

Faculty of science Chemistry Department



The protective role of turmeric, ginger and n-acetylcysteine on heat stress

A thesis

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Bу

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SUMMARY

HS causes physiological changes in broiler chickens like reduction in blood antioxidant status and increased the oxidative stress indicators. Heat exposure significantly increased the concentration of serum CORT, HSP70 and HSP90 α to help chickens coping with the damage from high temperature.

This study was carried out in summer season (August, 2018) on one hundred and sixty, apparently healthy, one-day old, unsexed broiler chicks (Cobb 500). All broilers had been obtained from El-Kahera Poultry Company, 10th of Ramadan City in Al-Sharkia governorate, Egypt.

After thorough cleaning and disinfecting, the chicks were housed in a constant environmental under hygienic measures following the guide for the care and the use of laboratory animals guide lines of the national institute of health (NIH). They were vaccinated and fed on a well-balanced ration throughout the experimental period (42 d).

On the day 21st d of age, all broilers randomly allocated in to 8 equal groups where each group contains 20 chicken and explained as follow:

A. Class 1: Thermoneutral (TN):

Non-Heat stressed birds reared under an ambient temperature of (22 \pm 4 °C) daily and relative humidity (55 \pm 3, RH) on day 21stuntil the end of the study (42 d).

- 1) **Group 1:** Thermoneutral control group (TNC) broilers were not received any supplement to the basal diet.
- Group 2: Thermoneutral turmeric group (TNT)birds were supplemented diet with turmeric powder (5 g/kg diet).

- 3) **Group 3**: Thermoneutral ginger group (TNG) birds were administered basal diet with ginger powder (7.5 g/kg diet).
- Group 4: Thermoneutral n-acetyl cysteine group (TNN) birds were administrated basal diet with antioxidant chemical n-acetyl cysteine (100mg/kg diet).

B. <u>Class 2: HS (HS)</u>:

Heat stressed (HS) birds reared under an ambient temperature $(34 \pm 2 ^{\circ}C)$ daily from 9:00 to 17:00 h. with a relative humidity 65% on day 21 until the end of the study (42 d).

1) **Group 5:** HS control (HSC) birds didn't receive any supplement to the basal diet.

2) Group 6: Heat stressed turmeric group (HST)birds administered with basal diet with turmeric powder (5 g/kg diet) with basal diet.

3) **Group 7:** Heat stressed ginger group (HSG)birds administered with ginger powder (7.5 g/kg diet).

4) **Group 8:** Heat stressed n-acetyl cysteine group (HSN)administrated with antioxidant chemical n-acetyl cysteine (100mg/kg diet) with basal diet.

Evaluation of general health and growth performance:

The clinical symptoms were recorded daily throughout the experimental period and 15 chickens from each group were weighted at the beginning of the experiment (21st day of age) then weekly weighted till the end of the study. Similarly, the diet consumed was calculated all over the study period.

Blood Samples:

After broilers' fast for 10 h., on 42 days of age, five birds from each group were selected randomly then blood samples were drawn from wing veins and divided in to 2 samples for hematological and biochemical studies.

- The first blood sample was collected in 1 test tube with stopper containing EDTA 50 I.U./ML blood as anticoagulant to measure hematological parameters.
- The second blood sample was collected in clean and dry tube with a rubber stopper left to clot then centrifuged at 2500 r. p. m for 10 minutes to allow serum separation. The clear sera were collected in dry Eppendorf tubes and then stored in deep freezer at -20°C until be used for the subsequent biochemical, MDA, SOD and hormones analysis.

Liver tissue samples:

Birds were slaughtered, liver specimens were removed and wrapped in aluminum foil and placed immediately in liquid nitrogen at -196°C for making snap freezing then stored in deep freezer at -80°C until be used for the subsequent detection of gene expression.

The obtained results showed that:

1- Clinical symptoms:

In thermoneutral condition $(22\pm 4^{\circ}C)$, all broiler chickens of control group and also diet supplemented with turmeric (0.5%), ginger (0.75%) and NAC (0.01%) groups were apparently healthy, viable or showing no clinical symptoms throughout the experimental period

In Heat stressed control group $(34 \pm 2 \text{ °C for 8 h})$, all broiler chickens were apparently lazy to reduce heat generated by activity, moving hardly towards diet to reduce feed intake spending more time drinking, lift their wings away from their bodies to reduce insulation and expose any areas of skin that have no feathers, more panting, opened mouths, moving away from other birds, Move away of block walls or into moving air streams. However, broiler chickens in Heat stressed groups $(34 \pm 2^{\circ}C \text{ for } 8 \text{ h})$ supplemented with turmeric (0.5%), ginger (0.75%) and NAC (0.01%) diet on 21st days of age displayed clinical signs which appeared after 24 to 72 hours post treatment represented by an increase of appetite, more activity, less waving wings, less panting when compared with HSC group.

