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Evaluation of some organic acids alternative to antibiotics for control of salmonella infection in broiler chickens

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Abstract

This study was conducted to compare the effects of two types of organic acids (caprylic acid and propionic acid) for control of salmonella infection in broiler chicks. A total of 165, one day-old (Cobb 500) broiler chicks were used. At day old, five chicks were sacrificed and examined bacteriologically to prove their freedom from *S.* Enteritidis infection. One hundred and sixty birds were divided into eight equal groups. Chickens in (G1) non infected-non treated birds, (G2) wasinfected non treated birds, (G3) was infected and treated with caprylic acid1%,(G4) non infected and treated with caprylic1%,(G5)infected and treated with propionic acid1.2%, (G6) non infected and treated with propionic acid 1.2%,(G7) infected and treated with caprylic1% and propionic acid1.2%,(G8) non infected and treated with caprylic1% and propionic acid1.2%. All birds in (G2, G3 G5, and G7) were challenged 10^5CFU/ml *S.*Enteritidis at 7days of age. All the groups were kept under complete observation for four weeks for recordingsigns, moralities, gross lesions, shedding rate of *S.* Enteritidis, the performance .Five birds from each group were euthanized on days 21and 35day of age and examined bacteriologically for re-isolation of *Salmonella* Enteritidis from cecum and crop (quantitative and qualitative), liver, spleen were tested qualitative .pH in crop and cecum were measured.Plasma samples were collected from the portal vein to determine medium chain fatty acids and short chain fatty acids. Results indicated that treatment with organic acids decreased reisolation of *Salmonella* Enteritidis from different organs, reduced colonization of*Salmonella* Enteritidis in the crop and cecumand fecal shedding. Birds supplemented with organic acidsshowed significantly ($P\leq0.05$) higher body weight, body weight gains and lower feed conversion ratio compared to control group. Chicks treated with acids had an increase MCFA (caprylic acid) and decrease in SCFA (acetic acid and propionic acid) in portal bloodthan the control group.

The present study was able to show that this organic acids were useful in controlling of *Salmonella* Enteritidis in infected chicks and this procedure can be important as part of a *Salmonella* control program.

Key words: Salmonella Enteritidis, organic acid, caprylicacid, propionic acid, growthperformance, broiler

Introduction

Salmonella is apathogenic organism and it is not a part of intestinal flora .It cause disease manifested by irritation of the intestinal wall and decrease villi number and length which in turn impair nutrient absorption **(Pelicano et al.,2005).** Furthermore, it may compete with their host for nutrients and produce substances such as ammonia or amines,which damage liver **(Krinke and Jamroz ,1996 *)***. *Salmonella entericserovarEnteritidis* is the most common serotype isolated **(Antunes et al.,2003)** . Despite progress in food safety through pathogen reduction programs,*Salmonella Entreritidis* remains one of the most common foodborne pathogens transmitted through consumption of poultry products.*Salmonella Enteritidis* colonizes various parts of the intestinal tract ,with the cecum being the most common site **(Stern,2008).**Cecal colonization eventually leads to fecal shedding , contamination of eggshells with infected feces ,carcass contamination during slaughter **(Gantois et al.,2009)**,so the some reduction in the number of *Salmonella Enteritidis* cells shed in feces could help control the spread of infection among the farms. Increased awareness of the potential problems associated with use of antibiotics has stimulated research efforts to identify alternatives to their use.Alternative strategies (including organic acids and their salts,probiotic, prebiotic and enzymes) were developed to cope with the removal of antibiotics as growth promoters .In recent years ,there is increasing trend to organic acids and mixtures ,as alternative to antibiotic growth promoters due to their inhibiting activity on the growth and development of pathogens in animal feed and gastrointestinal tract.**(Saki et al. ,2012)** reported that supplementation of organic acids in the diet increased LAB counts in the ileum and caecum of broiler chicken. This treatment also significantly decreased *Enterobacteriaceae* and *Salmonella* counts in the intestine of birds **(Cengiz et al., 2012)**. In terms of performance, feeding organic acids resulted in improved body weight gains and feed conversion ratio **(Adil et al., 2010)**.Their bacteriostatic activity is thought to drive from the penetration into the bacterial cell and their dissociation into anions and protons. The protons are responsible for the acidification of the cytoplasm ,interfering thus with most cellular function ,where as the anions may inhibit DNA syntheses to varying dgrees depending on the specific compound used **(Cherrington et al., 1991)**. It has been proposed that some of their activity drives from the creation of acidic environment in crop of the chicken,reducing the uptake of*Salmonella* **(Ricke, 2005)**.In addition, by modifying intestinal PH, organic acids also improve the solubility of the feed ingredients, digestion and absorption of the nutrient **(Skinner et al., 1991)**.

