# Intake, nutrient digestibility and nitrogen balance in lactating dairy cows fed diets containing sunflower cake

# Consumo, digestibilidade dos nutrientes e balanço de nitrogênio em vacas leiteiras alimentadas com dietas contendo torta de girassol

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# Abstract

The objective of this study was to evaluate the effect of different levels of sunflower cake (SC) inclusion in the diets of lactating dairy cows on dry matter (DM) intake, the apparent digestibility of dietary nutrients and nitrogen balance. Eight Holstein-Zebu cows with a milk yield of  $20 \pm 2$  kg/day were allotted to a  $4 \times 4$  replicated Latin square design, where were tested the effects of four levels of SC inclusion (0, 7, 14 and 21% of DM basis). Tifton-85 (*Cynodon dactylon*) hay was used as roughage in a 60:40 roughage:concentrate ratio. The SC inclusion had no effect on dry matter, crude protein, organic matter (OM) or total carbohydrate (TC) intake (kg/day). However, a quadratic effect (P < 0.05) was detected on neutral detergent fiber and non-fibrous carbohydrate (NFC) intake (kg/day). There were no effects of SC inclusion on OM, TC and NFC digestibility, as well as, for nitrogen balance. It was observed a linear effect for the basal endogenous nitrogen (g/day). Sunflower cake can be recommended as an ingredient in the rations of lactating dairy cows.

Key words: Agro-industry residues, dairy cow, fiber

# Resumo

O objetivo deste estudo foi avaliar o efeito de diferentes níveis de inclusão de torta de girassol (TG) na dieta de vacas em lactação sobre o consumo de matéria seca (MS), digestibilidade aparente dos nutrientes da dieta e balanço de nitrogênio. Oito vacas Girolanda com produção diária de leite de  $20 \pm 2$  kg/dia foram distribuídas em delineamento em quadrado latino duplo 4x4, em que foram testados os efeitos de quatro níveis de inclusão da TG (0, 7, 14 e 21% MS). Foi utilizado o feno de Tifton-85 (*Cynodon dactylon*) como volumoso em uma relação volumoso:concentrado de 60:40. A inclusão de TG não influenciou o consumo (kg/dia) de MS, proteína bruta, matéria orgânica (OM) e carboidratos totais (CT). Entretanto, foi observado efeito quadrático (P < 0,05) no consumo (kg/dia) de fibra em detergente neutro e carboidratos não-fibrosos (CNF). Não houve efeito da inclusão de TG na digestibilidade de MO, CT e CNF, assim como para o balanço de nitrogênio. Foi observado efeito linear para o nitrogênio endógeno basal (g/dia). A torta de girassol pode ser recomendada como um ingrediente de rações para vacas em lactação. **Palavras-chave:** Fibra, subprodutos agroindustriais, vacas leiteiras

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# Introduction

Lactating dairy cows require large amounts of dietary energy for maintenance, milk production and reproduction (RABELO et al., 2003). Feeding diets containing more than 50% of ration dry matter (DM) as forage to highly productive dairy cows has been shown to support similar levels of milk production (CHERNEY; CHERNEY; CHASE, 2004). High quality forages have high digestibility and can be consumed in greater quantities than low quality forages that have a high NDF and low digestibility (LLAMAS-LAMAS; COMBS, 1991). The main factors that might affect the partitioning of energy in lactating cows are energy intake, health status, environmental conditions and individual variability among cows with respect to the efficiency of energy utilization for maintenance and production (BROSH, 2007).

Appropriate feeding management and the use of intensive systems of exploitation may be favorable strategies for improving dairy production, especially during the dry season, which makes the use of nutritional and low cost foods essential (PEREIRA et al., 2009). Sunflower can consist in an alternative for energy and protein supplementation for animals in regions where soybean and maize could not produce successfully (NGONGONI et al., 2007), like in northeastern Brazil.

Agro-industry residues may be valuable sources of protein, fiber and energy. Sunflowers are grown for oil extraction from whole or dehulled seeds, generating large amounts of by-products. Different by-products are obtained according to extraction method: sunflower cake from mechanical oil extraction and sunflower meal from chemical extraction with solvents. The nutritional composition of sunflower by-products is variable and directly related to the oil extraction process and the amount of bark remaining (ENSMINGER; OLDFIELD; HEINEMANN, 1990).

