# Effect of sodium gluconate on performance, carcass characteristics, and intestinal morphometry of broilers from 22 to 42 days of age

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**ABSTRACT.** The objective of this experiment was to evaluate the effect of the sodium gluconate addition on performance (feed intake, body weight, weight gain, feed conversion, livability and productive efficiency), carcass characteristics (carcass, breast, thigh and drumstick, wing and back yield), and morphometry of duodenum, jejunal and ileum of broilers from 22 to 42 days of age. A total of 1,200 Cobb male broilers were distributed in a completely randomized design with five treatments (0.00, 0.10, 0.20, 0.30 and 0.40% sodium gluconate), and eight replications of 30 broilers each. The inclusion of sodium gluconate did not affect the broiler performance and carcass characteristics. However, the morphometry of duodenum and jejunal mucosa showed beneficial effects.

**Keywords:** butyrate, prebiotic, carcass characteristics, intestinal mucosa.

## Efeito do gluconato de sódio sobre o desempenho, rendimento de carcaça e morfometria da mucosa intestinal de frangos de corte de 22 a 42 dias de idade

**RESUMO.** O presente estudo teve por objetivo avaliar o efeito da utilização de níveis crescentes do gluconato de sódio sobre o desempenho, o rendimento de carcaça e de partes e a morfometria da mucosa intestinal de frangos de corte de 22 a 42 dias de idade. Foram utilizados 1200 frangos de corte da linhagem Cobb, distribuídos em um delineamento inteiramente casualizado, envolvendo cinco tratamentos (0,00; 0,10; 0,20; 0,30 e 0,40% de gluconato de sódio) com oito repetições de 30 aves cada parcela. As variáveis foram submetidas à análise de variância e em caso de significância estatística foram realizadas análises de regressão pelos modelos polinomial e quadrático. A inclusão do gluconato de sódio não afetou o desempenho e o rendimento de carcaça e de partes. Entretanto, exerceu efeito benéfico sobre a morfometria da mucosa intestinal do duodeno e do jejuno.

**Palavras-chave:** butirato, prebiótico, características de carcaça, mucosa intestinal.

## Introduction

Sodium gluconate is the sodium salt of gluconic acid produced by glucose fermentation. Gluconic acid can be used as a prebiotic, stimulating butyrate production in the large intestine (TSUKAHARA et al., 2002). This acid is able to bypass the small intestine digestion and absorption process, and 70% of gluconic acid reaches the large intestine (ASANO et al., 1997). Gluconic acid is fermented by bacteria of the *Lactobacillus* and *Bifidobacterium* genera (ASANO et al., 1994). Lactate and acetate are produced from gluconic acid by these lactic acid bacteria, and are converted to butyrate by acid-utilizing bacteria, such as *Megasphaera elsdenii* (ASANO et al., 1994, 1997; HASHIZUME et al., 2003; TSUKAHARA et al., 2002).

Butyrate is the minor component present in the lumen of the large intestine, when compared to the other short-chain fat acids (SCFA) (acetate and propionate). Nevertheless, butyrate is the main energy source for epithelial cells of the large intestine (ROEDIGER, 1995; SCHEPPACH et al., 1995), and this substance stimulates mucous release (SHIMOTOYODOME et al., 2000), epithelial cell proliferation (SAKATA, 1987; YOUNG; GIBSON, 1995), and mineral and water absorption from the intestinal lumen (HOLTUG et al., 1995; ROEDIGER; MOORE, 1981). Moreover, the inhibitory effect of butyrate on tumor development has been frequently discussed (D'ARGENIO et al., 1996; McINTYRE et al., 1993; PERRIN et al., 2001).

Saccharides are non-digestible materials that reach the large intestine (CAMPBELL et al., 1997; GIBSON; ROBERFROID, 1995; KIRIYAMA et al., 1992; LE BLAY et al., 1999a and b; MORTENSEN et al., 1988; PIVA et al., 1996; RISLEY et al., 1992), but only certain saccharides are able to stimulate butyrate production at that location (CAMPBELL et al., 1997; LE BLAY et al., 1999a and b; MORTENSEN et al., 1988). Gluconic acid is fermented in the large intestine (ASANO et al., 1997), but the fermentation process induced by this acid has not been elucidated.

