Growth performance and apparent total tract nutrient digestibility of limit-fed diets containing wet brewer's grains to Holstein heifers

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ABSTRACT: The objective of this study was to evaluate the growth performance and apparent total tract nutrient digestibility of Holstein heifers limit-fed diets containing different amounts of wet brewer's grains (WBG). A 12-wk randomized complete block study was conducted using 30 yearling Holstein heifers $[378 \pm 27 d \text{ of age},$ and body weight (BW) of 357.8 ± 27.6 kg (mean \pm SD)]. Treatments were 0%, 10% and 20% of WBG on a dry matter (DM) basis and diets were formulated to be limit-fed for dry matter intake (DMI) at 2.35% of BW and provided 15% crude protein (CP) and 2.27 Mcal metabolizable energy/ kg of DM. Dry matter intake was recorded daily, while BW and skeletal measurements were measured every 2 wk. During week 12, fecal samples were collected directly from the rectum over four consecutive days and composited by heifer to determine apparent total tract nutrient digestibility using acid detergent insoluble ash as a marker. Data were analyzed using the MIXED procedure of SAS. Dry matter intakes, BW, and average daily gain were not different among treatments (P = 0.2, P = 0.4, and P = 0.6, respectively). Dry matter

intakes ranged from 8.6 to 9.0 kg/d. Average BW were 404.4, 411.5, and 409.3 kg for heifers fed the 0%, 10%, and 20% WBG diets, respectively. Average daily gains were 1.03, 1.04, and 0.96 kg/d for heifers fed the 0%, 10%, and 20% WBG diets respectively. Skeletal measurements and body condition scores (BCS) were not different among treatments except for the change in heart girth (P < 0.01) and initial BCS (P < 0.01). Apparent total tract digestibilities of DM, organic matter, CP, fat, and hemicellulose were greater or tended to be greater in heifers fed 0% and 20% WBG treatments than heifers fed 10 % WBG (P = 0.04, P = 0.04, P = 0.06, P = 0.06, and P = 0.01, respectively). Neutral detergent fiber, acid detergent fiber, and fat digestibilities were similar among treatments (P = 0.2, P = 0.3, and P = 0.3, respectively). During the digestibility phase, DMI tended to be greater (P = 0.08) for the 10% WBG treatment. These results demonstrate that limit-feeding heifers with diets containing up to 20% WBG could replace soybean- and corn-based concentrates in diets without adverse consequences to the heifer growth performance.

Key words: dairy heifer, growth performance, wet brewer's grain

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INTRODUCTION

Raising dairy heifers from birth to the first parturition is an expensive cost to producers because of relatively low feed efficiency and absence of income until the onset of lactation (Zanton and Heinrichs, 2009). Due to this, one of the priorities of dairy producers is to decrease the cost of production through strategies that optimize the growth of heifers without sacrificing productivity (Tozer and Heinrichs, 2001). Moreover, the cost of energy and protein feeds especially corn and soybean meal can be expensive; because of this, the substitution of those ingredients for alternative feed sources is of great importance. The use of agroindustrial byproducts (AIBP) in cattle feeding has been investigated (Ajila et al., 2012), and AIBP represents a vast potential to reduce the cost associated with heifer production. Among various AIBP, wet brewer's grains (WBG), which are byproducts of the brewing industry, have gained popularity because of their high nutritional value, low cost, and availability in various regions of the country.

In terms of nutrition, the importance of WBG as a feed supplement may be attributed to its high protein content and low rumen solubility (Homm et al., 2008). Wet brewer's grains are characterized by having approximately 28% crude protein (CP), 47.1% neutral detergent fiber (NDF), and 5.2% ether extract, as well as high moisture content of 75–80% [National Research Council (NRC), 2001]. Compared to the energy content of ground corn (3.08 Mcal/kg of dry matter [DM]), WBG contains about 2.60 Mcal/kg DM (Frasson et al., 2018).

Wet brewer's grains have been successfully used in feedlot heifers (Homm et al., 2008), and data indicated that feeding 15% to 45% WBG on a DM basis in feedlot diets supports performance and carcass characteristics similar to or greater than that in cattle fed a typical high-moisture corn finishing diet. Research with lactating cows indicated that supplementing cows with WBG (15% and 30% of the diet) resulted in greater milk yield, fat, and protein contents than cows supplemented with soybean meal (Murdock et al., 1981; Faccenda et al., 2017). Similar results were observed when corn silage was substituted with WBG at 9% (Belibasakis and Tsirogogianni, 1996). Greater milk yields and milk components are influenced by a better synchronization and presence of essential amino acids for milk synthesis, such as lysine and methionine that are high in WBG (Faccenda et al., 2017). However, no research regarding the effect of feeding WBG to growing dairy heifers on their growth performance and nutrient utilization is available.

Another strategy to reduce the cost of raising dairy heifers is to use limit-feeding in which nutrient-dense diets are fed to meet but not to exceed nutrient requirements and reduce dry matter intake (DMI), and it has the potential to increase nutrient digestibility while maintaining growth performance (Hoffman et al., 2007; Zanton and Heinrichs, 2009). Therefore, the objective of this experiment was to evaluate the effect of feeding diets containing different amounts of WBG to replace corn- and soybean-based concentrates in a modified limit-fed heifer diet on the growth performance and apparent total tract nutrient digestibility of dairy heifers. The hypothesis was that WBG could partially or fully replace conventional concentrate feeds (corn based or soybean based) in dairy heifer's diets and provide similar growth performance and apparent total tract nutrient digestibility.

MATERIAL AND METHODS

Experimental Design

This experiment was reviewed and approved by the University of New Hampshire Animal Care and Use Committee (protocol # 170603). Thirty yearling Holstein heifers with a mean age of $378 \pm$ 27 d and BW of 357.8 ± 27.6 kg (mean \pm SD) were blocked by birth date and randomly assigned to one of three treatments (n = 10) in a randomized complete block design. Heifers entered the study when they were about 12 mo of age. There were 10 blocks of heifers and each block of heifers remained on the study for 12 wk. The experiment (for all 10 blocks of heifers) was completed in 10 mo from March 2018 to January 2019 because of the staggered starting time. Since WBG are readily consumed, treatments were fed immediately without any adaptation period.

