

Influence of beak trimming and inclusion of sodium butyrate in the diet on growth performance and digestive tract traits of brown-egg pullets differing in initial body weight

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ABSTRACT We studied the effects of beak trimming and sodium-butyrate inclusion in the diet on growth performance and gastrointestinal tract (GIT) traits of brown-egg pullets differing in initial BW. In experiment 1, a total of 6 treatments were organized as a 2 × 3 factorial with 2 BW at hatch (light, 33.9 g and heavy, 37.6 g) and 3 beak trimming protocols [mild (MI-0) or aggressive (AG-0) infrared power setting at hatch and traditional hot blade at 8 D of age (HB-8)] as main effects. Initial BW did not affect growth performance or GIT traits at any age. From hatch to 5 wk of age, HB-8 pullets had lower ADFI ($P < 0.01$) and ADG ($P < 0.05$) than MI-0 and AG-0 pullets but no differences were detected after this age. Beak trimming did not affect FCR, BW uniformity, GIT traits, or bacteria count in the excreta at any age. In experiment 2, a total of 12 treatments were organized as a 2 × 3 × 2 factorial,

with 2 BW at hatch, 3 beak trimming protocol (as per in experiment 1), and 2 levels of a sodium-butyrate additive (0 vs. 0.3%) as main effects. At 7 D of life, beak treatment reduced pullet growth and AG-0 procedure impaired pullet uniformity ($P < 0.001$) but the birds recovered completely by day 14 ($P < 0.001$ for the interaction with time). Cumulatively (0 to 6 wk of age), pullets beak treated at hatch (MI-0 and AG-0) had greater ADFI than HB-8 pullets ($P < 0.01$). Sodium butyrate tended to improve ADG ($P = 0.073$) and FCR ($P = 0.069$) with most of the benefits observed for the first 2 wk of life. In summary, initial BW and beak trimming procedure did not affect final pullet growth in any of the 2 experiments, or GIT traits in experiment 1. Sodium butyrate tended to improve growth and FCR from 0 to 6 wk of age but did not affect BW uniformity.

Key words: digestive tract pH, infrared beak treatment, hot-blade trimming, sodium butyrate, pullet uniformity

2019 Poultry Science 98:3937–3949
<http://dx.doi.org/10.3382/ps/pez129>

INTRODUCTION

The BW at hatch depends on numerous factors, including egg size and age of the breeder flock (Ulmer-Franco et al., 2010; Mendes et al., 2011). A positive relation between BW at hatch, gastrointestinal tract (GIT) development (Sklan et al., 2003; Tona et al., 2004), and BW at older ages (Proudfoot and Hulan, 1981; Wilson, 1991) has been documented in broilers. Based on this information, egg producers are reluctant to accept pullets with average BW below 36 g at the arrival of the birds to the commercial farms. As a result, small eggs produced by young breeders are not hatched, reducing the overall profitability of the breeder flock.

Beak trimming reduces the effects of aggressive behavior and feather pecking and decreases the incidence of mortality in poultry (Gentle, 1986; Glatz, 2005). Hot blade was the most traditional method used by the

industry for beak trimming of the pullets. However, this procedure damages beak tissues, causes stress, and pain; affects beak sensitivity and bird behavior (Gentle et al., 1997; Cheng, 2006); and compromises the welfare status of the bird during and after the trimming process (Duncan et al., 1989; Hughes and Gentle, 1995). Infrared beak treatment (IRBT) at hatch facilitates farm management, reduces stress, and decreases the risk of infection of the chicks (Dennis et al., 2009; Dennis and Cheng, 2010, 2012). Consequently, IRBT might be more convenient than hot blade technology in pullets.

The use of antibiotics as feed growth promoters was banned in the European Union in 2006 and since then, in many other countries. Consequently, new additives are needed to improve health and growth performance of the pullets (Mateos et al., 2002, 2012; Windisch et al., 2008). The inclusion of short-chain fatty acids in the diet has been shown to improve GIT development and help in the control of microbial growth in the GIT. In this respect, butyric acid elicits stimulatory, although inconsistent effects, on the integrity of the epithelial mucosa and the secretion of endogenous enzymes,

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Received October 17, 2018.

Accepted March 4, 2019.

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modifying positively the profile of the microbiota of the GIT (Ricke, 2003; Guilloteau et al., 2010; Zhang et al., 2011; Cerisuelo et al., 2014). As a consequence, the inclusion of butyric acid often increases growth performance in broilers (Hu and Guo, 2007; Adil et al., 2010; Kaczmarek et al., 2016). However, free butyrate can be easily absorbed in the upper part of the GIT, preventing the beneficial effects of butyric acid in the hindgut (Kaczmarek et al., 2016; Liu et al., 2017). In addition, when fed at a high dose, unprotected butyric acid, that is volatile and corrosive and has a penetrating smell and an unpleasant taste, reduces feed intake (**FI**) in chicks (Antongiovanni et al., 2007). Consequently, when used as a feed additive, the acid has to be protected to facilitate the handling, stability, and activity in the GIT (Ahsan et al., 2016). In this respect, the sodium salt of the acid reduces at a high extent all these problems. The protected sodium butyrate (**Na butyrate**) passes unchanged through the upper part of the GIT and dissociates into butyrate and hydrogen ions in the proximal part of the small intestine (**SI**), improving the development of the villi of the mucosa (Leeson et al., 2005; Jertzsele et al., 2012). Consequently, butyrate salts are commonly used to improve health and growth performance of the bird under commercial conditions (Patten and Waldroup, 1988; Ricke, 2003). However, most of the studies on the benefits of the inclusion of Na butyrate in the diet on growth performance have been conducted in broilers and not prior research studies have specifically focused on its effects in pullets.

The objective of this research was to study the effects of beak trimming protocol and the inclusion of Na butyrate in the diet on growth performance, BW uniformity, and GIT measurements of pullets sorted by initial BW at hatch. The hypothesis of this research was that the stress caused by the hot-blade trimming at 8 D of age could reduce FI, growth, and BW uniformity of the pullets as compared with IRBT at hatch, and that Na butyrate could improve pullet growth. Also, it was hypothesized that the negative effect of hot-blade trimming and the beneficial effects of Na-butyratesupplementation could be more evident in the young bird, and more pronounced in the lighter than in the heavier pullets.

MATERIALS AND METHODS

The experimental procedures used in this research were approved by the Animal Ethics Committee of the Universidad Politécnica de Madrid and were in compliance with the Spanish guidelines for the care and use of animals in research (BOE, 2013).

Experiment 1

Husbandry, Diets, and Experimental Design A total of 720 Lohmann Classic brown pullets were obtained at hatch from a commercial layer breeder flock

of 26 wk of age. The selected pullets were weighed individually at the hatchery and classified by initial BW into 2 groups: light (33.9 ± 1.3 g) and heavy (37.6 ± 1.2 g). Each BW group was divided into 3 subgroups with the same average BW. Two of the subgroups were beak trimmed at the hatchery applying a high-intensity beam of infrared energy source (Nova-Tech Engineering Inc., Willmar, MN). The infrared setting was adjusted according to 2 procedures: a) mild (**MI-0**) using a 27/23C plate size and a 48 power setting and b) aggressive (**AG-0**) using a 25/23C plate size and a 46 power setting. The pullets from the third subgroup (**HB-8**) were beak trimmed at the farm at 8 D of age, using a blade heated at 75°C.

At arrival to the experimental station, the pullets were allotted at random in groups of 10, within initial BW and beak trimming procedure, into 72 cages (100 cm × 45 cm; Alternative Design, Siloam Springs, AR). The cages were provided with 2 multidirectional low-pressure nipple drinkers (Lubing System S. R. L., Campodarsego, Italy) and an open trough feeder with a length of 65 cm. Management practices were as recommended by Lohmann (2017). Briefly, the pullets were vaccinated at hatch against Marek, Gumboro, and Infectious Bronchitis Disease, following standard commercial procedures. Room temperature at the farm was maintained at 35°C for the first 2 D of life and then reduced gradually to 31°C at 7 D of age and 20°C at 15 wk of age. Pullets were kept on a 24 h/d light program for the first 2 D of life and 18 h/d from 3 to 7 D. Then, the light period was decreased 2 h per week until reaching 10 h/d at 5 wk of age. From 6 to 15 wk of age, light was maintained constant at 9 h/d. The light intensity in the barn was maintained at 20 lux during the first 3 wk of age and reduced to 6 lux from 4 to 16 wk of age. Pullets had ad libitum access to feed and water throughout the experiment (0 to 15 wk). From 1 to 7 D of life, the wire floor of the cages was covered with a paper lining and small amounts of feed were distributed on top every day.

The feeding program consisted of 3 periods: starter (0 to 5 wk), grower (5 to 10 wk), and developer (10 to 15 wk of age). The diets were formulated according to the nutrient specifications of FEDNA (2018) and fed in mash form. All main ingredients, except the soybean meal, the macro minerals, and the premix, were ground prior to mixing using a hammer mill fitted with a 3-mm screen (model MRA 220, Rosal S.A., Barcelona, Spain).

The experimental design was completely randomized in a factorial arrangement with 2 initial BW of the pullets and 3 beak trimming procedures. Each of the 6 treatments had 12 replicates and the experimental unit was the cage with 10 pullets for all measurements.

