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Evaluation of a Commercially Available Probiotic and Organic Acid Blend Product on Production Parameters and Economics in Broiler Breeders

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Abstract

The effects of supplementing a blend of organic acids and a lactic acid bacteria based-probiotic on production parameters of broiler breeders was examined in the present study. Nine thousand females and one thousand males Cobb 500 breeders were used in three independent trials during the years 2013, 2014 and 2015. In each trial, during 10 consecutive weeks, starting from 25 to 35 weeks of age, breeders were divided into two groups: Control group, birds receiving regular water and as Treatment group, birds receiving organic acids plus probiotic in drinking water. No significant differences (P>0.05) on body weight and uniformity in females and males were observed between treatments in any of the three trials. A significant reduction in the percentage of deformed eggs, and weekly-cumulative mortality were observed in all three trials in breeders that received the blend of organic acids and probiotic compared to control non-treated birds. During the first trial in 2013, there was a cost benefit ratio of only 1:0.75 USD, represented by 474 extra hatched chicks in favor of the treated group when compared with the control group. However, in the following trials in 2014 and 2015, breeders that received the blend of organic acids followed by the probiotic had 5,465 and 5,629 extra hatched chicks when compared with the control group. This difference in the number of hatched chicks resulted in a cost benefit ratio of 1:4.41 and 1:4.40 USD respectively, in favor of the treated broiler breeders. When converted to cost benefit ratio, the numbers of all three trials suggest that for every U.S. dollar spent on the program of organic acids plus probiotic, producers may be able to recover on average 3.19 USD. These data suggest that the combination of organic acids and a probiotic may improve production and economic parameters by increasing the number of hatching chicks.

Keywords: Broiler breeder; Organic acid; Probiotic; Economics

Introduction

The GIT is more densely populated with microorganisms than any other organ and is an interface where the microflora may have a pronounced impact on animal biology [1]. More than 50 genera and at least 500–1,000 different species are distributed along the length of the gastrointestinal tract [2]. The bacterial population of the human cecum and colon is numerically ~10^{13} cfu/g [2], comprising to about 40–55% of solid stool matter and weights ~1 kg [3]. Presumably, the assembly of gut microflora is regulated by elaborate and combinatorial host–microbial and microbial–microbial interactions predicated on principles refined over the course of evolution [4]. Comparison of rodents raised without exposure to any microorganisms, to those colonized with an assembly of microbiota revealed a wide range of host functions affected by indigenous microbial communities. For example, the microbiota directs the assembly of the gut-associated lymphoid tissue [5], helps educate the immune system [6], affects the integrity of the intestinal mucosal barrier [7], modulates proliferation and differentiation of its epithelial lineages [8], regulates angiogenesis [9], modifies the activity of the enteric nervous system [10], and plays a key role in extracting and processing nutrients consumed in the diet [11]. The microflora can metabolize proteins and protein degradation products, sulfur-containing compounds, and endogenous and exogenous glycoproteins [12]. Some organisms grow on intermediate products of fermentation such as H_2, lactate, succinate, formate, and ethanol; converting these to end products including short chain fatty acids, a process which has direct benefits on digestive physiology [12].

