



## Performance evaluation and adaptability of lactating dairy cows fed soybean and its by-products

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**ABSTRACT.** Aiming to evaluate the lactation performance and adaptability of confined dairy cows fed diets containing soybean and its by-products, this study used 12 Holstein cows with initial production of 30 kg milk<sup>-1</sup> day<sup>-1</sup> in feedlot system distributed in a 4 x 4 Latin square design. The experimental period lasted 84 days. The dry matter intake (DMI) and meteorological variables were recorded daily. Milk production was measured from the 15<sup>th</sup> to the 21<sup>st</sup> day, with milk analysis twice in each period, and physiological variables collected on the 15, 17<sup>th</sup> and 21<sup>st</sup> days of each experimental period. The thermal comfort indices and rectal temperature were considered normal, however the respiratory frequency and heart rate were different between the periods. Total milk production and percentage of crude protein were not affected. The thermal environment had influence on the CMS and on the percentage of milk fat in warmer periods, but the mechanism of heat dissipation was efficient for the animals to maintain homeothermy without affecting milk production.

**Keywords:** production and composition of milk, rectal temperature, heat stress, thermoregulation.

## Avaliação de desempenho lactacional e adaptabilidade de vacas leiteiras alimentadas com soja e seus subprodutos

**RESUMO.** Objetivou-se avaliar o desempenho lactacional e adaptabilidade de vacas leiteiras confinadas que receberam dietas contendo soja e seus subprodutos. Foram utilizadas 12 vacas da raça Holandesa com produção média inicial de 30 kg leite<sup>-1</sup> dia<sup>-1</sup> em sistema de confinamento, distribuídas em um quadrado latino 4 x 4. O período experimental teve duração de 84 dias. O consumo de matéria seca (CMS) e as variáveis meteorológicas foram registrados diariamente. A produção de leite foi medida no 15<sup>o</sup> ao 21<sup>o</sup> dia, o leite analisado duas vezes em cada período e as variáveis fisiológicas coletadas no 15, 17 e 21<sup>o</sup> dias de cada período experimental. Os índices de conforto térmico e as temperaturas retais foram considerados dentro da normalidade. As médias das frequências respiratórias e cardíacas diferiram no período da tarde, mas a produção total de leite e o percentual de proteína bruta não foram influenciados. O ambiente térmico exerceu efeito no CMS e na percentagem de gordura do leite na época mais quente, contudo os mecanismos de dissipação de calor foram eficientes para que os animais conseguissem manter a homeotermia sem prejuízo para a produção de leite.

**Palavras-chave:** produção e composição do leite, temperatura retal, estresse térmico, termorregulação.

### Introduction

Intensive milk production systems have been increasingly adopted in many developing countries. The performance of these animals is evaluated with some precision by measuring some variables, such as weight gain, feed efficiency, quantity and quality of milk and responses to environmental factors that impose, in combination or alone, a certain degree of stress to animals, determined by the results of the dysfunctions observed in the homeothermy.

The adequacy of diets as primary adjuvant to mitigate the problems arising from heat stress can help minimize the heat increment resulting from the metabolism of nutrients.

Changes in digestibility of foods can provide higher caloric increase, reducing net energy for maintenance and production. Such information indicates the need to consider the adaptability of different breeds in the planning of feeding and environmental management.

Dairy cows when kept in warm, moist environment and fed high forage or high grain diets have lower ruminal pH, possibly due to decreased ruminal activity and reduced food intake (NETO et al., 2011). There are few studies on the effect of thermal environment on the digestibility of food by ruminants, mostly conducted in environmental chambers. Variations in feed composition affect and may

explain the retention time of food in the rumen, microbial activity or the reduction of food into fine particles, events that explain the increased digestibility.

Morais et al. (2008) state that a major obstacle is the association between productive genotypes with animals adapted to heat, this because, mainly the most productive ones have significant decreases in food consumption and production.

Moreover, there is limited information on the influence of excessive protein intake, variation in dietary protein degradability and the amount of metabolizable protein on physiological functions of lactating cows subjected to high ambient temperatures. Notwithstanding, Huber et al. (1994) reported that the content of lysine in the diet is a major determinant of milk yield in cows subjected to heat stress.

