

## Improvement of the Quality of Ojeoksan(Herbal Medicine) Meal Silage by Molasses Supplementation

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### 당밀의 첨가가 오적산박 사일리지의 품질 및 기호성에 미치는 영향

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#### 요 약

당밀의 첨가가 오적산박 사일리지의 품질 및 한국재래산양에 대한 기호성을 조사하기 위하여 두 가지 실험을 수행하였다. 실험 I에서는 오적산박에 당밀을 0, 0.5 및 1.0% 첨가하여 사일리지를 제조하였고, 실험 II에서는 오적산박 사일리지의 기호성을 조사하기 위하여 9두의 우리나라 재래산양을 공시하여 당밀 첨가수준별 및 옥수수 사일리지와의 섭취량을 비교하였다.

실험 I에서는 당밀의 첨가에 의하여 젖산함량이 유의하게 많아지고, pH와 낙산은 낮게 나타났다. 당밀의 첨가로 사일리지액의 유산균배지 배양시험에 있어 총균수는 증가하였고, 곰팡이 균주는 감소하였다. 그리고 당밀의 첨가에 따라 체외 건물 소실율이 증가하였다. 실험 II에서는 당밀의 첨가가 사일리지 건물섭취량에는 유의한 영향을 미치지 못 하였으며, 오적산박 사일리지는 옥수수 사일리지에 비하여 기호성이 좋았다. 결론적으로 오적산박 사일리지에 대한 당밀의 첨가수준은 1% 정도면 품질을 향상시킬 수 있으며, 체외 소화율도 개선할 수 있다고 생각된다.

**핵심어** : 한약제박 사일리지, 사일리지 품질, 기호성

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## I. Introduction

Nowadays herbal medicine is increasingly popular the world over. About £115 million were spent in UK for complementary medicines in 2000, 57% of these being derived from herbal medicines. In 2001, about US\$17.8 billion were spent in the U.S. health food market, 24% of these on herbs. In 1995, more than one thousand Chinese pharmaceutical factories produced about 400,000 M/T of herbal medicine, roughly equivalent to US\$ 2 billion (Dobes et al., 2005).

Herbal medicine meal is a by-product residue obtained from the extracting process of multi-mixed herbal medicine with boiling water. In Korea, the annual amounts of herbal medicine meal production is estimated to go beyond 50,000 M/T (Korea Ministry of Agriculture and Forestry, 2005). However, only a small portion of them is utilized as feed or fertilizer; most of them discarded causing environmental pollution.

A couple of studies have been reported on the feeding values of herbal medicine meal for ruminants. Choi (1993) investigated the ruminal degradation rate of nutrients in herbal medicine meal. Ko (1995) studied the digestion rate of herbal medicine meal and tofu cake in sheep. The fresh-wet herbal medicine meal is easily deteriorated in summer, presenting a preservation problem at farm level.

We investigated the feed value of Ojeoksan meal silage with or without molasses

supplementation. Ojeoksan meal was chosen in this experiment, because Ojeoksan is one of the popular multi-herbal medicine in Korea to treat bloated feeling, vomiting, diarrhea, and gastritis etc. Molasses were employed to provide some water-soluble carbohydrates, which is essential for satisfactory lactic fermentation.

## II. Materials and Methods

### **Experiment I: Graded levels of molasses supplementation to herbal medicine meal on silage quality**

#### **Silage material**

Ojeoksan was extracted with boiling water, and filtered to give Ojeoksan meal (Hankooksinyak Inc., Chungnam, Korea). The pH and chemical composition of Ojeoksan meal is shown in Table 1.

#### **Experimental design and preparation of Ojeoksan meal silage**

There were three treatments (0, 0.5, and 1.0% molasses supplementation levels), and three replicates (glass bottles) per treatment. Each 2-kg sample of fresh and wet Ojeoksan meal were mixed with relevant amounts of molasses and put into a 2.5 L glass bottle. They were sealed and stored at room temperature of 24 to 26°C for 45 d before being subjected to analyses.

