

Acta Scientiarum. Animal Sciences

ISSN: 1806-2636 ISSN: 1807-8672

Editora da Universidade Estadual de Maringá - EDUEM

Fonseca, Natália Vilas Boas; Cardoso, Abmael da Silva; Hoffmann, Alvair; Leite, Rhaony Gonçalves; Ferrari, Adriana Cristina; Fernandes, Marcia Helena Machado da Rocha; Reis, Ricardo Andrade Characterization and effects of DDG on the intake and digestibility of finishing bulls in feedlots Acta Scientiarum. Animal Sciences, vol. 43, e51877, 2021 Editora da Universidade Estadual de Maringá - EDUEM

DOI: https://doi.org/10.4025/actascianimsci.v43i1.51877

Available in: https://www.redalyc.org/articulo.oa?id=303168054030



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Doi: 10.4025/actascianimsci.v43i1.51877



RUMINANT NUTRITION

Characterization and effects of DDG on the intake and digestibility of finishing bulls in feedlots

Natália Vilas Boas Fonseca^{1*}, Abmael da Silva Cardoso¹, Alvair Hoffmann², Rhaony Gonçalves Leite¹, Adriana Cristina Ferrari¹, Marcia Helena Machado da Rocha Fernandes¹ and Ricardo Andrade Reis¹

¹Departamento de Zootecnia, Faculdade de Ciências Agrárias e Veterinárias, Universidade Estadual Paulista "Julio de Mesquita Filho", via de acesso Prof. Paulo Donato Castellane, km 5, 14884-900, Jaboticabal, São Paulo, Brazil. ²Departamento Técnico Trouw Nutrititon, Praça Capital, Campinas, São Paulo, Brazil. *Author for correspondence. E-mail: nataliafonseca0531@gmail.com

ABSTRACT. The aim of this study was to characterize four corn and sorghum co-products (DDG) in terms of their protein and carbohydrate fractions; we also evaluated the effects of substituting the protein source of the conventional supplement by DDG on consumption and nutrient digestibility in confined finishing cattle. Thirty-six male Nellore cattle with a mean age of 24 months were used. The treatments were: FA: concentrate with corn as an energy source and cottonseed meal as a protein source; DDG50: concentrate with a 50% substitution of the FA protein source by DDG; DDG100: concentrate with 100% substitution of the FA protein source by DDG. The experimental design was completely randomized with three treatments and three replicates (pens) containing four animals per pen. We found that the use of DDG in the finishing phase did not interfere with the animals' food intake or the digestibility of the nutrients (p > 0.05). Nutrients were used by the animals; therefore, DDGs may be viable substitutes of cottonseed meal. We conclude that the bromatological composition of this co-product is influenced a lot during processing; therefore, the nutritional values of this co-product present in the composition tables may not be true.

Keywords: cattle; co-products; confinement; corn ethanol; fractionation of nitrogen compounds; animal nutrition.

Received on January 21, 2020. Accepted on May 18 2020.

Introduction

Livestock farming is a very important part of the Brazilian agribusiness sector. The significant growth of livestock farming has led to the pursuit of alternatives in order to increase production and profitability, as traditional livestock farming systems have proved to be economically inefficient. The intensification of production systems appears to be a viable strategy (Romanzini, Bernardes, Munari, Reis, & Malheiros, 2018).

However, in intensive farming systems the cost of production increases, thus reducing the profitability of the system; therefore, alternatives that use less expensive products and nutritional quality in animal feed are becoming increasingly attractive.

The use of maize co-products from ethanol production, also known as dried distiller's grains (DDG) is a viable alternative as these co-products have high protein content and can be included in the animals' diets by substituting protein ingredients that have a high cost for farmers. These co-products have 26.0%-31.7% crude protein (CP), 47.0-53.0% rumen non-degradable protein (PNDR), 33.1-43.9% neutral detergent fiber (NDF), and 85-90% total digestible nutrients (TDN) (Li, Li, Yang, & Beauchemin, 2012; Valério Geron et al., 2017). However, a great challenge in the use of DDGs is to accurately determine their nutrient content that varies greatly, thus hindering their use in animal nutrition (Valério Geron et al., 2017).