The fragile composition of the gut microflora can be affected by various factors such as age, diet, environment, stress and medication [13]. Hence, global potential for poultry acidifiers, for both feed and water application are on the rise due to higher demand of top quality poultry products [14]. Organic acids (OA) based feed acidifiers have gained significance due to their high nutritional value and antimicrobial benefits [15,16]. Most of the research and subsequent applications have involved feed acidifiers as a preventive or treatment tool to control the presentation of enteric diseases or to improve poultry performance and welfare. The inclusion of various OA or their salts in animal diets has shown to improve growth performance by enhancing nutrient digestibility and affecting microbial populations in different parts of the digestive tract [15]. Using an OA blend in the drinking water at critical periods of poultry growth has been proved to be a viable practice to establish and maintain intestinal development by promoting stability of the intestinal microflora and eventually improving live production performance [17,18]. Previously, we have showed that a commercially available water treatment product (Optimizer™) significantly reduced carcass condemnation at the processing plant, mortality during transportation, and body weigh lost, suggesting that this OA product may improve animal welfare and economic concerns in the poultry industry [14,19,20]. Additionally, OA also have shown to have a profound impact on the intestinal microbiome [21]. The gut is colonized by different types of microorganisms that have dynamic and diverse symbiotic relationships providing multiple functions, which have a direct impact on the digestive physiology and the biology of metazoans [22]. The capacity to ferment complex polysaccharides to short chain fatty acids by intestinal microflora has a profound effect on energy homeostasis [11]. In exchange, their hosts have organs that enable microbial fermentation of non-digestible food stuff [23], revealing a symbiotic evolution over time, as indicated by concurrent phylogenetic trees [24]. Although the mechanisms by which the intestinal microflora assert these effects on the gastrointestinal tract remain essentially unknown, research in this area is focusing on elucidating these mechanisms as well as manipulating the bacteria and the gastrointestinal environment towards achieving optimal health through probiotics and prebiotics [25]. Utilization of selected probiotics may improve the metabolism of the host animals in various ways, including absorptive capacity, protein metabolism, energy metabolism, fiber digestion, and gut maturation [26,27]. Balanced gastrointestinal microflora and immune-stimulation are major functional effects attributed to the consumption of probiotics [28-30]. Many probiotic effects are mediated through immune regulation, particularly through balance control of pro-inflammatory and anti-inflammatory cytokines [29]. Conversely, several animal and human studies have provided unequivocal evidence that specific strains of probiotics are able to stimulate multiple aspects of innate immunity [31,32] as well as to increase humoral immunity [33]. During the last 15 years, our laboratories have worked toward the identification of probiotic candidates for use in poultry. FloraMax® B11 is a defined lactic acid bacteria (LAB)-based probiotic culture that has demonstrated accelerated development of normal microflora in chickens and turkeys, providing increased resistance to *Salmonella* spp. infections, improvement of performance parameters and production costs [17,30,34-37]. However, this is the first report of the utilization of both antibiotic growth promoter candidate alternatives in broiler breeder's production. Therefore, the objective of the present study was to evaluate the combination of both commercially available probiotic and organic acid products on production, performance and economic parameters in broiler breeders under commercial conditions.

Materials and Methods

Probiotic culture

FloraMax® B11 (Pacific Vet Group USA Inc., Fayetteville, AR 72703, USA) is a defined probiotic culture derived from gastrointestinal poultry origin, consisting of 2 strains of lactic acid bacteria isolates: *Lactobacillus salivarius* and *Pediococcus parvulus* that were selected based on their *in vitro* ability to inhibit enteropathogens [38]. Microbial identification has been previously confirmed by 16S rRNA sequence analyses (Microbial ID Inc., Newark, DE 19713, USA) [37]. This product was administered according to manufacturer's instructions. The final concentration delivered once diluted in the drinking water was 10^6 cfu/mL.

Organic acids

An OA product (Optimizer™, Pacific Vet Group-USA, Inc., Fayetteville, AR, 72703, USA) was used in the drinking water according to manufacturer's directions (4 L Optimizer™/1,000 L of water). This commercial OA product is a combination of five different OA (lactic, acetic, tannic, propionic, and caprylic acids) that contains proprietary flavoring agents. This OA product has been shown to reduce *Salmonella* colonization in crop and cecal tonsils without affecting water consumption in chickens [19,34,39].

