

Effects of dietary stevia and ginger extracts on fattening performance, organ weights and serum biochemical parameters in quails exposed to heat stress

T. Bayril*

Dicle University, Faculty of Veterinary Medicine, Department of Animal Science, 21280 Diyarbakir, Turkey

KEY WORDS: antioxidant status, heat stress, fattening performance, quails, organ weight

Received: 12 October 2022

Revised: 14 November 2022

Accepted: 13 January 2023

* Corresponding author:
e-mail: tbayril@hotmail.com

ABSTRACT. The aim of this study was to determine the effect of stevia and ginger extracts (SGE) on growth performance, serum biochemical parameters, organ weights and antioxidant activity in quails under heat stress (HS). A total of 180, 6-week-old male quails were divided into four groups, five quails each, with six replicates. The added extract doses were: 0 (control), 0.5 and 1.0% of the diet for treatments 1–6, respectively. The thermoneutral group (TN) was kept at a temperature of 21 ± 2 °C for 28 days, and the heat stress group (HS) was exposed to 21 ± 2 °C for 18 h and 33 ± 2 °C (10.00–16.00) for 6 h. HS exposure adversely affected performance parameters compared to quails kept at TN, as reflected by lower final body weight (FBW), body weight gain (BWG), cumulative feed intake (CFI) and cold carcass weight (CCW), as well as increased feed conversion ratio (FCR) ($P < 0.001$ and $P < 0.0001$, respectively). Raising dietary SGE level linearly increased FBW ($P < 0.011$), BWG ($P < 0.025$) and CCW ($P < 0.001$), while it decreased FCR ($P < 0.026$). Quails exposed to HS conditions had higher serum malondialdehyde (MDA) ($P < 0.0001$), creatine kinase (CK) ($P < 0.0001$), aspartate aminotransferase (AST) ($P < 0.006$) and triiodothyronine (T3) ($P < 0.0001$) levels than those reared under TN conditions. SGE supplementation decreased serum MDA ($P < 0.003$), glucose ($P < 0.0001$), CK ($P < 0.0001$), AST ($P < 0.0001$), T3 ($P < 0.001$) and thyroxine (T4) ($P < 0.024$) levels compared to the control groups. While heart and liver weights decreased ($P < 0.0001$; $P < 0.001$), an increase in testicular weight was recorded ($P < 0.0001$). The addition of SGE to the diet during periods of elevated environmental temperature can play an important role in reducing the negative effects of heat stress.

Introduction

In recent years, the progressing global warming and the negative impact of high environmental temperature on farm animals will intensify. It is known that poultry will be the most affected farm animal by the environmental temperature. Ventilation and cooling systems used in commercial poultry farming are designed to prevent heat stress, however, they are usually quite expensive. Free-range sys-

tems that have begun to be applied in recent years in laying hen breeding are at the higher level compared to those used in broiler rearing. It is very difficult to control the temperature, humidity and ventilation of the environment, in which broilers are housed in free-range systems. The use of these systems in broiler farming is both difficult and uneconomical. Therefore, it is necessary to develop alternative breeding programs for these systems (Pirgozliev et al., 2020).

Adding antioxidants to poultry diets and reducing the effect of raising environmental temperature with climate change has become an important issue. Increasing environmental temperature causes physiological and biochemical changes in birds and reduces feed consumption and growth performance (Attia et al., 2018). In addition, heat stress negatively affects meat oxidative state and aerobic metabolism, thereby deteriorating its quality (Kaplan and Köksal, 2021). Phytochemicals that are of natural origin, environmentally friendly and non-toxic are used to increase meat quality and growth performance of broiler chickens (Akbarian et al., 2016). Stevia (*Stevia rebaudiana*) is a plant that has been used for centuries as a natural sweetener in South America and is also currently applied in human diets (Lemus-Mondaca et al., 2012). Stevia and its extracts also show antimicrobial, diuretic, insulinotropic and hypotensive properties (Atteh et al., 2008). Ginger (*Zingiber officinale*) and its extracts have been extensively used in recent years in animal diets due to their antioxidant, hepatoprotective, antimicrobial, analgesic and anti-inflammatory effects. Most important bioactive components of ginger are phenolic compound known as gingerols (Alsherbiny et al., 2019).

In recent years, many studies have shown that the addition of stevia and ginger and their extracts to the diet positively affects many parameters such as heat stress (HS), growth performance, feed efficiency, blood serum levels, antioxidant status, as well as egg and meat yields (Habibian et al., 2016; Rehman et al., 2018; Xiong et al., 2022). The aim of this study was to determine the effect of stevia and ginger extracts (SGE) on fattening performance, serum biochemical parameters, organ weights and antioxidant activity in quails under HS.

