



## Use of dried brewers' grains instead of soybean meal to feed lactating cows

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**ABSTRACT** - The objective of this study was to determine the optimal level of dried brewers' grains (DBG) to replace soybean meal in diets for lactating Holstein cows. Five cows, around 88±28 days in milk, were distributed in a 5 × 5 Latin square design and fed diets containing different levels of DBG (0, 25, 50, 75, and 100%). The forage:concentrate ratio of the diet was 50:50. Feed intake, dry matter, nutrient digestibility, microbial synthesis, milk production and composition, and the economic viability of the diets were evaluated. There was reduction in dry matter intake and, consequently, in crude protein and non-fiber carbohydrates with increased levels of DBG. This occurred due to physical limitation of rumen caused by increased neutral detergent fiber intake. Ether extract intake also increased with levels of DBG due to higher concentrations of this nutrient in the diet. Apparent dry matter, organic matter, ether extract, and neutral detergent fiber digestibility increased with replacement of soybean meal by DBG. Milk production showed a quadratic effect and the levels of fat, protein, and total solids reduced linearly. Each 1% of soybean meal replaced by DBG in concentrate led to a reduction of 0.04, 0.02, and 0.06 g kg<sup>-1</sup> of milk fat, protein, and total solids, respectively. The milk production efficiency increased linearly and the microbial synthesis efficiency was not affected. The economic return increased along with the DBG levels. Thus, DBG levels replacing up to 75% of soybean meal can be used to feed lactating cows, since it provides improvements in digestibility, milk production efficiency, and economic return without affecting microbial efficiency.

Key Words: by-product, digestibility, intake, milk composition, protein, purine

### Introduction

In Brazil, the most commonly used ingredient as protein source in animal feed is soybean meal. However, the majority of soybeans are exported, mainly for the European market (Coronel et al., 2009), which has raised the grain and soybean meal cost in the domestic market. This scenario has motivated the search for alternative feed ingredients that can be stored, cost less, and have a comparable nutritional composition.

The use of agribusiness waste for ruminant feed provides a reduction in feed cost and allows for the reuse of organic material in animal production, thus preventing

waste accumulation and environmental pollution (Brochier and Carvalho, 2009). Among the various industrial wastes, brewers' grains has increased due the excessive supply used for increased beer production in Brazil (Albuquerque et al., 2011). The amount of wet brewers' grains is 32.02% higher than the amount of barley used for the initial process of brewing beer (Brochier and Carvalho, 2009).

High humidity of waste might limit its utilization on farms distant from the brewing industry, because the humidity prevents transportation and storage. Thus, the drying process is an interesting alternative, which also increases the lifespan of brewer's grains and reduces the storage volume (Aliyu and Bala, 2011). However, data using dried brewers' grains (DBG) in lactating cow diets are scarce. The use of brewer waste in the wet and fermented forms for lactating cows have showed positive results, because it does not affect the dry matter intake (Chiou et al., 1998) and milk production (West et al., 1994; Firkins et al., 2002; Geron et al, 2010; Imaizumi et al., 2015), but causes an increase in fat contents (Davis et al., 1983) and milk protein (West et al., 1994).

With a protein content ranging between 190 g kg<sup>-1</sup> and 308.0 g kg<sup>-1</sup> of dry matter (DM) (Russ et al., 2005; Valadares

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Filho et al., 2006), DBG is an alternative protein ingredient of lower cost, which can be used instead of soybean meal in ruminant feed, as ruminants have the capacity to convert plant waste into nutrients through the action of rumen microorganisms (Silva Filho et al., 1999). In this context, the objective of this study was to determine the optimal level of DBG for replacement of soybean meal in diets for lactating Holstein cows.

## Material and Methods

The experimental protocol was approved by the Ethic Committee on Animal Use (case no. 35/13) and performed following the ethical principles of animal experimentation. Five pregnant Holstein cows between the first and third lactation were used, with 88 + 28 days in milk, an average body weight of 626±31.9 kg (mean ± standard deviation), and an average initial milk yield of 27.0±4.86 kg d<sup>-1</sup>. The animals were assigned to a 5 × 5 Latin square design of 21 days, with 14 days for adaptation and seven days for data collection, totaling 105 days. The treatments were 0, 25, 50, 75, and 100% of DBG levels replacing soybean meal in the diets.

