Animal Feed Science and Technology 190 (2014) 38–46

Contents lists available at ScienceDirect

Animal Feed Science and Technology

journal homepage: www.elsevier.com/locate/anifeedsci

Impact of replacing soybean meal in beef cattle diets with inactive dry yeast, a sugarcane by-product of ethanol distilleries and sugar mills

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ARTICLE INFO

Article history:
Received 25 June 2013
Received in revised form 4 January 2014
Accepted 6 January 2014

Keywords:
Digestibility
Nutrient
Rumen-degradable protein

ABSTRACT

This study assessed the intake, total and partial apparent digestibility of nutrients, pH, ruminal ammonia concentration, nitrogen efficiency usage, and productive performance of beef cattle fed with different soybean meal replacement levels with inactive dry yeast (IDY 0, 250, 500, 750, and 1000 g/kg). The forage:concentrate ratio was 60:40 and the forage source was corn silage. Concentrates were formulated to comprise 220.0 g/kg CP independent of treatments. In the first experiment (EXP 1), 35 Nelore bulls with an initial average weight of 370 ± 42 kg were distributed across a completely randomized design, with five treatments and seven replicates to assess nutrient intake and performance. EXP 1 lasted 98 days and was divided into a 14-day adaptation period and three experimental periods of 28 days each. In the second experiment (EXP 2), five castrated Nelore steers with an initial average weight of 320 ± 39 kg were fistulated in the rumen and abomasum and distributed in a 5 × 5 Latin square design, balanced for residual effect. The purpose of this experiment was to assess the total and partial digestibility of nutrients, pH, ruminal ammonia nitrogen, and nitrogen efficiency of usage. EXP 2 lasted 90 days, divided into five experimental periods. Each period lasted 18 days and was divided into 10 days for adaptation to the diets and 8 days to collect samples. The intake of dry matter (DMI) decreased linearly (P = 0.03) with increased dietary IDY levels. Conversely, the intake of neutral detergent fiber assayed with a heat-stable amylase and corrected for ash and nitrogenous compounds [aNDFom(n)] in g/day (P = 0.043), and the g/kg body weight (P = 0.011) increased linearly as IDY was added to the concentrate. The experimental diets showed no effect (P > 0.05) on the total and partial apparent nutrient digestibility. IDY had no effect (P > 0.05) on ruminal pH, ruminal ammonia nitrogen, or dietary nitrogen efficiency. Additionally, IDY had no effect on productive performance variables, with the exception of average daily gain (ADG), which decreased linearly (P = 0.028) as IDY was added to the concentrate. IDY addition resulted in decreases

Abbreviations: RDP, rumen degradable protein; IDY, Inactive dry yeast; ADG, average daily gain; HCW, hot carcass weight; FLW, final live weight; CADG, carcass average daily gain; DM, dry matter; OM, organic matter; CP, crude protein; EE, ether extract; aNDFom(n), neutral detergent fiber assayed with a heat-stable amylase and corrected for ash and nitrogenous compounds; NDIN, neutral detergent insoluble nitrogen; ADIN, acid detergent insoluble nitrogen; NFC, non-fibrous carbohydrates; FDM, flow of fecal dry matter; ADM, abomasal dry matter; TDN, total digestible nutrients; NB, nitrogen balance; UUN, urinary urea nitrogen; NPN, non-protein nitrogen; PUN, plasma urea nitrogen; Emic, microbial efficiency; PD, purine derivatives; BW, body weight; ILW, the initial live weight; DRS, dressing percentage after slaughter; RCY, carcass yield; N_{mic}, microbial nitrogen compounds; NID, normal independent distribution.

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0377-8401/$ – see front matter
http://dx.doi.org/10.1016/j.anifeedsci.2014.01.003

in DMI and ADG for beef cattle in feedlots (EXP 2). However, the apparent digestibility of nutrients and microbial efficiency were not affected. In addition, IDY did not reduce feed conversion or carcass gain. The high market price of soybean meal might make feasible its total replacement by IDY, even considering the possibility of a small reduction in ADG.

1. Introduction

As a consequence of the high cost of protein supplements in feed concentrates, new non-conventional alternatives have been exploited in recent years. Both ethanol distilleries and beer breweries have surplus yeast co-products for use in animal feed. In feedlot diets for beef cattle, soybean meal is the most frequently used protein concentrate and is also one of the most expensive products. The search for alternative products that can replace soybean meal without altering body weight gain and carcass yield is important for achieving higher profits by reducing the costs of concentrate ingredients. Yeasts are unicellular microorganisms that grow during ethanol fermentation (Yara et al., 2006). Because of their protein composition (300 to 450 g/kg), yeasts are rich in limiting amino acids such as lysine, threonine, and methionine, in addition to vitamin B complex (Ezequiel et al., 2000).