Sodium gluconate is commonly used in human food industry as an acidifier, coagulant and carrier for mineral supplements (ASANO et al., 1994). Only two experiments have been performed to test the effects of sodium gluconate on broilers (GUO et al., 2009; RAFACZ-LIVINGSTON et al., 2005b), and the conclusion was that the addition of sodium gluconate in diet caused an increase in weight gain and tibia ash of starter broilers.

The objective of this experiment was to evaluate the effect of sodium gluconate on performance (feed intake, body weight, weight gain, feed conversion, livability and productive efficiency), carcass characteristics (carcass, breast, thigh and drumstick, wing and back yield), and morphometry of duodenum, jejunum and ileum of broilers from 22 to 42 days of age.

## Material and methods

A total of 1,200 Cobb male broilers were housed from 1 day of age in a completely randomized design, with 5 treatments (5 levels of sodium gluconate: 0.00, 0.10, 0.20, 0.30, and 0.40%) and 8 replications of 30 birds each. The broilers were vaccinated against Marek disease, Newcastle disease and Infectious Bronchitis at hatchery. The immunization against Gumboro and Newcastle diseases was done at 7 days of age, and the reinforcement against Gumboro disease at 14 days of age. The room temperature recorded during the experimental period was 27 ± 2.8ºC, and relative humidity was 78 ± 8.7%.

Water and feed were provided ad libitum. Broilers were fed with common starter diet until 21 days of age, in accordance with the nutrient recommendations (3,005 kcal ME $kg^{-1}$ and 21.6% of CP) of Rostagno et al. (2005). The broilers were weighed at 22 days of age, and then allocated according to weight in 40 boxes containing 30 birds each.

The basal diet (Table 1) was formulated following Rostagno et al. (2005) recommendations.

Broilers were housed over reused wood-shavings bedding to provide a sanitary challenge.

Body weight and feed intake values were recorded. Weight gain, feed conversion, livability, and productive efficiency {PE = [Daily mean weight gain (g) x livability (%)] / (feed conversion x 10)} were calculated for each pen at 42 days of age. The yield evaluation was done using 8 broilers per replication selected at random.

**Table 1.** Composition and nutritive values of basal diet.

| Ingredients | % |
|---|---|
| Corn | 66.48 |
| Soybean meal | 25.78 |
| Soybean oil | 3.03 |
| Dicalcium phosphate | 1.63 |
| Limestone | 0.96 |
| L-Lysine-HCl (78%) | 0.27 |
| DL-Methionine | 0.20 |
| L-Threonine | 0.11 |
| L- Arginine | 0.06 |
| L-Valine | 0.09 |
| L-Isoleucine | 0.05 |
| Vitamin and mineral premix[1] | 0.50 |
| Salt | 0.44 |
| Variable portion (sand)[2] | 0.40 |
| Total | 100.00 |
| Calculated nutrients | |
| CP (%) | 17.76 |
| ME (kcal $kg^{-1}$) | 3,175 |
| Calcium (%) | 0.87 |
| Sodium (%) | 0.21 |
| Total Phosphorus (%) | 0.63 |
| Available Phosphorus (%) | 0.41 |
| Digestible Lysine (%) | 1.07 |
| Digestible Methionine (%) | 0.47 |
| Digestible TSSA[3] (%) | 0.77 |
| Digestible Tryptophan (%) | 0.18 |
| Digestible Threonine (%) | 0.69 |
| Digestible Arginine (%) | 1.13 |
| Digestible Valine (%) | 0.82 |
| Digestible Isoleucine (%) | 0.72 |

[1]Vitamin-mineral premix supplied the following per kilogram of feed: vitamin A (8,000 IU), vitamin $D_3$ (1,800 IU), vitamin E (12 mg), vitamin $K_3$ (2 mg), vitamin $B_1$ (1 mg), vitamin $B_2$ (4 mg), vitamin $B_6$ (1 mg), vitamin $B_{12}$ (10 mcg), folic acid (0.40 mg), biotin (0.04 mg), niacin (28 mg), calcium pantothenate (11 mg), copper (6 mg), cobalt (0.10 mg), iodine (1 mg), iron (50 mg), manganese (65 mg), zinc (45 mg), selenium (0.21 mg), choline chloride 50% (500 mg), coccidiostat (60 mg), ethoxyquin (antioxidant) (12 mg). [2]Variable portion: consists of variable amounts of sand and sodium gluconate containing the levels of 0.00, 0.10, 0.20, 0.30, and 0.40% sodium gluconate. [3]TSAA = total sulfur amino acids (methionine + cystine).