Brewer's grains were acquired in the wet form from a brewery located at Toledo, Paraná, Brazil. The wet waste was passed through a 31-h dehydration process. Sixteen hours involving direct sun exposure, in which the waste was distributed on a concrete surface forming a layer of approximately 1.5 cm with a density of 0.998 kg of DM per m<sup>2</sup>. The material was spread and turned over every

hour. At the end of the dehydration process, the DBG was collected, packed in bags, and stored in a covered barn.

The ingredients used in the formulation of experimental diets were *Cynodon* sp. hay, corn silage, ground corn, DBG, soybean meal, and a mineral and inorganic phosphate supplement. Diets were formulated to be isonitrogenous, according to the NRC (2001) for dairy cows with a body weight of 626 kg that are producing 27 kg of milk per day. The forage:concentrate ratio was 50:50 (Table 1).

Animals were housed in individual stalls in a barn. At the beginning and end of each experimental period, the cows were weighed before the morning feeding. The feed was offered twice daily at 06.30 h and 04.30 h, with 700 g kg<sup>-1</sup> and 300 g kg<sup>-1</sup> of feed provided at each time, respectively. The refusals were weighed daily and adjusted for 100 g kg<sup>-1</sup> of DM offered. Dry matter and nutrient intake were determined by the difference between the offered and refused feed.

From the 15th to 21st day of the study period, diet samples were collected, including silage, hay, concentrate, and refusals, and stored at -20 °C. To determine the digestibility, fecal samples were collected directly from the rectum according to the following schedule: 15th day (08.00 h), 16th day (10.00 h), 17th day (12.00 h), 18th day (14.00 h), 19th day (16.00 h) and 20th day (18.00 h). Subsequently, samples of diets, refusals and feces were dried in a forced-ventilation oven (55 °C for 72 h) and ground to pass through a 1-mm mill screen. The pool of samples was collected to compose a representative sample of diets, refusals, and feces per animal per period.

Table 1 - Ingredients, nutritional composition (g kg<sup>-1</sup> of dry matter), and metabolizable energy (Mcal kg<sup>-1</sup> of dry matter) of the experimental diets

Ingredient	Replacement level (%)				
	0	25	50	75	100
Tifton 85 hay	400.00	400.00	400.00	400.00	400.00
Corn silage	100.00	100.00	100.00	100.00	100.00
Ground corn	382.74	360.72	332.09	280.52	176.47
Soybean meal	104.23	94.24	78.14	53.82	-
Dried brewers' grains	-	32.50	78.14	154.94	313.73
Mineral supplement <sup>1</sup>	9.77	9.75	9.77	9.79	9.80
Dicalcium phosphate	3.26	2.79	1.86	0.93	-
Nutritional composition					
Dry matter (g kg <sup>-1</sup> as fed)	817.27	818.67	820.65	822.58	828.36
Organic matter	941.80	940.72	940.97	940.69	939.19
Ether extract	20.32	23.43	26.17	26.92	36.32
Crude protein	145.12	143.42	145.08	145.07	148.33
Neutral detergent fiber	405.07	434.60	463.20	512.58	605.20
Acid detergent fiber	203.25	208.87	218.46	227.11	258.26
Non-fibrous carbohydrates	411.44	385.70	358.50	318.24	228.01
Metabolizable energy	2.68	2.66	2.62	2.56	2.43

<sup>1</sup> Chemical composition (quantities in kg<sup>-1</sup>): Ca, 215 g; P, 65 g; Co, 45 mg; Mg, 12 g; Mn, 425 mg; Zn, 1900 mg; Se, 35 mg; I, 65 mg; S, 10 g; F, 650 mg; Fe, 1700 mg; Cu, 800 mg; Na, 75 g (commercial product).

Samples were analyzed for DM, mineral matter (MM), crude protein (CP), and ether extract (EE) by AOAC (1990) methodology and neutral detergent fiber (NDF) and acid detergent fiber (ADF) quantification according to Van Soest et al. (1991). The amount of organic matter (OM) was calculated as the difference between the MM content and total DM. Non-fiber carbohydrate levels (NFC) were calculated according to equations proposed by Sniffen et al. (1992). Dietary metabolizable energy was calculated according to the equation from the NRC (2001).

