The Respective Effects of Shoot Height and Conservation Method on the Yield and Nutritive Value, and Essential Oils of Wormwood (*Artemisia montana* Pampan)

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ABSTRACT : This study was conducted to evaluate the shoot height at which the yield and nutritive value of wormwood (*Artemisia montana*) is optimized in order to provide information on its potential to support animal production (Experiment 1). A second objective was to determine how the essential oil (EO) concentration in wormwood hay and silage differ (Experiment 2). In Experiment 1, *Artemisia montana* was harvested at five different shoot heights (20, 40, 60, 80 and 100 cm) from triplicate 1.8×1.8 m plots. Dry matter (DM) yield was measured at each harvest date and the harvested wormwood was botanically separated into leaf, stalk and whole plant fractions and analyzed for chemical composition and *in vitro* dry matter digestibility (DMD). Values for total digestible nutrients (TDN), digestible energy (DE) and metabolizable energy (ME) were subsequently calculated using prediction equations. Dry matter yields of stalk and whole plant increased linearly ($p<0.001$) and leaf yield increased quadratically ($p<0.01$) with shoot height, whereas the leaf/stalk ratio decreased linearly ($p<0.001$). As shoot height increased, there was a linear increase ($p<0.001$) in leaf DM, ether extract (EE) and neutral detergent fiber (NDF) contents and a quadratic increase ($p<0.05$) in leaf acid detergent fiber (ADF) and nitrogen free extract (NFE) contents, and stalk and whole plant DM ($p<0.001$), organic matter (OM, $p<0.01$ and $p<0.05$), NDF ($p<0.001$ and 0.05) and NFE ($p<0.05$) contents. However, there were decreases in leaf crude protein content (CP, quadratic, $p<0.001$) and stalk and whole plant EE content (linear, $p<0.001$), CP (quadratic, $p<0.05$) and ash (quadratic, $p<0.05$) contents. Digestibility of DM and TDN, and DE and ME value in leaves were not affected by increasing shoot height, but these measures linearly decreased ($p<0.001$) in stalk and whole plant. In Experiment 2, the hay had higher DM and CP concentrations, but lower EE concentration than the silage. Essential oil (EO) content in wormwood silage (0.49 g/100 g DM) was higher ($p<0.05$) than that in wormwood hay (0.32 g/100 g DM). Wormwood hay contained 25 essentail oils (EO) including camphor (10.4 g/100 g), 1-borneol (11.6 g/100 g) and caryophyllene oxide (27.7 g/100 g), and wormwood silage had 26 EO constituents including 3-cyclohexen-1-ol (8.1 g/100 g), *trans*-caryophyllene (8.6 g/100 g) and γ-selinene (16.8 g/100 g). It is concluded that the most ideal shoot height for harvesting wormwood is 60 cm based on the optimization of DM yield and nutritive value. Wormwood silage had a greater quantity and array of EO than wormwood hay. ***(Asian-Aust. J. Anim. Sci. 2006. Vol 19, No. 6 : 816-824)***

Key Words : Wormwood, Maturity, Yield, Pepsin-cellulase Digestibility, Essential Oil

