



THE ROLE OF NUTRITIVE SOLUTIONS DURING EMBRYOGENESIS IN IMPROVING HATCHABILITY AND POST-HATCH GROWTH PERFORMANCE

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Received: 19/ 01/2016

Accepted: 09/02/2016

ABSTRACT: The purpose of this study was to testing the effects of spraying hatching eggs with some nutritive solutions i.e., vitamin C (Ascorbic acid, AA) and L-Carnitine (L-Car) on hatchability, embryonic mortalities, chick quality, some blood traits and post-hatch growth performance. A total number of 3600 hatching eggs were obtained from Cobb broiler breeder chickens at 35 wks of age. Eggs were randomly divided into 6 groups (600 eggs / group) with four replicates containing 150 for each. The eggs were sprayed during embryogenesis at 1st, 7th, 14th and 18th d of incubation. Eggs sprayed with AA and L-Car in: 1) no spraying without any supplementation (control group), 2) distilled water (sham group), 3) distilled water + 4 g/L AA., 4) distilled water + 6 g/L AA, 5) distilled water + 4 g/L L-Car, 6) distilled water + 6 g/L L-Car. At the end of hatching process, all un-hatched eggs were broken to estimate embryonic mortalities. Results revealed that spraying eggs with 6 g/L AA resulted in a significant ($P \leq 0.05$) improvement of hatchability traits. However, embryonic mortalities were almost significantly ($P \leq 0.05$) decreased due to application either AA or L-Car compared to control and sham groups. Furthermore, chick quality traits and subsequent growth performance markedly ($P \leq 0.05$) improved with application of nutritive solutions. Moreover, all blood traits were significantly ($P \leq 0.05$) affected due to positive effects of nutritive solutions, where the lowest corticosterone concentration observed for embryos resulted from eggs sprayed with either AA or L-Car, as judged by the decrease of stress. However, the highest RBC's count recorded for chicks produced from eggs sprayed with 4 g/L AA, while, the highest value of hemoglobin concentration detected for chicks resulted from eggs sprayed with 6 g/L AA compared to other experimental groups. Furthermore, subsequent growth performance was severely improved due to spraying eggs with nutritive solutions. Most of carcass traits significantly ($P \leq 0.05$) affected due to spraying eggs with L-Car and AA, especially regarding with abdominal fat percentage, which significantly decreased. It could be concluded that, spraying hatching Cobb broiler breeder eggs with either AA or L-Car with doses 4 or 6 g/L on 1st, 7th, 14th and 18th d of embryogenesis markedly improved hatchability, chick quality traits and performance of progeny. Hence spraying eggs with these nutrients could be used as a successful method to reducing embryonic mortalities and improving early post-hatch performance.

Key Words: Ascorbic acid, L-carnitine, hatchability, post-hatch growth performance

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INRODUCTION

It is well known that the supply of day-old chicks is very important for the success of the poultry production chain. However, fertility and hatchability are two major parameters that highly influence the supply of day-old chicks (**King'ori, 2011**). Furthermore, low hatchability negatively affects productivity and animal welfare in the poultry industry. Embryonic viability is influenced by a series of factors such as nutrition, hatching technology, egg quality and genetics. In this sense, macroscopically, the egg is a simple structure with three major components (shell, albumen, and yolk), demonstrating unique characteristics and playing specific roles during incubation, where each egg is built with a complete capacity to produce a perfect new organism. Avian embryos develop and grow from energy and nutrients stored in the egg by the hen (**Vieira, 2007**). The nutrient content of the hatching egg influences the development and growth of embryos during incubation and the post-hatch performance of chicks (**Shafey et al., 2013**). Unlike the mammalian embryo, the avian embryo has a finite amount of energy and nutrients for growth and development invested by the broiler breeder hen. Therefore, research showed that a solutions which includes macronutrients i.e., salts, minerals and vitamins elevates the nutrients in the egg and availability for the embryo. In *ovo* supplementation of nutrients may help late-term embryos to overcome the constraints of limited egg nutrients (**Foye et al., 2006**). In *ovo* feeding improves the hatchability of eggs, the viability and weight of chicks at hatch, intestinal development, growth and feed efficiency of hatched chicks (**Tako et al., 2004 and Shafey et al., 2012**), body weight at marketing age, and carcass quality (**Selim et al., 2012**). Therefore, providing sufficient nutrients in the egg is a good starting point for the hatching chicks.

However, spraying eggs during embryogenesis is one of the tools used to improve hatchability and chick quality at hatch. Vitamin C (ascorbic acid, AA) does not exist in a freshly laid egg and it appears only on 3rd - 4th d of incubation as a result of endogenous biosynthesis by the developing embryo. Spraying fertile Muscovy duck eggs with AA solution (30 g/L) twice times daily during the last 3 wks of embryogenesis had improve hatchability, immunity as well as growth performance of hatch (**Ghonim et al., 2009**). Also, **Nowaczewski et al. (2011)** indicated that in *ovo* injection of 6 mg of AA per egg on the 20th d of incubation (8 d before hatch) improved hatchability rate of duckling eggs. In a study reported by **Doaa et al. (2014)** showed that the embryonic mortality, fertility hatchability, WBC's count, body weight, gain and feed conversion were significantly improved by spraying eggs with 20 or 30 g AA compared to the control or distilled water during embryogenesis. However, L-Car is biosynthesized from the essential amino acids lysine and methionine in the presence of ferrous ions (Fe⁺⁺) and three vitamins: AA, niacin and pyridoxine that are required as cofactors for the enzymes involved in the metabolic pathway (**Rebouche, 1992**). The L- Car acts as an antioxidant that ultimately results in a decrease in reactive oxygen species (ROS) by removing excessive levels of intracellular acetyl CoA, which induces mitochondrial ROS production. Thereof, it may work as an antioxidant to scavenge free radicals (**Agarwal et al., 2005**). Chick embryo may have limited capability to synthesize L-Car during incubation (**Casillas and Newburgh, 1969**). Thus, the presence of L-Car in the fertile egg may decrease embryonic mortality by reducing oxidative stress during the hatch process, thereby increasing hatch rate (**Vicari and Calogero, 2001**). In addition to its role as an antioxidant, L-Car transports long chain

fatty acids across mitochondrial membranes for β -oxidation of fatty acids. In such situations, exogenous supplementation of L-Car could prove advantageous (Buyse *et al.*, 2001), and could in turn be used by the chick during hatching. For this purposes, the present study was undertaken to examine the effects of spraying Cobb broiler breeder eggs with either AA or L-Car during the embryogenesis on hatchability, embryonic mortality, chick quality traits, some blood parameters, carcass traits as well as subsequent progeny performance.

MATERIALS AND METHODS

Site and the aim of study: This study was carried out at Egyptian Poultry Company belonging to Giza Governorate, Egypt. The experiment was started on October 2014 and terminated on November 2014. The main objective was to examine the effects of spraying Cobb broiler breeder eggs with either AA or L-Car during the embryogenesis on hatchability, embryonic mortality, chick quality traits, some blood parameters, carcass traits as well as subsequent progeny performance.

Experimental procedure: A total number of 3600 eggs were collected from commercial flock of Cobb broiler breeder strain at 35 wks of age. All eggs were weighed on collection day and allocated to mean initial egg mass with similar for each group (60.48 g \pm 0.3). Eggs were divided into to 6 groups (600 eggs / group) with four replicates containing 150 for each. Eggs sprayed with AA and L-Car on 1st, 7th, 14th and 18th d during embryogenesis period as follows:

- T1:- 0 % AA and L-Car (without any treatment, negative control group)
- T2:- distilled water (sham group, positive control group)
- T3:- distilled water + 4 g/L AA
- T4:- distilled water + 6 g/L AA
- T5:- distilled water + 4 g /L L-Car
- T6:- distilled water + 6 g/L L-Car

Incubator and egg weight loss: The incubator used was a Chick Master Ultra Model, with digital temperature and humidity systems regulator and the setter was operated at 99.5 (\pm 0.2 $^{\circ}$ F) dry-bulb temperatures and 85.0 (\pm 0.2 $^{\circ}$ F) wet-bulb temperatures. Incubator was adjusted to turn the eggs once per hour at $^{\circ}$ 45, and stopped at 18 d of incubation. At the end of incubation period on 18th d, all eggs were transferred from setter trays to hatcher baskets. The Hatcher was operated at 98.5 (\pm 0.2 $^{\circ}$ F) dry bulb temperature and 90.0 (\pm 0.2 $^{\circ}$ F) wet bulb temperature from 19-21 d. The eggs were weighed before setting and examined to remove cracked and abnormal eggs. The eggs were weighted again at 18th d of incubation to measure egg weight loss. The weight loss calculated as a percentage of the initial egg weight. The following equation was used in calculation $(W_0 - W_{18}) / W_0 \times 100$, where W_0 is weight at setting and W_{18} is weight on d 18th of incubation (Tona *et al.*, 2001).

