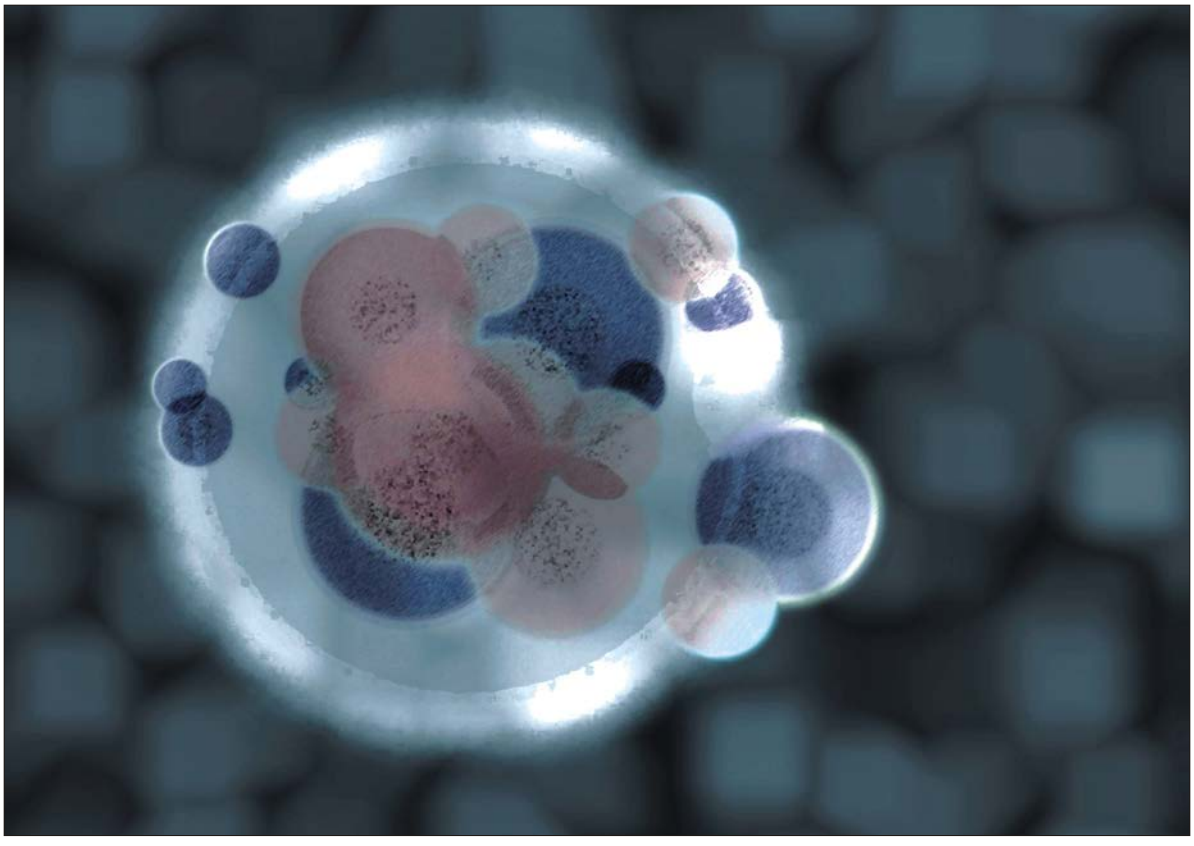

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Effect of Maturity on the Potential Nutritive Value of Burr Medic (*Medicago polymorpha*) Hay

[1]Rahim Aydin, [1]Adem Kamalak and [2]Onder Canbolat
[1]Department of Animal Science, Faculty of Agriculture,
Kahramanmaras Sutcu Imam University, Kahramanmaras, Turkey
[2]Department of Animal Science, Faculty of Agriculture, Bursa Uludag University, Bursa, Turkey

Abstract: The aim of this experiment was to determine the potential nutritive value of *Medicago polymorpha* hays harvested at vegetative, flowering and seeding stages using the chemical composition, *in vitro* gas production kinetics, Organic Matter Digestibility (OMD) and Metabolizable Energy (ME) content. Harvesting stage had a significant ($p<0.001$) effect on the chemical composition except for ash content, *in vitro* gas production kinetics, OMD and ME content of *Medicago polymorpha* hay. The cell wall contents of *Medicago polymorpha* hay were significantly increased with increasing maturity whereas crude protein content was decreased. The Acid Detergent Fibre (ADF) and Neutral Detergent Fibre (NDF) contents ranged from 31.16 to 43.69% and 41.66 to 66.23%, respectively. The CP content of *Medicago polymorpha* hays ranged from 13.67 to 22.05%. The OMD and ME content of *Medicago polymorpha* hays ranged from 40.66 to 79.76% and 10.42 to 11.78 MJ kg^{-1} DM. The most of the gas production kinetics, ME and OMD were negatively correlated with ADF and NDF content of *Medicago polymorpha* hay whereas the same parameters were positively correlated with CP content of *Medicago polymorpha* hay. Due to low level of cell wall contents and higher CP, OMD and ME *Medicago polymorpha* plant should be harvested or grazed at vegetative stage to obtain hay with higher nutritive value.

