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Evaluation of dried tomato pomace as a non-conventional feed: Its effect on growth, nutrients digestibility, digestive enzyme, blood chemistry and intestinal microbiota of growing quails

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Abstract

This work evaluated the effect of sun-dried tomato waste (SDTP) on growth, carcasses, digestibility of nutrients, digestive enzymes, blood chemistry and intestinal microbiota of 250 one-week-old growing Japanese quail. The birds were randomly distributed into 5 equal groups. Group 1 was fed on the basal diet (BD) (control), Groups 2, 3, 4 and 5 were fed on the BD containing 3, 6, 9 and 12% SDTP respectively. SDTP significantly improved the body weights and gains at 3 weeks of age compared with control, and the 6% SDTP level seems to be the optimal level used throughout the first 3-5 weeks of age. With 6, 9 or 12% SDTP, the percentages of liver and giblets and the meat percentages of moisture and crude protein were improved significantly relative to control or 3%. In quail diets, the percentage of ether extract was reduced with increasing the SDTP. The studied levels of SDTP did not significantly change the digestibility of DM, OM, CP or NFE. The highest levels of amylase and lipase enzymes were recorded in the 6% SDTP, opposite to control. SDTP at 9 and 12% significantly decreased the total cholesterol than all other treatments. The highest and the lowest HDL levels were shown in 12 and 9% SDTP respectively. The 12% SDTP showed the highest SOD, TAC, IgM, IgG and Complement 3 values among all groups. The different levels of SDTP significantly reduced the MDA content than control. The highest caecal E. coli and Salmonella spp counts were found in control and 6% inclusion level followed by 9% and finally 3 and 12% SDTP. In conclusion, dietary inclusion of SDTP can enhance the growth and health status of growing quails via improving the nutrient digestibility, digestive enzymes, and blood chemistry and reducing the intestinal pathogens.

K E Y W O R D S

digestive enzymes, growth, intestinal microbiota, quails, SDTP

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1 | INTRODUCTION

Due to human competition and lower-than-expected production, the monogastric ration market consumes over than 60% of the annually produced soybean and corn meals, and the essential elements in these diets, accounting for 80% of the final prices (Alagawany & Attia, 2015; Borges Rodrigues et al., 2004). As a result, interest in alternative foods that may be utilized in monogastric diets and do not damage performance has grown. These foods might potentially replace soybean and corn meal in ration, lower the costs and ensure adequate nutritional values of the diet (Alagawany et al., 2019; Borges Soares et al., 2007).

Primary or industrial processing of food designed for human and animal use has resulted in a large number of wastes which, despite their pollutant potential, have nutritional value and can be used to build monogastric diets (Alagawany et al., 2020; Bertocco Ezequiel et al., 2006; Di Cerbo et al., 2014). These wastes have been fed to animals, primarily ruminants, in areas of plant processing, regardless the content of nutrients or the best amount of inclusion (Cavalcante Lira et al., 2010).

According to Cavalcante Lira et al. (2010) rising costs of cereals and imported feedstuffs for chicken diets prompted a quest for alternative components available as by-products from local agricultural sectors. Furthermore, Agro-Industrial By-Products (AIBP) are one of the most talented and prospective sources of energy and protein for cattle (Salajegheh et al., 2012). The use of AIBP as a component of animal feed has high value, starting with lower production costs and ultimately enhancing livestock farmers' profit margins.

Because there is limited literature regarding the performance and carcass yield evaluation following the use of tomato waste, its utilization in monogastric feeding is feasible, especially for broiler chicks. Persia et al. used tomato wastes at levels of 0%, 5%, 10%, 15% and 20% in comparison with corn and soy meal in broiler chickens aged 8–21 days and found that the fowls' performance was unaffected in terms of weight increase and feed efficiency up to 15% (Persia et al., 2003). According to Alshatwi et al. (2010) tomatoes and tomato by-products are great sources of protein, fat, vitamin E and minerals such as nitrogen, iron and zinc.

Although tomato wastes and by-products have been investigated in poultry industry to a limited extent, few studies have been conducted to evaluate their nutritive value, protein quality, nutrient digestibility, digestive enzyme, immunity and intestinal microbiota so far.

Thus, this study aimed to explore the influence of diets containing various levels (3 to 12%) of sun-dried tomato waste (SDTP) on growth, weight, nutrient digestibility, digestive enzymes, blood chemistry and intestinal microbiota in Japanese quail chicks.

2 | MATERIALS AND METHODS

2.1 | Antimicrobial activity of tomato waste

2.1.1 | Disc diffusion assay

The disc diffusion method was used to measure the antibacterial and antifungal activities of tomato peels (Gulluce et al., 2007). 10 g of tomato peels were stirred for 1 h in 90 ml of sterilized distilled water to obtain a 1000 mg/ml concentration (stock solution), and then, 8 ml of the stock solution was diluted with 2 ml of sterilized distilled water to obtain an 800 mg/ml concentration; then, 6 ml of the stock solution was diluted with sterilized distilled water (4 ml) to obtain a 600 mg/ml concentration. The discs were drenched in sterilized distilled water as a control. The plates were cultured at 37°C for a day (bacteria) and at 28°C for five days (Candida and fungi). All of the experiments were carried out in triplicates. By using a ruler, the inhibition zones (mm) were measured (El-Saadony et al., 2021a; Hassan & Ullah, 2019).

Minimum bactericidal concentration (MBC) and minimum fungicidal concentration (MFC) were calculated according to the European Committee on Antimicrobial Susceptibility Testing, while the minimum inhibitory concentration (MIC) was calculated using the microdilution broth method. In test tubes containing 9 ml of Mueller Hinton broth (bacteria), Sabourard dextrose broth (Candida) and potato dextrose broth, 50 L of tomato peel extract concentrations (200, 400, 600, 800 and 1000 g/ml) were added (fungi). The tubes were filled with 100 L of bacterial and Candida inoculum (1.5108 CFU/ml) and the standard size of fungal spore solution (3103 CFU/l).