2- Effect on growth performance:

It was found that addition of TP, GP and NAC in normal conditions were significantly increased (P < 0.05) the body weight (BW), body weight gain (BWG) and feed intake (FI) of broilers at all experiments' measures with non-significant effect of TNN at 35 day when compared with TNC broilers. Diet supplemented with GP was significantly decreased the F.C.R on 28, 35 and 42 d while GP and NAC have significant (P < 0.05) decrease on 42 d of the experimental period when compared with TN control group.

In HS condition, the present investigation confirmed that broilers under chronic HS had a negative effect on the growth performance of broilers. However, broilers subjected to HS and supplemented with TP (0.5%), GP (0.75%) and NAC (0.01%) of diet have significant increase on BW and BWG and FI during 28-42 d with non-significant effect on FI of TP at 35 d of age than HSC. This study demonstrated that NAC addition on diet could attenuate the adverse effects of HS. It is important to note that dietary supplementation of ginger powder has provided the best outcomes for all growth efficiency indices of broilers.

3- Effect on hematological indices:

In HS condition, the PCV, HGB and RBCs were significantly decreased when compared with thermoneutral control group. While no

significant effect on platelet during high temperature. On the other side, the turmeric and ginger dietary additives have significant increase on PCV, HGB and RBCs while NAC affected PCV significantly than HSC at 42 of age.

4- Effect on lipid profile:

High ambient temperature produced significant increase in the data of serum total cholesterol, Triacylglycerol, LDL, VLDL and cholesterol ratio while non-significant decrease in HDL at the end of the experimental period compared with thermoneutral control group.

In comparison with the results of HSC chickens, dietary treatment with the powder form of turmeric additives have significant decrease in total cholesterol, Triacylglycerol, LDL, VLDL and cholesterol ratio with non-significant increase in HDL. The addition of ginger in diet have significant decrease in total cholesterol, Triacylglycerol, LDL, VLDL and cholesterol ratio with significant increase in HDL. Diet supplementation of NAC provided significant decrease in total cholesterol, LDL and cholesterol and VLDL with non-significant decrease in total cholesterol, LDL and cholesterol and VLDL with non-significant decrease in total cholesterol, LDL and cholesterol ratio while HDL has non-significant elevation.

5- Effect on serum gglucose, liver and kidney function tests:

HS have increased concentrations of Glc significantly then improved by supplementation of TP and GP significantly. In HS condition, overall data of total protein, albumin and A/G ratio showed significant decrease while non-significant decrease in globulin parameter of HSC group when compared with thermoneutral control group. On the other side, TP and GP dietary additives have significant increase in total protein and globulin while non-significant effect on albumin and A/G ratio of broilers subjected to HS. Broilers experimentally supplemented NAC in HS environment have non-significant increase in total protein, albumin, globulin and A/G on the 42nd day of chickens age.

In HS condition, overall data of AST, ALT and ALP were significantly increased when compared with thermoneutral control group. On the other side, Broilers experimentally supplemented TP, GP and NAC as dietary additives have significant decrease in serum AST, ALT and ALP on the 42nd day of age.

HS exposed broilers have significant increase in serum T-bill and Dbill that improved by dietary inclusion of TP.

In HSC group, broilers had significant increase (P< 0.05) in uric acid and creatinine levels on day 42 of age when compared with TNC group. Broilers experimentally supplemented with turmeric powder have nosignificant decrease in uric acid and creatinine concentration than HSC group.

6- Effect on MDA and SOD:

HS significantly elevated serum MDA content with significant decrease in serum SOD activity that improved significantly with dietary supplementation of TP and GP.

7- Effect on T3, T4 and CORT hormones:

HS caused significant decrease in T3 and T4 concentrations and a significant increase in CORT when compared with TNC group. The dietary intake of TP has no significant elevation effect on T3 and T4 while provided a significant improvement of CORT hormone. Addition of GP in diet significantly increased (P < 0.05) thyroid hormones while NAC affects in T3 by a significant increase compared with HSC group. Broiler chickens subjected experimentally to HS have significant increase in CORT hormone compared with HSC broilers.

8- Effect on HSP70 and HSP90α:

In liver, HSP70 and HSP90 α were elevated in HS broilers but have been ameliorated by dietary treatments of TP, GP and NAC. Our results indicated that natural additives have a better effect in alleviation of oxidative stress than NAC.