Effect of T-2 toxin and antioxidants on angel wing incidence and severity in White Roman geese

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ABSTRACT
This study investigates the effects of T-2 toxin and antioxidants on the incidence and severity of angel wing in White Roman geese. Twelve pens were used in this study, and half of them received dietary supplementation of T-2 toxin (10 ppm) and antioxidants (vitamin C 1000 ppm plus Se 0.3 ppm). Each pen contained birds from the normal wing line (NL), the selected angel wing line (AL), and a controlled commercial line (CL). The results showed that there was no significant difference in the body weight, body weight gain, and feed intake of goslings that were supplemented from birth to 6 weeks of age with T-2 toxin and antioxidants. The alkaline phosphatase level in the T-2 toxin group was lower than that in the control group at 4 and 6 weeks. The haemoglobin level in the T-2 toxin group was lower than that in the control group at 6 weeks. There was a significant interaction between T-2 toxin and antioxidants in the severity score of angel wing (SSAW) and incidence of angel wing (IAW) at 6 weeks. In conclusion, the results suggest that a diet supplemented with T-2 toxin does reduce alkaline phosphatase levels. When the diet contained T-2 toxin and antioxidants, the SSAW and IAW increased.

1. Introduction
Angel wing (AW) is an anomaly in certain growing waterfowl. Abnormal wing has been termed as slipped wing (Kreeger & Walser 1984), twisted wing (Grow 1972), AW (Francis et al. 1967), or airplane wing (Ritchie et al. 1994). The variety of names suggests the widespread nature of its abnormality around the world.

AW is characterized by a lateral torsion of the distal end of the forelimb, on either side or both sides. It occurs mostly at the carpometacarpus, where the limb begins to twist outward away from the body towards the extremity of the wing. Kuiken et al. (1999) observed the abnormal wing in double-crested cormorants, reporting that the primary feathers of the affected wing are held horizontally at 30–45° to the median plane, making the undersurface of the feathers face upwards. Furthermore, it was estimated that about 4.4% of Masked Booby chicks exhibited AW during March 2005. This coincided with a time of high nestling mortality that was apparently related to food shortage; this prompted speculation on the causal linkages between food shortages and AW (Wyatt et al. 1975).

AW occurs mostly at 6–14 weeks post-hatching in White Roman geese (Lin et al. 2006, 2008). In Taiwan, most geese raised for meat production are White Roman or one of its cross-breed. They are evaluated in market by their appearance and presentation. In a previous report on genetic selection, the incidence of angel wing (IAW) was 59.8% for 164 progenies comprising full sibs coming from 41 families; each included 1 sire and 4 dams, and only those that had both parents with AW were counted. The liability heritability of IAW, 0.39, was thereby estimated (Lin et al. 2006). A total of 1696 geese from both lines were used to estimate the liability heritability of AW incidence (LHIAW). LHIAWs in heavy body weight (BW) and high egg production lines were estimated separately at 0.39 and 0.03, respectively. The estimated pooled LHIAW was 0.31 (Lin et al. 2008). Lin et al. (2016) further indicated that differences were observed among the genetic stock; that is, SSAW and IAW are significantly higher in the AW line than in the normal or commercial lines.

T-2 toxin is a mycotoxin and part of a group of type A trichotheecenes produced by several fungal genera, including the Fusarium species. A previous report stated that feeding chickens T-2 toxin can inhibit their growth (Girish & Devegowda 2006) and cause abnormal feathering. Compared to the control group, the test chickens were sparsely covered, with short feathers protruding at odd angles. There were few feathers on the base of the neck, the anterior dorsal surface of the wing, or the side and back adjacent to the tail (Wyatt et al. 1975).

Vitamins with pyridoxine, ascorbic acid, and menadione are important for sustaining the integrity and health of bones. Pyridoxine strengthens collagen crosslinks and the crucial constituents of bone matrix, subsequently impacting the mechanical properties of a bone (Weber 1999). Selenium, ascorbic acid, and their precursors act as superoxide anion scavengers and protect against the toxic effects of mycotoxins.