At 42 days of age, the broilers were submitted to a 6h pre-slaughter fast, after which they were weighed individually, slaughtered and eviscerated. The yield was obtained from the entire carcass yield (without feet, head and neck), breast yield, thigh and drumstick yield, back yield and wing yield.

The morphometric analysis of the intestinal mucosa (duodenum, jejunum and ileum) was done in samples collected from two birds per replication. Samples measuring 1 cm were collected from the duodenum, jejunum and ileum. The duodenum sample was collected at the apex of this intestinal portion, the jejunum at the midway portion between the point of entry of the bill ducts and Meckel's diverticulum, and the ileum 10 cm before the cecal junction. The intestinal segments were fixed in Bouin's

solution during 48h. Then, the tissue fragments were dehydrated by immersion in growing concentrations of alcohols (from 70% to absolute), infiltrated with xylene and embedded in paraffin. A microtome was used to make 5 $\mu$m sections, which that were mounted on glass slides and stained with Masson's trichrome. Villus height and crypt depth ($\mu$m) were determined using an image analyzer (Kontron Elektronik GmbH, Poway, CA) coupled to a binocular microscope (Carl Zeiss, São Paulo, Brazil). Fifty readings of villus height and crypt depth were performed per treatment and per intestinal segment, comprising a total of 3,000 readings, which were documented in an Axioskop photomicroscope (Carl Zeiss, São Paulo State, Brazil). The criterion for villus selection was based on the presence of intact lamina propria. Villus height was measured from the tip of the villus to the villus-crypt junction, whereas crypt depth was defined as the depth of the invagination between adjacent villi.

The results were submitted to Analysis of Variance using the General Linear Model (GLM) procedure of the SAS Statistical Program (SAS, 2000). Pair-wise comparisons of means were made using Tukey's test procedure. A p value < 0.05 was considered significant. Regression analysis was used to estimate the optimum level of sodium gluconate (95% of the upper asymptote) when a significant quadratic response was detected.

## Results and discussion

The different levels of sodium gluconate did not influence performance (Table 2), when the values were evaluated by Analysis of Variance and regressions. These results are in disagreement with Guo et al. (2009), who added 2% sodium gluconate in ration, and observed a significant increase in weight gain from 8 to 42 days of age, and from 22 to 42 days of age. They detected a trend for better results of feed intake in the cited phases, and of feed conversion from 22 to 42 days of age. Rafacz-Livingston et al. (2005b) observed that the addition of 1.5 and 3.35% of sodium gluconate in a phosphorus-deficient diet with no phytase supplementation improved the weight gain and tibia ash percentage of broilers from 8 to 22 days of age. These results were similar to those found in the previous experiments by Rafacz-Livingston et al. (2005a) and Snow et al. (2004)

using 3% citric acid in phosphorus-deficient diets for broilers.

The effect of sodium gluconate in this present experiment was similar to the results found by Hofacre et al. (2003), who detected no significant differences in weight gain among the treatments using different growing promoters (probiotics and prebiotics). Waldroup et al. (2003), working with the addition of antibiotic, mannan oligosaccharides and a combination of antibiotic and mannan oligosaccharides, observed that body weight was not influenced by the treatments. However, Rodrigues et al. (2003) observed improvement in weight gain, feed conversion and production factor of broilers, while evaluating the mannan oligosaccharides prebiotic effect of rations containing corn of different nutritional qualities.