Many studies were concerned with the use of organic acids for control *salmonella* in poultry productions. Short chain fatty acids (SCFA), such as formic, acetic, propionic and butyric acids, have all been shown to have an inhibitory effect on *salmonella*growth **(Van Immerseel et al., 2006**). Propionic acid significantly decreases crop and cecal colonization by salmonella gallinarum**(Alshawabkenand Tabbaa**

2002). (Kwon et al.2003) reported that buffered propionic acid (BPA) markedly decrease the growth of *salmonella* and other intestinal anaerobic microbes by decreasing PH from 7 to 5 and maximum inhibitory effect was found at3% level of BPA in broilers .

Medium chain fatty acids(MCFA) (caproic,caprylic,capric and lauric) appear to be much more effective against *salmonella* than the short chain fatty acids**(Van Immerseel 2007**) .MCFA supplementation was equally effective with organic acids in decreasing the levels of colonization in ceca and improved growth performance .MCFA increased cecal SCFA concentrations and MCFA in portal blood .Therefore ,MCFA is one of the efficient additives appropriate for salmonella control in broilers **(Chotikatum et al.,2009**) .Direct acidification of the water with organic acids could significantly reduce the amount of recoverable salmonella on the carcass or in the crop and cecal tonsils when used during pre slaughter period **(Van Immerseel et al., 2006 ;Alali et al.,2010 ; Vandeplas et al.,2010) .(Sprong et al., 2001)** reported that caprylic and capric acids were bactericidal, but C14:0, C18:1 and C18:2 acids were not.Caprylic acid (CA) naturally present in breast milk, caprine milk, and coconut oil, is a Generally Recognized as Safe (GRAS) molecule approved by the FDA. In experiments with bacteria belonging to the *Enterobacteriaceae*, caprylic acid was more active against *Escherichia coli* and *Salmonella* than fatty acids with shorter or longer chain lengths **(Marounek*et al,*.2003; Skřivanova*et al.*,2004).** CA was effective in reducingcecal*Salmonella Enteritidis*populations in cecum, small intestine, cloaca, liver, and spleen and potentially reduces the pathogens ability to invade intestinal epithelial cells by downregulating key invasion genes,hilA and hilD**(Johny et al.,2009 andJohny et al.,2012). (Menoniet al., 2013**) reported that using organic acid mixture showed significant reduction in total number of *Salmonella Typhimurium*cecal positive tonsils, and reducing in its number in crop contents.

The objectives of this experiment were to evaluate the effect of both caprylic acid and propionic acid on eliminating *S. Enteritidis* colonization and shedding in broiler chickens. Moreover, the effect of these additives on growth performance, crop and cecal pH, level of volatile fatty acids in portal blood.

Material and methods

Salmonella strain: Salmonella*Enteritidis*resistance to novobiocin-nalidixic acid (NO 25 ug/ ml, /NA 20 ug /ml) was supplied by Animal Health Research Institute,Dokki, Giza, Egypt.

