The effects of green tea in the diet of broilers challenged with coccidiosis on their performance, carcass characteristics, intestinal mucosal morphology, blood constituents and ceca microflora

Keyvan Jelveh1 | Behrouz Rasouli1 | Isam T. Kadim2 | Marina Ivanovna Slozhenkina2 | Ivan Fedorovich Gorlov3 | Alireza Seidavi1 | Clive J. C. Phillips4,5

1Department of Animal Science, Rasht Branch, Islamic Azad University, Rasht, Iran
2Department of Biological Sciences and Chemistry, College of Arts and Sciences, University of Nizwa, Birkat Al-Mouz, Nizwa, Oman
3Department of Livestock Production, Volga Region Research Institute of Manufacture and Processing of Meat-and-Milk Production, Volgograd, Russia
4Curtin University Sustainability Policy (CUSP) Institute, Kent St, Bentley, Western Australia
5Estonian University of Life Sciences, Institute of Veterinary Medicine and Animal Sciences, Tartu, Estonia

Abstract

Background: Coccidiosis is an endemic protozoal disease of chickens normally controlled by ionophores. However, coccidiostats are also antibiotics, and evidence of resistance in both coccidia and bacteria may develop and reduce antibacterial activity in humans. This has led to a search for natural coccidiostats, such as green tea.

Objectives: To study the effects of supplementing broilers with various levels and types of green tea, in comparison to use of a conventional coccidiostat or a control, unsupplemented diet.

Methods: A total of 360 male, day-old Ross 308 broilers (days 1–42) were used to evaluate the gut morphology and performance when challenged with coccidiosis and fed varying dietary levels of green tea powder or extract. Treatments were Negative control (NC, unsupplemented control diet); positive control (PC, control diet + commercial coccidiostat); control diets with 0.2, 0.3 or 0.4 g/kg green tea extract (GTE 0.2, 0.3 and 0.4); and control diets with 1, 2 or 3 g/kg green tea powder (GTP 1, 2 and 3).

Results: Compared with NC, PC and all green tea treatments, but particularly GTE 0.4, increased feed intake and growth rate, with the best feed conversion ratio at GTE 0.4. As a proportion of carcass weight, higher inclusion rates increased intestine weight and decreased abdominal fat. The duodenum, jejunum and ileum of birds fed green tea, and particularly GTE 0.4, had longer, wider villi, and shallower crypts. Epithelium thickness was reduced by green tea and PC, compared to NC. Clostridium perfringens and coliform populations decreased in proportion to green tea inclusion rate and decreased in PC.
INTRODUCTION

Poultry production is growing in many regions, which contributes to increased protein requirements for humans (Dieye et al., 2010). However, the expanding poultry sector faces many challenges, including diseases. Antibiotics have widely been used for many decades, both as growth promoters and to prevent diseases (Phillips et al., 2004). Despite improving feed utilisation efficiency, there may be residue problems, and resistant bacteria, when consumed by humans in poultry products may lead to antibiotics being ineffective in poultry and humans (Gould, 2008; Van de Bogaard & Stobberingh, 2000). Therefore, many countries, including the EU, have made the inclusion of antibiotics in poultry feed illegal (Butaye et al., 2000; Catalá-Gregori et al., 2002). There is now a widespread search for alternatives to antibiotics.

Herbal products in poultry diets can enhance performance (Teteh et al., 2013) and reduce mortality (Kumar et al., 2003). Green tea (Camellia sinensis) is a natural, non-toxic product containing many bioactive compounds: alkaloids, polyphenols, polysaccharides, volatile oils, vitamin C and minerals (Karori et al., 2007; Khan, 2014). The leaves contain approximately 92.2% dry matter, 82.4% organic matter, 19.3% crude fibre, 8.7% ether extract, 9.8% ash, 18.1% crude protein, 36.2% nitrogen free extract and 3002 kcal/kg (Abdo et al., 2010). They have therapeutic properties: antimicrobial, antioxidant, immune modulatory (Erener et al., 2011; Ko & Yang, 2008; Nishida et al., 2006), and, most importantly for poultry, anticoccidial (Jang et al., 2007). Cocci
diosis is an endemic protozoal disease of the intestine transmitted between broilers through faeces. It causes diarrhoea unless controlled with an ionophore coccidiostat in the feed, which also controls some bacteria. These antimicrobials are classified as feed additives in the EU, as farmers want to retain use of ionophores for controlling coccidiosis, despite resistance in coccidia, but also resistance to bacitracine and avamycine, both antibiotics used in human medicine [Norwegian Scientific Committee for Food Safety (VKM) 2015]. When birds are fed coccidostats, the risk of exposure to coccidiostat-resistant bacteria is high in people handling manure and medium in people handling carcasses and raw meat. In the United States, coccidostats are classified as antibiotics and therefore not allowed in the rapidly developing antibiotic-free production systems (Swormink, 2019).

Green tea may enhance growth in poultry (Cross et al., 2007; Demir et al., 2003) and in humans, it has many health benefits: reduced obesity and blood pressure, blood glucose control, anti-hypercholesterolaemia (Liao et al., 2001). With these benefits, green tea may enhance production and replace antibiotic growth promoters in broilers (Cao et al., 2005; Kaneko et al., 2001; Khan, 2014). However, the physiological effects are largely unknown; hence, we studied these in broilers supplemented with green tea products, a conventional coccidiostat or no supplement.

MATERIALS AND METHODS

This study was conducted at a commercial poultry farm.

2.1 Birds, diets and experimental groups

A total of 360 male Ross 308 broiler day-old chicks were purchased from a local hatchery and randomly distributed into 8 treatments, with 3 replicates of 15 birds in eight treatments, in a completely randomised design:

- Group 1 (NC) (negative control): no supplement
- Group 2 (PC) (positive control): commercial coccidiostat (Salinomycin; C42H70O11) at 0.5 g/kg (Rooyan Darou Co)
- Group 3 (GTE0.2): PC and 0.2 g/kg green tea extract
- Group 4 (GTE0.3): PC and 0.3 g/kg green tea extract
- Group 5 (GTE0.4): PC and 0.4 g/kg green tea extract
- Group 6 (GTP1): PC and 1 g/kg green tea powder
- Group 7 (GTP2): PC and 2 g/kg green tea powder
- Group 8 (GTP3): PC and 3 g/kg green tea powder

Green tea was obtained from an autumn harvest of Chinese green tea (Camellia sinensis, Langeroud’s Nooshineh Co., http://www.nooshinehco.com). Green tea extract was prepared by heating 100 g dry tea leaves with 200 ml distilled water at 80°C for 10 min (Bombik et al., 2012). Green tea powder was obtained by grinding and passing through a 0.5 mm sieve. Chicks were reared from days 1 to 42, in starter (1–10 days), grower (11–24 days) and finisher (25–42 days) periods. Dietary ingredients and nutrient composition of experimental diets, fed as mash, are in Table 1. Feed and water were available ad libitum.