Yeast protein is classified as having high rumen degradable protein (RDP), which may result in increased use of readily available energy sources such as starch for the synthesis of microbial proteins, improving protein and energy synchronization. Yeast protein has a high ruminal degradability compared with soybean meal (990 g/kg vs. 790 g/kg of RDP) (Marcondes et al., 2009). As a consequence, Rufino et al. (2012) observed that the greatest amount of inactive dry yeast (IDY, *Saccharomyces cerevisiae*) used to substitute soybean meal resulted in the greatest ruminal ammonia production in lambs. IDY is marketed by the ethanol and sugar producing industries because the surplus from yeast mash can be used as an alternative ingredient in ruminant diets. Evaluations of nutrient use by livestock and the resulting rumen-microorganism interactions are needed to better understand the role of this potential ingredient. These data can be obtained through digestion studies, which analyze the possibility of replacing a high-cost conventional feed with a by-product that would otherwise be considered an environmental contaminant.

Thus, the present study aimed to evaluate the potential nutritive value of IDY as a protein ingredient in ruminant diets by assessing the intake, total and partial digestibility, pH, ruminal ammonia nitrogen, microbial efficiency (Emic), nitrogen balance (NB), average daily gain (ADG), carcass average daily gain (CADG), carcass yield, and feed conversion when using different soybean meal replacement levels in the diet of Nelore cattle.

2. Material and methods

2.1. Experimental area and climatic conditions

This experiment was conducted in the Experimentation, Research and Extension Center of Triângulo Mineiro (Central de Experimentação, Pesquisa e Extensão do Triângulo Mineiro, CEPET) of the Federal University of Viçosa (Universidade Federal de Viçosa, UFV), Brazil, from June to September of 2009. The CEPET is located at an average altitude of 620.2 m, 18.41°S latitude and 49.34°W longitude. The climate is classified by Köppen standards as Aw, i.e., hot and humid, with the temperature of the coldest month above 18 °C, a rainy season in the summer and a dry season in the winter, with an annual average precipitation of between 1,400 and 1,600 mm.

2.2. Experimental diets

The diets were formulated to meet beef cattle requirements of 1 kg of daily gain according to the National Research Council (NRC, 1996). The forage:concentrate ratio was 60:40 on a dry matter (DM) basis with corn silage as the forage. The diets consisted of five concentrate replacement levels of soybean meal by IDY, 0, 250, 500, 750, and 1000 g/kg on DM basis. The proportions of concentrate ingredients are shown in Table 1, and the chemical composition of the concentrates and corn silage is shown in Table 2.

2.3. Animals, management, and sample collection

The management and care of animals were performed in accordance with the guidelines and recommendations of the Committee of Ethics on Animal Studies at the Federal University of Vicosa (UFV), MG, Brazil. To determine nutrient intake and productive performance, 35 Nelore bulls with an average initial body weight (BW) of 370 ± 42 kg were distributed throughout a completely randomized experimental design, with five treatments and seven replicates (EXP 1). The trial lasted 98 days and was divided into a 14-day adaptation period and three experimental periods of 28 days each. Five animals were slaughtered at the end of the adaptation to estimate the initial carcass weights of all steers at the beginning of the experiment. They represented the mean of the total group.

Table 1
Ingredient proportion in experimental concentrate (g/kg, wet basis).

Ingredients	Replacement levels (IDY replacing SBM g/kg)				
	0	250	500	750	1000
Ground corn	774.7	776.9	779.0	781.2	783.4
Soybean meal (SBM)	184.2	138.5	92.7	46.5	–
Inactive dry yeast (IDY)	–	43.7	87.5	131.5	175.9
Urea/AS[a]	10.9	12.6	14.1	15.7	17.4
Wheat meal	12.5	10.7	9.0	7.3	5.5
Lime	5.5	5.5	5.5	5.5	5.6
Dicalcium phosphate	7.5	7.5	7.5	7.5	7.5
Sodium chloride	4.4	4.4	4.4	4.4	4.4
Mineral mix[b]	0.3	0.2	0.3	0.3	0.3

[a] Proportion between urea and ammonium sulphate (AS) was 9 parts of urea and 1 part of AS.
[b] Copper sulfate (225 g/kg), cobalt sulfate (14 g/kg), zinc sulphate (754 g/kg), potassium iodate (5 g/kg) and sodium selenite (2 g/kg).

Animals were weighed at the end of the adaptation period and, following 14 h of fasting, were also weighed after the last experimental period. Feed was provided in two daily meals at 8h00 and 15h00, and the orts were weighed daily to obtain a maximum of 50 to 100 g/kg of total feed provided as fresh feed. The orts were sampled, placed in labeled bags, and stored in the freezer for later analysis. The silage and concentrates were sampled three times per week.

