

Heat production, eating behavior and milk yield of lactating cows fed two rations differing in roughage content and digestibility under heat load conditions

G. Adin^b, R. Solomon^b, E. Shoshani^b, I. Flamenbaum^b, M. Nikbachat^a, E. Yosef^a,
A. Zenou^a, I. Halachmi^a, A. Shamay^a, A. Brosh^a, S.J. Mabjeesh^c, J. Miron^{a,*}

^a Agricultural Research Organization, P.O. Box 6, Bet-Dagan, 50250, Israel

^b Extension Service, Ministry of Agriculture, Israel

^c Faculty of Agriculture, The Hebrew University of Jerusalem, Israel

Received 19 September 2007; received in revised form 20 March 2008; accepted 24 March 2008

Abstract

The objective of this study was to measure the effect of feeding two total mixed rations (TMRs), differing in their roughage content and *in vitro* dry matter digestibility, on the respiratory rate, body temperature, eating behavior and energy balance of lactating cows. The partitioning of metabolizable energy intake (MEI) between heat production (HP) and retained energy (RE) of cows held under heat load conditions was measured. Forty-two lactating cows were divided into two similar sub-groups, each of 21 animals, and were fed either a control (CON) ration containing 18% roughage neutral detergent fiber (NDF) or an experimental (EXP) TMR that contained 12% roughage NDF and used soy hulls as partial wheat silage replacer. The *in vitro* DM digestibility of the CON and EXP TMRs was 75.3 and 78.6%, respectively ($P < 0.05$). The EXP diet reduced rectal temperature and respiratory rate of the cows while increasing their number of meals per day by 32.7% as compared with the CON fed cows, and these meals were shorter in duration and were eaten faster. The EXP diet increased total DM intake from 19.6 to 21.5 kg/cow/d, milk yield from 32.3 to 34.6 kg, and yield of energy corrected milk from 30.9 to 32.2 kg, as compared with the CON group. Cows fed the EXP TMR had increased RE in milk and body tissue, as compared with the CON group, but the diets had no effect on the measured HP that was maintained similar (121 vs. 127 MJ/cow/d) in the two groups. The measured MEI (MEI=RE+HP) and the efficiency of MEI utilization for RE production, were similar in the two dietary groups.

© 2008 Elsevier B.V. All rights reserved.

Keywords: Cow's eating behavior; Heat production; Energy retention; Heat load; Soy hulls; Roughage replacement; Milk yield

1. Introduction

It is well accepted in the literature that the main factors that might affect the partitioning of energy in lactating cow are: energy intake, environmental conditions and individual variability between cows with respect to the efficiency of energy utilization for maintenance and

* Corresponding author. Department of Dairy and Genetic Sciences, ARO, Israel. Tel.: +972 3 9683370; fax: +972 3 9604023.

E-mail address: jmiron@volcani.agri.gov.il (J. Miron).

production (Berman, 2005; Brosh, 2007). Heat load might affect both intake, heat production (HP), milk yield and body tissue gain (Brosh, 2007).

In many areas of the world including Israel the hot season has become longer and consequently a moderate heat stress condition often occurs in dairy barns, as reflected by their temperature humidity index (THI) of 70–80 (Flamenbaum et al., 1986; West, 2003).

Under heat stress conditions, lactating cows tend to reduce their dry matter intake (DMI) and milk production (West et al., 1999). Voluntary DMI can decrease by as much as 6% when ambient temperatures rise above 30 °C (Eastridge et al., 1998). Accordingly, Miron et al. (2003) suggested that the NRC (2001) recommendation of 18–19% minimal content in diets of neutral detergent fiber (NDF) from roughage might be too high. They suggested to reduce it to 12–15% of total mixed rations (TMRs) fed to high producing lactating cows that are exposed to hot weather conditions. Partial replacement of forage by starchy grain (Cummins, 1992; West et al., 1999) and addition of fat or protected fat to the diet (Chan et al., 1997; Huber et al., 1994; Serbester et al., 2005) have been tried as approaches to increase the energy consumption of cows under heat load conditions. These strategies have proven ineffective at increasing DMI, milk and milk fat production.

With this respect, we have demonstrated that partial replacement of dietary corn silage with soy hulls, that contain a high level of readily digestible NDF (Miron et al., 2001), increased the milk production of lactating cows by 10% under hot weather conditions (Miron et al., 2003; Halachmi et al., 2004). However, DMI was not reported to increase in that study and the changes in physiology and energy partitioning in the cows were not measured.

At present, in Israel wheat silage is the major roughage used in TMR fed to lactating cows, and only minor changes are made to the composition of TMR according to the season. Unfortunately, there is limited information in the literature concerning the effect of a feeding regime, which uses highly digestible soy hulls for replacing wheat silage, on the voluntary eating behavior, physiological response, energy status and performance of lactating cows experiencing heat load conditions.

A major part of the metabolizable energy intake (MEI) by cows is utilized as heat production (HP) and another part of the energy is retained in milk and body tissue (RE). Recently, the research group of Brosh and Aharoni, have developed a new method that can directly measure the HP and RE in milking cows held under commercial dairy barn conditions (Aharoni et al., 2005; Brosh, 2007). The HP is quantitatively assayed by measuring the volume of O₂ consumption (VO₂). In

mammals, most of the measured VO₂ is transferred to the tissues through the heart; therefore, multiplying of daily measured heart rate (HR) by short measurement of VO₂ per heart beat was used for direct measurement of cows' HP (Brosh, 2007). This new method provides a powerful tool to understand the effect of changing the *in vitro* digestibility and roughage content of the TMR, on the partitioning of the metabolizable energy intake (MEI) between HP and RE.