Measurements FI and BW of the pullets were determined by replicate at the end of each of the 3 feeding periods (5, 10, and 15 wk of age). Feed wastage was estimated to be low and was not measured. Mortality was recorded as produced. From these data, ADFI, ADG, and feed conversion ratio (**FCR**) were determined by

period and cumulatively. Birds that died or were culled during the experiment were weighed, and their estimated feed consumption was included in calculations of FCR. The BW uniformity was calculated by replicate at same ages, as indicated by Peak et al. (2000). Briefly, the CV of the individual BW of the pullets within each cage was generated and this parameter was used as an indirect measure of the uniformity.

After weighing the pullets for the growth performance control at 5, 10, and 15 wk of age, the birds were fasted for 60 min and then fed ad libitum for 4 h to maximize digesta content in the different segments of the GIT. Two birds per replicate were randomly selected, weighed, and euthanized by CO₂ inhalation. The GIT, from the pre-crop esophagus to the cloaca, including the liver, pancreas, spleen, and the digesta content, were removed aseptically and weighed. Then, the crop and the gizzard were carefully clamped to avoid digesta mixing, excised, and weighed, and the data expressed as a percentage of BW (relative weight). In addition, the pH of the crop, gizzard, ileum, and ceca contents was measured in situ in duplicate, using a digital pH meter fitted with a fine tip glass electrode (model 507, Crison Instruments S.A., Barcelona, Spain) as indicated by Jiménez-Moreno et al. (2009). The average value of the 2 measurements was used for further analysis. Then, the crop and the gizzard were flushed to eliminate digesta content, cleaned, dried with desiccant paper, and weighed again, and the fresh digesta content was calculated as the difference between the full and the empty weights. The data were expressed as the relative percentage (%) of full organ weight. Also, the lengths of the SI (duodenum, jejunum, and ileum) from the gizzard to the ileo-cecal junction and of the 2 ceca from the ostium to the tip of the ceca were measured on a glass surface using a flexible tape with a precision of 1 mm, and expressed relative to BW (cm/kg BW). Finally, the left tarsus of the pullets was removed and the length was measured using a digital caliper and expressed relative to BW.

At 34 D of age, clean pans were placed under the cages and 24 h later, representative samples of the excreta were collected from each cage, frozen, and stored at -20°C. Samples of the excreta were plated on specific selective growth medium for the quantification of *Lactobacilli*, Enterobacteriaceae, *Clostridium perfringens*, *Escherichia coli*, and total coliforms colonies according to accepted procedures (Harrigan, 1998). Briefly, the media used was MRS agar for Lactobacilli; violet red bile glucose agar for Enterobacteriaceae, SPS agar for *C. perfringens*, and a Coli-ID agar for *E. coli* and total coliforms counts. The results are expressed as log₁₀ cfu/g.

Experiment 2

Husbandry, Diets, and Experimental Design A total of 720 recently hatched pullets were obtained from

a Lohmann Brown Classic layer breeder flock at 27 wk of age and used in an experiment that lasted 6 wk. The management and distribution of the birds were similar to those described for experiment 1. Briefly, the pullets were weighed individually at the hatchery and classified by initial BW into a light (33.5 ± 1.2 g) and a heavy (37.5 ± 1.4 g) group. Each BW group was subdivided into 3 subgroups with similar average BW. At arrival to the experimental farm, the pullets were allotted at random in groups of 10, within each initial BW group and beak trimming procedure, into 72 cages as per in experiment 1. In addition, half of the subgroups were fed the same diet with or without 0.3% Na butyrate. The additive tested (Butirex C-4; Novation 2002 S. L., Madrid, Spain) was a Na protected salt that contained by analysis 54% butyrate and 21% sodium. The process consisted in the reaction of butyric acid with buffer salts, resulting after cooling in a protected chemical structure that avoided the release and absorption of the free acid in the upper part of the GIT. Room temperature and light program were similar to those indicated for experiment 1. Two diets were used: a control diet that was from the same batch used from 0 to 5 wk in experiment 1 and an experimental diet that had the same composition than the control diet but in which the Na-butyrate additive substituted the same amount of the whole diet.

The experimental design was completely randomized with 12 treatments in a factorial arrangement with 2 initial BW of the pullets, 3 beak trimming protocols, and 2 diets that differed exclusively in the inclusion or not of 0.3% of the Na-butyrate additive. Each treatment was replicated 6 times, and the experimental unit was the cage with 10 birds for all measurements.

Measurements FI and individual BW of the pullets were determined weekly by cage from 0 to 6 wk of age. Feed wastage was estimated to be low and was not measured. Mortality was recorded as produced and birds that died or were culled were weighed. From these data, ADFI, ADG, and FCR were determined by cage, weekly and cumulatively as indicated for experiment 1. The BW uniformity was estimated by replicate at the same ages, as indicated for experiment 1.

At 6 wk of age, after weighing the pullets for the growth performance control, 2 birds per replicate were randomly selected, weighed, and euthanized by CO₂ inhalation. Then, 2 g of the crop and ileum contents were collected from each bird, mixed immediately with 2 mL of 0.5 N HCl to avoid any fermentation process, frozen at -20°C, and stored until analysis of volatile fatty acids (VFA) contents.

Statistical Analysis In experiment 1, data on growth performance, BW uniformity, GIT traits, and tarsus length were analyzed as a completely randomized design with initial BW and beak trimming procedure as main effects, using the MIXED procedure of SAS as indicated by Littell et al. (1998). Data on bacteria count of the excreta were analyzed using the GLM procedure of SAS (SAS Institute, 2004). Sources of variation included main effects, time period, and the interactions.

When significant differences among treatments were detected, means were separated using the Tukey test. In experiment 2, data on growth performance and BW uniformity were analyzed as a completely randomized design with initial BW of the pullets, beak trimming procedure, and Na-butyrate supplementation as main effects, as indicated in experiment 1. Data on VFA of the crop and ileum contents were analyzed using the GLM procedure of SAS. Results in tables are presented as means, and differences were considered significant at $P < 0.05$.

Laboratory Analysis Particle size distribution of the diets, expressed as geometric mean diameter \pm geometric standard deviation, was determined in triplicate in 150 g samples using a Retsch shaker (Retsch, Stuttgart, Germany) provided with 8 sieves ranging in mesh from 5,000 to 40 μm (ASAE, 2003). Representative samples of the diets used in the 2 experiments were ground with a laboratory mill (Retsch Model Z-I, Stuttgart, Germany) equipped with a 0.75-mm screen and analyzed as indicated by AOAC International (2005). Briefly, DM was determined by oven-drying (method 930.15), total ash using a muffle furnace (method 942.05), nitrogen by Dumas (method 968.06) using a Leco analyzer (Model FP-528, Leco Corp., St. Joseph, MI), and crude fiber by sequential extraction with diluted acid and alkali (method 962.09). Neutral detergent fiber was determined as described by Van Soest et al. (1991) and expressed on an ash-free basis. Ether extract was analyzed after 3 N HCl acid hydrolysis (method 159 Am 5-04) as indicated by the AOCS (2004) using an Ankom XT10 extraction system 160 (Ankom Technology Corp. Macedon, NY). Gross energy was determined in an adiabatic bomb calorimeter (Model 1356, Parr Instrument Company, Moline, IL). The amino acid content of the diets was determined by ion-exchange chromatography (Hewlett-Packard 1100, Waldbronn, Germany) after acid hydrolysis, as indicated by De Coca-Sinova et al. (2008). In addition, the concentration of total VFA and their profile were determined in representative samples collected from the crop and the ileum by gas chromatography, using a Perkin Elmer Autosystem XL (Perkin Elmer Inc., Waltham, MA) equipped with a flame-ionization detector and a TR-FFAP capillary column (30 m \times 0.53 mm \times 1 μm ; Supelco, Barcelona, Spain) as described by Carro et al. (1992). Samples were thawed at 4°C for 4 h and homogenized. Then, 2 mL of the sample was centrifuged (13,000 \times g) at 4°C for 20 min, and 0.8 mL of the resulting supernatant was mixed with 0.5 mL of a deproteinized solution consisting of 20 g metaphosphoric acid and 0.6 g of crotonic acid per liter of distilled water and left overnight at 4°C. All the chemical analyses were conducted in duplicate. The ingredient composition, particle size distribution, and the calculated and determined analyses of the diets are shown in Table 1.

Table 1. Ingredient composition, chemical analyses (% as fed basis), and particle size distribution (μm) of the experimental diets¹: experiments 1 and 2.

