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Performance responses of broilers fed diets containing high-protein dried distillers' grains

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ABSTRACT - The objective of this study was to evaluate the inclusion of high-protein dried distillers' grains (HP-DDG) in broilers' diets. A total of 1,200 one-day-old male chicks were weighed and distributed among six treatments, with ten replicates of 20 birds each. The experimental period was divided into three phases: starter (1–21 days), grower (21–33 days), and finisher (33–43 days). The treatments consisted of six dietary levels of HP-DDG in the starter, grower, and finisher phases as follows: T1: 0%; T2: 3, 5, and 7%; T3: 5, 7, and 9%; T4: 7, 9, and 11%; T5: 9, 11, and 13%; and T6: 11, 13, and 15%. Body weight (BW), BW gain (BWG), feed intake (FI), and feed conversion ratio (FCR) were measured at 21, 33, and 43 days of age. In addition, carcass and cut yields were assessed at 43 days. Data were submitted to ANOVA and analyzed using Dunnett's test ($\alpha = 0.05$). In the starter phase, birds fed diets with 11% had lower BW ($P = 0.032$) and BWG ($P = 0.029$) than the control group. Overall performance from 1 to 33 days and from 1 to 43 days of age was not affected by HP-DDG levels ($P>0.05$). Carcass and cut yields at 43 days were not affected by HP-DDG ($P>0.05$). In conclusion, the gradual inclusion of HP-DDG in broiler diets up to 15% does not impair overall performance or carcass yield. However, inclusion levels should not exceed 9% during the starter phase to avoid initial growth loss.

Keywords: corn, ethanol coproducts, growth, HP-DDG, poultry, sustainability

1. Introduction

The global expansion of ethanol production, driven by the search for renewable energy sources and the depletion of fossil fuel reserves, has increased the availability of corn-based coproducts such as dried distillers' grains with solubles (DDGS) for use in animal feed (Kumar and Singh, 2019). This scenario represents not only a response to energy demand but also an opportunity to promote a circular economy and sustainability, by transforming residues from biofuel production into valuable inputs for the feed industry. In Brazil, corn ethanol production is rapidly growing, and the poultry industry is particularly interested in these coproducts as alternative ingredients that can reduce feed costs and improve the sustainability of intensive production systems.

However, conventional DDGS presents limitations in poultry nutrition due to its high fiber content and variable nutrient digestibility, which restrict its inclusion levels in broiler diets (Abudabos et al., 2017a,b; Shurson, 2018; Hristakieva et al., 2023). Negative effects on performance have been reported at higher dietary concentrations (above 9%), particularly during the starter phase when the digestive tract is still developing (Lumpkins et al., 2004; Abdel-Raheem et al., 2011; Loar et al., 2012).

Emerging technologies, such as fiber separation technology (FST™; ICM Inc., Colwich, KS, USA), remove fiber before fermentation, improving ethanol yield and creating a coproduct known as high-protein DDG (HP-DDG). Compared with conventional DDGS, HP-DDG presents a more concentrated nutritional profile, characterized by higher crude protein (CP; >38%), metabolizable energy (3060 kcal/kg), amino acids, and minerals (0.27% available phosphorus, 0.51% potassium, 0.07% sodium), and reduced fiber content (6.87%) (Applegate et al., 2009; Shurson, 2018, 2023; Dias et al., 2023; Calderano et al., 2024; VerBeek et al., 2024). These nutritional improvements suggest a potential for partial replacement of soybean meal (Fries-Craft and Bobeck, 2019), one of the costliest ingredients in poultry diets. Nonetheless, the nutrient composition of HP-DDG may vary depending on corn source and processing method, and its use in poultry diets depends on its actual digestible nutrient and energy content (Silva et al., 2022). In contrast to the United States, where the nutritional value of corn DDGS and HP-DDG has been more extensively characterized, Brazilian nutritionists still have limited familiarity with these coproducts. As a result, feeding recommendations in Brazil often rely on data generated in U.S. studies, which may not fully represent the characteristics of locally produced ingredients (Palowski et al., 2021).

This lack of knowledge represents a research gap that limits nutritionists' ability to fully incorporate HP-DDG into practical diet formulations. Generating scientific evidence on its effects is especially relevant in Brazil, where the rapid expansion of corn ethanol plants is expected to increase the availability of HP-DDG. In addition, evaluating this coproduct contributes to more sustainable poultry production, as it promotes the valorization of biofuel coproducts and reduces dependence on soybean meal.