The effect of the different sodium gluconate levels on carcass, thigh and drumstick, wing and back yield (Table 3) was not significant. However, the levels of sodium gluconate had a quadratic effect on breast yield (p < 0.05), following the equation BY = 39.373 - 23.261x + 53.224x$^2$ (r$^2$ = 0.70). This result is similar to thT obtained by Albino et al. (2006), who observed improved breast yield when the avilamycin, mannan oligosaccharides ST or mannan oligosaccharides AT, combined or not with avilamycin, were added to ration. Maiorka et al. (2001) used antibiotics, two types of probiotics, prebiotics and combinations of the cited items, and did not find significant differences in commercial cut yields, probably because they did not challenge the broilers excusing the use of these additives.

The results of intestinal epithelium morphometry from the duodenum, jejunum and ileum of broilers at 42 days of age are presented in Table 4. There was a quadratic effect (p < 0.05) of sodium gluconate levels on villus height (Y = 1813.19 – 242.85x – 44.10x$^2$; r$^2$ = 0.82), and on duodenum crypt depth (Y = 178.36 + 65.91x – 8.23x$^2$, r$^2$ = 0.48), presenting better values in the levels of 0.30 and 0.40% sodium gluconate, respectively. A linear reduction was detected (p < 0.05) in the duodenum villus:crypt ratio ($\hat{Y}$ = 7.059 – 0.448x; r$^2$ = 0.81) for higher levels of sodium gluconate. Prebiotic supplementation can adversely affect the duodenal villi structure, causing intestinal shedding, and consequently decreasing villus height and crypt depth.

**Table 2.** Performance characteristics of male broilers from 22 to 42 days of age treated with different levels of sodium gluconate.

| Variable[1] | Sodium gluconate levels (%) | | | | | CV | Regression |
|---|---|---|---|---|---|---|---|
| | 0.00 | 0.10 | 0.20 | 0.30 | 0.40 | (%) | |
| Feed Intake (kg) | 3.437 | 3.475 | 3.518 | 3.508 | 3.487 | 3.05 | NS |
| Body Weight (kg) | 2.636 | 2.657 | 2.666 | 2.651 | 2.654 | 2.27 | NS |
| Weight Gain (kg) | 1.762 | 1.792 | 1.805 | 1.781 | 1.779 | 3.60 | NS |
| Feed Conversion[2] (kg kg$^{-1}$) | 1.95 | 1.94 | 1.95 | 1.97 | 1.96 | 2.74 | NS |
| Livability (%) | 98.02 | 98.04 | 98.02 | 98.67 | 98.67 | 1.84 | NS |
| Productive Efficiency[3] | 319 | 324 | 323 | 320 | 322 | 4.42 | NS |

[1]Each mean represents 8 replications with 30 broilers each. [2]The feed conversion values were corrected considering the mortality. [3]PE = [daily mean weight gain (g) x livability (%)]/(feed conversion x 10).

**Table 3.** Carcass characteristics (carcass, breast, thigh and drumstick, wing and back yield) of male broiler chicks from 22 to 42 days of age treated with different levels of sodium gluconate.

| Variable (%)[1] | Sodium gluconate levels (%) | | | | | CV | Regression |
|---|---|---|---|---|---|---|---|
| | 0.00 | 0.10 | 0.20 | 0.30 | 0.40 | (%) | |
| Carcass yield | 73.69 | 74.08 | 77.65 | 74.96 | 76.49 | 4.78 | NS |
| Breast Yield[2] | 39.17a | 38.31a | 37.35b | 37.77ab | 37.37a | 3.75 | ⋆ |
| Thigh and Drumstick Yield | 29.37 | 29.56 | 29.42 | 30.12 | 29.57 | 4.99 | NS |
| Wing Yield | 10.38 | 10.39 | 10.31 | 10.23 | 9.96 | 4.70 | NS |
| Back Yield | 20.64 | 22.37 | 21.05 | 22.42 | 21.11 | 4.64 | NS |

⋆Quadratic effect ($p < 0.05$). [1]Each mean represents 8 replications with 8 broilers each. [2]$\hat{Y} = 39.373 - 23.261x + 53.224x^2$; $r^2 = 0.70$.