Key words: *Medicago polymorpha*, maturity, nutritive value, digestibility

INTRODUCTION

It is well known that forage has an important role in ruminant nutrition in terms of providing energy, protein and minerals. Burr medic (*Medicago polymorpha* L.) is one of self-generating legumes in the pasture in Mediterranean environment (Piano and Talamucci, 1996) and represent an important resource for grazing sheep during summer (Chriyaa *et al*., 1997).

Although the effect of maturity on the nutritive value of forages obtained from different plants is well established (Buxton, 1996; Kamalak *et al*., 2005a, b; Bal *et al*., 2006) there is little information about the nutritive value of hay obtained from *Medicago polymorpha* at different maturity stages.

Chemical composition, in combination with *in vitro* OMD and ME content are used to evaluate the potential nutritive value of the previously uninvestigated plants (Gulsen *et al*., 2004; Coskun *et al*., 2004; Kamalak *et al*., 2005a, b; Bal *et al*., 2006).

The aim of this experiment was to determine the potential nutritive value of *Medicago polymorpha* hays harvested at vegetative, flowering and seeding stages using the chemical composition, *in vitro* gas production kinetics, OMD and ME content

MATERIALS AND METHODS

Forage samples: *Medicago polymorpha* plants were harvested from plots established in the experiment unit of native pasture at vegetative, flowering and seeding stages in Kahramanmaras, Turkey, in 2004. Hay samples were shade-dried and milled in a hammer mill to pass through a 1 mm sieve for subsequent analysis.

Chemical analysis: Dry matter (DM) was determined by drying the samples at 105°C overnight and ash was determined by igniting the samples in muffle furnace at 525°C for 8 h. Nitrogen (N) content was measured by the Kjeldahl method (AOAC, 1990). Crude protein was calculated as N×6.25. Cell wall contents (NDF and ADF) of leaves were determined using the method described by Van Soest *et al*. (1991). All chemical analyses were carried

Corresponding Author: Rahim Aydin, Department of Animal Science, Faculty of Agriculture, Kahramanmaras Sutcu Imam University, Kahramanmaras, Turkey

out in triplicate in the laboratory of Department of Animal Science, Faculty of Agriculture, Kahramanmaras Sutcu Imam University.

***In vitro* gas production:** Fermentation of *Medicago polymorpha* hays samples were carried out with rumen fluid obtained from two fistulated sheep fed twice daily with a diet containing hay (60%) and concentrate (40%) following the method described by Menke and Steingass (1988). Approximately 200 mg hay samples were weighed into the glass syringes of 100 mL. The fluid-buffer mixture (30 mL) was transferred into the glass syringes of 100 mL. The glass syringes containing hay samples and rumen fluid-buffer mixture were incubated at 39°C. The syringes were gently shaken 30 min after the start of incubation. The gas production was determined after 3, 6, 12, 24, 48, 72 and 96 h of incubation. All samples were incubated in triplicate with three syringes containing only rumen fluid-buffer mixture (blank). The net gas productions for hay samples were determined by subtracting the volume of gas produced in the blanks. Gas production data were fitted to the model of Orskov and McDonald (1979).

$$Y = a + b\,(1-e^{-ct})$$

Where:
- a = The gas production (mL) from immediately soluble fraction
- b = The gas production (mL) from the insoluble fraction
- c = The gas production rate (%) for the insoluble fraction
- t = Incubation time (h)
- y = Gas produced at time t

The ME (MJ kg^{-1} DM) contents of *Medicago polymorpha* hays samples were calculated using equation of Menke *et al.* (1979) as follows:

$$ME\ (MJ\ kg^{-1}\ DM) = 2.20 + 0.136\ GP + 0.057\ CP$$

Where:
- GP = 24 h net gas production (mL/200 mg).
- CP = Crude protein (%)

Organic matter digestibility (OMD) (%) of *Medicago polymorpha* hays samples was calculated using equation of Menke *et al.* (1979) as follows:

$$OMD\ (\%) = 14.88 + 0.889\ GP + 0.45\ CP + 0.0651\ XA$$

Where:
- GP = 24 h net gas production (mL/200 mg).
- CP = Crude protein (%)
- XA = Ash content (%)

In vitro gas production measurements were carried out in the laboratory of Department of Animal Science, Faculty of Agriculture, Bursa Uludag University.