All steers were slaughtered at the end of the experiment to determine the dressing percentage, which was calculated as the percentage between the hot carcass weight (HCW) and the final live weight (FLW) after fasting. Thus, the carcass average daily gain (CADG) was calculated by using the following equation:

$$\text{CADG}\left(\frac{\text{kg}}{\text{day}}\right) = \frac{\left[\left(\text{FLW} \times \left(\frac{\text{DRS}f}{100}\right)\right) - \left(\text{ILW} \times \left(\frac{\text{DRS}i}{100}\right)\right)\right]}{n}$$

where FLW is the final live weight after fasting (kg), ILW is the initial live weight after fasting (kg), DRS*f* is dressing percentage after slaughter at the end of the experiment, DRS*i* is dressing percentage after slaughtered at the end of the adaptation period (beginning of the experimental periods), and "*n*" is the number of evaluated days.

Five cannulated Nelore steers were used to determine the total and partial digestibility, nitrogen use efficiency, and ruminal pH and ammonia levels. The animals were fistulated in the rumen and abomasum, and each steer had an average initial BW of 320 ± 39 kg. They were distributed across a 5 × 5 Latin square design (EXP 2), balanced for residual effects (Lucas, 1957). The feed was provided in two daily meals at 8h00 and 15h00 in a sufficient quantity to obtain 5 to 10% orts. The five experimental periods lasted 18 days and were divided as follows: 10 days for adaptation to the diets; 6 days to collect abomasal digesta, feces, supplied feed, and ort samples; 1 day to collect ruminal fluid at 0, 2, 4, and 6 h after the morning meal; and 1 day for urine spot sample, obtained by massage of the external genitalia of the steers. Blood collection was made by jugular vein puncture 4 h after the morning feeding. Orts were removed for each animal each day before the morning meal and were weighed, sampled, placed inside plastic bags, and frozen at −18 °C. The supplied feed was sampled three times each week and was also placed inside plastic bags and frozen at −18 °C.

Table 2
Chemical compositions of corn silage and concentrates of each treatment (g/kg DM).

Items[b]	Corn silage	Replacement levels (IDY replacing SBM g/kg)[a]				
		0	250	500	750	1000
DM	313.4	884.9	885.6	888.1	890.2	893.8
OM	966.0	956.4	958.3	945.8	956.3	952.4
CP	74.4	226.4	221.0	228.9	228.9	223.3
NDIN	93.7	118.3	117.4	118.7	113.0	73.2
ADIN	48.0	15.1	14.7	16.9	11.2	8.3
EE	22.6	30.9	28.3	26.4	24.8	20.0
NFC	316.8	524.9	517.6	509.7	540.0	609.2
NDF	567.6	221.6	233.8	261.5	248.7	183.8
aNDFom(n)	552.2	191.2	210.9	235.0	222.9	166.7
iNDF	214.0	20.8	20.9	18.1	19.0	14.7
ADF	302.1	35.5	30.8	28.0	21.7	13.4
HEM	265.5	186.1	203.0	233.5	227.0	170.4
CEL	257.3	30.3	23.2	19.5	16.8	10.1
LIG	42.8	4.2	6.3	7.2	3.7	2.7

[a] IDY–inactive dry yeast; SBM–soybean;
[b] DM: dry matter, OM: organic matter, CP: crude protein, NDIN: neutral detergent insoluble nitrogen (g/kg N), ADIN: acid detergent insoluble nitrogen (g/kg N), EE: ether extract, NFC: non-fibrous carbohydrates, NDF: neutral detergent fiber, aNDFom(n): neutral detergent fibre assayed with a heat stable amylase and corrected for ash and nitrogenous compounds, iNDF: indigestible NDF, ADF: acid detergent fiber, HEM: hemicellulose, CEL: cellulose and LIG: lignin.

Chromium oxide (Cr_2O_3) was used to determine the fecal excretion and abomasum flow of nutrients, and was administered in a single daily 15 g dose at 11h00 via rumen fistula on the fourth day of each experimental period. Daily collections of 500 mL of abomasal digesta and 200 g of feces were performed every 22 h between the 11th and 16th day of each experimental period. Feed samples, orts, feces, and abomasal digesta were placed in labeled plastic bags and stored at −18 °C for later analysis.

Ruminal fluid was collected to measure the pH and ammonia nitrogen ($N—NH_3$) concentrations, and these samples were collected at 0, 2, 4, and 6 h after the morning feeding on the 17th experimental day of each period. The pH was immediately measured after collection; 1 mL of 1:1 sulfuric acid (H_2SO_4) was added to the sample, which was then stored in the freezer at −18 °C for later ruminal $N—NH_3$ concentration analysis.

Four hours after the diet was consumed on the 18th day of each experimental period, blood was collected by jugular vein puncture in a test tube containing a separation gel with a coagulant activator (SST II Advance, BD Vacutainer, São Paulo city, São Paulo state, Brazil). These samples were stored at −15 °C for later urea analyses. Concomitantly, 50 mL urine spot samples were obtained for each animal. The urine was filtered, and 10 mL aliquots were removed and immediately diluted in 40 mL of 0.036 N H_2SO_4 to prevent bacterial destruction of the purine derivatives (PD) and uric acid precipitation. These samples were stored at −15 °C for later analysis of urea, creatinine, allantoin, and uric acid. An undiluted urine sample was stored for determining the total nitrogen compound yields.