The DM and nutrient digestibilities were estimated by the difference between the amount consumed and amount excreted. To estimate the daily fecal excretion, insoluble acid detergent fiber (iADF) was used as an internal marker. The iADF was estimated in samples of offered feed, refusals, and feces by an *in situ* digestibility procedure. The iADF was obtained after 264 h of incubation (Casali et al., 2008) in F57 filters (ANKOM® Technology Corporation) and then analyzed for ADF.

Cows were milked twice daily at 06:00 and 04:00 h. Milk production was recorded during the collection period using gauges attached to the milking equipment. The fat corrected milk (FCM) for 35 g kg<sup>-1</sup> of fat was calculated according to Sklan et al. (1992). Milk production efficiency (MPE) was calculated by dividing the milk yield average of each cow by its DM intake average in each experimental period.

Milk samples were collected on the 15th and 16th day of each period and were composed proportionally from the morning and afternoon milking. For chemical analysis, milk samples were placed in plastic vials containing Bronopol® (2-bromo-2-nitropropano-1,3-diol), which were mailed to the laboratory to determine the contents of fat, protein, lactose, and total solids.

To measure milk allantoin and urea nitrogen, milk samples were deproteinized on filter paper using 5 mL of 25% trichloroacetic acid to 10 mL of milk. Subsequently, the filtrate was used to determine allantoin by the same method used in determination of urine allantoin according to Chen and Gomes (1992). Milk urea nitrogen was measured using a commercial kit.

For evaluation of microbial synthesis, urine was collected on the 17th day as spot sampling 4 h after the morning feeding. Uric acid and creatinine concentrations were determined by the colorimetric method. The average daily excretion of creatinine, considered to indicate the urinary volume, was 24.05 mg kg<sup>-1</sup> body weight (Chizzotti et al., 2007). Total purine excretion was estimated by the sum of allantoin excreted in urine and milk plus uric acid excreted in the urine. Absorbed microbial purines (mmol d<sup>-1</sup>)

were estimated by the equation proposed by Verbic et al. (1990). The intestinal flow of microbial nitrogen (g d<sup>-1</sup>) was estimated from the equation by Chen and Gomes (1992).

To determine the economic viability of DBG, we considered the amount paid to the farmer for a liter of milk (US\$ 0.43 per L) and prices related to the ration ingredients practiced in Paraná in November, 2014: corn silage, US\$ 0.11 per kg of DM; Tifton hay, US\$ 0.17 per kg of DM; ground corn, US\$ 0.21 kg per of DM; soybean meal, US\$ 0.50 per kg of DM; DBG, US\$ 0.17 per kg of DM; mineral supplement, US\$ 1.02 per kg of DM; and inorganic phosphate, US\$ 0.84 per kg of DM. The value of US\$ 1.00 in the economic analysis period was equivalent to R\$ 2.557. The economic approach only considered the feed cost due to research purposes.

The ration cost per kilogram was calculated from the chemical composition of each ingredient in the diet and its cost. The feed cost was obtained by multiplying the ration cost by the animal's average intake of each treatment diet. Similarly, the gross revenue obtained for each treatment was calculated with the milk liter value and the milk production. The gross margin was calculated as the difference between gross revenue and feed cost. The equilibrium point was calculated by the ration cost divided by the milk liter value. The equilibrium point shows the exact production volume when there is zero return, i.e., when the gross revenue is equal to the feed cost.

The data were analyzed as a 5 × 5 Latin square design using the MIXED procedure of SAS (Statistical Analysis System, version 9.2.). The mathematical model used was:

$$\gamma_{ijk} = \mu + \tau_i + p_j + c_k + \varepsilon_{ijk},$$

in which  $\gamma_{ijk}$  = observation,  $\mu$  = population mean,  $\tau_i$  = diet effect ( $i = 1$  to 5),  $p_j$  = period effect ( $j = 1$  to 5),  $c_k$  = cow effect ( $k = 1$  to 5), and  $\varepsilon_{ijk}$  = residual error. Various covariance structures of errors were fitted. The first-order autoregressive structure (AR(1)) was selected based on the lowest Bayesian information criterion. The effects of DBG levels were evaluated by orthogonal polynomials testing linear and quadratic effects. Significance was declared at  $P \leq 0.05$ .