Organic acids: Caprylic acid and propionic acid (Sigma, aldrich)

Chickens: One hundred sixty five, one- day-old Cobb 500 broiler chicks were obtained from a commercial hatchery.At arrival, examined bacteriologically to prove their freedom from *S. Enteritidis* infection. Chicks were divided into 8 groups each group contain 20 chicks .Chicks were floor-reared and fed a commercial diet ,with consistant lighting at a mean temperature of 32º C with supplemental heat supply and relative humidity of 45%. Diet and water were cultured for the presence of Salmonella using a standred culture methods **(Andrew et l., 1978).**

Experimental design:

1st groupG1: Controlgroup,non infected and non treated.

2ndgroup G2: Infected with 10^5cfu of *Salmonella enteritidis*orally at7th day of age.

3rd groupG3: Infected with 10^5cfu of *Salmonella enteritidis*orally at7th day of age and treated with caprylic acid 1% in drinking water and continued until the 35th day of age consecutively .

4thgroupG4: Non infected with *salmonella enteritidis* and treated with caprylic acid 1%drinking water until35 thday of age consecutively .

5thgroupG5: Infected with 10^5cfu of *Salmonella enteritidis*orally at 7th day of age and treated with propionic acid 1.2% in drinking water until35th day of age consecutively.

6thgroup G6:Non infected with *Salmonella enteritidis* and treated with propionic acid 1.2% in drinking water until 35th day of age consecutively .

7thgroup G7: Infected with 10^5cfu of *Salmonella enteritidis*orally at 7th day of age and treated with caprylic acid 1% and propionic acid1.2% in drinking water until 35th day of age consecutively.

8thgroup G8:Non infected with *Salmonella enteritidis* and treated with caprylic acid 1% and propionic acid 1.2% in drinking water until35 th day of age consecutively .

Parameter measured: Clinical signs and mortalities were recorded.Clocal swabs were collectedfrom five chickens weekly after the challenge up to 5 weeks of age, examined bacteriologically (qualitative) for thepresence of *S. Enteritidis* organism . At 21 and 35 day of age five birds from each group were scarified, autopsied, and examined bacteriologically for re-isolation of SE.from caecum and crop(quantitative and qualitative) Liver, spleen were tested qualitative.The growth performance of broiler chickens was evaluated by recording body weight, body weight gain and feed conversion ratio .Weighing of the feed and chickens were made on a weekly basis.Cecal and crop PH were measured at 21 and 35 days of age .At 35 and21 day of age five serum samples were collected from each group for volatile fatty acid analysis.

Bacteriological Analysis: .1 g of crop and cecal contentswas used to make serial 10-fold dilution with BPW and spread on XLD agar plates (XLD) contain 25 µgnovobiocin/ml and 20 µgnalidixic. An additional 1 mL of the original solution and swabs from liver, spleen and cloaca were placed into 9 mL of TET broth for enrichment.The XLD agar plates and TET tubes were incubated for 18 to 24 h at 37°. The number of colony forming units was expressed colony- units per gram. Suspected colonies on XLDplates were confirmed biochemically on triple sugar iron agar and lysine iron agar **(Andrews *et al*., 1992).**

Volatile fatty acid analyses and determination: the collected serum samples were analysed to measurements of VFAby (**HLPC)** accordingto **(weast, 1971).**

Determination of crop and cecal pH:

Crop pH was measured *in situ* by inserting on electrode into an incision in the crop beforeremoving its contents. The pH of ceca were measured by collection of 0.2 g of cecal contents from each chick, suspended in 1.8 ml of sterile glass-distilled water and pH was measured immediately with glass electrode **(Nisbet et al., 1993).**

