



## Botanical traits, protein and carbohydrate fractions, ruminal degradability and energy contents of alfalfa hay harvested at three stages of maturity and in the afternoon and morning

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### ABSTRACT

Little information is available about nutrient profiles and availability of alfalfa harvested at different stages of maturity and harvested at different times during the day in relation to diet formulation for dairy cows. The objective of this study was to investigate the effect of stage of maturity and cutting time of alfalfa hay on botanical traits, nutrient profiles and *in situ* degradability of protein and carbohydrates and calculated energy content. Alfalfa was cut at early bud (June 15/16), late bud (June 26/27) and early flower stage (July 18/19) both in the afternoon (06:00 pm) and the next morning (06:00 am). With advancing maturity of alfalfa, leaf content, leaf:stem ratio, calculated energy values, crude protein (CP), *in situ* digestibility's (especially at 12 and 36 h of incubation) and nitrogen (N) to energy [organic matter (OM), carbohydrates (CHO)] ratios decreased ( $P<0.05$ ). While, neutral detergent fiber, acid detergent fiber, fiber associated CP (NDICP) and total CHO increased ( $P<0.05$ ) with advancing maturity of alfalfa. Protein and CHO fractions (defined according to Cornell net carbohydrate and protein system) associated with different degradation characteristics stayed consisted with advancing maturity, except for intermediate degradable protein (PB2), which decreased at the early flower stage compared with early and late bud stages ( $P=0.03$ ). Alfalfa harvested in the afternoon tended to have a higher leaf portion and leaf:stem ratio ( $P=0.06$ ) and contained 13 g/kg CHO more soluble carbohydrates (TESC, *i.e.* CA;  $P<0.01$ ), 27 g/kg CP more PB2 ( $P=0.02$ ), and 0.33, 0.33, 0.27 MJ/kg DM more ( $P<0.05$ ) net energy for maintenance, gain and lactation production, respectively, and had an improved

**Abbreviations:** ADF, acid detergent fiber; ADICP, acid detergent insoluble crude protein; aNDF, neutral detergent fiber; CA, soluble carbohydrate; CB1, rapidly degradable carbohydrate; CB2, intermediate degradable carbohydrate; CB3, slowly degradable carbohydrate; CC, undegradable carbohydrate; CHO, total carbohydrates; CNCPS, Cornell net carbohydrate and protein system; CP, crude protein; CT, cutting time; DM, dry matter; EE, ether extract; NDICP, neutral detergent insoluble crude protein; NE<sub>g</sub>, net energy for gain; NE<sub>lp</sub>, net energy lactation at production level of intake; NE<sub>m</sub>, net energy for maintenance; NFC, none fiber carbohydrates; NRC, national research council; OM, organic matter; PA, solublizable protein; PB1, rapidly degradable protein; PB2, intermediate degradable protein; PB3, slowly degradable protein; PC, undegradable protein; SM, stage of maturity; TESC, total ethanol soluble carbohydrate.

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rapidly degradable N to CHO ratio (PA:CA; 66 vs. 76 g/kg;  $P=0.02$ ) compared with alfalfa harvested in the morning. Cutting time had no impact on *in situ* degradability of alfalfa hay. In conclusion, nutrient availability of alfalfa hay, grown under semi arid climate condition, was not only influenced by stage of maturity but also by cutting time. In general, alfalfa harvested at early and late bud but in the afternoon had the highest nutrient levels for dairy production.

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## 1. Introduction

Cultivated alfalfa (*Medicago sativa* L.) is one of the major forage crops in the world (Hanson et al., 1988) and the most important forage crop for dairy rations in Iran (Kowsar et al., 2008). Alfalfa contains high nutrients levels, high digestibility, and unique proportion of structural to non-structural components (Yu et al., 2003). Botanical traits, nutritive value and crude protein (CP) and carbohydrate (CHO) fractions of alfalfa are influenced by cultivar, stage of maturity (SM) (Elizalde et al., 1999; Yu et al., 2003; Coblenz et al., 2008), climate condition (Weir et al., 1960; Lamb et al., 2003) and cutting time (CT) due to accumulation of non-structural CHO during the day (Burns et al., 2007; Brito et al., 2008, 2009).