Animals, housing and variables evaluated

The effects of supplementing the blend of OA and probiotic on performance and production parameters of broiler breeders were examined in this study. Nine thousand female and one thousand male Cobb 500 breeders were used in three independent trials, during three production cycles in the years 2013, 2014 and 2015. The trials were conducted in the same breeder house from an integrated commercial poultry company in the Province of Santa Fe, Argentina. Brooding management, maintenance and production phases, as well as feeding and lighting programs were conducted according to Cobb Breeder Management Guide. Birds were transfer to the production house at 22 weeks of age. The chicken house had a dimension of 140 meters long by 14 meters width, providing a density of 5 breeders per square meter, and it was divided in a longitude way in two equal compartments. The division separated treated groups while having them in the same environmental conditions. The chicken house possessed tunnel ventilation and spray system. Each section was equipped with automatic feeders and two lines of water with nipple and bell drinkers to administer water treatments. Feeders provided a minimum of 15 cm of feeding space per female for chain feeders and 12 females per pan to ensure that feed could be distributed in less than 3 minutes. Nipple drinkers were installed at the rate of 6 to 8 birds/nipple. Bell drinkers were installed at the rate of 60 birds per drinker. Each section was equipped with communal mechanical nests at a rate of 50 birds/m of nest floor area, allowing six birds per nest hole in single bird rollaway nests. During the 10 weeks of each trial, daily total mortality, culls, feed intake, total egg number, egg weight, hatching egg number, floor eggs and fertility were recorded. One hundred eggs were weighted immediately following the mid-morning collection, excluding only double-yolk and cracked eggs. Body weight and uniformity were evaluated weekly. Livability was recorded as a percentage of live birds daily, and the feed allowance was adjusted accordingly. Egg production performance was expressed as a percentage of hen-day egg production and recorded daily for each experimental sector. One hundred randomly sampled settable eggs for each treatment were collected. Eggs that were not dirty, cracked, broken, excessively small or large, or double-yolked were accepted as settable eggs. The settable egg rate and the extra large egg rate, which includes double-yolked eggs, were expressed as percentages of the total number of eggs laid per day and recorded daily. All animal handling procedures were in compliance with the Argentine Broiler Breeder company ENERCO, Unión Agrícola de Avellaneda Coop. Ltda., S3561AKR , Santa Fe, Argentina.

Experimental design

In each trial, every week during 10 consecutive weeks, starting at 25 weeks of age to 35 weeks of age, 10,000 birds were divided in two groups of 5,000 birds (4,500 females and 500 males in each group): control group, birds receiving only regular water and treatment group, birds receiving OA in the water at a concentration of 4 L/1,000 L of water (vol/vol) according to the manufacturer's directions. Twenty-four hours after OA administration, probiotic was administered in the drinking water according to the manufacturer's direction.

Formulas and estimated values

Money values were obtained in Argentinian pesos and changed to American dollars according to the dollar change rate of the Central Bank of Argentina during December 1st of each year. To obtain the CBR, the total value in USD of hatched chicks were considered, and the difference between treated and control groups was compared. The cost of each broiler chick in Argentina is equivalent to $0.48 USD.

Value of hatched chicks=Total number of hatched chicks × estimated at $0.48 USD/chicken

Difference in chick cost=Value of hatched chicks control – Value of hatched chicks treated

Total cost of Optimizer™=(cost of Optimizer™/L) × (L of Optimizer™ used)

Total cost of FloraMax B11®=(cost of FloraMax B11® × 10 envelopes, one for each week of treatment)

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Cost benefit ratio=Difference in chick cost/Optimizer™ and FloraMax B11® treatment cost

Cost of products per year

The costs of products per year are shown in table 1.

Data and statistical analysis

All data were subjected to ANOVA as a completely randomized design, using the GLM procedure of SAS [40]. Significant differences among the means were determined by Duncan's multiple-range test at $P<0.05$.

Results, Discussion and Conclusion

Today, the fields of immunology, microbiology, and nutrition converge in an astonishing way [41]. Balanced gastrointestinal microflora and immune-stimulation are major functional effects attributed to beneficial bacteria [42]. In this context, a short window of time during birth exists that enables the colonization of symbiotic bacteria to all mucosal surfaces, which may modify the future immune phenotype of the host [43]. Perhaps, a delayed microbial colonization of the gut mucosa, the largest immune organ of the body, could cause significant changes in the immune system possibly having long term impacts on systemic immunity [44]. For instance, some effects of the microbiome are mediated through immune regulation, particularly through balanced control of pro-inflammatory and anti-inflammatory cytokines [29,45]. Moreover, several animal and human studies have provided unequivocal evidence that specific bacterial strains are capable of stimulating multiple aspects of innate immunity [46] as well as to increase humoral immunity [47]. Very interestingly, through a process of "cross talk" with the mucosal immune system, the microbiota negotiates mutual growth, survival, and inflammatory control of the intestinal ecosystem and pathogen control [48]. On the other hand, concern about antimicrobial resistance has led to increased attention to alternatives for controlling infections and increasing performance in animal production.