The objective of this study was to measure the effect of partial wheat silage replacement by soy hulls, on the heat production, retained energy, eating behavior, DMI, physiology, milk yield and composition, and body condition scores (BCS) of lactating cows under heat load conditions.

2. Materials and methods

2.1. Cows, diets, environmental conditions and sampling procedures

Forty-two lactating multiparous Holstein cows were fed for a pre-experimental period of 2 weeks the control (CON) and experimental (EXP) TMRs are described below (Table 1). During this pre-experimental period the cows were gradually adapted within seven days from 7 times per day of evaporative cooling (in the milking yard) to non-cooling conditions. The cows were divided into two feeding groups similar in average (means±SE) age (2.9±0.1 lactations), days in milk (167±6.3), body weight (579±6.2 kg), daily milk yield (43.0±0.75 kg) and dry matter intake (DMI, 23±1 kg). The two TMRs were fed to the cows for two adaptation weeks plus additional six weeks of the experiment. The control TMR, fed to the CON group, contained 18% roughage NDF, whereas the experimental TMR, fed to the EXP group, contained just 12% roughage NDF, in which one third of the dietary wheat silage was replaced with soy hulls (Table 1). All the cows were housed at the Agricultural Research Organization (ARO) dairy farm, in one shaded corral with free access to water and without any cooling under the hot weather conditions.

The two TMRs were offered once daily at 1030 with *ad libitum* intake, allowing for 5–10%orts, and the cows were milked 3 times daily at 0500, 1200 and 2000 h. Cows were fed individually via a computerized monitoring system, designed to identify electronically individual cows and to control and record automatically each entry into the feed stalls. The 42 feeders were each mounted on weighing balances that enabled the voluntary feed intake during every meal for each individual cow to be monitored. Scientists from the Volcani Center developed this system, as described in Halachmi et al. (1998). A meal is defined as an identified cow visiting a feeder, where the visit lasts at least 2 min and at least 200 g of wet TMR is consumed. Once the electronic antenna located near the fodder registered a cow, feed intake and eating duration were measured for that meal and the number of meals per day recorded.

Table 1
Ingredients and chemical composition of the two TMRs

	% of DM		SEM
	EXP ¹	CON ¹	
<i>Ingredients</i>			
Wheat silage	18.7	29.7	
Soybean hulls	11.8	0.0	
Soybean meal (solvent extract)	8.4	9.2	
Vetch hay	4.3	4.3	
Ground corn grain	18.3	18.3	
Ground barley grain	17.4	17.4	
Whole cottonseeds	4.5	4.5	
Corn gluten feed	7.9	7.9	
Distillers dry grains	5.3	5.3	
NaHCO ₃	0.70	0.70	
NaCl	0.4	0.4	
CaCO ₃	1.3	1.3	
Protected fat	0.90	0.90	
Trace mineral+ vitamin mixture ²	0.10	0.10	
Water (liter/cow/d)	3.36	0.0	
<i>Chemical composition</i>			
DM (%)	64.6	63.2	0.5
OM (% of DM)	92.1	91.7	0.2
CP (% of DM)	16.0	16.0	0.1
Ether extract (% of DM))	4.6	4.6	0.1
NDF (% of DM)	36.0 ^a	30.8 ^b	0.02
Roughage NDF (% of DM)	12.0 ^b	18.0 ^a	0.02
<i>In vitro</i> DM digestibility (%)	78.6 ^a	75.3 ^b	0.02
<i>In vitro</i> NDF digestibility (%)	68.8 ^a	59.2 ^b	0.02
Calculated NE _L (MJ/kg DM)	7.53	7.32	–
Volume of the TMR (L/kg fresh TMR)	3.05 ^b	3.26 ^a	0.042

^{a,b}Means in the same row following by different superscripts differ at $P < 0.05$.

¹ EXP = experimental TMR containing 12% NDF of roughage origin; CON = control TMR containing 18% NDF of forage origin. SEM = standard error of the mean.

² The trace minerals + vitamins mix contained (g/kg DM): Zn, 24; Fe, 24; Cu, 12.8; Mn, 24; I, 1.44; Co, 0.32; Se, 0.32; Vit. A, 16,000,000 IU; Vit. D₃, 3,200,000 IU; Vitamin E, 48,000 IU.

Rations were sampled daily and pooled on a weekly basis to produce 6 samples for each dietary treatment. DMI was determined by oven drying (at 105 °C for 24 h) a portion of the weekly TMR samples. The rest of the weekly TMR samples were oven dried (at 60 °C for 48 h) and used for *in vitro* digestibility evaluation and chemical analyses.

The milk yield of each cow was recorded daily by automatic meters (Afimilk SAE Israel). Milk samples were collected during three sequential milkings on a weekly basis throughout the study. Each set of milk samples for each cow was stored at 4 °C in the presence of a preservative Bronopol (2-Bromo-2-nitropropane-1,3-Diol) tablet, until analyzed for content of fat, true protein, lactose and urea by infrared analysis (Israeli Cattle Breeders Association Lab., Caesaria, Israel, using Milkoscan 4000, Foss Electric, Hillerod, Denmark). Body weight (BW) data was recorded by an automated walk-over weigher (Afimilk Israel) each time the cow entered the milking parlor. The animal performance study was carried out according to the guidelines and under the supervision of the ARO Animal Care Committee. A trained veterinarian, the

same one throughout the study, measured body condition scores (BCS) on a weekly basis in a scale of 1 to 5 according to NRC (2001).