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Supplementation with tocopherol, ascorbic acid, and selenium functions as both an antioxidant system and a free radical scavenger, protecting the spleen and brain of rats from membrane damage caused by T-2 toxin and DON (deoxynivalenol) (Atroshi et al. 1995).

Although several studies have examined AW in geese or wild waterfowl, the aetiology of the abnormality remains unclear. Francis et al. (1967) indicated that if inheritance is involved in AW, then the phenotype must be affected by more than one gene rather than just a simple recessive. Feeding chickens T-2 toxin can inhibit growth and cause abnormal feathering (Wyatt et al. 1975), as well as destroy joint cartilage during the development and proliferation stages, eventually leading to osteoarthritis (Nascimento et al. 2001). This same mechanism may induce AW in geese. The objective of this study is, therefore, to supplement White Roman geese with T-2 toxin and antioxidants and examine the effects on the incidence and severity of AW.

2. Material and methods

2.1. Bird management

The care and use of all geese were according to the Institutional Animal Care and Use Panel (IACUP) at the Changhua Animal Propagation Station (Livestock Research Institute (CAPS-LRI, located at 23°51’N and 120°33’E), Council of Agriculture, Taiwan). The angel-winged line (AL) and normal-winged line (NL) were initially established in 2007 and were divergently selected thereafter by CAPS-LRI. The birds from a commercial farm were designated as the commercial line (CL).

Immediately after being hatched, the goslings’ gender was determined and their feet were banded. For the first two weeks, lighting and heat were supplied 24 h a day with an incandescent 60-Watt bulb hanging at a height that maintained an ambient temperature of 28°C; thereafter, no artificial heat source was provided. The goslings were moved from the nursery to the growing house when 2 weeks old.

The birds were fed ad libitum with commercial rations containing 20% crude protein plus 2900 kcal/kg metabolizable energy (ME) from 0 to 4 weeks before switching to feed with 15% crude protein plus 2800 kcal/kg ME (Table 1) from 5 to 6 weeks. The dietary content of the crude protein during weeks 0–6 was per National Research Council (NRC) guidelines (1994). The dietary ME content was per NRC guidelines (1994) during weeks 0–4, but was then lowered below the recommendation (3000 kcal/kg) for weeks 5 and 6, due to the high ambient temperature and humidity in Taiwan.

2.2. Experimental design

Twelve pens were randomly assigned to one of four treatments in a factorial arrangement (two T-2 toxin supplemented groups × two antioxidant supplemented groups). Birds from three different genetic backgrounds, that is, NL, AL, and CL, were nested within each pen. This study, therefore, was a 2 × 2 × 3 factorial experiment arranged in a split-plot design.

The T-2 toxin treatments (10 ppm) and antioxidants (ascorbic acid 1000 ppm and selenium 0.3 ppm) were administered from day one to week four. The 20 birds were kept in a 2 × 3 × 2 factorial experiment arranged in a split-plot design.

2.3. Production of T-2 toxin

The commercial laying geese feed was incubated at 25°C with a relative humidity of 86 ± 4% for 21 days. Without drying, the cultured feed was transferred to 200 mL Erlenmeyer flasks and extracted and homogenized with 100 mL 90% methanol for 2 minutes before being centrifuged for 5 min at 3000 g. After filtration through Whatman no. 1 filter paper, the filtrate was diluted with water at a ratio of 1:4 and then filtered again through a microfibre filter. Next, 10 mL of the diluted and filtered extract was passed through the T-2 toxin test™ column before being washed with 10 mL water. To obtain pure T-2 toxin, 10 mL high-performance liquid chromatography (HPLC) grade methanol was then passed through the column.

The pure T-2 toxin contained 218 mg T-2 toxin/kg, confirmed via T-2 toxin analysis by HPLC. Tso et al. (2012) previously described the extraction and analysis of pure T-2 toxin. The pure T-2 toxin was dissolved in 95% ethanol before being mixed into the birds’ diet.

2.4. Growth performance

The individual body weights and weight gains of the geese were measured biweekly. Feed consumption was recorded on a pen basis up to 6 weeks of age, and feed conversion ratios were then calculated.