	0 to 5 wk ²	5 to 10 wk	10 to 15 wk
Ingredient			
Corn	13.10	12.20	-
Wheat	45.09	48.41	49.99
Barley	4.50	7.85	24.50
Soybean meal (45.5% CP)	21.20	13.10	5.40
Sunflower meal (35% CP)	10.00	14.00	16.00
Soy oil	2.50	1.00	1.00
Calcium carbonate	1.00	1.20	1.39
Dicalcium phosphate	1.45	1.14	0.74
Sodium chloride	0.28	0.34	0.35
DL-methionine (99%)	0.22	0.12	0.05
L-lysine (78%)	0.26	0.24	0.18
Vitamin and mineral premix ³	0.40	0.40	0.40
Calculated analyses ⁴			
AME _n (kcal/kg)	2,900	2,820	2,790
Digestible amino acid			
Lys	0.98	0.84	0.65
Met	0.51	0.41	0.31
Met + Cys	0.79	0.69	0.58
Thr	0.59	0.55	0.46
Trp	0.21	0.20	0.17
Crude fiber	4.50	5.50	6.0
Neutral detergent fiber	11.9	14.0	15.3
Digestible phosphorus	0.44	0.40	0.34
Determined analyses ⁵			
Dry matter	89.6	91.1	93.0
Gross energy (kcal/kg)	4,020	4,040	4,030
Crude protein	20.3	18.9	17.3
Lys	1.10	0.96	0.75
Met	0.54	0.45	0.35
Met + Cys	0.89	0.79	0.67
Thr	0.70	0.65	0.55
Trp	0.25	0.23	0.21
Ether extract	4.40	3.00	2.80
Ash	4.96	5.01	4.74
Calcium	1.01	0.95	0.92
Phosphorus	0.72	0.68	0.60
Particle size distribution ⁶			
2,500	4.8	13.2	6.5
1,250	29.8	31.7	27.6
630	32.2	27.7	23.1
315	18.5	19.5	14.7
160	12.9	7.3	11.4
80	1.8	0.5	2.1
GMD \pm GSD ⁷	829 \pm 2.22	1,049 \pm 2.25	894 \pm 2.31

¹Within each period, 2 diets with or without 0.3% Na butyrate were used. The Na butyrate was included in the diet in substitution of barley.

²This diet was used from 0 to 5 wk of age in experiment 1 and from 0 to 6 wk of age in experiment 2.

³Supplied per kilogram of diet: vitamin A (trans-retinyl acetate), 10,000 IU; vitamin D₃ (cholecalciferol), 3,500 IU; vitamin E (all-*rac*-tocopherol-acetate), 35 mg; thiamine, 2 mg; riboflavin, 8 mg; pyridoxine, 4 mg; cyanocobalamin, 0.025 mg; vitamin K₃ (bisulphate menadione complex), 3 mg; choline (choline chloride), 270 mg; nicotinic acid, 60 mg; pantothenic acid (D-calcium pantothenate), 15 mg; folic acid, 1.5 mg; D-biotin, 0.15 mg; zinc (ZnO), 90 mg; manganese (MnO), 75 mg; iron (FeCO₃), 60 mg; copper (CuSO₄·5H₂O), 8 mg; iodine (KI), 2 mg; selenium (Na₂SeO₃), 0.3 mg; Roxazyme, 200 mg [1,600 IU of endo-1,4- β -glucanase (EC 3.2.1.4), 3,600 IU of endo-1,3 (4)- β -glucanase (EC 3.2.1.6), and 5,200 IU of endo-1,4- β -xylanase (EC 3.2.1.8)] supplied by DSM S.A., Madrid, Spain; and Natuphos 5000 [300 FTU/kg 6-phytase (EC 3.1.3.26), 60 mg, supplied by Basf Española S.A., Tarragona, Spain].

⁴According to FEDNA (2010).

⁵Data correspond to the average of the 2 experimental diets (with and without 0.30% Na butyrate). Differences in analytical values between the 2 diets were within acceptable ranges.

⁶Sieve diameter (μm). The percentages of particles smaller than 80 μm or bigger than 2,500 μm were negligible for all diets. Data presented correspond to the average of the 2 experimental diets.

⁷GMD = geometric mean diameter, GSD = Log normal SD.

Table 2. Influence of initial BW and beak trimming procedure on growth performance of the pullets: Experiment 1.

	Main effect					SEM ³	Probability ⁴	
	Initial BW ¹		Beak trimming ²				Initial BW	Beak trimming
	Light	Heavy	MI-0	AG-0	HB-8			
ADFI, g								
0 to 5 wk	19.4	19.4	19.8 ^a	19.3 ^{a,b}	19.1 ^b	0.15	0.898	0.003
5 to 10 wk	58.8	59.1	59.6	59.0	58.2	0.46	0.569	0.094
10 to 15 wk	70.7	71.4	72.0	71.0	70.2	0.74	0.450	0.270
ADG, g								
0 to 5 wk	8.9	8.9	9.0 ^a	8.8 ^b	8.8 ^b	0.08	0.897	0.023
5 to 10 wk	17.9	17.8	18.0	17.6	17.9	0.16	0.737	0.288
10 to 15 wk	11.2	11.4	11.3	11.3	11.2	0.16	0.153	0.826
FCR, g/g								
0 to 5 wk	2.19	2.19	2.20	2.20	2.18	0.04	0.971	0.942
5 to 10 wk	3.29	3.34	3.33	3.34	3.28	0.05	0.413	0.563
10 to 15 wk	6.36	6.29	6.38	6.31	6.30	0.05	0.202	0.471
Cumulative (0 to 15 wk)								
ADFI, g	49.6	50.0	50.5 ^a	49.8 ^{a,b}	49.2 ^b	0.35	0.445	0.038
ADG, g	12.6	12.7	12.8	12.6	12.6	0.09	0.541	0.246
FCR, g/g	3.95	3.94	3.97	3.95	3.92	0.03	0.755	0.458

^{a,b}Within a line, means without a common superscript differ significantly ($P < 0.05$).

¹Light = 33.9 ± 1.3 g BW; heavy = 37.6 ± 1.2 g BW.

²Infrared beak treatment at hatch with high (48; MI-0) and low (46; AI-0) setting or hot-blade beak trimming at the farm at 8 D of age (HB-8).

³ $n = 36$ for initial BW and $n = 24$ for beak trimming procedure.

⁴The interactions among main effects and between main effects and time period were not significant ($P > 0.05$).

Table 3. Influence of initial BW and beak trimming procedure on BW (g) and BW uniformity¹ (UNI) of the pullets: Experiment 1.

	Main effect					SEM ⁴	Probability ⁵	
	Initial BW ²		Beak trimming ³				Initial BW	Beak trimming
	Light	Heavy	MI-0	AG-0	HB-8			
BW, g								
5 wk	345	348	352 ^a	343 ^b	343 ^b	2.69	0.338	0.019
10 wk	969	970	982	961	967	7.15	0.905	0.099
15 wk	1,384	1,408	1,423	1,374	1,391	16.63	0.200	0.010
UNI, %								
0 wk	3.8	3.9	3.8	3.9	3.8	0.21	0.349	0.732
5 wk	8.9	9.8	9.4	9.8	8.9	0.54	0.138	0.545
10 wk	7.4	7.9	8.2	7.2	7.6	0.56	0.418	0.427
15 wk	7.6	7.6	8.1	7.3	7.5	0.58	0.995	0.594

^{a,b}Within a line, means without a common superscript differ significantly ($P < 0.05$).

¹Evaluated as the CV (%) of the individual BW of the pullets from each cage (Peak et al., 2000).

²Light = 33.9 ± 1.3 g BW; heavy = 37.6 ± 1.2 g BW.

³Infrared beak treatment at hatch with high (48; MI-0) and low (46; AI-0) setting or hot-blade beak trimming at the farm at 8 D of age (HB-8).

⁴ $n = 36$ for initial BW and $n = 24$ for beak trimming procedure.

⁵The interactions among main effects and between main effects and time period were not significant ($P > 0.05$).

RESULTS

Experiment 1

Mortality was 1.7% and no differences among treatments were detected throughout the experiment (data not shown). Most of the mortality (1.2%) occurred during the first week of the experiment. The percentage of pullets weighing less than 33.0 g was 23% and 0% for the light and heavy pullet groups, respectively. No interactions among main effects were detected for any of the variables studied and therefore, only main effects are presented.

Growth Performance Initial BW of the pullets did not have any effect on growth performance or BW uni-

formity of the birds in any of the periods considered (Tables 2 and 3, respectively).

From 0 to 5 wk of age, ADFI was higher for the MI-0 pullets than for the HB-8 pullets, with the AG-0 pullets being intermediate (19.8, 19.1, and 19.3 g, respectively; $P < 0.01$). In addition, MI-0 pullets had higher ADG than the AG-0 and HB-8 pullets (9.0 vs. 8.8 and 8.8 g; $P < 0.05$). From 5 to 10 wk and from 10 to 15 wk of age, previous beak trimming procedure had no effect on pullet ADFI, ADG, and FCR. As a result, from 0 to 15 wk of age, ADFI was greater for the MI-0 pullets than for the HB-8 pullets, with the AG-0 pullets being intermediate (50.5, 49.2, and 49.8 g, respectively; $P < 0.05$). However, ADG, FCR, and BW uniformity were not affected by beak trimming procedure.

Table 4. Influence of initial BW and beak trimming procedure on the relative weight (RW; % BW) and fresh digesta content (% organ weight) of the gastrointestinal tract (GIT): Experiment 1.