The use of sunflower seed as a source of biofuel provides interesting opportunities for farmers,

both in terms of oil production and interesting byproducts. The cake resulting from the grain pressing to obtain sunflower oil is an ingredient nutritionally rich for animal feed, containing high levels of protein, lipid and fiber (OLIVEIRA; CÁCERES, 2005). Considering other advantages of using byproducts and co-products from the biofuels industry, there is the possibility of obtaining gain from the carbon credit market, due reduction of emission of methane gas when using rations containing oil (ABDALLA et al., 2008). According to studies conducted in Australia and Canada, for each 1% increase of fat in the diet of ruminant can be reduced up to 6% the amount of methane produce per kg of dry matter consumed (LIMA, 2011).

Information about the use of sunflower cake in feeds is scarce (MATTII; PRIORI; TROMBETTA, 2009). Although research performed in several countries has provided some information about sunflower cake, an understanding of its nutritional properties is required in order to use these products in animal feeding. Therefore, the objective of this study was to evaluate intake, apparent digestibility and nitrogen compound balance in lactating cows fed diets containing different levels of sunflower cake.

# **Materials and Methods**

### Animals, experimental design and diets

The experiment was carried out in Quixeramobim, Ceara, Brazil, which has a semi-arid climate. Eight lactating Holstein-Zebu cows were used, with an average weight of  $515 \pm 21$  kg, milk production of  $20 \pm 2$  kg/d, 7 to 10 weeks postpartum and a parity of  $3.0 \pm 1$ . The animals were randomly distributed in a 4 × 4 replicated Latin square design. The animals were weighed at the beginning and at the end of each experimental period. Animal care and handling procedures were followed according to the University's animal care committee.

The animals were kept in a feedlot system

in individual covered pens, supplied with an individual feeder and drinker. Animals were fed a diet containing Tifton-85 (*Cynodon dactylon*) hay and concentrate (60 and 40%, respectively, on DM basis). The Tifton-85 hay presented low quality, with an average of CP and NDF of 96.88 g/kg of DM and 834.47 g/kg of DM, respectively (Table 1).

The treatments consisted of four levels of sunflower cake inclusion in the concentrate ration (0, 7, 14 and 21% of DM; Table 2). The diets were formulated to provide 13.0% of CP and to meet the nutritional requirements of a cow, with 550 kg, producing 20 kg of milk containing 3.5% fat (NRC, 2001).

Table 1. Chemical composition of Tifton-85 hay (TH), corn meal (CM), soybean meal (SBM), wheat bran (WB) and sunflower cake (SC), in g/kg DM.

Chemical analysis	Ingredients							
	TH	СМ	SBM	WB	SC			
Dry matter	843.10	816.60	836.41	799.01	836.55			
Crude protein	96.88	100.43	502.83	166.39	223.29			
Ether extract	16.21	46.72	25.34	56.48	116.28			
Neutral detergent fiber	834.47	217.29	167.70	352.55	537.71			
Acid detergent fiber	403.01	88.45	112.07	117.78	176.89			
Total carbohydrates	795.79	826.77	396.54	538.27	654.64			
Non-fibrous carbohydrates	33.93	646.52	275.98	255.39	183.47			

**Source:** Elaboration of the authors.

Table 2. Proportion (% DM) of ingredients (g/kg DM) of the diets.

	Inclusion levels of sunflower cake (%)							
Ingredients	0	7	14	21				
	Proportion of the ingredients in the diets (% DM)							
Tifton-85 hay	60.00	60.00	60.00	60.00				
Corn meal	23.08	22.39	22.73	21.63				
Soybean meal	9.26	7.26	5.13	3.33				
Wheat meal	6.68	6.62	5.80	5.86				
Sunflower cake	0.00	2.80	5.60	8.40				
Limestone	0.52	0.54	0.54	0.54				
Dicalcium phosphate	0.29	0.22	0.13	0.07				
Mineral mixture <sup>1</sup>	0.16	0.16	0.16	0.16				
Chemical analysis	Composition of diets (g/kg DM)							
Dry matter	835.61	835.22	834.04	838.29				
Crude protein	138.88	134.28	128.79	124.98				
Ether extract	27.14	29.79	31.15	35.41				
Neutral detergent fiber	781.61	784.82	802.77	794.07				
Acid detergent fiber	279.30	281.86	290.83	290.59				
Total carbohydrates	769.94	770.23	765.71	765.48				
Non-fibrous carbohydrates	241.67	211.55	191.71	198.87				
TDN (% of DM) $^2$	67.30	69.48	65.80	77.61				

<sup>1</sup>Composition: Ca – 7.5%; P – 3%; Fe – 16.500 ppm; Mn – 9.750 ppm; Zn – 35.000 ppm, I – 1.000 ppm; Se – 225 ppm; Co – 1.000 ppm.