### Table 1. The components and compositions of experimental diets (as fed basis).

| Item | Experimental diet |
|------|-------------------|
| | Starter (0–4 weeks) | Grower (5–6 weeks) |
| Ingredients, g/kg | | |
| Yellow corn | 614 | 668.5 |
| Soybean meal, 44% | 260 | 165 |
| Wheat bran | 20 | 50 |
| Fish meal, 65% crude protein | 50 | 25 |
| Molasses | 30 | 30 |
| Rice bran | – | 30 |
| Salt | 3 | 3 |
| Dicalcium phosphate, 22% phosphorous | 10 | 13 |
| Limestone, pulverized | 7 | 7 |
| Choline chloride, 50% | 1 | 1 |
| DL-methionine | 1 | 1 |
| Vitamin premix^a | 1 | 1 |
| Mineral premix^b | 2 | 2 |
| Calculated values | | |
| Crude protein, % | 20 | 15 |
| ME, kcal/kg | 2900 | 2800 |
| Calcium, g/kg | 8.20 | 7.30 |
| Available phosphorus, g/kg | 4.60 | 4.10 |
| Analysed values | | |
| Crude protein, % | 19.7 | 14.8 |

^aVitamin premix: Each kg contained retinol 3 g, cholecalciferol 0.05 g, D-α-tocopherol 18.2 g, thiamine 1 g, riboflavin 4.8 g, pyridoxine 3 g, cobalamin 0.01 g, biotin 0.2 g, menadione 1.5 g, D-calcium pantothenate 10 g, folic acid 0.5 g, and nicotinic acid 25 g.

^bMineral premix: Each kg contained copper 15.0 g, ferrum 80 g, zinc 50 g, manganese 80 g, cobalt 0.25 g, and iodine 0.85 g.
2.5. Serum biochemical parameters and blood routine index

On the first day of each month of the experimental period, serum and blood samples for biochemical determinations were collected from 6 randomly selected geese per pen (3 males and 3 females). Blood samples were processed approximately 4–5 h after collection by centrifuging at 3000 × g for 10 min at 4°C; serum samples were then stored at −4°C for up to 1 d until analysis (Lee, Ciou et al. 2013). Serum biochemical parameter and blood routine index analyses were performed by an Automatic Biochemical Analyser (Hitachi, 7150 auto-analyser, Tokyo, Japan).

2.6. Severity, severity scores, and IAW

Lin et al. (2016) defined severity, severity score, and IAW. A bird’s normal wing (NW) was defined as being covered with smooth, neat, and clean feathers along its body. AW, which may occur on one or both sides of the wing, was defined as a wing projecting away from the body surface at varying angles so that the undersurface of the primary feathers was facing outward and upward.

The severity of AW was categorized as slight, medium, or severe depending on the extent of primary feathers projecting away from the body at an angle of less than 30°, between 30° and 60°, and more than 60°, respectively. The severity score of AW (SSAW) ranged from 0 to 6, where 0 indicates NW; 1 indicates only one wing with slight AW; 2 indicates only one wing with medium AW; 4 indicates one wing with severe AW; 5 indicates one wing with slight AW and the other with medium AW; 6 indicates both wings with severe AW.

The IAW was expressed as a percentage and calculated as the number of birds with AW (SSAW = 1–6) divided by the number of birds in the pen multiplied by 100. Alternatively, it was obtained by deducting the percentage of NW from 100; the former value was calculated as the number of birds with NW (SSAW = 0) divided by the number of birds in the pen multiplied by 100 (Lin et al. 2016).

Goslings generally start to moult their down feathers when 2 weeks old, and their primary feathers will be full-fledged at 5–6 weeks. Therefore, SSAW and IAW of the geese were recorded at 6 weeks of age.

2.7. Statistical analyses

Since the obtained IAW values were between 30% and 70%, the data were converted to arcsine values for ANOVA. The data of the variables collected were statistically analysed using a MIXED procedure from SAS software (2004) following a factorial arrangement of treatments in a split-plot design, in which the three genetic lines of birds nested in each of the 12 pens were regarded as plots.