At the end of each experimental period, the feed samples, orts, feces, and abomasal digesta were thawed and subjected to pre-drying at 60 °C for 72 h and were then ground in a Wiley-type knife mill (TE-625, TECNAL, Piracicaba, São Paulo, Brazil) with a 1 mm mesh. Composite samples were saved for each animal on a dry weight basis from each time period.

2.4. Chemical analysis

DM, organic matter (OM), and crude protein (CP) analyses were performed according to the AOAC (1990), method number 934.01 for DM, 930.05 for OM, and 981.10 for CP. Ether extract (EE) was analyzed by Soxhlet extraction with petroleum ether, according to the AOAC (1990), method number 920.39. The concentration of neutral detergent fiber was assayed with a heat-stable amylase and corrected for ash and nitrogenous compounds [aNDFom(n)] by using techniques described by Mertens (2002), with corrections for protein according to Licitra et al. (1996) and added thermostable alpha-amylase (Ankon Tech. Corp., Fairport, NY, USA). Lignin (sa) was extracted with sulfuric acid 720 mL/L (Van Soest and Wine, 1967). Neutral detergent insoluble nitrogen (NDIN) and acid detergent insoluble nitrogen (ADIN) (Licitra et al., 1996) were measured using the Kjeldahl method. Non-fibrous carbohydrates (NFC) were calculated as follows according to Hall (2000): NFC (g/kg) = 1000−[(CP–urea derived CP + urea) + NDFap + EE + ash], where: CP = crude protein; NDFap = neutral detergent fiber corrected for ash and protein; and EE = ether extract.

The chromium oxide content of feces and abomasal digesta was determined according to Williams et al. (1962), with an atomic absorption spectrophotometer. The flow of fecal dry matter (FDM) and abomasal dry matter (ADM) was calculated as follows:

$$\text{FDM or ADM}\left(\frac{\text{kg}}{\text{day}}\right) = \frac{\text{ingested marker (g)}}{\text{fecal or abomasal marker concentration}} \times 100$$

The coefficients of ruminal and intestinal apparent digestibility of DM, OM, NDFap, and NFC were calculated in relation to the total digestible DM, whereas the ruminal and intestinal digestibility of CP and EE was calculated in relation to the amounts that arrived at each study site.

The ammonia nitrogen concentration of ruminal fluid samples was determined by using the colorimetric technique described by Chaney and Marbach (1962). Creatinine analysis was performed with acid picrate using a commercial kit (Labtest Diagnóstica, Uréia CE, Lagoa Santa, Minas Gerais, Brazil), according to the modified diacetyl method. Daily creatinine excretion was estimated based on the 27.76 mg/kg BW recommendations (Rennó et al., 2000). Daily urine volume was estimated by dividing daily creatinine excretion by its concentration in the urine spot sample. The determination of urea in urine was performed with an enzymatic colorimetric method using a commercial kit (Labtest Diagnóstica, Urea CE, Lagoa Santa, Minas Gerais, Brazil). The NB was calculated in g/day as follows: N intake–(fecal N + urine N).

Analyses of PD, allantoin, and uric acid were performed using a colorimetric method by Fujihara et al. (1987), which was described by Chen and Gomes (1992). PD excretion was calculated by multiplying the urine volume, which was estimated within 24 h, by the PD concentration of the spot urine samples. Absorbed purines (Y, mmol/day) were calculated from the PD excretion (X, mmol/day) by using the equation $Y = 0.85X + 0.385\ LW^{0.75}$, in which 0.85 is the recovery of absorbed purines as PD and $0.385\ LW^{0.75}$ is the endogenous contribution to purine excretion (Verbic et al., 1990). The production of microbial nitrogen compounds (N_{mic}) was calculated as:

$$N_{mic}\left(\frac{\text{gN}}{\text{day}}\right) = \frac{70 \times \text{absorbed } PD}{0.93 \times 100 \times 0.137}$$

where 70 is the N content of purines (mg N/mmol), 0.93 represents the true digestibility of purines, and 0.137 is the average N-purines:N-total ratio in the bacteria that were isolated from the rumen (Barbosa et al., 2011).

Table 3
Average daily intake of dry matter (DM) and nutrient and productive performance of growing beef cattle fed with inactive dry yeast (IDY) in concentrate (EXP 1).