Source: Elaboration of the authors.

<sup>&</sup>lt;sup>2</sup> Total digestible nutrients

#### Experimental procedures and sample collection

Cows were fed *ad libitum* twice a day (8:00 and 16:00 h) and milked twice daily at 6:00 and 16:00 h. Feed refusals were measured before morning feeding and the amount of feed offered was adjusted to allow 10% refusal. The animals had free access to water throughout the trial.

The experimental period lasted 64 days and each period consisted of 10 days of adaptation (FERREIRA et al., 2009) and 6 days of sample collection. Samples of experimental diets and refusal were weighed daily and stored (-20°C) for subsequent chemical analysis. Feces were collected from the rectum in the morning and afternoon of the collection period, in order to estimate the digestibility of dietary constituents. A sub-sample was taken, weighed and dried in a forced air oven at 55°C for 72 h, and then ground through a 1-mm screen (Wiley mill; Arthur H. Thomas, Philadelphia, PA). At the end of each experimental period, a composite sample was made for each animal, based on the pre-dried weight of each sampling day.

Indigestible NDF was used as the internal marker to calculate fecal excretion and the apparent digestibility of nutrients. The indigestible content of the composited total mixed ratio (TMR), orts and fecal samples was determined using *in situ* incubation (LYKOS; VARGA, 1995).

On the 16<sup>th</sup> day of each period, two urine samples (morning and afternoon) were collected from each animal by spontaneous urination approximately 4 hours after feed supply. The samples were stored at  $-20^{\circ}$ C prior to laboratory analysis. At the end of the experiment, samples were thawed and homogenized for preparation of a composite sample per cow per period for quantification of creatinine and urinary nitrogen.

#### Chemical analyses

Total dry matter was analyzed by drying the sample at 105°C to a constant weight (method

930.15; AOAC, 1990). Total nitrogen was measured using the Kjeldahl method (method 955.04; AOAC, 1990). Crude protein (CP) was calculated by multiplying %N by a factor of 6.25 (method 984.13; AOAC, 1990). Acid detergent fiber (ADF) was determined using acetyl-trimethyl ammonium bromide detergent in 0.5 M sulfuric acid (GOERING; VAN SOEST, 1970). Neutral detergent fiber (NDF) was determined using sodium sulfite and amylase (VAN SOEST; ROBERTSON; LEWIS, 1991).

Carbohydrates (TC) were determined using the following equation: TC = 100 - (%CP + %EE+ %MM) (SNIFFEN et al., 1992). Non-fibrous carbohydrates (A + B1) were determined according to the following equation: NFC = 100 - (%CP + %EE +%NDFap + %MM), in which NDFap is equivalent to the cell wall corrected for ash and protein. Total digestible nutrients (TDN) were calculated according to Sniffen et al. (1992), using the following equation: TDN = DCP + (DEE × 2.25) + DTC, in which, DCP = apparent digestible crude protein, DEE = apparent digestible ether extract and TDC = apparent digestible total carbohydrate.

Creatinine was determined with commercial kits (Labtest<sup>®</sup>, Lagoa Santa, Minas Gerais, Brazilian Industry), following the end-point method, using picrate and acidifier. Urine volume (UV) was estimated according to the equation: UV (L/d) =  $(29 \times \text{live weight, kg})/(\text{creatinine, mg/L})$  (VALADARES FILHO; VALADARES, 2001), in which, 29 = average daily creatine excretion in mg/ kg BW, obtained for dairy cows.

Nitrogen balance (NB) was calculated from the amounts of N (g/d) consumed and excreted in the feces, in the urine and in the milk, as follows: NB (g/d) = Nconsumed – Nfeces – Nurine – Nmilk. Nitrogen content in the milk was obtained by dividing the milk CP by conversion factor 6.38 (MCDONALD, 1993). Basal endogenous nitrogen (BEN) was calculated using the following equation: BEN (g/d) =  $(0.35 + 0.018) \times BW^{0.75}$  (AFRC, 1993).