The test bacteria were introduced into the broth medium in the control tubes. The MHB and SDB tubes (bacteria and Candida) were cultured at 37°C for 24 h, whereas the PDB tubes were incubated for five days at 28°C (fungi). The minimum inhibitory concentration (MIC) of tomato peel extract that suppresses microbiological growth was determined.

The minimum bactericidal concentration (MBC) was the lowest concentration that could kill bacteria, whereas the minimum fungicidal concentration (MFC) was the lowest concentration that could kill Candida and fungi (Clsi, 2013; El-Saadony et al., 2019; Reda et al., 2021). The MFC and MBC were calculated using the spread plate method, which involved in spreading a loopful of

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MIC tubes on new MHA, SDA and PDA plates, incubating at 37°C for 24 h (bacteria, Candida), and for 5 days at 28°C (fungi), and the microbial growth was observed (El-Saadony, Sitohy, et al., 2021b; Usman et al., 2014).

2.2 | Dietary microbial count

A 10^{-1} dilution of the supernatant was obtained. From prior dilutions, serial decimal dilutions were made up to 10^{-7} . 1 ml of each dilution was placed on sterilized plates, and then, appropriate media were added to count the bacteria in the food samples. After a 48-h incubation period at 37°C, the total bacterial count was determined on plate count agar. After five days of incubation at 25°C, the total yeast and mould count was determined on Rose Bengal Chloramphenicol agar.

After a 24-h incubation period at 37°C, total coliforms were counted on Violet Red Bile Agar (Biolife, Monza, Italy). After a 24-h incubation period at 37°C, *Escherichia coli* were counted on Tryptone Bile Glucuronide Agar. To make statistical analysis easier, the microbial count was transformed into log CFU (Alagawany et al., 2021).

2.3 | Animal husbandry and experimental design

A total of 250 one-week-old Japanese quails were employed in the experiment. The effects of varying dietary SDTP levels (0, 3, 6, 9 and 12%) on growth, carcasses, digestibility of nutrients, digestive enzymes, blood chemistry and intestinal microbiota of growing quails were investigated at the Poultry Department, Agriculture Faculty, Zagazig University, Zagazig, Egypt. The birds were divided into five treatment groups, each with 50 birds in five replicates of ten quails each.

For evaluating the five meals that incorporated into the experimental treatments, the chicks were distributed in a complete randomized design. The experimental meals were isoenergetic and isonitrogenous, based on maize and soybean meal, and included 0, 3, 6, 9 and 12 percent of tomato waste respectively (Table 1).

Quails were grown in a standard cage (50 cm x 30 cm x 50 cm of floor space) with free access to feed and water. During the trial, quails were subjected to a 17-h light/ seven-hour dark cycle. The experimental procedures were performed according to the guidelines set out by the Local Experimental Animal Care Committee. ZU–IACUC/2/F/56/2021 is the ethical approval code.

The tomato pomace was obtained from commercial processors (Hienz Company, 6th October City, Egypt). It was spread out on a plastic sheet and exposed to sunlight to dry. According to Yitbarek, waste particle size is reduced by pounding with a stick and hand crushing (Yitbarek, 2013).

2.4 | Meat composition, carcass characteristics and growth

Individual weights of quail were recorded at Weeks 1, 3 and 5 in order to compute body weight (g) and weight gain (g/day) over the course of the experiment. Throughout the trial, feed intake (g) was monitored and converted to g feed/g gain. At 35 days, 5 birds were chosen randomly from each group and slaughtered for carcass evaluation (Reda, Alagawany, et al., 2020; Reda, El-Saadony, et al., 2020).

Following treatment, the carcasses were chilled for 12 h at 1°C before being split into several pieces, including the breast and thigh. These pieces were kept frozen at -20 °C until they were analysed. The flesh from frozen pieces was removed from the bones and homogenized, and the crude fat, crude protein, moisture and ash content were all measured (AOAC, 2006).

2.5 | Digestibility trail

To measure the digestibility of each dietary item and the metabolizable energy (ME), another ten five-week-old quails were recruited. The quails were kept in separate cages and provided with appropriate experimental meals. The birds were given three days to adapt before being fed and having their excrement collected every 24 h for five days. After collecting the excreta, all quail feathers were removed, and the faecal samples were cleaned, weighed and dried for 36 h in ovens at 70°C.

Diet and faecal analyses were performed in accordance with AOAC guidelines (AOAC, 2006). ME was used to calculate nutritive values. The ME was 4.2 Kcal per gram TDN recommended by Titus (Titus, 1941).

2.6 | Digestive enzymes

At the end of the experiment, the activity of digestive enzymes (amylase, protease and lipase) was determined in the ileum of the birds (1 bird per replicate). The quail ileum was dissected from the Meckel's diverticulum to two centimetres above the ileocaecal junction, and the ileal contents were aseptically collected in screw-capped sterile specimen vials.

The activities of digestive enzymes were assessed according to the method of Najafi et al. (Najafi et al., 2005, 2006).

	Sun-dried to	omato waste	e (%-SDTW)		
Ingredient	Control	3	6	9	12
Maize 8.5%	51.80	50.50	47.90	46.4	44.6
Soybean meal 44%	36.70	33.11	32.04	29.34	26.80
Gluten meal 62%	5.21	7.00	7.00	8.00	9.00
Soybean oil	2.90	3.00	3.70	3.90	4.20
Limestone	0.70	0.70	0.70	0.70	0.70
Di-calcium phosphate	1.65	1.65	1.60	1.55	1.55
Salt	0.30	0.30	0.30	0.30	0.30
Premix ^a	0.30	0.30	0.30	0.30	0.30
L-Lysine	0.13	0.13	0.15	0.20	0.25
Dl-Methionine	0.11	0.11	0.11	0.11	0.10
Choline chloride	0.20	0.20	0.20	0.20	0.20
SDTW (22.70% CP)	0.00	3.00	6.00	9.00	12.00
Calculated composition ^b	(%)				
ME (Kcal/kg)	2999	2999	2999	2997	2977
Crude protein	24.04	24.04	24.00	23.99	24.01
Calcium	0.80	0.80	0.800	0.80	0.80
Nonphytate P	0.45	0.45	0.45	0.45	0.45
Lysine	1.30	1.30	1.27	1.27	1.27
TSAA	0.91	0.91	0.91	0.92	0.92
Analyzed ^c (as DM ^d)					
Crude protein	23.99	24.02	23.95	24.09	24.06
Fibre	3.78	4.50	5.36	6.13	6.91
Fat	5.29	5.55	6.38	6.75	7.21