INTRODUCTION

Wormwood (*Artemisia montana*) belongs to the family *Compositae* which includes about 300 species (Weyerstahl et al., 1987), 30 of which are found in South Korea (Lee, 1975). Due to the bitter taste of wormwood caused by sesquiterpene lactones (absinthin and anabsinthin), animals usually avoid eating fresh wormwood. Moreover wormwood contains volatile compounds like cineole and camphor which can inhibit the growth of neighboring grasses in the pasture (Schlattererr and Tisdale, 1969; McCahon et al., 1973). These factors have limited the use of wormwood as a forage source for ruminants. However, wormwood contains various bioactive compounds including phenolic acids, alkaloids and essential oils (EO); consequently it has been used in traditional European and Oriental folk medicine (Lee, 1975; Cosnier, 1989). Its anti-oxidant, anti-carcinogenic, anti-malarial and anti-bacterial properties have been documented (Hoffmann and Herrmann, 1982; Lee et al., 1999). There has been a lot of interest in using functional forages which have bioactive properties to improve human health, feed quality and to enhance the quality of animal products for consumers (Jang et al., 2004; Cho et al., 2006). Kim et al. (2002a) discovered that wormwood had a high CP (14-16%) content and when dry wormwood was fed to Hanwoo steers (Korean native beef cattle), their loin eye area was increased whereas back-fat thickness was decreased. Kim et al. (2002b, 2004) also reported that the omega 3 fatty acid concentration of the loin and top round lipid increased ($p<0.05$) when Hanwoo steers were fed dry wormwood or wormwood silage. Previously, Kim (1996) reported that South Korea wormwood contains 4.9-5.2 mg/g DM of EO. Recently, EO intake was found to increase live-weight gain, DM intake and N digestibility in steers at low inclusion rates (2-3 g/d DM), but decrease these measures at a higher inclusion rate (4 g/d DM; Benchaar et al., 2004). This study was

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Received November 29, 2005; Accepted February 24, 2006

conducted to determine the shoot height at which the nutritive value of wormwood is optimized and to determine how the EO content differs between wormwood hay and wormwood silage.

MATERIALS AND METHODS

Forage production

This study was conducted from March 2001 to June 2002 in South Korea which is a typical temperate region with hot, wet summers and dry winters. Wormwood (*Artemisia montana* Pampan) roots were transplanted (50 roots/m^2) from a nursery to twenty-five plots (1.8×1.8 m) at the research farm of Gyeongsang National University, Jin-ju (35°11'N lon., 128°5'E lat.), South Korea, in March 2001 and harvested and discarded in Nov. 2001. The soil at the experimental research farm was highly drained and loamy, and pH and organic matter content were 6.89 and 2.09%, respectively. Of the 25 plots that were planted, 18 were selected at random for the study. Fifteen plots were used for Experiment 1 and three plots were used for Experiment 2.

In Experiment 1, the wormwood regrowth in 2002 within three replicate plots was harvested manually with a sickle when shoot heights were 20, 40, 60, 80 and 100 cm. The respective harvest dates were on April 28, May 14, 23, and June 1 and 21. Shoot height had been monitored weekly between March and June 2002. Wormwood was harvested when the average shoot height in the plot reached the targets. Total biomass yield was measured and the forage was separated into leaves and stalks, and weighed.

In Experiment 2, wormwood was grown on three replicated plots as in Experiment 1, and harvested when the shoot height was 60-80 cm (25% DM). Half of the wormwood from each plot was air-dried under a covered barn for 7 d to make wormwood hay, and the other half was chopped to 3-4 cm lengths and ensiled without wilting in 3 kg experimental glass bottle silos for 60 d. These methods of conservation were used because fresh wormwood is not readily consumed by ruminant livestock, whereas ensiled, or dried wormwood have increased feed intake of Hanwoo steers and sheep (Kim et al., 2004, 2006a, b; Ko et al., 2006).

Chemical analysis

The dry matter (DM) content of harvested wormwood was determined in triplicate within an oven set to 105°C for 24 h. Samples were also dried in a 65°C forced draught oven for 48 h and then, ground to pass through a 1 mm screen with a Wiley mill. Ether extract (EE), crude protein (CP) and ash were analyzed using the AOAC (1990) method, and neutral detergent fibre (NDF) and acid detergent fibre (ADF) were quantified by the methods of Van Soest et al. (1991). Amylase and sulphite were used in the NDF procedure and the results were not expressed on an ash free basis.