The value of N retained (NR) was expressed as: NR (g/d) = NB - BEN.

#### Statistical analysis

Data for intake, the apparent digestibility of dietary constituents and nitrogen balance were subjected to ANOVA for a 4 x 4 replicated Latin square design. Terms in the model included square, period (square), cow (square), treatment, and the interaction of treatment and square. An orthogonal partition of the sum of squares of treatments into linear and quadratic degree effects was obtained following the analysis of variance. All statistical analyses were performed using the PROC GLM procedure in SAS 9.0 (SAS, 2005) and a probability level of 0.05, as the follow statist model.

$$Y_{ijkl} = \mu + S_i + T_l + P_{j(i)} + C_{k(i)} + ST_{il} + \varepsilon_{ijkl}$$
, where:

Y<sub>iikl</sub> is the dependent variable;

 $\mu$  is the overall mean;

 $S_i$  is the effect of square i;

 $T_1$  is the effect of treatment l;

 $P_{i(i)}$  is the effect of period j (within square i);

 $C_{k(i)}$  is the effect of cow k (within square i);

 $ST_{il}$  is the interaction between square i and treatment l;

 $\varepsilon_{iikl}$  is the residual error.

#### **Results and Discussion**

The sunflower cake is an alternative source of protein and energy, but can present extreme variation in lipid content (60.0 to 300.0 g/kg) and the high content of polyunsaturated lipids can improve the milk quality (OLIVEIRA; VIEIRA, 2004). The discovery using animal models and tissue culture that conjugated linoleic acid (CLA), which is found

in dairy products (LIN et al., 1995), possesses potential beneficial effects to human health is a positive health image of milk fat (PARODI, 1997).

There was no effect of sunflower cake inclusion on DM, OM, CP and TC intake expressed as kg/d, with average values of 14.06, 10.31, 1.8 and 10.36 kg/d, respectively (Table 3). The DM intake also did not present difference when expressed in %BW (2.74%) or g/kg<sup>0.75</sup> (130.89 g/kg<sup>0.75</sup>) (Table 3). According to NRC (2001) for cows with average milk production of 20 kg/day weighing 500 kg, the DM intake is 16.0 kg/day, value higher than obtained in this study. These differences may be related to the diet, environment and genotype, because crossbred Holstein-Zebu cows are more adapted to tropical conditions, present smaller frame size in maturity and therefore have lower dry matter intake, as was observed in this work. According to Mertens (1987), when the energy density of the diet is high (low NDF), in relation to requirements of the animal, intake may be limited by energy demand, not occurring rumen fill. For diets of low energy density (high content of NDF), the intake can be limited by the filling of the rumen reticulum.

Santos et al. (2009) studying the effect of inclusion of sunflower cake in the diet of lactating Holstein cows did not observed differences in intake with a mean value of 14.8 kg DM/day and 3.0% BW, similar to the obtained in this study.

A quadratic effect of SC levels in the diet on the NDF (P<0.05) and NFC (P<0.05) intakes (kg/d) was detected, with critical points (maximum response) at the SC levels of 14.01 and 13.02% DM, respectively. A similar pattern was also observed for NDF (P<0.05) intake, when expressed in %BW and g/kg<sup>0.75</sup>, with critical points (maximum response) at SC levels of 11.73 and 12.64% DM, respectively. On the other hand, ADF intake (kg/d) increased linearly (P<0.05) as the SC levels in the diet increased (Table 3).

The inclusion of SC at the levels of 0, 7, 14 and 21% provided intake of NDF of 16.01, 16.52,

18.35 and 16.97 g/kg BW, respectively. This can be attributed to the high levels of NDF in the experimental diets (781.61, 784.82, 802.77 and 794.07 g/kg DM, respectively) due the high levels of NDF in the Tifton-85 hay used (Table 1). This high level of indigestible NDF promoted the filling effect. The apparent digestibility of DM was higher than 69% for all experimental diets (Table 4), which may lead to incorrect interpretation of the physiological limitation of consumption. However, the model NDF-energy intake, formulated by Mertens (1994), provides that the intake is limited by the filling when the daily intake of NDF is greater than 11 to 13 g/kg BW. Thus, for the present experiment can be inferred that the intake was regulated by physical mechanisms, as the NDF intake ranged from 16.3 to 18.0 g/kg BW (Table 3).