**Table 1. The pH and Chemical composition of Ojeoksan<sup>1</sup> meal**

pH	Chemical composition					
	Moisture	Crude protein	Crude fat	Crude ash	NDF <sup>2</sup>	ADF <sup>3</sup>
	%	.....	%	DM	.....	
5.48	74.6	9.5	2.7	6.7	48.9	43.2

<sup>1</sup> Consist of *Atractylodis Lanceae*, *Ephedra sinica* Stapf, *Citrus unshiu*, *Magnoliae Cortex*, *Platycodon grandiflorum*, *Angelicae Radix*, dried *Zingiber officinale* Roscoe, *Paeoniae Radix Alba*, *Cnidium officinale* Makino, *Angelica dahurica*, *Pinelliae Tuber*, *Glycyrrhiza uralensis* Fisch

<sup>2</sup> Neutral detergent fiber.

<sup>3</sup> Acid detergent fiber.

**Measurements**

**Analyses of Ojeoksan meal** : Proximate compositions (AOAC, 1995) and detergent fibers (Van Soest, 1970) of Ojeoksan meal were analyzed. In order to measure pH, 100 g Ojeoksan sample and 100 mL distilled water are boiled and sealed, settled at 2 to 4°C for 24 h, and centrifuged at 3000 rpm for 20 min. After that, the most upper layer of the mixture was decanted. In order to measure organic acids content, 2 g homogenized sample was transferred to a test tube, and 4 mL pure water were added so that the silage could be submerged, and settled for 24 h. Then the mixture was centrifuged at 3000 rpm for 20 minutes, decanted the supernatant of the mixture, and passed through a syringe filter (0.2 μmpore). Aliquot of the filtrated liquid was taken and analyzed with HPLC (Waters 1260, USA) under following conditions.

**Counting total microbial cell in silages** : Aculture experiment was conducted to investigate the state of microbes in silage fermentation

process. Aliquot samples for organic acid analysis were taken and diluted with aseptic water to be diluted 100 times. These diluted samples, 0.5 mL each, were incubated on culture media such as MRS(Ventech-Bio Inc., Korea) or PDA(Ventech-Bio Inc., Korea) at 30°C for 72 h. After the incubation, total microbial cells were counted.

**HPLC condition for the analyses of organic acids**

Items	Conditions
Column	SUPELCOGEL C610H
Detector	UV, 210nm (Waters 2487)
Flow rate	0.5 mL/min
Solvent	0.1% phosphoric acid
Absorbance	210 nm
Injection volume	20 μl

**In vitro dry matter disappearance rate of Ojeoksan meal silage** : The method of Maeng et al. (1989) were adopted in this experiment. Rumen liquid was obtained from a mature female Hanwoo immediately after slaughter in a commercial slaughter house. Solution A

( $\text{KH}_2\text{PO}_4$ , 10.0 g;  $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ , 0.5 g; NaCl, 0.5 g;  $\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$ , 0.1 g; urea 0.5 g per 1 L of distilled water) was mixed with solution B ( $\text{Na}_2\text{CO}_3$  15.0 g;  $\text{Na}_2\text{S} \cdot 9\text{H}_2\text{O}$  1.0 g per 1 L of distilled water) at the rate of 5:1 to make 1600 mL buffer solution to reach pH 6.8 at 39°C, and thereafter mixed with filtrated 400 mL rumen liquid). Approx. 100 g of each Ojeoksan meal silage sample were dried and ground to pass through 1 mm sieve, and 0.5 g of each sample was put into a filter bag, replicated three times per sample. Blank bags with no sample were also prepared. When measuring digestion rate, artificial rumen (DAISY<sup>II</sup>, USA) was maintained at  $39.5 \pm 0.5^\circ\text{C}$ . After culturing for 48 hours, each filter bag was taken out of the artificial rumen, cleansed several times with cold tap water and dried in drying oven at  $105^\circ\text{C}$  until constant weight was reached. Dry matter disappearance rate was calculated as follows.

$$\text{Dry matter disappearance rate}(\%) = 100 - \{W3 - (W1 \times W4)\} \times 100 / W2$$

where

W1 = bag tare wt.