Pepsin-cellulase DM digestibility (DMD) was measured with pepsin (1:10,000, Sigma-aldrich, Co. P. O. Box 14508 St. Louis, MO 63178, USA, 314-771-5750) and cellulase (Sigma-aldrich, Co. P. O. Box 14508, St. Louis, MO 63178 USA, 314-771-5750) using the McLeod and Minson (1978) method. Dried and ground samples (0.5 g) were incubated in tubes for 48 h at 39°C with 50 ml of 0.1 N HCl containing 0.2% (w/v) pepsin (1:10,000). The contents of the tubes were centrifuged at 2,200×g for 10 min and the supernatant removed after filtration (Whatman 4). The residue was washed with 75 ml double distilled water, and incubated again with 50 ml of 0.05 M acetate buffer solution (pH 4.6) containing chloramphenicol (100 ug/ml) and 2.5% cellulose for 48 h. Afterwards the contents of the tubes were centrifuged at 2,200×g for 10 min, and the residues were collected with filter paper (Whatman 4), washed twice with 75 ml double distilled water and analyzed for DM content. Total digestible nutrients (TDN) were calculated using Abe and Horii's equation (1974), and digestible energy (DE) and metabolizable energy (ME) were estimated with NRC (1973) equations. These equations are as follows;

$$\text{TDN (\%)} = 14.9+0.737\ \text{DMD}$$

$$\text{DE (Kcal/g)} = \text{TDN (\%)}\times 4.409$$

$$\text{ME (Kcal/g)} = \text{TDN (\%)}\times 3.6155$$

The chemical composition of wormwood hay and silage in Experiment 2 was analyzed using the same methods as in Experiment 1. To extract silage fluid, 70 g of wormwood silage was mixed with 140 ml of double distilled water and the mixture was homogenized for 30 sec., and then stored at 4°C for 24 h. The mixture was filtered through four layers of cheese cloth and the pH was measured immediately with an electric pH meter, after which the silage extract was stored until analysis (-25°C). The silage extract was defrosted at room temperature and centrifuged at 1,465×g for 15 minutes, and the supernatant analyzed for NH_3-N by colorimetry (Chaney and Marbach, 1962), total volatile fatty acids (VFA) by steam distillation (Fenner and Elliot, 1963) and individual VFA by gas chromatography (GC, Erwin et al., 1961). The GC (Hewllett Packard GC-5890 series II) was equipped with a flame ionization detector (FID) and a HP-Innowax column (Crosslinked Polyethylene Glycol; 30 m×0.32 mm×0.5 um). The carrier gas was hydrogen, and the injector and detector temperatures were 250°C and 280°C, respectively. Initial oven temperature

Table 1. Meteorological data collected during the growing period (March to June, 2002)

Items		March	April	May	June
Temperature (°C)	Highest	16.4	20.8	23.7	28.5
	Lowest	1.1	7.2	11.9	15.8
	Mean	8.7	14.1	17.5	22.0
	10-year mean	10.7	15.4	17.5	25.8
Precipitation (mm)		100.7	152.1	150.6	81.7
	10-year mean	102.7	167.5	155.3	88.7

Table 2. Effects of shoot height on dry matter yield of botanical fractions and leaf to straw ratio of wormwood (*Artemisia montana*)

	Shoot heights, cm					SEM	Contrasts[1]	
	20	40	60	80	100		L	Q
Cutting dates	April 28	May 14	May 23	June 1	June 12			
Leaf (t DM /ha)	2.2	2.5	2.6	2.8	3.4	0.10	***	**
Stalk (t DM /ha)	0.9	1.0	3.0	3.7	4.4	0.11	***	NS
Whole plant (t DM /ha)	3.1	3.5	5.6	6.5	7.8	0.20	***	NS
Leaf/stalk ratio	2.5	2.4	0.9	0.8	0.8	0.07	***	***

[1] L: Linear effect, Q: Quadratic effect.
SEM: Standard error of the mean. NS: Not significant. * $p<0.05$, ** $p<0.01$, *** $p<0.001$.

was 120°C for 1 min. Following sample injection, the oven temperature was increased by 10°C/min to 180°C and held for 10 min. Individual VFAs were identified by comparison of retention times to those of pure standards. Essential oils (EO) in wormwood hay and silage from Experiment 2 were extracted by simultaneous steam distillation (Schultz et al., 1977), and analyzed using a gas chromatograph/mass spectrometer (Hewllet Packard GC-5890A) equipped with a flame ionization detector (FID) and a DB-5 column (60 m× 0.32 mm×0.25 um). The carrier gas was nitrogen, and the temperature of injector and detector was 230°C. Initial oven temperature was 50°C for 5 min, and this was increased by 4°C/min to 200°C, and held for 18 min. The mass spectrometer was a Joel JMS-700 (Joel Ltd., West Yorkshire, UK), and Wiley 138 library was used to identify each peak.

