

## EFFICACY OF SALICYLIC AND ASCORBIC ACIDS TO REDUCE THE EFFECT OF TRANSPORT STRESS ON LIVE WEIGHT, SOME BLOOD CHARACTERISTICS AND INTESTINAL VILLI OF BROILER CHICKENS

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### ABSTRACT

This experiment was conducted and aimed to reducing mortality and losing the weight of birds transported to the slaughter house through reducing or Transport stress removal and thus to reduce financial losses. 150 birds used almost equal weights that were divided into three treatment groups and by five replicates for each treatment is control treatment (T1) without giving anything and treatment with salicylic acid concentration 1 g / liter of drinking water (T2) and treatment with ascorbic acid (T3) concentration 0.5 g / Liter of drinking water provided 24 hours before its transport. The transport process resulted in a highly significant decrease ( $P \leq 0.01$ ) in body weight in treatment T1 as it reached 4.66%, while it reached 1.68 and 1.09% in treatment T2 and T3, respectively. The transport process led to an increase in body temperature, H / L ratio, the concentration of glucose, aspartate transaminase (AST) and alanine transaminase (ALT) in the blood serum, as the transport process led to a very significant decrease ( $P \leq 0.01$ ) in the cholesterol concentration, Uric acid, intestinal villi length and crypts depth. T2 and T3 treatments showed their ability to reduce transport stress, especially T3, where they outperformed T1 in most traits. It is concluded from the study that adding ascorbic acid by 0.5 g / L drinking water 24 hours before transporting the chicks to the slaughter house reduces the transport stress and lessen financial losses, so it is recommended to use it.

Keywords: H/L, aspartate transaminase, alanine transaminase, cholesterol, uric acid

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فعالية حامضي السالسليك و الاسكوربيك للتقليل من تأثير اجهاد النقل على الوزن الحي ، بعض صفات الدم والزغابات المعويه

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### المستخلص

اجريت هذه التجربة التي هدفت الى خفض الهلاكات وفقد وزن الطيور المنقولة الى المجزرة و ذلك من خلال تخفيف او ازالة اجهاد النقل و بالتالي لخفض الخسائر الماليه. استخدم 150 طير متجانسة الاوزان التي تم تقسيمها الى ثلاث مجاميع من المعاملات و بواقع خمس مكررات لكل معاملة هي معاملة السيطرة (T1) بدون اعطائها شيئا و المعاملة بحامض السالسليك تركيز 1 غم / لتر ماء (T2) و المعاملة بحامض الاسكوربيك (T3) تركيز 0.5 غم / لتر ماء قدم قبل 24 ساعة من نقلها. ادت عملية النقل الى انخفاض عالي المعنوية ( $P \leq 0.01$ ) في وزن الجسم في المعاملة T1 اذ بلغت نسبته 4.66 % في حين بلغت نسبته 1.68 و 1.09 % في المعاملة T2 و T3 على التوالي. عملية النقل ادت الى زيادة درجة حرارة الجسم ، نسبة H/L ، تركيز الكلوكوز و انزيمات aspartate transaminase (AST) و alanine transaminase (ALT) في مصل الدم ، كما ادت عملية النقل الى انخفاض عالي المعنوية ( $P \leq 0.01$ ) في تركيز الكولسترول، حامض اليوريك ، طول الزغابات و عمق الخبايا . أظهرت معاملات T2 و T3 قدرتها في التقليل من اجهاد النقل خصوصا T3 حيث تفوقتا على T1 في اغلب الصفات. يستنتج من الدراسه ان اضافة حامض الاسكوربيك بمقدار 0.5 غم/لتر ماء شرب 24 ساعه قبل نقل الافراخ الي المجزرة خفف من اجهاد النقل وقلل الخسائر الماليه لذا ينصح باستخدامه.