Replacing 50% of cottonseed meal with this co-product cottonseed mealleads to improved performance, digestibility coefficient, and economic efficiency and increases the dry matter intake, feed conversion rate, and average daily gain of the animals (Omer et al., 2015).

The productivity response of the animals depends on the consumption and the digestibility of the nutrients (Silva et al., 2009). In diets with high digestibility, the consumption may be lower, because when the food is more digestible the animal's required energy intake will be satisfied by a lower feed intake, owing to the energy density of the diet (National Academies of Sciences, Engineering, and Medicine, 2016). In a study published by Walter, McAllister, Yang, Beauchemin, & McKinnon (2012), the authors reported that the use of corn DDG increased the CP and fiber digestibility; the latter increase was attributed to the low lignin content of the diet. This

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improvement is probably due to an increase in the ruminal fermentation power of DDG. DDG fiber is potentially more soluble and fermentable than other concentrated foods because it represents the portion of the cereal grain envelope (Association of Official Analytical Chemists [AOAC], 1990).

Although DDG production in Brazil is still small compared to the US, it has grown by more than 1000% in recent years. The nitrogen and carbohydrate fractions of DDGs produced in Brazil have not yet been evaluated; it is, therefore, necessary to characterize the DDGs in order to develop supplementation strategies in finishing cattle and increase their performance.

Our aim in this study was to characterize four corn and sorghum DDGs in terms of their protein and carbohydrate fractions; we also evaluated the effects of substituting the protein source of the conventional supplement (cottonseed meal) by DDG on the consumption and the digestibility of nutrients by confined finishing cattle. We hypothesized that the replacement of cottonseed meal by DDG will not interfere with nutrient intake and digestibility.

Material and methods

Experimental site

All procedures described were in accordance with the Ethical Principles on Animal Experimentation, adopted by the National Council for the Control of Animal Experimentation (CONCEA) and approved by the Committee on Ethics in Animal Use (CEUA) of UNESP (protocol no. 12703/15).

The study was conducted during the finishing phase of the animals, for a total of 120 days from May to July 2016, in the feedlot of the Forage Sector of the College of Agricultural and Veterinary Sciences (FCAV) of the São Paulo State University (UNESP), Jaboticabal campus, São Paulo, Brazil.

Animals and treatments

Thirty-six male Nellore cattle (*Bos taurus indicus*) were used, with an average body weight of 410 kg \pm 35.2 and an average age of 24 months. The animals were allocated to collective pens and were fed a diet with a 30:70 roughage to concentrate ratio and corn silage as a bulk feed. Diets were formulated for a gain of 1.6 kg day⁻¹ according to National Research Council (NRC, 1996) and were offered at 6:00 a.m. and 3:00 p.m., when nutrient intake and digestibility were evaluated.

The experimental design was completely randomized with three replicates. The animals were submitted to three treatments: FA: concentrate with corn as an energy source and cotton bran as a protein source; DDG50: concentrate with a 50% substitution of the FA protein source by DDG; DDG100: concentrate with 100% replacement of the FA protein source by DDG (Table 1).

	Diet composition		
	FA	DDG50	DDG100
Ingredients g kg ⁻¹ DM			
Corn silage	300.0	300.0	300.0
Cottonseed meal	184.6	92.3	0.0
Corn	489.1	475.3	461.4
DDG	0.0	92.3	184.6
Urea/Sulfate	0.0	3.7	8.3
Salt	1.4	1.4	1.4
Limestone	6.5	6.5	6.5
Premix*	18.5	18.5	18.5
Caulim	0.0	10.2	19.4
Chemical composition of the diet g kg ⁻¹ DM			
DM	711.0	713.0	715.0
CP	138.5	136.8	137.5
TDN	740.0	741.0	742.0
NDF	295.7	326.1	356.5
Starch	460.2	448.7	437.2
EE	30.9	32.4	33.9

Table 1. Percentage of inclusion of the ingredients and chemical composition of the diet.