Treatment diets were formulated on a DM basis: 1) a control (0% WBG), 2) a diet containing 10% WBG, and 3) a diet containing 20% WBG. Diets were formulated to be isonitrogenous (15% CP) and isocaloric [2.27 Mcal metabolizable energy (ME)/kg of DM] using the NRC (2001) software to provide 0.9 kg/d of average daily gain (ADG). With the fact that WBG is high in protein content, and WBG is a potential fiber source, it was challenging to keep the ratio of grass silage:corn silage and the ratio of the roughage:concentrate constant across treatments. Therefore, as WBG was increased, grass silage, corn silage, and mineral mix were changed within the diets along with the corn and soybean meal-based energy and protein mixes (Table 2). The content of corn silage and grass silage in the diets varied by no more than 5% and the mineral mix varied by <0.65%. This may have confounded some of the results. However, it was attempted to

keep dietary nutrient content consistent across treatments.

Animal Care and Feeding

Heifers were group-housed in a naturally ventilated free-stall barn bedded with mattresses. Because heifers did not enter the study at the same time, one pen $(15.9 \times 4.8 \text{ m})$ having the capacity to host 16 heifers was used. Heifers were fed once daily at 0900 hours using the Calan gate feeding system (American Calan Inc., Northwood, NH) in individual feed tubs to allow for feed intakes and refusals (if any). The feed was mixed and distributed using a motorized feeding vehicle (Data Ranger, American Calan Inc. Northwood, NH). Before feeding, orts (if any) were collected from individual feed tubs and recorded. Samples of the total mixed ration (TMR) for each treatment were taken every day and stored at -20 °C for future nutrient analysis. Rations were limit-fed to 2.35% of BW (DM basis) and adjusted or corrected for nutrient content every 2 wk after BW measurements or nutrient analysis of the feed ingredients.

Heifers had ad libitum access to water through automatic refilling water troughs. The health status of heifers was observed every day according to the routine management of the research center.

Animal Measurements

Every Tuesday before feeding, throughout the study, BW and skeletal measurements were recorded on weeks 0, 2, 4, 6, 8, 10, and 12, resulting in a total of seven measurements. Heifer measurements that were taken on week 0 (initial measurements) served as covariates in our statistical model. Skeletal measurements included body length (distance between the points of the shoulder and rump), hip height (length from the base of the rear feet to the hook bones), withers height (distance from the base of the front feet to the top of the withers), heart girth (circumference of the chest), paunch girth (circumference of the belly), hip width (distance between the points of hook bones), and body conditions score (BCS) based on the scale described by Wildman et al. (1982) with 1 = emaciated and 5 = obese.

For BW, heifers were weighed on a platform scale (Cardinal, Northeast Scale Co. Inc., Hooksett, NH). Hip and withers heights were measured using a sliding height stick with a bubble level. Heart and paunch girth, as well as body length, were measured using a weight tape (Coburn Co, Inc., Whitewater, WI).

Feed and Fecal Samples Analysis

Frozen samples of the TMR and orts (orts were rare and only occurred during times of elevated environmental temperatures) were thawed and samples from four consecutive weeks were composited on an as-fed basis for each treatment as a monthly composite. Composites of samples were then dried in duplicate for 48 h in a forced-air oven (Binder, Bohemia, NY) to dry at 55 °C and ground through a 1-mm screen Wiley mill (model 3, Arthur H. Thomas, Philadelphia, PA). All samples were sent to a commercial laboratory (Rock River Laboratory Inc., Watertown, WI) for nutrient analysis. Samples were analyzed for NDF (method 6 in an Ankom Fiber Analyzer A2000 with α -amylase and Sodium sulfite, Ankom Technology; solutions as in Van Soest et al., 1991), acid detergent fiber (ADF) (method 5 in an Ankom Fiber Analyzer A2000, Ankom Technology, Fairpoint, NY; method 973.18; AOAC International, 1998). Hemicellulose was calculated as NDF% - ADF%. Nitrogen was analyzed via Dumas combustion (AOAC International 2002; method 968.06) on a Rapid N cube (Elementar Analysensystem, GmbH, Hanau Germany). Nitrogen was then multiplied by 6.25 to calculate the CP. Starch concentration was analyzed using a modified method of glucose analysis (Bach Knudsen, 1997) completed on a YSI 2700 select Biochemistry Analyzer (YSI Biochemistry analyzer, YSI Inc., Yellow Spring, OH) and fat was analyzed with ether extraction technique (method 2003.05; AOAC International, 2006). Ash content was determined by incinerating 1 g of sample for 8 h at 450 °C in a muffle furnace (AOAC International, 2002; method 942.05). Organic matter (OM) was calculated as OM = 100 - % ash. Mineral composition analysis included Ca, P, Mg, K, Na (AOAC International, 1998; method 985.01), and S (AOAC International, 1998; method 923.01).

Digestibility Measurements

Acid detergent insoluble ash (ADIA) was used as an internal digesta marker to estimate 24-h fecal excretion, and total tract nutrient digestibility was determined by calculations. Feeds, orts (if any) and feces were analyzed for ADF using the filter bag technique (method 5, Ankom Technology) followed by determination of acid-insoluble ash according to Van Keulen and Young (1977). The equation used to estimate apparent nutrient digestibility was:

Apparent total tract nutrient digestibility (%) =

$$100 - 100 \times \frac{\% \text{ AIA in feed} \times \% \text{ Nutrient in feces}}{\% \text{ AIA in feces} \times \% \text{ Nutrient in feed}}$$

Each of the 30 heifers underwent the digestibility phase from day 77 of study until day 84 of the study. Total mixed ration samples were collected on days 77–81. Individual orts (if any) were collected on days 78–82. Fecal samples were collected on days 80–84. Orts and TMR samples were then composited over the sampling days. Rectal fecal grab samples (~200 g/sample) were collected via gloved hand by collecting directly from the rectum for the last 4 d every 12 h to represent a 24-h period (day 80: 0900 and 2100 hours; day 81: 1200 and 0000 hours; day 82: 1500 and 0300 hours; day 83: 1800 through 0600 hours of day 84.

Fecal samples collected from day 80 to day 84 were combined to obtain a single composite and were frozen at -20 °C. Fecal samples were then thawed at room temperature and emptied into aluminum trays to be dried in a forced-air convection oven (Binder, Bohemia, NY) at 55 °C for at least 96 h until completely dried. The dried TMR, orts, and fecal samples were ground through a 1-mm screen using a Wiley mill (model 3, Arthur H. Thomas, Philadelphia, PA). Ground samples were sent to Rock River Laboratory (Watertown, WI) for nutrient analysis. Fecal samples were analyzed for CP, ADF, NDF, fat, OM, starch, ash, and ADIA as previously described for feed samples. Neutral detergent fiber intake (NDFI) and NDF/ BW, forage NDF intake, and forage NDF/BW were determined during the digestibility phase (week 12) to determine if there were differences due to varying amounts of NDF in the diets.