W2 = sample wt.

W3 = final bag wt.

W4 = blank bag correction (final blank bag wt. / original bag wt.)

### Statistical analysis

Data were subjected to one-way ANOVA using GLM of SAS/STAT 6.03 Package

(SAS, 1996). When significant at the level 5%, treatment means were compared with Duncan's new multiple range tests (Duncan, 1955).

### Experiment II: Palatability of herbal medicine meal silage in goats

Palatability of Ojeoksan meal silage was measured by a cafeteria-style feeding trial (Morimoto, 1971). Nine 1.5-yr-old Korean native goats of approx. 15 kg BW were acclimatized for 10 d, followed by 3 d feeding period. Each goat was housed in an individual metabolism cage equipped with four-sectioned roughage feeder and a separate concentrates bucket. To each section one of the four silages (0, 0.5, and 1.0% molasses supplemented Ojeoksan meal silages, and corn silage) was randomly allotted at 09:00, 300 g of each silage. At 15:00 500 g of a commercial concentrates for fattening were offered to each goat through the bucket. The amounts of each silages consumed during 09:00-09:30 and 09:00-15:00 were measured daily for 3 d.

Data were subjected to one-way ANOVA, four treatments with nine replicates per treatment, of GLM in SAS/STAT 6.03 Package (SAS, 1996). When significant at 5%, Duncan's new multiple range tests (1955) were employed to compare the treatment means.

## III. Results and Discussion

### Experiment I: Graded levels of molasses

supplementation to herbal medicine meal on silage quality

**Chemical composition, pH and organic acid contents of Ojeoksan meal silage**

The molasses supplementation did not affect the proximate composition, and detergent fiber contents of Ojeoksan meal silages (Table 2). Moisture contents were somewhat high, so that nutrient loss due to the drain of juice and reduction of DM intake could be occurred. Similar results were observed by Kim et al. (2006), in that Daeshotang meal, also one of the herbal medicine meal in Korea, was used instead of Ojeoksan meal.

The pH of the Ojeoksan meal silage decreased gradually as the molasses supplementation level increased, probably reflecting the increased lactic acid contents. As molasses supplementation level increased, acetic acid content decreased, and butyric acid vice versa. These results are well agreed with those of Kim et al. (2006), who used herbal medicine meal silage. Several

workers (Pathak and Lee, 1987; El-Yassin et al. 1991 Kwak and Park, 2003) also reported beneficial effects of molasses supplementation in silage fermentation, probably due to its high water-soluble carbohydrates contents. McDonald (1981) reported that the achievement of a pH value of about 4.0 will prevent clostridial activity and stable preservation of silage by lactic acids fermentation requires not only low buffering capacity but also a certain level of content.

**Total microbial cell counts**

The total microbial cell counts in MRS culture medium, which was targeted for *Lactobacillus*, increased as molasses supplementation level increased (Table 3). However, the number of fungi and yeast, as shown in PDA culture medium, decreased as molasses level gradually increased. Apparently, molasses provided advantageous condition for lactic acids fermentation, and disadvantageous condition for butyric acid bacteria and fungi, indicating that molasses supplementation

**Table 2. Chemical composition, pH and organic acid of Ojeoksan meal silage**

Molasses levels	Moisture	pH	Chemical composition					
			Crude protein	NDF	ADF	Acetic acid	Butyric acid	Lactic acid
	... % ...		..... % DM.....					
0%	76.6	4.95a	9.3	53.5	48.9	0.90 <sup>b</sup>	0.62 <sup>a</sup>	1.19 <sup>c</sup>
0.5%	76.9	4.53b	9.5	50.1	45.1	0.93 <sup>ab</sup>	0.39 <sup>b</sup>	2.15 <sup>b</sup>
1.0%	77.0	4.16c	9.5	51.0	46.5	0.95 <sup>a</sup>	0.37 <sup>b</sup>	2.46 <sup>a</sup>
SEM <sup>1</sup>	0.12	0.23	0.07	1.02	1.11	0.01	0.08	0.38

<sup>1</sup> Standard error of the mean.