The mathematic model is:

\[ Y_{ijkl} = \mu + T_i + A_j + (TA)_{ij} + e_{ijk} + L_k + S_m + (LS)_{km} + (TLS)_{ilm} + (ALS)_{jm} + (TALS)_{ijm} + e_{ijklm}, \]

where \( Y_{ijkl} \) = the observed response of bird line in a pen; \( \mu \) = the overall mean; \( T_i \) = the fixed effect of toxin supplementation; \( A_j \) = the fixed effect of antioxidant supplementation; \( (TA)_{ij} \) = the interaction effect of toxin supplementation × antioxidant supplementation; \( e_{ijk} \) = the residual error when the pen is regarded as an experimental unit, \( e_{ijk} \sim N(0, \sigma^2_e) \); \( L_k \) = the fixed effect of the bird line; \( S_m \) = the fixed effect of the bird sex; \( (LS)_{km} \) = the interaction effect of line × sex; \( (TLS)_{ilm} \) = the interaction effect of toxin supplementation × line × sex; \( (ALS)_{jm} \) = the interaction effect of antioxidant supplementation × line × sex; and \( e_{ijklm} \) = the residual error when lines nested in a pen are regarded as an experimental unit, \( e_{ijklm} \sim N(0, \sigma^2_e) \). The mean values were compared between the T-2 toxin supplementations, the antioxidant supplementations, and among the three lines using least squares means, with the significance level at \( p < .05 \).

3. Results

3.1. Growth performance

Table 2 shows the effects of T-2 toxin and antioxidants on the growth for grower geese at the age of 6 weeks. There was no significant effect between the control and T-2 toxin groups on body weight at the age of 6 weeks. There was no significant effect between the control and antioxidant groups on body weight at the age of 6 weeks. The body weight in CL was heavier than that in AL and NL at birth and 2 weeks old (\( p < .05 \); Table 2).

There was no difference observed among the body weight of the three lines at ages after 6 weeks old. There was no difference in body weight between male and female geese at birth and 2 weeks old. In contrast, compared to female geese, male geese had a heavier body weight at ages after 6 weeks old (\( p < .01 \)). There was a significant interaction between line and sex on body weight at 4 weeks old (\( p < .01 \)).

There was no significant effect between the control and T-2 toxin groups in body weight gain (Table 3). The body weight gain in the male group was higher than that of the female group at the age of 6 weeks (\( p < .05 \)). There was no significant effect between the control and T-2 toxin or antioxidant groups’ feed consumption and feed conversion at the age of 6 weeks (Table 4).

3.2. Serum biochemical parameters and blood routine index

The alkaline phosphatase (ALK) in the T-2 toxin group was lower than that of the control group at the age of 4 and 6 weeks (\( p < .05 \); Table 5). The haemoglobin in the T-2 toxin group was heavier than that in AL and NL at birth and 2 weeks old (\( p < .05 \) and \( p < .01 \), respectively). There was a significant interaction between line and sex on body weight at 4 weeks old (\( p < .05 \)).

There was no significant effect between the control and T-2 toxin groups in body weight gain (Table 3). The body weight gain in the male group was higher than that of the female group at the age of 6 weeks (\( p < .05 \)). There was no significant effect between the control and T-2 toxin or antioxidant groups’ feed consumption and feed conversion at the age of 6 weeks (Table 4).
Table 2. Effect of T-2 toxin and antioxidants on the body weight of grower geese.

| Age  | T   | A   | Line | Sex   | Significance |
|------|-----|-----|------|-------|--------------|
|      | 0   | 10  | NIL  | YES   |              |
|      | SEM1| SEM2| AL   | CL    | NL           |
| Body weight, kg/bird | 0.10 | 0.10 | 0.01 | 0.10 | 0.001 |
| 2 weeks | 0.09 | 0.09 | 0.09 | 0.09 | 0.09 |
| 4 weeks | 0.38 | 0.38 | 0.38 | 0.38 | 0.38 |
| 6 weeks | 0.70 | 0.70 | 0.70 | 0.70 | 0.70 |
| Body weight gain, kg/bird | 1.83 | 1.85 | 0.037 | 1.82 | 1.86 |
| 5–6 weeks | 0.89 | 0.91 | 0.017 | 0.90 | 0.90 |
| 0–6 weeks | 2.72 | 2.72 | 0.030 | 2.73 | 2.75 |