Material and methods

Animal management and treatments

All procedures used in the present study were approved by the Institutional Animal Care and Use Committee at the Faculty of Veterinary Medicine, Dicle University (Diyarbakir, Turkey). Quails (*Coturnix coturnix japonica*) used in the study were purchased from the poultry unit at the Faculty of Veterinary Medicine, Dicle University (Diyarbakir, Turkey). One-day old male chicks (180) were randomly divided into six groups. The experiment was conducted in 6 replicated cages (60 × 120 × 30 cm), 5 birds each. During the first days, the birds were housed in a poultry room with a temperature of 35 ± 2 °C and 24-hour lighting period. Room temperature was

gradually reduced to 21 ± 2 °C until day 15. The birds were housed in two separate rooms at two different ambient temperatures during the 28-day trial. The thermoneutral (TN) and heat stress (HS) groups were kept at a temperature of 21 ± 2 °C for 28 days, and a long-term temperature of 21 ± 2 °C for 18 h, and 33 ± 2 °C (10.00–16.00) for 6 h, respectively. The relative humidity in the TN group was around 52%. The relative humidity in the HS group was maintained at 52 ± 2%. The illumination period in both groups was similar for 32 days (10–42 days) (18L:6D).

Animal feeding and data collection

All birds were fed the same basal diet for 42 days. Stevia (*S. rebaudiana* 20 000 mg/kg; Sigma-Aldrich, Interlab, Istanbul, Turkey) and ginger (*Z. officinale* 5 000 mg/kg; Calbiochem, Interlab, Istanbul, Turkey) extract mixtures (0, 0.5 and 1%) were added daily to the diets of each subgroup of the TN and HS groups. Feed and fresh water were offered *ad libitum* throughout the experiment. The nutritional and chemical compositions of the ration are given in Table 1. Cumulative feed consumption, changes in full body weight (FBW),

Table 1. Ingredient and nutrient composition of the basal diet¹

Ingredients, %	Grower (15–28 d)	Finisher (29–42 d)
Corn	55.9	60.69
Soybean meal (44%)	34.3	29.6
Soy oil	5.5	5.7
Limestone	1.7	1.5
Dicalcium phosphate	1.5	1.4
Sodium chloride	0.4	0.41
DL-methionine	0.2	0.2
Vitamin-mineral premix ²	0.5	0.5
Chemical analyses, dry matter basis, %		
crude protein	22.25	19.62
crude fat	6.12	6.26
crude fiber	4.76	3.83
calcium	1.15	0.95
phosphorus	0.77	0.73
Calculated compositions ³		
metabolizable energy, kcal/kg	2918	3029
lysine, %	1.12	1.09
methionine + cysteine, %	0.86	0.81

¹ mix was added into the basal diet at a dosage of 0, 0.5 and 1 g/100 g;

² vitamin premix provides the following per kg: mg: all-trans-retinyl acetate 1.8, cholecalciferol 0.025, all- α -tocopherol acetate 1.25, menadione (menadione sodium bisulphate) 1.1, riboflavin 4.4, thiamine (thiamine mononitrate) 1.1, pyridoxine 2.2, niacin 35, Ca-pantothenate 10, vitamin B₁₂ 0.02, folic acid 0.55, d-biotin 0.1; mineral premix provides the following per kg: mg: Mn (from MnO) 40, Fe (from FeSO₄) 12.5, Zn (from ZnO) 25, Cu (from CuSO₄) 3.5, I (from KI) 0.3, Se (from NaSe) 0.15, choline chloride 175; ³ value calculated according to tabulated values listed for the feed ingredients

body weight gain (BWG) and feed conversion ratio (FCR) were calculated weekly. At the end of the experiment, 12 randomly selected quails (2 quails from each cage) from each group were slaughtered to determine cold carcass and visceral weights. Carcasses were obtained by separating the internal organs, feet and feathers. Cold carcass weight (CCW) and cold carcass yield (CCY) were calculated by keeping the carcasses at 4 °C for 24 h. For the analysis of blood serum parameters, blood samples were collected during slaughter into biochemical tubes. Blood samples were centrifuged at 3 000 rpm for 10 min at 4 °C. The obtained blood sera were stored at -20 °C for analysis.

Results

Growth performance

Table 2 shows the effects of HS and SGE supplementation on quail performance parameters, including FBW, BWG, CFI, FCR, CCW and CCY. HS exposure adversely affected performance parameters compared to quails kept at TN conditions, as reflected by reduced FBW, BWG, CFI and CCW, and increased FCR ($P < 0.001$, $P < 0.001$, $P < 0.0001$, $P < 0.0001$ and $P < 0.0001$, respectively). Increments in dietary SGE levels linearly increased FBW ($P < 0.011$), BWG ($P < 0.025$) and CCW ($P < 0.001$), and decreased FCR ($P < 0.026$), whereas CFI and CCY were not affected.