	Main effect					SEM ³	Probability ⁴	
	Initial BW ¹		Beak trimming ²				Initial BW	Beak trimming
	Light	Heavy	MI-0	AG-0	HB-8			
Full GIT, RW								
5 wk	21.3	21.3	21.0	21.4	21.5	0.444	0.934	0.731
10 wk	16.7	16.2	16.0	16.5	16.8	0.444	0.377	0.410
15 wk	13.4	13.6	13.5	13.5	13.6	0.213	0.367	0.939
Crop, RW								
5 wk	4.01	3.98	4.03	3.90	4.06	0.369	0.934	0.949
10 wk	3.84	3.14	3.32	3.40	3.75	0.377	0.113	0.689
15 wk	2.00	1.96	1.93	2.06	1.96	0.383	0.911	0.966
Crop content, %								
5 wk	45.2	45.5	45.8	44.4	45.7	3.531	0.941	0.954
10 wk	59.0	58.0	56.0	56.7	62.4	4.782	0.822	0.593
15 wk	57.5	58.0	55.2	60.7	57.4	3.023	0.881	0.426
Gizzard, RW								
5 wk	5.02	5.08	5.01	5.11	5.03	0.071	0.463	0.539
10 wk	4.39	4.48	4.42	4.53	4.36	0.067	0.216	0.216
15 wk	3.50	3.59	3.56	3.52	3.55	0.071	0.269	0.905
Gizzard content, %								
5 wk	35.7	35.6	35.1	36.0	35.8	0.868	0.972	0.698
10 wk	29.8	30.3	29.2	30.2	30.8	0.890	0.596	0.420
15 wk	28.8	29.4	29.4	28.6	29.1	0.903	0.531	0.811

¹Light = 33.9 ± 1.3 g BW; heavy = 37.6 ± 1.2 g BW

²Infrared beak treatment at hatch with high (48; MI-0) and low (46; AI-0) setting or hot-blade beak trimming at the farm at 8 D of age (HB-8).

³n = 36 for initial BW of the pullets and n = 24 for beak trimming procedure.

⁴The interactions among main effects and between main effects and time period were not significant ($P > 0.05$).

Table 5. Influence of initial BW and beak trimming procedure on the pH of the crop, gizzard, ileum, and cecum: Experiment 1.

	Main effect					SEM ³	Probability ⁴	
	Initial BW ¹		Beak trimming ²				Initial BW	Beak trimming
	Light	Heavy	MI-0	AG-0	HB-8			
Crop								
5 wk	5.67	5.65	5.60	5.70	5.68	0.079	0.944	0.583
10 wk	4.95	4.91	4.94	4.85	5.01	0.082	0.705	0.374
15 wk	4.31	4.39	4.26	4.38	4.41	0.083	0.460	0.426
Gizzard								
5 wk	2.95	2.97	2.99	3.00	2.90	0.076	0.767	0.555
10 wk	2.78	2.85	2.84	2.80	2.81	0.078	0.508	0.811
15 wk	2.57	2.55	2.64	2.60	2.44	0.079	0.771	0.156
Ileum								
5 wk	6.77	6.78	6.80	6.81	6.72	0.045	0.822	0.254
10 wk	7.22	7.25	7.26	7.23	7.21	0.048	0.566	0.756
15 wk	6.92	6.92	7.00	6.88	6.88	0.049	0.892	0.107
Cecum								
5 wk	5.33	5.35	5.28	5.37	5.37	0.060	0.753	0.537
10 wk	5.95	6.07	6.02	6.00	5.99	0.062	0.108	0.968
15 wk	6.21	6.14	6.16	6.24	6.13	0.063	0.273	0.440

¹Light = 33.9 ± 1.3 g BW; heavy = 37.6 ± 1.2 g BW.

²Infrared beak treatment at hatch with high (48; MI-0) and low (46; AI-0) setting or hot-blade beak trimming at the farm at 8 D of age (HB-8).

³n = 36 for initial BW of the pullets and n = 24 for beak trimming procedure.

⁴The interactions among main effects and between main effects and time period were not significant ($P > 0.05$).

GIT Traits and Shank Length The main effects of initial BW and beak trimming procedure on GIT, crop, and gizzard weights, and pH of the organs of the GIT, are shown in Tables 4 and 5, respectively. Data on GIT and shank length are shown in Table 6. Neither initial BW of the pul-

lets nor beak trimming procedure affected any of the GIT traits or body measurements studied at any age.

Bacterial Count in the Excreta Treatment did not affect bacteria counts in the excreta at 5 wk of age (Table 7).

Table 6. Influence of initial BW and beak trimming procedure on the relative length (cm/kg) of the cecum, small intestine (SI), and tarsus of the pullets: Experiment 1.

	Main effect						Probability ⁴	
	Initial BW ¹		Beak trimming ²			SEM ³	Initial BW	Beak trimming
	Light	Heavy	MI-0	AG-0	HB-8			
Cecum								
5 wk	29.3	29.3	28.9	29.6	29.4	0.405	0.991	0.378
10 wk	29.0	29.6	29.0	29.3	29.6	0.415	0.191	0.597
15 wk	23.5	23.1	22.9	23.6	23.4	0.421	0.362	0.423
SI								
5 wk	275.1	272.0	270.6	273.9	276.2	2.833	0.348	0.368
10 wk	116.0	118.4	115.1	117.7	118.8	2.906	0.461	0.648
15 wk	87.2	85.2	86.2	86.8	85.5	2.948	0.556	0.956
Tarsus								
5 wk	14.1	13.9	14.0	14.0	14.0	0.125	0.192	0.988
10 wk	7.72	7.79	7.73	7.82	7.72	0.128	0.619	0.835
15 wk	5.91	5.67	5.75	5.92	5.69	0.129	0.106	0.432

¹Light = 33.9 ± 1.3 g BW; heavy = 37.6 ± 1.2 g BW.

²Infrared beak treatment at hatch with high (48; MI-0) and low (46; AI-0) setting or hot-blade beak trimming at the farm at 8 D of age (HB-8).

³n = 36 for initial BW of the pullets and n = 24 for beak trimming procedure.

⁴The interactions among main effects and between main effects and time period were not significant ($P > 0.05$).

Table 7. Influence of initial BW and beak trimming procedure on bacteria count (log₁₀ cfu/g) in the excreta of pullets at 5 wk of age: Experiment 1.

	Main effect					SEM ³	Probability ⁴	
	Initial BW ¹		Beak trimming ²				Initial BW	Beak trimming
	Light	Heavy	MI-0	AG-0	HB-8			
Lactobacilli	7.56	7.60	7.61	7.51	7.62	0.137	0.779	0.806
<i>Clostridium perfringens</i>	2.08	1.92	1.88	2.08	1.99	0.252	0.576	0.745
Enterobacteriaceae	7.95	7.96	8.08	7.77	8.03	0.116	0.968	0.135
Total coliforms	7.74	7.73	7.81	7.56	7.81	0.115	0.893	0.309
<i>Escherichia coli</i> ⁵	7.75	7.72	7.87	7.53	7.81	0.121	0.830	0.116

¹Light = 33.9 ± 1.3 g BW; heavy = 37.6 ± 1.2 g BW.

²Infrared beak treatment at hatch with high (48; MI-0) and low (46; AI-0) setting or hot-blade beak trimming at the farm at 8 D of age (HB-8).

³n = 36 for initial BW of the pullets and n = 24 for beak trimming procedure.

⁴The interactions among main effects were not significant ($P > 0.05$).

⁵β-glucuronidase (+).

Experiment 2

Mortality was 1.4% and no differences among treatments were detected throughout the experiment (data not shown). Most of the mortality (1.1%) occurred during the first week of the experiment. The percentage of pullets weighing less than 33.0 g was 28% and 0% for the light and heavy pullets, respectively.

Growth Performance Data on pullet performance and BW uniformity by week are shown in Tables 8 and 9, respectively. Cumulatively (0 to 6 wk of age), ADFI was higher ($P < 0.01$) for pullets beak treated at hatch than for pullet beak trimmed at the farm (Table 8). However, the ADG and FCR were not affected by beak trimming procedure.

Several interactions on growth performance traits between beak trimming and age were detected (Figure 1). For the first week of life, pullets of the HB-8 group (not trimmed at hatch) showed higher ADFI and ADG ($P < 0.001$) and better FCR ($P < 0.01$) than pullets of the MI-0 and AG-0 groups (trimmed at hatch). From 1 to 2 wk of age, however, an opposite ef-

fect was observed, with pullets beak trimmed at hatch (MI-0 and AG-0 groups) showing greater ADFI and ADG and better FCR ($P < 0.001$) than pullets beak trimmed on day 8 at the farm (HB-8 group). As a consequence, from 0 to 4 wk of age, heavier pullets at hatch had greater BW than lighter pullets (254 vs. 249 g; $P < 0.05$) but no interactions were observed after this age.

BW uniformity was not affected by beak trimming procedure at any age, except at 7 D of age in which non-trimmed pullets (HB-8 group) showed better uniformity than AG-0 pullets, with MI-0 pullets being in an intermediate position (7.3, 10.9, and 9.4%, respectively; $P < 0.001$) (Table 9).

The inclusion of Na butyrate in the diet tended to improve ADG ($P = 0.073$) and FCR ($P = 0.069$) from 0 to 6 wk of age (Table 8) but BW uniformity was not affected (Table 9). Most of the benefits of Na-butyrate inclusion on growth performance and feed efficiency were observed during the first 2 wk of life.

VFA in the Crop and Ileum Contents Total VFA production and VFA profile in the crop and ileum are

Table 8. Influence of initial BW of the pullets, beak trimming procedure, and inclusion of Na butyrate in the diet, and period on growth performance from 0 to 6 wk of age: Experiment 2.