Statistical Analysis

Initial BW and skeletal measurements served as covariates for their respective variables of interest. Growth characteristics (BW, body length, hip and withers height, heart and paunch girth, hip width, and BCS) were analyzed as randomized complete block design with week as repeated measure and heifer (block) as subject using the Mixed procedure of SAS 9.4 (SAS Institute Inc., Cary, NC). The model included block, treatment, week, and treatment × week interaction according to the following model:

$$Y_{ijkl} = \mu + B_i + T_k + W_l + TW_{kl} + X_{ijk} + E_{ijkl}$$

where Y_{iikl} = the dependent variable; μ = the overall mean; $\mathbf{B}_i =$ the random effect of block i (i = 1, ..., 10); T_k = the fixed effect of the *k*th treatment (k = 0%, 10%, and 20% WBG); W_1 = the fixed effect of the *l*th week of the study (l = 2, 4, 6, 8, 10, and12); T_{wkl} = the fixed effect of interaction between the kth inclusion rate of WBG and the lth week; X_{iik} = the covariate measurement; and the E_{iikl} = the residual error. Compound symmetry, unstructured, first-order autoregressive, variance components, and Toeplitz covariance structures were determined and the structure with the lowest Bayesian information criterion was chosen. Degrees of freedom were calculated using the Kenward–Roger approximation option of the Mixed procedure. Single degree of freedom contrasts for linear and quadratic effects were determined. If the covariate analysis resulted in a probability >0.25, the covariate was removed from the model. Dry matter intake, ADG, and gain-to-feed ratio were analyzed similarly but

Initial and final BW, hip and withers heights, heart and paunch girth, hip width, and BCS were analyzed as randomized complete block design using the Mixed procedure of SAS 9.4 (SAS Institute Inc.) according to the following model:

without using any covariate.

$$\mathbf{Y}_{\mathrm{ijk}} = \mathbf{\mu} + \mathbf{B}_i + \mathbf{T}_j + \mathbf{X}_{\mathrm{ij}} + \mathbf{E}_{\mathrm{ijk}}$$

where Y_{ijk} = the dependent variable; μ = the overall mean; B_i = the random effect of block *i* (*i* = 1,, 10); T_j = the fixed effect of the *j*th inclusion rate of WBG (0%, 10%, and 20%); X_{ij} = the covariate measurement; and E_{ijk} = the residual error.

Changes over time for the growth parameters were calculated for the 2-wk intervals and averages analyzed using repeated measures in the Mixed procedure of SAS. The gain-to-feed ratio was calculated as the ratio of ADG to DMI for each treatment. Apparent total tract digestibility data was analyzed using the Mixed procedure of SAS 9.4 (SAS Institute, Inc.) according to the following model:

$$Y_{ijk} = \mu + B_i + T_j + E_{ijk}$$

where Y_{ijk} = the dependent variable; μ = the overall mean; B_i = the random effect of block i (i = 1, ..., 10); T_j = the fixed effect of the *j*th inclusion rate of WBG (0%, 10%, and 20% WBG); and E_{ijk} = the residual error.

For all variables, the least-square means for each treatment were reported. Significant treatment and interaction effects were declared at $P \le 0.05$ and a tendency was declared at $0.05 < P \le 0.10$.

RESULTS AND DISCUSSION

Feed Nutrients and Diet Composition

The average nutrient composition of major feed ingredients used in this study is presented in Table 1. Because of different entrance dates of different groups of heifers (10 blocks in total), the whole experiment that included all 30 heifers was completed in a period of 10 mo, and every block of three heifers stayed in the experiment for 12 wk. Therefore, the nutrient composition of feed ingredients is shown with SDs, indicating feed ingredient nutrient variation throughout the experiment.

The ingredient and nutrient composition of experimental diets is listed in Table 2. The dietary roughage content (grass silage and corn silage) was 80.7%, 78.79%, and 74.05% for diet 0%, 10%, and 20% WBG, respectively. The 20% WBG diet had a lower roughage content due to increased levels of WBG in diets. The contribution of WBG as a fiber source is not negligible (Firkins et al., 2002), which is why reducing the amount of roughage concentration has been considered with increasing levels of WBG in diets. The fact that WBG has high moisture and high protein content, as well as a high digestible fiber content makes it hard to make a balanced diet, especially with high-fiber diets, in which large amounts of WBG need to be consumed to provide adequate nutrients (Hersom, 2006). Hence, our diets exhibited changes in forage to concentrate ratio to provide similar nutrient content across treatments. The ratio of grass silage:corn silage was 1.58, 1.87, and 1.82 for 0%, 10%, and 20% WBG diet, respectively, which made NDF intake less for heifers fed 0% WBG and forage NDF intake less for heifers fed 20% WBG.

Energy mix was removed in the 10% and 20% WBG diet, while the protein mix was decreased by 46.6% in the 10% WBG and removed in the 20% WBG to keep protein and energy levels balanced across treatments (Table 2). The DM content of diets decreased as the inclusion of WBG increased due to the high moisture content of WBG. On average, our diets provided 2.29 ME Mcal/kg DM, which was similar to the targeted energy intake of 2.27 ME Mcal/kg DM.

Dietary OM was similar across treatments, while CP content was slightly lower (14.6% CP) than the target of 15% CP for the 0% WBG treatment. Crude protein content in 10% WBG diet was on target, while CP content was slightly greater (15.9% CP) in the 20% WBG diet. Wet brewer's grains varied in the DM and nutrient content throughout the study as it was purchased in different batches. The variation in the nutrient content of WBG suggests a constant nutrient analysis for better inclusion in diets because grain varieties and brewing processes have an important influence on nutrient composition of WBG (Robertson et al., 2010; Muthusamy, 2014). Moreover, small batches of WBG were used

		C 1	WDC	Energy	Protein
Item, % DM	Grass silage"	Corn silage	WBG	mix ^o	mix
DM	30.1 ± 3.5	31.9 ± 2.9	24.9 ± 2	87.9	88.5
СР	13.6 ± 2.7	7.4 ± 0.6	31.8 ± 0.3	7.8	53.2
NDF	58.9 ± 5.9	41.2 ± 3.4	46.3 ± 1.2	21.8	14.8
ADF	38.6 ± 3.7	24.8 ± 2	21 ± 0.03	12.5	9.3
Lignin	5.7 ± 0.6	3.1 ± 0.3	-	_	_
Starch	1.1 ± 0.3	32.1 ± 3	2.1 ± 0.04	_	6.4
NE _m (Mcal/kg DM)	1.4 ± 0.1	1.68 ± 0.06	1.75 ± 0.1	1.2	_
NE _g Mcal/kg DM)	0.76 ± 0.13	1.06 ± 0.05	1.12 ± 0.1	1.8	_
Fat	4.2 ± 0.5	3.0 ± 0.2	7.2 ± 0.05	3.5	2.6
Ash	8.4 ± 0.9	3.4 ± 0.4	4.7 ± 0.03	3.0	_
Ca	0.68 ± 0.06	0.19 ± 0.2	0.21 ± 0	0.40	0.55
Р	0.43 ± 0.18	0.22 ± 0.01	0.63 ± 0	0.22	0.83
K	2.6 ± 0.12	0.9 ± 0.4	0.07 ± 0	0.58	2.12
Mg	0.1 ± 0.02	0.1 ± 0.02	0.19 ± 0	0.16	0.40
S	0.2 ± 0.02	0.1 ± 0.0	0.32 ± 0	0.17	0.48

Table 1. Average nutrient composition of major ingredients used in the experiment

 $NE_m = Net energy of maintenance; Ne_g = Net energy of gain.$

^aGrass silage was primarily comprised of Orchard grass and Timothy.