The animals in each group were kept in 1.2 x 1.2 m pens, bedded with wood shavings. A standard thermo-neutral ambient temperature was maintained, in accordance with standard rearing practices.
Feeding ingredients and nutrient analysis of the diets used during the starter (days 1–10), grower (days 11–24) and finisher (days 25–42) periods

| Ingredient (%) | Days 1–10 | Days 11–24 | Days 25–42 |
|----------------|-----------|------------|------------|
| Maize (Zea mays) | 57.8 | 58.6 | 61.6 |
| Soybean meal (44% P) | 36.7 | 35.47 | 31.66 |
| Soybean oil | 1.60 | 2.20 | 3.17 |
| Limestone | 1.26 | 1.24 | 1.09 |
| Dicalcium phosphate | 1.56 | 1.30 | 1.15 |
| NaCl | 0.20 | 0.25 | 0.32 |
| Mineral and vitamin mixture* | 0.50 | 0.50 | 0.50 |
| DL-Methionine | 0.20 | 0.25 | 0.30 |
| L-Lysine hydrochloride | 0.04 | 0.05 | 0.07 |
| Total | 100 | 100 | 100 |
| Calculated nutrients | | | |
| Energy (ME) (kcal/kg) | 2900 | 2950 | 3000 |
| Crude protein | 21.0 | 20.5 | 18.75 |
| Crude fat (%) | 2.42 | 4.64 | 2.86 |
| Linoleic acid (%) | 2.81 | 2.13 | 1.46 |
| Calcium (%) | 0.94 | 0.87 | 0.78 |
| Available phosphorus (%) | 0.42 | 0.38 | 0.35 |
| Sodium (%) | 0.19 | 0.17 | 0.15 |
| Lysine (%) | 1.30 | 1.10 | 1.10 |

*Containing: calcium pantothenate: 4 mg/g; niacin: 15 mg/g; vitamin B6: 13 mg/g; Cu: 3 mg/g; Zn: 15 mg/g; Mn: 20 mg/g; Fe: 10 mg/g; K: 0.3 mg/g; vitamin A: 5000 IU/g; vitamin D3: 500 IU/g; vitamin E: 3 mg/g; vitamin K3: 1.5 mg/g; vitamin B2: 1 mg/g.

(Aviation 2007). Lighting was provided for 23 h on days 1–7 and 40–42, and for 20 h/day on days 8–39.

Routine vaccination and deworming were applied according to a programme certified by the regional veterinary authority. At day 9 of age, birds were vaccinated against infectious bronchitis and on days 9, 18 and 31 against Newcastle disease, all vaccinations being administered via the drinking water.

2.2 Coccidiosis challenge

All chickens were challenged with coccidiosis according to standard protocols (Ott et al., 2018). At day 21, all treatments except for NC were orally gavaged with 1.5 ml/chick suspension containing 200,000 live spored oocysts, including *Eimeria tenella*, *E. negatrix*, *E. maxima* and *E. acervulina*.

2.3 Broiler measurements

Feed intake and weight gain were recorded over two periods (days 1–21 and 22–42). Feed conversion ratio was calculated as feed intake/weight gain, and measurements were made, based on standard methods (Giannenas et al., 2011; Shabani et al., 2015). In brief, at day 42 after 4 h of fasting to evacuate the gastrointestinal tract, one bird with a representative body weight for each replicate was slaughtered for measuring carcass yield and distribution of meat and gastrointestinal tract characteristics. Feathers were removed by dry plucking. Neck, wingtips and feet at the tibio-tarsal joint were separated from the carcass. The gastrointestinal tract (from oesophagus to rectum), heart, kidney and liver were removed, and the empty or edible carcass weighed.

The different parts and the digestive tract of the carcasses were dissected, and the economically relevant parts weighed: first, breast muscle, including skin and sternum, then legs (thighs and drumsticks, dissected by disarticulation of the hip joint and dissecting tissue from the iliac bone). Cecum content was collected to evaluate microflora. All dimensions were rounded to whole integers. The weights of all dissected parts were related to the totally eviscerated carcass [(weight of component(s)/eviscerated carcass weight) × 100]. Three samples of 1 cm length from the centre of three intestinal segments were taken: first, the middle of the duodenum from gizzard outlet to the end of the pancreatic loop, second, middle of the jejunum from the pancreatic loop to Meckel's diverticulum, and third, 5 cm section from Meckel's diverticulum to the ileo-caecocolic junction. After washing with soluble phosphate-buffered saline (PBS), samples were fixed in 6 ml 10% buffered formalin for 2 days, embedded in paraffin, sectioned at 3 μm and stained the haematoxylin-eosin and periodic acid-Schiff methods. One cm long intestine segments were taken from the centre of each sample for morphometric studies under light microscopy. Three researchers independently measured villus length and width, crypt depth, epithelium thickness and number of goblet cells in the epithelium in each sample. Using ten villi and the corresponding crypts per section, villous length was estimated as the vertical distance from the tip to the crypt junction level, crypt depth as the vertical distance from villous-crypt junction to crypt base (Nahavandinejad et al., 2014).

2.3.1 Blood samples

Measurements were based on methods described previously (Nahavandinejad et al., 2014). Briefly, at the end of the 42-day experiment, one bird from each replicate was randomly selected for morning blood sampling – 3 birds per group. Prior to blood collection and slaughter, feed was removed for 4 h to stabilise plasma constituents. Samples (~5 ml/bird) were collected from the wing vein (Vena cutanea ulnaris) into tubes with 10 mg of the anticoagulant ethylenediaminetetraacetic acid (EDTA) for plasma separation, and immediately transferred to the laboratory. Plasma was harvested after centrifugation (3000 rpm, for 10 min at room temperature) and stored at ~20°C until analysed.

2.3.2 Determination of blood parameters

Blood parameters analysed were glucose (GI), total cholesterol (Chol), triglycerides (TG), high-density lipoprotein (HDL), low-density lipoprotein (LDL), very-low-density lipoprotein (VLDL), albumin (Alb), total...
protein (TP) and alkaline phosphatase (AP). Blood plasma parameters were analysed using a Roche Cobas Integra autoanalyzer (Roche Diagnostics, GmbH, Mannheim, Germany), based on standard protocols prescribed by the manufacturer of commercial kits (Pars Azmoon Co., Tehran, Iran) and described previously (Nahavandinejad et al., 2014; Shabaniet al., 2015).