2.2. Chemical analyses and *in vitro* digestibility measurements

Replicate samples of the weekly composites of each TMR and of theorts of the individual cows were assayed in triplicate for DM content (drying at 105 °C for 24 h) and residual ash (4 h at 600 °C). Dry TMR samples (at 60 °C for 48 h) were ground through a 1 mm sieve, and analyzed for crude protein (CP) content according to the Kjeldahl method (AOAC, 2001) and for the content of NDF (without sodium sulfite and with heat stable amylase, Van Soest et al., 1991). The Ankom apparatus (Ankom 220, Fairport, NY USA) was used for extracting and filtering the NDF. Net energy of lactation (NE_L) content of each TMR was calculated based on the NE_L content of the individual feeds (NRC, 2001).

In vitro digestibility of DM and NDF in the weekly composites of TMR was analyzed in triplicate for each sample. The

procedure involved incubating 0.5 g dry plant material with rumen fluid for 48 h and then with 0.1 N HCl and 0.2% pepsin for another 48 h, according to the two-stage fermentation technique of Tilley and Terry (1963). Rumen fluid was obtained before morning feeding via rumen fistula from 3 dry cows fed a mixture of the CON and EXP diets. Residual NDF in the *in vitro* tubes was determined according to Van Soest et al. (1991).

2.3. Physiological measurements

Rectal temperature was measured with a digital electronic thermometer (Toshiba) and respiration rate (RR) was visually measured in each cow during three days per week of the experiment at 0700 and 1700 h. The heart rate (HR) and heat production (HP) of the individual cows were measured twice, at three week intervals, during the experimental period as described previously by Aharoni et al. (2005) and Brosh (2007).

In order to measure HP we recorded the HR continuously for 4 d and measured the volume of oxygen consumption during 10–15 min by using a face-mask. Heart rate was measured with a Polar instrument (Polar Electro Oy, Kempele, Finland), a model T51H HR transmitter, and a watch model S610 data logger and receiver. The devices were attached to the thorax behind the forelegs by means of a specifically designed elastic belt (Pegasus, Eli-ad, Israel). The data logger was programmed to record HR at 1-min intervals. Volume of oxygen consumption (VO_2) was measured in each cow with a face-mask open-circuit respiratory system simultaneously with the HR for 10 to 15 min for each cow, after adaptation of 20 min to the mask, from 0900 to 1100 after 4 d HR measurement. The HP ($\text{kJ kgBW}^{-0.75} \text{d}^{-1}$) was calculated by multiplying the means of HR measured throughout 3–4 days by the VO_2 per heartbeat O_2 pulse (O_2P), and by 20.47 kJ per liter VO_2 (Nicol and Young, 1990). For the non-cooled cows O_2P during the hot h was corrected for the effect of temperature humidity index (THI) on O_2P , as suggested by

Aharoni et al. (2003). The HP and the energy retained in the milk and BW gain were measured for each cow in each treatment. The equation used to calculate HP was (Brosh, 2007): Daily HP (MJ/cow/d) = specific HP * $\text{kgBW}^{-0.75}$
Where: Specific HP ($\text{kJ} \cdot \text{kgBW}^{-0.75} \cdot \text{d}^{-1}$) = $HR * O_2P * 20.47 / 1000 * 60 * 24$

HR = Heart rate (beats/min)
 $O_2P = L O_2 / (\text{beat} * \text{kgBW}^{0.75})$
 $20.47 = \text{kJ/L } O_2 \text{ consumption}$

Environmental climate data in the barn including ambient temperature, humidity, wind speed and directions were monitored continuously during the experimental period using a weather monitoring station (RWMS-8, Rotem Computerized Controllers — Israel).

The temperature humidity index (THI) was calculated (Aharoni et al., 2003) with the equation:

$$THI = td - (0.55 - 0.55RH) * (td - 58)$$

Where: td is the dry bulb temperature ($^{\circ}\text{C} * 9/5 + 32$). RH is the relative humidity expressed as a decimal.

The average diurnal pattern of ambient air temperature, relative humidity and temperature humidity index (THI) in the barn is presented in Fig. 1. Data given in this figure summarize the pattern of an average day of the six weeks experimental period.

2.4. Calculations and statistical methods

Yield of energy corrected milk (ECM) was calculated according to the NRC (2001) equation: $\text{ECM (kg/d)} = \text{Milk yield (kg/d)} * \{ (0.3887 * \% \text{ milk fat}) + (0.2356 * \% \text{ milk protein-urea}) + (0.1653 * \% \text{ milk lactose}) \} / 3.1338 \text{ MJ/kg}$. The yield of 4% FCM (fat corrected milk) was calculated

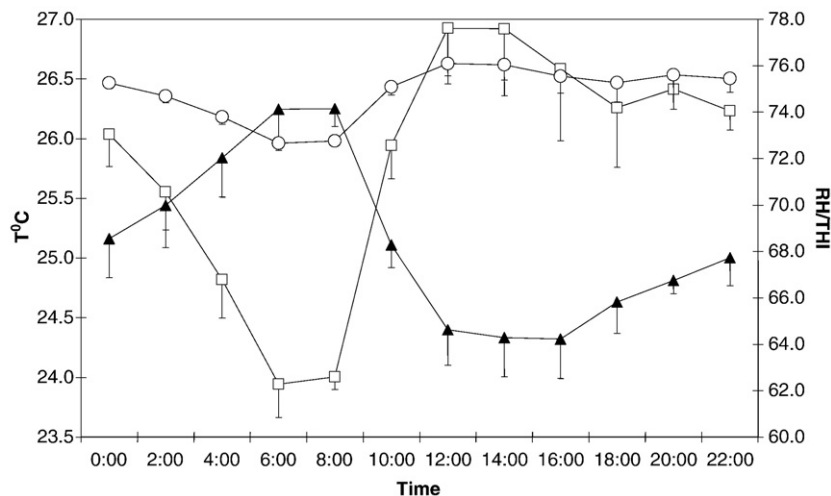


Fig. 1. Average diurnal pattern of ambient air temperature ($T^{\circ}\text{C} \pm \text{se}$) \square ; relative humidity ($\text{RH} \pm \text{se}$) \blacktriangle ; and temperature humidity index ($\text{THI} \pm \text{se}$) \circ ; in the barn.