^bContained 5% pellet mill molasses; 45.79% fine corn meal; 15.2% steam-flaked corn; and 33.99% whole beet pulp. Delivered in large batches and no variations in nutrient content.

Contained 7.28% distillers; 69.14% soybean meal; 21.83% canola; and 1.75% urea.

		Treatments ^a		
	0% WBG	10% WBG	20% WBG	
Ingredient composition, % of DM				
Energy mix ^b	2.17	0.00	0.00	
WBG	0.00	10.00	20.00	
Protein mix ^c	11.82	5.51	0.00	
Grass silage	49.44	51.36	47.84	
Corn silage	31.26	27.43	26.21	
Mineral mix ^d	5.32	5.69	5.95	
Nutrient composition, % DM				
DM	36.7 ± 2.2	34.3 ± 2.1	31.2 ± 1.1	
OM	88.1 ± 0.2	88.8 ± 0.1	88.9 ± 0.1	
СР	14.6 ± 0.2	15.2 ± 0.2	15.9 ± 0.3	
aNDF	48.3 ± 0.3	51.2 ± 0.4	49.2 ± 0.2	
ADF	29.7 ± 0.8	30.9 ± 0.6	29.6 ± 0.8	
Fat ^e	3.0 ± 0.0	3.2 ± 0.3	3.6 ± 0.0	
Starch	13.6 ± 0.0	12.1 ± 0.5	10.7 ± 0.3	
NFC	30.5 ± 0.6	27.0 ± 0.4	26.9 ± 0.1	
ME ^g , Mcal/kg DM	2.29 ± 0.3	2.29 ± 0.3	2.29 ± 0.4	
Ash	11.8 ± 0.1	11.2 ± 0.1	11.1 ± 0.1	
Ca	1.09 ± 0.00	1.01 ± 0.05	1.09 ± 0.02	
Р	0.78 ± 0.01	0.74 ± 0.00	0.80 ± 0.00	
K	1.86 ± 0.05	1.69 ± 0.03	1.46 ± 0.01	
Mg	0.37 ± 0.00	0.35 ± 0.00	0.35 ± 0.00	
Cl	1.26 ± 0.00	1.13 ± 0.00	1.19 ± 0.00	
S	0.32 ± 0.00	0.30 ± 0.00	0.33 ± 0.00	

Table 2. Ingredient and nutrient composition (% DM) of experimental diets containing 0%, 10%, or 20% WBG limit-fed to yearling heifers

aNDF, α-amylase NDF.

^aFormulated according to the NRC (2001).

^bContained 5% pellet mill molasses; 45.79% fine corn meal; 15.2% steam-flaked corn; and 33.99% whole beet pulp.

^eContained 7.28% distillers; 69.14% soybean meal; 21.83% canola; and 1.75% urea.

^dContained 19.05% Ca; 6.01 % P; 3.51% Mg; 20.00% Salt; 7.80% Na; 0. 26 % Fe; 0.26% Zn; 0.26 % Mn; 12.30% Cl; 602 mg/kg Cu; 15.0 mg/kg Co; 25.09 mg/kg Se; and 15.00 mg/kg I; 267,800 IU/kg vitamin A; 111,071 IU/kg vitamin D; and 2,207 IU/kg vitamin E.

^eEther extract.

 $\frac{100}{100}$ non-fiber carbohydrate (NFC) = 100 - [CP% + (NDF% - neutral detergent insoluble CP%) + fat% + ash%].

^gME calculated using NRC (2001).

in this study to avoid nutrient losses observed when large batches of WBG are delivered and spoiled by yeast and mold, especially when they are not preserved with commercial preservatives (Marston et al., 2009), commercial preservative or salt (Hatungimana and Erickson, 2019), microbial inoculants (Lilly et al., 1980; Schneider et al., 1995), or ensiling (Nishino et al., 2003). The high moisture content of WBG is the main limitation for long period storage and utilization in livestock feeding (Wang et al., 2014).

Average NDF and ADF were consistent in all diets, while starch and NFC were greater in the control diet (0% WBG) because of energy mix inclusion. Fat content was slightly greater in the 20% WBG diet due to the amount of WBG used in the diet and the high-fat content of WBG (7.2% fat).

Variation in the nutrient composition of the rations over time was observed; however, rations provided adequate nutrients to heifers and were comparable to typical diets fed to growing heifers. Phosphorous concentrations were elevated in diets due to the feeding rate of the mineral mix (limit-fed heifer diet) and a greater concentration of P in the WBG.

Heifer Growth Performance

Results for BW, DMI, ADG, and feed efficiency (gain:feed) are presented in Table 3. Treatment effect and treatment × week interactions were not found for those parameters. The initial BW of heifers and the average and final BW were not different among treatments. No treatment × week interaction was found for BW, which means that heifers

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Table 3. DMIs, BW, and gain-to-feed for dairy heifers limit-fed diets containing 0%, 10%, or 20% WBG

]	Treatment (WBG %)				P-values			
Item	0%	10%	20%	SEM	Trt	Week	Trt × Week	L	Q
Age $(day \pm SD)^a$	362.2 ± 22.8	372.0 ± 29.2	365.1 ± 24.1						
BW, kg									
Mean	404.4	411.5	409.3	3.77	0.41	< 0.01	0.19	0.38	0.32
Initial	359.4	357.4	352.2	5.4	0.64				
Final	440.2	445.7	438.2	5.2	0.58			0.80	0.31
DMI, kg	8.8	9.0	8.6	0.14	0.22	< 0.01	0.69	0.47	0.12
ADG	1.03	1.04	0.96	0.06	0.59	< 0.01	0.22	0.40	0.53
Gain:feed	0.12	0.12	0.11	0.006	0.79	< 0.01	0.14	0.56	0.81

aInitial age at the start of the study

consumed similar amounts of nutrients leading to similar growth rates. Overall, heifers fed the control diet 0%, 10%, and 20% WBG increased their BW by 22.5%, 24.7, and 24.4%, respectively.