The magnitude of environmental stress can be measured directly by changes in hormones and/or in blood flow, heart rate, body temperature as well as indirectly through the animal response in terms of productivity.

Given the above, this study evaluated the lactation performance and adaptability of Holstein cows in feedlot systems fed diets containing soybean and its by-products.

## Material and methods

The survey was conducted at the Unit for Teaching, Research and Extension in Dairy Cattle (UEPE-GL), Department of Animal Science, Federal University of Viçosa (UFV), Viçosa, Minas Gerais State. The municipality is located in the Zona da Mata Norte region, at 20°45'14" south latitude and 42°52'55" west longitude, with an altitude of 648 meters. The climate is Cwb type, according to the Köppen classification, i.e., mesothermal with hot, rainy summers and cold, dry winters. The average annual temperature is 21.8°C and annual rainfall of 1221.4 mm (INMET, 2009).

It was used 12 black and white Holstein cows with approximately 570 kg body weight, maintained in permanent housing system, in free stall. The experimental period lasted 84 days during fall. The first 14 days were used for adaptation of cows to diets; data were collected on the subsequent seven days. Cows were grouped according to the initial milk production (30 kg day<sup>-1</sup>), average of 160 lactation days, body condition score three and a half and on the 4<sup>th</sup> order of lactation, distributed in a 4 x 4 Latin square, each with four animals undergoing four treatments in four distinct periods.

Diets were formulated to be isonitrogenous with 25% crude protein (CP) as feed concentrates and

approximately 15.5% crude protein in the dry matter (Table 1). We used corn silage as forage for all diets. Chemical analysis was performed according to Silva and Queiroz (2002). Dry matter intake (DMI) was measured daily, milk production from the 15<sup>th</sup> to the 21<sup>st</sup> day, and milk composition twice in each period, on the 17<sup>th</sup> and 19<sup>th</sup> day. For all physiological variables, data collection took place on the 17, 19<sup>th</sup>, and 21<sup>st</sup> days.

**Table 1.** Chemical composition in percentage of DM of experimental diets.

Samples <sup>2</sup>	Diets <sup>1</sup>			
	SM	RS	RoS	SMU
DM	56.20	56.80	56.80	56.43
OM	94.01	93.66	93.66	95.02
CP	15.23	15.76	15.76	15.32
ADIN	3.77	3.48	3.48	3.05
EE	2.54	6.74	6.74	2.70
RDP <sup>3</sup> (%CP)	63.50	65.48	65.48	76.67
RUP <sup>3</sup> (%CP)	36.50	34.52	34.52	23.33
NDF	40.47	40.68	40.68	43.79
NDF <sub>cp</sub>	34.27	34.25	34.25	38.33
NFC	40.74	35.36	35.36	41.24

<sup>1</sup>SM (soybean meal); RS (raw soybean); RoS (roasted soybean); FSU (soybean meal plus 5% urea). <sup>2</sup>DM (dry matter); OM (organic matter); CP (crude protein); ADIN (acid detergent insoluble nitrogen); EE (ether extract); <sup>3</sup>rumen degraded protein (RDP), rumen undegraded protein (RUP) estimated by the equation of NRC (2001); NDF (neutral detergent fiber); NDF<sub>cp</sub> (neutral detergent fiber corrected for ash and protein); non-fibrous carbohydrates (NFC) = 100 - (%NDF<sub>cp</sub> + %CP + %EE + %ash); LIG (lignin).

Psychrometers and globe thermometers were installed inside and outside the stall, at 1.70 from the ground. Wind speed was measured with a portable anemometer every 2 hours from 6h to 18h. From the data of dry and wet bulb temperatures, we calculated the relative humidity, expressed as a partial vapor pressure. With the temperature of the globe thermometer and the wind speed, we calculated the Globe Temperature and Humidity Index (BGHI) for dairy cows, and estimated the radiant heat load (RHL) according to Silva (2008). This measure was calculated to check the influence of heat exchange by radiation between the environment and the animal.