Table 2. Effect of dietary stevia and ginger extract (SGE) supplementation on performance in heat-stressed quails

Variables	ET						SEM	P-value		
	TN			HS				ET	SGE	ET x SGE
	0	0.5	1	0	0.5	1				
FBW, g	227.7	232.9	240.4	215.1	224.6	227.2	3.951	0.001	0.011	0.812
BWG, g	213.2	218.4	225.9	200.7	210.3	212.7	3.956	0.001	0.025	0.791
CFI, g	847	862	877	807	798	789	17.760	0.0001	0.568	0.905
FCR, g	3.97	3.94	3.88	4.02	3.79	3.70	0.054	0.0001	0.026	0.979
CCW, g	142.2	146.0	151.1	131.2	137.6	141.3	2.447	0.0001	0.001	0.878
CCY, %	62.4	62.7	62.9	61.0	61.3	62.2	0.749	0.063	0.525	0.867

ET – environmental temperature, TN – thermoneutral, HS – heat stress, FBW – final body weight, BWG – body weight gain, CFI – cumulative feed intake, FCR – feed conversion ratio, CCW – cold carcass weight, CCY – cold carcass yield, SEM – standard error of the mean; 0, 0.5, 1 – levels of SGE supplementation (%); $P < 0.05$ indicates significantly different data

Laboratory analyses

Malondialdehyde (MDA) levels in the homogenate and serum were determined using the single heating method of Yoshioka et al. (1979) based on thiobarbituric acid (TBA) reactivity. Serum aspartate aminotransferase (AST) (Archem, A2212, Istanbul, Turkey) activity and cholesterol, glucose and creatine kinase (CK) (Archem, A2091, Istanbul, Turkey) levels were measured using commercially available kits on a D280 biochemistry auto analyzer (Sinnowa, Nanjing, JS, China). Thyroid hormones, triiodothyronine (T_3 , ng/ml) and thyroxine (T_4 , ng/dl) were analyzed by the RIA method according to Akiba et al. (1982).

Statistical analysis

Data were analysed by 2-way ANOVA using the PROC GLM procedure implemented in the SPSS software (version 16.0 for Windows; Chicago, IL, USA). Differences between treatment means were evaluated by Duncan's multiple range test. Statistical significance was considered at $P < 0.05$.

Serum biochemical parameters

Quails exposed to the HS conditions had higher serum levels of MDA ($P < 0.0001$), CK ($P < 0.0001$), AST ($P < 0.006$) and T_3 ($P < 0.0001$) than those reared in TN (Table 3). SGE supplementation decreased serum levels of MDA ($P < 0.003$), glucose ($P < 0.0001$), CK ($P < 0.0001$), AST ($P < 0.0001$), T_3 ($P < 0.001$) and T_4 ($P < 0.024$) compared to the control groups. On the other hand, the addition of SGE did not induce any changes in serum cholesterol levels in both the HS and TN groups.

Organ weights

The effects of HS and SGE supplementation on liver, heart and testis weights are shown in Table 4. While liver ($P < 0.0001$), heart ($P < 0.001$) and testis ($P < 0.0001$) weights in quails exposed heat stress decreased, SGE supplementation increased testis weight ($P < 0.034$) and did not affect liver and heart weights.

Table 3. Effect of dietary stevia and ginger extract (SGE) supplementation on serum malondialdehyde (MDA), glucose, creatine kinase (CK), cholesterol, aspartate aminotransferase (AST), triiodothyronine (T₃) and thyroxine (T₄) levels in heat-stressed quails

Variables	ET						SEM	P-value		
	TN			HS				ET	SGE	ET x SGE
	0	0.5	1	0	0.5	1				
MDA, nmol/ml	4.13	3.38	3.50	5.95	4.62	4.69	0.302	0.0001	0.003	0.523
Glucose, mg/dl	333 ^a	321 ^b	323 ^b	337 ^a	326 ^b	290 ^c	5.314	0.080	0.0001	0.002
CK, U/l	1245 ^c	729 ^d	661 ^d	2246 ^a	2193 ^{ab}	1842 ^b	45.088	0.0001	0.0001	0.0001
Cholesterol, mg/dl	208	206	200	220	218	206	6.347	0.093	0.243	0.856
AST, U/l	278	264	258	307	267	272	6.196	0.006	0.0001	0.136
T ₃ , ng/ml	2.62	2.43	2.08	2.17	1.84	1.67	0.124	0.0001	0.001	0.756
T ₄ , µg/ml	0.55	0.54	0.52	0.49	0.47	0.46	0.020	0.277	0.024	0.335

ET – environmental temperature, TN – thermoneutral, HS – heat stress, SEM – standard error of the mean; 0, 0.5, 1 – levels of SGE supplementation (%); $P < 0.05$ indicates significantly different data; ^{a-d} – means in the same row with different superscript are significantly different

Table 4. Effect of dietary stevia and ginger extract (SGE) supplementation on organ (liver, heart and testis) weights in heat-stressed quails