We hypothesized that HP-DDG could be included in broiler diets up to a certain practical level without impairing growth performance or carcass yield when compared with a corn-soybean meal control diet. Therefore, the objective of this study was to evaluate the effects of increasing dietary HP-DDG levels by comparing each level with a 0% inclusion control, to determine the maximum inclusion that maintains broiler performance from 1 to 43 days of age.

2. Material and methods

2.1. Ethics Committee

Research on animals was conducted according to the Ethics Committee on Animal Use of the Universidade Federal de Viçosa (protocol number 034/2021).

2.2. Birds' husbandry

The experiment was conducted at the Research & Extension Sector for Poultry Production and Nutrition of the Animal Science Department, Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil.

Male broiler chicks (Cobb500™) were obtained from a commercial hatchery (Rivelli Alimentos SA, Matheus Leme, MG, Brazil), where all chicks were vaccinated against Marek's disease, Newcastle disease, and infectious bronchitis.

A total of 1,200 one-day-old male chicks (47.3 g ± 2.43 g) were weighed and distributed into six treatments with ten replicates of 20 birds each (35 kg/m^2). The experiment was conducted as a completely randomized design, with pens randomly assigned to one of the six dietary treatments. All pens were subjected to the same environmental and management conditions. Birds were reared in floor pens (1 × 2 m) covered with fresh wood shavings in a conventional poultry house equipped with lateral curtains and fans. Each pen was equipped with one tubular feeder and five in-line nipple drinkers. Experimental diets and water were provided *ad libitum* throughout the entire experimental period. The temperature and lighting programs were based on the breeder's guideline. Specifically, the temperature was initially set at 33 °C and then gradually reduced to 20 °C. The internal temperature of the house was monitored five times daily using digital thermometers placed inside the pens at the birds' height. The lighting schedule began with 24 hours of light for the first week and then was reduced to 16 hours per day from day 7 to day 43.

2.3. Experimental period and diets

The same batch of HP-DDG evaluated by Dias et al. (2023) was used in the present study. Its nutrient composition is shown in Table 1. Analyses were carried out by CBO Análises Laboratoriais (Valinhos,

Table 1 - Analyzed composition of high-protein dried distillers' grains (HP-DDG) on an as-fed basis

	HP-DDG[1]
Dry matter (%)	91.7
Crude protein (%)	42.6
Gross energy (kcal/kg)	5,358
Metabolizable energy (kcal/kg)[2]	3,058
Ether extract (%)	13.0
Starch (%)	5.48
Crude fiber (%)	6.06
Neutral detergent fiber (%)	24.1
Acid detergent fiber (%)	16.8
Ash (%)	2.21
Calcium (%)	0.005
Total phosphorus (%)	0.49
Sodium (%)	0.07
Magnesium (%)	0.10
Potassium (%)	0.56
Copper (mg/kg)	10.4
Iron (mg/kg)	157.5
Manganese (mg/kg)	6.23
Zinc (mg/kg)	49.7
Lysine (%)	1.36 (80.3)
Methionine (%)	0.91 (90.5)
Methionine + Cystine (%)	1.68 (85.9)
Threonine (%)	1.64 (75.6)
Arginine (%)	1.85 (89.6)
Histidine (%)	1.27 (84.9)
Isoleucine (%)	1.56 (86.4)
Leucine (%)	5.03 (90.6)
Valine (%)	2.10 (85.8)
Phenylalanine (%)	2.14 (88.7)
Phenylalanine + Tyrosine (%)	3.85 (94.7)
Alanine (%)	3.27 (88.0)
Aspartic acid (%)	2.78 (77.7)
Cystine (%)	0.77 (79.3)
Glutamic acid (%)	7.51 (88.9)
Glycine (%)	1.66 (75.4)
Serine (%)	2.12 (80.5)
Glycine + Serine (%)	3.78 (78.2)
Tyrosine (%)	1.71 (88.6)
Proline (%)	3.89 (87.0)

[1] Values in parentheses represent the digestibility coefficients determined by Dias et al. (2023).
[2] Nitrogen-corrected; determined through the total excreta collection method by Dias et al. (2023).