## Results

The use of DBG instead of soybean meal had a linear effect, decreasing the body weight, DM, OM, CP, and NFC intake in kg d<sup>-1</sup> ( $P < 0.05$ ) (Table 2). Dry matter intake g kg<sup>-1</sup> d<sup>-1</sup> and g kg<sup>-0.75</sup> d<sup>-1</sup> had a quadratic effect ( $P < 0.05$ ). The maximum intake was estimated using the replacement levels of 22.5% and 18.4% of DBG, respectively. Ether extract intake in kg d<sup>-1</sup>, NDF intake in kg d<sup>-1</sup>, g kg<sup>-1</sup> d<sup>-1</sup>

and  $\text{g kg}^{-0.75} \text{d}^{-1}$  showed a positive linear effect with the increase of DBG in the diet ( $P < 0.05$ ). The ADF intake and metabolizable energy did not show any effect by the replacement levels ( $P > 0.05$ ).

Apparent DM and OM digestibilities were linearly increased ( $P < 0.05$ ) with DBG levels (Table 3). A positive linear effect was also obtained for EE and NDF digestibilities with an increasing level of DBG in the concentrate ( $P < 0.05$ ). The CP digestibility showed a quadratic effect with minimal digestibility estimated when there was 11.2% of DBG in

the concentrate replacing soybean meal ( $P < 0.05$ ). The ADF and NFC digestibility was not affected ( $P > 0.05$ ) by the treatments (Table 3).

Milk production and fat corrected milk showed quadratic effect ( $P < 0.05$ ) (Table 4). Milk production efficiency increased linearly ( $P < 0.05$ ). Milk fat, protein, and total solids expressed in  $\text{g kg}^{-1}$  showed a decreasing linear effect ( $P < 0.05$ ). Milk fat, lactose, and total solids in  $\text{kg d}^{-1}$  showed a quadratic effect ( $P < 0.05$ ) with the production peak estimated at 23.7%, 23.7%, and 31.5% of

Table 2 - Body weight, daily dry matter, and nutrient intake of Holstein cows fed diets containing different levels of dried brewers' grains replacing soybean meal

Variable	Replacement level (%)					P-value		SEM
	0	25	50	75	100	Linear	Quadratic	
BW (kg) <sup>1</sup>	623.40	623.60	614.40	621.80	599.40	0.002	0.110	5.91
DMI (kg d <sup>-1</sup> ) <sup>2</sup>	19.68	19.37	18.49	18.30	15.28	<0.001	0.016	0.75
DMI (g kg <sup>-1</sup> d <sup>-1</sup> ) <sup>3</sup>	31.76	31.37	30.39	29.64	24.21	<0.001	0.007	1.17
DMI (g kg <sup>-0.75</sup> d <sup>-1</sup> ) <sup>4</sup>	158.41	156.34	150.91	147.70	121.32	<0.001	0.008	5.84
OMI (kg d <sup>-1</sup> ) <sup>5</sup>	18.57	18.25	17.42	17.26	14.40	<0.001	0.014	0.70
EI (kg d <sup>-1</sup> ) <sup>6</sup>	0.41	0.47	0.51	0.50	0.60	<0.001	0.806	0.03
CPI (kg d <sup>-1</sup> ) <sup>7</sup>	2.88	2.81	2.72	2.68	2.29	<0.001	0.101	0.13
NDFI (kg d <sup>-1</sup> ) <sup>8</sup>	7.61	7.92	8.76	9.08	9.04	0.009	0.571	0.62
NDFI (g kg <sup>-1</sup> d <sup>-1</sup> ) <sup>9</sup>	12.27	12.82	14.38	14.69	14.33	0.013	0.304	0.98
NDFI (g kg <sup>-0.75</sup> d <sup>-1</sup> ) <sup>10</sup>	61.22	63.90	71.43	73.27	71.79	0.012	0.359	4.90
ADFI (kg d <sup>-1</sup> ) <sup>11</sup>	3.79	3.79	3.81	3.97	3.77	0.695	0.749	0.19
NFCI (kg d <sup>-1</sup> ) <sup>12</sup>	8.50	7.90	6.99	6.10	3.69	<0.001	<0.001	0.30
MEI (Mcal d <sup>-1</sup> ) <sup>13</sup>	39.26	40.60	41.93	41.02	37.16	0.471	0.066	2.38

BW - body weight; DMI - dry matter intake; OMI - organic matter intake; EI - ether extract intake; CPI - crude protein intake; NDFI - neutral detergent fiber intake; ADFI - acid detergent fiber intake; NFCI - non-fibrous carbohydrate intake; MEI - metabolizable energy intake; SEM - standard error of the mean.