Variables	ET						SEM	P-value		
	TN			HS				ET	SGE	ET x SGE
	0	1	2	0	1	2				
Liver, g	3.4	3.5	3.5	3.2	2.7	3.0	0.126	0.0001	0.282	0.085
Heart, g	2.0	1.9	1.9	1.7	1.8	1.6	0.061	0.001	0.315	0.131
Testis, g	2.6	3.0	2.9	2.2	2.5	2.4	0.114	0.0001	0.034	0.657

ET – environmental temperature, TN – thermoneutral, HS – heat stress, SEM – standard error of the mean; 0, 0.5, 1 – levels of SGE supplementation (%); $P < 0.05$ indicates significantly different data

Discussion

Growth performance

Heat stress exerts a very strong effect on poultry. Feed consumption, live weight gain and feed efficiency decreases in poultry exposed to heat stress (Biswal et al., 2020; Vandana et al., 2021). Comparing the groups in our study, the FBW and BWG ratios in the TN group were found to be higher. In addition, it was observed that SGE added to both the TN and HS groups had a significant positive effect on FBW and BWG. It has been previously reported that FBW increased significantly in groups supplemented with stevia (Khalifa et al., 2021; Xiong et al., 2022). Similarly, it was demonstrated that the addition of ginger extract to the diet in broilers exposed to heat stress increased growth performance compared to control groups (Rehman et al., 2018). The addition of dried fermented ginger was found to have a strong effect on broiler body weight increase (El-Kashef, 2022). These positive outcomes could be due to the beneficial effects of herbal stevia and ginger extracts on growth, digestibility, strong antibacterial potential and health status (Rehman et al., 2018; Khalifa et al., 2021). However, some researchers reported that

ginger supplementation had no effect on body weight (Herve et al., 2018). They explained these results by changes in the phenolic and flavonoid compound levels in the digestive tract of the animal caused by the ginger extract composition. Feed consumption in the TN group was higher than in the HS group. Recent studies reported that heat stress reduced feed consumption, which was consistent with the results of the present study (Pirgozliev et al., 2020; Woods et al., 2021). Insufficient thermoregulatory capacity of quails under high temperature conditions results in lower food intake and nutrient absorption from the intestines (Sohail et al., 2012). There was an increase in CFI and FCR in the TN group fed the herbal extract. Although CFI decreased in the experimental group under HS, FCR increased. In other words, FCR in the experimental groups in both TN and HS increased compared to the control groups. In a study where stevia was added to broiler diets, it was reported that CFI and FCR increased (Atteh et al., 2008). Moreover, a study conducted on pigs found that the addition of different dietary stevia doses elevated CFI and FCR (Xiong et al., 2022). Similarly, other authors reported that the addition of ginger to broiler diets increased CFI and FCR (Kafi et al., 2017; Safiullah et al., 2019; Asghar

et al., 2021). The effect of ginger on performance is due to the fact that it contains two types of enzymes, i.e. protease and lipase (Zhang et al., 2009). These enzymes enhance food digestion and absorption of birds by positively affecting gastric secretion, as well as enterokinase and digestive enzyme activities (Demir et al., 2003). The current study determined that the effect of heat stress and dietary SGE addition on cold carcass yield was significant, but cold carcass parameters were not affected. Similarly, another study showed that heat stress affected cold carcass yield (Habibian et al., 2016); however, other authors reported that the addition of herbal extract exerted no effect on cold carcass yield (Asghar et al., 2021; Kaplan and Köksal, 2021).

Serum biochemical parameters

MDA levels in the HS group were higher compared to the TN group. However, the concentration of MDA in both experimental groups was lower than in the control groups. MDA is one of the most important indicators of oxidative damage in skeletal muscles. It is the main oxidation product of peroxidized polyunsaturated fatty acids. Its level raises with increasing temperature (Lehtonen et al., 1979), and bird exposure to high temperatures elevates lipid peroxidation and MDA concentration (Hangalapura et al., 2003). The increase in serum MDA levels may be due to the higher activity of mitochondrial respiratory chains (Kalmath and Swamy, 2013). The health status of birds is mostly determined by the level of antioxidants, and ginger and its products show hepatoprotective antioxidant, anti-inflammatory and antimicrobial properties (Ogbuwu, 2013). A previous study found that MDA levels were lowered due to the antioxidant properties of ginger (Zhang et al., 2009). Stevia extract, added to pig diets at different doses, was demonstrated to linearly reduce MDA levels and exhibit high antioxidant activity (Xiong et al., 2022). Additionally, Herve et al. (2018) reported that ginger essential oil added to quail rations significantly lowered MDA levels in the experimental groups compared to the control group. The antioxidant activity is one of the body's defense mechanisms against the harmful effects of reactive oxygen species in birds. Bioactive substances in the ginger structure protect the cells against free radicals through their antioxidant activity (Haksar et al., 2006). Glucose levels in the present study were similar in the TN and HS groups. However, there was a difference between all experimental and control groups, and an interaction between ambient