الكلمات المفتاحيه: نسبة الخلايا المتغايره/ الخلايا اللمفيه، انزيم اسبارتيت ترانزامينيز، انزيم الانين ترانزامينيز، كولسترول، حامض اليوريك

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## INTRODUCTION

Poultry transport is one of the key links in the poultry industry (30). As all types of birds used in intensive production systems are transported at least twice during their lifetime and distances ranging from several kilometers to tens of hours' trips (39, 28, 23, 38). Birds during this period are exposed to several stressful factors that lead to the reduce immunity defense of body that adversely affect the productive efficiency of these birds (2, 17, 26, 10) which leads to economic losses such as mortalities, Weight loss, low nutritional value and specific quality of meat produced as a result of exposure to wounds, fractures, bruises, and dehydration (13, 40, 16). Any practical solution to the problems of the transportation process should include improving ventilation systems and controlling temperature during transportation and that by using modern transport vehicles that have a Mechanical ventilation and temperature control systems (15). The obstacle preventing the use of such vehicles is their high cost (18). Physiological transport stress can be reduced by using some vitamins and minerals Such as ascorbic acid and vitamin E or the use of some tranquilizers that have low prices and are easy to obtain and use (1, 21, 22, 2, 27, 31, 8). This study aims to know the effectiveness of the use of salicylic acid and ascorbic acid in reducing the negative effects of physiological stress resulting from transportation and thus reducing financial and economic losses.

## MATERIALS AND METHODS

One hundred and fifty Ross 308 broiler chickens were used in this experiment, 35 days of age, mostly equal weights. They were divided into three groups with five replications per treatment (10 birds per replicate) as follow: The first treatment is T1, control without addition, the second treatment, T2, the addition of salicylic acid with drinking water at a concentration of 1 g / litre, the third treatment, T3 addition of ascorbic acid with drinking water at a concentration of 0.5 g / l water. Both compounds are added 24 hours before the transport. Feed was removed from the birds 6 hours before the transport, and the birds were then placed in plastic cages for transporting. The birds were transported for a distance of 120 km for a period of 150

minutes. During the transport process, temperature and humidity were recorded every 15 minutes using an electronic thermometer (Figure 1). The weight of the birds was taken before and after the transport by a ground electronic balance and the lost weight and its percentage were calculated using the following equations: lost weight during transportation = weight before transportation - weight after transportation, lost weight ratio = lost weight during transportation / weight of birds before transport x 100. As for the economic losses, it was calculated on the basis of the price of 1 kg live body weight equal to 2500 Iraqi dinars. As for the physiological characteristics, it was measured for 10 birds per treatment before and after the transport, where the temperature was measured by inserting a digital thermometer sensor type TCH-4 Mini 5 cm depth inside the rectum. When the packed cell volume was measured according to (11) and hemoglobin using the Drabkins reagent produced by the Swiss company Agappe, the differential count of white blood cells was measured according to (14). The blood serum traits were measured according to the instructions of the German human company produced kits. Statistical analysis of the data was performed using the (34) and using the complete Randomized Design (CRD). The data were analyzed according to one-way variance analysis and to test the differences between the factors in the experiment, used the Duncan's Multiple Range Test (20).

## RESULTS AND DISCUSSION

Table 1 shows that there was no significant differences between treatments in body weight before transport, while significantly ( $P \leq 0.01$ ) decreased after transport in control treatment (T1) only. T2 and T3 were significantly superior to T1 in body weight after transport, lost weight, weight loss and lower economic losses. . Table 2 indicates a highly significant ( $P \leq 0.01$ ) increase in body temperature after transport, amount and percentage of high body temperature and heat tolerance factor in T1 and T2 compared to T3. Heterophil percentage and H/L ratio exhibited significant differences in favor of T2 and T3 comparing with T1 before and after transportation, and the percentage of change, whereas this did not happen in the lymphocytes. In general,