Hatchability, egg candling and embryonic mortality: At 7th and 14th d of incubation all eggs were candled and all clear eggs or early dead embryos were removed from the trays, opened and examined macroscopically. At the end of 18th d of incubation all eggs were candled again and those with evidence of living embryos were transferred from the incubation trays to the hatcher baskets. All un-hatched eggs were removed from the hatcher and broken for macroscopic examination to determine embryonic mortality. Embryonic mortalities were classified as early dead (1-7 d), middle dead (8-18 d) and late dead plus piped eggs (19-21 d). Early dead were differentiated by the absence of an egg tooth, and late dead were differentiated by evidence of the yolk sac entering the body cavity and the beak positioned to pip the air cell. Hatchability of total egg percentage was calculated as the number of hatched chicks per 100 eggs set for each replicate. Fertile hatchability percentage was calculated as

the number of hatched chicks per 100 fertile eggs set for each replicate.

Chick quality traits: All hatched chicks at the end of incubation period were recorded and wing banded individually after the entire batch of chicks hatched. All chicks were weighed and examined to score them for quality. Commercial chick quality (saleable chicks, grade A) was defined as being clean, dry and free from deformities (no skin lesion, well-formed beak, normal conformation of legs), completely sealed navel, and no yolk sac or residual membrane protruding from the navel area (Tona *et al.*, 2004). The un-saleable chicks were examined to score them for classification into grade B to the previous traits. Chick weight percentage was calculated as = [(chick weight / fresh egg weight) × 100], set for each replicate. All day-old chicks were scored with a Tona score as described by Tona *et al.* (2003).

Blood sampling: Blood samples were taken from embryo at 18th d of incubation. Each blood sample from each individual was divided into two samples in Eppendorf tubes. One was heparinized test tube by using Ethylenediamine-tetraacetate (EDTA) as an anticoagulant to study blood hematological parameters including white blood cells (WBC's), red blood cells (RBC's) and hemoglobin (Hb). The other part was non-heparinized to study blood biochemical constituents including total serum glucose (TSG), total serum aspartate transaminase (AST) and total serum alanine transaminase (ALT), total serum creatinine (TSC), total serum urea (TSU) and total serum corticosterone (CS) concentration by using the commercial kits.

Growth performance: A total number of 360 newly hatched chicks were obtained from all treatment groups. Chicks were randomly divided into 6 treatment groups (60 chicks / group) to measure their growth performance up to 35 d of age with four replicates containing 15 for each. All chicks were fed the same stander diet

According to **Cobb 500 broiler performance guide supplement (2012)**. Mean live body weight (LBW), body weight gain (BWG), feed intake (FI) and feed conversion ratio (FCR) was calculated for each group for 1, 14, 28 and 35 d of age. For each time period, (BWG) was calculated and expressed as gm per bird. Also, FCR (g FI /g BWG) was calculated by dividing total FI by total BWG in each group alone.

Carcass traits: At the end of 35 d of age 6 birds were randomly selected from each treatment to determine carcass traits. Chickens were fasted for approximately 12 h and then individually weighed, slaughtered (by severing the jugular vein), feathered and eviscerated. Weight of carcass, and internal organs were weighed and the corresponding percentages (% of body weight) were calculated.

Statistical analysis: The experiment was arranged in a complete randomized design. Then 1-way ANOVA was employed using the SPSS procedure (**SPSS for Windows Release 16, SPSS Inc. 2010**). The differences among groups were evaluated by Duncan's (**Duncan's, 1955**) multiple comparison tests. Differences were considered statistically significant at ($P \leq 0.05$). Statistical analysis of traits presented as percentages was carried out for arcsine values of their estimates. All obtained data were analyzed using the following model, $Y_{ij} = \mu + S_j + e_{ij}$

Where: Y_{ij} = response variable, μ = is the overall mean, S_j = fixed effects of nutritive solutions nutrients ($i=1-4$), e_{ij} = residual error

RESULTS

Egg weight loss, hatchability and embryonic mortalities: Table 2 shows the data concerning the average of egg weight loss during the setting phase of incubation (0-18d), hatchability and embryonic mortalities due to spraying eggs with different nutrient solutions. It is clear that fresh egg weight ranged from 60.43 to

60.53 g, with insignificant differences observed among the experimental groups. It is noted that loss of egg weight was significantly ($P < 0.05$) increased due to spraying eggs with either AA or L-Car compared to control and sham groups. However, there were significant ($P < 0.05$) differences observed among the experimental groups with regard to total hatchability and fertility hatchability, where the highest values were recorded for eggs sprayed with 6 g /L AA compared with other groups. However, embryonic mortalities including early, middle, late and total mortalities were characterized by the highest values of control and sham groups vs. eggs sprayed with different nutrient solutions. It is interesting to note that spraying eggs with neither AA nor L-Car improved hatchability and lowered embryonic mortalities.

Chick Quality traits: Data of chick quality traits are presented in Table 3. It is appeared that average of chick weight had ranged from 39.95 to 41.04 g, where the highest chick weight observed for chicks hatched from eggs sprayed with different nutritive solutions, competed to chicks hatched from either control or sham groups. Also, the relative weight exhibited significantly ($P < 0.05$) higher values for chicks treated with AA and L-Car than both control groups. The values of Tona score also indicated that there was improvement in chicks traits compared to control and sham groups. On the other hand, there were insignificant differences observed among groups with respect to the commercial chick quality traits including grade A and B. Form the present results it is interesting to note that better chick quality traits recorded for eggs sprayed with different nutrient solutions than control groups.

Chick blood parameters: Table 4 shows the effects of spraying eggs with different doses of nutritive solutions during embryogenesis on some blood parameters of embryo at d 18 of incubation. It is

observed that spraying eggs with 4 g/L AA significantly ($P \leq 0.05$) increased RBCs count circulation compared with other different nutrient solutions or control groups. While, WBC's count significantly ($P \leq 0.05$) recorded higher values for eggs sprayed with 6 g/L AA, than other groups. Results revealed that eggs sprayed with 6 g/L AA embryos have high level of Hb concentration, whereas the lowest was recorded for control groups. However, the lowest levels of TSG observed for eggs sprayed with either 4 or 6 g/L L-Car compared to AA or control groups. On the other hand, insignificant differences were observed for AST, ALT and TSC concentrations among the experimental groups. While, TSU concentration recorded higher ($P \leq 0.05$) value for eggs sprayed with 6 g/L AA, followed by 4 g/L AA, 4 g/L L-Car, distilled water, control and 6g/L-Car respectively. Concerning CS concentration the results indicated that due to application of spraying method lead to significant ($P \leq 0.05$) decreases of CS level compared to control or sham groups, where the lowest CS concentration observed for embryo sprayed with 6 g/L AA. It is interesting to note that most of measured blood traits were severely affected due to application of either AA or L-Car.

Subsequent growth performance: Results presented in Table 5 show the growth performance of chicks produced from eggs sprayed with different nutritive solutions for different growing periods. The data indicated that LBW at hatch, 28 and 35 d recorded the highest ($P \leq 0.05$) values for eggs sprayed with either AA or L-Car compared to control or sham groups. However, at 14 d chicks resulted from eggs sprayed with either 4 or 6 g/L L-Car exhibited higher LBW than other groups. Concerning the values of BWG, at 1-14 d chicks produced from eggs sprayed with either 4 or 6 g/L L-Car showed the highest ($P \leq 0.05$) values compared to either AA or control groups. While, at 15-28 d chicks resulted from eggs sprayed with 4 g/L AA

have high values compared to other experimental groups. However, at 29-35 d, BWG significantly recorded ($P \leq 0.05$) higher values for chicks produced from eggs sprayed with different nutritive solutions or distilled water group than chicks resulted from control group. For the whole period (1-35 d), the same trend was observed, where chicks produced from eggs sprayed with different nutritive solutions recorded significantly ($P \leq 0.05$) higher value than control or sham groups. Concerning FI, the chicks produced from eggs sprayed with 4 g/L AA consumed more feed compared to other treatments for the first period of fattening (1-14d). However, in the later period (15-28 d) chicks produced from eggs sprayed with 4 or 6 g/L L-Car consumed more feed compared to other experimental groups. While, at 15-28 d FI recorded higher values for chicks produced from eggs treated with either 4 or 6g/L-Car than other groups. On the other hand insignificant differences were observed for FI in the period between 29-35 d of age. For the whole period (1-35 d), chicks resulted from eggs sprayed with different nutritive solutions exhibited significantly ($P \leq 0.05$) higher value of FI than control or sham groups. Concerning, FCR the worst values observed for chicks produced from eggs treated with 4g/L and distilled water compared to other treatments. From 15-28 d the best value showed for chicks produced from control group compared to either AA or L-Car groups. While at 29-35 d, the chicks resulted from eggs treated with either 4 or 6 g/L L-Car significantly exhibited best value compared to AA or control groups. For the whole period (1-35 d) the best FCR observed for chicks produced from eggs sprayed with 6 g/L AA compared to other nutritive groups or control groups. From all this, it is obvious that spraying eggs during embryogenesis lead to improve subsequent growth performance.