São Paulo, Brazil) using the following methods: crude protein determined by the Dumas method (N × 6.25); ether extract, crude fiber, and ash determined by gravimetry (specific to each nutrient); gross energy by bomb calorimetry; calcium, sodium, potassium, copper, iron, manganese, and zinc by atomic absorption; phosphorus by colorimetry; and amino acid profile by high-performance liquid chromatography (HPLC). The metabolizable energy value was determined using the total excreta collection method, whereas amino acid digestibility coefficients were obtained using the ileal digesta method, as described in detail by Dias et al. (2023). All results are expressed on an as-fed basis. The coproduct was obtained from a Brazilian company (F.S. Bioenergia Inc., Lucas do Rio Verde, Mato Grosso, Brazil) and produced using FST™ technology (ICM Inc., Colwich, KS, USA).

The experimental period was divided into three phases: 1) starter (1 to 21 days), 2) grower (21 to 33 days), and 3) finisher (33 to 43 days). Diets were based on corn and soybean meal and formulated

Table 2 - Experimental diets used and included high-protein dried distillers' grains (HP-DDG, %) in the starter (1 to 21 days), grower (21 to 33 days), and finisher (33 to 43 days) phases

Item	Starter		Grower		Finisher	
	0%	11%	0%	13%	0%	15%
Ingredient (%)						
Corn	52.1	50.1	57.5	53.1	63.7	57.0
Soybean meal	41.2	32.6	35.1	26.6	29.7	21.5
HP-DDG	0.00	11.0	0.00	13.0	0.00	15.0
Soybean oil	3.05	2.49	4.23	3.93	3.80	3.75
Dicalcium phosphate	1.24	1.04	0.93	0.68	0.54	0.25
Limestone	1.01	1.21	0.79	1.02	0.76	1.02
Salt	0.52	0.50	0.49	0.47	0.46	0.44
DL-Methionine, 99%	0.32	0.27	0.27	0.20	0.23	0.14
L-Lysine HCl, 78.5%	0.14	0.28	0.28	0.49	0.31	0.47
L-Threonine, 98%	0.05	0.05	0.04	0.02	0.03	0.00
Mineral premix[1]	0.13	0.13	0.13	0.13	0.13	0.13
Vitamin premix[2]	0.13	0.13	0.13	0.13	0.13	0.13
Choline chloride, 60%	0.10	0.10	0.10	0.10	0.10	0.10
Salinomycin, 12%	0.06	0.06	0.06	0.06	0.06	0.06
Phytase 50 g/t	0.01	0.01	0.01	0.01	0.01	0.01
BHT[3]	0.01	0.01	0.01	0.01	0.01	0.01
Total	100	100	100	100	100	100
Calculated composition						
Crude protein (%)	23.2	23.8	20.8	22.1	18.9	20.8
Metabolizable energy (kcal/kg)	3,000	3,000	3,150	3,150	3,200	3,200
Calcium (%)	0.937	0.937	0.758	0.758	0.634	0.634
Available phosphorus (%)	0.440	0.440	0.374	0.374	0.296	0.296
Sodium (%)	0.218	0.218	0.208	0.208	0.197	0.197
Potassium (%)	0.923	0.823	0.829	0.735	0.752	0.664
Chlorine (%)	0.374	0.358	0.362	0.342	0.349	0.325
Crude fiber (%)	2.90	3.12	2.70	3.00	2.55	2.94
Neutral detergent fiber (%)	12.8	14.0	12.7	14.1	12.8	14.4
Acid detergent fiber (%)	4.84	5.96	4.54	5.93	4.32	5.99
SID Lysine (%)	1.256	1.256	1.124	1.124	1.014	1.014
SID Methionine (%)	0.624	0.598	0.554	0.516	0.492	0.443
SID Methionine + Cystein (%)	0.929	0.929	0.832	0.832	0.750	0.750
SID Threonine (%)	0.829	0.829	0.742	0.742	0.669	0.676

[1] Trace mineral premix provided per kg of premix: Mn, 58.36 g; Fe, 41.68 g; Zn, 54.21 g; Cu, 8.31 g; I, 0.84 g; Se, 0.25 g.
[2] Vitamin premix provided per kg of premix: vitamin A, 9,638,000 IU; vitamin D3, 2,410,000 IU; vitamin E, 36,100 IU; vitamin B1, 2.60 g; vitamin B2, 6.45 g; vitamin B6, 3.61 g; vitamin B12, 15.9 mg; vitamin K3, 1.94 g; pantothenic acid, 12.95 g; nicotinic acid, 39.20 g; folic acid, 0.90 g; biotin, 89.80 mg.
[3] Antioxidant butylhydroxytoluene.

according to Rostagno et al. (2017) to meet all nutritional requirements for each phase. The treatments were determined through different inclusion levels of HP-DDG in each phase: starter phase – 0, 3, 5, 7, 9, and 11%; grower phase – 0, 5, 7, 9, 11, and 13%; and finisher phase – 0, 7, 9, 11, 13, and 15%. For each phase, two basal diets were formulated: a control diet (0% HP-DDG) and a diet with the maximum level of HP-DDG for that phase (Table 2). The intermediate treatment diets were then obtained by mixing these two basal diets in specific proportions to achieve the target inclusion levels.