Statistical analysis: One-way analysis of variance (ANOVA) was carried out to compare chemical composition, gas production kinetics, OMD and ME content of *Medicago polymorpha* hays samples with stage as the main factor using General Linear Model (GLM) of Statistica for Windows (1993). Significance between individual means was identified using the Tukey's multiple range test (Pearse and Hartley, 1966). Mean differences were considered significant at $p<0.05$. Standard errors of means were calculated from the residual mean square in the analysis of variance. A simple correlation analysis was used to establish the relationship between chemical composition and gas production kinetics or OMD and ME content of *Medicago polymorpha* hays samples.

RESULTS AND DISCUSSION

The maturity stage had a significant ($p<0.001$) effect on the chemical composition of *Medicago polymorpha* hays except for DM and ash content (Table 1). The CP content of *Medicago polymorpha* hay harvested at vegetative stage was significantly ($p<0.001$) higher than those obtained at flowering and seeding stages whereas ADF and NDF contents of *Medicago polymorpha* hays obtained at vegetative stage were significantly ($p<0.001$) lower than those obtained at flowering and seeding stages. The CP contents of *Medicago polymorpha* hays ranged from 13.67 to 22.05%. The CP content of *Medicago polymorpha* hay harvested at vegetative stage was similar to those reported by Porqueddu (2000), but lower than that reported by Sitzia *et al.* (2000). The ADF and NDF content ranged from 31.16 to 43.69 and 41.66 to 66.23%, respectively. The cell wall (ADF and NDF) contents of *Medicago polymorpha* hay harvested at vegetative stage was lower than those reported by Porqueddu (2000) and Sitzia *et al.* (2000). On the other hand the CP, NDF and ADF contents of *Medicago polymorpha* hay harvested at seeding stage was similar to those reported by Fois *et al.* (2000). It is well established that the cell wall content of forages increase with increasing maturity (Gulsen *et al.*, 2004; Kamalak *et al.*, 2005a, b).

The maturity stages had a significant effect on the cumulative gas production (Fig. 1). The gas production after 96 h ranged from 69.17 and 73.83 mL per 200 g of hay. As can be seen from Fig. 1 the gas production decreased with increasing maturity. This result is in agreement with findings of Zinash *et al.* (1996), Lee *et al.* (2000) and Kamalak *et al.* (2005a, b).

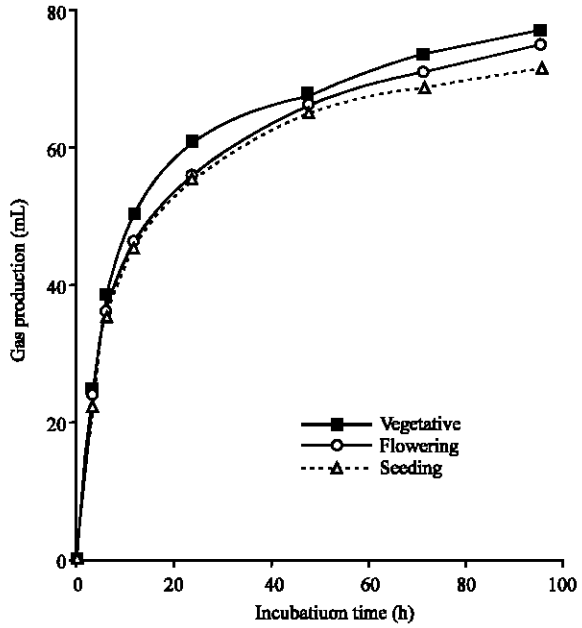

Fig. 1: The effect of maturity on the gas production of *Medicago polymorpha* hays

Table 1: The chemical composition (%) of *Medicago polymorpha* hays harvested at different maturity stages

	Maturity stages				
Parameters	Vegetative	Flowering	Seeding	SEM	Sig.
DM	97.15	97.49	97.00	0.233	NS
Ash	8.98	8.45	8.48	0.145	NS
CP	22.05^{b}	16.89^{a}	13.67^{a}	0.650	***
ADF	31.16^{a}	35.38^{b}	43.69^{c}	0.739	***
NDF	41.66^{a}	52.39^{b}	66.23^{c}	1.254	***

Means within the same row with various superscripts are significant; DM: Dry Matter; CP: Crude Protein; ADF: Acid Detergent Fibre; NDF: Neutral Detergent Fiber; SEM = Standard Error Mean; Sig = Significance level; NS = Non-significant; ***p<0.001

The maturity had a significant (p<0.001) effect on the gas production kinetics except for gas production rate (Table 2). There are no significant differences among *Medicago polymorpha* hays in terms of gas production rate. This result is in agreement with finding of Kamalak *et al.* (2005a) but is not in agreement with finding of Kamalak *et al.* (2005b).