Carcass traits: Data of carcass traits at 35 d of age as affected by spraying eggs with

different nutritive solutions are outlined in Table 6. Results of this study indicated that there were significant ($P \leq 0.05$) differences observed for the most carcass traits due to spraying eggs with either AA or L-Car, with exception, of spleen weight, dressing weight, total edible and total inedible parts weight percentages. The highest ($P \leq 0.05$) value of LBW observed for chickens produced from eggs sprayed with different nutritive solutions compared to both control groups. However, the highest liver weight percentage ($P \leq 0.05$) observed for chicks produced from eggs sprayed with 6 or 4 g/L AA compared to other groups. While, gizzard weight recorded significant ($P \leq 0.05$) higher values for control and sham groups followed by eggs sprayed with 6 g/L AA, 4 g/L L-Car, 6 g/L L-Car and 4 g/L AA respectively. Heart weight also exhibited higher value for both control groups than values recorded for nutritive solutions groups. The highest giblets parts weight observed for chicks produced from sham group and 6 g/L AA, while the lowest for chicks produced from eggs sprayed with 6 g/L L-Car. Moreover, there were severely ($P \leq 0.05$) decreases for abdominal fat percentage, where chicks produced from eggs sprayed with L-Car and AA recorded the lowest value compared to control and sham groups. The highest small intestine and cecum length observed for chicks sprayed with 4 g/ L L-Car, while the lowest detected for control and sham groups. It is observed that spraying eggs with either AA or L-Car progressively affected most carcass traits measured at 35 d of age, especially concerning abdominal fat percentage.

DISCUSSIONS

Egg weight loss, hatchability and embryonic mortalities: Results finding from this study indicate that spraying eggs with AA and L-Car solutions positively affected egg weight loss during embryogenesis. Water loss is a normal

process during incubation; where it is one of the physical factors that determine the success of incubation and optimum hatchability. Usually 12 to 14% of water is lost in broilers and turkeys eggs (**Rahn et al., 1981**). However, low or high water loss influences embryo development (**Rahn and Ar, 1974**), and consequently, egg hatchability (**Meir et al., 1984**). The highest egg loss observed due to application of AA and L-Car may be attributed to the increasing in egg shell conductance, where change the properties of cuticle of eggs were observed. One of the basic biological functions of the egg shell is movement of water vapor and respiratory gases. It is well known that, AA is a weak acid and the ability of diluted acid to interact with the egg shell cuticle, which may have cause a thinner cuticle or some physical changes in their morphology (**Burley and Vadehra, 1989**). The positive effect of L-Car is to increase water loss during embryogenesis. Further, the best hatchability obtained when egg weight loss is 10.71 % of their fresh weight from the time of lay to the time of embryo pipe out of the shell. However, weight loss smaller than 10 % and greater than 15 % of their fresh weight decreases hatchability (**Tullett, 1995**). This finding confirmed by different researchers indicated that the best hatchability are obtained when eggs loss 12 % of their fresh weight from the time of lay to the time the embryo pips the shell (**Meir and Ar, 1991**). **Askar (2012)** who indicated that quail eggs dipped in solution contains 8 g/L AA had the highest percentage of lost weight compared to untreated eggs. However, Hatchability traits markedly improved by application either AA or L-Car, where spraying eggs with these nutritive solutions were sufficient to support the hatching process. The improvement of hatchability may be due to the increasing of embryonic viability and decreasing the embryonic mortality. It is well known that AA may act as an anti-stress agent led to the reduction of CS

which has a negative impact in collagen synthesis and the metabolism of minerals and vitamin D (**Lohakare et al., 2005**). Also, AA could improve the biological functions of the egg shell to allow adequate movement of water vapor and respiratory gases, which helps embryos to exchange of respiratory gases and to break egg shell at hatch (**Tullett, 1990**). These results are consistent with **Ipek et al. (2004)** who found that the treated eggs with 3.0 mg/egg AA at different times of incubation improved hatchability. Moreover, **Ghonim et al. (2009)** who found that spraying Muscovy duck eggs with AA solution (30 g/L) twice times daily during the last 3 wks of incubation improve hatchability percentage. However, because eggs contain little or no L-Car (**Chiodi et al., 1994**), it has been suggested that the enhancement of L-Car in yolk would be beneficial for the development of the embryo (**Broquist, 1994**). Low levels of L-Car synthesis may make spraying it on eggs improves the hatchability percentage. It was also documented that the positive effect of L-Car may be attributed to chick embryos require a high requirement, where yolk contains low level and have a limited capacity to synthesize L-Car during incubation (**Casillas and Newburgh, 1969**). So, L-Car would promote lipid circulation from fat storage areas such as the yolk sac, leading to an increase in fatty acid catabolism may help to overcome the constraint of limited egg nutrients (**Foye et al., 2006**). Furthermore, L-Car has a specific role in facilitating the uptake of fatty acyl groups from the egg yolk into the tissues of embryonic chicks via the yolk sac membrane, which in turn increases their efficiency of energy utilization (**Casillas and Newburgh, 1969**). **Rinaudo et al. (1991)** who indicated that the ratio of esterified short-chain L-Car to free L-Car reaches a maximum in tissues (liver, brain and heart) of the embryos on the 18th d of incubation, thereby suggesting that the fatty acid oxidation is inevitable for energy

production in embryos. This finding is in accordance with, **Oso et al. (2014)** who indicated that L-Car has been reported to positively affect the hatchability of eggs. In such situations, exogenous supplementation of L-Car could be advantageous (**Buyse et al., 2001**), and could in turn be used by the chick during hatching. For these reasons the spraying eggs with L-Car improved hatchability. This finding is consistent with those reported by **Rabie et al. (2015)** who indicated that hatchability of fertile eggs was significantly increased when eggs injected with L-Car on 14 d of incubation. The same trend observed for embryonic mortality, where spraying eggs with AA and L-Car resulted in decreases of embryonic mortalities. This attributed to AA protect embryo from any stresses during incubation (**Tag El-Din et al., 2004**). The increase mortality during incubation especially in middle period (8-18 d) of incubation usually ascribed to different factors i.e., vitamins or minerals deficiency (**Leeson and Summers, 1991**), improper incubator conditions, contamination, and lethal genes (**Wilson, 2004**), so the spraying eggs with AA and L-Car maximize nutrient required during embryogenesis and overcome of these negative factors. Chick embryos may be subjected to stress caused by excessive production of metabolic heat during the latter part of incubation, so AA solution acting as an anti-stress agent may be beneficial for embryos viability and protecting them from many stress during incubation (**Tullett, 1990**). **Mohammed et al. (2011)** who indicated that dipping eggs in 5.0 g AA/L for 2 minutes reduced embryonic mortality at 1-18, 19-21 and 1-21 d of incubation. Also, L-Car has beneficial effect in decreasing embryonic mortalities, this may be due to embryonic tissues contain high amounts of polyunsaturated fatty acids, which are essential components of cell membrane phospholipids (**Rabie and Szilagyi, 1998**).

Polyunsaturated fatty acids are susceptible to lipid peroxidation caused by free radical, which are produced by mitochondria because of the high metabolic rate of rapidly developing embryos (**Surai, 1999**). Moreover, L-Car may work as an antioxidant to scavenge free radical and reducing oxidative stress during embryogenesis (**Deniz and Turkmen 2007**), thereby reducing embryonic mortality. **Oso et al. (2014)** who found that dietary L-Car supplementation for turkey breeder hens reduced the late embryonic mortality.