^aProvides per kg of diet: vitamin A, 12,000 I.U; vitamin D3, 5000 I.U; vitamin E, 130.0 mg; vitamin K3, 3.605 mg; vitamin B1 (hiamin), 3.0 mg; vitamin B2v (riboflavin), 8.0 mg; vitamin B6, 4.950 mg; vitamin B12, 17.0 mg; niacin, 60.0 mg; D-Biotin, 200.0 mg; calcium D-pantothenate, 18.333 mg; folic acid, 2.083 mg; manganese, 100.0 mg; iron, 80.0 mg; zinc, 80.0 mg; copper, 8.0 mg; iodine, 2.0 mg; cobalt, 500.0 mg; and selenium, 150.0 mg.

^bCalculated according to NRC (1994).

^cAccording to AOAC (2006).

^dDry matter.

2.7 | Blood biochemistry

At 5 weeks of age, 10 birds from each group were chosen randomly, weighed and slaughtered to collect blood samples in test tubes containing EDTA. The test tubes were then gently shaken to combine the anticoagulants and the blood. Isolated plasma was obtained by centrifuging the whole blood at 3000 rpm for 20 minutes and then stored at -20 °C until analysis.

Total protein (g/dl) was determined according to the method proposed by Armstrong and Carr (Armstrong & Carr, 1965). Albumin concentrations (g/dl) were calculated using a calorimetric method. The globulin concentrations (g/dl) were calculated by subtracting albumin concentrations from total protein concentrations. Triglycerides and

total cholesterol were measured according to the method proposed by Allain et al. (1974).

High-intensity lipoprotein levels (HDL) and lowdensity lipoprotein (LDL) were measured according to the methods proposed by Cooper et al. (1988) and Friedewald et al. (1972) respectively. The enzymes in the liver were also studied. The activities of amylase, lipase and protease enzymes were determined according to Somogyi (1960), Tietz and Fiereck (1966) and Lynn and Clevette Radford (1984), respectively.

The total antioxidant capacity (TAC) was assessed as described in Koracevic et al. (2001). Superoxide dismutase (SOD) was measured spectrophotometrically following the method of Nishikimi et al. (1972). Malondialdehyde (MDA) was measured according to the method proposed

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by Mihara and Uohiyama (1978). IgG was estimated according to the method proposed by Bianchi et al. (1995).

2.8 | Microbiological analysis

At the end of the experiment, samples (about 10 g) were collected from the quail caecum (five samples /treatment) and transferred to a 250 ml Erlenmeyer flask containing 90 ml of 0.1% peptone in saline solution (0.85% NaCl) and mixed thoroughly. Total bacteria, total yeasts and moulds, *Salmonella*, *E. coli*, Enterococcus and coliforms counts were all assessed using the methods of Reda, Alagawany, et al. (2020) and Reda, El-Saadony, et al. (2020).

2.9 | Statistical analysis

All microbiological experiments have been done in triplicate according to Steele et al. (1997). The data were analysed using SAS (SAS, 2002) and a o ne-way analysis of variance (ANOVA) with the least significant difference (LSD) at a level of probability of *p < 0.05. SAS software (Cary, NC, USA) was used to carry out all of the statistical approaches.

The Tukey's test was used to analyse all data (performance, carcass, blood metabolites, immunity, digestibility of nutrients, digestive enzymes, antioxidants and caecal microbiota) (*p < 0.05).

3 | **RESULTS AND DISCUSSION**

3.1 Antibacterial activity of SDTP

Table 2 demonstrates that the widths of inhibitory zones grew significantly in a concentration-dependent manner. The inhibition diameter zones (IDZs) ranged from 9.2 to 28.5 mm for gram-positive (G+) bacteria, 7.8–24.2 mm for gram-negative (G-) bacteria, and 7.2–21.9 mm for the studied fungi.

The most resistant G+ and G-bacteria to tomato peel concentrations were *Staphylococcus aureus* and *Salmonella Typhi*, whereas the most sensitive bacteria were *Streptococcus pyogenes* and *Escherichia coli*. On the contrary, *Candida glabrata* was the most resistant fungus, while *Aspergillus flavus* was the most sensitive. The examined bacteria were inhibited in the range of 130–170 g/ ml, and the tested fungi were inhibited in the range of 140–165 g/ml.

As shown in Table 3, the microbial count in diet samples was significantly lower in the treated groups than in the control group ($p^* < 0.05$).

The dietary group supplemented with 12% tomato peel powder, on the contrary, outperformed the other treatment groups in terms of microbial count reduction in a time-independent way. In comparison with control, the addition of tomato peels considerably reduced microbial count in diet samples, with a relative decrease of 20% in total bacterial count (TBC), 13% in total yeasts and moulds count (TYMC), 18% in *Escherichia coli* count and 25% in coliform count.

3.2 | Effect on growth

Table 4 summarizes the effect of dietary SDTP inclusion on the growth performance indicators of Japanese quail chicks.

The results showed that the use of SDTP in the diet of quail chicks resulted in a significant (p < 0.05) improvement in body weight and weight gains at 3 weeks of age when compared to those fed a control diet. While using 6% SDTP seems to be the optimal level to use throughout the first 3–5 weeks of life, it resulted in the highest body weight gain when compared to other levels (3, 9 and 12%).

3.3 Effect on carcasses and meat analysis

The effects of SDTP on carcass characteristics and meat composition are shown in Table 5.

Apart from the percentages of liver and giblets, there were no significant variations in carcass features among treatment groups. However, with 6, 9 or 12% SDTP, the percentages of liver and giblets were significantly improved ($p^* < 0.05$) as compared to control or 3%.

Apart from ash, the percentages of moisture and crude protein in meat were enhanced (p = 0.0366 and 0.0172) with 6, 9 or 12% SDTP compared with control or 3% (p = 0.0366 and 0.0172). In quail diets, the percentage of ether extract was reduced with increasing the level of SDTP.