heterophil and H/L ratio were increased, whereas lymphocyte decreased after transportation (Table 3). The treatments with salicylic and ascorbic acids resulted in reducing the stress that birds are exposed to before transport and during transportation in a highly significant way ( $P \leq 0.01$ ), which can be inferred from H/L ratio. , glucose, cholesterol, and uric acid significantly decreased while increased total protein before transport in T2 and T3 compared to T1, while glucose increased and cholesterol, total protein and uric acid decreased in the three treatments after transfer (Table 4). The AST decreased significantly before the transport and the ALT significantly increased after the transport in the T2 and T3 compared to the T1 (Table 5). The length of villi and the Crypts depth in T2 and T3 were significantly lower after transport compared to T1, whereas villi length decreased significantly after transport compared to before transport in all three treatments (Table 6). The process of fasting the chicks before transportation reduces the glycogen and impedes the formation of liver proteins due to stress hormones in addition to destruction the body tissues, thereby reducing body weight (3, 5). The exposure of chickens to stressful factors and high temperatures that cause an increase in the numbers of mature and immature heterophil cells (37, 35) and a decrease in the number of lymphocytes and thus an increase in the H/L ratio, as well as an increase in the level of catechol amines and corticosteroids and glucagon hormones , which works to break down the glycogen by glycogenolysis process, also works to form glucose from non-carbohydrate sources by the process of gluconeogenesis, which leads to high blood glucose level and low proteins (6, 7,

24). The reason for the decrease in the level of cholesterol and uric acid in the blood serum is due to the fasting of broiler before transport and consequently the low concentrations of these substances (19, 29, 32, 41). And that the reason for increasing the effectiveness of ALT and AST liver enzymes and increasing their concentration in the blood serum may be due to the destruction of cells and tissues, and thus the transmission of these enzymes to the blood serum or the reason for the high concentration may be due to high temperatures (25, 4). As for the reason for the decrease in villi length, it may be due to stopping the secretion of mucin by the action of the corticosteroids, which leads to the protection of villi, that leads to an increase in the number of harmful coliform bacteria in the gut and decrease numbers of beneficial anaerobic bacteria that produce a group of vitamins, mineral elements, amino acids and volatile fatty acids, which are a source of food for the intestinal cells, for their growth, maintenance, regeneration and increased secretion of the myosin layer that protects them from external and internal toxins. The presence of short-chain fatty acids and persistence Its production by beneficial bacteria is essential for sustaining and vitality of crypts of Lieberkuhn (33, 12, 9), or the reason for this significant reduction may be due to pre-transport fasting (36). This experiment confirmed the results of previous relevant studies, the roles of ascorbic and salicylic acids in overcoming stress factors, including transport stress that birds were exposed to during transportation. We suggest providing ascorbic acid 0.5 g / l drinking water or salicylic acid 1 g / l drinking water 24 hours before transporting broiler chickens to the slaughterhouse.