Table 2
Physiological status of cows fed the two TMRs under heat load conditions

Variable	Treatments ¹		
	EXP	CON	SEM
<i>Respiratory rate/min</i>			
Morning	69.1 ^b	78.4 ^a	1.88
Afternoon	84.5 ^b	93.6 ^a	2.15
<i>Rectal temperature °C</i>			
Morning	38.5 ^b	38.8 ^a	0.05
Afternoon	39.1 ^b	39.4 ^a	0.06
Heart rate (beat/min)	76.6	76.4	1.70
Pedometer (steps/h)	91.3	92.0	4.18

SEM = standard error of the mean.

^{a,b}Means in the same row following by different superscripts differ at $P < 0.05$.

¹ EXP = experiment TMR containing 12% roughage NDF; CON = control TMR containing 18% roughage NDF.

using the Equation: 4% FCM (kg) = $0.4 \times \text{milk (kg)} + 15.0 \times \text{milk fat (kg)}$, NRC (2001).

Comparison between the two feeding groups with respect to physiology, intake behavior parameters, performance and energy status parameters was carried out according to the repeated measurement Proc-mixed model of SAS (SAS, 1996), and presented in Tables 2–5 as the daily mean for the 6 week experimental period. *F*-test was used to differentiate between means (SAS, 1996).

Differences between the two TMRs (6 weekly composites of each TMR) with respect to composition and *in vitro* digestibility were tested for significance using an analysis of variance (SAS, 1996).

3. Results and discussion

The distribution in the barn of ambient air temperature, relative humidity and THI data over an average day and

Table 3
Eating behavior, voluntary intake, rectal temperature, and rumen pH of cows fed the two TMRs under heat load conditions

Variable/feeding treatment	Treatments ¹		
	EXP	CON	SEM
Meals per day	8.88 ^a	6.69 ^b	0.16
Meal intake (DM, kg/meal)	2.42 ^b	2.93 ^a	0.08
Meal duration (min/meal)	15.7 ^b	20.5 ^a	0.30
Daily eating duration (min)	139	137	2.65
Rate of meal intake (DM, g/min)	154 ^a	143 ^b	2.85
Daily DMI (kg/day)	21.5 ^a	19.6 ^b	0.28
Daily NDF intake (kg/day)	7.05 ^a	6.04 ^b	0.09
Rumen pH	6.56	6.75	0.088

^{a,b}Means in the same row following by different superscripts differ at $P < 0.05$.

¹ EXP = experiment TMR containing 12% roughage NDF; CON = control TMR containing 18% roughage NDF.

Table 4
Performance of cows fed the two TMRs under heat load conditions

Variable	Treatments ¹		
	EXP	CON	SEM
Milk yield, kg/d	34.6 ^a	32.3 ^b	0.15
Milk fat, %	3.53 ^b	3.76 ^a	0.010
Milk protein, %	3.17 ^a	3.14 ^b	0.004
Milk lactose, %	4.85	4.84	0.005
Milk urea-N, %	0.030	0.033	0.001
Milk fat, yield, kg/d	1.22	1.21	0.006
Milk protein, yield, kg/d	1.10 ^a	1.01 ^b	0.005
4% Fat corrected milk, kg/d	32.1	31.5	0.17
Energy corrected milk, kg/d	32.2 ^a	30.9 ^b	0.16
Production efficiency (kg milk/kg DMI)	1.61	1.65	0.018
Body condition score change/d (units 1–5)	0.0048	0.0039	0.0004

SEM = standard error of the mean.

^{a,b}Means in the same row following by different superscripts differ at $P < 0.05$.

¹ EXP = experiment TMR containing 12% roughage NDF; CON = control TMR containing 18% roughage NDF.

night of the experimental period is illustrated in Fig. 1. This figure is important due to the environmental effects on eating behavior and physiology changes during the day and night.

Environmental conditions in the barn were similar for the two dietary groups and stable throughout the 6 weeks of the experiment. Ambient temperature was high (26–27 °C) during most of the day hours and early evening, with daily averages of 25.7 °C.

Table 5
Energy partitioning in cows fed the two TMRs under heat load conditions

Variable	Treatments ¹		
	EXP	CON	SEM
Retained energy ² in Milk (MJ/Cow/d)	100.9 ^a	96.8 ^b	0.50
Retained energy ² in BCS change (MJ/Cow/d)	4.96 ^a	3.76 ^b	0.10
Total retained energy (MJ/Cow/d)	105.9 ^a	100.6 ^b	0.14
Heat production ³ (MJ/Cow/d)	126.9	121.2	3.35
Measured MEI ⁴ (MJ/Cow/d)	232.8	221.8	3.85
Energy efficiency for production (total retained energy/measured MEI)	0.455	0.454	0.008

SEM = standard error of the mean.