Various CP and CHO fractions present in feed differ in rate and extent of ruminal degradation. These fractions influence the amount of CP and CHO degraded in the rumen and escaping to the lower digestive tract (Lanzas et al., 2007a,b; Jonker et al., 2011). Furthermore, the knowledge of these CP and CHO fractions and degradation is used in modern diet formulation programs such as Cornell net carbohydrate and protein system (CNCPS; Lanzas et al., 2007a,b) and National Research Council (NRC, 2001) to formulate ruminant diets. Therefore, information about CP and CHO fractions and degradability (NRC, 2001; Lanzas et al., 2007a,b) and predicted energy values (NRC, 2001) should be taken in to consideration when formulating diets for ruminants. However, information about the effect of alfalfa CT at different stages of maturity on these nutritional properties is lacking. Our objectives were to investigate botanical traits, protein and carbohydrate fractions, *in situ* ruminal degradability and energy content of alfalfa harvested at three stages of maturity in the afternoon and next morning.

## 2. Materials and methods

### 2.1. Alfalfa plots management

A second year alfalfa field (20 m × 24 m) seeded with cv. Ranger at the Research Farm of Ferdowsi University of Mashhad (Mashhad, Iran; 36° 17' 52.8" N, 59° 36' 20.52" E) was used in this study. The whole field was harvested before the experiment at April 6, 2010 and irrigated every 10 days during experiment. The first cut at May 11, 26 and 30 for early bud, late bud and early flower, respectively was not used for this study.

Six plots (4 m × 4 m each) within 5 replicate blocks were randomly assigned to 6 treatments in a factorial arrangement (3 SM × 2 CT). The three SM were early bud, late bud and early flower and two CT were at 06:00 pm and the next morning at 06:00 am. The SM was determined according to Kalu and Fick (1981). Briefly, a quadrat (250 cm<sup>2</sup>) was randomly thrown in each plot (one time) and all stems above 3 cm stubble height inside the quadrat (ca. 70–80 stems) were used to calculate the mean SM for each plot. In total, there were 10 plots for each SM from which half was cut at 06:00 pm and the other half at 06:00 am, when alfalfa reached the appropriate SM (Table 1). At each harvest, an area of 3 m × 3 m was manually clipped using a small scythe at ca. 5 cm above the soil surface.

Immediately after cutting, twenty stems were randomly selected from each plot to separate leaf and stems by hand. Alfalfa leaf, stem, and whole plant dry matter (DM) content were determined by oven drying for 48 h at 60 °C. Remaining fresh alfalfa harvested from each plot was air dried in the shade (ca. 10–15 days). After air drying, alfalfa hay samples were chopped using a hay chopper with 20 mm screen (Agri-Equip, Nasr Co., Isfahan, Iran). The hay from the first, second and third blocks were pooled to one sample and hay from the fourth and fifth blocks were pooled to another sample to generate sufficient material for chemical analysis and *in situ* degradability measurements.

**Table 1**

Date and climate condition in Mashhad, Iran during alfalfa cutting in 2010.

Date of cutting	Min <sup>a</sup>	Max	GDD	TTC	Sunrise	Sunset
Early bud, 06:00 pm (June 15)	18	35	22	28	05:14	19:51
Early bud, 06:00 am (June 16)	18	35	22	19	05:14	19:51
Late bud, 06:00 pm (June 26)	19	36	23	27	05:16	19:54
Late bud, 06:00 am (June 27)	19	37	23	20	05:17	19:54
Early flower, 06:00 pm (July 18)	24	36	25	27	05:28	19:48
Early flower, 06:00 am (July 19)	23	36	24	24	05:29	19:48

<sup>a</sup> Min: minimum temperature (°C); Max: maximum temperature (°C); GDD: growing degree day was calculated daily by subtracting 5 °C from the average of the maximum and minimum temperatures for that day (Coblenz et al., 2008); TTC: temperature at time of cutting; the time of sunset and sunrise at that day; there was no rainfall at cutting dates; data for climate condition were collected from weather station located close to the experimental field (Mashhad Meteorological Network Station, Mashhad, Iran).

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