$$^1 \hat{Y} = 626.4800 - 0.1992x.$$

$$^2 \hat{Y} = 20.1971 - 0.0395x.$$

$$^3 \hat{Y} = 31.4326 + 0.0453x - 0.0011x^2.$$

$$^4 \hat{Y} = 156.8702 + 0.1989x - 0.0054x^2.$$

$$^5 \hat{Y} = 19.0440 - 0.0373x.$$

$$^6 \hat{Y} = 0.4169 + 0.0016x.$$

$$^7 \hat{Y} = 2.9404 - 0.0053x.$$

$$^8 \hat{Y} = 7.6760 + 0.0161x.$$

$$^9 \hat{Y} = 12.5044 + 0.0239x.$$

$$^{10} \hat{Y} = 62.2232 + 0.1220x.$$

$$^{11} \hat{Y} = 3.826.$$

$$^{12} \hat{Y} = 8.9202 - 0.0456x.$$

$$^{13} \hat{Y} = 39.994.$$

Table 3 - Apparent dry matter and nutrient digestibility (g kg<sup>-1</sup> of dry matter) of Holstein cow diets containing different levels of dried brewers' grains replacing soybean meal

Variable	Replacement level (%)					P-value		SEM
	0	25	50	75	100	Linear	Quadratic	
DMD <sup>1</sup>	630.75	643.85	636.26	652.80	667.79	0.049	0.563	17.02
OMD <sup>2</sup>	654.06	665.88	658.83	675.98	691.33	0.036	0.468	15.98
EED <sup>3</sup>	551.73	630.02	684.78	662.35	786.02	<0.001	0.824	22.50
CPD <sup>4</sup>	597.74	617.65	617.95	649.64	713.08	<0.001	0.026	17.51
NDFD <sup>5</sup>	538.98	573.38	596.23	622.96	663.41	<0.001	0.646	18.04
ADFD <sup>6</sup>	478.09	510.62	476.94	471.50	514.56	0.656	0.593	35.15
NFCD <sup>7</sup>	762.69	764.66	765.65	760.03	748.91	0.521	0.431	20.08

DMD - dry matter digestibility; OMD - organic matter digestibility; EED - ether extract digestibility; CPD - crude protein digestibility; NDFD - neutral detergent fiber digestibility; ADFD - acid detergent fiber digestibility; NFCD - non-fibrous carbohydrate digestibility; SEM - standard error of the mean.

$$^1 \hat{Y} = 629.6845 + 0.3321x.$$

$$^2 \hat{Y} = 652.2879 + 0.3386x.$$

$$^3 \hat{Y} = 562.7931 + 2.0037x.$$

$$^4 \hat{Y} = 603.5974 - 0.3029x + 0.0135x^2.$$

$$^5 \hat{Y} = 539.3026 + 1.1936x.$$

$$^6 \hat{Y} = 490.342.$$

$$^7 \hat{Y} = 760.388.$$

DBG, respectively. Milk urea nitrogen concentrations were not affected by DBG levels ( $P>0.05$ ).

Allantoin and urine uric acid showed linear and quadratic effects ( $P<0.05$ ) respectively, with the increase of DBG in the concentrate, while milk allantoin ( $P<0.05$ ) showed no effect (Table 5). As a result of the decline in urine allantoin concentrations, total purines also showed

a linear decrease ( $P<0.05$ ). Microbial purines absorbed in  $\text{mmol d}^{-1}$ , N-microbial production, and microbial CP expressed in  $\text{g d}^{-1}$  showed a linear effect with different DBG levels in the concentrate ( $P<0.05$ ). However, efficiency of microbial protein synthesis ( $\text{g of CP-mic kg}^{-1} \text{d}^{-1}$  of total digestible nutrients) did not differ ( $P<0.05$ ) between the different DBG levels (Table 5).