2.3.3 Composition of cecum microbiota

Measurements were based on methods described previously (Dibaji et al., 2014). Briefly, agar plates were streaked with ceca contents for determination of bacterial growth from colony counts. Collecting tubes were weighed, wrapped in aluminium foil and autoclaved for 10 min. Culture mediums were prepared 24 h before collecting samples. Tryptose-sulphite Cycloserin Egg Yolk agar-TSC (CM 587 oxoid) was used for *Clostridium perfringens*, MacConkey agar (105465.0500) to culture coliforms, and MRS agar (Man Rogosa Sharpe agar, 1.10660.500) to culture *Lactobacilli*. Samples were transferred to the laboratory and weighed, with sample weight the difference between these two values. Tubes for bacteria isolated from gastrointestinal contents and preparation of suspension were shaken for 30 minutes. One millilitre was removed from the suspension and added to 9 ml buffer phosphate saline (PBS). The suspension was prepared from 10^{-1} dilutions and serial dilutions to 10^{-2}, 10^{-3}, 10^{-4}, 10^{-5} and 10^{-6}. A 100 μl sample was then removed from 10^{-4}, 10^{-5} and 10^{-6} dilutions and poured into the Petri dish previously prepared with the medium and completely distributed. *Lactobacilli* bacteria were incubated at 37°C in anaerobic conditions for 72 h, then bacteria were counted by a colony counter (Digital Magnificent Colony Counter, P118, S.ELC).

2.4 Statistical analysis

Results are presented as means ± standard error of the mean. The Shapiro–Wilk test confirmed normal distribution of the data, except bacterial counts, which were transformed to logarithm_{10} no. bacteria/g. Significance of treatment differences was analysed using ANOVA, followed by a Tukey’s post hoc test to separate means, using IBM SPSS Statistics 21 software for Windows® (SPSS 1997). p Values ≤ 0.05 were regarded as statistically significant.

3 RESULTS

All birds remained healthy throughout the study, with no evidence of diarrhoea and no mortality.

3.1 Productive performance

In the first half of the study, feed intake was least in NC, increased in PC and further increased by GTE0.4 (Table 2). In green tea powder treatments, however, feed intake was intermediate between NC and PC. Weight gain was least in NC, increased in PC and green tea treatments and was highest in GTE0.4. FCR was least in GTE0.4, and reduced for all green tea levels and PC, compared with NC.

In the second half of the study, feed intake was increased in GTE0.3 and 0.4 and PC, compared with other treatments. Weight gain was

| Treatment | Feed intake (g/chick) | Weight gain (g/chick) | Feed conversion ratio |
|-----------|-----------------------|-----------------------|----------------------|
| First half of study (days 1–21) | | | |
| NC | 978^c | 641^d | 1.52^a |
| PC | 1031^b | 740^b | 1.39^bc |
| GTE0.2 | 1036^ab | 736^b | 1.40^bc |
| GTE0.3 | 1047^a | 743^b | 1.40^c |
| GTE0.4 | 1030^b | 726^ac | 1.37^c |
| GTP1 | 1027^b | 715^c | 1.44^b |
| GTP2 | 1025^b | 710^c | 1.44^b |
| GTP3 | 1025^b | 710^c | 1.44^b |
| p Value | 0.0001 | 0.0001 | 0.0001 |
| SEM | 4.13 | 7.33 | 0.0097 |

| Second half of study (days 22–42) | | | |
| NC | 3792^b | 2009^d | 1.89 |
| PC | 3937^a | 2145^ab | 1.84 |
| GTE0.2 | 3841^b | 2098^bc | 1.83 |
| GTE0.3 | 3934^a | 2147^ab | 1.83 |
| GTE0.4 | 3943^a | 2192^a | 1.80 |
| GTP1 | 3798^b | 2074^d | 1.83 |
| GTP2 | 3823^b | 2070^d | 1.85 |
| GTP3 | 3833^b | 2065^d | 1.86 |
| p Value | 0.0001 | 0.0007 | 0.1219 |
| SEM | 13.755 | 13.097 | 0.0071 |

| Total study | | | |
| NC | 4770^c | 2650^d | 1.80^a |
| PC | 4968^a | 2885^a | 1.72^c |
| GTE0.2 | 4872^a | 2833^c | 1.72^c |
| GTE0.3 | 4970^a | 2890^a | 1.72^c |
| GTE0.4 | 4990^a | 2953^a | 1.69^c |
| GTP1 | 4828^b | 2800^c | 1.72^c |
| GTP2 | 4850^b | 2785^b | 1.74^b |
| GTP3 | 4858^b | 2775^c | 1.75^b |
| p Value | 0.0001 | 0.0001 | 0.0001 |
| SEM | 16.2950 | 18.8301 | 0.0071 |

*Means within each column with common superscripts do not differ significantly (p < 0.05).
NC: negative control; PC: positive control; GTE: green tea extract; GTP: green tea powder; SEM: standard error of mean.
TABLE 3  Carcass weight as a % of total weight, and the weight of breast, drumstick, ventriculus, proventriculus, intestine and body fat, as % of carcass weight, at day 42 of age in Ross 308 male broilers (n = 24) fed different levels of green tea extract (GTE) or powder (GTP), a neutral control (NC) or a positive control (PC)

| Treatment | Eviscerated carcass (% total weight) | Breast (% CW) | Drumstick (% CW) | Ventriculus (% CW) | Proventriculus (% CW) | Intestine (% CW) | Abdominal fat (% CW) |
|-----------|-------------------------------------|---------------|------------------|-------------------|---------------------|-----------------|---------------------|
| NC        | 64.86                               | 28.68         | 24.90            | 0.94b             | 0.49               | 5.84d           | 1.36a               |
| PC        | 65.51                               | 28.08         | 24.26            | 0.94b             | 0.43               | 5.46d           | 1.42a               |
| GTE0.2    | 65.77                               | 29.11         | 24.62            | 0.94b             | 0.47               | 5.78d           | 1.21b               |
| GTE0.3    | 66.55                               | 29.24         | 24.91            | 0.94b             | 0.48               | 5.77d           | 1.21b               |
| GTE0.4    | 66.75                               | 29.90         | 25.43            | 0.88b             | 0.45               | 5.49d           | 1.03c               |
| GTP1      | 65.75                               | 28.46         | 24.39            | 1.17a             | 0.43               | 7.02bc          | 0.95cd              |
| GTP2      | 65.35                               | 29.51         | 25.27            | 1.17a             | 0.43               | 7.42bc          | 0.88d               |
| GTP3      | 65.55                               | 28.57         | 25.18            | 1.18a             | 0.48               | 8.92a           | 0.89d               |
| p Value   | 0.37                                | 0.07          | 0.25             | 0.0003            | 0.63               | 0.0002          | 0.0001              |
| SEM (std error of mean) | 0.224   | 0.164         | 0.132            | 0.0267            | 0.0099             | 0.268           | 0.423               |

*Means within each column with common superscript do not differ significantly at *p* < 0.05.
NC: negative control (diets not supplemented); PC: positive control (diets supplemented commercial coccidiostat); GTE: green tea extract; GTP: green tea powder.

highest in these treatments, and least at the higher powder inclusion rates and NC. FCR was not affected by treatment.