Feed was given as a complete mixture ad libitum twice daily (50%) after the morning milking (8h) and the remaining on the afternoon (16h) to allow leftovers from 5 to 10% of the natural matter. Every day, forage and concentrate feed supplied to each animal were weighed and recorded to determine individual intake. Leftovers were weighed and discarded, before the morning meal, for purposes of adjusting the amount to be supplied.

Milk production per cow was monitored and recorded from the 15<sup>th</sup> to the 21<sup>st</sup> day of each period during the morning (6h) and afternoon (16h) milkings, expressed in kg. Individual milk samples were collected twice on the 17<sup>th</sup> and 19<sup>th</sup> day, and combined to form a composite sample proportional

to the productions of the morning and afternoon for analyzing the composition of fat and protein.

Respiratory rate was measured in each cow (through the movements of the flanks/minute) using a stop watch for 30 second-period and multiplying by two to obtain the result in minutes. Heart rate was obtained with a stethoscope placed directly on the left thoracic region, by counting the number of beats for 30 seconds and multiplying the value by two, thus determining the beats per minute. Rectal temperature was measured with digital clinical thermometers. Data on respiratory rate, heart rate and rectal temperature were collected from 8h to 9h and from 15 to 16 hours on the 17, 19<sup>th</sup>, and 21<sup>st</sup> days.

To test the effect of collection periods during the experiment, we used a Latin square design, and to test the effect of collection times on physiological data, we added this effect in subplot:

$$Y_{iklm} = \mu + T_i + Q_j + P_{k(i)} + A_{l(i)} + e_{ikl} + H_{lm} + TH_{ilm} + PH_{k(i)lm} + \varepsilon_{iklm}$$

which:

$Y_{ijk}$  is the observation of the variable relating to the treatment  $i$ , in the Latin square  $j$ , in the period  $k$ , in the animal  $l$  in the time  $m$ ;  $\mu$  is the overall constant;

$T_i$  is the effect relating to the treatment  $i$ ;

$Q_j$  is the effect of the Latin square  $j$ ;

$P_k$  is the effect of the period  $k$  nested within the Latin square  $j$ ;

$A_l$  is the effect of the animal  $l$  nested within the Latin square  $j$ ;

$e_{ijkl}$  is the residual effect of the experimental plots;  $H_{lm}$  is the effect of the collection time  $lm$ ;  $TH_{ilm}$ ;  $PH_{k(i)lm}$ ; and  $\varepsilon_{ijklm}$  is the random error associated with each observation, assuming NID (0;  $\sigma^2$ ). Pearson correlation was also run for the variables studied. All data were analyzed with the software SAS (2001) at 5% probability, and means were compared by Tukey's test.

### Results and discussion

Mean values of air temperature (Tar) inside the stall by the morning and afternoon showed small variations. The maximum temperature (25.40°C) was observed in the afternoon, in April, and the minimum (17.11°C) in the morning, in June. Wind speed (WS) was on average 0.40 km h<sup>-1</sup> (Table 2).

The relative humidity expressed in atmospheric pressure (Pp {ta}) was similar in the months studied, with the highest means verified in April, 1.98 (morning) and 2.04 (afternoon) associated with higher temperatures. It is likely that this may have hindered the process of heat dissipation and caused

stress in animals. Matarazzo et al. (2007) reported that, under conditions of high humidity, saturated air inhibits water evaporation from the skin and respiratory system providing more stressful environment for the animal.

**Table 2.** Meteorological variables and thermal comfort indices registered during the experimental period.

Season	Month	Meteorological variables <sup>1</sup>					
		Tar (°C)	WS (m s <sup>-1</sup> )	Pp{ta} (KPa)	PPt (mm)	ITGU	RHL (W m <sup>-2</sup> )
Morning							
Fall	April	22.37	0.37	1.98	1.9	72.60	443.78
Fall	May	18.03	0.32	1.73	0.2	66.43	426.58
Fall	June	17.11	0.30	1.78	0.7	66.96	426.18
Afternoon							
Fall	April	25.40	0.81	2.04	1.9	73.15	463.98
Fall	May	19.33	0.30	1.63	0.2	68.72	444.66
Fall	June	23.76	0.35	2.02	0.7	72.99	453.11

<sup>1</sup>Tar (air temperature); WS (wind speed); Pp{ta} (atmospheric pressure); PPt (rainfall) obtained from the meteorological station of Viçosa, Minas Gerais State); ITGU (globe temperature and humidity index); RHL (radiant heat load).