Items[a]	Replacement levels (g/kg)						Effect (P-value)	
	0	250	500	750	1000	SEM	Linear	Quadratic
DM, kg/d	9.6	9.5	9.4	9.2	8.4	0.14	0.003	0.110
CP, kg/d	1.3	1.2	1.2	1.1	0.9	0.03	<0.001	0.308
aNDFom(n), kg/d	3.5	3.6	3.5	3.9	3.8	0.06	0.043	0.795
DM, g DM/kg BW	21.4	21.1	21.4	20.9	19.2	0.02	0.004	0.065
aNDFom(n), g DM/kg BW	8.0	8.1	8.1	9.0	8.9	0.02	0.011	0.694
ADG, kg/d	1.3	1.3	1.3	1.2	1.1	0.04	0.028	0.531
CADG, kg/d	0.9	0.9	1.0	0.9	0.9	0.02	0.709	0.144
Dressing, g/kg	558	567	572	556	562	2.7	0.904	0.219
FE, g gain/kg DMI	141	139	130	128	128	0.25	0.282	0.687

[a] DM: dry matter; CP: crude protein; aNDFom(n): neutral detergent fibre assayed with a heat stable amylase and corrected for ash and nitrogenous compounds; ADG: average daily gain; CADG: carcass average daily gain; CY: carcass yield (%); FE: feed efficiency.

2.5. Statistical analyses

The nutrient intake and animal performance analyses in EXP 1 were subjected to analyses of variance in a completely randomized design with five treatments, namely, 0, 250, 500, 750, and 1000 g/kg soybean meal replacement with IDY and seven replicates. The initial BW of steers was considered to be a covariate in the statistical model, which employed the PROC MIXED option in SAS software (version 9.1), according to the following model:

$$Yi = \mu + Ti + \beta(Xi - \bar{X}) + ei$$

where: Yi = dependent variable corresponding to experimental observations; μ = general mean; Ti = fixed effect of treatments i; β = regression coefficient or functional relationship with covariate; Xi = observed value of covariate applied to experimental unit; $\bar{x}$ = mean value for covariate; and ei = random error assuming normal independent distribution (NID) (0; $\sigma 2\varepsilon$).

In EXP 2, which assessed total and partial digestibility, nitrogen efficiency, and ruminal pH and ammonia nitrogen, the analyses were conducted in a 5 × 5 Latin square design with the PROC MIXED option in SAS software (version 9.1), according to the following model:

$$Y_{ijk} = \mu + Ti + Aj + Pk + eijk$$

where Y_{ijk} = dependent variable measured in animal j that was subjected to the i treatment in period k; μ = general mean, Ti = fixed effect of treatment, Aj = random effect of animal j, Pk = random effect of period k, and $eijk$ = random error assuming NID (0; $\sigma 2\varepsilon$).

In this case, the fixed effect was represented by soybean meal replacement levels by IDY and the random effects were represented by animal and period effects. The fixed effects for evaluating ruminal pH and ammonia nitrogen in the 5 × 5 Latin square were the replacement level of soybean meal by IDY, collection time (T), and the interaction between these two factors ($Y \times T$). Period and animal were considered to be random effects within the model. A scheme of repeated time measurements was used (Littell et al., 1998), with collection times (0, 2, 4, and 6 h after feeding) repeated once within each experimental unit (animal × period).

Replacement level comparisons followed the decomposition of orthogonal polynomials in linear, quadratic, cubic, and quartic effects and were conducted using PROC MIXED in SAS software (version 9.1). Homogeneity of variances between treatments was assumed and the degrees of freedom were estimated by using the Kenward–Roger method. The regression models were adjusted according to the significance of the β_1, β_2, β_3, and β_4 parameters by using a restricted maximum likelihood method in PROC MIXED, and the parameter estimates were obtained through PROC REG in SAS software (version 9.1). The same procedure was performed to obtain the linear, quadratic, and cubic effects of collection times on the ruminal pH and ammonia nitrogen data. All statistical procedures were conducted by using 0.05 as the critical probability level for a type I error.

3. Results

3.1. Nutrient intake (EXP 1)

The intake of DM ($P = 0.003$) and CP ($P < 0.001$) in kg/day and DM in g/kg BW decreased ($P = 0.004$) linearly as the dietary IDY increased (Table 3). Conversely, the average intake of NDFap in g/day ($P = 0.043$) and g/kg BW increased ($P = 0.011$) as IDY was added to the concentrate. The intake of NFC and total digestible nutrients (TDN) showed no difference ($P > 0.05$) between the treatments (Table 3).

Table 4
Average total, ruminal and intestinal apparent digestibilities in growing beef cattle fed with inactive dry yeast (IDY) in concentrate (EXP 2).