**Table 3.** Least squares means and mean square error (MSE) for average daily intake of dry matter (DM), organic matter (OM), crude protein (CP), ether extract (EE), neutral detergent fiber (NDF), acid detergent fiber (ADF), total carbohydrate (TC), non-fibrous carbohydrate (NFC) and total digestible nutrients (TDN) according to sunflower cake levels in the diet

Variables —	Inclu	sion levels of	sunflower c	DNE	MCE		
	0	7	14	21	r>r	WISE	
			(k	g/d)			
$DM^a$	13.62	13.87	14.74	14.01	ns	0.66	
$OM^b$	10.25	10.09	10.65	10.24	ns	0.43	
CP <sup>c</sup>	1.85	1.82	1.83	1.70	ns	0.01	
EE <sup>d</sup>	0.39	0.59	0.84	1.09	0.001	0.004	
NDF <sup>e</sup>	8.28	8.51	9.45	8.74	0.030	0.30	
$\mathrm{ADF^{f}}$	3.71	3.80	4.14	3.94	0.013	0.05	
TC <sup>g</sup>	10.41	10.51	9.96	10.57	ns	0.34	
NFC <sup>h</sup>	1.80	2.28	2.20	2.15	0.001	0.03	
TDN <sup>i</sup>	9.18	9.66	9.70	10.88	0.001	0.49	
			(%	BW)			
$\mathbf{D}\mathbf{M}^{\mathrm{j}}$	2.69	2.80	2.80	2.69	ns	0.03	
$NDF^{k}$	1.63	1.73	1.80	1.69	0.020	0.01	
$TC^1$	2.05	2.09	1.87	2.01	ns	0.02	
NFC <sup>m</sup>	2.31	2.31	2.03	2.22	0.01	0.02	
			(g/k	(g <sup>0.75</sup> )			
$DM^n$	127.48	132.70	134.33	129.07	ns	63.12	
NDF°	77.40	82.05	86.45	81.06	0.020	27.56	
TDN <sup>p</sup>	85.90	90.99	87.59	98.81	0.001	46.54	
${}^{a}\tilde{Y} = 14.0609$				${}^{j}\tilde{Y} = 2.7456$			
${}^{b}\tilde{Y} = 10.3050$				${}^{k}\hat{Y} = 1.6250 + 0.0$	0258X - 0.0011X2 (H	$R^2 = 0.72$ )	
$^{\circ}\tilde{Y} = 1.5900$				$^{1}\tilde{Y} = 2.0050$			
$^{d}Y = 0.3754 + 0.0334X (R^2 = 0.99)$			$^{11}Y = 2.3018 - 0.0081X (R^2 = 0.84)$ $^{11}X = 120.8025$				
${}^{\circ}Y = 8.1584 + 0.1345X - 0.0048X2 (R^{2} = 0.64)$ ${}^{\circ}Y = 130.8925$ ${}^{\circ}\hat{Y} = 76.0200 + 1.2070X - 0.0512X^{2} (R^{2} = 0.64)$					$R^2 = 0.61$ )		
$s \tilde{Y} = 10.3625$	$\hat{V}_{1} = 1 - \frac{1}{2} \frac{1}{$						
$h\hat{Y} = 1.8286 + 0.0$	703X - 0.0027	$X^2 (R^2 = 0.87)$		1 00.0270 0			
$\hat{Y} = 9.0841 + 0.07$	$734X (R^2 = 0.8)$	5)					
Source: Elaborati	on of the autho	rs.					

The digestibility coefficient of DM and NDF showed a linear response (P<0.05) to increasing levels of SC inclusion, therefore, the ether extract content were not high enough to alter the fiber digestion. The CP and EE showed a quadratic response (P<0.05), with the maximum effect at 12.24 and over 21% of SC, respectively. High levels of ADF in the diet can

affect the digestibility of nutrients, given that the lignin fraction (indigestible) represents the largest proportion of ADF (EASTRIDGE, 1997). Despite the high ADF content of sunflower cake, there was no reduction in the digestibility of nutrients, which can be considered a satisfactory result for the studied levels (Table 1 and 4).

**Table 4.** Least square means and mean square error (MSE) for apparent digestibility coefficients (%) of dry matter (DM), organic matter (OM), crude protein (CP), ether extract (EE), neutral detergent fiber (NDF) and total carbohydrate (TC) and non-fibrous carbohydrate (NFC) according to sunflower cake levels in the diet.