Over the entire study, feed intake was greatest for in GTE0.3 and 0.4 and PC, least for NC and intermediate in other treatments. Weight gain was greatest in the GTE0.4, least in NC and was greater in PC than the 1–3 g/kg powder treatments. FCR was least in GTE0.04 and greatest in NC.

3.2 Carcass composition

There were no significant treatment effects on the carcass weight, or on breast, drumstick or proventriculus weights, as % of carcass weight (Table 3). However, ventriculus weight was increased at the three highest green tea inclusion rates, as powder, compared with the other treatments. Intestine weight was greatest at the highest powder inclusion rate and was also greater at the second highest inclusion rate than the controls and lower inclusion rates. Abdominal fat was greatest in the two control treatments and decreased with inclusion rate of green tea.

3.3 Gut mucosa morphology

3.3.1 Duodenum

Villus length was longest for GTE0.04% extract treatment and least in the two control treatments (Table 4); it declined at higher inclusion rates of powder. Villus width increased, and epithelium thickness decreased, in the green tea treatments compared with NC, and intermediate in PC. Crypt depth was decreased at all levels of green tea inclusion, compared with NC, with PC intermediate. The ratio of villus length to crypt depth was greatest in the GTE0.4, then the lower extract concentrations, then the higher, powder, concentrations and, finally, least in NC and PC. Treatment did not affect the number of goblet cells.

3.3.2 Jejunum

Villus length and width were increased, and crypt depth was decreased, by green tea treatments, compared with NC and intermediate in PC (Table 5). Villus length to crypt depth ratio increased in all green tea treatments, compared with NC, and increased in all green tea treatments except the highest powder concentration, compared with PC. Epithelial thickness was greatest in NC and least in PC and green tea treatments up to 1 g/kg, and intermediate in GTP2 and 3. Goblet cells were least in NC and PC and increased for green tea treatments, with a peak for GTE 0.4.

3.3.3 Ileum

Villi were taller in all the green tea treatments compared with the two control treatments, which did not differ (Table 6). GTE0.4 had taller villi than other green tea treatments. Villus width increased and crypt depth decreased in the green tea treatments, compared with NC, with PC intermediate. Villus length to crypt depth ratio was increased in all green tea treatments, compared with NC and PC, and it increased in GTE0.4 compared with other green tea treatments. Epithelial thickness was less in the green tea treatments and PC, compared with NC. Goblet cells were greatest in GTE0.4, then other green tea treatments, then PC and least in NC.
### TABLE 4  Duodenal morphology at day 42 of age in Ross 308 male broilers (n = 24) fed different levels of green tea extract (GTE) or powder (GTP), neutral control (NC) or positive control (PC)

| Treatment | Villus length (µm) | Villus width (µm) | Crypt depth (µm) | Villus length to crypt depth ratio | Thickness of epithelium (µm) | Number of goblet cells in epithelium |
|-----------|-------------------|-------------------|------------------|-----------------------------------|-----------------------------|----------------------------------|
| NC        | 1700d             | 148c              | 321a             | 5.31c                             | 41.8a                       | 5                                |
| PC        | 1910d             | 170bc             | 270b             | 7.08c                             | 30.1c                       | 8                                |
| GTE0.2    | 2393abc           | 197a              | 231bc            | 10.42ab                           | 35.3b                       | 10                               |
| GTE0.3    | 2390ab            | 199a              | 213c             | 11.36ab                           | 34.2bc                      | 13                               |
| GTE0.4    | 2443abc           | 213a              | 200c             | 12.31a                            | 33.3bc                      | 15                               |
| GTP1      | 2310bc            | 193ab             | 233bc            | 10.01b                            | 34.3b                       | 11                               |
| GTP2      | 2320bc            | 191ab             | 239bc            | 7.79ab                            | 34.1b                       | 11                               |
| GTP3      | 2290bc            | 188ab             | 240bc            | 9.56b                             | 33.3bc                      | 11                               |
| p Value   | 0.0001            | 0.001             | 0.0002           | 0.0001                            | 0.002                       | 0.35                             |
| SEM (standard error of mean) | 53.35 | 4.57 | 8.24 | 0.486 | 0.69 | 0.93 |

*Means within each column with common superscript do not differ significantly at p < 0.05.
NC: negative control; PC: positive control; GTE: green tea extract; GTP: green tea powder.

### TABLE 5  Jejunum morphology at day 42 of age in Ross 308 male broilers (n = 24) fed different levels of green tea extract (GTE) or powder (GTP), a neutral control (NC) or a positive control (PC)

| Treatment | Villus length (µm) | Villus width (µm) | Crypt depth (µm) | Villus length to crypt depth ratio | Thickness of epithelium (µm) | Number of goblet cells in epithelium |
|-----------|-------------------|-------------------|------------------|-----------------------------------|-----------------------------|----------------------------------|
| NC        | 990d              | 141c              | 217a             | 4.61c                             | 43.2a                       | 5c                               |
| PC        | 1180abcd          | 170bc             | 190abc           | 6.22bc                            | 30.2c                       | 10c                              |
| GTE0.2    | 1400ab            | 188ab             | 155c             | 9.04a                             | 33.1bc                      | 18abc                            |
| GTE0.3    | 1461ab            | 195ab             | 155c             | 9.57a                             | 32.0bc                      | 19abc                            |
| GTE0.4    | 1483a             | 209a              | 148c             | 10.01a                            | 30.2c                       | 24a                              |
| GTP1      | 1383abc           | 189ab             | 158bc            | 8.83a                             | 34.3bc                      | 19abc                            |
| GTP2      | 1378bc            | 190ab             | 163bc            | 8.65a                             | 34.7b                       | 17b                              |
| GTP3      | 1381abc           | 190ab             | 163bc            | 8.47ab                            | 35.0b                       | 17b                              |
| p Value   | 0.0009            | 0.03              | 0.003            | 0.001                             | 0.002                       | 0.0001                           |
| SEM (standard error of mean) | 34.85 | 5.13 | 5.41 | 0.388 | 0.91 | 1.26 |

*Means within each column with common superscript do not differ significantly at p < 0.05.
NC: negative control (diets not supplemented); PC: positive control (diets supplemented commercial coccidiostat); GTE: green tea extract; GTP: green tea powder.

### 3.4 Blood composition

Blood glucose was reduced in GTP2 and 3, compared with the two control treatments, with other green tea treatments intermediate (Table 7). Total cholesterol was reduced in all green tea treatments and PC compared with NC, and further decreased at the highest green tea inclusion rate, GTP3. Triglycerides were reduced at this highest rate compared to NC and PC and the lowest inclusion rate, GTE0.2. HDL cholesterol was not affected by treatment, but LDL cholesterol was reduced by all the green tea treatments, and particularly the highest level, compared with NC. PC was intermediate and greater than all green tea treatments except GTE0.2. VLDL was reduced by most green tea inclusion rates, but particularly the highest level, compared with NC and PC.