BGHI was virtually unchanged, with mean values from 66.43 to 72.60 (morning) and 68.72 to 73.15 (afternoon), and higher values also in April and RHL between 426.18 and 463.98 w/m<sup>2</sup>. In the present study, both BGHI and RHL were low and considered normal for protection of pure Holstein cattle (BUFFINGTON et al., 1981).

Regarding the highest (25.40°C) and lowest (17.11°C) Tar, the interior of the free stall had an ideal microclimate for the animals. According to Azevedo and Alves (2009), there is a wide variation with respect to thermoneutral range, ranging from 18°C to 21°C for cattle. Thermal comfort also depends on the relative humidity and the adaptation and metabolic rate of the animal, which considers the nutritional plan and production level.

The higher mean values of meteorological variables and BGHI by the afternoon April probably have triggered some change in the physiological behavior of animals due to the intensive milk production. Despite of the higher mean values of BGHI in April, 73.15/afternoon, the environmental conditions inside the free stall appeared to be satisfactory for thermal comfort. Possibly these animals reared in tropical environment have an adaptation to environmental variation. This result corroborates Silva (2008), who observed that the thermal comfort index up to 74 is ideal for dairy cows.

The physiological data of RR and HR, independent of the diet, remained above the reference values, with the highest mean values observed in the afternoon (p < 0.05) showing that the animals had to use the mechanism of respiratory evaporative heat loss as a way to dissipate heat (Table 3).

**Table 3.** Respiratory rate, heart rate and rectal temperature in the different diets and times of collection.

	Variáveis fisiológicas <sup>1</sup>			
	RR (mov. min. <sup>-1</sup> )	HR (beat min. <sup>-1</sup> )	RT (°C)	DMI (kg day <sup>-1</sup> )
	Diets <sup>2</sup>			
SM	38.96 ± 0.35	78.58 ± 9.33	38.48 ± 0.08	19.71 ± 0.50
RS	40.58 ± 0.39	77.25 ± 9.21	38.40 ± 0.08	18.99 ± 0.48
RoS	40.92 ± 0.41	77.42 ± 8.09	38.45 ± 0.06	19.06 ± 0.51
FSU	44.54 ± 0.43	78.92 ± 9.30	38.42 ± 0.07	19.63 ± 0.48
CV(%)	-	-	-	3.86
Times				
Morning	38.25 ± 0.20 <sup>a</sup>	75.92 ± 9.60 <sup>a</sup>	38.39 ± 0.06 <sup>a</sup>	
Afternoon	44.25 ± 0.65 <sup>b</sup>	80.17 ± 10.04 <sup>b</sup>	38.49 ± 0.05 <sup>a</sup>	

<sup>1</sup>RR (respiratory rate); HR (heart rate); RT (rectal temperature). <sup>2</sup>SM (soybean meal); RS (raw soybean); RoS (roasted soybean); FSU (soybean meal plus 5% urea). Means followed by the same letter in the column are not significantly different by Tukey-Kramer test at 5% probability.

The overall average of RR was 41.30 mov. min.<sup>-1</sup> close to that registered by Morais et al. (2008) of 45.5 mov. min.<sup>-1</sup> studying the behavior of thermoregulatory characteristics in dairy cows. Azevedo et al. (2005) stated that the respiratory rate is an indicator of heat stress better than the rectal temperature. This because animals absorb heat and change the temperature of the skin, activating the homeostatic mechanism, which lead to increased respiratory rate and prevent rise in rectal temperature before reaching a critical point.