2.4. Broiler performance and carcass yield

Birds, feed, and leftover feed were individually weighed on days 1, 21, 33, and 43 to determine body weight (BW, g/bird), body weight gain (BWG, g/bird), feed intake (FI, g/bird), and feed conversion ratio (FCR, g/g). In cases of broiler mortality, dead bird was removed from its experimental unit, and the feed was weighed to correct FI values.

At 43 days, two birds per replicate with a BW within ±5% of the pen's average were selected and subjected to a 12 h feed withdrawal period. After that, birds were stunned by electronarcosis and slaughtered at the Unit of Teaching, Research, and Extension "Frigorífico Escola" to measure carcass and cut yields. After the removal of feathers, viscera, head, neck, and feet, the carcasses were weighed, and carcass weight was divided by live body weight to determine carcass yield (CY, %). Subsequently, breast fillet (BY, %), wings (WY, %), drumsticks (DY, %), and thighs (TY, %) were removed and weighed to determine their respective yields.

2.5. Statistical analysis

Data were analyzed by one-way ANOVA using the Exp.Des.pt package from the R statistical software (R software v.4.0.4). Prior to analysis, the assumptions of normality of residuals and homogeneity of variances were verified by the Shapiro-Wilk and Bartlett's tests, respectively. Dunnett's test (P≤0.05) was used to compare each HP-DDG treatment against the control group. The statistical model was:

$$Y_{ij} = \mu + T_i + \varepsilon_{ij} \quad (1)$$

in which Y_{ij} is the observation in the j-th replicate of the i-th treatment, μ is the overall mean, T_i is the fixed effect of treatment, and ε_{ij} is the random error. For performance parameters, each replicate was considered an experimental unit, whereas for carcass and cut yields, 2 birds per replicate were the experimental unit.

3. Results

Birds fed 11% HP-DDG in the starter diet had a 3% lower BWG (P = 0.011) and a 2.8% lower BW (P = 0.011) compared to those in the control treatment (Table 3). However, no effects were observed when analyzing the performance from 1 to 33 days and from 1 to 43 days (P>0.05). Additionally, carcass and cut yields at 43 days were not affected by treatments (P>0.05; Table 4).

4. Discussion

The starter phase is crucial for skeletal and gastrointestinal development, which directly impacts broiler growth. In this study, birds fed 11% HP-DDG had a 2.8% lower BW and a 3.0% lower BWG compared with the control group during the starter phase, indicating that high levels of inclusion in early life may compromise initial growth. This effect is likely due to the combination of higher dietary fiber from HP-DDG and the immature gastrointestinal tract in young chicks, which can transiently reduce nutrient utilization efficiency. Similar findings were reported by Fries-Craft and Bobeck (2019), who observed impaired performance with 15% HP-DDG during the starter phase. However, they also reported no differences with 10% inclusion, aligning with our findings that performance was maintained up to 9% inclusion. On the other hand, Jung and Batal (2010) reported no differences in broilers' performance when fed HP-DDG up to 16% during the starter phase.

Table 3 - Feed intake (FI), body weight gain (BWG), body weight (BW), and feed conversion ratio (FCR) of broilers fed different levels of high-protein dried distillers' grains (HP-DDG) from 1 to 43 days of age