Notes: T: T-2 toxin; A: antioxidants; AL: angel-winged line; NL: normal-winged line; CL: commercial line; T × A: the interaction of T-2 toxin and antioxidants; Line × Sex: the interaction of line and sex; T × Line × Sex: the interaction among T-2 toxin, line, and sex; A × Line × Sex: the interaction among antioxidants, line, and sex. SEM1: standard error of means of T-2 toxin. SEM2: standard error of means of antioxidants. SEM3: standard error of means of genetic line. SEM4: standard error of means of sex. a, b Means in the same row under either genetic line or sex without the same superscripts differ significantly (p < .01).

Table 3. Effect of T-2 toxin and antioxidants on body weight gain of grower geese.

| Age  | T   | A   | Line | Sex   | Significance |
|------|-----|-----|------|-------|--------------|
|      | 0   | 10  | NIL  | YES   |              |
|      | SEM1| SEM2| AL   | CL    | NL           |
| Body weight gain, kg/bird | 1.83 | 1.85 | 0.037 | 1.82 | 1.86 |
| 5–6 weeks | 0.89 | 0.91 | 0.017 | 0.90 | 0.90 |
| 0–6 weeks | 2.72 | 2.72 | 0.030 | 2.73 | 2.75 |

Notes: AL: angel-winged line; NL: normal-winged line; CL: commercial line; T: T-2 toxin; A: antioxidants; AL × A: the interaction of T-2 toxin and antioxidants; Line × Sex: the interaction of line and sex; T × Line × Sex: the interaction among T-2 toxin, line, and sex; A × Line × Sex: the interaction among antioxidants, line, and sex. SEM1: standard error of means of T-2 toxin. SEM2: standard error of means of antioxidants. SEM3: standard error of means of genetic line. SEM4: standard error of means of sex. a, b Means in the same row under line without the same superscripts differ significantly (p < .05). x, y Means in the same row under sex without the same superscripts differ significantly (p < .05).
3.3. SSAW and IAW

IAW in the T-2 toxin group tended to be higher than that in the control group at the age of 6 weeks \((p = .0732)\). There was a significant interaction between T-2 toxin and antioxidants on SSAW and IAW at 6 weeks old \((p < .05; \text{Table 7})\). SSAW in the T-2 toxin and antioxidant groups was higher than that in the single T-2 toxin or antioxidant group at the age of 6 weeks \((p < .05)\). IAW in the T-2 toxin and antioxidant groups was higher than that in the other groups at the age of 6 weeks \((p < .05)\).

SSAW and IAW in the male group were lower than that in the female group at the age of 6 weeks \((p < .05)\).

4. Discussion

The variables measured were not significantly affected by toxin supplementation or antioxidant supplementation; there were two-way interactions between toxin and antioxidant supplementation; there were three-way interactions among toxin

### Table 4. Effect of T-2 toxin and antioxidants on the feed consumption and feed conversion ratio of grower geese.

| Age Group | T | SEM1 | A | SEM2 | Significance |
|-----------|---|------|---|------|-------------|
| 0–4 weeks | FEED CONSUMPTION | kg feed/bird | 3.61 | 3.68 | 0.086 | 3.66 | 3.63 | 0.086 | 0.5727 | 0.8516 | 0.7188 |
| 5–6 weeks | FEED CONVERSION RATIO | kg diet/kg body weight gain | 8.05 | 8.17 | 0.199 | 8.15 | 8.07 | 0.199 | 0.6943 | 0.7858 | 0.2673 |

Notes: T: T-2 toxin; A: antioxidants; T × A: the interaction of T-2 toxin and antioxidants. SEM1: standard error of means of T-2 toxin. SEM2: standard error of means of antioxidants.