Dry matter (DM), crude protein (CP), total digestible nutrients (TDN), neutral detergent fiber (NDF), ether extract (EE). *Guaranteed analysis: Calcium (Min/Max) 120/145 g kg⁻¹; Phosphorus 30 g kg⁻¹; Sodium 80 g kg⁻¹; Potassium 50 g kg⁻¹; Magnesium 68 g kg⁻¹; Sulfur 25 g kg⁻¹; Zinc 1.220 mg kg⁻¹; Copper 330 mg kg⁻¹; Fluorine 500 mg kg⁻¹; Manganese 950 mg kg⁻¹; Cobalt 20 mg kg⁻¹; Iodine 24 mg kg⁻¹; Selenium 6 mg kg⁻¹; Vitamin A (Min) 67.000 IU Kg⁻¹; Vitamin D3 (Min) 9.500 IU Kg⁻¹; Vitamin E (Min) 950 IU kg⁻¹; Monensin 650 mg kg⁻¹; DM - Dry matter; CP - Crude Protein; TDN- Total Digestible Nutrients; NDF- Neutral Detergent Insoluble Fiber; EE - Ethereal Extract.

Evaluation of nutrient intake and digestibility

Nutrient intake was calculated daily by measuring the difference between the diet supplied and the leftovers in each pen (there were three pens per treatment). The leftovers were removed from the stall and weighed before the first feeding in the morning. Samples were obtained from the remains in the pens, which were dried in an oven at 55°C for 72 hours. Subsequently, they were milled through 2 or 1 mm screen sieves.

The fecal dry matter excretion was estimated with the internal indicator technique (Casali et al., 2008), with indigestible neutral detergent fiber (iNDF) being the indicator adopted. The iNDF contents of the feces samples as well as the food (bulk and concentrate ingredients) and leftovers, were determined by the *in situ* incubation procedure for 240 hours (Casali et al., 2008).

To estimate the digestibility of the diet, fecal samples were collected once a day for three days at different times (8:00, 12:00 and 4:00 a.m.). Spot fecal samples were collected from each animal individually and were dried in an oven at 55°C for 72 hours. Afterwards, they were ground in a mill with 2 and 1 mm sieves for NDF determination and other analyses, respectively (Casali et al., 2008).

Dry matter (DM), CP, ethereal extract (EE), ash (AOAC, 1990), and NDF were determined from the samples milled in a 1 mm mesh sieve as described by Delevatti et al. (2019); crude energy was calculated with the use of an adiabatic calorimetric pump (PARR Instrument Company 6300, IL, USA).

Using the nutrient intake and the excretion of these nutrients in the feces, the total digestibility was calculated by the following formula: Dig = (CN - EF)/CN, where Dig = total apparent digestibility of dry matter and nutrients (%), CN = consumption of total dry matter and nutrients (kg day⁻¹), and EF = fecal excretion (kg day⁻¹) [EF = (iNDF ingested/iNDF feces) \times 100].

DDG characterization

Four different products were analyzed: sorghum DDG for 2015, corn DDG for 2015 and 2016, and corn DDG for 2017, all obtained from the Sipal Group (Campos Júlio, Mato Grosso, Brazil). The four co-products were used in the forage and pasture sector of UNESP, Campus de Jaboticabal, São Paulo, Brazil, in scientific experiments. The DDG of 2016 was used as a substitute of cottonseed meal in this experiment cotton seed meal.

From these samples, we calculated carbohydrate fractionation, protein, rumen-degraded protein (RDP), and rumen non-degraded protein (URP).

The fractionation of proteins was analyzed according to the methodology described by Licitra, Hernandez and Van Soest (1996), in which fraction A refers to the nitrogen soluble in trichloroacetic acid (TCA), fraction B3 is determined by the difference between the levels of neutral detergent insoluble nitrogen (NIDN) and acid detergent insoluble nitrogen (NIDA), and fraction C refers to NIDA. To determine fractions B1 and B2, borate-phosphate buffer and a sodium azide solution were added to the sample, which, after standing for 3 hours, was washed with cold distilled water. The remaining residue was fraction B2; the soluble fraction B1 was calculated by the crude protein difference while all other fractions were already calculated.