Reference values of HR for adult cattle are between 50 and 70 beats per minute. In the present study, the values varied from 77.25 to 78.92 beat minutes<sup>-1</sup>. The increase in HR is related to several factors, including ambient temperature, age, individuality, temperament and degree of excitement of the animal. In turn, RT ranged from 38.39 to 38.49°C, within the normal range, suggesting that the thermoregulation mechanisms used were sufficient to maintain homeothermy. These results agree with those of Perissinotto et al. (2009), which used fuzzy classification and concluded that simultaneous values of RT and RR smaller or equal to 38.7°C and 54 mov. min.<sup>-1</sup>, respectively, indicate a very good thermal comfort.

Dry matter intake (DMI) ranged from 18.99 to 19.71 kg day<sup>-1</sup>, without differences between diets ( $p > 0.05$ ). However, these values are lower than recommended by NRC (2001) of 21 kg day<sup>-1</sup> for dairy cows producing 30 kg milk day<sup>-1</sup> corrected for 3.5% fat and with approximate weight of 576 kg.

The increase in RR and HR in the afternoon ( $p < 0.05$ ) was expected, since the combination of climatic variables and BGHI in this period may have imposed a greater discomfort to animals and hindered the processes of heat dissipation. In the treatment with the diet containing RS, we observed a lower intake. Possibly the effect of elevated environmental temperature in the afternoon in the diet containing RS of higher energy density has

caused a decrease in the intake and also for presenting high levels of degradable protein with excessive ammonia release in the rumen (Table 1) interfering negatively with the consumption. These results corroborate the study of Barletta et al. (2012) that reported that the inclusion of raw soybean at 24% reduces the dry matter intake.

Only RR correlated with RHL and WS. We verified a relationship of BGHI, Pp{ta} and Tar ( $p < 0.05$ ) with the other physiological variables and with the DMI. Tar also correlated ( $p < 0.05$ ) with the percentage of milk fat (Table 4).

**Table 4.** Pearson correlation between meteorological, physiological and production variables.

	Meteorological <sup>1</sup>				
	RHL(W m <sup>-2</sup> )	BGHI	Pp{ta} (KPa)	Tar(°C)	WS (m s <sup>-1</sup> )
	Physiological				
RR(mov. min. <sup>-1</sup> )	0.2212 <sup>a</sup> (0.0303) <sup>5</sup>	0.4216 ( $< 0.0001$ )	0.5437 ( $< 0.0001$ )	0.5613 ( $< 0.0001$ )	0.2428 (0.0171)
HR(beat min. <sup>-1</sup> )	-0.0482 (0.6406)	0.2905 (0.0041)	0.5097 ( $< 0.0001$ )	0.3023 (0.0028)	-0.0359 (0.7283)
RT (°C)	0.0507 (0.6236)	0.3169 (0.0017)	0.3645 (0.0003)	0.3670 (0.0002)	-0.0111 (0.9141)
Production <sup>3</sup>					
DMI (kg day <sup>-1</sup> )	-0.0965 (0.0772)	-0.1492 (0.0062)	-0.1285 (0.0184)	-0.1559 (0.0042)	-0.0692 (0.2054)
TMP (kg day <sup>-1</sup> )	0.0168 (0.7585)	0.0331 (0.5449)	0.0400 (0.4646)	0.0317 (0.5626)	-0.017 (0.7544)
Protein (%)	-0.2077 (0.1566)	-0.2726 (0.0608)	-0.2475 (0.0899)	-0.2325 (0.1117)	-0.2238 (0.1263)
Fat (%)	-0.0055 (0.9705)	-0.1712 (0.2446)	-0.2725 (0.0610)	-0.2901 (0.0455)	-0.2230 (0.1276)

<sup>1</sup>RHL (radiant heat load); BGHI (globe temperature and humidity index); Pp{ta} (partial vapor pressure); Tar (air temperature); WS (wind speed). <sup>2</sup>RR (respiratory rate); HR (heart rate); RT (rectal temperature). <sup>3</sup>DMI (dry matter intake); TMP (total milk production). <sup>4</sup>Correlation coefficient. <sup>5</sup>P-value.

There was a positive quadratic relationship between RR and all meteorological variables, indicating that in warmer periods, animals used the same physiological mechanisms of heat loss to avoid hyperthermia. This increased RR (tachypnea) is required to promote heat loss, in order to maintain body temperature within physiological limits.