Chick quality traits: The present study showed that spraying eggs with either AA or L-Car improved newly hatched chick quality traits including, chick weight, relative weight and Tona score in comparison with the control groups. On the contrary, it is observed that commercial hatched chicks including grade A and B not affected by the treatment groups. Accordingly, it is obvious that spraying eggs with AA and L-Car led to improvement in chick quality at hatch. An increased post hatch weight was the only indication that these nutrients had passed through the shell and was ultimately accessible to the embryo. Presumably, spraying eggs with former nutrients were confined to the egg periphery until chorioallantoic circulation conveyed them to the established embryo. Egg shell permeability to oxygen at the onset of incubation is negligible because the pores and membrane matrix contain water (**Lomholt, 1976**). Subsequent alterations in albumen osmotic pressure clear these channels and a concurrent development of the chorioallantoic membrane permits gas exchange and aerobic respiration ensues (**Fancsi and Feher, 1979**). Previous studies demonstrated that the weight of newly hatched chickens is an important predictor of market weight. Although this correlation between hatching weight and market weight may differ among strains, the effect of hatch weight on market weight

apparently increases as broiler breeding companies continue to select for ever increasing growth rates (**Havenstein et al., 2003**). This finding are confirmed by **Wilson (1991)** who indicated that each 1 g of increase in body weight at hatch resulted in 8 to 13 g increase in body weight at market age. In this study, we showed that a 1 g of increase in body weight at hatch due to spraying nutrients increased BW by about 46 to 132 g on d 35. However, spraying eggs could improve the nutritional and health status of the embryo and enhance nutrient uptake, increase in activity of the intestinal enzymes and post hatch growth due to the role of nutrients to increase size and surface area of the intestinal villi (**Tako et al., 2004**) and enhanced intestinal function and maturation prior to hatching (**Foye et al., 2007**). The other reason for this case is that accumulation of yolk-derived lipids increases in embryo tissues and absorption in this stage at last week of incubation (**Pulikanti et al., 2010**). It is well known that AA participate in several biochemical processes and functions is also related to reversible oxidation and reduction characteristics in the cells endogenous. Moreover, AA has an important position not only in reacting with all aggressive reactive oxygen species but also in transferring radical equivalents from lipid phases to hydrophilic compartments (**Gey, 1998**). Results are consistent with **Zakaria and Al-Anezi (1996)** who found that eggs injected by a dose of 3 mg of AA at the 15th d of incubation increased BW of chicks compared to the control. However, L-Car acts as an antioxidant that absorbed prior to pipping (**Uni et al., 2005**), thereby improving embryogenesis, and overcome the constraint of limited egg nutrients (**Foye et al., 2006**). Rapid development, high energy requirement, and low level of L-Car synthesis may make supplementation was beneficial to chicken embryos. We hypothesize that administration of L-Car will improve fatty

acid β -oxidation and suppress lipid peroxidation, thus increasing chicks quality traits. Also, **Salmanzadeh et al. (2013)** who indicated that in *ovo* injection of L-Car on d 6 of incubation improved embryonic development and weight of newly hatched chicks.

Blood parameters: Results from this study indicate that spraying eggs with AA and L-Car had a significant effect on most measured blood traits at d 18 of incubation. The values of RBCs, WBCs and Hb concentration were markedly improved, due to application of nutrient solutions, this could attributed to AA increase heterophils percentage, resulted from decreases in vessel permeability, which limits leucocytes infiltration and the increase passing of the heterophils from the bone marrow to blood circulation (**Lohakare et al., 2005**). Spraying eggs during embryogenesis may be a good way to improve blood hematology. These findings are in agreement with those reported by **Ghonim et al. (2009)** who indicate that the treated duck eggs with AA solution during incubation resulted in an increase of hemoglobin content and WBCs count. Similar results were reported by **Askar (2012)** who found that hematological traits were differed in response to AA treatment, where the values of Hb and RBCs count for eggs sprayed by AA solution or those dipped in 4 g/L AA solution were higher than those dipped in 8 g/L of AA or eggs of the control group. Lymphocytes percentage was increased in chicks of eggs sprayed by AA solution or those dipped in 8 g/L AA of solution than those of the control group or those dipped in 4 g/L AA solution. However, the glucose concentration severely increased for embryo produced from eggs sprayed with AA or chicks for the control groups. Whereas the lowest concentration recorded for L-Car groups, this may be due to L-Car can buffer the acetyl CoA/ CoA ratio, thus preventing the negative feedback of high acetyl-CoA level on PDH activity. As a consequence, the

breakdown of glucose can proceed, leading to enhanced glucose utilization and hence lower plasma glucose level (Feller and Rudman, 1988). These findings are in agreement with Buysse *et al.* (2001) reported that L-Car supplementation significantly reduced plasma glucose concentration on d 17 of embryo. Also, Wang *et al.* (2012) who indicated that L-Car supplementation significantly reduced serum glucose of broiler on d 35. The values of ALT and AST were not affected by spraying eggs with AA and L-Car solutions. Yalcin *et al.* (2005) who found that no differences in ALT and AST among groups of layer chickens fed diet supplemented with 0.1 g/kg L-Car. Concerning kidney function, creatinine insignificantly affected due to spraying eggs with application of different nutritive solutions, while urea level showed the highest value for embryo produced from eggs sprayed with 6g/L AA and the lowest for eggs sprayed with 6g/L L-Car. These findings are consistent with Greenwood *et al.* (2001) who indicated that L-Car supplementation tends to increase the plasma urea concentration, where the urinary excretion increases as the supplementation level increases, and this was observed in terms of the elevated plasma urea concentration when intermediate levels of supplemental L-Car were provided. Therefore, the significant decrease in urea levels may indicate that embryo bodies were free of oxidative stress. Urbaityte *et al.* (2006) who indicated that L-Car is capable of eliminating of reactive oxygen species. Creatinine is the metabolite key of skeletal muscles and originates from metabolic transformations of creatine. The latter serves to accumulate and release energy indispensable for the course of many chemical processes ongoing in cells (Pawłowska-Góral *et al.*, 2003). The enhanced production of L-Car is due to physical effort which is linked with increased demands of cells for oxygen and

with high quantities of reactive oxygen species generated in the body (Moffarts *et al.*, 2005). The use of L-Car in spraying eggs, caused a decrease in urea level in the terminal period of incubation, which also points to a low level of reactive oxygen species and thus to the balance of antioxidative processes. However, it is observed that corticosterone level was severely decreased due to spraying eggs with AA and L-Car. The decrease of CS was an evidence of judged decreased stress response due to AA, which may act as an anti-stress agent led to the reduction of corticosterone, which has a negative impact in collagen synthesis and the metabolism of minerals and vitamin D (Lohakare *et al.*, 2005). The incubator temperature during incubation was between 32 °C and 37.7 °C and the metabolic heat production of the developing embryo is sufficient to raise the internal egg temperature by 1.5 °C to 2 °C (Tullett, 1995) that is above the incubator temperature. This may contribute the high percent of death at later stage. Moreover, either AA or L-Car was known to suppress corticosterone synthesis and /or release from adrenal cortex, which in turn play an important role in alleviating different stressor effects and hence increased the heterophils ratio. These results may ensure that AA acted as a good anti-stressors agent. Since, the alteration in temperature degree during incubation or the high effort exerted by chicks during egg shell breakage may cause some stress on the embryo (Askar, 2012).

Subsequent growth performance: It appears from results presented in Table 5 that spraying eggs with AA or L-Car had a positive effect on subsequent growth performance. The finding showed that each g difference in BW at hatch due to spraying eggs with nutritive solutions resulted in an increase of BW at d 35. This may be attributed to there was a strong positive correlation between LBW at hatch and body at market age. In this sense, spraying eggs just before internal pipping likely

provide the embryo the caloric resources to fuel the hatching process and early development. Increasing LBW due to spraying eggs with AA could be attributed to it plays a major role in the biosynthesis of CS (Bains, 1996), a primary glucocorticoid hormone involved in gluconeogenesis to enhance energy supply during stress (Frandsen, 1986). However, spraying eggs with L-Car improving growth in chickens (Zhai *et al.*, 2008), due to an increased efficiency in fatty acid oxidation may, likewise, reduce the dependency of the embryo upon gluconeogenesis, thereby sparing muscle tissue protein in the post-hatched chick. Reduced utilization of glucose may spare muscle protein mobilization for gluconeogenesis during late term embryonic development, thus increasing LBW. The enhanced muscle growth is due, in part, to higher proliferation of myoblasts during embryonic development and to the presence of more satellite cells in early post-hatch development (Halevy *et al.*, 2004). These results suggest an increased efficiency in fatty acid oxidation may reduce the embryo's dependency upon gluconeogenic pathways and spare muscle tissue protein in the post-hatch chick, which could subsequently lead to an increase in muscle yield during grow-out (Keralapurath *et al.*, 2010). We hypothesized that spraying eggs with L-Car may provide energy for embryo activity and consequently increase growth performance. An increased post-hatch weight was the only indication that these nutrients had passed through the shell and was ultimately accessible to the embryo. Presumably, spraying method may cause changes in the pores, which in turn may affect the gas exchange properties of the shell. Therefore, accelerated embryo development improved nutritional status and improved hatching weight and growth rate (Bhanja *et al.*, 2004). The results of this study agree with the findings of Wilson (1991) who indicated that each 1 g

increase in body weight at hatch resulted in 8 to 13 g increase in body weight at market age. Furthermore, Zhai *et al.* (2008) who indicated that supplementation of L-Car to the embryo would affect subsequent grow-out performance and slaughter yield of broilers at d 47 of post-hatch. Also, chicks produced from eggs sprayed with either AA or L-Car consumed more feed compared to control and sham groups. However, FCR recorded the best for chickens produced from eggs treated with different nutritive solutions. This suggestion is supported in *ovo* feeding improves feed efficiency of hatched chicks (Tako *et al.*, 2004). Also, Salmanzadeh *et al.* (2013) who found that the *in ovo* injection of L-Car in the yolk sac can be seen as an effective tool to increase the weight gain and feed conversion ratio of newly hatched chicks. However, Rabie *et al.* (2015) who indicated that *in ovo* injection with L-Car particularly with dose 8.0 or 12 mg 100 μ L saline resulted in significant improvements in LBW and FCR of broilers during the period from the 2nd to the 7th wks of life as compared to control. The improvement in FCR of broilers in response with L-Car may partly explain by the improved final LBW and cumulative BWG, since feed intake of birds was not affected.