Carbohydrate fractionation was analyzed according to the method described in Sniffen, O'Connor, Van Soest, Fox and Russell (1992). The RDP and UDP values were calculated based on the association of the obtained CP fractions with their respective passage and digestion rates (Vieira et al., 2020). Thus, the PDR (as a percentage of CP) was calculated as: $PDR = A + B1 \left(\frac{kdB1}{kdB1} + kp\right) + B2 \left(\frac{kdB2}{kdB2} + kp\right)$), where A, B1, B2, and B3 are the proportions of the protein fractions of the protein, kdB1, kdB2, and kdB3 are the ruminal degradation rates of the respective fractions, and kp is the ruminal passage rate (0.02 and 0.06% h⁻¹). The baseline ruminal passage rate was obtained by Fox et al. (2003). The PNDR was estimated by the following equation: (UDP = 1 - RDP).

Data analysis

The statistical design was completely randomized (CRD) and consisted of three treatments with three replicates (pens with 4 animals each) per treatment (n = 12). Each pen was considered as an experimental unit.

The statistical model was: $yij = \mu + \tau i + \epsilon ij$

where:

Yij = observation j in the treatment i

u = general mean

 τi = fix effect in the treatment i

εij = random error of observation j in the treatment i

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The assumptions for the analysis of variance test (ANOVA) [error normality (Cramér-von Mises) and homoscedasticity (Box-cox)] were determined using the PROC UNIVARIATE procedure of the SAS software 2008 (Statistical Analysis System [SAS], 2008). The means were generated by the LSMEANS command; when the differences between the means were significant, Tukey's test was used to compare the averages by using the PDIFF option in SAS

Results

Consumption and digestibility

There was no statistical difference in dry matter intake, NDF, energy, CP, OM, EE, total carbohydrates (TC), and non-fibrous carbohydrates (NFC) between diets; there were also no significant differences among nutrient digestibilities (p > 0.05, Table 2).

Table 2. Consumption of dry matter and nutrients (kg day⁻¹) and apparent digestibility (%) of the nutrients of the experimental diets.

	Treatments					
	FA	DDG50	DDG100100	SEM	P-value	
Intake kg day¹						
DMI	10.8	10.7	10.2	0.27	0.324	
NDF	2.33	2.34	2.35	0.06	0.977	
CP	1.38	1.38	1.37	0.03	0.96	
OM	9.32	9.28	9.29	0.23	0.992	
EE	0.34	0.34	0.34	0.01	0.994	
TC	7.59	7.56	7.57	0.19	0.995	
NFC	5.26	5.23	5.22	0.13	0.973	
Energy MJ kg DM ⁻¹	9.55	9.49	9.48	0.24	0.974	
Digestibility g kg ⁻¹						
OM	729.80	726.80	725.50	0.70	0.501	
CP	662.70	657.30	655.20	1.10	0.81	
NDF	436.60	436.90	438.50	1.80	0.178	
EE	922.70	922.10	921.80	2.41	0.057	
TC	878.00	876.50	875.70	0.52	0.562	
ME MJ kg DM ⁻¹	7.00	6.93	6.90	0.87	0.97	

DMI (dry matter intake), NDF (neutral detergent fiber), CP (crude protein), OM (organic matter), EE (ether extract), TC (total carbohydrates), NFC (non-fibrous carbohydrates).

DDG characterization

Table 3 shows the characterization of the four DDGs studied.

Table 3. Carbohydrate fractionation, protein fractionation, and ruminal degradable protein (RDP) and undegradable protein (URP) calculations of four distinct DDG crops.