Results and discussion

Organic acids are widely distributed in nature as normal constituents of plants or animal tissues. They are also products of microbial fermentation of carbohydrates especially in the caeca of birds. A wide range of organic acids with variable physical and chemical properties are available for poultry, of which many are used in the drinking water or mixed with the feed (**Huyghebaert et al., 2011).** In the market, organic acids can be found in the form of single or in combination **(Menconi et al., 2014).**The supplementation of organic acids to poultry diets was shown to suppress the growth of certain species of bacteria, mainly acid-intolerant species, such as *Salmonella*, *E. coli***. (Van Immerseel et al., 2002)**. In chickens, the cecum is a major colonization site for *Salmonella* Enteritidis, and the pathogen usually is present in large numbers ***(*Cerquettiand Gheradi 2000; Van Immerseel *et al*., *2004).*** *Salmonella* Enteritidis also colonizes the small intestine **(Khan et al.,2003; Li et al.,2003***)* and cloaca ***(*Van Immerseel *et al*., *2004),***through which the pathogen is horizontally transmitted. In addition to these sites, *Salmonella* Enteritidis also has been recovered from the crop, although in lower numbers ***(*Avila et al.,2003;Durant et al.,1999).**The pathogen reaches the liver and spleen by lymphatic or circulatory systems ***(*Cerquetti and Gheradi2000;Van Immerseel *et al*., *2004).*** This study focused on the efficacy of different types of organic acids for reducing *Salmonella* Enteritidis populations in all of these organs and their effect on growth performance parameters. Characteristics of organic acids such as chain length, side chain composition, pkA values and hydrophobicity could be factors that effect biocidal activity **(Van Immerseel *et al*., 2006).** For these purpose, a mixture of organic acids was tested to reduce Salmonella **.** Diarrhea, depression, ruffled feather, sunken eyes and decreased feed intake were observed in all infected groups from the 3rd day PI. Diarrhea was severe in G2, followed by G5 and G3 while G7showed less signs. Dead birds at 1st week after infection showed sever congestion in the carcasses, liver and heart muscles. Intestinal blood vessels were engorged with blood, ballooning in intestine, abdominal air–sacculitis were also seen. Liver severely congested, at 2nd week white chalky diarrhea in large intestine, enlarged gall bladder was very clear especially in G2 thickening in wall of intestine mainly duodenum with clear pancreatitis, Caecal core were mainly manifested in groups G2,G3 and G5. By the 3rd week post infection clinical signs and Post mortem examination were minimized in comparison with 1st and 2nd weeks except G2. Mortality of chickens were observed and detected in percentages of 20 %, 10%, 10% and 5% in G2,G3, G5andG7 respectively(table1). These results decleared that oral administration of organic acids reduced clinical symptoms, anatomicopathological changes in the digestive tract of infected birds and mortalities.Our results were confirmed by previous reports of (**Ezzeldeen and Zouelfakar 2003; Ellakany et al. 2004; Zohair 2006, Franiszek et al., 2013 and Hamed and Hassan 2013)** reported that treated birds with acidifier could minimize both symptoms and postmortem lesions and reduced mortalitis. Results tabulated in table (2) showed that the rate of reisolation of salmonella from different organs were decreased in treated groups(G3,G5 andG7) than non treated group(G2).The lowest rate of re isolation were (2.5%), (7.5%) and (12.5%) inG7, G3 and G5 respectively.**(Van Immerseel et al. , 2003andVan Immerseel et al. , 2004)**reported that C6 to C10 acids were bacteriostatic to a Salmonella Enteritidis**.(Sprong et al.,2001)** reported that caprylic and capricacids were bactericidal.Caprylic acid at 0.7 or 1% decreased *Salmonella* Enteritidis populations in cecum, small intestine, cloaca, liver and spleen**(Johny et al.,2012).(Aishawabkeh and Tabbaa 2002**) who reported that propionic acid is likely to be antibacterial effect on *Salmonella gallinarum* both in the crop as well as the ceca . The results of colonization of crop and cecal of broilers at day 21 and day 35 of the experiment in table (3)showed that there were significant difference between infected non treated group and infected treated groups. Among the treated groups there were significant reduction ($P \leq 0.05$) in crop and cecal bacterial count between the medicated groups. The highest reduction were in G7 followed by G3 and G5 respectively.At 35 day treatment could eliminate colonization in crop in all treated groups and in cecum of G7 and G3.Our results clearly showed that organic acid supplementation controlled the crop and cecal colonization of *Salmonella* .Several other studies suggested that organic acid supplementation to poultry diets acted as a bactericidal for *Salmonella* in the crop ,cecum ,small intestine and ileum **(Thompson and Hinton, 1997,Parker et al.,2011,Cengiz et al.,2012,Menconi et al., 2013andAbudabos et al.,2014).**We noticed that acids has more effect on salmonella in crop than cecum and this resultsagree with**(Skřivanová, et al.,2014)**. The results of the fecal shedding rate of *S.* Enteritidis in broiler chickens after infection with *Salmonella* entritidisand treatment with acids in (table4) declared that there were differences between the infected treated groups and the infected non treated one along four weeks observation period. Gradual decrease in the shedding rate was observed within each group until reached the last week of observation period. The rate of fecal shedding was (10%, 15% and 20%) forG7,G3 and G5 respectively. Some explanations for the SE shedding in these groups may reside firstly in the ability of *Salmonella* to be affected withthe environmental conditions and some specific conditions that are required to promote adhesion and invasion; secondly, it could be due the capacity of organic acids to specifically alter the expression of the *HilA*gene **(Van Immerseel *et al*., 2003; Van Immerseel *et al*., 2004, Borsoi et al., 2011).** Our results showed that mixed acids more effective than single acids**.** It has been suggested that combinations of organic acids are more effective than supplements that contain only one type of acid. This is because different types of organic acid diffuse through the bacterial cell wall and membrane and into the cell cytoplasm at different rates. These acids dissociate to form a conjugate base and a free hydrogen ion at different rates and respective pKa values **(Novus International Inc. 2006).** Also there were difference between the effect of propionic acid and caprylic acids and this results agree with (**Nakai and Siebrt,2002)** who reported that the antibacterial activity of the MCFA appears higher than that of the activity of the SCFA (formic, acetic, propionic, and butyric acid) against both gram-positive and gram-negative bacteria, as well as antimicrobial activity against *Salmonella*, even at low concentrations.