**Table 4.** Morphometric characteristics (villus height, crypt depth, and villus height crypt[-1] depth ratio) of duodenum, jejunum and ileum from male broilers at 42 days of age, treated with different levels of sodium gluconate.

| Variable[1] | Sodium gluconate levels (%) | | | | | CV | Regression |
|---|---|---|---|---|---|---|---|
| Duodenum | 0.00 | 0.10 | 0.20 | 0.30 | 0.40 | (%) | |
| Villus Height ($\mu m$)[2] | 1618.7 | 1512.8 | 1429.3 | 1608.1 | 1478.7 | 12.67 | ⋆ |
| Crypt Depth ($\mu m$)[3] | 251.07 | 233.37 | 290.33 | 298.89 | 300.79 | 17.05 | ⋆ |
| Villus Height Crypt[-1] Depth Ratio ($\mu m$)[4] | 6.58 | 6.53 | 5.16 | 5.38 | 4.92 | 17.50 | ⋆⋆ |
| Jejunum | | | | | | | |
| Villus Height ($\mu m$) | 1158.5 | 1231.6 | 1323.8 | 1091.6 | 1124.5 | 11.12 | NS |
| Crypt Depth ($\mu m$)[5] | 190.45 | 193.49 | 247.43 | 276.87 | 257.51 | 21.58 | ⋆⋆ |
| Villus Height Crypt[-1] Depth Ratio ($\mu m$)[6] | 6.39 | 6.44 | 5.43 | 4.02 | 4.43 | 18.71 | ⋆⋆ |
| Ileum | | | | | | | |
| Villus Height ($\mu m$) | 812.8 | 714.3 | 751.5 | 856.7 | 797.2 | 17.48 | NS |
| Crypt Depth ($\mu m$) | 224.67 | 197.26 | 225.58 | 300.89 | 236.97 | 17.65 | NS |
| Villus Height Crypt[-1] Depth Ratio ($\mu m$) | 3.73 | 3.72 | 3.35 | 2.88 | 3.55 | 27.24 | NS |

⋆Quadratic effect ($p < 0.05$), ⋆⋆Linear effect ($p < 0.05$). [1]Each mean represents 8 replications with 2 broilers each. [2]$\hat{Y} = 1813.19 - 242.85x - 44.10x^2$; $r^2 = 0.82$. [3]$\hat{Y} = 178.36 + 65.91x - 8.23x^2$; $r^2 = 0.48$. [4]$\hat{Y} = 7.059 - 0.448x$; $r^2 = 0.81$.

Poeikhampha and Bunchasak (2010) concluded that sodium gluconate supplementation as prebiotic is beneficial because this additive is the main substrate used by saprophyte bacteria for butyrate production in poultry and swine. Ribeiro et al. (2002) affirmed that viable cells (probiotics) compete with pathogenic microorganisms for a location to attach to the intestinal mucosa, decreasing the incidence of diarrhea and improving the absorption of available nutrients. This mechanism has direct influence on the reestablishment of intestinal mucosa, increasing villus height.

Crypt depth increased ($p < 0.05$) ($\hat{Y} = 167.90 + 21.75x$; $r^2 = 0.83$), and the villus:crypt ratio decreased ($p < 0.05$) linearly ($\hat{Y} = 7.25 - 0.63x$; $r^2 = 0.82$) in the jejunum portion for higher levels of sodium gluconate. These results are in disagreement with Oliveira et al. (2009), who did not observe any effect of mannan oligosaccharide and enzymatic complex addition on villus height and crypt depth of jejunum. Likewise, Schwarz et al. (2002) compared the effect of diets with and without antibiotics, and with fructooligosaccharide (0.2%) and mannan oligosaccharide (0.2%), and did not detect significant differences in duodenum villus height and crypt depth.

The different levels of sodium gluconate did not cause significant effect on ileum morphometry of broilers at 42 days of age (Table 4).

## Conclusion

The use of sodium gluconate until the level of 0.4% did not affect the performance of broilers from 22 to 42 days of age. Moreover, the inclusion of sodium gluconate caused no effect on carcass characteristics, but improved the morphometric characteristics of the duodenum and jejunum.

## Acknowledgements

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