	Main effect								Probability ⁴		
	Initial BW ¹		Beak trimming ²			Na butyrate		SEM ³	Initial BW	Beak trimming	Na butyrate
	Light	Heavy	MI-0	AG-0	HB-8	0%	0.3%				
ADFI, g											
0 to 1 wk	8.9	9.0	8.7 ^b	8.6 ^b	9.6 ^a	9.0	9.0	0.178	0.573	0.001	0.826
1 to 2 wk	14.8	14.6	15.2 ^a	15.2 ^a	13.7 ^b	14.6	14.8	0.307	0.435	0.001	0.474
2 to 3 wk	17.7	17.8	18.1 ^a	17.8 ^a	17.3 ^b	17.7	17.8	0.181	0.578	0.003	0.507
3 to 4 wk	24.1	24.5	24.7 ^a	24.4 ^a	23.7 ^b	24.2	24.4	0.204	0.109	0.005	0.498
4 to 5 wk	31.4	31.4	32.1	31.2	31.0	31.5	31.4	0.390	0.970	0.137	0.884
5 to 6 wk	41.6	41.1	42.1 ^a	41.5 ^{a,b}	40.5 ^b	41.1	41.6	0.415	0.276	0.026	0.278
ADG, g											
0 to 1 wk	3.69	3.64	3.49 ^b	3.45 ^b	4.06 ^a	3.62	3.71	0.062	0.482	<0.001	0.226
1 to 2 wk	6.53	6.62	7.07 ^a	6.99 ^a	5.68 ^b	6.43	6.73	0.088	0.376	<0.001	0.004
2 to 3 wk	9.19	9.11	9.16	9.08	9.20	9.11	9.18	0.110	0.529	0.743	0.612
3 to 4 wk	11.3	11.5	11.6	11.3	11.4	11.4	11.5	0.106	0.176	0.292	0.658
4 to 5 wk	13.4	13.5	13.7	13.3	13.4	13.4	13.5	0.210	0.832	0.367	0.583
5 to 6 wk	17.3	17.3	17.6	17.1	17.4	17.0	17.6	0.312	0.999	0.625	0.098
FCR, g/g											
0 to 1 wk	2.43	2.50	2.52 ^a	2.50 ^a	2.37 ^b	2.50	2.42	0.036	0.114	0.008	0.074
1 to 2 wk	2.29	2.21	2.15 ^b	2.18 ^b	2.42 ^a	2.28	2.22	0.036	0.071	<0.001	0.161
2 to 3 wk	1.92	1.96	1.98	1.97	1.88	1.94	1.94	0.036	0.428	0.084	0.993
3 to 4 wk	2.12	2.13	2.14	2.15	2.08	2.12	2.13	0.036	0.874	0.406	0.911
4 to 5 wk	2.35	2.33	2.34	2.36	2.31	2.36	2.33	0.036	0.708	0.651	0.487
5 to 6 wk	2.41	2.39	2.41	2.44	2.35	2.43	2.37	0.037	0.554	0.271	0.225
Cumulative (0 to 6 wk)											
ADFI, g	23.1	23.1	23.5 ^a	23.1 ^a	22.6 ^b	23.0	23.2	0.163	0.856	0.003	0.397
ADG, g	10.3	10.3	10.4	10.2	10.2	10.2	10.4	0.098	0.794	0.187	0.073
FCR, g/g	2.25	2.25	2.26	2.27	2.24	2.27	2.24	0.016	0.928	0.472	0.069

^{a,b}Within a line, means without a common superscript differ significantly ($P < 0.05$).

¹Light = 34.2 g BW; heavy = 37.9 g BW.

²Infrared beak treatment at hatch with high (48; MI-0) and low (46; AI-0) setting or hot-blade beak trimming at the farm at 8 D of age (HB-8).

³ $n = 18$ for Na-butyrate inclusion and initial BW of the pullets and $n = 24$ for beak trimming procedure.

⁴The interactions among main effects were not significant ($P > 0.05$).

shown in Table 10. Initial BW and beak trimming procedure did not affect VFA concentration or VFA profile of the crop or ileum contents at 6 wk of age. The inclusion of 0.3% Na butyrate in the diet, however, increased ($P < 0.001$) the proportion of butyrate in the crop.

DISCUSSION

Experiments 1 and 2

Growth Performance BW at hatch is affected by numerous factors, including species, egg size and egg composition, and environmental conditions during incubation and hatching (Shanawany, 1987; Wilson, 1991; Vieira and Moran, 1999). Most studies in broilers have shown that larger eggs result in heavier chicks both at hatch (Shanawany, 1984; Lopez and Leeson, 1994; Vieira and Moran, 1999) and at slaughter (Stringhini et al., 2003; Gomes et al., 2008). The ratio between pre-hatch egg weight and chick weight at hatch is approximately 67 to 68% in both broilers (Shanawany, 1987; Pinchasov, 1991; Lopez and Leeson, 1994) and pullets (Rodado et al., 2015). Wilson (1991) reported that each extra gram of weight at hatch led to 13 g increase in BW at 56 D of age. Mendes et al. (2011) reported that from 1 to 42 D of age, FI and BW gain were

2% greater for chicks that weighed 40.3 g at hatch than for chicks that weighed 34.8 g, although FCR was not affected. Similar results were observed by Sklan et al. (2003) and Gomes et al. (2008). The information available on the effects of BW at hatch on BW of mature pullets, however, is very limited. Based on field data, most managers of brown-egg layer operations recommend a minimum BW of the pullets at hatch of 32 to 33 g for optimal performance. However, the scientific evidence for this recommendation in flocks that are well managed during the rearing period is not apparent. In fact, Deaton et al. (1979) reported that a BW at hatch of 32.2 g vs. a BW of 35.2 g did not affect BW at 18 wk of age in pullets reared in cages. In the current research, initial BW did not affect pullet growth at any age, probably because the BW at hatch of the light pullets was not excessively low (average of 33.9 ± 0.5 g and 33.5 ± 0.5 g in experiments 1 and 2, respectively). In fact, the proportion of pullets weighing less 33 g was of 23 and 28% for the light pullets and 0% for the heavy pullets in experiments 1 and 2, respectively. Moreover, in the current research, pullets were sorted by BW at hatch, resulting in a high BW uniformity within each replicate in the 2 experimental BW groups. In fact, independent of the initial BW, growth and BW uniformity of the pullets were similar for all groups after the second week of age. Consequently, the potential

Table 9. Influence of initial BW, beak trimming procedure, and inclusion of Na butyrate in the diet on BW (g) and BW uniformity¹ (UNI) of the pullets: Experiment 2.

	Main effect											
	Initial BW ²		Beak trimming ³			Na butyrate		SEM ⁴	Probability ⁵			
	Light	Heavy	MI-0	AG-0	HB-8	0%	0.3%		Initial BW	Beak trimming	Na butyrate	
BW ⁴ , g												
1 wk	60	63	60 ^b	60 ^b	64 ^a	61	62	0.60	<0.001	<0.001	0.110	
2 wk	106	110	110 ^a	109 ^a	104 ^b	106	109	1.02	0.001	<0.001	0.015	
3 wk	170	173	174 ^a	172 ^{a,b}	169 ^b	170	173	1.32	0.027	0.018	0.032	
4 wk	249	254	255	252	249	250	254	1.83	0.028	0.058	0.070	
5 wk	344	349	351	345	343	344	348	2.75	0.115	0.097	0.135	
6 wk	465	469	473	463	464	463	471	4.23	0.375	0.164	0.096	
UNI, %												
0 wk	3.8	3.9	3.7	3.9	3.9	3.8	3.9	0.20	0.497	0.743	0.855	
1 wk	9.2	9.1	9.4 ^{a,b}	10.9 ^a	7.3 ^b	9.1	9.2	0.50	0.915	<0.001	0.807	
2 wk	11.5	10.4	10.9	11.6	10.4	11.2	10.7	0.69	0.177	0.434	0.555	
3 wk	10.0	10.5	10.5	10.7	9.7	10.2	10.4	0.53	0.422	0.390	0.707	
4 wk	9.8	10.4	10.1	10.7	9.5	10.1	10.0	0.53	0.316	0.277	0.843	
5 wk	9.0	9.8	9.6	9.8	8.9	9.5	9.3	0.55	0.230	0.535	0.709	
6 wk	8.8	9.8	9.4	9.3	9.2	9.7	8.9	0.57	0.125	0.964	0.245	

^{a,b}Within a line, means without a common superscript differ significantly ($P < 0.05$).

¹Evaluated as the CV (%) of the individual BW of the pullets from each cage (Peak et al., 2000).

²Light = 33.9 ± 1.3 g BW; heavy = 37.6 ± 1.2 g BW.

³Infrared beak treatment at hatch with high (48; MI-0) and low (46; AI-0) setting or hot-blade beak trimming at the farm at 8 D of age (HB-8).

⁴n = 36 for Na-butyrate inclusion and initial BW of the pullets and n = 24 for beak trimming method.

⁵ β -glucuronidase (+).

competition for feed and space among individual pullets, within each individual replicate, was limited. In this respect, Bray (1983) suggested that broiler growth was optimized and BW variability at slaughter was reduced, when the eggs were segregated by size at hatch and the birds were reared separately, consistent with the results reported herein. Moreover, Hearn (1986) reported that broilers from small hatching eggs reared separately according to BW, which presumably reduced competition, showed similar final BW than broilers from heavier eggs, in spite of differences in initial BW. Similar results have been reported by Wilson (1991) in pullets. Consequently, the segregation of the pullets by initial BW at hatch, together with the rearing the birds under good management practices, might facilitate the utilization of small eggs produced by young breeders, improving overall breeder flock profitability.

Untrimmed birds showed increased impact of feather pecking and cannibalism than properly trimmed birds (Sun et al., 2014). Beak trimming modifies the feeding behavior of the birds which need to adapt picking practices to the new form of the beak. If beak trimming is not conducted properly, the stress and pain caused might affect FI, reducing bird performance (Cheng, 2006). In this respect, the IRBT may result in less aggressive behavior of the birds than the hot-blade procedure and consequently, IRBT might be a good alternative to traditional hot-blade technology in commercial operations (Glatz, 2000; Dennis et al., 2009; Dennis and Cheng, 2010).