	Inclusions of HP-DDG[1]						SEM	P-value
	1 to 21 days							
	0%	3%	5%	7%	9%	11%		
FI (g/bird)	1364	1383	1359	1372	1359	1371	0.006	0.877
BWG (g/bird)	1083	1093	1073	1060	1063	1050*	0.004	0.011
BW (g/bird)	1129	1139	1119	1107	1109	1097*	0.004	0.011
FCR (g/g)	1.260	1.265	1.267	1.294	1.279	1.306	0.005	0.085
	1 to 33 days							
	0%	3-5%	5-7%	7-9%	9-11%	11-13%		
FI (g/bird)	3338	3360	3369	3366	3351	3359	0.010	0.954
BWG (g/bird)	2368	2375	2393	2342	2365	2341	0.007	0.166
BW (g/bird)	2414	2422	2457	2388	2412	2387	0.007	0.195
FCR (g/g)	1.410	1.415	1.409	1.438	1.418	1.436	0.004	0.224
	1 to 43 days							
	0%	3-5-7%	5-7-9%	7-9-11%	9-11-13%	11-13-15%		
FI (g/bird)	5525	5501	5521	5532	5517	5543	0.016	0.985
BWG (g/bird)	3527	3467	3486	3505	3504	3489	0.007	0.254
BW(g/bird)	3574	3513	3549	3552	3551	3536	0.008	0.332
FCR (g/g)	1.567	1.587	1.585	1.578	1.575	1.588	0.004	0.669

SEM - standard error of the mean.
[1] Inclusion levels of HP-DDG (%) in the starter (1-21 d), grower (21-33 d), and finisher (33-43 d) phases, respectively: 0, 0, 0%; 3, 5, 7%; 5, 7, 9%; 7, 9, 11%; 9, 11, 13%; 11, 13, 15%.
* Means followed by an asterisk differ from the treatment without HP-DDG (0%) by Dunnett's test (α = 0.05).

Table 4 - Carcass yield (CY), breast fillet yield (BY), wings yield (WY), drumsticks yield (DY), and thighs yield (TY) of broiler chickens fed different levels of high-protein dried distillers' grains (HP-DDG) at 43 days of age

	0%	3-5-7%	5-7-9%	7-9-11%	9-11-13%	11-13-15%	SEM	P-value
CY (%)	79.7	79.7	79.2	80.0	80.9	79.4	0.261	0.531
BY (%)	34.7	34.7	34.8	33.7	34.7	34.5	0.209	0.681
WY (%)	9.41	9.61	9.56	9.47	9.48	9.55	0.046	0.852
DY (%)	14.7	14.8	14.7	14.7	14.7	14.9	0.094	0.965
TY (%)	12.7	12.6	12.7	12.9	12.7	12.7	0.068	0.930

SEM - standard error of the mean.
Inclusion levels of HP-DDG (%) in the starter (1-21 d), grower (21-33 d), and finisher (33-43 d) phases, respectively: 0, 0, 0%; 3, 5, 7%; 5, 7, 9%; 7, 9, 11%; 9, 11, 13%; 11, 13, 15%.

Interestingly, VerBeek et al. (2024) reported that turkey poults fed HP-DDG showed a temporary reduction in FI from 1 to 14 days, which was attributed to delayed pancreatic enzyme secretion during early development. However, this reduction did not affect BW and FI normalized in the subsequent phase (15 to 32 days). While caution is warranted when extrapolating across species, it is reasonable to suggest that a similar transient adaptation could have occurred in our broilers before 14 days of age. By 21 days, FI may have already normalized, which would explain why cumulative FI was not affected in our study. Therefore, the BWG reduction at 21 days can be interpreted as a short-term effect of digestive adaptation rather than a persistent impairment. Nonetheless, because BWG in broilers is particularly sensitive during the starter phase due to the accelerated development of the gastrointestinal tract and other organs (Shani and Irani, 2024), even a brief reduction in nutrient utilization efficiency at this stage could manifest as the modest reductions in BW and BWG observed at 21 days, without causing long-term impairment in overall performance.

Fries-Craft and Bobeck (2019) associated poor performance in the grower and finisher phases and throughout the overall period with the reduction in performance observed in the starter phase. However, our results showed that birds with lower BW and BWG in the starter phase did not experience performance impairment in the subsequent rearing phases. This suggests that birds were able to compensate for the initial growth reduction once the digestive tract matured and nutrient absorption normalized.

The inclusion of HP-DDG up to 16% decreased the performance of broiler chickens from 0 to 33 days old when compared with the control group without without HP-DDG inclusion (Jung and Batal, 2010). Moreover, it was reported that 15% and 20% HP-DDG negatively affected performance parameters in the grower phase (Fries-Craft and Bobeck, 2019). These findings diverge from the results of the present study and those of VerBeek et al. (2024) in turkey poults, where increasing levels of HP-DDG or its inclusion at 15% in the rearing phases had no negative effect when the period from 1 to 33 days of age was evaluated.