Cost	2013	2014	2015
Optimizer™	US $8.12/L	US $7.87/L	US $7.08/L
FloraMax B11®	US $19.48/L	US $22.81/L	US $27.69/L

Table 1: Cost of products per year

Probiotics and organic acids have gained attention as options in poultry industry. Our laboratory has been working in the selection of lactic acid bacteria, mainly from the genus *Lactobacillus*, as potential probiotic candidates. Previous data indicates that these selected probiotic bacteria are able to reduce *Salmonella* infection and improve performance in broiler and turkey under experimental and commercial trials in the USA. The selected probiotic organisms were used in field trials to evaluate their efficacy in commercial conditions. Probiotic supplementation to broiler breeders has been shown to stimulate the release of reproductive and metabolic hormones and improve digestion and absorption of nutrients [49]. Previous results published in our laboratory have shown that the administration of selected probiotic candidate bacteria in combination with OA may reduce environmental *Salmonella* in turkey houses prior to live haul, and that this practice could help to reduce the risk of *Salmonella* cross contamination in the processing plant and reduced weight lost during transportation to the processing plant [14,17-20]. In the present study, the use of an OA blend followed by the administration of a probiotic for ten consecutive weeks, from week 25 to week 35, on growth performance parameters are summarized in table 2. No significant differences ($P>0.05$) on body weight and uniformity in females and males were observed between treated or control groups in any of the three trials (Table 2). Table 3 shows the results of the broiler breeder production parameters evaluated with or without supplementation of organic acids and probiotic. A significant reduction ($P<0.05$) in the percentage of deformed eggs, and weekly-cumulative mortality were observed in all three trials in the birds that received every week (25 to 35 weeks) the blend of OA and probiotic when compared with control non treated birds (Table 3). The results of the evaluation of the CBR of organic acids and probiotic in broiler breeder production parameters are summarized in the table 4. During the first trial in 2013, there was a CBR of only $0.75 USD, represented by 474 extra chicks, hatched in favor of the treated group when compared with control non-treated group. However, in the following trials in 2014 and 2015, birds that received once a week during 10 consecutive weeks, the blend of OA followed by the probiotic, had 5,465 and 5,629 extra hatched chicks when compared with control birds. This difference in the number of hatched chicks resulted in a CBF of $4.41 and $4.40 respectively in favor of the treated breeders (Table 4). In conclusion, the CBR of all three

Performance parameters	Trial 1 (2013)		Trial 2 (2014)		Trial 3 (2015)	
	Control	OA+Probiotic	Control	OA + Probiotic	Control	OA+Probiotic
Female body weight (g)	3,597.82 ± 62.22	3,669.64 ± 53.17	3,663.68 ± 60.70	3,645.32 ± 63.64	3,683.64 ± 101.18	3,666.00 ± 94.25
Female uniformity (%)	84.18 ± 1.43	82.09 ± 0.56	85.00 ± 1.16	82.00 ± 1.51	81.59 ± 1.10	83.86 ± 1.24
Male body weight (g)	4,251.45 ± 46.54	4,139.27 ± 36.55	4,028.55 ± 69.45	4,146.05 ± 88.70	4,327.05 ± 74.60	4,370.77 ± 71.98
Male uniformity (%)	70.09 ± 1.77	71.73 ± 2.27	81.55 ± 2.00	83.18 ± 1.63	82.64 ± 2.07	83.05 ± 1.27

Table 2: Broiler breeder growth performance parameters evaluated with or without supplementation of organic acids (OA) and probiotic[1]
[1]Data expressed as mean ± standard error