¹EXP = experiment TMR containing 12% roughage NDF; CON = control TMR containing 18% roughage NDF.

²RE = Retained energy in milk and/or BCS increment according to Aharoni et al. (2006).

³Total heat production (HP) = $\text{HR} \times \text{O}_2\text{P} \times 20.47 / 1000 \times 60 \times 24$.

⁴Measured metabolizable energy intake (MEI) = total retained energy + heat production.

^{a,b}Means in the same row following by different superscripts differ at $P < 0.05$.

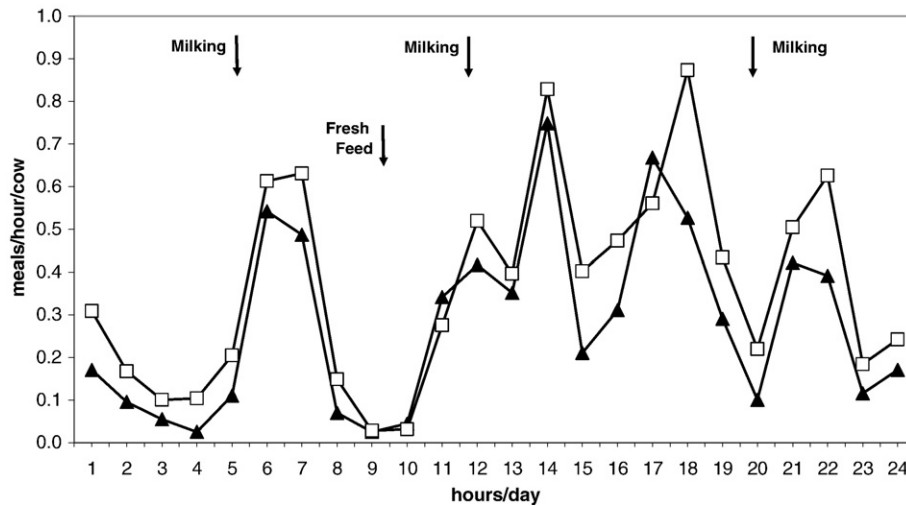


Fig. 2. Pattern of meal distribution over 24 h of an average day for cows fed the EXP (□) and CON (▲) TMRs. The arrows indicate time of dispensing a fresh TMR or cows' return from milking.

Relative humidity (RH) was highest in the early morning hours and its daily average was 66.6%. Thus, THI was confined within a narrow range (74–76) during the experimental period. Since the critical THI of high producing cows is 68 (Flamenbaum et al., 1986; Berman, 2005), the cows in this study were indeed under heat load conditions.

The *in vitro* digestibility of the EXP TMR was significantly higher than the CON TMR (78.6 vs. 75.3, Table 1) and reflected the large difference in NDF digestibility of the two TMRs (68.8 vs. 59.2%). This difference in the digestibility and volume of the TMRs affected the eating behavior, respiratory rate (RR) and

rectal temperature of the cows (see Tables 2 and 3) and their voluntary DMI (Table 3). Cows fed the EXP TMR responded by a decrease of their RR by 12% in the morning and by 10% in the afternoon hours, as compared with the CON group. A positive significant effect of the feeding regime on reducing rectal temperature of the non-cooled cows by 0.3 °C was manifested both in the morning and afternoon measurements (Table 2). However, the effect of the feeding regime on HR and daily steps (activity) was non-significant.

Feeding behavior data is presented in Table 3 and Figs. 2 and 3, and the milk production data is shown in Table 4. Cows fed the soy hulls-rich (EXP) TMR

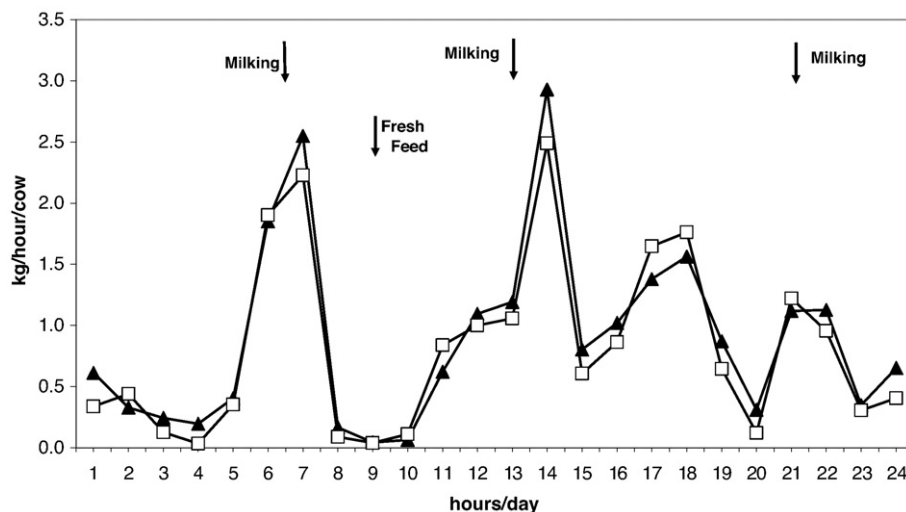


Fig. 3. Diurnal pattern of feed intake (kg DM/h) distribution over 24 h of an average day, in cows fed the EXP (□) and CON (▲) TMRs. The arrows indicate time of dispensing a fresh TMR or cows' return from milking.

consumed 32.7% more meals per day compared with the CON fed cows, these meals were shorter in duration and were eaten faster ($P < 0.05$, Table 3). Two explanations are proposed here to clarify such differences. One possibility is that the relatively higher DM and NDF digestibility of the EXP TMR as well as its lower volume relative to the CON TMR (Table 1) slows the mechanism regulating the short term full-stomach feeling, which is responsible for inhibiting eating rate (Allen, 1996; Van Soest, 1994). Additionally, it is possible that the EXP TMR is more palatable to the cows and therefore is consumed more willingly and rapidly than the CON TMR. These data and explanations concur with the results of a previous study (Halachmi et al., 2004; Miron et al., 2003), which reported similar eating behavior for lactating cows fed a TMR containing digestible soy hulls as corn silage replacement.