Statistical analysis

The data in Experiment 1 were analyzed as a completely randomized design using the General Linear Model procedure of SAS (1990). Polynomial contrasts were used to determine the influence of increasing wormwood shoot height in Experiment 1. The Tukey test was used to identify differences ($p<0.05$) between means in Experiment 1 and Experiment 2.

RESULTS

The meteorological data collected during the harvesting period (March 2002-June 2002) are shown in Table 1. The average temperature and precipitation were 15.6°C and 121.3 mm, respectively.

DM yields of stalk and whole plant increased ($p<0.001$) linearly with increasing shoot height while leaf yields increased quadratically ($p<0.01$) (Table 2). The ratio of leaf to stalk decreased ($p<0.001$) quadratically as shoot height increased.

The chemical composition of leaf, stalk and whole plant from the harvested wormwood is shown in Table 3. Increasing the shoot height did not affect organic matter (OM) and ash contents in leaves, but linearly increased leaf EE ($p<0.001$) and NDF ($p<0.01$) contents, quadratically increased DM ($p<0.001$), ADF ($p<0.05$) and nitrogen free extract (NFE, $p<0.05$) contents, and quadratically decreased CP ($p<0.001$). The contents of DM ($p<0.001$), OM ($p<0.01$), NDF ($p<0.001$) and NFE ($p<0.05$) in the stalk were increased quadratically with increasing shoot height, whereas CP (quadratic), EE (linear) and ash (quadratic) contents decreased ($p<0.001$). As shoot height increased, the contents of DM, OM, NDF and NFE in the whole plant increased ($p<0.01$) quadratically while ADF content increased ($p<0.05$) linearly. The contents of CP and ash in the whole plant decreased ($p<0.001$) quadratically while EE content decreased ($p<0.001$) linearly. The shoot height-related decreases in CP and EE, and increases in NDF and ADF were particularly pronounced in plants that were between 60 and 80 cm tall.

Shoot height did not affect pepsin-cellulase dry matter digestibility (DMD) and concentration of total digestible nutrients (TDN), digestible energy (DE) and metabolizable energy (ME) in leaves (Table 4). However, these measures decreased ($p<0.001$) linearly with increasing shoot height in the stalk and whole plant samples. In particular, stalk and whole plant DMD, TDN, DE and ME values decreased substantially ($p<0.05$) between 60 and 80 cm.

Wormwood hay had higher contents of DM (89.4 vs. 25.6 g/100 g, $p<0.001$) and CP (13.6 vs. 10.8 g/100 g DM, $p<0.05$) than silage, whereas wormwood silage had higher

Table 3. Effect of shoot height on the chemical composition (g/100 g DM basis) of botanical fractions of wormwood (*Artemisia montana*)