3.4 Effect on digestion coefficients and digestive enzymes

Data in Table 6 represent the effect of different dietary levels of tomato waste meal on digestion coefficient and nutritive values of Japanese quail.

There was no significant change in the digestibility of DM, OM, CP or NFE following the inclusion of tomato waste in the quail diet at different percentages. On the contrary, the other values were highly affected and inclusion of tomato waste at 6% achieved the highest digestibility of

TABLE 2 Antimicrobial activity of tomato peels aqueous extract concentrations against tested bacteria and fungi

	Items	Toma	to waste con	centrations	(µg/ml)				
	Microorganisms	200	400	600	800	1000	<i>p</i> value	MIC	MBC
G+	Bacillus cereus	9.8e	12.3d	16.5c	21.6b	26.8a	0.001	140b	280b
	Staphylococcus aureus	10.7e	14.4d	17.8c	22.7b	28.5a	0.006	130c	260c
	Streptococcus pyogenes	9.2e	11.8d	15.3c	19.6b	25.8a	0.008	160a	320a
	Main effect	9.9E	12.8D	16.5C	21.3B	27.0A			
	<i>p</i> value			0.004				>0.001	>0.001
G-	Escherichia coli	7.8e	10.7d	13.6c	17.4b	22.6a	0.008	170a	340a
	Salmonella Typhi	8.6e	11.6d	14.3c	18.5b	24.2a	0.01	160b	320b
	Klebsiella pneumoniae	8.4e	10.4d	13.8c	17.6b	23.3a	0.007	165ab	230c
	Main effect	8.3E	10.9D	13.9C	17.8B	23.4A			
	<i>p</i> value			0.009				>0.001	>0.001
								MIC	MFC
Fungi	Candida tropicalis	8.2a	10.3b	13.4b	17.2b	20.8b	0.0008	150bc	300b
	Candida albicans	7.5b	9.8c	12.3c	16.5bc	19.6c	0.004	160ab	320a
	Candida glabrata	8.7a	11.2a	14.6a	18.2a	21.9a	0.0001	140c	280c
	Aspergillus flavus	7.2c	9.4c	12.1c	15.8c	17.9d	0.005	165a	320a
	Fusarium oxysporum	7.5b	9.9c	13.4b	16.8bc	19.6c	0.001	155b	300b
	Aspergillus niger	7.6b	10.3b	13.8b	17.5b	20.5b	0.0008	150bc	300b
	Main effect	13.98E	13.14D	14.92C	12.48B	13.44A			
	<i>p</i> value			0.0007				>0.001	>0.001

Note: Mean values with different lowercase in the same raw indicate significant difference between tomato waste concentrations at $p^* < 0.05$.

EE, CP, TDN and ME among all other groups, while the lowest values were recorded by the control group.

Regarding the digestive enzymes, the effect of dietary tomato waste meal on digestive enzymes of Japanese quail is illustrated in Table 7.

Results showed that the highest levels of amylase and lipase enzymes were recorded in the 6% tomato waste meal, while the control group showed the lowest levels compared with other groups. On the contrary, protease activity did not significantly differ among the different dietary treatments.

3.5 | Effect on haematobiochemical parameters

The results in Table 8 show that the total protein content (TP) did not significantly change among the different dietary treatment groups except for 12% tomato waste meal that showed the highest TP content.

The 12% tomato waste meal also recorded the highest ALB level among other groups, and the lowest level was

shown in control. The highest GLOB level was observed also in 12% tomato waste meal followed by 3% compared with control and other groups which had comparable values, while the A/G ratio was significantly higher in 6 and 9% tomato waste meal compared with other groups which were comparable to the control.

Data also revealed that the highest ALT activity was observed in 3 and 9% tomato waste meal, while inclusion at 6% showed the lowest values. The 9% tomato waste meal also showed the highest AST value followed by 3%, 12% and control and finally the 6% tomato waste which showed the lowest value.

The highest level of creatinine was found in 3% tomato waste meal group and control, while the lowest value was recorded in 6% meal. On the contrary, the urea level did not significantly differ by the different dietary inclusion levels of tomato waste.

The inclusion of tomato waste at 9% and 12% significantly decreased the total cholesterol than control and other treatments, while the highest triglyceride level among different groups was obtained in 3% tomato waste meal. The highest and the lowest HDL levels were shown in 12% and