### 3.5 Cecum bacterial content

Populations of *Clostridium perfringens* were reduced in proportion to green tea inclusion rate, compared with NC, with PC the same as all green tea inclusion rates except GTP3 (Table 8). Coliforms were decreased in approximate proportion to green tea inclusion rate,
### TABLE 6  
Ileal morphology at day 42 of age in Ross 308 male broilers (n = 24) fed different levels of green tea extract (GTE) or powder (GTP), a neutral control (NC) or a positive control (PC)

| Treatment | Villus length (µm) | Villus width (µm) | Crypt depth (µm) | Villus length to crypt depth ratio | Thickness of epithelium (µm) | Number of goblet cells in epithelium |
|-----------|---------------------|-------------------|-----------------|-------------------------------|-----------------------------|--------------------------------|
| NC        | 880<sup>a</sup>     | 120<sup>c</sup>   | 256<sup>a</sup> | 3.43<sup>c</sup>          | 41.2<sup>a</sup>                      | 5<sup>f</sup>               |
| PC        | 990<sup>c</sup>     | 143<sup>bc</sup>  | 185<sup>bc</sup>| 5.61<sup>c</sup>          | 30.0<sup>b</sup>                      | 10<sup>c</sup>             |
| GTE0.2    | 1150<sup>b</sup>    | 175<sup>ab</sup>  | 173<sup>bc</sup>| 6.78<sup>b</sup>          | 33.2<sup>b</sup>                      | 18<sup>b</sup>             |
| GTE0.3    | 1175<sup>b</sup>    | 180<sup>a</sup>   | 168<sup>bc</sup>| 7.02<sup>b</sup>          | 33.1<sup>b</sup>                      | 19<sup>b</sup>             |
| GTE0.4    | 1300<sup>c</sup>    | 195<sup>a</sup>   | 143<sup>c</sup> | 9.11<sup>a</sup>          | 30.1<sup>b</sup>                      | 24<sup>a</sup>             |
| GTP1      | 1145<sup>ab</sup>   | 168<sup>b</sup>   | 175<sup>bc</sup>| 6.57<sup>b</sup>          | 34.2<sup>b</sup>                      | 19<sup>b</sup>             |
| GTP2      | 1110<sup>b</sup>    | 171<sup>ab</sup>  | 183<sup>ab</sup>| 6.21<sup>b</sup>          | 34.1<sup>b</sup>                      | 18<sup>b</sup>             |
| GTP3      | 1110<sup>b</sup>    | 173<sup>ab</sup>  | 180<sup>bc</sup>| 6.45<sup>b</sup>          | 33.4<sup>b</sup>                      | 17<sup>b</sup>             |
| p Value   | 0.0001              | 0.005             | 0.03            | 0.0006                       | 0.004                       | 0.0001                     |
| SEM (standard error of mean) | 27.2               | 5.55              | 7.81            | 0.354                        | 0.816                        | 1.21                        |

*Means within each column with common superscript do not differ significantly at p < 0.05. NC: negative control; PC: positive control; GTE: green tea extract; GTP: green tea powder.

### TABLE 7  
Blood parameters at day 42 of age in Ross 308 male broilers (n = 24) fed different levels of green tea extract (GTE) or powder (GTP), a neutral control (NC) or a positive control (PC)

| Treatment | Glucose (mg/dl) | Total cholesterol (mg/dl) | Triglycerides (mg/dl) | HDL Cholesterol (high-density lipoproteins) (mg/dl) | LDL Cholesterol (low-density lipoproteins) (mg/dl) | VLDL (mg/dl) |
|-----------|-----------------|---------------------------|-----------------------|---------------------------------------------------|---------------------------------------------------|-------------|
| NC        | 238<sup>ab*</sup> | 201<sup>a</sup>           | 119<sup>a</sup>      | 61                                                 | 117<sup>a</sup>                                    | 23.8<sup>a</sup>|
| PC        | 240<sup>c</sup>  | 173<sup>b</sup>           | 117<sup>a</sup>      | 58                                                 | 91<sup>b</sup>                                     | 23.3<sup>a</sup>|
| GTE0.2    | 219<sup>abc</sup>| 168<sup>b</sup>           | 108<sup>bc</sup>     | 67                                                 | 79<sup>bc</sup>                                     | 16.5<sup>ab</sup>|
| GTE0.3    | 213<sup>abc</sup>| 169<sup>b</sup>           | 85<sup>lcd</sup>     | 75                                                 | 78<sup>c</sup>                                     | 17.0<sup>abcd</sup>|
| GTE0.4    | 203<sup>abc</sup>| 160<sup>b</sup>           | 75<sup>c</sup>       | 74                                                 | 71<sup>c</sup>                                     | 15.0<sup>cd</sup>|
| GTP1      | 198<sup>bc</sup>| 164<sup>b</sup>           | 103<sup>abc</sup>    | 66                                                 | 78<sup>c</sup>                                     | 20.6<sup>abc</sup>|
| GTP2      | 193<sup>c</sup>| 165<sup>b</sup>           | 93<sup>abcd</sup>    | 67                                                 | 79<sup>c</sup>                                     | 18.6<sup>abcd</sup>|
| GTP3      | 183<sup>c</sup>| 133<sup>c</sup>           | 74<sup>d</sup>       | 70                                                 | 49<sup>d</sup>                                     | 14.8<sup>d</sup>|
| p Value   | 0.04            | 0.0001                    | 0.008                | 0.18                                               | 0.0001                                           | 0.008        |
| SEM (standard error of mean) | 5.39             | 4.05                      | 4.30                 | 1.74                                               | 3.83                                              | 0.859        |

*Means within each column with common superscript do not differ significantly at p < 0.05. NC: negative control; PC: positive control; GTE: green tea extract; GTP: green tea powder.

compared with NC, but PC was the same as higher levels of green tea. Lactobacilli were increased by green tea inclusion, more at GTP2 and 3, compared with NC and particularly PC.

#### 4 DISCUSSION

Green tea increased length and width of villi in the different intestinal segments, as well as reducing crypt depth, compared with NC. This led to increased growth rates and feed conversion ratio, as well as increased feed intake, particularly at the lower inclusion rates. The improvement in weight gain of experimental birds fed with GTE0.4, compared with PC, may be due to the action of flavonoids, known to have anticoccidial effects due to antioxidants (Chen et al., 2008; Jang et al., 2007).