	Shoot heights (cm)					SEM	Contrasts[1]	
	20	40	60	80	100		L	Q
Leaf								
Dry matter	26.98	17.22	24.00	31.03	28.99	0.49	***	***
Organic matter	91.83	91.30	91.15	91.50	91.49	0.34	NS	NS
Crude protein	25.00	21.82	20.43	16.98	17.40	0.99	***	*
Ether extract	3.46	3.89	4.07	4.05	4.17	0.15	***	*
Crude ash	8.17	8.70	8.85	8.50	8.51	0.34	NS	NS
Neutral detergent fiber	29.30	28.48	32.60	31.66	32.72	1.39	**	NS
Acid detergent fiber	35.62	34.58	36.88	37.30	32.52	1.58	NS	*
Nitrogen free extract	55.48	49.40	49.98	53.71	51.74	2.19	NS	*
Stalk								
Dry matter	18.99	15.14	17.00	33.03	27.99	0.70	***	***
Organic matter	93.60	93.00	93.85	96.04	96.19	0.30	***	**
Crude protein	9.41	8.20	6.58	4.58	4.78	0.44	***	*
Ether extract	1.91	2.09	1.31	0.59	0.62	0.12	***	NS
Crude ash	6.4	7.00	6.14	3.96	3.81	0.30	***	**
Neutral detergent fiber	36.86	40.57	51.26	47.26	51.29	1.38	***	***
Acid detergent fiber	35.16	39.40	39.07	39.56	44.29	1.24	***	NS
Nitrogen free extract	60.00	56.36	51.97	59.44	55.67	2.14	NS	*
Whole plant								
Dry matter	24.13	16.46	19.70	32.13	28.42	0.56	***	***
Organic matter	92.32	91.88	92.58	94.06	94.17	0.29	***	*
Crude protein	20.63	17.23	13.10	9.97	10.22	0.48	***	***
Ether extract	3.02	3.29	2.61	2.09	2.15	0.09	***	NS
Crude ash	7.68	8.12	7.42	5.94	5.83	0.29	***	*
Neutral detergent fiber	31.42	32.55	42.48	40.48	43.30	1.31	***	*
Acid detergent fiber	35.49	36.20	38.04	38.58	39.22	1.32	**	NS
Nitrogen free extract	56.78	51.75	51.04	56.95	52.98	2.21	NS	*

[1] L: Linear effect, Q: Quadratic effect.
SEM: Standard error of the mean. NS: Not significant, * p<0.05, ** p<0.01, *** p<0.001.

contents of EE (3.1 vs. 2.4 g/100 g DM, p<0.05) than hay (Table 5). The NFE content in wormwood hay tended (p = 0.069) to be higher than that in silage. The pH, and concentrations of NH_3-N and lactic acid in wormwood silage were 4.1, 32.9 g/kg DM and 28.1 g/kg DM, and the concentrations of total VFA (excluding lactic acid), acetate, propionate and butyrate in wormwood silage were 29.5, 11.5, 6.2 and 11.7 g/kg DM, respectively. The ratio of NH_3-N to total N was 1.2 in wormwood silage.

The essential oil (EO) constituents in wormwood hay and silage are shown in Table 6. Total EO in wormwood silage (0.49 g/100 g DM) was higher (p<0.05) than that in wormwood hay (0.32 g/100 g DM). Twenty-five EO were identified in wormwood hay, and the main components were Camphor (10.4 g/100 g of EO), 1-borneol (11.6 g/100 g of EO) and caryophyllene oxide (27.7 g/100 g of EO). In wormwood silage, 26 EO were identified and 7 unknown peaks were detected. The main constituents were 3-cyclohexen-1-ol (8.1 g/100 g of EO), *trans*-caryophyllene (8.6 g/100 g of EO) and γ-selinene (16.8 g/100 g of EO). Concentrations of 1.8-cineole, isocyclocitral, camphor, 1-borneol, *trans*-caryophyllene, α-humulene, bezene, caryophyllene oxide, (-)-isolongifolol and δ-cadinol in wormwood hay were higher (p<0.05) than in wormwood silage. However concentrations of 3-cyclohexen-1-ol, γ-selinene, torreyol and Δ-cadinene in wormwood silage were higher (p<0.05) than those in wormwood hay, and γ-gurjunene and hexadecanoic acid were detected in wormwood silage but not in wormwood hay.

DISCUSSION

The wormwood species used in the present study grows widely in South Korea and shoot heights of 60-120 cm are common (Kim, 1996). However, Rho and Seo (1994) reported that DM yield and plant height of wormwood (*Artemisia montana*) were 600 kg/ha and 7 cm, respectively in March in South Korea due to slow early growth. This study shows that yields and shoot heights increase greatly between April and June. The DM yield of leaves increased slightly with increasing shoot height while those of the stalk and whole plant increased considerably. Classically the ratio of leaf/stalk decreases with increasing plant maturity (Merchen and Bourquin, 1994). This trend was evident in

Table 4. Effects of shoot height on *in vitro* dry matter digestibility and energy value (DM basis) of botanical fractions of wormwood (*Artemisia montana*)