temperature and SGE was recorded. Glucose levels in the groups supplemented with the SGE mixture were lower than in the control group. Similar results were reported in birds under HS, where dietary SGE addition also lowered serum glucose levels (Saeid et al., 2010; Wu et al., 2019; Khalifa et al., 2021), and that there was no interaction between environmental temperature and SGE (Rehman et al., 2019). In another study on HS stress, blood glucose levels in broilers fed herbal preparations were lower than in the control group (Rao et al., 2021). Regulation of glucagon secretion and blood sugar by the herbal mixture can be explained by increased insulin production and improved glucose tolerance, and thus lower blood sugar levels and carbohydrate absorption in birds (Chen et al., 2005). Heat stress is an environmental factor that affects the welfare and even vital activities of livestock. CK levels in the HS-treated group were found to be higher in the present experiment; however, they were lower in all experimental groups compared to the control group. In addition, there was an interaction observed between ambient temperature and SGE. An increase in plasma creatine kinase levels determines the occurrence of muscle damage and its extent. It is well known that higher plasma creatine kinase activity is indicative of myopathy. Therefore, high plasma creatine kinase levels may serve as a marker of heat stress occurrence (Sandercock et al., 2006). A study on broilers reported that high environmental temperature increased blood creatine kinase levels (Tao et al., 2011). Evaluating the findings of our study, it can be observed that dietary SGE addition reduces blood CK levels. It can be concluded that the lower the CK level, the less damage to the muscles, and this result may be an important finding in terms of animal welfare and health. In the present study, serum AST levels were found to be higher in the HS group. However, AST concentrations in the treatment groups were lower compared to the control groups. It has been found that an increase in ambient temperature may cause damage to serum metabolites by increasing the formation of reactive oxygen species at the cellular level. Liver is one of the organs most severely affected by heat stress. Chronic exposure of the liver to heat causes cell damage and their excessive secretion. This in turn results in higher ALT and AST enzyme levels in the blood (Mohamed et al., 2012). AST levels in a study on quails fed different levels of ginger essential oil were similar to those obtained in our study (Herve et al., 2018). The addition of ginger to the diet of broilers exposed to heat

stress was previously shown to significantly reduce AST levels compared to the control group (Rehman et al., 2019; El-Kashef, 2022). However, a numerical decrease in AST levels in pigs fed different doses of stevia residue extract was previously reported (Xiong et al., 2022). This could be due to the different effect of stevia derivatives in individual species. Although the cholesterol level in the HS group was numerically higher than in the thermoneutral group, the difference was not statistically significant. Similarly, cholesterol levels in the treatment groups were numerically lower compared to the control group. This indicated that the SGE mixture exerted a positive effect on the concentration of blood cholesterol. Some researchers reported that the addition of ginger to the diet reduced cholesterol concentration (Rehman et al., 2019; Asghar et al., 2021; El-Kashef, 2022). The increase in serum cholesterol levels caused by heat stress may be due to hyperactivity of the adrenal glands, which produce adrenocorticotropin hormones (Malik et al., 2013). Many studies have demonstrated that heat stress lowers serum T_3 concentrations. The T_3 value in the HS group was higher than in the TN group and these values were similar to those reported earlier (Mack et al., 2013). Blood T_3 values in birds fed SGE decreased compared to the control group. Atteh et al. (2008) found that T_3 levels were lower in birds fed stevia leaves and stevioside, which was consistent with our study. However, previous works also reported different results regarding plasma T_4 concentrations under high temperature stress. Some researchers recorded an increase (Elnagar et al., 2010), some a decrease (Attia et al., 2016), and yet others found no changes in this parameter (Wan et al., 2017). Like in the latter case, long-term temperature stress did not have any effect on plasma T_4 concentration.

Organ weights

When assessing the effect of HS and SGE on organ weights, it was found that heat stress exerted a significant impact on this parameter. SGE only affected the weight of the testicles. Heat stress can lead to fertility disorders in livestock and birds. Depending on the temperature, disruptions in the release of gonadotropic hormones and lower sperm motility can occur, leading even to full infertility in birds (Bonato et al., 2014). In our study, testis weights in the experimental groups were higher than in the control group, which was in line with the results of Herve et al. (2018). On the other hand, heart and liver weights were lower, which could

be related to lower feed consumption and nutrient absorption due to chronic heat stress (de Souza et al., 2016).

Conclusions

High ambient temperatures cause great economic losses in the poultry industry as they negatively affect growth and reproductive performance. The addition of SGE at different doses to the poultry diets increased FBW, BWG, CCW and FCR in quails. In addition, MDA, glucose, CK, AST and T_3 levels in the experimental groups fed the SGE mixture were lower compared to the control group. There was no effect on liver and heart weights, but an increase in testicular weight was noted. The latter result may have a positive effect on semen quality. The addition of SGE to the diet during periods of increased environmental temperature may play an important role in reducing the negative effects of heat stress.