Conc. (µml)	TBC by	weekly						TYMC by	weekly				
Treatments	Fresh	1 week	2 weel	ζ 3 we	ek 4	week	5 week	Fresh	1 week	2 week	3 week	4 week	5 week
Control	5.47 ^a	5.55 ^a	5.73 ^a	5.98 ^a	6.	13a	6.21 ^a	3.13^{a}	3.19^{a}	3.27^{a}	3.39 ^a	3.55 ^a	3.67^{a}
3% Mix	5.29 ^b	5.36^{b}	5.45 ^b	5.58 ^b	5.	72b	5.85 ^b	3.07^{ab}	3.14^{ab}	3.22^{ab}	3.31^{ab}	3.42 ^b	3.59 ^b
6% Mix	5.19 ^{bc}	5.28^{b}	5.39 ^{bc}	5.48 ^c		61c	5.72 ^c	2.92 ^b	3.08^{b}	3.12 ^b	3.21 ^b	3.34°	3.45 ^c
9% Mix	5.05 ^c	5.12 ^c	5.19 ^c	5.26 (cd 5.	38d	5.43 ^d	2.83 ^c	2.92^{bc}	3.07 ^{bc}	3.12 ^c	3.22 ^d	3.38 ^d
12% Mix	4.87 ^c	4.95 ^c	5.04^{d}	5.13 ^d	5.	21 ^e	5.33 ^e	2.76 ^d	2.83 ^c	2.91 ^c	3.04^{d}	3.11^{e}	3.26 ^e
<i>p</i> value	0.008	0.01	0.01	0.000	1 0.	008	0.02	0.02	0.01	0.02	0.001	0.003	0.007
Conc. (mg/100 m	(I)	Coliform cou	unt by week					Esche.	richia coli cou	unt by week			
Treatments		Fresh	1w	2w	3w	4w	5w	Fresh	1w	2w	3w	4w	5w
Control		1.61^{a}	1.68^{a}	1.75 ^a	1.87^{a}	1.93^{a}	2.06 ^a	1.34^{a}	1.39^{a}	1.46^{a}	1.52^{a}	1.59 ^a	1.63^{a}
3% Mix		1.52^{ab}	1.59^{b}	1.67^{b}	1.74^{b}	1.82 ^b	1.92 ^b	1.27 ^b	1.31 ^b	1.38^{b}	1.44^{b}	1.52 ^{ab}	1.61 ^{ab}
6% Mix		1.42 ^b	1.51^{bc}	$1.61^{\rm bc}$	1.69 ^c	1.77^{c}	1.85 ^c	$1.21^{\rm bc}$	1.26^{bc}	1.33^{bc}	$1.42^{\rm bc}$	1.50^{b}	1.57^{b}
9% Mix		1.33°	1.41 ^c	1.53 ^c	1.63 cd	1.69^{d}	1.75 ^d	1.16°	1.19 ^c	1.25 ^c	1.31°	1.37^{c}	1.45 ^c
12% Mix		1.25 ^d	1.37^{d}	1.43 ^d	1.52 ^d	1.61 ^d	1.67 ^e	1.11 ^d	1.17 ^d	1.21 ^d	1.27^{d}	1.33 ^d	1.39 ^d
<i>p</i> value		0.001	0.01	0.006	0.02	0.005	0.002	0.001	0.02	0.008	0.03	0.04	0.02
Note: Mean values with	h different l	owercase in the	same column i:	ndicate signific	sant difference	e at $p^* < 0.0$.	5.						

centrations on total bacterial count, total veasts and moulds, coliform and *Eschenichia coli* count in diet 000 TABLE 3 Effect of tomato peels extract mixture 7 of 15

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FABLE 4	Japanese quail's growt	n performance variation	according to different	t dietary tomato waste	meal percentages
		*			

	Tomato waste	meal level (%)					
Items	0	3	6	9	12	SEM	p value
Body weight (g)							
1 week	29.25	29.36	29.45	29.27	29.55	0.226	0.9355
3 week	102.76 ^c	114.55 ^a	105.66 ^{bc}	110.00^{ab}	112.45 ^a	1.369	0.0023
5 week	211.36	213.5	215.37	211.5	217.75	2.180	0.3151
Body weight gain (g/	/day)						
1–3 week	5.25 ^c	6.08 ^a	5.44 ^{bc}	5.77 ^{ab}	5.92 ^a	0.088	0.0019
3–5 week	7.76 ^{ab}	7.07 ^d	7.84 ^a	7.25 cd	7.52 ^{bc}	0.084	0.0006
1–5 week	6.50	6.58	6.64	6.51	6.72	0.077	0.3748
Feed intake (g/day)							
1–3 week	13.39 ^c	15.59 ^b	14.04 ^c	16.93 ^{ab}	17.45 ^a	0.472	0.0006
3–5 week	25.56 ^d	27.50 ^c	26.80 cd	30.15 ^b	31.83 ^a	0.340	< 0.0001
1–5 week	19.48 ^c	21.54 ^b	20.42 ^{bc}	23.54 ^a	24.64 ^a	0.318	< 0.0001
Feed conversion rati	o (g feed/g gain)						
1–3 week	2.55 ^b	2.56 ^b	2.58 ^b	2.94 ^a	2.95 ^a	0.084	0.0169
3–5 week	3.30 ^c	3.89 ^b	3.42 ^c	4.16 ^a	4.23 ^a	0.046	< 0.0001
1–5 week	2.99 ^c	3.28 ^b	3.07 ^c	3.62 ^a	3.67 ^a	0.057	< 0.0001

Note: Means in the same raw with no superscript letters after them or with a common superscript letter following them are not significantly different (p < 0.05).

TABLE 5 Japanese quail's carcass traits and meat quality variation according to different dietary tomato waste meal percentages

	Tomato wast	e meal level (%)	1				
Items	0	3	6	9	12	SEM	p value
Carcass %	71.79	74.84	73.48	71.19	74.93	1.140	0.1594
Liver %	2.14 ^b	2.12 ^b	2.33 ^{ab}	2.53 ^a	2.56 ^a	0.091	0.0171
Gizzard %	1.92	1.84	2.48	2.12	2.19	0.155	0.1806
Heart %	0.87	0.85	0.91	0.88	0.95	0.094	0.9445
Giblets %	4.93 ^{bc}	4.81 ^c	5.72 ^a	5.53 ^{ab}	5.69 ^a	0.170	0.0274
Dressing %	76.72	79.65	79.2	76.72	80.62	1.222	0.1618
Chemical body compositi	on						
Moisture %	71.10 ^c	71.83 ^{bc}	72.88 ^{ab}	73.45 ^a	72.63 ^{ab}	0.459	0.0366
CP %	20.70 ^c	21.61 ^{bc}	22.26 ^{ab}	22.57 ^{ab}	23.35 ^a	0.432	0.0172
Ether extract %	14.20 ^a	13.49 ^b	12.98 ^c	10.92 ^e	11.17 ^d	0.059	< 0.0001
Ash %	3.61	3.32	3.45	3.18	3.33	0.155	0.4419

Note: Means in the same raw with no superscript letters after them or with a common superscript letter following them are not significantly different (p < 0.05).

9% tomato waste meals, respectively, compared with control and other groups, while the highest LDL value was observed in control and it was decreased by increasing the inclusion level of tomato waste to be the lowest in 12% group.

3.6 | Effect on immunity, antioxidants and oxidative stress biomarkers

The VLDL level did not significantly change among different treatments except in 3% inclusion level which was significantly higher than the others.