Table 4 - Milk production, composition, and urea nitrogen of Holstein cows fed diets containing different levels of dried brewers' grains replacing soybean meal

Variable	Replacement level (%)					P-value		SEM
	0	25	50	75	100	Linear	Quadratic	
Milk production ( $\text{kg d}^{-1}$ ) <sup>1</sup>	24.40	24.25	25.00	24.40	22.40	0.0206	0.047	0.96
3.5% Fat corrected milk ( $\text{kg d}^{-1}$ ) <sup>2</sup>	25.31	25.34	25.21	24.64	20.92	<0.001	0.004	0.83
Milk production efficiency <sup>3</sup>	1.26	1.29	1.36	1.34	1.47	0.007	0.746	0.05
Fat ( $\text{g kg}^{-1}$ ) <sup>4</sup>	36.97	36.04	35.46	36.01	31.52	0.002	0.072	1.08
Fat ( $\text{kg d}^{-1}$ ) <sup>5</sup>	0.91	0.90	0.88	0.87	0.70	<0.001	0.001	0.03
Protein ( $\text{g kg}^{-1}$ ) <sup>6</sup>	31.92	31.54	30.52	31.13	29.52	0.006	0.713	0.65
Protein ( $\text{kg d}^{-1}$ ) <sup>7</sup>	0.78	0.79	0.77	0.75	0.65	0.005	0.107	0.04
Lactose ( $\text{g kg}^{-1}$ ) <sup>8</sup>	44.80	45.23	45.41	45.43	45.34	0.364	0.341	0.32
Lactose ( $\text{kg d}^{-1}$ ) <sup>9</sup>	1.10	1.13	1.15	1.10	1.01	0.022	0.038	0.04
Total solids ( $\text{g kg}^{-1}$ ) <sup>10</sup>	123.58	122.71	121.25	122.34	116.14	<0.001	0.065	1.33
Total solids ( $\text{kg d}^{-1}$ ) <sup>11</sup>	3.03	3.07	3.05	2.96	2.58	0.002	0.016	0.12
Milk urea nitrogen ( $\text{mg dL}^{-1}$ ) <sup>12</sup>	13.31	13.30	13.66	14.67	14.31	0.205	0.844	1.14

SEM - standard error of the mean.

$$^1 \hat{Y} = 24.3314 + 0.0479x - 0.0007x^2.$$

$$^2 \hat{Y} = 25.0453 + 0.0528x - 0.0009x^2.$$

$$^3 \hat{Y} = 1.2484 + 0.0019x.$$

$$^4 \hat{Y} = 37.3860 - 0.0437x.$$

$$^5 \hat{Y} = 0.8926 + 0.0019x - 0.00004x^2.$$

$$^6 \hat{Y} = 31.9680 - 0.0208x.$$

$$^7 \hat{Y} = 0.8096 - 0.0012x.$$

$$^8 \hat{Y} = 45.242.$$

$$^9 \hat{Y} = 1.0940 + 0.0019x - 0.00004x^2.$$

$$^{10} \hat{Y} = 124.2540 - 0.0610x.$$

$$^{11} \hat{Y} = 3.0091 + 0.0063x - 0.0001x^2.$$

$$^{12} \hat{Y} = 13.820.$$

Table 5 - Purine derivative excretions and microbial synthesis of lactating Holstein cows fed diets containing different levels of dried brewers' grains replacing soybean meal

Variable	Replacement level (%)					P-value		SEM
	0	25	50	75	100	Linear	Quadratic	
Purine derivative excretions ( $\text{mmol d}^{-1}$ )								
Urine allantoin <sup>1</sup>	315.99	306.51	304.02	298.83	264.96	0.024	0.324	17.18
Milk allantoin <sup>2</sup>	31.49	37.29	27.75	34.93	28.99	0.743	0.433	5.30
Uric acid <sup>3</sup>	32.01	32.75	31.00	25.94	19.87	0.004	0.004	2.49
Total purine <sup>4</sup>	379.50	376.55	362.77	359.70	313.82	0.033	0.150	20.03
Microbial purines ( $\text{mmol d}^{-1}$ )								
Absorbed purines <sup>5</sup>	389.97	386.48	370.91	366.79	314.35	0.038	0.163	23.77
Microbial production ( $\text{g d}^{-1}$ )								
Microbial N <sup>6</sup>	283.53	280.99	269.67	266.68	228.55	0.038	0.163	17.28
Microbial CP <sup>7</sup>	1772.06	1756.19	1685.41	1666.72	1428.42	0.038	0.163	108.02
Microbial CP $\text{kg}^{-1}$ TDN <sup>8</sup>	163.44	156.90	152.59	151.60	144.78	0.070	0.833	10.49

CP - crude protein; TDN - total digestible nutrients; SEM - standard error of the mean.