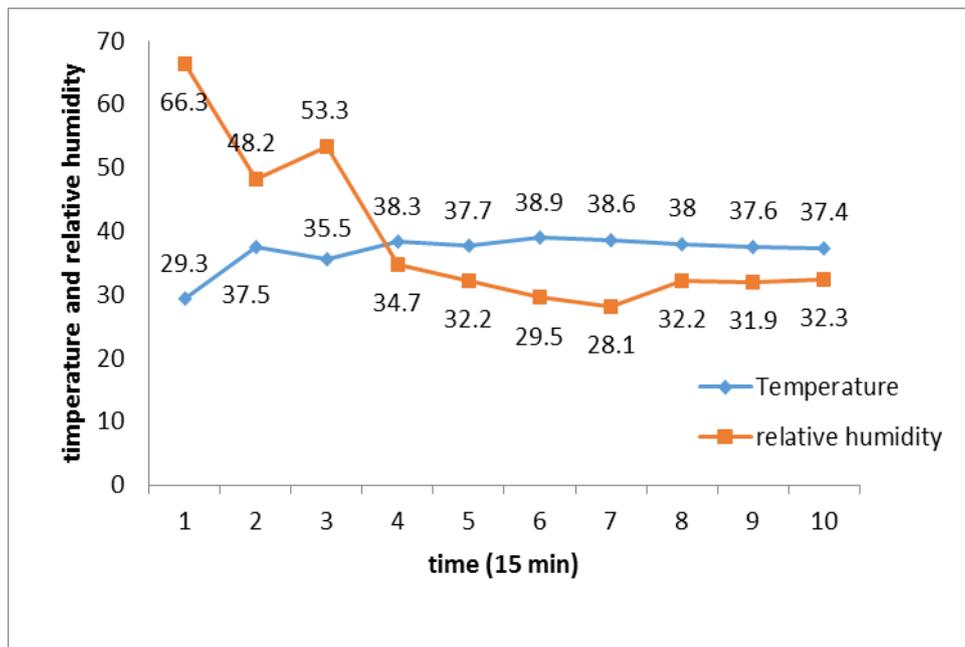


Figure 1. Temperature (°C) and relative humidity (%) recorded each 15 minutes during transportation of birds

Table 1. Using of salicylic and ascorbic acids to reduce the effect of transport stress on live weight (gm) of broiler chickens

| Treatments | Weight           |                            | Weight loss (g)          | Weight loss (%)        | Economical loss (ID)      | Sig. level |
|------------|------------------|----------------------------|--------------------------|------------------------|---------------------------|------------|
|            | Before transport | After transport            |                          |                        |                           |            |
| T1         | 2198.0 ± 19.7    | 2095.4 ± 19.3 <sup>b</sup> | 102.6 ± 4.2 <sup>c</sup> | 4.7 ± 0.2 <sup>c</sup> | 256.5 ± 10.5 <sup>c</sup> | *          |
| T2         | 2208.8 ± 28.4    | 2171.6 ± 27.5 <sup>a</sup> | 37.2 ± 5.5 <sup>b</sup>  | 1.7 ± 0.3 <sup>b</sup> | 93.0 ± 13.9 <sup>b</sup>  | N.S        |
| T3         | 2218.4 ± 11.5    | 2194.0 ± 11.1 <sup>a</sup> | 24.4 ± 1.7 <sup>a</sup>  | 1.1 ± 0.1 <sup>a</sup> | 61.0 ± 4.2 <sup>a</sup>   | N.S        |
| Sig. level | N.S              | **                         | **                       | **                     | **                        | ---        |

Mean ± standard error, T1, T2 and T3 means control treatment and addition of salicylic acid (1 g) and ascorbic (0.5 g) / liter of water 24 hours before transport, respectively, N.S. And \* and \*\* mean that there are no significant and no significant effects ( $P \leq 0.05$ ) and ( $P \leq 0.01$ ) respectively. Different small letters indicate the presence of significant differences, I.D means Iraqi Dinar

Table 2. Effect of salicylic and ascorbic acids to reduce the effect of transport stress on body temperature (° C) of broiler chickens

| treatments  | Body temperature  |                         | Amount of increase   | Increase (%)         | Heat tolerance       | Sig. levels |
|-------------|-------------------|-------------------------|----------------------|----------------------|----------------------|-------------|
|             | before transports | After transport         |                      |                      |                      |             |
| T1          | 41.7±0.1          | 42.0±0.1 <sup>a</sup>   | 1.2±0.2 <sup>a</sup> | 2.9±0.4 <sup>a</sup> | 0.5±0.1 <sup>a</sup> | *           |
| T2          | 41.4±0.2          | 42.3 ± 0.2 <sup>b</sup> | 0.9±0.2 <sup>a</sup> | 2.3±0.5 <sup>a</sup> | 0.4±0.1 <sup>a</sup> | *           |
| T3          | 41.4±0.2          | 41.8±0.2 <sup>c</sup>   | 0.4±0.1 <sup>b</sup> | 1.0±0.2 <sup>b</sup> | 0.2±0.0 <sup>b</sup> | N.S         |
| Sig. levels | N.S               | **                      | **                   | **                   | **                   | —           |