Conflict of interest

The Authors declare that there is no conflict of interest.

References

- Akbarian A., Michiels J., Degroote J., Majdeddin M., Golian A., De Smet S., 2016. Association between heat stress and oxidative stress in poultry; mitochondrial dysfunction and dietary interventions with phytochemicals. *J. Anim. Sci. Biotechnol.* 7, 37, <https://doi.org/10.1186/s40104-016-0097-5>
- Akiba Y., Jensen L.S., Bart C.R., Kraeling R.R., 1982. Plasma estradiol, thyroid hormones and liver lipids determination in birds. *J. Nutr.* 112, 299–308, <https://doi.org/10.1093/jn/112.2.299>
- Alsherbiny M.A., Abd-Elsalam W.H., El badawy S.A., Taher E., Fares M., Torres A., Chang D., Li C.G., 2019. Ameliorative and protective effects of ginger and its main constituents against natural, chemical and radiation-induced toxicities: a comprehensive review. *Food Chem. Toxicol.* 123, 72–97, <https://doi.org/10.1016/j.fct.2018.10.048>
- Asghar M.U., Rahman A., Hayat Z., Rafique M.K., Badar I.H., Yar M.K., Ijaz M., 2021. Exploration of *Zingiber officinale* effects on growth performance, immunity and gut morphology in broilers. *Braz. J. Biol.* 83, e250296, <https://doi.org/10.1590/1519-6984.250296>
- Atteh J.O., Onagbesan O.M., Tona K., Decuyper E., Geuns J.M.C., Buyse J., 2008. Evaluation of supplementary stevia (*Stevia rebaudiana*, bertonii) leaves and stevioside in broiler diets: effects on feed intake, nutrient metabolism, blood parameters and growth performance. *J. Anim. Physiol. Anim. Nutr.* 92, 640–649, <https://doi.org/10.1111/j.1439-0396.2007.00760.x>
- Attia Y.A., Al-Harhi M.A., Elnaggar A.S., 2018. Productive, physiological and immunological responses of two broiler strains fed different dietary regimens and exposed to heat stress. *Ital. J. Anim. Sci.* 17, 686–697, <https://doi.org/10.1080/1828051X.2017.1416961>