$$^1 \hat{Y} = 320.0112 - 0.4389x.$$

$$^2 \hat{Y} = 32.090.$$

$$^3 \hat{Y} = 32.1171 + 0.0691x - 0.0019x^2.$$

$$^4 \hat{Y} = 388.1098 - 0.5928x.$$

$$^5 \hat{Y} = 399.8893 - 0.6838x.$$

$$^6 \hat{Y} = 290.7376 - 0.4971x.$$

$$^7 \hat{Y} = 1817.1116 - 3.1070x.$$

$$^8 \hat{Y} = 153.862.$$

The economic viability (Table 6) shows the gross margin of greater value and the lowest equilibrium point that occurred to treatment of 100% DBG.

## Discussion

Body weight decreased with the increase in the level of DBG because the diets with DBG had lower energy density. Animals fed diets with higher levels of DBG showed reduction in dry matter intake and mobilized body reserves to maintain their milk production. Animals fed diets containing 100% DBG consumed 4.4 kg DM per day less than the control treatment (Table 2). Using DBG above these levels may have limited the DM intake due to its high NDF (663 g kg<sup>-1</sup> DM), which led to intake regulation by physical filling. Davis et al. (1983) also observed that DM intake was reduced when 30 or 40% of DM from wet brewers' grain was used in the feed.

Ether extract and NDF intake increased along with DBG level in the diet (Table 1). Geron et al. (2010) also showed an increase in EE intake (g kg<sup>-0.75</sup> d<sup>-1</sup>) of around 8% when lactating cows were fed 15% brewer's grain silage compared with cows fed a diet with 0% brewers' grain silage. Mertens (1992) suggested that NDF intake of an animal up to 12 g kg<sup>-1</sup> d<sup>-1</sup> of body weight is the main mechanism of physical regulation of DM intake. Thus the NDF intake observed for the treatments with 50%, 75%, and 100% DBG was 14.4, 14.7, and 14.3 g kg<sup>-1</sup> d<sup>-1</sup> of body weight, respectively, confirming the fact that high levels of DBG in the concentrate limited feed intake.

Organic matter, CP, and NFC intake decreased as a result of a reduction in DM intake. In the case of NFC, this reduction also occurred due to the low NFC concentration in diets with greater amounts of DBG.

Apparent DM and OM digestibility increased with DBG levels (Table 3), which may be related to an increased feed retention time in the rumen due to higher NDF in diets with DBG. Doreau et al. (2003) reported that the main cause of digestibility variation in the diet is the retention time of particles in the rumen (i.e., a lower DM intake leads

to a reduction in passage rate and a consequent increase in its apparent digestibility). The increase in EE and NDF digestibility can be attributed to a slower passage rate and a greater concentration of these nutrients in the diets with higher DBG levels. The greater digestibility of NDF can also be associated with an improved ruminal condition, since the use of fibrous byproducts increases rumen motility and maintains a higher pH, which favors the fibrolytic microorganisms.

The CP digestibility was high because, although the brewers' grains present low ruminal degradability, high intestinal digestibility (Santos et al., 1984) leads to a high total apparent digestibility. This was also reported by Geron et al. (2007), who found larger amounts of rumen undegradable protein but degradable in the small intestine with fermented beer residue (394 g kg<sup>-1</sup> CP) compared with soybean meal (187 g kg<sup>-1</sup> CP).

Milk production and fat corrected milk showed quadratic effect with maximum production estimated at 34.2% and 29.3% of DBG replacing soybean meal, respectively. This corroborates with the results of Imaizumi et al. (2015), who demonstrated higher fat corrected milk with 20% of wet brewers' grains in the diet DM. This estimated maximum production may have occurred due to better nutrient synchronization and the presence of essential amino acids for milk synthesis such as lysine and methionine. According to the NRC (2001), DBG contains 10.4 g of lysine per 100 g of CP and 4.3 g of methionine per 100 g of CP, while soybean meal has 13.9 g of lysine per 100 g of CP and 3.2 g of methionine per 100 g of CP.

Milk production efficiency increased along with a reduction in DM intake with increased levels of DBG. However, it should be noted that this increase in milk production efficiency disregards the reduction in body weight observed in cows consuming larger amounts of DBG, which, in the long term, may adversely affect production and reproduction. Davis et al. (1983) also assessed feed efficiency and obtained an apparent increase in this ratio due to body weight loss along with increasing wet brewers' grain levels in the diet.