It should be noted, that most of the voluntary meals, whatever the dietary treatment, were consumed by the cows during late afternoon and evening hours (1700 to 2300) and in the early morning hours (0400 to 0800) (Fig. 2), when the environmental temperatures were more moderate (Fig. 1). Only a small portion of the total daily intake occurred in the cows during night hours (between 2300 and 0400) or during the hot hours of the day (between 0900 and 1600; Fig. 1), where it was generally in response to external stimulating events such as the return from milking or at the dispensing time of the basic TMR (Fig. 2).

The greater number of meals per day compensated for the smaller meal size and shorter meal duration of EXP TMR fed cows. Therefore, total daily DMI of cows fed EXP TMR relative to cows fed CON TMR was higher by 9.7% (21.5 vs. 19.6 kg/d/cow, $P < 0.05$, Table 3). This higher DMI of the EXP cows might be attributable partially to their better heat balance status reflected in lower rectal temperature and RR (Table 2), and to improved digestibility and increased rate of passage for the soy hull containing diet. Similar daily DMI differences in mid-lactating cows were reported in previous studies employing soy hulls as corn silage replacement under hot weather conditions (Miron et al., 2003; Halachmi et al., 2004). The higher DMI observed for cows fed the EXP TMR and the higher DM and NDF *in vitro* digestibility of this TMR as compared with the CON diet (Table 1) were reflected in concomitant increase of 7.1% in milk production and 4.2% in ECM yield of the cows ($P < 0.05$, Table 4). Also, the EXP TMR stimulated milk protein production ($P < 0.05$, Table 4). It appears that the higher digestibility of the EXP TMR (Table 1) encourages milk and milk protein production more efficiently than milk fat production, as this latter parameter was almost un-affected by diet (Table 4). Nevertheless, the similar

rumen pH values observed for cows fed each dietary treatment (Table 3) suggests that despite its higher *in vitro* digestibility, the EXP TMR enables appropriate and sufficient activity of cellulolytic bacteria in the rumen as required to produce metabolites and precursors for high levels of milk fat production in the mammary gland. These data are in accord with results from earlier studies that used soy hulls as corn silage replacer in TMR for lactating cows (Halachmi et al., 2004; Miron et al., 2003). However, other studies that employed starchy grain (Cummins, 1992; West et al., 1999) or fat supplementation (Chan et al., 1997; Huber et al., 1994; Serbester et al., 2005) used to increase the energy content of TMR, failed to enrich DMI, milk and milk protein production up to the levels obtained in the present study.

Energy partitioning data of cows fed the two dietary treatments under heat load conditions is given in Table 5. The significant differences in digestibility, DMI and ECM yield between the two dietary treatments (Tables 1–4) were expressed in a 5.3% increase ($P < 0.05$, Table 5) in RE for milk + body tissue production of the cows fed the EXP TMR, as compared with the CON group. This increase in RE was not followed by any significant increase in the heat production (HP) of the cows, that was maintained at a constant level of 121–127 MJ/cow/d. Similar constant levels of total HP in the range of 123–135 MJ/cow/d were measured also in previous studies with lactating un-cooled Holstein cows, held under various feeding regimes, climate conditions, and over different stages of the lactation (Aharoni et al., 2005, 2006). It should be noted here, that in a parallel study, by feeding these two TMRs to cows that were held under heat load conditions, but received evaporative cooling for 6 h per day, we found similar heat production values (~130 MJ/cow/d), but higher milk and ECM values as compared with the present study (Miron et al., *in press*). The finding of this study and of the previous ones, that shower cooling and various feeding regimes, hardly affect the HP of mid-lactation Holstein cows, deserves further investigation. Total HP is actually the sum of HPp (HP for milk and body tissue production) plus HPm (HP for maintenance including pregnancy and embryo needs). The constant level of HPp+HPm = 121–130 MJ/cow/d, found in this study, in accord with previous data (Aharoni et al., 2005, 2006; Miron et al., *in press*), suggests that any increase in HPp of the cows is enabled by a concomitant decrease in their HPm. Thus, under severe heat load conditions without external cooling, HPm of lactating cows might increase (see Berman 2005) and therefore HPp should be concomitantly decreased to maintain the total HP level. Whereas under external evaporative cooling or feeding of more digestible TMR, the HPm might decrease as

demonstrated previously (Berman, 2005; Miron et al., 2008) allowing for concomitant increase in HPP. Based on this study and on the previous ones (Aharoni et al., 2005, 2006; Miron et al., 2008) we suggest that a HP level around 130 MJ/cow/d is the upper limiting range of heat disposal capability that a high producing mid-lactation cow (yielding 32–44 kg milk/d) can handle under Israeli heat load conditions, while maintaining her body rectal temperature at a level below 40 °C. Consequently the cows respond to external heat load conditions, by reducing level of production and HPP in order to maintain their internal HP level.