In the current research, trimming method did not affect pullet growth or BW uniformity at the end of the 2 experiments. In experiment 1, pullets beak trimmed at the farm (HB-8 group) or by the aggressive IRBT pro-

cedure at hatch (AG-0 group) showed lower ADG at 5 wk of age than pullets beak trimmed using the mild IRBT procedure (MI-0 group) but the birds recovered with time and no effects were detected at 10 or 15 wk of age. In experiment 2, pullets beak treated at hatch (MI-0 and AG-0 groups) showed a reduction in growth performance and BW uniformity for the first week of life compared to pullets of the HB-8 group. A similar loss in performance was observed in the HB-8 pullets the week after the trimming process took place. In both cases, the pullets recovered quickly, showing a compensatory growth within a week after beak treatment. In fact, all pullets had similar BW, FCR, and BW uniformity at 6 wk of age, irrespective of the method used and the age at beak treatment. These results indicate that pullets recover rapidly from the decrease in FI caused by beak trimming, provided that the procedure is conducted properly and the pullets are kept under good management practices during the rearing period.

In experiment 1, ADFI and ADG from 0 to 5 wk of age were lower for the AG-0 pullets than for the MI-0 pullets but no differences were observed after this age. Similar results were observed in experiment 2, although in this case the differences were not significant. The AG-0 protocol produced a more pronounced reduction of the length of the upper and lower parts of the beak, causing greater stress to the birds than the MI-0 protocol (Dennis and Cheng, 2012). Consequently, a more pronounced reduction in growth should be expected the week after trimming for the AG-0 group compared to the MI-0 group.

Numerous reports have shown positive effects of the inclusion of protected butyrate on intestinal microbiota profile, nutrient absorption, and growth performance in

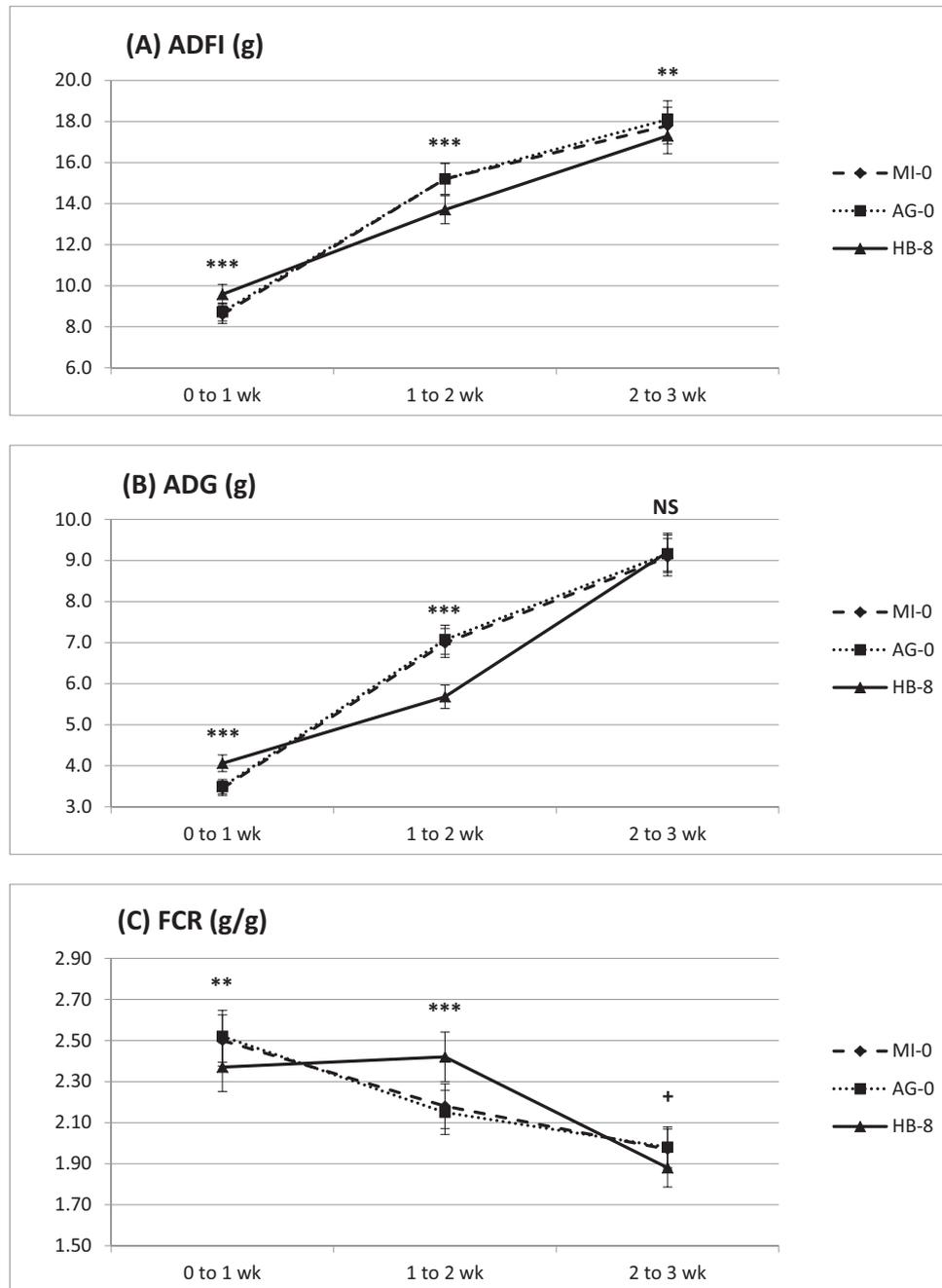


Figure 1. Interaction between beak trimming and time on ADFI (A), ADG (B), and FCR (C): Experiment 2. NS, no significant; + $P < 0.10$; * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$.

broilers (Antongiovanni et al., 2007; Adil et al., 2010; Chamba et al., 2014; Kaczmarek et al., 2016). However, the authors have not found any report on the potential benefits of Na-butyrate inclusion in pullets. In the current research, Na butyrate tended to improve ADG and FCR from 0 to 6 wk of age, with most of the benefits detected during the first 2 wk of life. This information is consistent with data of Hu and Guo (2007), who reported in broilers a linear increase in ADG from 0 to 21 D of age with 0.20% Na-butyrate supplementation.

GIT Traits and Tarsus Length Bird performance is influenced by the development of the GIT during the

first stages of life (Frikha, et al., 2009; Kimiaetalab et al., 2017). Sklan et al. (2003) reported that BW at hatch (43 g vs. 53 g) affected the development of the GIT of the bird but had no effects on BW and FCR at slaughter. The authors have not found any report on the effects of BW at hatch on the posterior development of the GIT in pullets. In the current research, initial BW did not affect the relative weight, length, or pH of the different organs of the GIT at any age. These results are consistent with the data on tarsus length and BW uniformity that were similar for both groups of pullets at all ages in experiment 1. Overall, the data reported suggest that light pullets at hatch, if properly

Table 10. Influence of initial BW, beak trimming procedure, and inclusion of Na butyrate in the diet on volatile fatty acid (VFA) profiles and total VFA (mmol/mL) in the crop and ileum of the pullets at 6 wk of age: Experiment 2.

	Main effect								Probability ⁴		
	Initial BW ¹		Beak trimming ²			Na butyrate		SEM ³	Initial BW	Beak trimming	Na butyrate
	Light	Heavy	MI-0	AG-0	HB-8	0%	0.3%				
Crop											
Acetate	9.10	10.5	9.08	9.28	11.0	10.5	9.13	1.324	0.358	0.502	0.371
Propionate	0.67	0.69	0.68	0.63	0.72	0.72	0.63	0.055	0.752	0.496	0.135
Butyrate	2.16	2.55	2.16	2.08	2.82	1.01	3.69	0.267	0.212	0.115	<0.001
Isovalerate	0.10	0.08	0.09	0.09	0.10	0.09	0.09	0.012	0.211	0.809	0.584
Total VFA	12.1	13.9	12.1	12.1	14.8	12.4	13.6	1.391	0.273	0.288	0.448
Ileum											
Acetate	9.08	10.9	10.1	10.7	9.14	10.5	9.46	1.311	0.232	0.694	0.485
Propionate	0.18	0.15	0.17	0.17	0.17	0.20	0.13	0.041	0.628	0.999	0.131
Butyrate	0.03	0.02	0.04	0.02	0.02	0.02	0.03	0.019	0.658	0.654	0.559
Isovalerate	0.004	0.009	0.01	0.01	0.00	0.009	0.004	0.006	0.404	0.390	0.404
Total VFA	9.48	11.1	10.3	11.0	9.56	10.9	9.62	1.406	0.310	0.764	0.400

¹Light = 33.9 ± 1.3 g BW; heavy = 37.6 ± 1.2 g BW.

²Infrared beak treatment at hatch with high (48; MI-0) and low (46; AG-0) setting or hot-blade beak trimming at the farm at 8 D of age (HB-8).

³n = 36 for Na-butyrate inclusion and initial BW of the pullets and n = 24 for beak trimming method.