Dry matter intakes were similar among treatments (P = 0.22) and a treatment × week interaction was not observed. However, DMI increased over time because heifers were limit-fed based on a percentage of BW. Similar DMI were reported by Hoffman and Armentano (1988), when feeding up to 25% dried brewer's grains to lactating cows. However, Davis et al. (1983) observed a decrease in DMI when 30% to 40% of WBG were fed to lactating dairy cows. In contrast to our results, Homm et al. (2008) observed greater DMI and ADG in beef heifers fed 15% and 30% WBG than beef heifers fed the control and 45% WBG.

Average daily gains of heifers (1.03, 1.04, and 0.96 kg/d for heifers fed 0%, 10%, and 20% WBG diets, respectively) were slightly greater than the targeted gain of 0.90 kg/d. However, ADG and feed efficiency were similar among treatments and decreased over time as nutrient requirements for maintenance increased with BW (Anderson et al., 2015b). Dietary energy intake was on average 2.29 Mcal/kg DM, which was close to the target of 2.27 Mcal/kg DM, and heifers had greater ADG than recommended, and we think that the NRC (2001) model we used to formulate diets may have overestimated the energy requirements of growing heifers or underestimated the energy provided by feed ingredients. Similar results were reported by Anderson et al. (2015a, 2015b) and Manthey et al. (2016) who limit-fed diets containing distiller's grains with different forage to concentrate ratios to growing dairy heifers. Our diets provided greater energy than expected, which would have probably caused greater ADG than was expected. Regardless of feeding strategy, Zanton and Heinrichs (2005) recommend dairy heifers to be fed energy to allow the ADG of 0.8 to 0.9 kg/d.

Skeletal measurements and BCS are presented in Table 4. Similar to BW results, heifers' skeletal measurements were not different among treatments and increased throughout the study. No difference was observed in change per day for the skeletal measurements except for hip width change that had a treatment \times week effect (P = 0.03). This effect was not expected as there were no treatment or treatment \times week effects on overall mean hip width. These findings suggest that the heifers were consuming adequate amounts of nutrients to promote growth throughout the study. Initial withers height and BCS were different (P = 0.02 and P = 0.01 respectively), heifers with the greatest wither's height being on the 10% WBG treatment. Initial paunch girth tended to be different (P = 0.10), and heifers fed the 20% WBG had the least paunch girth. However, there were no differences in final skeletal measurements indicating similar growth performance among treatments. Body weight gain per centimeter of hip height gain (kg/cm/d) was calculated to check if heifers were growing or fattening and those ratios were similar among treatments. No treatment or treatment × week effect was observed on BW gain per centimeter of hip height gain.

On average, heifers grew approximately 9.7 cm in body length, 6.8 cm withers height, and 5.8 cm in hip height throughout the experiment. Body condition scores slightly increased for all heifers but were similar across treatments. The similarity of frame growth agrees with findings from other research on limit-feeding when heifers have similar energy intakes(Zanton and Heinrichs 2007).

Dry Matter Intakes and Total Tract Nutrient Digestibility as Measured During Week 12

The DMI, NDFI, and forage NDFI, as well as DMI as a percentage of BW, NDFI as a percentage of BW, and forage NDF as a percentage of BW, and the total tract nutrient digestibility of diets measured during week 12 are presented in Table 5.

Table 4. Skeletal measurements for dairy heifers limit-fed diets containing 0%, 10%, or 20% WBG^a

	Trea	Treatments (WBG %)					P-value		
Item	0%	10%	20%	SEM	Trt	Week	Trt × Week	L^b	Qc
Withers height									
Mean, cm	137.4	137.1	136.8	0.36	0.40	< 0.01	0.37	0.18	0.92
Initial	132.3	135.1	131.4	1.3	0.02			0.51	0.01
Final	140.0	139.6	139.6	0.49	0.72			0.48	0.76
Change ^d , cm/d	0.09	0.08	0.08	0.01	0.16	0.15	0.87	0.25	0.42
Hip height									
Mean, cm	140.2	140.6	140.4	0.34	0.75	< 0.01	0.54	0.66	0.55
Initial	137.3	137.4	136.4	0.64	0.47			0.32	0.47
Final	142.6	143.2	142.6	0.49	0.56			0.98	0.29
Change ^d , cm/d	0.06	0.07	0.06	0.007	0.62	0.97	0.58	0.96	0.33
Body length									
Mean, cm	127.2	126.7	128.1	0.66	0.34	< 0.01	0.68	0.34	0.26
Initial	122.9	123.0	120.8	0.98	0.20			0.13	0.33
Final	133.1	131.3	131.3	1.17	0.44			0.28	0.53
Change ^d , cm/d	0.14	0.10	0.14	0.03	0.5	0.1	0.77	0.94	0.25
Heart girth									
Mean, cm	174.7	173.9	174.2	0.82	0.78	< 0.01	0.41	0.66	0.59
Initial	167.2	168.8	165.5	1.40	0.27			0.40	0.17
Final	181.1	179.1	179.3	1.23	0.45			0.30	0.50
Change ^d , cm/d	0.22	0.14	0.13	0.02	0.01	0.47	0.46	0.01	0.19
Paunch girth									
Mean, cm	205.4	205.0	207.5	1.30	0.36	< 0.01	0.85	0.26	0.34
Initial	197.8	198.0	192.8	0.91	<0.10			0.07	0.26
Final	211.5	209.6	212.3	1.9	0.61			0.60	0.26
Change ^d , cm/d	0.18	0.22	0.18	0.04	0.73	0.17	0.25	0.85	0.44
Hip width									
Mean, cm	47.5	47.8	47.7	0.17	0.31	< 0.01	0.14	0.44	0.18
Initial	46.5	46.3	46.1	0.26	0.42			0.19	0.95
Final	48.4	48.9	48.3	0.26	0.26			0.78	0.11
Change ^d , cm/d	0.02	0.03	0.02	0.003	0.23	0.62	0.03	_	_
BCS									
Mean, cm	3.6	3.6	3.6	0.04	0.9	< 0.01	0.61	0.70	0.88
Initial	3.5	3.4	3.4	0.04	< 0.01			0.16	0.01
Final	3.7	3.8	3.6	0.07	0.32			0.52	0.23

^{*a*}WBG = a byproduct of the beer brewing industry.

 ${}^{b}L$ = Linear effect.

 $^{c}Q = Quadratic effect.$

^dCalculated based on change per 2-wk interval.