Caracas traits: It has been clearly demonstrated in the present study that spraying eggs with AA and L-Car improved carcass traits, where the highest effect observed for abdominal fat percentage and small intestine. The positive effect of L-Car may be attributed to embryos require a high level, where yolk contains low level of L-Car would promote lipid circulation from fat storage areas such as the yolk sac, leading to an increase in fatty acid catabolism. This could subsequently lead to an increase in muscle yield during grow-out. In this regard, Quarles and Adrian (1989) who indicated that when vitamin C was supplemented at a level of 976 ppm/128 gal in the drinking

improved slaughter carcass yield. However, **Salmanzadeh et al. (2013)** who found that *in ovo* administration of L-Car has significantly increased the breast muscle size. In addition, **Salmanzadeh et al. (2012)** who reported *in ovo* administration of L-Car into turkey eggs with 20 and 30 mg L-Car significantly improve carcass characteristics compared to the control. Furthermore, the L-Car decreased carcass fat and increased breast meat. **Kidd et al. (2005)** who showed that dietary L-Car induced significant decreases in abdominal fat and more specifically in carcass fat deposits and significant increases in breast meat yield in the corresponding chickens fed with high nutrient density diets. Also, **Rabie et al. (2015)** who indicated that *in ovo* injection

with L-Car resulted in a significant increase in the relative weight of front parts including and significantly decreased the percentages of liver and gizzard.

CONCLUSION AND RECOMENDATION

In conclusion, under our experimental condition, spraying eggs with either AA or L-Car at different doses during embryogenesis (1, 7, 14 and 18th d of incubation) may be used as a successful method to improve hatchability and post-hatch performance. Therefore, it is suggested that applications of these nutritive solutions during embryogenesis can serve as a potential regulatory agent to reduce embryonic mortalities.

Ascorbic acid, L-carnitine, hatchability, post-hatch growth performance.

Table (1): Composition and chemical analysis of grow-out experimental diets.

Ingredients	Diets*		
	Starter	Grower	Finisher
Yellow corn (8.5%)	55.4	60.6	62.8
Soybean meal (44 %)	33.3	27.8	24.4
Corn gluten meal (60%)	3.00	3.20	4.20
DL-Methionine	0.24	0.24	0.20
Lysine-Hcl	0.18	0.24	0.16
Soybean oil	3.66	3.83	4.33
calcium phosphate (CaHPO ₄)	1.64	1.58	1.49
Pre-mix**	0.30	0.30	0.30
Choline chloride	0.10	0.10	0.10
Calcium carbonate (CaCO ₃)	1.66	1.61	1.59
Sodium Chloride (Na Cl)	0.35	0.30	0.30
Sodium bicarbonate	0.08	0.12	0.10
Anti Coccidiosis drug.	0.05	0.05	0.05
Total (kg)	100	100	100
<i>Calculated analysis:</i>			
ME (Kcal / kg)	3033	3108	3180
Crude protein %	21.5	19.7	18.8
Ether extract %	2.65	2.7	2.77
Crude fiber %	3.02	2.94	2.80
Lysine %	1.30	1.20	1.05
Methionine %	0.61	0.59	0.55
Threonine %	0.85	0.78	0.75
Calcium (%)	1.00	1.00	1.00
Total phosphorus (%)	0.75	0.72	0.69
Av. phosphorus	0.50	0.48	0.42
Sodium (%)	0.17	0.16	0.16
Chlorine (%)	0.19	0.17	0.17
<i>Chemical analysis:</i>			
Crude protein %	21.4	19.6	18.9
Ether extract %	2.85	2.50	2.90
Calcium (%)	1.10	1.05	1.03
Total phosphorus (%)	0.73	0.71	0.68

*Requirement according to Cobb 500 broiler performance guide supplement (2012) and NRC (1994)

** pre-mix each 3 kg of vitamin and mixture contains: 13000000 IU Vit. A, 5000000 IU D3 8000 mg E, 4000 mg K, 5000 mg B1, 9000mg B2, 4000 mg B6, 20 mg B12, 15000 mg pantothenic acid, 6000 mg Nicotinic acid, 2000 mg Folic acid, 150 mg Biotin, 40000 mg Choline chlorine, 20000 mg Copper sulphate, 1000 mg Ca iodide, 50000 mg ferrous sulphate, 10000 mg Manganese oxide, 100000 mg Zinc oxide and 300 mg Sodium selenite.

Table(2): Egg weight loss, hatchability and embryonic mortalities of eggs as affected by nutritive solutions (Means \pm SE).

Items	Main treatment effects:						SEM	Sig.*
	Control ¹	Distilled water ²	Vitamin C 4 g/L	Vitamin C 6 g/L	L-Carnitine 4 g/L	L-Carnitine 6 g/L		
Fresh egg weight (g)	60.48	60.45	60.43	60.45	60.48	60.53	0.02	NS
Egg weight at d 18 (g)	54.33 ^a	54.38 ^a	53.95 ^{bc}	53.83 ^c	54.25 ^{ab}	54.33 ^a	0.06	*
Loss of Egg weight (%)	10.17 ^{bc}	10.05 ^c	10.71 ^{ab}	10.96 ^a	10.29 ^{bc}	10.24 ^{bc}	0.09	*
Fertility (%)	95.61	95.75	95.91	95.47	95.76	95.60	0.08	NS
Total hatchability (%)	83.18 ^e	83.79 ^{de}	85.91 ^b	86.97 ^a	84.70 ^c	84.09 ^{cd}	0.29	*
Fertility hatchability (%)	86.87 ^c	87.52 ^c	89.58 ^a	90.33 ^a	88.13 ^{bc}	89.05 ^{ab}	0.29	*
Early mortality (%)	2.85 ^a	2.68 ^{ab}	2.37 ^{abc}	2.32 ^{abc}	1.59 ^c	1.91 ^{bc}	0.13	*
Middle mortality (%)	5.85 ^a	5.85 ^a	4.42 ^{bc}	4.00 ^c	6.01 ^a	5.55 ^{ab}	0.23	*
Late mortality (%)	4.43 ^a	3.95 ^{ab}	3.63 ^{ab}	3.35 ^b	4.27 ^{ab}	3.49 ^{ab}	0.13	*
Total mortalities (%)	13.13 ^a	12.48 ^a	10.42 ^c	9.67 ^c	11.87 ^{ab}	10.95 ^{bc}	0.29	*

a-c Means within a row that do not share common superscript differ significantly ($P \leq 0.05$)

1 = Negative control group

2 = Sham control group

* = significant at 0.05

Ascorbic acid, L-carnitine, hatchability, post-hatch growth performance.

Table(3): Chick quality traits as affected by nutritive solutions (Means \pm SE).

Items	Main treatment effects:							SEM	Sig.
	Control	Distilled water	Vitamin C 4 g/L	Vitamin C 6 g /L	L-carnitine 4 g/L	L-carnitine 6 g/L			
Chick weight (g)	39.95 ^b	40.22 ^b	41.04 ^a	40.85 ^a	41.03 ^a	40.76 ^a	0.11	*	
Relative weight	66.06 ^c	66.53 ^{bc}	67.91 ^a	67.57 ^a	67.85 ^a	67.35 ^{ab}	0.18	*	
Tona score (%)	93.75 ^{bc}	93.25 ^c	94.00 ^{abc}	94.25 ^{ab}	94.75 ^a	94.25 ^{ab}	0.14	*	
<i>Commercial chick quality:</i> Chick grade A (%)	98.25	98.37	98.75	98.75	98.75	98.63	0.07	NS	
Chick grade B (%)	1.75	1.63	1.25	1.25	1.25	1.37	0.07	NS	

a-c Means within a row that do not share common superscript differ significantly ($P \leq 0.05$)

Table (4): Some blood parameter of embryo at d 18 of incubation as affected by nutritive solutions (Means \pm SE).