⁴The interactions among main effects and between main effects and time period were not significant ($P > 0.05$).

reared, will have a GIT development similar to that of heavy pullets and consequently, hatching eggs from brown hens weighing more than 48.0 to 49.0 g, corresponding to pullets with an initial BW of 32 to 33 g, should not be discarded at hatch.

In the current research, beak trimming technique did not affect any GIT measurements at any age, consistent with growth performance data that was similar for both groups. The authors have not found any report on the effects of beak trimming of pullets on the posterior development of the GIT to compare with the data reported herein. Shank length has been used as a tool to predict future growth and performance of commercial broilers (Mendes et al., 2007; Willemsen et al., 2008; Van Roovert-Reijrink, 2013) and pullets (Guzmán et al., 2015; Saldaña et al., 2015). An increase in shank length suggests better bone development (Cleasby et al., 2011), which might be of benefit for future egg production (Senar and Pascual, 1997; Mendes et al., 2008). However, in the current research, none of the main factors tested affected shank length, consistent with the lack of effects on BW and BW uniformity reported at the end of the 2 experiments.

Bacterial Count in Excreta In experiment 1, treatment did not affect microbial count of the excreta at 5 wk of age. The authors have not find any report on the effects of initial BW or beak trimming procedure on microbial counts to compare with the data of the current research.

VFA in the Crop and Ileum Content In experiment 2, initial BW of the pullets and beak trimming procedure did not affect total VFA production or VFA profile in the crop or ileum at 6 wk of age. The inclusion of 0.3% Na butyrate in the diet had no effects on total VFA concentration or VFA profile in the ileum but increased butyrate concentration in the crop. The increase observed was unexpected because the Na butyrate used was protected and consequently,

butyric acid concentration should be similar for the Na-butyrate supplemented and non-supplemented groups. In the current research, crop contents were acidified immediately after collection by adding 0.5 N HCl to the sample to stop fermentation processes (Carro et al., 1992). The methodology used dropped the pH of the crop sample below 2.5. At low pH, the encapsulation of the Na butyrate is not effective, resulting in the release of free butyric acid. In the ileum, no differences in butyric acid concentration between diets were detected, indicating that the butyric acid contained in the commercial product was released and absorbed in the proximal section of the SI. These data confirm that the commercial product used was correctly formulated and processed.

In summary, growth performance and BW uniformity of the pullets at 5, 10, and 15 wk of age in experiment 1 and at 6 wk of age in experiment 2 were not affected by the initial BW of the birds, suggesting that healthy pullets, weighing 32 to 33 g at hatch, might not differ on later growth performance compared to pullets weighing 37 to 38 g, if properly managed during the rearing period. Beak treatment decreased bird performance for the first days after the process took place but the birds recovered quickly with age, and the negative effects on growth performance or BW uniformity disappeared at later ages. The beak treatment at hatch by infrared technology might be a recommended commercial practice because it facilitates flock management and reduces the chances of potential microbial contamination of the flock at the farm. The benefits of the inclusion of 0.3% Na butyrate in the diet on pullet growth were more evident in the early stages of life.

ACKNOWLEDGMENTS

We acknowledge the help of Yolanda Alegre and Dolores Carro of the Department of Animal Science in the

formal presentation and discussion of the results of the manuscript, respectively.

REFERENCES

- Adil, S., T. Banday, G. A. Bhat, M. S. Mir, and M. Rehman. 2010. Effect of dietary supplementation of organic acids on performance, intestinal histomorphology, and serum biochemistry of broiler chicken. *Vet. Med. Int.* 10:4061–4067.
- Ahsan, U., Ö. Cengizi, I. Raza, E. Kuter, M. F. A. Chacher, Z. Iqbal, S. Umar, and S. Çakir. 2016. Sodium butyrate in chicken nutrition: the dynamics of performance, gut microbiota, gut morphology, and immunity. *Worlds Poult. Sci. J.* 72:265–276.
- Antongiovanni, M., A. Buccioni, F. Petacchi, S. Leeson, S. Minieri, A. Martini, and R. Cecchi. 2007. Butyric acid glycerides in the diet of broiler chickens: effects on gut histology and carcass composition. *Italian J. Anim. Sci.* 6:19–25.
- AOAC International. 2005. *Official Methods of Analysis*. 17th edn. AOAC, Gaithersburg, MD.
- AOCS, 2004. *Official Methods and Recommended Practices of the AOCS*, 5th ed. Am. Oil Chem. Soc., Champaign, IL.
- ASAE. 2003. *Method of Determining and Expressing Fineness of Feed Materials by Sieving*. ASAE standard S319.2. Agriculture Engineers Yearbook of Standards. ASAE, St. Joseph, MO.
- BOE (Boletín Oficial del Estado). 2013. Ley 53/2013 de 1 de febrero por el que se establecen las normas básicas aplicables para la protección de los animales utilizados en experimentación y otros fines científicos, incluyendo la docencia. *BOE* 34:11, 370-11,421.
- Bray, T. S. 1983. Broiler chick weight—Does it matter? *Gleadthorpe Experimental Husbandry Farm. Poult. Booklet* 11: 17–20.
- Carro, M. D., P. Lebzien, and K. Rohr. 1992. Effects of yeast culture on rumen fermentation, digestibility and duodenal flow in dairy cows fed a silage based diet. *Anim. Feed. Sci. Technol.* 32:219–229.
- Cerisuelo, A., C. Marin, F. Sanchez-Vizcaino, E. A. Gomez, J. M. de la Fuente, R. Duran, and C. Fernandez. 2014. The impact of a specific blend of essential oil components and sodium butyrate in feed on growth performance and Salmonella counts in experimentally challenged broilers. *Poult. Sci.* 93:599–606.
- Chamba, F., M. Puyalto, A. Ortiz, H. Torralba, J. J. Mallo, and R. Riboty. 2014. Effect of partially protected sodium butyrate on performance, digestive organs, intestinal villi and *E. coli* development in broilers chickens. *Int. J. Poult. Sci.* 13:390–396.
- Cheng, H. W. 2006. Morphopathological changes and pain in beak trimmed laying hens. *Worlds Poult. Sci. J.* 62:41–52.
- Cleasby, I. R., T. Burke, J. Schroeder, and S. Nakagawa. 2011. Food supplements increase adult tarsus length, but not growth rate, in an island population of house sparrows (*Passer domesticus*). *BMC Res Notes* 4:431.
- De Coca-Sinova, A. D., G. Valencia, E. Jiménez-Moreno, J. M. González-Alvarado, R. Lázaro, and G. G. Mateos. 2008. Apparent ileal digestibility of nitrogen, amino acids, and energy of soybean meals from different origins in broilers. *Poult. Sci.* 87:2613–2623.
- Deaton, J. W., J. L. McNaughton, and F. N. Reece. 1979. Relationship of initial chick weight to body weight of egg-type pullets. *Poult. Sci.* 58:960–962.
- Dennis, R. L., A. G. Fahey, and H. W. Cheng. 2009. Infrared beak treatment method compared with conventional hot-blade trimming in laying hens. *Poult. Sci.* 88:38–43.
- Dennis, R. L., and H. W. Cheng. 2010. A comparison of infrared and hot blade beak trimming in laying hens. *Int. J. Poult. Sci.* 9:716–719.
- Dennis, R. L., and H. W. Cheng. 2012. Effects of different infrared beak treatment protocols on chicken welfare and physiology. *Poult. Sci.* 91:1499–1505.
- Duncan, I. J. H., G. S. Slee, E. Seawright, and J. Breward. 1989. Behavioural consequences of partial beak amputation (beak trimming) in poultry. *Br. Poult. Sci.* 30:479–488.
- FEDNA (Fundación Española para el Desarrollo de la Nutrición Animal). 2010. *Necesidades Nutricionales para Avicultura: Pollos de Carne y Aves de Puesta*. R. Lázaro, and G. G. Mateos, eds. FEDNA, Madrid, Spain.
- FEDNA (Fundación Española para el Desarrollo de la Nutrición Animal). 2018. *Necesidades Nutricionales para Avicultura: Pollos de Carne y Aves de Puesta*. R. Lázaro, and G. G. Mateos, eds. FEDNA, Madrid, Spain.
- Frikha, M., H. M. Safaa, M. P. Serrano, X. Arbe, and G. G. Mateos. 2009. Influence of the main cereal and feed form of the diet on performance and digestive tract traits of brown-egg laying pullets. *Poult. Sci.* 88:994–1002.
- Gentle, M. J. 1986. Beak trimming in poultry. *World's Poult. Sci. J.* 42:268–275.
- Gentle, M. J., B. O. Hughes, A. Fox, and D. Waddington. 1997. Behavioural and anatomical consequences of two beak trimming methods in 1- and 10-d-old domestic chicks. *Br. Poult. Sci.* 38:453–463.
- Glatz, P. C. 2000. Beak trimming methods. *Asian-Australas. J. Anim. Sci.* 13:1619–1637.
- Glatz, P. C. 2005. *Poultry Welfare Issues: Beak Trimming*. Nottingham University Press, Nottingham, England.
- Gomes, G. A., L. F. Araújo, J. A. Prezzi, D. Savietto, J. R. S. Junior, and J. Valerio. 2008. Period of feeding a pre-starter diet for broiler chickens with different body weights at housing. *Braz. J. Poult. Sci.* 37:1802–1807.
- Guilloteau, P., L. Martin, V. Eeckhaut, R. Ducatelle, R. Zabielski, and F. Van Immerseel. 2010. From the gut to the peripheral tissues: the multiple effects of butyrate. *Nutr. Res. Rev.* 23: 366–384.
- Guzmán, P., B. Saldaña, H. Mandalawi, A. Pérez-Bonilla, and G. G. Mateos. 2015. Productive performance of brown-egg laying pullets from hatching to 5 weeks of age as affected by fiber inclusion, feed form, and energy concentration of the diet. *Poult. Sci.* 94:249–261.
- Harrigan, W. F. 1998. *Laboratory Methods in Food Microbiology*. 3rd ed. Academic Press, San Diego.
- Hearn, P. J. 1986. Making use of small hatching eggs in an integrated broiler company. *Br. Poult. Sci.* 27:498–504.
- Hu, Z., and Y. Guo. 2007. Effects of dietary sodium butyrate supplementation on the intestinal morphological structure, absorptive function and gut flora in chickens. *Anim. Feed Sci. Technol.* 132:240–249.
- Hughes, B. O., and M. J. Gentle. 1995. Beak trimming of poultry: its implications for welfare. *Worlds Poult. Sci. J.* 51:51–61.
- Jerzsele, A., K. Szeke, R. Csizinszky, E. Gere, C. Jakab, J. J. Mallo, and P. Galfi. 2012. Efficacy of protected sodium butyrate, a protected blend of essential oils, their combination, and *Bacillus amyloliquefaciens* spore suspension against artificially induced necrotic enteritis in broilers. *Poult. Sci.* 91:837–843.
- Jiménez-Moreno, E., J. M. González-Alvarado, A. de Coca-Sinova, R. Lázaro, and G. G. Mateos. 2009. Effects of source of fibre on the development and pH of the gastrointestinal tract of broilers. *Anim. Feed Sci. Technol.* 154:93–101.
- Kaczmarek, S. A., A. Barri, M. Hejdysz, and A. Rutkowski. 2016. Effect of different doses of coated butyric acid on growth performance and energy utilization in broilers. *Poult. Sci.* 95:851–859.
- Kimiaetalab, M. V., L. Cámara, S. Mirzaie Goudarzi, E. Jiménez-Moreno, and G. G. Mateos. 2017. Effects of the inclusion of sunflower hulls in the diet on growth performance and digestive tract traits of broilers and pullets fed a broiler diet from zero to 21 d of age. A comparative study. *Poult. Sci.* 96:581–592.
- Leeson, S., H. Namkung, M. Antongiovanni, and E. H. Lee. 2005. Effect of butyric acid on the performance and carcass yield of broiler chickens. *Poult. Sci.* 84:1418–1422.
- Littell, R. C., P. R. Henry, and C. B. Ammerman. 1998. Statistical analysis of repeated measures data using SAS procedures. *J. Anim. Sci.* 76:1216–1231.
- Liu, J. D., H. O. Bayir, D. E. Cosby, N. A. Cox, S. M. Williams, and J. Fowler. 2017. Evaluation of encapsulated sodium butyrate on growth performance, energy digestibility, gut development, and Salmonella colonization in broilers. *Poult. Sci.* 96:3638–3644.
- Lohmann. 2017. *Management Guide*, Lohmann Brown Classic. Lohmann Tierzucht. Cuxhaven, Germany.