	Shoot heights (cm)					SEM	Contrasts[1]	
	20	40	60	80	100		L	Q
Leaf								
DMD[2] (g/100 g)	46.50	47.81	47.75	46.14	47.55	1.58	NS	NS
TDN (g/100 g)	49.17	50.13	50.10	48.90	49.95	1.17	NS	NS
DE (kcal/g)	2.17	2.21	2.21	2.16	2.20	0.05	NS	NS
ME (kcal/g)	1.78	1.81	1.81	1.77	1.81	0.04	NS	NS
Stalk								
DMD (g/100 g)	51.44	48.64	46.91	44.21	42.01	0.88	***	NS
TDN (g/100 g)	52.81	50.75	49.47	47.48	45.86	0.65	***	NS
DE (kcal/g)	2.33	2.24	2.18	2.09	2.02	0.03	***	NS
ME (kcal/g)	1.91	1.83	1.79	1.72	1.66	0.02	***	NS
Whole plant								
DMD (g/100 g)	47.85	48.06	47.15	45.05	44.42	1.19	***	NS
TDN (g/100 g)	50.16	50.32	49.68	48.10	47.64	0.87	***	NS
DE (kcal/g)	2.21	2.22	2.19	2.12	2.10	0.04	***	NS
ME (kcal/g)	1.81	1.82	1.80	1.74	1.72	0.03	***	NS

[1] L: Linear effect, Q: Quadratic effect.
[2] DMD: Dry matter digestibility; TDN: Total digestible nutrients; DE: Digestible energy; ME: Metabolizable energy.
SEM: Standard error of the mean.
NS: Not significant, * p<0.05, ** p<0.01, *** p<0.001.

Table 5. Chemical composition (DM basis) of wormwood (*Artemisia montana*) hay and silage

	Wormwood hay	Wormwood silage	SEM	p value
Dry matter (g/100 g)	89.43	25.57	1.566	***
Organic matter	92.04	91.34	1.622	NS
Crude protein	13.56	10.78	0.876	*
Ether extract	2.41	3.12	0.279	*
Crude ash	7.96	8.66	1.078	NS
Neutral detergent fiber	52.12	51.45	1.591	NS
Acid detergent fiber	42.76	42.75	1.344	NS
Nitrogen free extract	37.51	35.17	1.166	NS
pH		4.11	0.176	
NH_3-N (g/kg)		32.9	4.28	
Lactic acid (g/kg)		28.1	2.51	
Total VFA (g/kg)		29.5	4.33	
Acetic acid (g/kg)		11.5	1.68	
Propionic acid (g/kg)		6.2	1.19	
Butyric acid (g/kg)		11.7	4.41	
NH_3-N/total N ratio		1.2	0.21	

SEM: Standard error of the mean.
NS: Not significant, * p<0.05, ** p<0.01, *** p<0.001.

the present study.

Rho and Seo (1993) reported that CP content of *Artemisia montana* leaves was 17-19 g/100 g DM at 109-133 cm shoot height. They also reported in another study (1994) that CP content in *Artemisia montana* was 16-19 g/100 g DM when it was harvested on March 25. Kim (1995) reported that when wormwood was harvested in April, the CP content was 19 g/100 g DM. These results are similar to those in the present study. Maturity is usually considered as the primary factor that determines the chemical composition and nutrient quality of grass (Nelson and Moser, 1994). Generally, the components of cell walls such as hemicellulose, cellulose and lignin increase whereas WSC, total and soluble N concentrations decrease with maturity (Merchen and Bourquin, 1994; Hill and Leaver, 1999; Saddul et al., 2005). These typical trends were also evident in this study.