Mean ± standard error, T1, T2 and T3 means control treatment and addition of salicylic acid (1 g) and ascorbic (0.5 g) / liter of water 24 hours before transport, respectively, N.S. And \* and \*\* mean that there are no significant and no significant effects ( $P \leq 0.05$ ) and ( $P \leq 0.01$ ) respectively. Different small letters indicate the presence of significant differences, Sig. Level means significant levels

**Table 3. Efficacy of ascorbic and salicylic acids to reduce the impact of transport stress on lymphocytes, heterophil and heterophil/lymphocyte ratio**

| treatments                     | Before transport      | After transport        | Amount of change     | Change (%)            | Sig. levels |
|--------------------------------|-----------------------|------------------------|----------------------|-----------------------|-------------|
| <b>heterophil (%)</b>          |                       |                        |                      |                       |             |
| T1                             | 35.0±0.6 <sup>a</sup> | 43.1±1.0 <sup>a</sup>  | 8.0±0.5 <sup>a</sup> | 22.8±2.1 <sup>a</sup> | **          |
| T2                             | 33.5±0.4 <sup>a</sup> | 39.1±0.7 <sup>b</sup>  | 5.5±0.8 <sup>b</sup> | 16.5±2.4 <sup>b</sup> | **          |
| T3                             | 31.3±0.9 <sup>b</sup> | 33.2±0.9 <sup>c</sup>  | 1.8±0.3 <sup>c</sup> | 5.9±1.1 <sup>c</sup>  | NS          |
| Sig. levels                    | **                    | **                     | **                   | **                    | —           |
| <b>lymphocyte (%)</b>          |                       |                        |                      |                       |             |
| T1                             | 55.1±1.1              | 50.8±1.0 <sup>ab</sup> | 4.2±0.5              | 7.6±1.0               | *           |
| T2                             | 55.8±0.6              | 49.7±0.8 <sup>b</sup>  | 6.2±0.1              | 11.0±1.7              | **          |
| T3                             | 56.5±0.7              | 53.0±0.7 <sup>a</sup>  | 3.5±0.7              | 6.2±1.2               | *           |
| Sig. levels                    | NS                    | *                      | NS                   | NS                    | —           |
| <b>heterophil : lymphocyte</b> |                       |                        |                      |                       |             |
| T1                             | 0.6±0.0 <sup>a</sup>  | 0.8±0.0 <sup>a</sup>   | 0.2±0.0 <sup>a</sup> | 32.9±1.3 <sup>a</sup> | **          |
| T2                             | 0.6±0.0 <sup>b</sup>  | 0.8±0.0 <sup>b</sup>   | 0.2±0.1 <sup>a</sup> | 31.0±2.4 <sup>a</sup> | **          |
| T3                             | 0.6±0.1 <sup>c</sup>  | 0.6±0.0 <sup>c</sup>   | 0.1±0.0 <sup>b</sup> | 12.9±1.5 <sup>b</sup> | *           |
| Sig. levels                    | **                    | **                     | **                   | **                    | —           |

Mean ± standard error, T1, T2 and T3 means control treatment and addition of salicylic acid (1 g) and ascorbic (0.5 g) / liter of water 24 hours before transport, respectively, N.S. And \* and \*\* mean that there are no significant and no significant effects ( $P \leq 0.05$ ) and ( $P \leq 0.01$ ) respectively. Different lowercase letters indicate the presence of significant differences, Sig. Level means significant levels