- Attia Y.A., Abd el Hamid E.A., Abedalla A.A., Berika M.A., Al-Harhi M.A., Kucuk O., Sahin K., Abou-Shehema B.M., 2016. Laying performance, digestibility and plasma hormones in laying hens exposed to chronic heat stress as affected by betaine, vitamin C, and/or vitamin E supplementation. SpringerPlus 5, 1619, <https://doi.org/10.1186/s40064-016-3304-0>
- Bonato M., Malecki I.A., Rybnik-Trzaskowska P.K., Cornwallis C.K., Cloete S.W.P., 2014. Predicting ejaculate quality and libido in male ostriches: effect of season and age. Anim. Reprod. Sci. 151, 49–55, <https://doi.org/10.1016/j.anireprosci.2014.09.015>
- Biswal J., Kennady V., Rahman H., 2020. Impact of methane emission and associated environmental stress on livestock production systems in India. J. Entomol. Zool. Stud. 8, 113–116
- Chen T.H., Chen S.C., Chan P., Chu Y.L., Yang H.Y., Cheng J.T., 2005. Mechanism of the hypoglycemic effect of stevioside, a glycoside of *Stevia rebaudiana*. Planta Med. 71, 108–113, <https://doi.org/10.1055/s-2005-837775>
- Demir E., Sarica S., Ozcan M.A., Suicmez M., 2003. The use of natural feed additives as alternatives for an antibiotic growth promoter in broiler diets. Brit. Poult. Sci. 44, 44–45, <https://doi.org/10.1080/713655288>
- de Souza L.F.A., Espinha L.P., de Almeida E.A., Lunedo R., Furlan R.L., Macari M., 2016. How heat stress (continuous or cyclical) interferes with nutrient digestibility, energy and nitrogen balances and performance in broilers. Livest. Sci. 192, 39–43, <https://doi.org/10.1016/j.livsci.2016.08.014>
- El-Kashef M.M., 2022. Evaluation of using ginger (*Zingiber officinale*) on growth performance, carcass characteristics, blood biochemistry and immune responses of quail birds. Egypt. Poult. Sci. J. 42, 199–212, <https://doi.org/10.21608/epsj.2022.249546>
- Elnagar S.A., Scheideler S.E., Beck M.M., 2010. Reproductive hormones, hepatic deiodinase messenger ribonucleic acid, and vasoactive intestinal polypeptide-immunoreactive cells in hypothalamus in the heat stress-induced or chemically induced hypothyroid laying hen. Poult. Sci. 89, 2001–2009, <https://doi.org/10.3382/ps.2010-00728>
- Habibian M., Ghazi S., Moeini M.M., 2016. Effects of dietary selenium and vitamin E on growth performance, meat yield and selenium content and lipid oxidation of breast meat of broilers reared under heat stress. Biol. Trace Elem. Res. 169, 142–152, <https://doi.org/10.1007/s12011-015-0404-6>
- Haksar A., Sharma A., Chawla R. et al., 2006. *Zingiber officinale* exhibits behavioral radioprotection against radiation-induced CTA in a gender-specific manner. Pharmacol. Biochem. Behav. 84, 179–188, <https://doi.org/10.1016/j.pbb.2006.04.008>
- Hangalapura B.N., Nieuwland M.G., de Vries Reilingh G., Heetkamp M.J., van den Brand H., Kemp B., Parmentier H.K., 2003. Effects of cold stress on immune responses and body weight of chicken lines divergently selected for antibody responses to sheep red blood cells. Poult. Sci. 82, 1692–1700, <https://doi.org/10.1093/ps/82.11.1692>
- Herve T., Raphaël K.J., Ferdinand N., Laurine Vitrice F.T., Gaye A., Outman M.M., Willy Marvel N.M., 2018. Growth performance, serum biochemical profile, oxidative status, and fertility traits in male Japanese quail fed on ginger (*Zingiber officinale*, Roscoe) essential oil. Vet. Med. Int. 2018, 7682060, <https://doi.org/10.1155/2018/7682060>
- Kafi A., Uddin M.N., Uddin M.J., Khan M.M.H., Haque M.E., 2017. Effect of dietary supplementation of turmeric (*Curcuma longa*), ginger (*Zingiber officinale*) and their combination as feed additives on feed intake, growth performance and economics of broiler. Int. J. Poult. Sci. 16, 257–265, <https://doi.org/10.3923/ijps.2017.257.265>
- Kalmath G.P., Swamy M.N., Yathiraj S., 2013. Effect of summer stress and supplementation of vitamin E and selenium on serum lipid profile in Hallikar cattle. Int. J. Sci. Res. 4, 95–97
- Kaplan C., Köksal B.H., 2021. Effect of dietary supplementation with a herbal extract on growth performance and meat quality in quails raised under thermal-neutral and heat stress conditions. Poult. Sci. J. 9, 73–84, <http://doi.org/10.22069/psj.2021.18502.1649>
- Khalifa A.M., El-Saadany A.S., Hassan M.I., Kashyout W.A., Dosoky W.M., 2021. Impact of stevioside supplementation as feed additive in finisher broiler diets on growth performance, carcass traits, meat quality, selected biochemical parameters, and caecum microflora. Adv. Anim. Vet. Sci. 9, 2168–2175, <https://doi.org/10.17582/journal.aavs/2021/9.12.2168.2175>
- Lehtonen M.A., Savolainen M.J., Hassinen I.E., 1979. Hormonal regulation of hepatic soluble phosphatidate phosphohydrolase. Induction by cortisol *in vivo* and in perfused rat liver. FEBS Lett. 99, 162–165, [https://doi.org/10.1016/0014-5793\(79\)80270-7](https://doi.org/10.1016/0014-5793(79)80270-7)
- Lemus-Mondaca R., Vega-Gálvez A., Zura-Bravo L., Ah-Hen K., 2012. *Stevia rebaudiana* Bertoni, source of a high-potency natural sweetener: a comprehensive review on the biochemical, nutritional and functional aspects. Food Chem. 132, 1121–1132, <https://doi.org/10.1016/j.foodchem.2011.11.140>
- Mack L.A., Felver-Gant J.N., Dennis R.L., Cheng H.W., 2013. Genetic variations alter production and behavioral responses following heat stress in 2 strains of laying hens. Poult. Sci. 92, 285–294, <https://doi.org/10.3382/ps.2012-02589>
- Malik H.E.E., Ali O.H.A., Mohamed E.A.A., Yousif I., 2013. Effect of season and dietary protein level on immune response of three exotic broiler strains in Sudan. J. Anim. Feed Res. 3, 31–35
- Mohamed E.A.A., Ali O.H.A., Malik H.E.E., Yousif I.A., 2012. Effect of season and dietary protein level on some haematological parameters and blood biochemical compositions of three broiler strains. Int. J. Poult. Sci. 11, 787–793, <https://doi.org/10.3923/ijps.2012.787.793>
- Ogbuewu P., 2013. Studies on the physiological responses of rabbits to ginger rhizome powder. Ph. D Thesis. Department of Animal Science and Technology, Federal University of Technology, Owerri (Nigeria), 90–91
- Pirgozliev V., Westbrook C., Woods S. et al., 2020. Feeding dihydroquercetin and vitamin E to broiler chickens reared at standard and high ambient temperatures. Arch. Anim. Nutr. 74, 496–511, <https://doi.org/10.1080/1745039X.2020.1820807>
- Rao M.H., Gurram S., Raghunandan T., Kumar K.P., 2021. Efficacy evaluation of herbal preparations and antioxidants on the growth, immunity, antioxidant status and *Escherichia coli* counts of broilers under heat stress. Indian J. Anim. Sci. 91, <https://doi.org/10.56093/ijans.v91i8.115926>
- Rehman Z., Chand N., Khan R.U., Naz S., Alhidary I.A., 2018. Serum biochemical profile of two broiler strains supplemented with vitamin E, raw ginger (*Zingiber officinale*) and L-carnitine under high ambient temperatures. South Afr. J. Anim. Sci. 48, 935–942, <https://doi.org/10.4314/sajas.v48i5.13>
- Rehman Z., Chand N., Naz S., Khan R.U., Mobashar M., 2019. Comparative serum biochemical profile of two broilers strains supplemented with L-carnitine, alpha-tocopherol and raw ginger under high ambient temperatures. J. Anim. Plant Sci. 29, 412–418
- Saeid J.M., Arkan B.M., Al-Baddy M.A., 2010. Effect of aqueous extract of ginger (*Zingiber officinale*) on blood biochemistry parameters of broiler. Int. J. Poult. Sci. 9, 944–947, <https://doi.org/10.3923/ijps.2010.944.947>
- Safiullah N.C., Rifat U.K., Shabana N., Munib A., Sina G., 2019. Effect of ginger (*Zingiber officinale* Roscoe) and organic selenium on growth dynamics, blood melanodialdehyde and paraoxonase in broilers exposed to heat stress. J. Appl. Anim. Res. 47, 212–216, <https://doi.org/10.1080/09712119.2019.1608211>