Dry matter intake tended to be different among treatments (P = 0.08) and had a quadratic response (P = 0.04) with less DMI in heifers fed the 20% WBG diet due probably to a greater moisture content of WBG. Regardless of small particle size, Firkins et al. (2002) found that WBG could also be an effective forage NDF source, which could have contributed to more gut fill in heifers fed 20% WBG. From these results, feeding more than 20% WBG in the diet of growing heifers would likely decrease DMI. Moreover, Schingoethe et al. (1988) found that the moisture content of WBG can affect the level of intake in cattle, particularly when it is fed in combination with silages. The lower DMI observed in heifers fed 20% WBG may be a response to gut fill and distension caused by the structural volume of WBG water held within the cell wall (Balch and Campling, 1962). However, DMI as a percentage of BW was similar across treatments.

Total NDF intake was greater for heifers fed 10% and 20% WBG (P < 0.01) and had a quadratic response (P < 0.01) than heifers fed 0% WBG. Forage NDF intake as provided by grass silage and corn silage was different among treatments (P < 0.01) with a linear and quadratic effect (P < 0.01 and P = 0.01, respectively) and was less for heifers fed 20% WBG. This was expected due to the increased amount of WBG in the diet. According to Hersom

	Treatment (WBG)			<i>P</i> -value					
Item	0%	10%	20%	SE	Treatment	L^a	Q^b		
DMI ^c , kg/d	9.57	10.0	9.23	0.2	0.08	0.28	0.04		
NDFI, kg/d	4.32	4.83	4.57	0.01	0.01	0.1	0.01		
Forage NDFI, kg/d	4.15	4.27	3.72	0.09	< 0.01	0.001	0.01		
DMI, % BW	2.12	2.20	2.15	0.05	0.21	0.76	0.24		
NDFI, % BW	0.98	1.07	1.05	0.02	0.01	0.03	0.03		
Forage NDFI, % BW	0.94	0.95	0.85	0.01	< 0.01	< 0.01	0.02		
$\mathrm{D}\mathrm{M}^{d},\%$	53.6	44.8	51.2	2.3	0.04	0.46	0.01		
OM^e , %	57.9	49.7	55.2	2.1	0.04	0.36	0.01		
CP ^{<i>f</i>} , %	45.3	39.5	50.1	2.9	0.06	0.25	0.03		
NDF ^g , %	47.0	40.7	45.2	2.5	0.22	0.62	0.09		
$\mathrm{ADF}^{h},\%$	37.4	30.9	35.7	3.1	0.29	0.70	0.13		
Fat ⁱ , %	66.8	65.6	72.3	2.2	0.06	0.05	0.15		
Starch ^{<i>i</i>} , %	98.5	98.2	98.7	0.2	0.28	0.47	0.15		
Hemicellulose ^k , %	58.2	50.8	55.9	1.7	0.01	0.36	0.007		

Table 5. DMI and apparent total tract nutrient digestibility of nutrients for heifers limit-fed diet containing 0%, 10%, or 20% WBG during week 12

 $^{a}L = Linear effect.$

 ${}^{b}Q = Quadratic effect.$

 $^{\rm c}\text{DMI}$ (kg/d) during the digestibility portion of the experiment.

^dDry matter digestibility.

^eOrganic matter digestibility.

^fCrude protein digestibility.

^gNeutral detergent fiber digestibility.

^hAcid detergent fiber digestibility.

'Fat digestibility.

Starch digestibility.

^{*k*}Hemicellulose digestibility.

(2006), the effective fiber of WBG is useful in dairy rations as it is utilized to replace some portion of the forage in the rations. Because most of the starch has been fermented away, WBG can be considered as a moderate source of fiber.

Neutral detergent fiber intake as a percentage of BW was different among treatments (P = 0.01) and was less in heifers fed the control diet (0% WBG). According to Hoffman and Kester (2013), dairy heifers consume a near-constant 1.0% of BW as NDF, which agrees with our results (Table 5). Neutral detergent fiber intakes conform to gut fill theories of intake regulation according to Mertens (1994) who suggests that gut fill regulation of NDFI occurs in lactating dairy cows at 1.2% of BW when fed diets containing >30% NDF. Mertens et al. (1994) fed diets containing 36.1% to 49% NDF (greater than NDF in diets typically fed to lactating cows) to heifers and found that NDF gut fill regulation occurred near 1% of BW.

The digestibility of DM and OM was different among treatments (P = 0.04 and P = 0.04, respectively) and had a quadratic response (P = 0.01 and P = 0.01). Greater DM and OM digestibility were observed in heifers fed the 10% WBG diet. Crude protein digestibility tended to be different among treatments (P = 0.06) and had a quadratic effect (P = 0.1) and was greater in heifers fed the 0% WBG and the 20% WBG diet compared to heifers fed 10% WBG diet (P < 0.03). Neutral detergent fiber and ADF digestibility were similar among treatment.

According to Colucci et al. (1982), the rate at which digesta moves through the gastrointestinal tract, the rate of fermentation of the feed, and the amount of DM consumed are the major factors that determine how much of the nutrient will be digested, absorbed, and utilized in the animal. Alteration of one of those factors generally changes the other two. The lower DM digestibility observed in heifers limit-fed diets with 10% WBG would likely be attributed to greater forage NDF content in the diet (Pino et al., 2018). Including WBG in heifers' diets increased fiber content because WBG is a good source of forage NDF despite its small particle size (Firkins et al., 2002). Greater NDF content in the diet causes a slower passage rate and increased retention time in the rumen, which may allow microbial growth and more nutrient absorption (Pino et al., 2018).

This may explain the similar growth performance of heifers fed 10% WBG despite less nutrient digestibility observed in this group.

Results of DM digestibility of diets from this study were lower (53.6%, 44.8%, and 51.2% for the 0%, 10%, and 20% WBG, respectively) compared to 77% DM digestibility when WBG was included up to 15% in diets composed of corn silage, ryegrass silage, and concentrated feed, which were provided to Holstein cows (Geron et al., 2010). Greater CP digestibility in the 20% WBG diet may have been influenced by higher rumen undegradable protein (RUP) content of WBG. According to Clark et al. (1987), approximately 50% of the protein found in WBG is RUP.

Digestibilities of NDF, ADF, and starch were similar among treatments. Fat digestibility tended to be different among treatment (P = 0.06) and increased linearly (P = 0.05) with increasing levels of WBG in diets. Fat digestibility was greater in heifers fed the 20% WBG. Fat from WBG is bound to feed particles and slowly introduced to the rumen and has fewer negative effects compared with other dietary fat sources. Hemicellulose digestibility was different among treatments (P < 0.01) and was greater in heifers fed the 0% WBG and the 20% WBG diets compared to heifers fed the 10% WBG diet. Overall, the digestibility of DM, OM, CP, fat, and hemicellulose was less in heifers fed 10% WBG probably because of greater roughage: concentrate ratio leading to greater NDF and forage NDF intake.