The present study showed that 12% tomato waste meal showed the highest IgG, IgM and complement 3 levels among all studied groups, while the different inclusion

TABLE 6 Japanese quail's digestion coefficient and nutritive value variation according to different dietary tomato waste meal percentages

	Tomato waste m	eal level (%)					n
Items	0	3	6	9	12	SEM	<i>p</i> value
DM	78.25	79.96	82.81	81.05	82.24	1.394	0.2337
OM	80.59	81.9	84.65	82.75	84.39	1.352	0.2604
СР	86.13	86.36	85.88	87.05	86.60	0.522	0.5989
EE	71.19 ^c	74.78 ^b	79.50 ^a	76.81 ^b	77.05 ^b	0.721	0.0001
CF	25.52 cd	26.72 ^{bc}	29.40 ^a	24.53 ^d	27.87 ^{ab}	0.580	0.0015
NFE	78.27	79.9	83.91	80.26	83.34	1.655	0.1734
TDN	71.69 ^c	73.72 ^{bc}	78.07 ^a	73.33 ^{bc}	76.39 ^{ab}	1.284	0.0372
ME	3011 ^c	3096 ^b	3278 ^a	3080 ^b	3208 ^{ab}	53.94	0.0372

Note: Means in the same raw with no superscript letters after them or with a common superscript letter following them are not significantly different (p < 0.05).

Abbreviations: CF, rude fibre; CP, crude protein; DM, dry matter; EE, ether extract; ME, metabolic energy; NFE, nitrogen free extract; OM, organic matter; TDN, total digestible nutrients.

TABLE 7 Japanese quail's digestive enzyme (unit) variation according to different dietary tomato waste meal percentages

	Tomato waste	meal level (%)					n
Items	0	3	6	9	12	SEM	value
Amylase	5.45 ^d	11.41 ^c	19.51 ^a	13.79 ^{bc}	16.51 ^{ab}	1.415	0.0005
Lipase	6.04 ^c	13.58 ^b	25.30 ^a	4.30 ^c	15.20 ^b	1.704	0.0001
Protease	0.57	0.64	0.63	0.42	0.95	0.120	0.1022

Note: Means in the same raw with no superscript letters after them or with a common superscript letter following them are not significantly different (p < 0.05).

levels did not significantly change the lysozyme activity compared with control (Table 8).

Regarding the effect of different dietary treatments on the antioxidants of Japanese quails, the highest SOD activity was observed in 12% inclusion level followed by 6 and 9% and the lowest activity was found in control and 3% tomato waste meal. However, the different inclusion levels did not significantly influence the GPx activity. Furthermore, the results of oxidative stress biomarkers showed that the different levels of tomato waste have similar reducing effect on the level of MDA compared with control. On the contrary, 12% inclusion level showed the highest TAC level followed by other treatments and the lowest TAC was observed in control. Tomatoes and tomato by-products are great sources of iron, zinc, carotene and lycopene with some of them having antioxidant characteristics.

3.7 Effect on caecal microbiota

Bacteriology of Japanese quail as affected by dietary tomato waste meal was represented in Table 9.

The 6% inclusion level and control groups showed the highest total bacterial count, while the lowest one was

recorded in 3% inclusion group. The highest total yeasts and moulds count was found in control group followed by 6%, then 3 and 9%, and finally 9% which was the lowest one. The highest *Escherichia coli* and *Salmonella* spp counts were found in control and 6% inclusion level followed by 9%, and finally 9 and 12% tomato waste meals that showed the lowest count. The *Enterococcus* spp showed the highest count in control group followed by 3% tomato waste meal group compared with other treatments which were comparable to each other. Additionally, the highest coliform count was observed in control and 6% tomato waste groups, while the lowest count was in the 12% tomato waste inclusion group.

4 | DISCUSSION

According to Gaafar et al., (2015) tomato pomace extract had an excellent antibacterial activity against grampositive bacteria and yeast, with MICs ranging from 147 to 154 ppm. The manufacture of protein in *Bacillus subtilis* cells was inhibited by aqueous extract of tomato pomace. The aqueous extract of tomato pomace was more effective than the ethanol and acetone extracts against bacteria, fungi and yeasts. With 600 ppm, the highest TABLE 8 Japanese quails haematobiochemical parameter variation according to different dietary tomato waste meal percentages

	Tomato waste r	neal level (%)					
Items	0	3	6	9	12	SEM	P value
TP (g/dL)	2.97 ^b	3.49 ^b	3.16 ^b	3.14 ^b	4.01 ^a	0.150	0.0057
ALB (g/dL)	1.61 ^d	1.83 ^c	2.11 ^{ab}	1.98 ^{bc}	2.23 ^a	0.045	< 0.0001
GLOB (g/dL)	1.36 ^{bc}	1.66 ^{ab}	1.05 ^c	1.16 ^c	1.78 ^a	0.111	0.0049
A/G (%)	1.18 ^b	1.11 ^b	2.08 ^a	1.77 ^a	1.26 ^b	0.116	0.0042
ALT (IU/L)	8.60 ^{ab}	9.97 ^a	5.25 ^c	11.29 ^a	5.76 ^{bc}	0.857	0.004
AST (IU/L)	160.05 ^c	202.55 ^b	124.85 ^d	222.60 ^a	177.25 ^c	5.889	< 0.0001
Creatinine (mg/dl)	0.61 ^a	0.63 ^a	0.41 ^c	0.46 ^{bc}	0.57 ^{ab}	0.039	0.0259
Urea (mg/dl)	5.94	6.32	4.57	7.35	4.19	0.775	0.0932
TC (mg/dl)	247.79 ^a	232.41 ^a	230.57 ^a	196.40 ^b	210.60 ^b	5.694	0.0011
TG (mg/dl)	255.60 ^b	309.18 ^a	272.60 ^b	247.95 ^b	253.50 ^b	8.461	0.0041
HDL (mg/dl)	38.55 ^{bc}	30.69cd	42.81 ^{ab}	27.27 ^d	48.19 ^a	2.158	0.0009
LDL (mg/dl)	158.12 ^a	139.89 ^{ab}	133.24 ^{bc}	119.55cd	111.71 ^d	5.881	0.0021
VLDL (mg/dl)	51.12 ^b	61.84 ^a	54.52 ^b	49.59 ^b	50.70 ^b	1.692	0.0041
IgG (mg/dl)	0.88 ^c	1.04 ^{bc}	1.14 ^{ab}	1.02 ^{bc}	1.31 ^a	0.068	0.0182
IgM (mg/dl)	0.70 ^{bc}	0.77 ^b	0.76 ^b	0.51 ^c	1.00^{a}	0.056	0.0038
Lysozyme (U/ml)	0.43	0.42	0.39	0.44	0.51	0.044	0.5526
Complement 3	119 ^b	59 ^c	74 ^c	67 ^c	153 ^a	7.034	< 0.0001
SOD (U/ml)	0.35 ^c	0.32 ^c	0.49 ^b	0.50 ^b	0.71 ^a	0.027	< 0.0001
GPx (ng/ml)	0.42	0.38	0.4	0.39	0.51	0.042	0.2532
MDA (nmol/ml)	0.37 ^a	0.20 ^b	0.22 ^b	0.24 ^b	0.15 ^b	0.031	0.0103
TAC (ng/ml)	0.21 ^c	0.36 ^b	0.32 ^b	0.33 ^b	0.49 ^a	0.029	0.0014