- Lopez, G., and S. Leeson. 1994. Nutrition and broiler breeder performance: a review with emphasis on response to diet protein. *J. Appl. Poult. Res.* 3:303–311.
- Mateos, G. G., E. Jiménez-Moreno, M. P. Serrano, and R. Lázaro. 2012. Poultry response to high levels of dietary fiber sources varying in physical and chemical characteristics. *J. Appl. Poult. Res.* 21:156–174.
- Mateos, G. G., R. Lázaro, and M. I. Gracia. 2002. The feasibility of using nutritional modifications to replace drugs in poultry feeds. *J. Appl. Poult. Res.* 11:437–452.
- Mendes, M., D. Ecmel, and E. Arslan. 2007. Profile analysis and growth curve for body mass index of broiler chickens reared under different feed restrictions in early age. *Arch. Tierz.* 50:403–411.
- Mendes, A. S., S. J. Paixão, R. Restelatto, R. Reffatti, J. C. Possenti, D. J. de Moura, G. M. Z. Morello, and T. M. R. de Carvalho. 2011. Effects of initial body weight and litter material on broiler production. *Rev. Bras. Cienc. Avic.* 13:165–170.
- Mendes, M., A. Pala, and E. Dincer. 2008. Body mass index slopes of growth and fat content under different feed restrictions in broiler chickens. *Arch. Gefl.* 72:41–45.
- Patten, J. D., and P. W. Waldroup. 1988. Use of organic acids in broiler diets. *Poult. Sci.* 67: 1178–1182.
- Peak, S. D., T. J. Walsh, C. E. Benton, and J. Brake. 2000. Effects of two planes of nutrition on performance and uniformity of four strains of broiler chicks. *J. Appl. Poult. Res.* 9:185–194.
- Pinchasov, Y. 1991. Relationship between the weight of hatching eggs and subsequent early performance of broiler chicks. *Br. Poult. Sci.* 32:109–115.
- Proudfoot, F. G., and H. W. Hulan. 1981. The influence of hatching egg size on the subsequent performance of broiler chickens. *Poult. Sci.* 60:2167–2170.
- Ricke, S. C. 2003. Perspectives on the use of organic acids and short chain fatty acids as antimicrobials. *Poult. Sci.* 82:632–639.
- Rodado, S. M., B. Saldaña, P. Guzmán, H. A. Mandalawi, R. Rodríguez, L. Cámara, and G. G. Mateos. 2015. Influence of body weight at hatching and inclusion of oat hulls in the diet on growth performance and digestive tract traits of brown-egg laying pullets from 0 to 16 wk of age. *Poult. Sci.* 104 (Suppl. 1):222 (Abstr.).
- SAS Institute. 2004. SAS STATs User's Guide. Version 9.0, SAS Institute Inc., Cary, NC.
- Saldaña, B., P. Guzmán, H. M. Safaa, R. Harzalli, and G. G. Mateos. 2015. Influence of the main cereal and feed form of the rearing phase diets on performance, digestive tract, and body traits of brown-egg laying pullets from hatch to 17 weeks of age. *Poult. Sci.* 94:2650–2661.
- Senar, J. E., and J. Pascual. 1997. Keel and tarsus length may provide a good predictor of avian body size. *Ardea* 85:269–274.
- Shanawany, M. M. 1984. Inter-relationship between egg weight, parental age and embryonic development. *Br. Poult. Sci.* 25:449–455.
- Shanawany, M. M. 1987. Hatching weight in relation to egg weight in domestic birds. *World's Poult. Sci. J.* 43:107–115.
- Sklan, D., S. Heifetz, and O. Halevy. 2003. Heavier chicks at hatch improves marketing body weight by enhancing skeletal muscle growth. *Poult. Sci.* 82:1778–1786.
- Stringhini, J. H., A. Resende, M. B. Cafe, N. S. M. Leandro, and M. A. Andrade. 2003. Effect of one-day chicks weight and pre-starter diets on broiler performance. *Braz. J. Poult. Sci.* 32:353–360.
- Sun, Y., E. D. Ellen, J. J. van der Poel, H. K. Parmentier, and P. Bijma. 2014. Modelling of feather pecking behavior in beak-trimmed and non-beak-trimmed crossbred laying hens: variance component and trait-based approach. *Poult. Sci.* 93:773–783.
- Tona, K., O. Onagbesan, B. De Ketelaere, E. Decuyper, and V. Bruggeman. 2004. Effects of age of broiler breeders and egg storage on egg quality, hatchability, chick quality, chick weight, and chick posthatch growth to forty-two days. *J. Appl. Poult. Res.* 13:10–18.
- Ulmer-Franco, A. M., G. M. Fasenko, and E. E. O'Dea Christopher. 2010. Hatching egg characteristics, chick quality, and broiler performance at 2 breeder flock ages and from 3 egg weights. *Poult. Sci.* 89:2735–2742.
- Van Roover-Reijrink, I. 2013. Incubation affects chick quality. *World's Poult.* 29:22–23.
- Van Soest, P. J., J. B. Robertson, and A. Lewis. 1991. Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. *J. Dairy Sci.* 74:3583–3597.
- Vieira, S. L., and E. T. Moran, Jr. 1999. Effects of egg of origin and chick post-hatch nutrition on broiler live performance and meat yields. *World's Poult. Sci. J.* 55:125–142.
- Willemsen, H., N. Everaert, A. Witters, L. De Smit, M. Debonne, F. Verschuere, P. Garain, D. Berckmans, E. Decuyper, and V. Bruggeman. 2008. Critical assessment of chick quality measurements as an indicator of posthatch performance. *Poult. Sci.* 87:2358–2366.
- Wilson, H. R. 1991. Interrelationships of egg size, chick size, posthatching growth and hatchability. *World's Poult. Sci. J.* 47:5–20.
- Windisch, W., K. Schedle, C. Plitzner, and A. Kroismayr. 2008. Use of phytogetic products as feed additives for swine and poultry1. *J. Anim. Sci.* 86:E140–E148.
- Zhang, W. H., Y. Jiang, Q. F. Zhu, F. Gao, S. F. Dai, J. Chen, and G. H. Zhou. 2011. Sodium butyrate maintains growth performance by regulating the immune response in broiler chickens. *Br. Poult. Sci.* 52:292–301.