The increase in feed intake contrasts with decreases reported previously (Alimohammadi-Saraei et al., 2014; Biswas & Wakita, 2001), but these authors included green tea at much higher doses. The smaller increase in intake at our higher inclusion rates suggests a dose dependent effect. Others (Hrnčár & Bujko, 2017) reported no effect of green tea supplementation on feed intake in broilers. Observed reductions in feed intake (Alimohammadi-Saraei et al., 2014; Biswas & Wakita, 2001), attributed to suppression of lipid metabolism, reduced broilers’ growth, probably unacceptable to the industry. Yang et al. (Yang et al., 2003) also observed a reduction in weight gain of broilers fed 10 g/kg of their diet as green tea by-product, high in tannins. Biswas and Wakita
TABLE 8  Mean (± SEM) cecum microbial populations at day 42 in Ross 308 male broilers (n = 24) fed different levels of green tea extract (GTE) or powder (GTP), a neutral control (NC) or a positive control (PC)

| Treatment   | Clostridium perfringens (log_{10} CFU/g) | Coliforms (log_{10} CFU/g) | Lactobacillus (log_{10} CFU/g) |
|-------------|----------------------------------------|-----------------------------|-------------------------------|
| NC          | 2.59a                                  | 7.65a                       | 7.54f                         |
| PC          | 2.25bcd                                | 7.40d                       | 6.48d                         |
| GTE0.2      | 2.34b                                  | 7.56b                       | 7.65a                         |
| GTE0.3      | 2.31bc                                 | 7.54ab                      | 7.73ab                        |
| GTE0.4      | 2.22d                                  | 7.40d                       | 7.78a                         |
| GTP1        | 2.28bcd                                | 7.48bc                      | 7.67b                         |
| GTP2        | 2.20d                                  | 7.39d                       | 7.81e                         |
| GTP3        | 1.80e                                  | 7.34d                       | 7.82e                         |
| p Value     | 0.0001                                 | 0.0007                      | 0.0001                        |
| SEM (standard error of mean) | 0.0437                               | 0.0234                      | 0.0873                        |

*Means within each column with common superscript do not differ significantly at p < 0.05.
NC: negative control; PC: positive control; GTE: green tea extract; GTP: green tea powder; CFU: colony forming units.

(Biswas & Wakita, 2001) found that green tea powder included at 5, 7, 10, and 15 g/kg only tended to decrease weight gain at the highest dose. The optimum inclusion rate for production performance, at which FCR is least, may be 0.4 g/kg. Differing effects may also be due to the type of green tea used, in particular catechin content. Improved FCR could be attributed to the most abundant catechin, epigallocatechin gallate. Other green tea components, alkaloids, carotenoids, minerals, amino acids (especially l-theanine), and volatiles compounds, decrease pathogens and improve nutrient absorption, leading to better FCR (Engelhardt, 2010).

4.1 | Carcass characteristics

We observed no effects on dressing %; neither did Biswas and Wakita (Biswas & Wakita, 2001) when they added green tea to broilers’ diet at 5–15 g/kg. In contrast, Guray et al. (2011) observed increased dressing percentage. A reduction in abdominal fat has also been reported by Yang et al. (2003) and Guray et al. (2011). The increase in intestine weight at our two highest levels probably reflects changes in villi. Contrary to our study, Uuganbayar (2004), when supplementing with 0.5% green tea, recorded lighter small intestines compared to a control diet.

4.2 | Intestinal morphology

Similar results were obtained from the three sections of the intestine. Villi length and width were increased by green tea, especially in GTE0.4. In Hassanpour et al.’s (2010) study, jejunal but not duodenal or ileal villus length increased, with no increase in villus width. In our study, the proportional increase in villus length was similar in the three intestinal segments (+55%, 48% and 44% for the jejunum, ileum and duodenum, respectively). Epithelial tissue was thinner and there was a tendency for the number of goblet cells to increase, at least in the jejenum and ileum, and again especially in GTE0.4. Thinner epithelial tissue may facilitate nutrient absorption, also longer villi. Intestinal villi are the main site of nutrient absorption (Ray et al., 2002). There is a positive relationship between between villus length, nutrient absorption and digestibility (Mekbungwan et al., 2002). Thus, it was noticeable that birds in treatment GTE0.4 gained most weight, had biggest villi and most goblet cells. Goblet cells secrete mucin glycoproteins that protect the epithelium and they sense microbiota, sampling bacteria and transferring them to dendritic cells, thereby actively participating in immune responses to pathogens (Shira & Friedman, 2018). Shorter villi and deeper crypts will reduce nutrient absorption. According to Yason et al. (1987), deep crypts indicate fast renewal of the villi in response to tissue inflammation or toxins produced by pathogens. Thus, it is likely that it was the changes in crypt depth and thickness of the epithelium, which caused the slower weight gain observed in NC.

4.3 | Haematological parameters

As has been demonstrated, green tea is hypocholesterolemic, particularly of LDL cholesterol. Green tea reduces lipoprotein lipase and adipose triglyceride lipase (Mohammadpour et al., 2021). This action of green tea may be link to polyphenols, which enhance reverse-cholesterol transport: cholesterol is removed from peripheral tissues and delivered to the liver, reducing cholesterol absorption (Tebib et al., 2004). Other researchers have found that 1.5 g/kg of green tea significantly decreased plasma triglyceride and total cholesterol, compared to a control group (El-Deek et al., 2012).

4.4 | Cecal microbial population

Coliforms and Clostridium perfringens populations, respectively 7.65 CFU/g and 2.59 CFU/g, were greater for NC, and the lactobacillus
population (7.54 CFU/g) for the same group was less (p < 0.05), compared to the other groups. Coliforms and Clostridium perfringens decreased as green tea level increased. The least value in the GTE group was 2.22 CFU/g while 1.8 CFU/g was the least value in GTP group. Green tea increased lactobacilli (Table 8), thus increasing useful intestinal microflora, and inhibiting growth of enteropathogenic strains of Coliforms and Clostridium perfringens. The better nutrient utilisation and improved growth and development of chickens, particularly in GTE0.4, may be a direct result of this. The stimulation of useful and inhibition of pathogenic microflora leads to improved absorption and utilisation of nutrients. Thus, green tea can inhibit and kill a wide range of pathogenic bacteria (Hamilton-Miller, 1997).

5 | CONCLUSIONS

We found benefits of including green tea in the diet of broilers from increased weight gain, improved feed conversion, reduced LDL and VLDL cholesterol, and reduced pathogenic bacteria in the cecum. At least some of these beneficial effects of green tea may stem from direct effects on intestinal mucosa morphology, since we observed that it supported the growth of villi and goblet cells and reduced the thickness of the epithelium. The results also confirm the beneficial effects of green tea as a feed additive, as it increased production performance and enhanced health status of the birds. The most beneficial effects were observed with green tea included in the diet at a concentration of 0.04 g/kg.