Items[a]	Replacement levels (g/kg)[b]						Effect (P-value)	
	0	250	500	750	1000	SEM	Linear	Quadratic
	Total apparent digestibility (g/kg)[c]							
DM	620.9	671.1	647.1	658.9	660.1	12.6	0.731	0.766
OM	640.3	691.5	670.8	681.7	685.9	12.2	0.652	0.762
CP	622.0	632.3	605.0	611.3	612.6	15.3	0.998	0.876
EE	736.8	726.0	734.3	731.1	698.5	20.3	0.949	0.919
aNDFom(n)	593.3	620.6	552.7	636.0	602.9	16.5	0.745	0.760
NFC	745.4	764.5	794.2	716.8	776.8	10.8	0.933	0.907
	Ruminal apparent digestibility (g/kg)[d]							
DM	554.1	57.56	555.6	562.5	587.2	11.1	0.220	0.982
OM	635.6	626.1	604.3	632.5	644.9	10.0	0.309	0.693
CP	68.3	53.1	24.7	45.0	25.6	6.70	0.088	0.770
EE	204.1	207.4	220.9	227.3	210.2	11.8	0.183	0.771
aNDFom(n)	821.5	804.6	853.9	791.3	832.3	14.1	0.886	0.851
NFC	615.6	512.0	532.5	519.6	633.5	21.4	0.326	0.186
	Intestinal apparent digestibility (g/kg)[e]							
DM	445.9	424.4	444.4	437.5	412.8	11.1	0.220	0.982
OM	364.4	373.9	395.7	367.5	355.1	10.0	0.309	0.693
CP	595.3	611.9	595.5	594.2	603.4	14.7	0.660	0.809
EE	669.5	658.6	658.7	650.9	610.9	25.4	0.756	0.939
aNDFom(n)	178.5	195.4	146.1	208.7	167.7	14.1	0.886	0.851
NFC	384.4	488.0	467.5	480.4	366.5	21.4	0.326	0.186

[a] DM: dry matter, OM: organic matter, CP: crude protein, EE: ether extract, NFC: non-fibrous carbohydrates, aNDFom(n): neutral detergent fibre assayed with a heat stable amylase and corrected for ash and nitrogenous compounds.
[b] Five experimental units per treatment (Latin square 5 × 5).
[c] Total apparent digestibility (g/kg) = [(Nutrient intake − Nutrient excretion)/Nutrient intake] × 1000.
[d] Ruminal apparent digestibility (g/kg) = [(Nutrient intake − Nutrient abomasal flow)/Nutrient intake] × 1000.
[e] Intestinal apparent digestibility (g/kg) = [(Nutrient abomasal flow − Nutrient excretion)/Nutrient abomasal flow] × 1000.

3.2. Animal performance (EXP 1)

IDY levels had no effect (P > 0.05) on productive performance variables, with the exception of the ADG, which linearly decreased (P = 0.028) as IDY was added to the concentrate (Table 3). The fitted equation for this variable was as follows: ADG = 1.39451 − 0.00269X (r^2 = 0.1490).

3.3. Digestibility, ruminal pH, and ammonia concentration (EXP 2)

The experimental diets had no effect (P > 0.05) on the total and partial apparent digestibility of nutrients (Table 4). No effects were found for the IDY level (P > 0.05) or the interaction between IDY level and collection time after feeding (P > 0.05) on N—NH_3 and pH (Table 5). Conversely, collection time after feeding had linear (P < 0.001) and quadratic effects (P < 0.001) on pH and N—NH_3 variables, respectively (Table 5). pH showed a decreasing linear trend (pH 6.15576 − 0.11732X) from time-point 0 (pH 6.18) until 6 h after feeding (pH 5.47). N—NH_3 exhibited quadratic behavior as a function of collection time

Table 5
Averages for pH, ruminal N-ammonia (N—NH_3, mg/dL) and efficiency of nitrogen usage in growing beef cattle fed with inactive dry yeast (IDY) in concentrate (EXP 2).

Items[a]	Replacement levels (g/kg)[b]						Effect (P-value)	
	0	250	500	750	1000	SEM	Linear	Quadratic
pH[c]	5.80	5.72	5.66	5.97	5.85	0.15	0.192	0.352
N—NH_3 mg/dL[d]	12.94	12.41	12.89	12.16	11.14	0.68	0.144	0.848
PUN, mg/dL	14.23	15.82	17.16	14.20	15.94	0.61	0.749	0.965
UUN, g/d	48.27	43.72	47.42	41.64	47.90	0.74	0.239	0.064
N_{mic}, g/d	91.37	81.96	87.78	82.66	84.36	1.58	0.064	0.308
Emic, gCP/kg TDN	138.43	122.26	157.15	154.05	166.42	8.85	0.572	0.293
NB, g/d	30.74	23.69	20.54	24.60	26.63	0.78	0.339	0.747

[a] Plasma urea nitrogen (PUN). Urinary urea nitrogen (UUN). Microbial nitrogen (N_{mic}). Microbial efficiency (Emic) and nitrogen balance (NB).
[b] Five experimental units per treatment (Latin square 5 × 5).
[c] Main effects relative to pH: replacement level (*L*) P = 0.342, collection time (*T*) P < 0.001 and *L* × *T* = 0.147.
[d] Main effects relative to NNH3: replacement level (*L*) P = 0.391, collection time (*T*) P < 0.001 and *L* × *T* = 0.944.
Slicing the effect relative to collection time (*T*) for pH, pH: linear (P < 0.001) quadratic (P = 0.1053) and cubic (P = 0.9612). Effects relative to collection time (*T*) for N—NH_3: linear (P < 0.001) quadratic (P < 0.001) and cubic (P = 0.5290).