Table 6 - Economic analysis of the use of increasing levels of dried brewers' grains to replace the soybean meal concentrate in the diet of lactating Holstein cows

Variable	Replacement level (%)				
	0	25	50	75	100
Ration cost (US\$ kg <sup>-1</sup> dry matter)	0.22	0.22	0.21	0.20	0.18
Feed cost (US\$ d <sup>-1</sup> )	4.39	4.24	3.92	3.69	2.75
Gross revenue (US\$ d <sup>-1</sup> )	10.50	10.43	10.75	10.50	9.64
Gross margin (US\$ d <sup>-1</sup> )	6.10	6.19	6.83	6.81	6.89
Equilibrium point (kg of milk d <sup>-1</sup> )	10.61	9.85	9.11	8.58	6.39

The reduction in milk fat may have occurred as a result of the reduction in DM intake and also because DBG is a rich source of polyunsaturated fatty acids (Geron et al., 2007), which can promote milk fat depression. For protein, each 1% of soybean meal replaced by DBG in the concentrate lead to a reduction of 20.8 mg kg<sup>-1</sup> and 1.2 g d<sup>-1</sup> of milk protein. This differed from West et al. (1994) and Cozzi and Polan (1994), who found an increase in the milk protein content of cows fed brewers' grains in both wet and dry forms.

Concentrations of milk urea nitrogen were not affected, although DBG and soybean meal showed some differences regarding their protein use by animals. After ruminal incubation for 16 h, Geron et al. (2007) noted values of rumen degradable protein of 436 g kg<sup>-1</sup> of DM for wet brewers' grains and 797 g kg<sup>-1</sup> of DM for soybean meal, while the rumen undegradable protein was 564 g kg<sup>-1</sup> CP for wet brewers' grains and 204 g kg<sup>-1</sup> CP for soybean meal. As the diets in the present study were isoproteic, these factors were not enough to characterize differences in urea nitrogen concentrations. Additionally, the milk urea nitrogen values obtained are near the range between 10 and 14 mg dL<sup>-1</sup>, which was described by Powell et al. (2011) as the ideal, since it demonstrates adequate synchronization between proteins and carbohydrates in the diet.

N-microbial production and microbial CP in g d<sup>-1</sup> decreased with increasing DBG levels. Santos and Pedroso (2011) emphasized that the reduction in microbial production may be related to lower carbohydrate availability and lower dietary protein degradability for protein synthesis. This can explain the results of the current study because, according to Santos et al. (1984), brewers' grains generate a smaller contribution of ammonia N due to lower ruminal degradable CP present in this feed. Additionally, diets with higher DBG levels also had lower corn proportions (Table 1), which reduced the availability of rapidly fermenting carbohydrates, limiting microbial growth and protein synthesis. However, microbial efficiency was not affected by treatments regardless of the difference in rumen degradable protein content of diets (628.5 g of rumen degradable protein kg<sup>-1</sup> of CP for diet containing 0% DBG and 575.8 g of rumen degradable protein kg<sup>-1</sup> of CP for diet containing 100% DBG). This effect may be due to the lower DM intake and consequently higher retention time in the rumen of diets with higher DBG, which probably allowed a more efficient protein degradation of this diet (Table 5). Microbial efficiency for all levels of DBG was greater than the 130 g kg<sup>-1</sup> of total digestible nutrients recommended by the NRC (2001).

The economic viability showed that the total replacement of soybean meal by DBG reduces the cost

in US\$ by \$0.04 per kg of dietary DM, due to the 64.6% lower cost of this waste compared with the soybean meal (Table 6). The average daily cost of feed also decreased with increasing DBG levels due to the reduction in the cost per kg of dietary DM and to the decline in DM intake.

The gross margin of greater value and the lowest equilibrium point occurred with 100% DBG replacement, making this the most advantageous diet over the short term. However, treatment with 75% DBG showed a gross margin similar to treatment with 100% DBG, the latter causing a marked reduction in DM intake as well as reduced body weight of the animals.

## Conclusions

Dried brewers' grains can be used instead of soybean meal in dairy cow feed as it provides improvements in digestibility, milk production efficiency, and economic return to the producer without affecting microbial efficiency. However, due to the decrease in dry matter intake, body weight, milk production, milk fat, and milk protein, the use of DBG is not recommended to replace more than 75% of soybean meal.

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