Items	Main treatment effects:						SEM	Sig.
	Control	Distilled water	Vitamin C 4 g/L	Vitamin C 6 g /L	L-Carnitine 4 g/L	L-Carnitine 6 g/L		
RBC ^S (cells/ $10^6 \times \text{mm}^3$)	1.13 ^d	1.15 ^d	1.66 ^a	1.49 ^b	1.33 ^c	1.35 ^c	0.38	*
WBC ^S (cells/ $\times \text{mm}^3$)	37.65 ^f	38.79 ^e	43.36 ^b	46.27 ^a	40.99 ^d	41.53 ^c	0.59	*
Hb (g/ 100 mL)	12.15 ^e	12.51 ^d	14.02 ^b	14.65 ^a	13.51 ^c	13.52 ^c	0.18	*
Glucose (Mg/ 100 mL)	177.25 ^a	178.50 ^a	178.50 ^a	178.00 ^a	170.50 ^b	169.75 ^b	0.99	*
AST(U/L)	168.25	164.50	168.50	164.25	166.25	165.50	0.72	NS
ALT(U/L)	14.50	15.25	15.75	15.00	14.00	14.25	0.40	NS
Creatinine (mg/100mL)	0.34	0.33	0.35	0.36	0.37	0.34	0.01	NS
U Urea (mg/dL)	27.25 ^{bc}	30.50 ^{ab}	30.00 ^{abc}	32.00 ^a	30.50 ^{ab}	26.50 ^c	0.59	*
Corticosterone (ug/dL)	0.39 ^a	0.38 ^a	0.34 ^b	0.29 ^c	0.33 ^b	0.33 ^b	0.01	*

a-c Means within a row that do not share common superscript differ significantly ($P \leq 0.05$)

Table (5): Growth performance of broiler chickens as affected by nutritive solutions

Items	Ages (d)	Main treatment effects:						SEM	Sig.
		Control	Distilled water	Vitamin C 4 g/L	Vitamin C 6 g/L	L-Carnitine 4 g/L	L-Carnitine 6 g/L		
Live body weight (g)	1	39.95 ^b	40.22 ^b	41.05 ^a	40.85 ^a	41.03 ^a	40.75 ^a	0.11	*
	14	417 ^c	415 ^c	438 ^b	438 ^b	446 ^a	445 ^a	2.7	*
	28	1335 ^b	1327 ^b	1380 ^a	1360 ^a	1380 ^a	1374 ^a	5.2	*
	35	1868 ^c	1914 ^{bc}	1960 ^{ab}	1972 ^a	2000 ^a	1996 ^a	11.6	*
Body weight gain daily (g)	1-14	26.95 ^c	26.79 ^c	28.32 ^b	28.33 ^b	28.91 ^a	28.89 ^a	0.19	*
	15-28	65.57 ^{bc}	65.09 ^c	67.34 ^a	65.89 ^{abc}	66.71 ^{ab}	66.36 ^{abc}	0.23	*
	29-35	76.11 ^b	83.86 ^a	82.83 ^a	87.43 ^a	88.57 ^a	88.79 ^a	1.22	*
	1-35	52.23 ^b	53.17 ^b	54.83 ^a	55.18 ^a	55.96 ^a	55.86 ^a	0.34	*
Feed intake (g/d)	1-14	32.73 ^d	33.45 ^{dc}	36.70 ^a	34.59 ^{bc}	35.57 ^{ab}	35.70 ^{ab}	0.32	*
	15-28	112.09 ^c	112.94 ^{bc}	113.80 ^{bc}	114.57 ^b	119.59 ^a	117.98 ^a	0.62	*
	29-35	171.93	174.46	177.04	175.29	172.57	175.11	0.79	NS
	1-35	92.62 ^b	94.75 ^{ab}	96.19 ^a	94.75 ^{ab}	96.82 ^a	97.05 ^a	0.44	*
Feed conversion ratio (g feed /g gain)	1-14	1.21 ^b	1.25 ^{ab}	1.29 ^a	1.22 ^b	1.23 ^b	1.23 ^b	0.01	*
	15-28	1.71 ^{cd}	1.73 ^{bcd}	1.69 ^d	1.74 ^{bc}	1.79 ^a	1.78 ^{ab}	0.01	*
	29-35	2.27 ^a	2.08 ^{bc}	2.15 ^{ab}	2.01 ^{bc}	1.96 ^c	1.97 ^{bc}	0.03	*
	1-35	1.77 ^{ab}	1.78 ^a	1.76 ^{ab}	1.72 ^b	1.73 ^{ab}	1.74 ^{ab}	0.01	*

(Means \pm SE).a-c Means within a row that do not share common superscript differ significantly ($P \leq 0.05$)

Ascorbic acid, L-carnitine, hatchability, post-hatch growth performance.

Table (6): Some carcass traits of broiler chickens as affected by nutritive solutions (Means \pm SE).

Items	Main Treatment effects:							SEM	Sig.
	Control	Distilled water	Vitamin C 4 g/L	Vitamin C 6 g/L	L-Carnitine 4 g/L	L-Carnitine 6 g/L			
Live body weight (g)	1880 ^b	1885 ^b	2002 ^a	2015 ^a	2012 ^a	1977 ^a	16.31	*	
Liver weight (%)	2.42 ^d	2.49 ^{bc}	2.54 ^{ab}	2.59 ^a	2.44 ^{cd}	2.41 ^d	0.02	*	
Gizzard weight (%)	2.85 ^a	2.86 ^a	2.76 ^b	2.81 ^{ab}	2.81 ^{ab}	2.78 ^b	0.01	*	
Heart weight (%)	0.54 ^a	0.54 ^a	0.48 ^{ab}	0.47 ^{ab}	0.47 ^{ab}	0.45 ^b	0.01	*	
Spleen weight (%)	0.28	0.26	0.28	0.27	0.28	0.28	0.01	NS	
Giblets weight (%)	6.09 ^{ab}	6.15 ^a	6.06 ^{abc}	6.14 ^{ab}	6.00 ^{bc}	5.92 ^c	0.02	*	
Dressing weight (%)	67.50	66.31	66.64	66.47	66.37	66.70	0.17	NS	
Total edible parts weight (%)	73.59	72.46	72.70	72.62	72.37	72.62	0.18	NS	
Total inedible parts weight (%)	26.41	27.54	27.30	27.38	27.63	27.38	0.18	NS	
Abdominal fat weight (%)	2.95 ^a	2.98 ^a	2.76 ^b	2.70 ^b	2.57 ^c	2.49 ^d	0.04	*	
Small intestine length(cm)	145.00 ^d	149.12 ^c	151.38 ^{ab}	153.12 ^{ab}	153.00 ^a	149.50 ^{bc}	0.63	*	
Cecum length(cm)	13.00 ^b	13.25 ^{ab}	13.37 ^{ab}	13.50 ^{ab}	13.75 ^a	13.25 ^{ab}	0.08	*	

a-c Means within a row that do not share common superscript differ significantly ($P \leq 0.05$)

REFERENCES

- Agarwal, A., S. A. Prabakaran, and T. M. Said (2005).** Prevention of oxidative stress injury to sperm. *J. Androl.* 26:654–660.
- Askar, A. A. (2012).** Influence of the manipulation of incubation Japanese quail eggs by ascorbic acid on the embryo weight, hatchability and hematological parameters of the chicks. *Egypt. J. Anim. Prod.* 49(2):187-194.
- Bains, B. S. (1996).** The role of vitamin C in stress management. *World's Poul. Sci.*, 12: 38- 42.
- Bhanja S. K., A. B. Mandal and T. K. Goswami. (2004).** Effect of in ovo injection of amino acids on growth, immune response, development of digestive organs and carcass yield of broiler. *Ind. J. of Poul. Sci.*, 39: 212-218.
- Broquist H. P. (1994).** *Modern Nutrition in Health and Disease.* Eds. M. E. Shils,