Performance parameters	Trial 1		Trial 2		Trial 3	
	Control	OA+Probiotic	Control	OA+Probiotic	Control	OA+Probiotic
Total eggs/hens housed (week 35)	56.9 ± 0.28	57.12 ± 0.28	54.68 ± 0.28	54.87 ± 0.28	53.9 ± 0.28	55.28 ± 0.28
Total eggs/hens housed (weekly)	5.07 ± 0.39	5.10 ± 0.40	4.91 ± 0.53	4.95 ± 0.53	4.85 ± 0.58	4.98 ± 0.57
Peak egg production (%)	84.28 ± 0.33	85.88 ± 0.33	85.76 ± 0.33	86.66 ± 0.33	90.06 ± 0.33	90.03 ± 0.33
Age at peak egg production (days)	214 ± 3.97	219 ± 3.97	197 ± 3.97	208 ± 3.97	217 ± 3.97	209 ± 3.97
Egg production (%)	72.33 ± 5.55	72.71 ± 5.75	69.99 ± 7.49	70.58 ± 7.59	69.21 ± 8.30	71.09 ± 8.16
Hatching eggs (%)	83.16 ± 2.40	82.19 ± 2.49	84.87 ± 2.51	85.42 ± 2.33	82.99 ± 3.70	83.23 ± 3.45
Dirty eggs (%)	0.88 ± 0.09	0.78 ± 0.08	1.05 ± 0.13	1.17 ± 0.18	0.39 ± 0.05	0.36 ± 0.05
Double yolk eggs (%)	1.83 ± 0.38	1.61 ± 0.33	1.32 ± 0.30	1.44 ± 0.31	1.53 ± 0.32	1.52 ± 0.29
Deformed eggs (%)	2.39 ± 0.31	2.10 ± 0.31*	2.42 ± 0.37	2.18 ± 0.35*	1.24 ± 0.13	1.09 ± 0.13*
Egg weight (g)	61.50 ± 1.52	60.61 ± 1.46	61.12 ± 1.43	61.10 ± 1.46	58.25 ± 1.90	58.57 ± 2.07
Density (%)	93.77 ± 0.86	92.81 ± 1.99	83.42 ± 1.92	85.47 ± 2.90	95.36 ± 0.91	95.45 ± 1.03
Uniformity (%)	82.09 ± 0.56	84.18 ± 1.43	82.00 ± 1.51	85.00 ± 1.16	83.86 ± 1.24	81.59 ± 1.10
Mortality (week %)	0.45 ± 0.07	0.39 ± 0.05*	0.29 ± 0.03	0.22 ± 0.03*	0.22 ± 0.02	0.16 ± 0.02*
Cumulative mortality (%)	5.00 ± 0.02	4.34 ± 0.02*	3.12 ± 0.02	2.43 ± 0.02*	2.38 ± 0.02	1.79 ±02*

Table 3: Broiler breeder production parameters evaluated with or without supplementation of organic acids (OA) and probiotic[1]
[1]Data expressed as mean ± standard error
*Mean values significantly differ between experimental treatments ($P<0.05$).

Item	Trial 1 (2013)		Trial 2 (2014)		Trial 3 (2015)	
	Control	OA+Probiotic	Control	OA+Probiotic	Control	OA+Probiotic
Hatching eggs	251,990	252,547	255,117	261,547	253,192	259,815
Hatched chicks	214,191	214,665	216,849	222,315	215,213	220,842
Difference in hatched broiler chicks	474		5,465		5,629	
Value of hatched chicks ($0.48 USD/chick)	$102,811.68	$103,039.92	$104,087.52	$106,711.12	$103,302.24	$106,004.16
Difference in chick cost	$227.52		$2,623.68		$2,701.92	
OA+Probiotic treatment cost*	$303.57		$597.22		$616.00	
Cost benefit ratio	$0.75		$4.41		$4.40	
Average cost benefit ratio of all trials			**$3.19**			

Table 4: Evaluation of the cost benefit ratio of organic acids (OA) and probiotic in broiler breeder production parameters
*Cost benefit ratio=Difference in chick cost/Optimizer™ and FloraMax B11® treatment cost

trials suggest that for every U.S. dollar spent on the program of OA plus probiotic products, producers may be able to recover on average 3.19 U.S. dollars. These data suggest that the combination of OA and probiotic used in the present study may improve economics by increasing the number of hatching chicks from broilers breeders supplemented with these additives.

Conflict of Interest Statement

The authors declare that the research was con-ducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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