The pH of crop, and caecum reduced significantly (P≤0.05) in all treatments compared with control group at day 21and day 35of the experiment as shown in table (5) this indicated that the pH decreased by acid supplementation, and this lower pH inhibited the growth of *Salmonella* in the crop before degradation or absorption occurred in the intestine. The organic acids resulted in production of sufficient undissociated acid molecules which remained in contact with bacteria for enough time, and this undissociated acid diffused into the bacterial cells caused a lower pH inside the cell **(Hume *et al*., 1993)** These results were in harmony with the result of the(**Al-Tarazi and Alshawabkeh 2003**) who reported that adding 0.5-1.5% FA to broiler diet reduced significantly crop and cecalpH. However, **Hernadez *et al*. (2006) and Al-Natour and Alshawabkeh (2005**) found insignificant reduction in the intestinal pH for broiler when used 0.5-1.5% FA**.**

The results of SACF volatile fatty acids(acetic acid and propionic acid) analysis in portal blood in table (6) showed that organic acid treatment cause reduction in its level in infected treated groups as compared with control. There were high significant difference (P≤0.05) within infected and treated groups .This was refered to the infection cause stress so liver could not change this acids to their products, while liver of treated groups could change these acids to other metabolites. The results of MCFA showed increase level of it in portal blood of infected and treated groups .This results are agree with results of***(Chotikatum* et al., 2009)**who reported that supplementation of medium chain fatty acids had increased medium chain fatty acid concentrations in the portalblood of broiler chicks infected with salmonella enteritidis and treated with organic acids.