Overall, HP-DDG inclusion was not reported to affect broilers' performance from 0 to 42 days (Applegate et al., 2009), except for the studies by Fries-Craft and Bobeck (2019) and Mustafa et al. (2017). In the former, the authors stated that poor performance was linked to HP-DDG inclusion and its amino acid content. They observed that increasing levels of HP-DDG decreased Lys and Met + Cys levels in the diets, leading to a deficiency, which can occur due to the lower Lys content in HP-DDG compared with soybean meal (Dias et al., 2023). Therefore, there is a need for supplemental amino acids, especially Lys, when replacing soybean meal with HP-DDG. In the present study, diets were formulated to account for this requirement for supplemental amino acids, as we increased Lys supplementation as HP-DDG levels increased in the diets, which could explain why we did not find any negative effects on overall performance. In other words, the adequate adjustment of amino acid balance likely prevented the reductions observed in other studies. However, Mustafa et al. (2017) reported better BW, BWG, and FCR in broilers fed 7.5% HP-DDG at 42 days of age, which was attributed to improvements in villus height and crypt depth that increased the absorptive surface area.

Energy, AA, and fiber content differences can be observed among corn coproducts. Regarding AA and CP contents, soybean meal and HP-DDG used in the present study had similar CP content. On the other hand, both feedstuffs differed considerably in fiber content. The HP-DDG contained 6.06% CF, 24.14% NDF, and 16.88% ADF, whereas the soybean meal contained 4.82% CF, 11.6% NDF, and 7.07% ADF (Dias et al., 2023; Calderano et al., 2024). It is a characteristic of corn coproduct feedstuffs, in which the high dietary fiber content is mainly composed of non-starch polysaccharides (Knudsen, 2014; Sanchez et al., 2021). In poultry nutrition, dietary fiber is often considered antinutritive because of its limited contribution to energy supply and its disruption of digestive processes by increasing digesta viscosity in the intestinal tract and reducing nutrient availability and the overall nutritive value of the diets (Mateos et al., 2012; Sanchez et al., 2021). Therefore, it could impair broilers' growth by decreasing nutrient utilization, especially in the starter phase when the gastrointestinal tract is developing.

However, some authors suggest that structural materials, like fiber, stimulate the development and physiology of the gastrointestinal tract, especially the gizzard, and stimulate the secretion of HCl, bile acids, and enzymes (Mateos et al., 2012; Njeri et al., 2023). Njeri et al. (2023) found that broilers fed a diet with high fiber content, represented by the addition of 10% DDGS, had higher gizzard weight and better FCR, which the authors indicated was due to the fiber source effectively promoting gizzard development. Although increasing fiber content through HP-DDG could have impaired the starter performance in the present study, its gradual addition may have conditioned the digestive tract of birds for the next rearing phases and maintained growth performance. Thus, the initial challenge imposed by fiber may have ultimately contributed to a more efficient digestive capacity later on.

The results regarding carcass and cut yields were consistent with the performance results, as no differences were observed. Mustafa et al. (2017) observed that broilers fed 7.5% HP-DDG had higher breast, thigh, and drumstick weights at 42 days compared with the group fed no HP-DDG, which was attributed to the physiological and morphological improvements previously discussed.

5. Conclusions

The gradual inclusion of HP-DDG in broiler diets up to 15% does not impair overall growth performance or carcass yield. However, inclusion levels should not exceed 9% during the starter phase to avoid initial growth loss.

Data availability

The entire dataset supporting the results of this study was published in the article itself.

Author contributions

Conceptualization: Dias, K. M. M.; Lima, I. L.; Rostagno, H. S. and Calderano, A. A. **Data curation:** Dias, K. M. M. and Oliveira, C. H. **Formal analysis:** Dias, K. M. M. and Oliveira, C. H. **Investigation:** Dias, K. M. M.; Oliveira, C. H. and Fernandes, W. C. **Methodology:** Dias, K. M. M.; Lima, I. L.; Rostagno, H. S. and Calderano, A. A. **Project administration:** Rostagno, H. S. and Calderano, A. A. **Resources:** Lima, I. L. and Mike, B. **Supervision:** Rostagno, H. S. and Calderano, A. A. **Writing – original draft:** Dias, K. M. M. **Writing – review & editing:** Oliveira, C. H.; Lima, I. L.; Mike, B. and Calderano, A. A.

Conflict of interest

The authors declare no conflict of interest.

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