- Sandercock D.A., Hunter R.R., Mitchell M.A., Hocking P.M., 2006. Thermoregulatory capacity and muscle membrane integrity are compromised in broilers compared with layers at the same age or body weight. *Brit. Poult. Sci.* 47, 322–329, <https://doi.org/10.1080/00071660600732346>
- Sohail M.U., Hume M.E., Byrd J.A., Nisbet D.J., Ijaz A., Sohail A., Shabbir M.Z., Rehman H., 2012. Effect of supplementation of prebiotic mannan-oligosaccharides and probiotic mixture on growth performance of broilers subjected to chronic heat stress. *Poult. Sci.* 91, 2235–2240, <https://doi.org/10.3382/ps.2012-02182>
- Tao X., Dong H., Zhang H., Xin H., 2011. Sex-based responses of plasma creatine kinase in broilers to thermoneutral constant and cyclic high temperatures. *Brit. Poult. Sci.* 52, 800–806, <https://doi.org/10.1080/00071668.2011.628639>
- Vandana G.D., Sejian V., Lees A.M., Pragna P., Silpa M.V., Maloney S.K., 2021. Heat stress and poultry production: impact and amelioration. *Int. J. Biometeorol.* 65, 163–179, <https://doi.org/10.1007/s00484-020-02023-7>
- Wan X., Jiang L., Zhong H., Lu Y., Zhang L. Wang, T., 2017. Effects of enzymatically treated *Artemisia annua* L. on growth performance and some blood parameters of broilers exposed to heat stress. *Anim. Sci. J.* 88, 1239–1246, <https://doi.org/10.1111/asj.12766>
- Woods S.L., Rose S.P., Whiting I.M., Yovchev D.G., Ionescu C., Blanchard A., Pirgozliev V., 2021. The effect of selenium source on the oxidative status and performance of broilers reared at standard and high ambient temperatures. *Brit. Poult. Sci.* 62, 235–243, <https://doi.org/10.1080/00071668.2020.1824292>
- Wu X., Yang P., Sifa D., Wen Z., 2019. Effect of dietary stevioside supplementation on growth performance, nutrient digestibility, serum parameters and intestinal microflora in broilers. *Food Funct.* 10, 2340–2346, <https://doi.org/10.1039/C8FO01883A>
- Xiong Y., Liu S., Xiao H., Wu Q., Chi L., Zhu L., Fang L., Li Y., Jiang Z., Wang L., 2022. Dietary stevia residue extract supplementation improves the performance and antioxidative capacity of growing-finishing pigs. *J. Sci. Food Agric.* 102, 4724–4735, <https://doi.org/10.1002/jsfa.11833>
- Yoshoika T., Kawada K., Shimada T., Mori M., 1979. Lipid peroxidation in maternal and cord blood and protective mechanism against active oxygen toxicity in the blood. *Am. J. Obstet. Gynecol.* 135, 372–376, [https://doi.org/10.1016/0002-9378\(79\)90708-7](https://doi.org/10.1016/0002-9378(79)90708-7)
- Zhang G.F., Yang Z.B., Wang Y., Yang W.R., Jiang S.Z., Gai G.S., 2009. Effects of ginger root (*Zingiber officinale*) processed to different particle sizes on growth performance, antioxidant status and serum metabolites of broiler chickens. *Poult. Sci.* 88, 2159–2166, <https://doi.org/10.3382/ps.2009-00165>