The results in Table (7) showed that. the average of live body weight differed significantly (P≤0.05) between infected treated groups and non infected treated groups. Among infected groups, the infected treated groups (G3, G5&G7) showed significantly (P≤0.05) higher body weight than infected groups (G2). The highest live body weight (1560) was recorded for group G7 followed by G3 (1512) andG5 (1462).Between non infecteted treated groups, the body weight differed significantly (P≤0.05) between treatments and the maximum improvement in body weight was achieved in groups (G8&G4) supplemented with mixed acids and caprylic acid followed by (G6) supplemented with propionic acid. Table (8) showed that there was significance difference ($P \leq 0.05$) between body weight gain and food conversion ratio (FCR) between infected treated groups and non infected treated groups. The highest body weight gain and the lowest food conversion ratio were recorded for (G7&G3) in infected treated groups and for (G8&G4) in non infected treated groups. Our findings correspond to(**Marcq *et al.* 2011and Vandeplas *et al.*,2009)** who reported a decline in growth performance in broilers that were challenged with *Salmonella* and this could decrease BWG from14 to33%and increase FCR by 5 to 20**%(Vandeplas *et al.*,2009,Chalghoumi,et al.,2009).(Wilson *et al*. 2005)**suggested that the growth suppressing effect of intestinal bacteria was due to the production of toxic metabolites that irritate the gut mucosa, thereby inhibiting nutrient absorption.This results indicated that treatment with acidswas able to restore the effect of the bacterial challenge and this agree with(**Abudabos et al., 2014)**. Organic acids, such as lactic, acetic, tannic, fumaric, propionic, caprylic acids, etc., have been shown to exhibit beneficial effects on the intestinal health and performance of birds (**Saki et al., 2012; Menconi et al., 2014).**Feeding organic acids resulted in improved body weight gains and feed conversion ratio **(Adil et al., 2010)**. Inclusion of organic acids seems to have direct effects on the histomorphology of the gut by increasing the height of villus **.**Here in, supplementation of organic acids may facilitate the nutrient absorption and that in turn growth performance in broiler chicken **(Adil et al., 2010).**

The present study was able to show that this organic acids were useful in controlling the colonization and fecal shedding of *Salmonella* Enteritidis frominfected chicks and this procedure can be important as part of a *Salmonella* control program as agrememt withthe results obtained by**. Gornowicz and Dziadek (2002), Wolfenden et al. (2007) Abd El-Hakim et al. (2009) and Hassan et al.,(2010)** who concluded that organic acids could be used in poultry, not only as a growth promoter but also as a meaningful tool of controlling intrinsic pathogenic bacteria (*E. coli* and *Salmonella*).

Table (1) Mortality of broiler chickens in numbers and percentage in different groups after *Salmonella enteritidis* infection and treatment with organic acids

WPI	Tested groups							
	G1	**G2**	**G3**	**G4**	**G5**	**G6**	**G7**	**G8**
1st WPI	0	2	1	0	1	0	0	0
2nd WPI	0	1	1	0	1	0	1	0
3rd WPI	0	1	0	0	0	0	0	0
4th WPI	0	0	0	0	0	0	0	0
Total	0	4/20	2/20	0	2/20	0	1/20	0
%	0	20	10	0	10	0	5	0

WPI: week post infection

N.B Symbls in all tables donets

G1: Non infected non treated group.

G2: Infected non treated group.

G3: Infected and treated with caprylic acid 1%.

G4: Non infected and treated with caprylic acid 1%.

G5: Infected and treated with propionic acid 1.2%.

G6: Non infected and treated with propionic acid 1.2%.

G7: Infected and treated with caprylic acid 1% and propionic acid 1.2%.

G8: Non infected and treated with caprylic acid 1% and propionic acid 1.2%.