- J.A. Olson, M. Shike, Philadelphia, Lea and Febiger, V1, 459-465.
- Burley, R. W. and D. V. Vadehra (1989).** The eggshell and shell membranes properties and synthesis. In: R.W. Burley and D.V.Vadehra (Eds).The Avian Egg: Chemistry and Biology, John Wiley, New York, USA, 25 –64.
- Buyse, J., G. P. Janssens, and E. Decuypere (2001).** The effects of dietary L-carnitine supplementation on the performance, organ weights and circulating hormone and metabolite concentrations of broiler chickens reared under a normal or low temperature schedule. *Br. Poult. Sci.*, 42:230–241.
- Casillas, E. R., and R. W. Newburgh (1969).** L-carnitine and derivatives in embryonic chick tissue. *Biochim. Biophys. Acta*, 184:566–577.
- Chiodi, P., B. Ciani, S. Kentroti, F. Maccari, A. Vernadakis, L. Angelucci and M. T. Ramacci (1994).** L-Carnitine and derivatives in the central nervous system of chick embryo. *Int. J. Bioc.*, 26:711–720.
- Cobb 500 performance broiler and nutrition supplement (2012).** Cobb-vantress.com.
- Deniz G.Y. and I. I. Turkmen (2007).** Effect of an addition of Hepabial Carnitine ®in drinking water on the performance of broilers under stocking stress. *Rev. Med. Vet.*, 158: 13-18.
- Doaa M. M. yassein; S. M. M. Mousa and A. M. A. Bealish (2014).** Effect of spraying hatching eggs by ascorbic acid during incubation on hatchability, post-hatch chick growth and physiological parameters in a local strain of chickens. *Egypt. Poult. Sci.* (34) (III): (715-733).
- Duncan, B. D. (1955).** Multiple range and multiple-F tests. *Biometrics*, 11:1-42.
- Fancsi, T., and G. Feher. (1979).** Ultrastructural studies of chicken embryo chorioallantoic membrane during incubation. *Anat. Histol. Embryol*, 8:151–159.
- Feller, A. G. and D. Rudman. (1988).** Role of carnitine in human nutrition. *J. Nutr.*, 118:541- 547.
- Frandsen, R. D. (1986).** Anatomy and physiology of farm animals, Chapter 32: Endocrinology, 481-507, Lea and Febiger Publisher, Philadelphia, USA.
- Foye O. T., P. R. Ferket and Z. Uni (2006).** Effect of in ovo feeding egg white protein, betahydroxy-beta-methylbutyrate, and carbohydrates on glycogen status and neonatal growth of turkeys. *Poultry Sci.*, 85: 1185-1192.
- Foye, O. T., P. R. Ferket and Z. Uni (2007).** The Effects of In Ovo Feeding Arginine, -hydroxy-methyl-butyrate, and Protein on Jejunal Digestive and Absorptive Activities Embryonic and Neonatal Turkey Poults. *Poult. Sci.*, 86: 2343-2349.
- Geng, Z. Y., and X. L. Wang (1990).** Relationship of hatchability and the percentage of egg weight loss and shell pore concentration during incubation. *Chin. J. Anim. Sci.*, 26:12-14.
- Gey, K.F. (1998).** Vitamins E plus C and interacting co nutrients required for optimal health. *Biofactors* 7:113-174.
- Ghonim, A. I. A.; A. L. Awad, M. H. A. Fattouh and A. M. El-Shhat (2009).** Comparative study of ascorbic acid treatment methods on hatchability traits and growth performance of ducklings. *Egyptian Poult. Sci.*, 29:1085-1099.
- Greenwood, R. H., E. C. Titgemeyer and G. L. Stokka (2001).** Effects of L-carnitine on nitrogen retention and blood metabolites of growing steers and performance of finishing steers. *J. Anim. Sci.*, 79:254 – 260.
- Halevy O., Y. piestun, M. Z. Allouh, B. W. Rosser, Y. Rinkevich, R. Reshef, I. Rozenboim, M. Wleklinskilee, Z. Yablonkareuveni (2004).** Pattern of Pax7 ex- pression during myogenesis in the post hatch chicken establishes a model for satellite cell differentiation and renewal. *Dev. Dyn.* 231: 489-502.

- Havenstein, G. B., P. R. Ferket, and M. A. Qureshi. (2003).** Carcass composition and yield of 1957 versus 2001 broilers when fed representative 1957 and 2001 broiler diets. *Poult. Sci.*, 82:1509–1518.
- Ipek, U. Sahan and B. Yilmaz (2004).** The effect of in ovo ascorbic acid and glucose injection in broiler breeder eggs on hatchability and chick weight. *Arch. Geflügelk.*, 68: 132–135.
- Keralapurath, M. M., A. Corzo, R. Pulikanti, W. Zhai and E. D. Peebles (2010).** Effects of in ovo injection of L-carnitine on hatchability and subsequent broiler performance and slaughter yield. *Poult. Sci.*, 89: 1497-1501.
- Kidd, M. T., C. D. Mc- Daniel, E. D. Peebles, S. J. Barber, A. Corzo, S. L. Branton and J. C Woodworth (2005).** Breeder hen dietary L-carnitine affects progeny carcass traits. *Br. Poult. Sci.*, 46, 91-103.
- King 'ori, A. M. (2011).** Review of the Factors That Influence Egg Fertility and Hatchability in Poultry. *Int. J. Poult. Sci.*, 10 (6): 483-492.
- Leeson, S. and J. D. Summers (1991).** *Commercial Poultry Nutrition.* University Books, Guelph, 141- 144, 207-212 and 237-245.
- Lohakare, J. D., M. H. Ryu, T. W. Hahn, J. K. Lee and B. J. Chae (2005).** Effects of supplemental ascorbic acid on performance and immunity of commercial broilers. *J. Appl. Poult. Res.* 14:10 -19.
- Lomholt, J. P. (1976).** The development of the oxygen permeability of the avian eggshell and its membranes during incubation. *J. Exp. Zool.*, 198:177 184.
- Meir, M. and A. Ar (1991).** Compensation for seasonal change in eggshell conductance and hatchability of goose eggs by dynamic control of egg water loss. *Brit. Poult. Sci.*, 32: 723-732.
- Moffarts, B., N. Kirschvink, T. Art, J. Pincemail, and P. Lekeux (2005).** Effect of oral antioxidant supplementation on blood antioxidant status in trained thoroughbred horses. *Vet. J.*, 169: 65- 74.
- Mohammed, K.A., Attiat. El-Boghdady, M.A.H. Soliman, M.A. Abd-Al-Galil and N.M. Abd-Al-Aleem (2011).** The effect of both pre-incubation dipping eggs in vitamin c and cooling eggs during incubation period on embryonic and hatchability parameters in two local chicken strains. *Egypt. Poult. Sci.*, 31: 379-392.
- Nowaczewski, S., K. Stuper K., T. Szablewski and H. Konteka (2011).** Microscopic fungi in eggs of ring-necked pheasants kept in aviaries. *Poult. Sci.*, 90: 2467-2470.
- N.R.C, Nutrient requirements of poultry (1994).** 9th ed., National Academy of Science, National Research Council. Washington, D. C. U.S. A.
- Oso, A. O., A. O. Fafiolu, M. A. Adeleke, O. A. Ladokun and R. A. Sobayo (2014).** Effect of dosage and application mode of L-carnitine on plasma lipid and egg-yolk cholesterol of turkeys, hatchability of eggs and post-hatch growth of their offsprings. *J. Anim. Physiol. Anim. Nutr.*, 98: 766-774.
- Pawlowska-Góral, K., A. Kalamarz, and M. Wardas (2003).** The total antioxidation potential, methods of its measurement and its clinical usefulness. *Diagn. Lab.*, 39:327-338.
- Pulikanti, R., E. D. Peebles, R. W. Keirs, L. W. Bennett, M. M. Keralapurath, and P. D. Gerard. (2010).** Pipping muscle and liver metabolic profile changes and relationships in broiler embryos on days 15 and 19 of incubation. *Poult. Sci.*, 89(5):860- 865.
- Quarles, C. L. and W. J. Adrian (1989).** Evaluation of ascorbic acid for increasing carcass yield in broiler chickens. The role of vitamin C for poultry stress management. F. Hoffmann. Laroche and Co. AG, Switzerland.