Theoretically, lignification of forages is the primary factor influencing the extent of fiber digestion, though it has no inhibitory effect on ruminal degradation of the soluble components (Kozlosiki et al., 2005). Kim et al. (1989) reported that *in vitro* pepsin cellulase DMD, TDN, DE and ME of Korean domestic herbage like *Agropyron tsukushience*, *Digitaria sanguinalis*, *Arundinella hirta* decreased with maturity. Cerrillo et al. (1999) noted that the ruminal digestibility of DM, NDF, ADF and CP decreased

Table 6. The essential oil constituents (DM basis) of wormwood (*Artemisia montana*) hay and silage

Volatile components (g/100 g of wormwood)	Wormwood hay	Wormwood silage	SEM	p value
Total essential oil	0.32	0.49	0.046	**
Composition of EO (g/100 g of EO)				
2,5,5-trimethyl-3,6-heptadien-2-ol	1.33	1.19	0.360	NS
1,8-cineole	3.84	1.02	0.359	***
Isocyclocitral	1.08	0.49	0.249	*
Camphor	10.38	0.55	0.849	***
1-borneol	11.64	4.88	1.774	**
3-cyclohexen-1-ol	0.69	8.14	1.357	**
Bicyclo[3.1.1]hep-3-en-2-one	1.53	1.61	0.579	NS
1-α-terpineol	0.46	0.86	0.311	NS
Naphthalene	2.94	2.90	0.303	NS
Trans-caryophyllene	7.75	8.64	0.960	NS
α-humulene	3.24	0.39	0.251	***
γ-selinene	1.33	16.83	1.051	***
Benzene	2.00	0.72	0.327	**
Torreyol	1.03	5.89	0.391	***
Δ-cadinene	1.78	3.82	0.345	**
(-)-caryophyllene oxide	3.13	3.89	0.699	NS
Caryophyllene oxide	27.7	1.00	2.667	***
Veridiflorol	0.85	0.58	0.292	NS
(-)-isolongifolol	6.84	2.07	0.818	**
Fonenol	1.00	0.66	0.427	NS
δ-cadinol	3.59	1.22	0.939	*
Trans-cardinol	2.39	3.01	0.896	NS
Lendene	0.55	0.71	0.412	NS
Campherenone	0.71	0.76	0.281	NS
Spathulenol	0.86	0.90	0.421	NS
Γ-gurjunene	-	2.84	0.622	**
Unknown 1	-	0.38	0.159	*
Unknown 2	-	5.27	0.7937	**
Unknown 3	-	0.37	0.125	*
Unknown 4	-	5.05	0.768	**
Unknown 5	-	6.04	0.636	***
Unknown 6	-	0.35	0.178	NS
Unknown 7	-	3.01	0.477	**
Hexadecanoic acid	-	0.86	0.381	NS

SEM: Standard error of the mean.
NS: Not significant, * $p<0.05$, ** $p<0.01$, *** $p<0.001$.

with maturity in alfalfa (*Medicago sativa* 1) and smooth bromegrass (*Bromus inermus* 1) hay. Van Soest (1994) also observed the existence of a negative curvilinear relationship between lignin content and NDF digestibility. Moreover, Van Soest (1996) reported a negative linear relationship between ADF content and fiber digestibility of forages growing during the spring. Generally, lignin contents increase with increasing forage maturity, and there is negative relationship between lignin content and DMD (French, 1957; Minson, 1971). Therefore, although lignin content was not analyzed in the present study, the linear decrease in whole plant DMD, TDN, DE and ME with maturity, and the rapid decrease of these measures at 60-80 cm shoot height in the stalk and whole plant would be most likely due to increasing lignification.