Table (2) the rate of re -isolation of *Salmonella enteritidis* from different organs of sacrified chicks after treatment with organic acids

Groups	**Re isolation rate**										
	At the end of 2 nd WPI					**At the end of 4th WPI**					**Total**
	Examined organs										
	Liver	**Spleen**	**Crop**	**Cecum**	**Total**	**Liver**	**Spleen**	**Crop**	**Cecum**	**total**	
G1	0	0	0	0	0	0	0	0	0	0	0
G2	2/5	1/5	1/5	4/5	8/20 40%	1/5	0/5	1/5	2/5	4/20 20%	12/40 30%
G3	0/5	0/5	0/5	2/5	2/20 10%	0/5	0/5	0/5	1/5	1/20 5%	3/40 7.5%
G4	0	0	0	0	0	0	0	0	0	0	0
G5	1/5	0/5	0/5	2/5	3/20 15%	1/5	0/5	0/5	1/5	2/20 10%	5/40 12.5%
G6	0	0	0	0	0	0	0	0	0	0	0
G7	0/5	0/5	0/5	1/5	1/20 5%	0/5	0/5	0/5	0/5	0/20 0%	1/40 2.5%
G8	0	0	0	0	0	0	0	0	0	0	0

Table (3) Cecal and crop colonization of *salmonella enteritids* in broiler chicks infected and treated with organic acid

groups	Cecum		Crop	
	Day21	Day35	Day21	Day 35
	Mean ±SE	Mean ±SE	Mean ±SE	Mean ±SE
G1	0	0	0	0
G2	$1.65*10^{5a}\pm138.71$	$1.48*10^{4}a\pm22.75$	$2.04*10^{3a}\pm6.43$	$1.4*10^{3a}\pm4$
G3	$6.6*10^{3b}\pm42.65$	0^{b}	$2.4*10^{2b}\pm15.36$	0^{b}
G4	0	0	0	0
G5	$1.44*10^{4b}\pm51.31$	$2.6*10^{2b}\pm15.78$	$3.0*10^{2b}\pm36.62$	0^{b}
G6	0	0	0	0
G7	$3.2*10^{3b}\pm37.8222$	0^{b}	$2.0*10^{2b}\pm9.45$	0^{b}
G8	0	0	0	0

A-b = Means with the same letter in each column are not significantly different at P≤0.05.

Table (4) Rate of fecal shedding of *salmonellaenteritids* from different groups after treatment with organic acids

groups	14day	21day	28 day	35 day	Total&%
G1	0	0	0	0	0
G2	4/5	3/5	2/5	1/5	10/20(50%)
G3	2/5	1/5	0/5	0/5	3/20(15%)
G4	0/5	0/5	0/5	0/5	0
G5	2/5	1/5	1/5	0/5	4/20(20%)
G6	0/5	0/5	0/5	0/5	0
G7	2/5	0/5	0/5	0/5	2/20(10%)
G8	0/5	0/5	0/5	0/5	0/20

Table (5) Results of PH of crop and cecum of broiler chicks infected with *salmonella enteritidis* and treated with organic acids.

Groups	G1	G2	G3	G4	G5	G6	G7	G8
Crop								
21day	$5.7\pm o.02^{a}$	6.2 ± 0.02^{b}	5.4 ± 0.02^{a}	5.2 ± 0.01^{c}	5.45 ± 0.0^{c}	5.1 ± 0.01^{d}	5.5 ± 0.013^{c}	5.2 ± 0.01^{d}
35day	5.6 ± 0.02^{b}	6.6 ± 0.025^{a}	5.1 ± 0.021^{c}	5.01 ± 0.01^{c}	5.2 ± 0.01^{c}	4.95 ± 0.01^{d}	5.1 ± 0.01^{c}	4.8 ± 0.009^{d}
Cecum								
21day	6.4 ± 0.015^{b}	6.9 ± 0.016^{a}	6.4 ± 0.013^{b}	6.3 ± 0.013^{b}	6.5 ± 0.014^{b}	6.2±0.012c	6.4 ± 0.014^{b}	6.1 ± 0.01^{c}
35day	6.8±0.016a	7.2 ± 0.017^{a}	6.1 ± 0.014^{b}	5.9 ± 0.012^{c}	6.2 ± 0.013^{b}	6.01 ± 0.01^{b}	6.1 ± 0.01^{b}	5.8 ± 0.01^{c}