AUTHOR CONTRIBUTIONS

Conceptualisation: K.J., B.R., A.S. Methodology: K.J., B.R., A.S. Formal analysis: K.J., B.R., I.T.K., A.S. Investigation: K.J., B.R., I.T.K., M.S., I.G., A.S., C.J.C.P. Resources: K.J., B.R., I.T.K., M.S., I.G. Data curation: K.J., B.R., I.T.K., M.S., I.G. Writing—original draft preparation: I.T.K., M.S., I.G. Writing—review and editing: C.J.C.P. Supervision: B.R., A.S. Project administration: K.J., M.S., I.G. Funding acquisition: K.J., M.S., I.G., C.J.C.P.

AUTHOR DECLARATIONS

All authors are either employed by, or associated with, a government agency or university, whose primary function is research and education.

ANIMAL ETHICAL STATEMENT

All procedures involving animals were approved by the Animal Ethics Committee of the Islamic Azad University (Approval number 11750103931001) and conducted in accordance to the EU’s International Guidelines for Research involving Animals (Directive 2010/63/EU).

ACKNOWLEDGEMENTS

The authors are grateful to staff at the commercial poultry farm at Amirabad, Lahijan, Iran, and to Rasht Branch, Islamic Azad University, Rasht, Iran (grant number 17.16.3.16303). The research study proposed was conducted under the grant of the RSF No. 22-16-00041, 21-16-00025, GNU NIIMMP.

CONFLICT OF INTEREST

The authors declare that there are no conflicts of interest.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

PEER REVIEW

The peer review history for this article is available at https://publons.com/publon/10.1002/vms.923.

ORCID

Alireza Seidavi https://orcid.org/0000-0002-1903-2753

REFERENCES

Abdo, Z. M. A., Hassan, R. A., Amal, A. E., & Shahinaz, A. H. (2010). Effect of adding green tea and its aqueous extract as natural antibiotics to laying hen diet on productive, reproductive performance and egg quality during storage and its content of cholesterol. Egyptian Poultry Science Journal, 30, 1121–1149.

Alimohammadi-Saraei, M. H., Seidavi, A. R., Dadashbeiki, M., Laudadio, V., & Tufarelli, V. (2014). Effect of dietary supplementation with different levels of green tea powder and fish oil or their combination on carcass characteristics in broiler chickens. Pakistan Journal of Zoology, 46, 1767–1773.

Aviagen. (2007). Ross 308 BROILER: Nutrition specification (p. 8). Scotland (UK): Aviagen.

Biswas, M. A. H., & Wakita, M. (2001). Effect of dietary Japanese green tea powder supplementation on feed utilization and carcass profiles in broilers. Journal of Poultry Science, 38, 50–57.

Bombik, T., Bombik, E., Frankowska, A., Trawińska, B., & Saba, L. (2012). Effect of herbal extracts on some haematological parameters of calves during rearing. Bulletin of the Veterinary Institute of Pulawy, 56, 655–658.

Butaye, P. K., Van Damme, L. A., Devriese, L., Van Damme, M., Bael, S., & Haesebrouck, F. (2000). In vitro susceptibility of Enterococcus faecium isolated from food to growth promoting and therapeutic antibiotics. International Journal of Food Microbiology, 54, 181–187.

Cao, B. H., Karasawa, Y., & Guo, Y. M. (2005). Effects of green tea polyphenols and fructo-oligosaccharides in semi-purified diets on broilers’ performance and caecal microflora and their metabolites. Asian-Australasian Journal of Animal Sciences, 18, 85–89.

Catalá-Gregori, P., Mallet, S., Travel, A., & Lessire, M. (2002). Efficiency of a prebiotic and a plant extract on broiler performance and intestinal physiology. Proceedings of the 16th European Symposium on Poultry Nutrition; 2002; Strasbourg. France: World Poultry Science Association; 2007; https://www.cabi.org/Uploads/animal-science/worlds-poultry-science-association/WPSA-france-2007/32.pdf, Accessed 17 January, 2022.

Chen, H., Zhang, M., Qu, Z., & Xie, B. (2008). Antioxidant activities of different fractions of polysaccharide conjugates from green tea (Camellia sinensis). Food Chemistry, 106, 559–563.

Cross, D. E., McDevitt, R. M., Hilman, K., & Acamovic, T. (2007). The effect of herbs and their associated essential oils on performance, dietary digestibility and gut microflora in chickens from 7–28 days of age. British Poultry Science, 48, 496–506.

Demir, E., Sarica, S., Ozcan, M. A., & Suıcemz, M. (2003). The use of natural feed additives as alternatives for antibiotic growth promoters in broiler diets. British Poultry Science, 44, 544–545.

Dibaji, S. M., Seidavi, A. R., Asadpour, L., & Moreira da Silva, F. (2014). Effect of a symbiotic on the intestinal microflora of chickens. Journal of Applied Poultry Research, 23, 1–6. https://doi.org/10.3382/japr.2012-00709

Dieye, P. N., Missohou, N. A., & Faye, A. (2010). L’aviculure familiale: Un levier pour améliorer les revenus des éleveurs pauvres au Sud du

JELVEH ET AL.
Kaneko, K., Yamasaki, Y., Tagawa, M., Tokunaga, M., Tobisa, & Furus, M. (2010). Chemistry of tea. Comprehensive natural products II: Chemistry and biology (vol. 9, pp. 999–1032). Amsterdam, The Netherlands: Elsevier Science.

Erener, G., Ocak, N., Altop, A., Cankaya, S., Aksoy, H. M., & Ozturk, E. (2011). Growth performance, meat quality and caecal coliform bacteria count of broiler chicks fed with green tea extract. Asian-Australasian Journal of Animal Sciences, 24, 1128–1135.

Gianinhas, I., Tsialle, E., Chronis, E., Mavridis, S., Tontis, D., & Kyriazakis, I. (2011). Consumption of Agaricus bisporus mushroom affects the performance, intestinal microbiota composition and morphology, and antioxidant status of turkey poulets. Animal Feed Science and Technology, 165, 218–229. https://doi.org/10.1016/j.anifeedsci.2011.03.002

Gould, I. M. (2008). The epidemiology of antibiotic resistance. International Journal of Antimicrobial Agents, 32(1 Supplement 1), S2–S9.

Guray, E., Ocak, N., Altop, A., Cankaya, S., Aksoy, H. M., & Ozturk, E. (2011). Growth performance, meat quality and caecal coliform bacteria count of broiler chicks fed with green tea extract. Asian-Australasian Journal of Animal Sciences, 24, 1128.

Hamilton-Miller, J. M. T. (1997). Microbial properties of tea infusions. In R. Schubert & M. Spiro (Eds), Chemical and biological properties of tea infusions (pp. 63–74). Frankfurt, Germany: U & M.