In Experiment 1, the yield of pepsin-cellulase digestible dry matter increased quadratically ($p<0.01$) to 1.48, 1.68, 2.64, 2.92 and 3.46 ton/ha DM, respectively with increasing shoot height indicating the high yield potential of wormwood. The ratio of leaf to stalk at the 60 cm shoot height (0.9) decreased greatly from that at 40 cm (2.4, Table 2). Furthermore, NDF concentration in the whole plant harvested at 60 cm shoot height was 31% higher than that at 40 cm shoot height (Table 3). The quadratic increase in yield of digestible DM implies that the yield and nutritive value of wormwood were optimized at the 60 cm shoot height. Yield was compromised at shoot heights below 60 cm while nutritive value was compromised at longer shoot heights. Since Korean farmers already rely on rice straw as a cheap source of dietary fiber and roughage, wormwood utilization in livestock rations will be enhanced if it is harvested at shoot heights below 80 cm. In particular, harvesting at 60 cm shoot height should be the target in order to optimize yield and nutritive value. These guidelines should be used for wormwood destined for hay or silage production since fresh wormwood is not readily eaten by ruminant livestock due to the presence of sesquiterpene lactones (absinthin and anabsinthin) which are responsible for the well-known bitter taste of wormwood (Skyles and Sweet, 2004). Kim et al. (2004) reported that when rice straw was replaced with wormwood silage in the diet of Hanwoo Korean native steers, feed intake and growth performance increased. Kim et al. (2006a) also demonstrated that replacing rice straw with wormwood silage increased feed intake and nitrogen utilization in sheep. Further, Kim et al. (2006b) showed that replacing concentrates with dry wormwood increased N retention, microbial N yield and EE digestibility in sheep.

The differences between the chemical compositions of wormwood hay and silage in the present study are similar to observations in other forages. In general, hay contains more CP and cell wall contents than silage (Thomas et al., 1968), suggesting that the hay would be a more appropriate protein supplement than the silage. The higher fat and volatile fatty acid concentration of the silage, suggests that it would be a better energy supplement than the hay.

Essential oils are mixtures of lipophylic liquids and terpenoid compounds (Hirasa and Takemasa, 1998). Kim (1996) observed that Korean wormwood (*Artemisia sp.*) contains 4.9-5.3 g/kg DM of EO and these EO had great volatility. Cho and Chiang (2001) reported that the EO concentrations in wormwood differed greatly between wormwood species. On DM basis, they found 0.27-0.5 g/100 g EO and 24 EO constituents in *Artemisia capillaries*, 0.4-1.2 g/100 g EO and 27 EO constituents in *Artemisia argyi* and 0.5-1.4 g/100 g EO and 24 EO constituents in *Artemisia princes,* respectively. These concentrations and numbers of EO constituents were similar to those of the dry wormwood (0.32 g/100 g EO, 25 constituents) and wormwood silage (0.49 g/100 g EO, 26 constituents+7 unknown constituents) in this study. Across species, Korean wormwood (*Artemisia sp.*) has been found to contain over 100 different EO like borneol, β-caryophyllene, camphor and Cineole (Kim, 1996). In the present study, these compounds were also identified from wormwood hay and silage. Kim et al. (1994) reported that EO content in fresh wormwood was higher than those in dried wormwood, and some EO that were not detected in fresh wormwood like 4-dione, butanamide, nonanal were identified in parched wormwood. They attributed this difference to volatilization or thermal degradation. The greater number and concentration of total EO in wormwood silage than hay may also be attributable to thermal degradation.

Relatively, little is known about the effects of EO on the performance of ruminants. Some EO have inhibited bacterial growth (Hirasa and Takemasa, 1998), but others seem to have improved nutrient utilization. When different levels of individual EO were applied to alfalfa pellets in sheep diets, intake was linearly reduced by camphor and α-pinene, quadratically increased by borneol, unaffected by β-caryophyllene, cis–jasmone and limonene (Estell et al., 1998). In contrast, a commercial EO product containing thymol, eugenol, vanillin and limonene increased steer liveweight gain and DM intake, and increased N digestibility at low inclusion rates (2-3 g/d DM), but decreased it at a higher inclusion rate (4 g/d DM; Benchaar et al., 2004). A different product containing thymol, guajacol and limonene has decreased ruminal protein degradability in some studies (Newbold, et al., 2003; Molero et al., 2004) but increased rumen VFA production without affecting CP digestion in another study (Castillejos et al., 2004). Further studies are required to clearly elucidate the effects of EO, particularly those in wormwood on nutrient metabolism in ruminants.

CONCLUSION

This study indicates that the nutritive value and yield of wormwood is optimized when the shoot height is 60 cm. Wormwood silage had higher EE concentration, but lower CP concentration than wormwood hay. Total EO concentration in wormwood silage was higher than in wormwood hay, and this was partly because there were more types of EO in the silage than the hay.

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