### Table 5. Effect of T-2 toxin and antioxidants on serum biochemical parameters in grower geese.

| Age Group | T | SEM1 | A | SEM2 | Significance |
|-----------|---|------|---|------|-------------|
| 4 weeks | GOT, U/L | 61.5 | 66.1 | 3.21 | 71.3 | 56.3 | 3.21 | 0.3422 | 0.0107 | 0.7099 |
| 6 weeks | GPT, U/L | 26.1 | 27.9 | 0.92 | 28.1 | 25.8 | 0.92 | 0.2102 | 0.1045 | 0.3974 |
| 4 weeks | TP, g/dL | 4.11 | 4.19 | 0.055 | 4.14 | 4.17 | 0.055 | 0.3260 | 0.7259 | 0.1598 |
| 6 weeks | ALB, g/dL | 1.89 | 1.95 | 0.034 | 1.93 | 1.91 | 0.034 | 0.2397 | 0.7380 | 0.9109 |
| 4 weeks | ALK, IU/L | 709 | 581 | 33.7 | 638 | 653 | 33.7 | 0.0282 | 0.7550 | 0.1766 |
| 6 weeks | GLU, mg/dl | 143 | 140 | 11.5 | 152 | 130 | 11.5 | 0.8490 | 0.2164 | 0.7301 |
| 4 weeks | BUN, mg/dl | 106 | 107 | 8.26 | 112 | 102 | 8.26 | 0.9248 | 0.4183 | 0.9835 |
| 6 weeks | UA, mg/dl | 1.36 | 1.75 | 0.109 | 1.98 | 1.13 | 0.110 | 0.0374 | 0.0096 | 0.4628 |
| 6 weeks | CREA, mg/dl | 1.00 | 1.09 | 0.068 | 1.06 | 1.03 | 0.068 | 0.3670 | 0.7581 | 0.6355 |

Notes: T: T-2 toxin; A: antioxidants; T × A: the interaction of T-2 toxin and antioxidants. GOT: Glutamic Oxaloacetic Transaminase, GPT: Glutamic Pyruvic Transaminase, TP: Total protein, ALB: Albumin, ALK: Alkaline phosphatase, GLU: Glucose, BUN: Blood urea nitrogen, UA: Uric acid, CREA: Creatinine, TG: Triglycerides, TC: Total cholesterol, HDL: high density lipoprotein, LDL: low density lipoprotein. SEM1: standard error of means of T-2 toxin. SEM2: standard error of means of antioxidants.
supplementation, line, and sex, as well as antioxidant supple-
movement, line, and sex; or there were four-way interactions
between toxin supplementation, antioxidant supplementation,
line, and sex. Therefore, this study only presents the effects of
line, sex, or line and sex.

Yellow-feathered broiler chickens fed 0.5 mg/kg of Ochra-
toxin A and 1 mg/kg of T-2 toxin had lighter final BW, average
daily gain, and average daily feed intake, but there was no sta-
tistical significance on the efficiency of feed utilization (Wang
et al. 2009). Weber et al. (2010) reported that broiler chickens
supplemented with 1.04 mg T-2 toxin and 0.49 mg HT-2
toxin A and 1 mg/kg of T-2 toxin had lighter final BW, average
daily BW gain, feed conversion ratios, and survival rate after
90 days than those not fed selenium. Dvorska et al. (2007) pre-
icted a major protective effect of the mycotoxin binder in
combination with organic selenium against the detrimental
consequences of T-2 toxin-contaminated feed consumed by
growing chickens. In the current study, however, there was no
significant difference in feed consumption for the group that
received antioxidant supplementation (Table 4). The resulting
feed conversion ratios were evaluated for groups both with
and without antioxidants, revealing that birds fed diets sup-
plemented with antioxidants demonstrated no significant
differences in the efficiency of feed conversion.