Variables	Inclusion levels of sunflower cake (%)	D>E	MSE			
variables	0	7	14	21	<b>r&gt;r</b>	IVISE
DM <sup>a</sup>	69.96	71.81	72.07	72.43	0.020	3.35
$OM^{b}$	65.20	65.93	66.16	66.76	ns	6.20
CP <sup>c</sup>	75.06	75.47	77.34	75.42	0.030	1.86
EE <sup>d</sup>	69.17	80.12	85.65	89.50	0.001	2.08
NDF <sup>e</sup>	59.85	62.39	63.21	64.13	0.002	4.96
$\mathrm{TC}^{\mathrm{f}}$	70.80	71.99	68.87	72.49	ns	2.64
NFC <sup>g</sup>	87.29	88.97	87.08	88.73	ns	2.68

 $^{a}\hat{Y} = 70.4172 + 0.1099X (R^{2} = 0.81)$ 

 ${}^{b}\tilde{Y} = 66.01$ 

 $^{\circ}\hat{Y} = 74.7980 + 0.2914X - 0.0119X^{2} (R^{2} = 0.86)$  $^{\circ}\hat{Y} = 69.3544 + 1.7117X - 0.0363X^{2} (R^{2} = 0.99)$   ${}^{e}\hat{Y} = 60.3485 + 0.1951X (R^{2} = 0.92)$  ${}^{f}\tilde{Y} = 71.0361$ 

 ${}^{g}\tilde{Y} = 88.0169$ 

Source: Elaboration of the authors.

The amount of N excreted in milk was associated quadratically (P<0.05) with the SC levels in the diet, with the maximum at 7.17% SC (Table 5). Basal endogenous nitrogen (BEN) (g/d) showed a linear response to SC. According to Riis (1983), the protein deposition rate increases up to a certain

level as the supply of energy, protein and other nutrients increases. The highest intakes of TDN were recorded in diets with high levels of SC. This suggests the availability of nutrients for the deposition and renewal of tissue proteins, which was supported by the analysis of N retention by animals, indicating net N available for gain.

Variables -	Inclus	sion levels of	D> E	MOE		
	0	7	14	21	- r>r	WISE
MP (kg/day) <sup>a</sup>	18.88	19.92	20.56	19.29	0.001	0.50
MUN (mg/dL) <sup>b</sup>	13.04	13.15	12.40	11.65	0.001	0.49
NC $(g/d)^{c}$	238.78	241.53	250.06	234.53	ns	163.42
NF $(g/d)^d$	68.17	64.82	65.76	65.22	ns	33.85
NU $(g/d)^e$	1.55	1.40	1.16	1.48	ns	0.26
NM $(g/d)^{f}$	9.59	9.64	9.75	8.66	0.002	0.19
NB $(g/d)^g$	159.47	165.67	173.40	159.17	ns	179.16
BEN $(g/d)^h$	39.42	39.23	40.78	40.51	0.001	0.08
NR $(g/d)^i$	120.05	126.44	132.62	118.66	ns	181.52

**Table 5.** Least square means and mean square error (MSE) for milk production (MP), milk urea nitrogen (MUN), nitrogen consumed (NC), nitrogen excreted in feces (NF), in the urine (NU) and in the milk (NM), nitrogen balance (NB), basal endogenous nitrogen (BEN) and nitrogen retention (NR) according to sunflower cake levels in the diet

 $^{a}\hat{Y} = 19.6311 + 0.0002X (R^{2} = 0.82)$  by Pereira et al. (2011)

 ${}^{b}\hat{Y} = 13.2963 - 0.0702X (R^{2} = 0.84)$  by Pereira et al. (2011)

 $^{\circ}\tilde{Y} = 241.2258$ 

 ${}^{d}\tilde{Y} = 65.9906$ 

 $^{\circ}\tilde{Y} = 1.3986$ 

Source: Elaboration of the authors.

Conclusions

Sunflower cake can be recommended as an ingredient in the rations of lactating dairy cows.

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 $^{f}\hat{Y} = 9.5264 + 0.0846X - 0.0059X^{2} (R^{2} = 0.90)$ 

 ${}^{g}\tilde{Y} = 164.4260$ 

 $^{h}\hat{Y} = 39.2609 + 0.0690 X (R^{2} = 0.65)$ 

 ${}^{i}\tilde{Y} = 124.4403$ 

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