Analyzing the effects of meteorological variables with the physiological ones, the results for RT showed a quadratic effect ( $p < 0.05$ ) for BGHI and linear ( $p < 0.05$ ) for Pp{ta} and Tar. The positive coefficients between BGHI and physiological variables indicate a quadratic effect ( $p < 0.05$ ) for RR, HR and RT. A quadratic effect was found of Pp{ta} ( $p < 0.05$ ) for RR and HR and linear ( $p < 0.05$ ) for RT. Tar indicated a quadratic effect ( $p < 0.05$ ) for RR and linear ( $p < 0.05$ ) for HR and RT. The increase in BGHI and in these meteorological elements cause thermal discomfort reflected in greater increases in these physiological variables.

Comparing the thermal environment and its effects on DMI and the percentage of milk fat, BGHI, Pp{ta}

and Tar revealed a linear effect ( $p < 0.05$ ). Unfavourable climatic conditions are partially responsible for the decline in food consumption and changes in milk constituents. Pinarelli (2003) observed that, for cows kept at low temperatures, the content of fat, protein and lactose were 3.47, 3.07 and 5.08%, for animals kept at intermediate temperatures, 3.46, 3.02 and 5.06%, and 3.17, 2.89 and 5.01%, respectively, for cows maintained at high temperatures. Barbosa et al. (2004) investigated Holstein cows in the shade, and observed that higher meteorological variables influenced the content of protein and fat. In this way, animals tend to reduce food intake in an attempt to decrease metabolic rate and hence the heat production. In accordance with NRC (2001), food intake begins to decrease when the ambient temperature is between 25 and 26°C; at 30°C is observed a more pronounced drop, and at 40°C, food intake reduces by 40% in lactating Holstein cows. The literature indicates that the reduction in food intake in cows undergoing heat stress is mainly due to the reduction in forage intake, which is aggravated when the animal cannot select the food. The greater participation of the forage in the diet, the greater the heat production per unit of metabolizable energy. In this study, cows were given feed completely mixed with 30:70 (corn silage: concentrate) and small possibility of selection. This feeding system probably contributed to increased heat production per unit of metabolizable energy or through other uncontrolled factors, and not necessarily due to climate variables.

There was a positive correlation between DMI and RT, with a quadratic effect ( $p < 0.05$ ) with all physiological variables studied (Table 5).

**Table 5.** Pearson correlation coefficient between physiological variables and milk production variables.

Production variables <sup>1</sup>	RR	HR	RT
DMI (kg day <sup>-1</sup> )	- 0.3685 <sup>3</sup> (0.0002) <sup>4</sup>	- 0.7859 ( $< 0.0001$ )	0.9603 ( $< 0.0001$ )
TMP (kg day <sup>-1</sup> )	- 0.3314 ( $< 0.0001$ )	0.1017 (0.1603)	- 0.0614 (0.3973)

<sup>1</sup>DMI (dry matter intake); TMP (total milk production). <sup>2</sup>RR (respiratory rate); HR (heart rate); RT (rectal temperature). <sup>3</sup>Correlation coefficient. <sup>4</sup>P-value.

The highest correlation coefficient was found between RT and DMI ( $r = 0.9603$ ). TMP was negatively correlated with RR, with a linear effect ( $p < 0.05$ ). The quantification of the direct environmental effects on milk production is complex, once this variable is largely affected by other factors, such as nutritional management or mobilization of body energy. The main reason for the decrease in milk production is the reduction of food intake in hot environments. And in situations of heat stress, it is possibly due to the action of heat, increasing RR and, as

a consequence, reduced activity of the gastrointestinal tract, which result in slow passage rate and rapid filling of the rumen, reduction in grasping, chewing and rumination of food causing decreased milk production. Also according to Porcionatto et al. (2009) due to hypofunction of the thyroid by the energy spent to eliminate excess body heat.

## Conclusion

The thermal environment influenced the dry matter intake and milk fat percentage in the warmer period, but the heat dissipation mechanisms were efficient and enabled animals to maintain homeothermy without affecting milk production.

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