Oxidative stress, an imbalance condition when the reactive
oxygen species (ROS) formation exceeds cellular antioxidant
capacity (Wang et al. 2017), has become a major issue and
the subject of production concerns and related research in
the domestic animal industry (Lin et al. 2017). ROS are con-
stantly produced in aerobic organisms as by-products of
normal oxygen metabolism. Chapple (1997) pointed out that
inflammatory diseases may be subjected to oxidative stress
caused by their oxidative metabolism and rapid growth
associated with the production of large quantities of free rad-
icals or other reactive oxygen metabolites. In animals, in order
to combat and neutralize the deleterious effects of those ROS,
the cells develop various mechanisms for maintaining redox
homeostasis, which could be classified into two types of anti-
oxidants. First, direct antioxidants have redox activity and
short half-lives that should be regenerated or supplemented
during the process (Jung & Kwak 2010; Lin et al. 2017). More-
ever, they should be administrated frequently and in relatively
high dosages to sustain their physiological efficacy; whereas
indirect antioxidants act through the augmentation of cellular
antioxidant capacity by enhancing specific genes encoding
antioxidant proteins through the key transcription factor
Nrf2, which have been a problem in the use of high-dose vitamin E
therapy (Mamede et al. 2011). Nrf2 is a basic leucine zipper-

| Table 6. Effect of T-2 toxin and antioxidants on blood routine index in grower geese. |
|-----------------|----------|--------|--------|--------|--------|----------|--------|
| Age      | T        | SEM1 | NIL | YES | SEM2 | T  | A  | T×A |
| WBC, 10³/µL |           |      |     |     |      |    |     |     |
| 4 weeks  | 276⁴     | 264⁵ | 3.64 | 271  | 268  | 3.64 | 0.0495 | 0.5458 | 0.4327 |
| 6 weeks  | 272      | 281  | 2.88 | 279  | 273  | 2.88 | 0.0625 | 0.2517 | 0.5577 |
| RBC, 10⁹/µL |           |      |     |     |      |    |     |     |
| 4 weeks  | 1.49     | 1.48 | 0.022 | 1.50 | 1.46 | 0.022 | 0.7771 | 0.2489 | 0.6860 |
| 6 weeks  | 1.58     | 1.61 | 0.029 | 1.62 | 1.57 | 0.029 | 0.5259 | 0.2748 | 0.9845 |
| HB, μg/dL |           |      |     |     |      |    |     |     |
| 4 weeks  | 9.46     | 9.66 | 0.084 | 9.39³ | 9.72⁺ | 0.084 | 0.1264 | 0.0239 | 0.6055 |
| 6 weeks  | 10.2⁴    | 10.0⁵ | 0.061 | 10.1 | 10.2 | 0.061 | 0.0498 | 0.1624 | 0.6655 |
| HT, %    |           |      |     |     |      |    |     |     |
| 4 weeks  | 25.6     | 25.7 | 0.475 | 25.8 | 25.5 | 0.475 | 0.9202 | 0.6469 | 0.1801 |
| 6 weeks  | 27.3     | 27.8 | 0.543 | 28.1 | 27.1 | 0.543 | 0.5468 | 0.2269 | 0.8610 |
| MCV, fl  |           |      |     |     |      |    |     |     |
| 4 weeks  | 173      | 174  | 1.46 | 172  | 175  | 1.46 | 0.5209 | 0.3093 | 0.0406 |
| 6 weeks  | 173      | 173  | 1.61 | 174  | 173  | 1.61 | 0.9518 | 0.6553 | 0.7937 |
| MCH, pg  |           |      |     |     |      |    |     |     |
| 4 weeks  | 63.8     | 65.7 | 0.896 | 62.8³ | 66.7⁺ | 0.896 | 0.1668 | 0.0142 | 0.4434 |
| 6 weeks  | 65.2     | 62.3 | 0.988 | 62.6 | 65.2 | 0.988 | 0.1017 | 0.1005 | 0.7879 |
| MCHC, g/dL|           |      |     |     |      |    |     |     |
| 4 weeks  | 37.0     | 37.8 | 0.736 | 36.5 | 38.2 | 0.736 | 0.4656 | 0.1157 | 0.1570 |
| 6 weeks  | 37.6     | 36.2 | 0.652 | 36.0 | 37.8 | 0.652 | 0.1623 | 0.0913 | 0.7163 |
| PLT, 10³/µL|           |      |     |     |      |    |     |     |
| 4 weeks  | 8.31     | 12.2 | 2.17 | 11.0 | 9.47 | 2.17 | 0.2440 | 0.6321 | 0.4057 |
| 6 weeks  | 7.67     | 17.3 | 9.900 | 19.8 | 5.22 | 9.900 | 0.5106 | 0.3297 | 0.3475 |