The gas production from quickly soluble fraction of *Medicago polymorpha* hay harvested at flowering stage was significantly (p<0.001) higher than those obtained at vegetative and seeding stages whereas gas production (b) from the insoluble fraction and potential gas production (a+b) of *Medicago polymorpha* hay harvested at vegetative stage was significantly (p<0.001) higher than those flowering and seeding stages. The OMD and ME contents of *Medicago polymorpha* hay harvested at vegetative stages were also significantly (p<0.001) higher than those obtained at flowering and seeding stages.

Table 2: The effect of maturity stage on the gas production kinetics, organic matter digestibility (%) and metabolizable energy (MJ kg^{-1} DM) content of *Medicago polymorpha* hay

	Maturity stages				
Parameters	Vegetative	Flowering	Seeding	SEM	Sig.
c	0.104	0.090	0.092	0.004	NS
a	3.31^{a}	4.50^{b}	3.57^{a}	0.189	***
b	68.73^{b}	65.44^{a}	63.97^{a}	0.410	***
a+b	72.09^{c}	69.94^{b}	67.55^{a}	0.456	***
ME	11.78^{b}	10.78^{a}	10.42^{a}	0.083	***
OMD	79.76^{b}	72.81^{a}	70.66^{a}	0.640	***

c = Gas production rate (%); a = Gas production (mL) from quickly soluble fraction; b = Gas production (mL) from the insoluble fraction; a+b = Potential gas production (mL); OMD: Organic Matter Digestibility; ME: Metabolic Energy; Means within the same row with various superscripts are significant; SEM = Standard Error Mean; Sig: Significance level; ***p<0.001

Table 3: Correlation coefficient (r) relationship of chemical composition with gas production kinetics and some estimated parameters

Parameters	DM	Ash	CP	ADF	NDF
c	-0.126^{NS}	0.361^{NS}	0.677*	-0.527^{NS}	-0.524^{NS}
a	0.533^{NS}	-0.390^{NS}	-0.295 ^{NS}	0.014^{NS}	0.083^{NS}
b	0.182^{NS}	0.569^{NS}	0.956***	-0.856***	-0.926***
a+b	0.351^{NS}	0.490^{NS}	0.930***	-0.906***	-0.960***
ME	0.071^{NS}	0.604^{NS}	0.971***	-0.864***	-0.907***
OMD	0.048^{NS}	0.610^{NS}	0.961***	-0.863***	-0.888***

c = Gas production rate (%); a = Gas production (mL) from quickly soluble fraction; b = Gas production (mL) from the insoluble fraction; a+b = Potential gas production (mL); OMD: Organic Matter Digestibility; ME: Metabolic Energy; DM: Dry Matter; CP: Crude Protein; ADF: Acid Detergent Fiber; NDF: Neutral Detergent Fiber; NS: Non-significant; ***p<0.001; NS: Non-significant (p>0.05)

It is well known that gas production is associated with volatile fatty acid production following fermentation of substrate so the more fermentation of a substrate the grater the gas production (Blummel and Orskov, 1993). Vegetative stage of growth all part of plants are highly digestible, but during stem elongation and flowering there is more rapid decline in digestibility of stem than of leaf (Terry and Tilley, 1964). The decrease in digestibility is due to increase in concentration of cell wall contents (Wilson *et al.*, 1991) and lignin content as plant matures (Morrison, 1980) and decrease in leaf/stem ratio (Hides *et al.*, 1983). The reason why gas production and most of the estimated parameters such as a, b, a+b, OMD and ME decreased with increasing maturity is the increase in cell wall content, lignin content of *Medicago polymorpha* hay with increased maturity (Table 1). Another possible cause of decrease in the as production could be the decrease in CP content of *Medicago polymorpha* hay with increasing maturity. As can be clearly seen from Table 3, the most of the gas production kinetics, ME and OMD were negatively correlated with ADF and NDF content of *Medicago polymorpha* hay whereas the same parameters were positively correlated with CP content of *Medicago polymorpha* hay. These results are in consistent with finding of Kamalak *et al.* (2005a, b).

CONCLUSIONS

The results obtained in this experiment clearly indicated that maturity (harvesting time) resulted in significant differences in nutritive value of *Medicago polymorpha* hay in terms of chemical composition and microbial fermentability. The nutritional quality deteriorates when harvesting stage is delayed. Therefore, it seems to be desirable to harvest or graze *Medicago polymorpha* hay at vegetative stage to obtain a high quality forage with a high level of CP, ME and low level of cell wall contents.

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