A-d = Means with the same letter in each column are not significantly different at P≤0.05

Table (6) Results of volatile fatty acid in portal blood of chickens infected with salmonella entreitidis and treated with organic acids

Groups / Tested organic acids	G1	G2	G3	G4	G5	G6	G7	G8
Acetic acid								
21day	3.472^{a}	12.953^{b}	3.839^{b}	3.724^{b}	6.881^{b}	3.824^{b}	5.088^{b}	3.251^{a}
35day	3.215^{a}	10.051^{b}	3.015^{c}	3.542^{c}	6.213^{b}	3.621^{c}	5.910^{b}	3.125^{a}
Propionic acid								
21day	2.015^{a}	4.013^{c}	3.651^{c}	2.815^{b}	3.512^{c}	2.751^{b}	2.981^{b}	2.351^{a}
35day	2.001^{a}	4.030^{c}	3.214^{c}	2.621^{b}	3.215^{c}	2.521^{b}	2.895a	2.121^{a}
Caprylic acid								
21day	10.10^{a}	8.531^{a}	22.15^{b}	34.21^{c}	25.151^{b}	31.52^{c}	28.12^{b}	35.10^{c}
35day	12.30^{a}	9.512^{a}	23.18^{b}	37.75^{c}	26.181^{b}	33.45^{c}	27.13^{c}	38.16^{c}

A-c = Means with the same letter in each row are not significantly different at P≤0.05

Table (7) Average of body weight of broiler infected with *salmonella enteritids* and treated with organic acids

group	DAY1	DAY7	DAY14	DAY21	DAY28	DAY35
	Mean±SE	**Mean±SE**	**Mean±SE**	**Mean±SE**	**Mean±SE**	**Mean±SE**
G1	40.0^{a}±0.34	130.0^{a}±0.88	332.2^{b}±0.53	695.0^{d}±0.88	1112.0^{d}±0.89	1543.0^{de}±2.94
G2	40.0^{a}±0.37	130.0^{a}±0.70	265.0^{f}±0.88	591.0^{h}±1.19	975.0^{h}±1.05	1384.0^{g}±0.98
G3	39.4^{a}±0.36	130.0^{a}±0.90	293.0^{d}±1.02	636.0^{f}±1.13	1062.0^{f}±1.29	1512.0^{e}±1.2
G4	39.0^{a}±0.37	130.2^{a}±0.89	342.0^{ab}±0.75	742.0^{b}±0.75	1216.0^{b}±0.79	1714.0^{b}±1.34
G5	38.8^{a}±0.47	130.0^{a}±0.67	279.0^{e}±0.98	615.0^{g}±1.39	1034.0^{g}±0.79	1462.0^{f}±1.23
G6	40.0^{a}±0.39	130.0^{a}±0.61	339.4^{ab}±0.61	727.0^{c}±0.75	1163.0^{c}±1.29	1646.0^{c}±0.98
G7	40.0^{a}±0.39	129.6^{a}±0.74	308.0^{c}±1.01	665.0^{e}±0.78	1094.0^{e}±0.78	1560.0^{d}±0.96
G8	40.6^{a}±0.45	129.0^{a}±0.66	349.0^{a}±0.79	761.0^{a}±0.98	1265.0^{a}±1.05	1782.0^{a}±1.16

A-h = Means with the same letter in each column are not significantly different at P≤0.05.

Table (8) the average body weight gain and FCR for broiler chicksinfected with *salmonella entreitidis* and treated with organic acids

group	Body weight gain	FCR
G1	1500[b]	1.57[b]
G2	1344[a]	1.73[d]
G3	1472[b]	1.62[c]
G4	1675[c]	1.52[b]
G5	1423.2[a]	1.66[c]
G6	1600[c]	1.56[b]
G7	1520[b]	1.59[b]
G8	1741.4[d]	1.47[a]

A-d = Means with the same letter in each column are not significantly different at P≤0.05

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