Note: Means in the same raw with no superscript letters after them or with a common superscript letter following them are not significantly different (p < 0.05).

Abbreviations: A/G, albumin/globulin ratio; Alb, albumin; ALT, alanine aminotransferase; AST, aspartate aminotransferase; GLOB, globulin; GPx, glutathione peroxidase; HDL, high density lipoprotein; IgA, immunoglobulin A; IgG, immunoglobulin G; IgM, immunoglobulin M; LDL, low-density lipoprotein; MDA, malondialdehyde; SOD, superoxide dismutase; TAC, total antioxidant capacity; TC, total cholesterol; TG, triglycerides; TP, total protein; VLDL, very low-density lipoprotein.

TABLE 9 Japanese quail's bacteriology (Log CFU/g) variation according to different dietary tomato waste meal percentages

	Tomato v	vaste meal lev	vel (%)				
Items	0	3	6	9	12	SEM	<i>p</i> value
Total bacterial count	6.76 ^a	5.85 ^c	6.59 ^a	6.45 ^{ab}	6.02 ^{bc}	0.158	0.0103
Total yeasts and moulds count	4. 78 ^a	4.18 ^{bc}	4.50 ^{ab}	4.29 ^{bc}	4.14 ^c	0.103	0.0076
E. coli	5.56 ^a	4.44 ^c	5.37 ^{ab}	5.17 ^b	4.46 ^c	0.106	< 0.0001
Salmonella spp	3.86 ^a	2.12 ^c	3.52 ^{ab}	3.17 ^b	2.16 ^c	0.118	< 0.0001
Enterococcus spp.	5.70 ^a	5.09 ^b	4.63 ^c	4.56 ^c	4.66 ^c	0.110	0.0002
Coliform	6.14 ^a	4.93 ^{bc}	5.91 ^a	5.23 ^b	4.76 ^c	0.097	< 0.0001

Note: Means in the same raw with no superscript letters after them or with a common superscript letter following them are not significantly different (p < 0.05).

inhibitory zone was obtained against *Staphylococcus aureus* (20.23 mm).

Hassanien et al. (2020) discovered that the tomato fruit aqueous extract demonstrated more antibacterial activity at both the green and mature stages, as well as that gram-positive bacteria such as *Bacillus thuringiensis* were more sensitive than gram-negative bacteria such as *Escherichia coli*, and that the MIC of tomato fruit extract against *Bacillus thuringiensis* was 200 and 4 at both the green and mature stages.

Tomato peels' high phenolic content had an antagonistic effect on the bacterial load in the diet. The antibacterial activity of tomato peel extract is attributed to polyphenols, which may be due to interactions between polyphenols and a variety of cell membrane locations (Bouarab-Chibane et al., 2019; Cushnie & Lamb, 2011; Sikkema et al., 1995).

Persia et al. (2003) employed tomato residues at levels of 0%, 5%, 10%, 15% and 20% in relation to corn and soy meal and found that the fowls' performance was unaffected in terms of weight gain and feed efficiency up to 15%.

When compared to the control group, chicks fed a diet containing various levels of SDTP had considerably higher feed consumption over all study periods. However, owing to the best feed conversion ratio compared with other levels, the addition of SDTP in quail diets up to 6% seems to be the ideal amount. Our findings are consistent with those of a previous study, which found that tomato waste enhanced feed intake in chicken aged 1 to 7, 8 to 14 and 29 to 36 days (Cavalcante Lira et al., 2010).

The significant increase in liver and giblets coincides with that of Leke et al., who claimed that dietary tomato waste levels had little effect on the heart and gizzard (Leke et al., 2018).

However, they found that using of 12% tomato meal enhanced the final body weight, carcass, breast meat, wings, thighs and drumsticks. Yatbarek (2013) reported that carcass yield was higher for birds fed on 5% tomato pomace compared with other treatments, while dried tomato pomace up to 15% in the diet had no effect on the carcass features of broiler chicks (King & Zeidler, 2004).

The ether extract percentage reduction observed with the increase in the level of SDTP is supported by the results of Leke et al., (2018) who reported that meat moisture and crude protein percentages were much higher, whereas meat crude fat was significantly lower. Tomato pomace could be used up to 20% in broiler chicken diets, according to Squires et al. (1992).

Tomato pomace has been reported to have alpha-, beta-, gamma- and delta tocopherols with an antioxidant effect; therefore, adding dried tomato pomace (just separated tomato seeds) to broiler chicken meals results in a prolonged shelf life for poultry products. The findings of protein, fat, moisture and ash of meat in varied amounts of dried tomato pomace up to 10% were dramatically increased, according to Asadollahi et al. (2014).

When the analysed characteristics were compared, it was shown that raising the amount of dried tomato meal utilized by 12% boosted both water and protein content while lowering the meat fat content of the Japanese quail, resulting in higher overall consumer acceptability. In general, the treatment with 12% dried tomato meal was considered to be the best and was advised to be added to the quail diet based on the results of each of the five parameters.

