnim pivskim tropom, cijelom biljkom kukuruza, mješavina 1 i mješavina 2 (P<0.01). Suha tvar i probavljivost sirovih bjelančevina iznosili su: 65,0; 70,5; 70,0; 67,10%; 71,5; 55,8; 58,7; 62,3%, redosljedom. Dobiveni rezultati pokazuju da je vlažni pivski trop nusproizvod koji povoljno djeluje na process siliranja, a, pomiješan s cijelom biljkom kukuruza, poboljšava fermentacijsku kvalitetu i stabilnost, smanjujući aerobno kvarenje.*

**Ključne riječi:** *vlažni pivski trop, cijela biljka kukuruza, fermentacija, aerobna stabilnost, probavljivost*

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