**Table 4. Effect of salicylic and ascorbic acids on some blood serum traits of transport stressed broiler chickens**

| Treatments                        | Before transport       | After transport        | Amount of change      | Change (%)            | Sig. levels |
|-----------------------------------|------------------------|------------------------|-----------------------|-----------------------|-------------|
| <b>Glucose (mg / decL.)</b>       |                        |                        |                       |                       |             |
| T1                                | 165.1±3.2 <sup>a</sup> | 205.3±2.8 <sup>a</sup> | 40.3±1.3 <sup>a</sup> | 24.5±1.1 <sup>a</sup> | **          |
| T2                                | 150.5±2.5 <sup>b</sup> | 174.3±1.1 <sup>b</sup> | 23.8±1.6 <sup>b</sup> | 15.9±1.3 <sup>b</sup> | **          |
| T3                                | 119.1±3.3 <sup>c</sup> | 129.0±4.6 <sup>c</sup> | 9.9±1.4 <sup>c</sup>  | 8.3±1.0 <sup>c</sup>  | N.S         |
| Sig. levels                       | **                     | **                     | **                    | **                    | —           |
| <b>Cholesterol (mg / decL.)</b>   |                        |                        |                       |                       |             |
| T1                                | 229.4±3.7 <sup>a</sup> | 142.5±4.3 <sup>a</sup> | 87.0±1.4 <sup>a</sup> | 38.0±1.0 <sup>a</sup> | **          |
| T2                                | 182.3±4.6 <sup>c</sup> | 110.7±8.3 <sup>c</sup> | 71.6±4.1 <sup>b</sup> | 39.6±3.1 <sup>a</sup> | **          |
| T3                                | 196.9±3.3 <sup>b</sup> | 167.0±4.9 <sup>b</sup> | 29.9±2.7 <sup>c</sup> | 15.2±1.5 <sup>b</sup> | **          |
| Sig. levels                       | **                     | **                     | **                    | **                    | —           |
| <b>Total protein (mg / decL.)</b> |                        |                        |                       |                       |             |
| T1                                | 2.10±0.14 <sup>b</sup> | 1.72±0.13 <sup>b</sup> | 0.4±0.0 <sup>a</sup>  | 17.9±1.5 <sup>a</sup> | **          |
| T2                                | 2.5±0.4 <sup>b</sup>   | 2.2±0.1 <sup>b</sup>   | 0.3±0.0 <sup>a</sup>  | 12.1±0.6 <sup>b</sup> | **          |
| T3                                | 3.6±0.3 <sup>a</sup>   | 3.4±0.3 <sup>a</sup>   | 0.2±0.0 <sup>b</sup>  | 6.2±1.0 <sup>c</sup>  | N.S         |
| Sig. levels                       | **                     | **                     | **                    | **                    | —           |
| <b>Uric acid (mg / decL.)</b>     |                        |                        |                       |                       |             |
| T1                                | 13.3±0.6 <sup>a</sup>  | 6.5±0.6                | 6.8±0.2 <sup>a</sup>  | 50.3±2.4 <sup>a</sup> | **          |
| T2                                | 10.6±0.3 <sup>b</sup>  | 6.3±0.3                | 4.2±0.2 <sup>b</sup>  | 40.3±2.2 <sup>b</sup> | **          |
| T3                                | 10.0±0.55 <sup>b</sup> | 6.9±0.8                | 3.1±0.5 <sup>c</sup>  | 31.4±5.1 <sup>c</sup> | *           |
| Sig. levels                       | **                     | N.S                    | **                    | **                    | —           |

Mean ± standard error, T1, T2 and T3 means control treatment and addition of salicylic acid (1 g) and ascorbic (0.5 g) / liter of water 24 hours before transport, respectively, N.S. And \* and \*\* mean that there are no significant and no significant effects ( $P \leq 0.05$ ) and ( $P \leq 0.01$ ) respectively. Different lowercase letters indicate the presence of significant differences, Sig. Level means significant levels.