Different tomato cultivars, growing circumstances and processing methods could explain the differences in CP, fat, Ca and P levels between our study and others (i.e. the number of seeds, pulp and skins in the waste by-product). Tuoxunjiang et al. (2020) found that DM digestibility in tomato group was significantly higher than in the control group, but CP digestibility was not affected by treatments. Also, Abdollahzadeh and Pirmohammadi (2010) reported that dry matter intake was increased when animals were fed diets containing mixed apple pomace and ensiled tomato at levels of 30 or 15%, respectively, in comparison with control.

The increase in dry matter intake can be explained by the high palatability of tomato by-product. On the contrary, So et al. (2019) stated that apparent digestibility, nutrients intake, DM, crude protein and organic matter digestibility were increased in animals fed rations containing tomato pomace. Diets containing lycopene such as tomatoes can improve the nutritive value of animal products and human health (Czauderna et al., 2020).

The activities of the intestinal enzymes have crucial effects on the life activities and animal's physiological processes (Di Cerbo et al., 2013; Cho et al., 2012). Sarker et al. (2020) found that dietary supplementation of lycopene significantly improved lipase and amylase activities in broiler chickens compared with the other group.

Depending on the different tomato waste meal percentage, different effects were recorded on blood biochemical parameters of quails, as observed by Rahmatnejad et al., (2009) that found that feeding dried tomato pomace to broiler chicks at inclusion rates of 16% and 24% enhanced mean HDL-cholesterol values and decreased serum cholesterol and LDL cholesterol. According to Moundras et al., (1997) crude fibre's ability to modify faecal excretion of cholesterol and bile acids may explain its serum cholesterol-lowering impact.

According to Burr et al., (1985) dietary fibre content and blood cholesterol levels have a negative relationship. It is likely that a larger quantity of dried tomato pomace in laying-hen diets is required for a significant reduction in blood cholesterol. According to Mahata et al., (2016) varying quantities of cooked tomato waste powder up to 12% in diets had no effect on total cholesterol, LDL cholesterol and HDL cholesterol in laying-hen blood serum, as well as the fat content of egg yolk.

Frederiksen et al., (2007) on the contrary, found that dietary supplementation with a lycopene-rich tomato extract had no effect on cholesterol and triglycerol levels in rabbit plasma. The effects of feeding dried tomato pomace to laying hens on plasma cholesterol and low-density

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lipoproteins were inconclusive (Nobakht & Safamehr, 2007).

The use of antioxidants produced from natural sources has been shown to quench free radicals, increase antioxidant enzyme activity and minimize lipid peroxidation. Dry tomato pomace supplementation at the level of 5 per cent improved growth performance, and the activities of SOD and GPx, while decreased HDL-cholesterol and triglyceride levels and MDA concentration in broilers subjected to heat stress (Hosseini-Vashan et al., 2016). Many activities, including scavenging reactive oxygen species (ROS) and upregulating the production of antioxidant enzymes such as SOD, GPx and CAT, have been shown to have significant potential for maintaining oxidative balance in the host body (Martinez et al., 2008). Since the late 1950s, researchers have been aware of the health-promoting and antioxidant properties of lycopene (Lauretani et al., 2008). Given the high concentration of conjugated dienes in lycopene, it has a scavenging potential that is twice that of-tocopherol and ten times that of alfa-carotene, making it a powerful antioxidant (Palozza et al., 2012). Sahin et al. (2008) found that supplementing Japanese quail raised under heat stress with lycopene-rich tomato powder increased feed intake, weight gain and decreased levels of MDA in the muscles, liver and serum by a significant amount. It has been furtherly observed that dietary lycopene had similar effects on antioxidant status and growth performance in Japanese quails that had been exposed to high temperatures (Sahin et al., 2006). For the same reason, dietary addition of lycopene-enriched tomato by-product at a level of 5% in feed improved overall growth performance and, even at lower levels (1%), increased total antioxidant capacity while simultaneously lowering MDA levels in broilers under heat stress (Selim et al., 2013).

As far as concerns the tomato lycopene supplementation, it is known to protect the animal gut from damage when it comes into contact with hazardous agents such as pathogens, poisons or any other foreign antigen during its lifetime. Anwar et al. revealed that supplementing lycopene along with vitamins A, C and E could help to prevent intestinal damage in rats exposed to radiation (Anwar et al., 2013). A number of previous researches (Saada et al., 2010; Yucel et al., 2016) have established the favourable effects of dietary supplementation of lycopene in animals' diets on the improvement of intestinal structure. Moreover, tomato pomace contains alpha-, beta-, gamma- and delta tocopherols with high antioxidant effects; therefore, tomato pomace could be used up to 20% in broiler chicken diets (Squires et al., 1992).

There are several possible mechanisms by which SDTP can improve growth rate and physiological indices: the first is through the enhancement of the antioxidant system due to the presence of phytochemical compounds, the second is through the reduction in pathogenic microbiota, and the third may be through the enhancement of immunity and the increase in feed efficiency. In a related study, Tuoxunjiang et al. reported that the serum IgG and IgA levels in tomato pomace group (10%) were markedly higher than in the control group (Tuoxunjiang et al., 2020).

The findings of this study support the use of SDTP, a novel feed additive derived from natural plants, in the production of broiler chickens. Using SDTP as a dietary supplement can improve the growing quail's performance, lipid profile, immunity and antioxidant indices, while decrease the intestinal infections, resulting in an overall improvement in their health state. SDTP could be a viable component of chicken feed. Consequently, tomato waste could be employed as a functional food ingredient and should be evaluated as a potential nutraceuticals resource.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest regarding the publication of this paper.

AUTHOR CONTRIBUTIONS

M.A. and F.M.R planned the research and supervised the project. M.A., M.T.E.S., F.M.R. and A.D.C. conducted the experimental work and analysed the data. M.A., M.T.E.S., T.K.E.R., M.M., A.R.L. and A.D.C wrote the manuscript with the input of all the other authors.

INSTITUTIONAL REVIEW BOARD STATEMENT

The experimental procedures were performed according to the guidelines set out by the Local Experimental Animal Care Committee. ZU–IACUC/2/F/56/2021 is the ethical approval code.

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