Hassanpour, H., Moghaddam, A. K. Z., Yazdani, A., & Bashi, M. C. (2010). Evaluation of intestinal morphology and nitric oxide metabolites in broiler chickens supplemented by green tea. Comparative Clinical Pathology, 19, 43–47. https://doi.org/10.1007/s00580-009-0831-x

Hrnčar, C., & Bujko, J. (2017). Effect of different levels of green tea (Camellia sinensis) on productive performance, carcass characteristics and organs of broiler chickens. Potravinarstvo Slovack Journal of Food Sciences, 11, 623–628.

Jang, S. I., Jun, M. H., Lillehoj, H. S., Dalloul, R. A., Kong, I. K., Kim, S., & Min, W. (2007). Anticoccidial effect of green tea-based diets against Eimeria maxima. Veterinary Parasitology, 144, 172–175.

Kaneko, K., Yamasaki, Y., Tagawa, M., Tokunaga, M., Tobisa, & Furus, M. (2001). Effect of dietary Japanese green tea powder on the growth, meat ingredient and lipid accumulation in broilers. Japanese Poultry Science, 38, 77–85.

Karori, S. M., Wachira, F. N., Wanyoko, J. K., & Ngure, R. M. (2007). Antioxidant capacity of different types of tea products. African Journal of Biotechnology, 6, 2287–2296.

Khan, S. H. (2014). The use of green tea (Camellia sinensis) as a phytogenic substance in poultry diets. Journal of Veterinary Research, 81, 1–8. https://doi.org/10.4102/jvrr.v8i1.706

Ko, S. Y., & Yang, C. J. (2008). Effect of green tea probiotics on the growth performance, meat quality and immune response in finishing pigs. Asian-Australasian Journal of Animal Sciences, 21(9), 1339–1347.

Kumar, M., Choudhary, R. S., & Vaishnav, J. K. (2003). Effect of supplemented prebiotics, probiotic and turmeric in diet on the performance of broiler chicks during summer. International Journal Poultry Science, 40, 137–141.

Liao, S., Yao, K. H., & Hiipakka, R. A. (2001). Green tea: Biochemical and biological basis for health benefits. Vitamins and Hormones, 62, 1–94.

Mekburganw, A., Yamauchi, K. E., & Thongwitaya, N. (2002). Intestinal morphology and enteral nutrient absorption of pigeon pea seed meal in piglets. Animal Science Journal, 73, 509–516.

Mohammadpour, F., Darmani-kahi, M., Mohit, A., & Sohani, M. (2021). Effects of dietary fat source and green tea (Camellia sinensis) extract on genes associated with lipid metabolism and inflammatory responses in female broiler chickens. Italian Journal of Animal Science, 20, 578–586. 10.1080/1828051X.2021.1898292

Nahavandinejad, M., Seidavi, A., Asadpour, L., & Payan-Carreira, R. (2014). Blood biochemical parameters of broilers fed differently thermal processed soybean meal. Revista MVZ Córdoba, 19, 4301–4315.

Nishida, T., Eruden, B., Hosoda, K., Nakagawa, K., Miyazawa, T., & Shiyo, S. (2006). Effects of green tea (Camellia sinensis) waste silage and polyethyleneuronic fermentation and blood components in cattle. Norwegian-Australasian Journal of Animal Sciences, 19, 1728–1736.

Norwegian Scientific Committee for Food Safety (VKM). (2015). The risk of development of antimicrobial resistance with the use of coccidiostats in poultry diets. Opinion of the Panel on Animal Feed of the Norwegian Scientific Committee for Food Safety, ISBN: 978-82-8259-185-0, Oslo, Norway.

Ott, C. P., Omara, I. L., Persia, M. E., & Dalloul, R. A. (2018). The impact of β-glucans on performance and response of broiler chickens during a coccidiosis challenge. Poultry Science, 97(8), 2713–2721.

Phillips, I., Casewell, M., Cox, T., De Groot, B., Friis, C., Jones, R., Nightingale, C., Preston, R., & Waddell, J. (2004). Does the use of antibiotics in food animals pose a risk to human health? A critical review of published data. Journal of Antimicrobial Chemotherapy, 53, 28–52.

Ray, E. C., Avissar, N. E., & Sax, H. C. (2002). Growth factor regulation of enterocyte nutrient transport during intestinal adaptation. American Journal of Surgery, 183, 361–371.

Shabani, S., Seidavi, A., Asadpour, L., & Corazzin, M. (2015). Effects of physical form of diet and intensity and duration of feed restriction on the growth performance, blood variables, microbial flora, immunity, and carcass and organ characteristics of broiler chickens. Livestock Science, 180, 150–157.

Shira, E. B., & Friedman, A. (2018). Innate immune functions of avian intestinal epithelial cells: Response to bacterial stimuli and localization of responding cells in the developing avian digestive tract. PLoS One, 13(7), e0200393.

SPSS (1997). SPSS Base 7.5 for Windows. SPSS, Chicago, IL.

Swormink, (2019). Coccidiostats: Antibiotics or feed additives? Poultry World, Mar 8, 2019.

Tebib, K., Bitri, L., Besancon, P., & Rouanet, J. M. (2004). Polymeric grape seed tannins prevent plasma cholesterol changes in high-cholesterol-fed rats. Food Chemistry, 49, 403–406.

Teteh, A., Lawson, E., Tona, K., Decuyper, E., & Gbeassor, M. (2013). Moringa oleifera leaves: hydro-alcoholic extract and effects on growth performance of broilers. International Journal of Poultry, 12, 401–405.

Uuganbayar, D. (2004). A study on the utilization of green tea for laying hens and broiler chicks. Dissertation thesis. Suncheon, Korea: Suncheon National University.

Van de Bogaard, A. E., & Stobberingh, E. E. (2000). Epidemiology of resistance to antibiotics, Links between animals and humans. International Journal of Antimicrobial Agents, 14, 322–335.

Yang, C. J., Yang, I. Y., Oh, D. H., Bae, I. H., Cho, S. G., Kong, I. G., Uuganbayar, D., Noy, I. S., & Choi, K. S. (2003). Effect of green tea by-product on performance and body composition in broiler chicks. Asian-Australasian Journal of Animal Sciences, 16, 867–872.

Yason, C. V., B. A., Summers, & Schat, K. A. (1987). Pathogenesis of rotavirus infection in various age groups of chickens and turkeys pathology. American Journal of Veterinary Research, 48, 927–938.

How to cite this article: Jelveh, K., Rasouli, B., Kadim, I. T., Slozhenkina, M. I., Gorlov, I. F., Seidavi, A., & Phillips, C. J. C. (2022). The effects of green tea in the diet of broilers challenged with coccidiosis on their performance, carcass characteristics, intestinal mucosal morphology, blood constituents and ceca microflora. Veterinary Medicine and Science, 8, 2511–2520. https://doi.org/10.1002/vms3.923