From an economic standpoint, the cost of concentrate feeds used in our study, mainly energy mix and protein mix was estimated at \$252/metric ton and \$387/metric ton respectively (Poulin Grain, Inc., Newport, VT), while the cost of WBG was \$190/ metric ton on an as-dry basis. Nationwide, the price for corn was \$147.5 per metric ton, while soybean price was \$333.5 per metric ton (Indexmundi, 2019a, 2019b). Based on all feed ingredient prices, the cost of our treatment diets was on average \$0.26, \$0.22, and \$0.20/kg of DM for 0%, 10%, and 20% WGB diet, respectively. Considering the ADG of heifers fed different amounts of WBG in diets, the feed cost per kilogram of ADG was \$2.21, \$1.90, and \$1.79 for heifers fed 0%, 10%, and 20% WBG diet, respectively. Hence, feeding diets containing 20% WBG to dairy heifers would be more economical than feeding diets supplemented with conventional concentrates.

CONCLUSIONS

In agreement with our hypothesis, dairy heifers performed equally when limit-fed diets containing different levels of WBG. However, we suggest not to feed more than 20% WBG in diets of dairy heifers as we observed a decreasing tendency in the performance of heifers fed a greater amount of WBG. Body weight and frame growth were similar among treatments but ADG was slightly greater than the target. Nutrient digestibilities were mostly greater for heifers fed the 0% and 20% WBG diet suggesting that grass silage and WBG used could have influenced the overall digestibility as they contributed to greater NDF content in the 10% WBG. This research indicates that including WBG at a rate of 20% DM in heifer's diets may be more economical, and WBG can replace soybean- and corn-based concentrates without compromising heifer's growth performance.

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LITERATURE CITED

- Ajila, C. M., S. K. Brar, M. Verma, R. D. Tyagi, S. Godbout, and J. R. Valéro. 2012. Bio-processing of agro-byproducts to animal feed. Crit. Rev. Biotechnol. 32:382–400. doi:10. 3109/07388551.2012.659172
- Anderson, J. L., K. F. Kalscheur, J. A. Clapper, G. A. Perry, D. H. Keisler, A. D. Garcia, and D. J. Schingoethe. 2015a. Feeding fat from distillers dried grains with solubles to dairy heifers: II. Effects on metabolic profile. J. Dairy Sci. 98:5709–5719. doi:10.3168/jds.2014-9163.
- Anderson, J. L., K. F. Kalscheur, A. D. Garcia, and D. J. Schingoethe. 2015b. Feeding fat from distillers dried grains with solubles to dairy heifers: I. Effects on growth performance and total-tract digestibility of nutrients. J. Dairy Sci. 98:5699–5708. doi:10.3168/jds.2014-9162.
- AOAC International. 1998. Official methods of analysis. 16th ed. Washington, DC: AOAC International.
- AOAC International. 2002. Official methods of analysis. 17th ed. Gaithersburg, MD: AOAC International.
- AOAC International. 2006. Official methods of analysis. 18th ed. Gaithersburg, MD: AOAC International.

- Bach Knudsen, K. E. 1997. Carbohydrate and lignin contents of plant materials used in animal feeding. Anim. Feed Sci. Technol. 67:319–338. doi:10.1016/S0377-8401(97)00009-6.
- Balch, C. C., and R. C. Campling. 1962. Regulation of voluntary food intake in ruminants. Nutr. Abstr. Rev. 32:669–686.
- Belibasakis, N. G., and D. Tsirogogianni. 1996. Effects of wet brewer's grains on milk yield, milk composition, and blood components of dairy cows in hot weather. Anim. Feed Sci. Technol. 57:175–181.
- Clark, J. H., M. R. Murphy, and B. A. Crooker. 1987. Supplying the protein needs of dairy cattle from by-product feeds. J. Dairy Sci. 70:1092–1109. doi:10.3168/ jds.S0022-0302(87)80116-9.
- Colucci, P. E., L. E. Chase, and P. J. Van Soest. 1982. Feed intake, apparent diet digestibility and rate of particulate passage in dairy cattle. J. Dairy Sci. 65:1445–1456. doi:10.3168/jds.S0022-0302(82)82367-9.
- Davis, C. L., D. A. Grenawalt, and G. C. McCoy. 1983. Feeding value of pressed brewers' grains for lactating dairy cows. J. Dairy Sci. 66:73–79. doi:10.3168/jds. S0022-0302(83)81755-X.
- Faccenda, A., M. A. Zambom, D. D. Castagnara, A. Sanches de Avila1, T. Fernandes, E. I. Eckstein, F. A. Anschau, and C. R. Schneider. 2017. Use of dried brewers' grains instead of soybean meal to feed lactating cows. Rev. Bras. Zootec. 46(1):39–46. doi:10.1590/ s1806-92902017000100007.
- Firkins, J. L., D. I. Harvatine, J. T. Sylvester, and M. L. Eastridge. 2002. Lactation performance by dairy cows fed wet brewers grains or whole cottonseed to replace forage. J. Dairy Sci. 85:2662–2668. doi:10.3168/jds. S0022-0302(02)74351-8.
- Frasson, M. F., S. C. V. Jaurena, A. M. Menegon, M. M. Severo., J. H. Motta, and W. S. Teixeira. 2018. Intake and performance of lambs finished in feed lot with wet brewer's grains. J. Anim. Sci. Technol. 60:12. doi:10.1186/ s40781-018-0166-8.
- Geron, L. J. V., L. M. Zeoula, J. A. Erkel, I. N. Prado, E. Bublitz, and O. P. P. Prado. 2010. Consumo, digestibilidade dos nutrientes, produção e composição do leite de vacas alimentadas com resíduo de cervejaria fermentado. Acta Sci. Anim. Sci. 32:69–76. doi:10.4025/ actascianimsciv32i1.6990.
- Hatungimana, E., and P. S. Erickson. 2019. Effect of storage of wet brewer's grains treated with salt or a commercially available preservative on the prevention of spoilage, in vitro and in situ dry matter digestibility and intestinal protein digestibility. Appl. Anim. Sci. 35:464–475. doi:10.15232/aas.2019-01857.
- Hersom, M. J. 2006. By-product feed utilization for forage diets. In: Proceedings of the 55th Annual Florida Beef Cattle Short Course; May3-5, 2006. University of Florida Institute of Food and Agricultural Sciences, Gainesville, FL. p. 5–14.
- Hoffman, P. C., and L. E. Armentano. 1988. Comparison of brewer's wet and dried grains and soybean meal as supplements for dairy cattle. Nutr. Rep. Int. 38:655–663.
- Hoffman, P. C., and K. Kester. 2013. Estimating dry matter intake of dairy heifers. Forage focus. Madison: University of Wisconsin-Madison. Available from www.midwestforage.org > pdf > 819.pdf.pdf [accessed February 4, 2020].
- Hoffman, P. C., C. R. Simson, and M. Wattiaux. 2007. Limit feeding of gravid Holstein heifers: effect on growth,

manure nutrient excretion, and subsequent early lactation performance. J. Dairy Sci. 90:946–954. doi:10.3168/jds. S0022-0302(07)71578-3.