(Table 5). By calculating the derivative of equation $N—NH_3 = 7.54381 + 2.72635X - 0.25516X^2$, the peak was obtained at 5.3 h with an expected maximum response of 14.82 mg of $N—NH_3$ per dL of ruminal fluid.

3.4. Nitrogen use efficiency (EXP 2)

No effect of IDY ($P > 0.05$) on variables related to dietary nitrogen efficiency was found (Table 5). A quartic effect was found for urinary urea nitrogen (UUN); however, this effect was not relevant because of its lack of biological significance.

4. Discussion

4.1. Nitrogen intake (EXP 1)

A linear decrease in DM intake with increasing replacement levels of soybean meal by IDY can be explained by the very fine texture of the yeast, which most likely hindered feed intake by the steers, as noted in the experimental phase, making the concentrate with high yeast levels more sticky relative to the concentrate containing only soybean meal. Tegbe and Zimmerman (1977) also found that including IDY in the diet modified the texture of pig feed, giving a sticky consistency in the pigs' mouths and hindering ingestion. Prado et al. (2000) assessed DM intake in finishing heifers that received diets in which cottonseed meal was replaced by IDY, and they also found a reduction in DM intake with an increase in IDY levels.

Linear decreases also occurred in OM and CP intakes. These results are most likely due to the DM intake and also to the experimental diet composition (Table 2), which displayed lower concentrations of true CP and EE with increased amounts of IDY in the concentrate. The inclusion of yeast decreased the proportion of true CP because of the lower concentration of yeast relative to soybean meal. It is worth noting that the inclusion of a higher proportion of urea (Table 1) in diets with higher IDY levels was used to formulate isoproteic diets. The increasing levels (109, 126, 141, 157, and 174 g/kg DM) of urea + ammonium sulphate (9:1) were used to make up the total protein content. However, the higher non-protein nitrogen (NPN) and true protein ratio in the diets with higher IDY inclusion levels seem to have significantly contributed to a reduction in the intake of DM and CP. Urea could probably depress intake by reducing the palatability of the diets with higher inclusion levels of IDY and urea.

The NDFap intake increased with an increase of IDY in the diet. The reduction of DM intake at the highest IDY levels most likely occurred through a selective reduction in concentrate intake. The likely maintenance of forage intake with a reduction in concentrate acceptability could explain the linear increase in NDFap intake, through a simple dilution effect. The NFC intake remained unchanged due to a greater NFC content in the concentrates (Table 2), which resulted in a lower intake for greater IDY inclusion levels.

Although increased IDY levels affected the intake of almost all nutrients (Table 3), this increase did not result in differences in apparent digestibility coefficients. According to Van Soest (1994), the digestibility of feeds is directly influenced by factors such as ingestion and feed composition, feed processing, the protein:energy ratio, degradability rate, and inherent animal factors. It is known that NDF exerts a significant effect on intake regulation and is responsible for the physical limitation of dietary intake. However, when dietary NDF is below 500–600 g/kg DM, the intake is not highly correlated with digestibility (Mertens, 1994) which may have occurred in the present experiment.

4.2. Animal performance (EXP 1)

Animal performance parameters such as meat and milk production are direct functions of DM intake, as a consequence of a greater or lesser supply of nutrients (Mertens, 1994). The linear decrease in ADG with increased IDY in the diets directly reflects the effect found for DM intake. However, a linear decrease in DM intake showed no effect on the average carcass gain, carcass yield, or feed conversion.

The CADG is an economically important variable for producers of beef cattle because the producer's compensation by the packing plant is based on the HCW. The carcass yield and feed conversion also showed no differences between treatments. Therefore, one can infer that yeast can potentially replace soybean meal in the diets of finishing Nelore cattle, although this replacement is conditioned by economic factors.

4.3. Digestibility, ruminal pH, and ammonia concentration (EXP 2)

Average ruminal digestibility values of 567.0 and 628.7 (g/kg) were found for DM and OM, respectively. The ruminal digestibility of OM was close to the average value of 650 g/kg that was found by the British Agricultural Research Council (ARC) (ARC, 1984) for different feeds. Although yeast is a source of RDP, no difference in the ruminal digestibility of this nutrient was observed, which can be explained by its low intake by the animals, possibly preventing greater levels of degradation. The average rumen CP digestibility of 43.5 g/kg indicates that fluid losses of nitrogen components in the rumen were low and that nitrogen sources were used to a greater extent by microorganisms.

Fregadolli et al. (2001) found that ruminal and intestinal digestibility were not affected by the inclusion of a highly degradable nitrogen source, as in the present study. These authors found average values of 541 and 639 g/kg for the ruminal

digestibility of DM and OM, respectively. As for CP, a negative value was found for ruminal digestibility, with an average value of −185.0 g/kg; those authors attributed this value to the endogenous N recycling in animal rumens as well as to microbial protein synthesis.