	DDG 1	DDG 2	DDG 3	DDG 4	SEM
Carbohydrate fractions g kg ⁻¹ DM				-	
A+B1	164.70	148.60	115.20	44.00	2.677
B2	699.80	754.80	795.80	845.30	3.086
С	135.50	96.60	89.00	110.70	1.023
Nitrogen fractions g kg ⁻¹ CP				_	_
A	118.00	89.00	94.40	75.00	0.895
B1	46.70	78.50	96.70	87.50	1.087
B2	525.60	576.30	560.20	569.70	1.128
В3	100.20	74.90	71.40	83.40	0.642
С	209.50	181.30	177.20	181.30	0.745
RDP g kg ⁻¹	458.00	486.00	501.00	481.00	0.009
URP g kg ⁻¹	542.00	514.00	499.00	519.00	0.009

DM (dry matter), CP (crude protein), RDP (ruminal degradable protein), URP (undegradable protein) DDG 1: sorghum DDG; DDG 2: maize DDG used in 2015; DDG 3: maize DDG used in 2016; DDG 4: maize DDG used in 2017.

Discussion

Consumption and digestibility

Animal performance is determined by nutrient intake, which can be regulated by several factors such as food (fiber content, energy density, and volume), animal (weight, production level, and physiological state), and feeding (food condition, frequency of feeding) (Reis, Ruggieiri, Casagrande, & Páscoa, 2009). According to Nardone, Ronchi, Lacetera, Ranieri and Bernabucci (2010), food intake is responsible for 70% of the variation in the potential of animal production; the remaining 30% is attributed to the digestibility and efficiency of food use.

The inclusion of DDGs with a lower starch content and a higher amount of fiber can alter nutrient consumption. Studies have reported a decrease in dry matter intake, with an increase in the percentage of DDG in the diet (Griffin et al., 2012). In contrast, Benchaar et al. (2013) reported an increase in consumption and a reduction in rumination when DDG was used as a substitute for soybean meal.

DDG is a more fibrous co-product in relation to other concentrates (crude fiber 5.4-10.4%, NDF 33.1-43.9%, and acid detergent fiber 11.4-20.8%) and this characteristic may possibly interfere with dry matter and nutrient consumption; the fiber content, however, is variable, mainly owing to the processing conditions of DDG (Li, Li, Yang, & Beauchemin, 2012). The absence of the effects of including DDG on dry matter intake and nutrient consumption may be related to the amount of NDF present in the co-product, which possibly did not interfere with these analyzed variables.

Digestibility is related to the nutritional value of the food provided to the animals; it refers to the capacity to utilize the available nutrients in a greater or lesser degree, which is a characteristic of the food and not of the animal, therefore facilitating the choice of the ingredients used (Silva et al., 2009).

During the drying stage of the ethanol extraction process to obtain DDG, nutrient digestibility can be compromised. If carried out in high temperatures, this procedure can cause changes in some constituents, thus making them unavailable for degradation by rumen microorganisms and, consequently, for the animals (NRC, 2012). However, the digestibility of the nutrients in the present study did not vary, which can be a positive factor. Even when DDGs were processed in high temperatures, the animals were able to take advantage of the nutrients. Therefore, DDGs may be substitutes for cottonseed meal, since some of these co-products have similar URP values to cottonseed meal.

The inclusion of DDGs in the diet is an option that reduces feed-related costs if it does not alter animal performance (function of consumption and digestibility) (Griffin et al., 2012). Based on our results, the replacement of cottonseed meal by DDG was a viable alternative, as it did not interfere with the consumption of DM or nutrients and with the apparent digestibility of the nutrients. Moreover, DDG is a coproduct that can be cost effective for the production system.

DDG characterization

With regard to nitrogenous compounds, fraction A consists of non-protein nitrogenous compounds (NNP), amino acids, and peptides and has high rumen digestibility and rapid degradation rates. Fraction B1 consists of soluble proteins rapidly degraded in the rumen, fraction B2 consists of proteins of lower solubility with an intermediate degradation rate in the rumen, and fraction B3 consists of insoluble proteins associated with the FDN fraction with a slow degradation rate in the rumen; fraction C consists of insoluble lignin-associated proteins that are indigestible in the rumen and intestines (Sniffen et al., 1992). In addition, the nitrogen content of the soil is dependent on the CP content, which, in turn, varies with plant age, crop management, fertilization, and overall management.