- Homm, J. W., L. L. Berger, and T. G. Nash. 2008. Determining the corn replacement value of wet brewer's grain for feedlot heifers. Prof. Anim. Sci. 24:47–51. doi:https://doi.org/10.15232/S1080-7446(15)30809-3.
- IndexMundi. 2019a. Commodity price indices. Available from https://www.indexmundi.com/commodities/?commodity=soybeans [accessed November 14, 2019].
- IndexMundi. 2019b. Commodity price indices. Available from https://www.indexmundi.com/commodities/?commodity=corn [accessed November 14, 2019].
- Lilly, V., M. Birch, and B. Garscadden. 1980. The preservation of spent brewer's grains by the application of intermediate moisture food technology. J. Sci. Food Agric. 31:1059– 1065. doi:10.1002/jsfa.2740311014.
- Manthey, A. K., J. L. Anderson, and G. A. Perry. 2016. Feeding distillers dried grains in replacement of forage in limit-fed dairy heifer rations: effects on growth performance, rumen fermentation, and total-tract digestibility of nutrients. J. Dairy Sci. 99:7206–7215. doi:10.3168/jds.2015-10785
- Marston S. P., D. A. Spangler, N. L. Whitehouse, and P. S. Erickson. 2009. Case study: addition of a silage preservative reduces spoilage in wet brewer's grain. Appl. Anim. Sci. 25:388–392. doi:10.15232/s1080-7446(15)30730-0.
- Mertens, D. R. 1994. Regulation of forage intake. In: G. C. Fahey Jr, editor, Forage quality, evaluation, and utilization. ASA, CSSA, Madison, WI. p. 450–493.
- Murdock, F. R., A. S. Hodgson, and R. E. Riley Jr. 1981. Nutritive value of wet brewer's grains for lactating dairy cows. J. Dairy Sci. 64:1826:1832. doi:10.3168/jds. s0022-0302(81)82771-3.
- Muthusamy, N. 2014. Chemical composition of brewer's spent grain. A review. Int. J. Sci. Environ. Technol. 3:2109–2112.
- National Research Council. 2001. Nutrients requirements of dairy cattle. National Academic Press, Washington, DC. 381 p.
- Nishino, N., H. Harada, and E. Sagaguchi. 2003. Evaluation of fermentation and aerobic stability of wet brewer's grains ensiled alone or in combination with various feeds as a total mixed ration. J. Sci. Food Agric. 83:557–563. doi:10.1002/jsfa.1395.
- Pino, F., L. K. Mitchell., C. M. Jones and A. J. Heinrichs. 2018. Comparison of diet digestibility, rumen fermentation, rumen rate of passage, and feed efficiency in dairy heifers fed ad libitum versus precision diets with low- and high-quality forages. J. Appl. Anim. Res. 46(1):1296–1306. doi:10.1080/09712119.2018.1498788.
- Robertson, J. A. I., K. J. A. Anson., J. Treimo., C. B. Faulds., T. F. Brocklehurst., V. G. H. Eijsink, and K. W. Waldron. 2010. Profiling brewer's spent grain for composition and microbial ecology at the site of production. LWT Food Sci. Technol. 43:890–896. doi:10.1016/j.lwt.2010.01.019.
- Schingoethe, D. J., F. M. Byers, and G. T. Schelling. 1988. Nutrient needs during critical periods of the life cycle. In: D. C. Church, editor, The ruminant animal: digestive physiology and nutrition. Upper Saddle River (NJ): Prentice-Hall, Inc.; p. 421–447.
- Schneider, R. M., J. H. Harrison, and K. A. Loney. 1995. The effect of bacteria inoculant, beet pulp, and propionic acid on ensiled wet brewer's grains. J. Dairy Sci. 78:1096–1105. doi:10.3168/jds.s0022-0302(95)76726-1.

- Tozer, P. R., and A. J. Heinrichs. 2001. What affects the costs of raising replacement dairy heifers: a multiple-component analysis. J. Dairy Sci. 84:1836–1844. doi:10.3168/jds. S0022-0302(01)74623-1.
- Van Keulen, J., and B. A. Young. 1977. Evaluation of acid-insoluble ash as a natural marker in ruminant digestibility studies. J. Anim. Sci. 44:282–287. doi:10.2527/ jas1977.442282x.
- Van Soest, P. J., J. B. Robertson, and B. A. Lewis. 1991. Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. J. Dairy Sci. 74:3583–3597. doi:10.3168/jds. S0022-0302(91)78551-2.
- Wang, B., Y. Luo, K. H. Myung, and J. X. Liu. 2014. Effect of storage duration and temperature on the chemical composition, microorganism density, and in vitro rumen fermentation of wet brewer's grains. Asian-Australas. J. Anim. Sci. 27:832–840. doi:10.5713/ajas.2013.13668.

- Wildman, E. E., G. M. Jones, P. E. Wagner, R. L. Boman, H. F. Troutt Jr, and T. N. Lesch. 1982. A dairy cow body condition scoring system and its relationship to selected production characteristics. J. Dairy Sci. 65:495–501. doi:10.3168/jds.s0022-0302(82)82223-6.
- Zanton, G. I., and A. J. Heinrichs. 2005. Meta-analysis to assess effect of prepubertal average daily gain on Holstein heifers on firstlactation production. J. Dairy Sci. 88:3860– 3867. doi:10.3168/jds.S0022-0302(05)73071-X.
- Zanton, G. I., and A. J. Heinrichs. 2007. The effects of controlled feeding of a high-forage or high-concentrate ration on heifer growth and first-lactation milk production. J. Dairy Sci. 90:3388–3396. doi:10.3168/jds. 2007-0041.
- Zanton, G. I., and A. J. Heinrichs. 2009. Limit-feeding with altered forage-to-concentrate levels in dairy heifers' diets. Appl. Anim. Sci. 25:393–403. doi:10.15232/ s1080-7446(15)30740-3.