**Table 5. Effect of salicylic and ascorbic acids on alanine transaminase (ALT) and aspartate transaminase (AST) levels in blood serum (IU / L) of transport stressed-broiler chickens**

| Treatments | before transport      | After transport        | Amount of decrease   | Increase (%)          | Sig. level |
|------------|-----------------------|------------------------|----------------------|-----------------------|------------|
| <b>ALT</b> |                       |                        |                      |                       |            |
| T1         | 117.1±3.1             | 126.7±1.1 <sup>a</sup> | 9.6±2.2 <sup>a</sup> | 8.4±2.0 <sup>a</sup>  | **         |
| T2         | 110.4±0.7             | 114.3±3 <sup>b</sup>   | 3.9±0.8 <sup>b</sup> | 3.6±0.7 <sup>b</sup>  | *          |
| T3         | 111.0±0.8             | 114.5±1.6 <sup>b</sup> | 3.5±1.5 <sup>b</sup> | 3.1±0.8 <sup>b</sup>  | N.S        |
| Sig. level | N.S                   | **                     | *                    | *                     | —          |
| <b>AST</b> |                       |                        |                      |                       |            |
| T1         | 10.8±0.4 <sup>a</sup> | 14.0±0.7 <sup>a</sup>  | 3.2±0.6 <sup>a</sup> | 30.2±6.5 <sup>a</sup> | **         |
| T2         | 9.4±0.2 <sup>b</sup>  | 12.7±0.3 <sup>b</sup>  | 3.3±0.4 <sup>a</sup> | 35.7±5.2 <sup>a</sup> | **         |
| T3         | 8.8±0.2 <sup>b</sup>  | 9.9±0.2 <sup>c</sup>   | 1.1±0.2 <sup>b</sup> | 11.9±2.6 <sup>b</sup> | *          |
| Sig. level | **                    | **                     | **                   | **                    | —          |

Mean ± standard error, T1, T2 and T3 means control treatment and addition of salicylic acid (1 g) and ascorbic (0.5 g) / liter of water 24 hours before transport, respectively, N.S. And \* and \*\* mean that there are no significant and no significant effects ( $P \leq 0.05$ ) and ( $P \leq 0.01$ ) respectively. Different lowercase letters indicate the presence of significant differences, Sig. Level means significant levels

**Table 6. Effect of salicylic and ascorbic acids on villi length and crypts depth (Mic.) in the jejunum of the transported stressed-broiler chickens**

| Treatments             | Before transport | After transport         | Amount of decrease      | Decrease (%)          | Sig. level |
|------------------------|------------------|-------------------------|-------------------------|-----------------------|------------|
| <b>Length of villi</b> |                  |                         |                         |                       |            |
| T1                     | 684.8 ±22.0      | 346.8±18.9 <sup>b</sup> | 338.0±25.2 <sup>a</sup> | 49.3±2.8 <sup>a</sup> | **         |
| T2                     | 729.0±33.1       | 405.0±30.8 <sup>b</sup> | 324.0±22.6 <sup>a</sup> | 44.5±2.7 <sup>a</sup> | **         |
| T3                     | 712.8±30.3       | 502.7±14.2 <sup>a</sup> | 210.1±39.6 <sup>b</sup> | 28.8±4.2 <sup>b</sup> | **         |
| Sig. level             | N.S              | **                      | *                       | **                    | —          |
| <b>crypts depth</b>    |                  |                         |                         |                       |            |
| T1                     | 184.0±15.6       | 124.2±10.8 <sup>b</sup> | 59.8±9.5 <sup>a</sup>   | 32.1±3.9 <sup>a</sup> | *          |
| T2                     | 200.0±13.1       | 181.0±19.0 <sup>a</sup> | 19.0±9.7 <sup>b</sup>   | 10.2±5.7 <sup>b</sup> | N.S        |
| T3                     | 181.6±11.8       | 157.4±5.7 <sup>ab</sup> | 24.2±6.5 <sup>b</sup>   | 12.5±3.3 <sup>b</sup> | N.S        |
| Sig. level             | N.S              | *                       | *                       | **                    | —          |

Mean ± standard error, T1, T2 and T3 means control treatment and addition of salicylic acid (1 g) and ascorbic (0.5 g) / liter of water 24 hours before transport, respectively, N.S. And \* and \*\* mean that there are no significant and no significant effects ( $P \leq 0.05$ ) and ( $P \leq 0.01$ ) respectively. Different lowercase letters indicate the presence of significant differences, Sig. Level means significant levels

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