The A + B1 carbohydrate fractions correspond to the soluble fraction that consists of sugars that degrade rapidly in the rumen and to starch and pectin, respectively; fraction B2, which has a slower ruminal degradation rate, is the digestible portion of the cell wall, while fraction C is the indigestible portion of the cell wall (Sniffen et al., 1992).

DDGs have a high NDF content (33.1-43.9%), but this composition may vary depending on the product used (maize or sorghum), the quality control during ethanol production, and conditions such as differences in drying time and temperature (AOAC, 1990).

The main cause of such oscillation is likely a variation in the composition that is related to the dry milling process (Pecka-Kiełb et al., 2015). In this study, the EE and CP compositions were similar, but the

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coefficient of variation of the EE data was considerably high. Therefore, this large variation in the EE content may influence the A + B1 carbohydrate fractions because these are represented by non-fibrous carbohydrates (NFC = Total Carbohydrates - Fibrous Carbohydrates); in order to calculate the total carbohydrates, we must take into account the ethereal extract percentage (TC = 100 - (CP% + EE% + ash%)).

The amylopectin and amylose proportions may vary between maize hybrids and may affect the fermentation efficiency, apparently owing to differences in starch composition (Reddy, Lakshmi, Raju, Kishore, & Anil, 2017; Beck, Truong, & Stains, 2016). Therefore, these proportions can influence the A + B1 and B2 carbohydrate fractions. In the dry milling process, chemical products can be added to maintain optimum fermentation conditions (Liu, 2017); this is another factor that could influence the composition of DDG.

Nitrogen (N) analysis is a quicker and easier way to determine the CP content of a sample, since the N factor is appropriate for a specific biomass. A nitrogen factor of 6.25 is generally recommended for most biomasses, except for wheat grains; it may also not be suitable for distillers dried grains since these are processed by-products subject to multiple heat treatments and drying processes (Pekel et al., 2020).

When heat treatments are more intense they can provoke the Maillard reaction, which is a result of the amination of the amine groups in the amino acids with the reductive sugars, making the protein unavailable and incorporating it into the acid detergent fiber (ADF). This fraction is known as acid detergent insoluble nitrogen (ADIN) (Bueno, Jobim, Ribeiro, & Oliveira, 2017). The ADIN fraction has been used as an indicator of the amount of heat damage and formation of the Maillard product in DDGs (Li, Li, Yang, & Beauchemin, 2012).

Drying is the last processing step for DDG production, where temperature conditions can significantly influence protein quality (Khose, Manwar, Dhore, Kuralkar, & Waghmare, 2017). When a DDG undergoes a drying process, overheating may occur and potentially cause the Maillard reaction, which is detrimental to the quality of the animal feed (Liu, 2017). These factors can influence the protein fractions. As fraction A is represented by non-protein nitrogen compounds, it may occur during the Maillard reaction and, thus, influence the other protein nitrogen fractions (B1, B2, and B3).

The RDP consists of fraction A and the part of fraction B that is degraded in the reticulum-rumen, whereas the URP is formed by fraction C and the part of fraction B that passes through the reticulum-rumen without being fermented (Vieira et al., 2020). By modifying these fractions, one could influence the RDP and URP values.

The URP of cottonseed meal and soybean meal is 43.00 and 39.89%, respectively (Chizzotti, Tedeschi, & Valadares Filho, 2008). The results of this study confirmed that DDG has a higher URP percentage compared to the traditional protein sources mentioned.

Conclusion

The use of DDGs in the feed of finishing young bulls did not interfere with the food intake of the animals and the digestibility of the nutrients; it proved to be a positive factor and could be used as a substitute of traditional protein sources. In order to meet the animals' requirements, the characterization of the coproduct should, ideally, be carried out with each new acquisition.

Acknowledgements

The authors thank the members of UnespFOR (Unesp Jaboticabal Forage Team) for the contributions during the field trial setup. This work was supported by the Fundação de Amparo à Pesquisa do Estado de São Paulo (FAPESP, grant 2015/16631-5; 2017/11274-5); the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES); and the Conselho Nacional de Desenvolvimento Científico e Tecnológico. We would like to thank Editage (www.editage.com) for English language editing.

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