The possible mechanism involved in the control of voluntary DMI by the lactating cow under heat load vs. external cooling conditions is still unknown. Forbes (1986) suggested that temperature receptors occur in many parts of the cow body, including in the skin and anterior hypothalamus as well as in the rumen wall and the abdomen, with the afferent fibers in the splanchnic nerves, and this may be the way in which heating or cooling affects the short term control of voluntary DMI. Based on the present study findings we suggest that the cow's heat loss capability, limits her total heat production, which indirectly affects her voluntary intake and milk production level.

The constant level of total HP found in this study, suggests that since calculated $MEI = RE + HP$, there should be a linear correlation between RE and calculated MEI. Indeed, in this study the r^2 value of the correlation between MEI and RE was 0.93. This finding suggests that changes in the composition and/or digestibility of the TMR, that affect DMI and may increase MEI, affect directly the level of production expressed by RE and has a minor effect, if any, on the HP.

Milk production efficiency (kg milk/kg DMI, Table 4), and efficiency of energy utilization for milk and tissue production (total RE/measured MEI, Table 5) were hardly influenced by the dietary treatments, despite the higher *in vitro* digestibility of the EXP TMR (Table 1). This finding implies that the increased DM and NDF potential digestibility (*in vitro*) of EXP TMR is expressed mostly in increasing daily DMI, which in turn elevates milk and ECM production, and is not manifested by improved feed utilization efficiency along the digestive tract. This premise is supported by the high coefficient of correlation for the linear regression in this study between DMI and ECM yield ($r^2 = 0.94$). This high correlation supports the important influence of DMI on milk production, as described also in previous reports (Van Soest, 1994).

One possible explanation for the non-consistent direct effect of *in vitro* TMR digestibility on ECM yield (Table 4) is that the higher DM and NDF voluntary intake by cows fed the EXP TMR (Table 3), increased in turn the

passage rate of particles and NDF from the rumen, causing reduced rate and extent of NDF digestion along the gastrointestinal tract (Van Soest 1994; NRC, 2001). Similar findings have been documented in previous studies that compared various diets differing in their potential *in vitro* DM digestibility, where a great effect on voluntary DMI was observed, but with little influence on milk production efficiency (Solomon et al., 2005; Miron et al., 2004a,b).

The data of this study, based on direct measurements of total HP and total RE enables comparison between the predicted NEL value of the diets which was 7.53 and 7.32 MJ/kg DMI in the EXP and CON diets, respectively, (Table 1, based on NRC, 2001 data) and the direct measurements of RE in the two treatments (Table 5). This comparison shows under-estimation of 37 and 33% in the total RE measured in this study as compared with the predicted NEL values (NRC, 2001). This gap between estimated and measured NEL values probably originated from the higher HP values measured in this study (121–127 MJ/cow/d operated under hot summer conditions) as compared with lower HP estimations (~90 MJ/cow/d) used by (NRC, 2001). Additional explanation to this gap is the lower quality of Israeli forages as compared with NRC, which contribute to higher heat production in the digestive tract that might further increase HP level. This gap between predicted and measured NEL values suggests that the methodology used for the estimation of the nutritive value of forages in Israel [NEL estimation according to NRC (2001) Tables], should be re-evaluated and adapted to the quality of Israeli forages and to the actual HP values measured under Israeli climate conditions.

4. Conclusions

This study demonstrated that feeding with soy hulls as partial roughage replacer increased DMI by 9.7% and milk production by 7.1%, ($P < 0.05$). This improvement was due to the higher potential (*in vitro*) digestibility of the EXP diet and to the better physiology parameters of the EXP cows under heat load conditions. The feeding regime hardly affected the efficiency of energy utilization for ECM and BCS production, suggesting that the EXP treatment affected mostly intake level rather than the *in vivo* digestibility along the gastrointestinal tract. This was reflected by increase in total RE for milk and body tissue reserves by 4.5%, while the HP was maintained fairly constant at a level of 121–127 MJ/cow/d. Based on the present study findings, supported by previous studies, we can hypothesize that the cow's total heat production capability (upper level of about 130 MJ/cow/d) limits her voluntary intake, level of production and RE.

Acknowledgements

We wish to express our appreciation to the Volcani Center dairy farm team for their efforts in cooling and feedings the cows, and to Mr. Zwi Sarid and his team from the Yavne Feeding Center for preparing and delivering the two TMRs. This study was supported by a research grant # 362-071-06 of the Israeli Milk Council.