An average value of 830 g/kg for NDFap ruminal digestibility is within the range commonly found in the literature; for example, Chizzotti et al. (2005) observed an average value of 890 g/kg in diets exhibiting a forage:concentrate ratio of 60:40. The digestibility of NDFap in the intestines had an average value of 179.3%, which was expected because NDFap digestibility is usually low. Messana et al. (2009) also found that yeast had effect on total and ruminal nutrient digestibility coefficients in Nelore cattle diets containing cottonseed processing by-products as forage.

An average value of 12.31 mg/dL NH_3—N in ruminal fluid (Table 5) was found in this study for the different treatments, a value that is above the minimum concentration of 5 mg/dL recommended by Satter and Slyter (1974) as an adequate value for satisfactory *in vitro* microbial growth. Hoover (1986) proposed values of 3.3 and 8.0 mg NH_3—N for the maximization of microbial growth and OM digestion in the rumen, respectively. Mehrez et al. (1977) found that the minimum ammonia concentration for maximum microbial growth should be 23 mg/dL. Conversely, Leng and Nolan (1984) found that concentrations above 5–10 mg/dL of ruminal fluid did not lead to an increase in microbial protein production. Sampaio et al. (2009) and Lazzarini et al. (2009) assessed the effects of nitrogen supplementation in animals fed with low-quality forage and found ruminal ammonia concentration values of 9.64 and 15.33 mg/dL, respectively; these values maximized the voluntary feed intake of the animals and consequently affected their performance.

The pH decreased linearly with rumen fluid collection time (Table 5), with estimated values of 6.15 and 5.45 at time-points 0 and 6 h after feeding, respectively. pH values below 6.2 can inhibit digestion rate and increase the lag time for cell wall degradation (Van Soest, 1994). However, Hoover (1986) proposed that only pH values below 5.0–5.5 could inhibit the development of cellulolytic microorganisms. Nevertheless, the ruminal digestibility of NDFap was not affected by increasing levels of yeast (Table 4), indicating that there was likely satisfactory growth of cellulolytic microorganisms and that these low pH values seemed to not affect fiber degradation in any of the diets under study.

4.4. Nitrogen use efficiency (EXP 2)

Urea is the primary form of N excretion in mammals. The urea concentration of blood plasma is well established as a parameter for assessing dietary CP use inefficiency. The presence of high proportions of nitrogen in body fluid indicates that, in the rumen at least, there is low ammonia utilization and that it is being absorbed by the epithelium of the organ (Broderick and Clayton, 1997). This study's average plasma urea nitrogen (PUN) concentration of 15.55 mg/dL is within the normal limits for cattle. Urea is one metabolite of dietary protein that is formed from detoxification of NH_4 by the liver. Levels of urea in the plasma or serum (PUN or SUN) are reflective of the quantity and degradability of the protein consumed, of the severity of negative energy balance in fasted animals, or of the combination of protein feeding and negative energy balance. PUN concentrations have often been used as a correlate for dietary protein level and fertility (Elrod and Butler, 1993). Ferguson et al. (1991) have suggested that when SUN concentrations are > 20 mg/dL, fertility will be impaired. Although an effect of different diets on CP intake and ruminal ammonia concentration was detected, and these two parameters are the main factors that influence the amount of nitrogen present in body fluids, the PUN concentration was not altered.

When the synthesis rate of ammonia exceeds its utilization by microorganisms, an increase in ruminal ammonia concentration occurs, with a consequent increase in urinary urea excretion (Russell et al., 1992). The lower CP intake that was recorded in animals receiving higher IDY levels seems to have been compensated for by a numerically greater effect on Emic in these animals, which was reflected in the similar concentrations of ruminal PUN, UUN, and NH_3—N for animals in the different treatment groups. An average value of 157.1 g CP/kg TDN was estimated for Emic in the present study, which is greater than the value of 120 g CP/kg TDN that was established as a reference for tropical conditions by Valadares Filho et al. (2010) and is also greater than the value of 130 g CP/kg TDN reported by the US NRC (2001).

5. Conclusions

The replacement of soybean meal by IDY in feed concentrate has emerged as a feasible alternative for beef cattle producers. However, its use should be conditional on product availability and price. Although increased levels of IDY in the concentrate resulted in reduced intake and ADG, IDY allowed satisfactory feed conversion and carcass gain. IDY altered DM intake in beef cattle in the feedlot as well as ADG, without affecting the apparent digestibility of nutrients or microbial efficiency.

Acknowledgements

The authors wish to thank the Conselho Nacional de Pesquisa e Desenvolvimento Científico e Tecnológico (CNPq), Fundação de Amparo à Pesquisa do Estado de Minas Gerais (FAPEMIG), and Instituto Nacional de Ciência e Tecnologia Ciência Animal (INCT-CA) for financial support.

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