Notes: T-2 toxin; A: antioxidants; T × A: the interaction of T-2 toxin and antioxidants. WBC: White blood cells, RBC: Erythrocytes, HB: Haemoglobin, HT: Haematocrit, MCV: HT/RBC, MCH: HB/RBC, MCHC: HB/ HT, PLT: Platelets. SEM1: standard error of means of T-2 toxin. SEM2: standard error of means of antioxidants.
Table 7. Effect of T-2 toxin and antioxidants on severity and IAW in grower geese at week 6.

| Treatment | Line | Sex | Significance |
|-----------|------|-----|--------------|
| T-2       | TxA  | Line×Sex | 0.3402  |
| T-2       | TxA  | Sex   | 0.1697   |
| T-2       | TxA  | A     | 0.7373   |
| Antioxidant | CL×Sex | Line×Sex | 0.3713  |
| Antioxidant | CL×Sex | Sex | 0.3713   |
| Antioxidant | CL×Sex | A | 0.3713   |

Notes: CON: control; AN: antioxidants; T2: T-2 toxin; AL: angel-winged line; NL: normal-winged line; CL: commercial line; T: T-2 toxin; A: antioxidants; Line × Sex: the interaction of line and sex; T × Line × Sex: the interaction among T-2 toxin, line, and sex; A × Line × Sex: the interaction among antioxidants, line, and sex. SEM1: standard error of means of T-2 toxin and antioxidants. SEM2: standard error of means of genetic line. SEM3: standard error of means of sex. a, b Means in the same row under line without the same superscripts differ significantly (p < 0.05). p Means in the same row under sex without the same superscripts differ significantly (p < 0.05). e, f Means in the same row under T-2 toxin and antioxidants without the same superscripts differ significantly (p < 0.05).
ganders to 4 angel-winged geese and found that among the goslings from normal-winged parents, 232 (85.3%) were normal-winged and 40 (14.7%) were angel-winged; of those from angel-winged parents, 16 (47.0%) were normal-winged and 18 (53.0%) were angel-winged. The authors, therefore, conclude that if inheritance is involved, then the character (AW) must be affected by more than one pair of genes. Fan et al. (2006) indicated that the overall average of IAW was 8.7%. There was no significant difference in IAW among the stocking densities. IAW was higher in White Chinese geese (13.6%) than in White Roman geese (6.9%).

The LHIAWs \((h^2 = (\chi_R - \chi_I)/i)\) of body weight and egg production lines were 0.39 and 0.03, respectively. A pooled LHIAW of 0.31 was also estimated. These results imply that selection for heavy body weight might induce IAW (Lin et al. 2008). The studies showed that different lines exhibited significant differences in IAW, as seen in Table 7. AL birds had a significantly higher SSAR and IAW than NL birds at 6 weeks. The results support previous observations that SSAR and IAW increased when birds were selected by appearance. Compared to males, female geese had higher SSAR and IAW values at 6 weeks (Table 7). This study is consistent with previous observations that elevated SSAR and IAW values, especially due to selection, increase the selection intensity of geese in such a manner as to decrease IAW. According to radiographs, abnormal ligaments at the carpal joint might be attributed to the twisted wings (Fan et al. 2006).

5. Conclusion

ALK plays a role in the calcification of cartilage and bone. The ALK level in the T-2 toxin group was lower than that of the control group. SSAR in the T-2 toxin and antioxidant groups was higher than that of the single T-2 toxin or antioxidant group at 6 weeks of age. IAW in the T-2 toxin and antioxidant groups was higher than that of the other groups at 6 weeks. The SSAR and IAW values in the male group were lower than those of the female group at 6 weeks.

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Disclosure statement

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