References

- Aharoni, Y., Brosh, A., Kourilov, P., Arieli, A., 2003. The variability of the ratio of oxygen consumption to heart rate in cattle and sheep at different hours of the day and under different heat load conditions. *Livest. Prod. Sci.* 79, 107–117.
- Aharoni, Y., Brosh, A., Harari, Y., 2005. Night feeding for high-yielding dairy cows in hot weather: effects on intake, milk yield and energy expenditures. *Livest. Prod. Sci.* 92, 207–219.
- Aharoni, Y., Brosh, A., Kafchuk, I., 2006. The efficiency of utilization of metabolizable energy for milk production: a comparison of Holstein with F1 Montbeliarde 3 Holstein cows. *Anim. Sci.* 82, 101–109.
- Allen, M.S., 1996. Physical constraints on voluntary intake of forages by ruminants. *J. Anim. Sci.* 74, 3063–3075.
- AOAC, Association of official analytical chemists, 2001. *Official Methods of Analysis*. AOAC publications, Washington, DC.
- Berman, A., 2005. Estimates of heat stress relief needs for Holstein dairy cows. *J. Anim. Sci.* 83, 1377–1384.
- Brosh, A., 2007. Heart rate measurement as an index of energy expenditure and energy balance in ruminants: a review. *J. Anim. Sci.* 85, 1213–1227.
- Chan, S.C., Huber, J.T., Chen, K.H., Simas, J.M., Wu, Z., 1997. Effects of ruminally inert fat and evaporative cooling on dairy cows in hot environmental temperatures. *J. Dairy Sci.* 80, 1172–1178.
- Cummins, K.A., 1992. Effect of dietary acid detergent fiber on responses to high environmental temperature. *J. Dairy Sci.* 75, 1465–1471.
- Eastridge, M.L., Bucholtz, H.F., Slater, A.L., Hall, C.S., 1998. Nutrient requirements for dairy cattle of the National Research Council vs some commonly used software. *J. Dairy Sci.* 81, 3049–3062.
- Flamenbaum, I., Wolfenson, D., Maman, M., Berman, A., 1986. Cooling dairy cattle by a combination of sprinkling and forced ventilation and its implementation in the shelter system. *J. Dairy Sci.* 69, 3140–3147.
- Forbes, J.M., 1986. *The Voluntary Food Intake of Farm Animals*. Butterworth & Co. (Publishers) Ltd, London, p. 47.
- Halachmi, I., Edan, Y., Maltz, E., Peiper, U.M., Brukental, I., Moalem, U., 1998. A real-time control system for individual dairy cow food intake. *Comput. Electron. Agric.* 20, 131–144.
- Halachmi, I., Maltz, E., Livshin, N., Antler, A., Ben-Ghedalia, D., Miron, J., 2004. Effects of replacing roughage with soy hulls on feeding behavior and milk production of dairy cows under hot weather conditions. *J. Dairy Sci.* 87, 2230–2238.
- Huber, J.T., Higginbotham, G., Gomez-Alarcon, R.A., Taylor, R.B., Chen, K.H., Chan, S.C., Wu, Z., 1994. Heat stress interactions with protein, supplemental fat, and fungal cultures. *J. Dairy Sci.* 77, 2080–2090.
- Miron, J., Yosef, E., Ben-Ghedalia, D., 2001. Composition and in vitro digestibility of monosaccharide constituents of selected byproduct feeds. *J. Agric. Food Chem.* 49 (5), 2322–2326.
- Miron, J., Yosef, E., Maltz, E., Halachmi, I., 2003. Soybean hulls as a replacement of forage neutral detergent fiber in total mixed rations of lactating cows. *Anim. Feed Sci. Technol.* 106, 21–28.
- Miron, J., Yosef, E., Nikbachat, M., Zenou, A., Maltz, E., Halachmi, I., Ben-Ghedalia, D., 2004a. Feeding behavior and performance of dairy cows fed pelleted nonroughage fiber byproducts. *J. Dairy Sci.* 87, 1372–1379.
- Miron, J., Nikbachat, M., Zenou, A., Ben-Ghedalia, D., Solomon, R., Shoshani, E., Halachmi, I., Livshin, N., Antler, A., Maltz, E., 2004b. Lactation performance and feeding behavior of dairy cows supplemented via automatic feeders with soy hulls or barley based pellets. *J. Dairy Sci.* 87, 3808–3815.
- Miron, J., Adin, G., Solomon, R., Nikbachat, M., Zenou, A., Shamay, A., Brosh, A., Mabweesh, S.Y., in press. Heat production and retained energy in lactating cows held under hot summer conditions with evaporative cooling and fed two rations differing in roughage content and in vitro digestibility. *Animal*.
- National Research Council, 2001. *Nutrient Requirements of Dairy Cattle*, 7th ed. National Academy Press, Washington, DC.
- Nicol, A.M., Young, B.A., 1990. Short-term thermal and metabolic responses of sheep to ruminal cooling: effects of level of cooling and physiological state. *Can. J. Anim. Sci.* 70, 833–843.
- SAS, 1996. *SAS/STAT Software Changes and Enhancements*. SAS institute, Cary, NC.
- Serbester, U., Görgülü, M., Kutlu, H.R., Yurtseven, S., Arieli, A., Kowalski, Z.M., 2005. The effects of sprinkler+fan, SH meal or dietary fat on milk yield and milk composition of dairy cows in mid lactation during summer. *J. Anim. Feed Sci.* 14 (4), 634–654.
- Solomon, R., Adin, G., Mabweesh, S.J., Nikbachat, M., Yosef, E., Ben-Ghedalia, D., Miron, J., 2005. Digestibility in lactating cows of diets containing whole Pima treated with sodium hydroxide versus Akala or Pima cottonseed. *J. Dairy Sci.* 88, 1745–1751.
- Tilley, J.M., Terry, R.M., 1963. A two stage technique for the in vitro digestion of forage crops. *J. Br. Grassl. Soc.* 18, 104–111.
- Van Soest, P.J., 1994. *Intake, Nutrition Ecology of the Ruminants*, 2nd ed. Cornell University Press, Ithaca, NY, pp. 337–353.
- Van Soest, P.J., Robertson, J.B., Lewis, B.A., 1991. Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. *J. Dairy Sci.* 74, 3583–3597.
- West, J.W., 2003. Effects of heat stress on production in dairy cattle. *J. Dairy Sci.* 86, 2131–2144.
- West, J.W., Hill, G.M., Fernandez, J.M., Mandevu, P., Mullinix, B.G., 1999. Effect of dietary fiber on intake, milk yield, and digestion by lactating dairy cows during cool or hot, humid weather. *J. Dairy Sci.* 82, 2455–2465.