<sup>a-c</sup> Means within a column with same superscripts are not significantly different(P>0.05).

**Table 3. Total microbial cell counts in Ojeoksan meal silage**

Molasses level	Total microbial cells in, log cfu/g	
	MRS	PDA
0%	3.81 <sup>b</sup>	2.4 <sup>a</sup>
0.5%	4.18 <sup>ab</sup>	2.18 <sup>ab</sup>
1.0%	4.30 <sup>a</sup>	1.10 <sup>b</sup>
SEM	0.14	0.10

<sup>a,b</sup> P<0.05.**Table 4. In vitro DM disappearances of Ojeoksan meal silage**

Molasses level	Dry matter disappearance, %
0%	43.7 <sup>b</sup>
0.5%	44.9 <sup>ab</sup>
1.0%	45.3 <sup>a</sup>
SEM	0.53

<sup>a,b</sup> P<0.05.**Table 5. DM matter intake of Ojeoksan meal silage and corn silage in Korean native goats**

Silages	DM intake, g/head		
	for 30 min <sup>2</sup>	for 6 h <sup>3</sup>	
Ojeoksan meal silage	0% molasses	9.5 <sup>ab</sup>	38.5 <sup>a</sup>
	0.5% molasses	9.6 <sup>a</sup>	39.1 <sup>a</sup>
	1.0% molasses	10.2 <sup>a</sup>	40.3 <sup>a</sup>
Corn silage <sup>1</sup>		8.9 <sup>b</sup>	32.9 <sup>b</sup>
	SEM	0.22	0.53

<sup>1</sup> Corn silage was prepared without molasses supplementation. Its lactic acid content was 2.5%, and pH 4.2.

<sup>2</sup> For the first 30 min right after the silage was offered (09:00-09:30).

<sup>3</sup> DM intake between 09:00 and 15:00, when concentrates was offered.

<sup>a,b</sup> P<0.05.

could be a useful tool to improve preservation of herbal medicine silage.

### ***In vitro* DM disappearance of Ojeoksan meal silage**

The 1.0% molasses treatment displayed significantly higher level of DM disappearance compared to the 0% control treatment (Table 4), and the 0.5% molasses treatment in between. Cho et al.(1989) also observed the beneficial effect of molasses on the DM disappearance rate of silage. Kim et al.(2006) reported that as molasses supplementation level increased, digestibility of DM, NDF, and ADF of herbal medicine meal increased. The molasses supplementation also improved the OM digestibility in sheep (Martin et al., 1981), and *in vitro* trial (Petersen et al., 1981).

### **Experiment II: Palatability of herbal medicine meal silage in goat**

As shown in Table 5, molasses supplementation did not exert any influence on the DM intakes of Ojeoksan meal silages in Korean Native goats. It is of interest to note that the Ojeoksan meal silages showed significantly better palatability than the corn silage regardless of times measured, or molasses supplementation. However, Jeon et al. (1999) reported that the Formosan deer preferred corn silage to herbal medicine meal silage, indicating that the palatability of forages might be different among various ruminant animals.

It appears that the molasses supplementation at 1.0% level could enhance the quality of Ojeoksan meal silage by stabilizing the silage material sooner, and improving the digestibility of Ojeoksan meal silage. It turned out that the palatability of Ojeoksan meal silage was better than that of corn silage in goats, warranting further studies in this regard.

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