- Rabie, M. H., F. S. A. Ismail and A. A. S. Ahmed (2015).** Effect of In ovo Injection of L-Carnitine at Different Incubational Ages on Egg Hatchability in Broiler Breeders and Post-Hatch Performance. *Asian J. of Ani. and Vet. Adva*, 10: 875-884.
- Rabie, M. H. and M. Szilagyi (1998).** Effects of L-Carnitine supplementation of diets differing in energy levels on performance, abdominal fat content and yield and composition of edible meat of broilers. *Brit. J. of Nut*, 80: 391-400.
- Rahn H. and A. Ar (1974).** The avian egg: Incubation time and water loss. *Condor*; 76:147-152.
- Rahn H, V.L. Christensen and F.W. Edens (1981).** Changes in shell conductance, pores, and physical dimensions of egg and shell during the first breeding cycle of turkey hens. *Poultry Science*; 60:2536-2541.
- Rebouche, C. J. (1992).** L-Carnitine function and requirements during the life cycle. *FASEB J.* 6:3379–3386.
- Rinaudo, M. T., M. Curto, R. Bruno, M. Piccinini and C. Marino (1991).** Acid soluble, short chain esterified and free carnitine in the liver, heart, muscle and brain of pre- and post-hatched chicks. *Int. J. Biochem.*, 74: 152-160.
- Salmanzadeh, M. Y. Ebrahimnezhad, H. Aghdam and H. A. Shahryar (2012).** Effects of in ovo administration of L-carnitine on hatchability, subsequent performance, carcass traits and blood cholesterol of turkey poults. *Revu Méd. Vét*, 163, 10: 448-453.
- Salmanzadeh, M., Y. Ebrahimnezhad and H. A. Shahryar (2013).** The Effects of in ovo Injection of L - carnitine on hatching traits, growth Performance, and carcass Characteristics of turkey Poults. *India Bulletin of Environment, Pharmacology and Life Sciences Bull. Env. Pharmacol. Life Sci.*, 2 (11) 125 -1 28.
- Selim, S. H. A., K. M. Gaafar and S. S. El-ballal (2012).** Influence of in-ovo administration with vitamin E and ascorbic acid on the performance of Muscovy ducks. *Emir. J. Food Agric.*, 24: 264-271.
- Shafey, T. M., Alodan, M.A., Al-Ruqaie, S.I.M. and M.A. Abouheif (2012).** In ovo feeding of carbohydrates and incubated at a high incubation temperature on hatchability and glycogen status of chicks. *African J. of Anim. Sci.*, 42, 3.
- Shafey, T. M., A. S. Same and M. A. Abouheif (2013).** Effects of in ovo feeding of L-glutamine on hatchability performance and hatching time of meat-type breeder eggs. *JAVA*, 12: 135-139.
- SPSS 16 .0(2010).** Statistical package in Social Science for Windows. Statistical innovation INC., Chicago, USA
- Surai, P. F. I. (1999).** Tissue-specific changes in the activities of antioxidant enzymes during the development of the chicken embryo. *Br Poult. Sci. J.*, 40(3):397-405.
- Tag El-Din, H. T., A. A. El-Serwy, E. H. Abou-Egla, I.A. Homuda and A. L. Awad (2004).** Effect of in-ovo injection material in duck eggs on hatchability ,embryonic mortality and ducklings performance . *Egypt. J. Anim. Prod.*, 41:459 – 469.
- Tako, E., P. R. Ferket and Z. Uni (2004).** Effects of in ovo feeding of carbohydrates and beta hydroxy betamethyl butyrate on the development of chicken intestine. *Poult. Sci.*, 83, 2023: 2028.
- Tona, K., E. Decuypere and W. Coucke (2001).** Effects of strain, hen age and transferring eggs from turning to stationary trays after 15 to 18 days of incubation. *Br. Poult. Sci.*, 42: 663-667.
- Tona, K., F. Bamelis, B. De Ketelaere, V. Bruggeman, V. M. B. Moraes, J. Buyse, O. Onagbesan, and E. Decuypere (2003).** Effects of egg storage time on spread of hatch, chick quality, and chick juvenile growth. *Poult. Sci.*, 82, 736: 741.

- Tona, K. O., O. Onagbesan, B. De Ketelaere, E. Decuypere and V. Bruggeman (2004).** Effects of age of broiler breeders on egg quality, hatchability, chick quality, chick weight and chick post-hatch growth to 24 days. *J.Appl. Poult.Res.*, 13:10-18.
- Tullett, S. C. (1990).** Science and the art of incubation. *Poult. Sci.*, 69:1-15.
- Tullett, S. G. (1995).** Incubation. In: P. Hunton (ed.). *World Animal Science C9, Poultry Production, the Ontario Egg Producers' Marketing Board*, Mississauga, Ontario, Canada.
- Uni, Z., P. R Ferket, E Tako, and Kedar (2005).** In ovo feeding improve status of late-term chicken embryos. *Poult.Sci.* 84:764-770.
- Urbaityte R., L. Danyla, A. Sederevicius and H. Jeroch (2006).** Effect of dietary L-carnitine supplementation on sows performance. *Med. Wet.*, 62 (5), 527–530
- Vicari, E., and A. E. Calogero. (2001).** Effects of treatment with L-carnitine in infertile patients with prostato-vesiculopididymitis. *Hum. Reprod.*, 16:2338–2342.
- Vieira, S. L. (2007).** Chicken Embryo Utilization of Egg Micronutrients. *Brazilian J. Poult. Sci.* Vol. 9: 01 – 08.
- Wang, Y. W., Y. M. Guo, D. Liu, Y. Yan and D. Ning. (2012).** Changes of biochemical parameters and proteomics in liver of broiler chickens with cold-induced ascites. *Poult. Sci. Assoc. Annual Meeting*, Athens, Georgia, America, 149.
- Wilson, J. H. (1991).** Bone strength of caged layers as affected by dietary calcium and phosphorus concentrations, reconditioning, and ash content. *Br. Poult. Sci.*, 32:501–508.
- Wilson, H. R. (2004).** Hatchability Problem Analysis I. Web:<http://edis.ifas.ufl.edu/AA204>. Accessed in June, 2006.
- Yalcin, S., A. Ergün, H. Erol, S. Yalçın and B. Özsoy (2005).** Use of L-carnitine and humate in laying quail diets. *Acta Vet. Hung.*, 53:361-370.
- Zakaria, A.H. and M.A. El-Anezi (1996).** Effect of ascorbic acid and cooling during egg incubation on hatchability, culling, mortality and the body weights of broiler chickens. *Poult. Sci.*, 75:1204–1209.
- Zhai, W., S. Neuman, M. A. Latour and P.Y. Hester. (2008).** The effect of in ovo injection of L-carnitine on hatchability of White Leghorns. *Poult. Sci.*, 87: 569- 572.

الملخص العربي

دور المحاليل المغذية أثناء التطور الجنيني في تحسين الفقس والأداء الإنتاجي للكتاكيت
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تهدف هذه الدراسة إلى اختبار تأثير رش بيض التفريخ ببعض المحاليل الغذائية مثل فيتامين ج - والكارنيتين على نسبة الفقس، والنفوق الجنيني، وصفات جودة الكتكوت، وبعض صفات الدم ونمو الكتاكيت الفاقسة. استخدم عدد 3600 بيضة من سلالة دجاج أمهات التسمين التجارية (كب) عمر 35 أسبوع. تم تقسيم البيض عشوائيا إلى 6 مجموعات (600 بيضة / مجموعة) كل مجموعة احتوت على 4 مكررات واحتوت كل مكرره على 150 بيضة. تم رش البيض أثناء التطور الجنيني في اليوم الأول، السابع، الرابع عشر، و الثامن عشر من التطور الجنيني. وكانت المعاملات كالاتي: 1- (مجموعة الكنترول السلبي بدون رش فيتامين ج والكارنيتين)، 2- الماء المقطر (مجموعة الكنترول الزائف)، 3- الماء المقطر + 4 جرام فيتامين ج لكل لتر، 4- الماء المقطر + 6 جرام فيتامين ج لكل لتر، 5- الماء المقطر + 4 جرام كارنيتين لكل لتر، 6- الماء المقطر + 6 جرام كارنيتين لكل لتر. في نهاية عملية الفقس، تم كسر كل البيض الغير فاقس لتقدير مراحل النفوق الجنيني. أشارت النتائج أن رش البيض بمعدل 6 جرام فيتامين ج لكل لتر ماء أدى لتحسين نسبة الفقس معنويا. انخفض النفوق الجنيني بشكل ملحوظ معنويا مع المجموعات المرشوشة بفيتامين ج والكارنيتين مقارنة مع مجموعات الكنترول. بالإضافة إلى ذلك تحسنت جودة الكتكوت و النمو بعد الفقس بشكل ملحوظ معنويا مع تطبيق نظام رش المحاليل المغذية. وعلاوة على ذلك، تأثرت معظم صفات الدم بشكل ملحوظ معنويا بسبب الآثار الإيجابية لرش البيض بفيتامين ج والكارنيتين حيث انخفض تركيز هرمون الكورتيكوستيرون و الذي لوحظ في الأجنة نتيجة رش البيض بفيتامين ج والكارنيتين. سجلت كرات الدم الحمراء ارتفاعا ملحوظا في الاجنه الناتجة من البيض المرشوش ب 4 جرام فيتامين ج ، في حين سجلت أعلى قيمة لتركيز الهيموجلوبين في اجنه البيض المرشوش ب 6 جرام فيتامين ج مقارنة مع المجموعات التجريبية الأخرى. وعلاوة على ذلك تحسن النمو بعد الفقس مع رش البيض بالمحاليل المغذية. معظم صفات الذبيحة تأثرت بشكل ملحوظ معنويا بسبب رش البيض بفيتامين ج والكارنيتين خصوصا فيما يتعلق بنسبة الدهون في منطقة البطن حيث انخفضت بشكل ملحوظ معنويا. ويمكن أن نستخلص من هذه الدراسة إلى أن رش البيض المخصب الناتج من سلالة اللحم التجارية (كب) بفيتامين ج والكارنيتين بجرعات 4 أو 6 جرام / اللتر في اليوم الأول و السابع و الرابع عشر و الثامن عشر من التطور الجنيني أدى الى تحسن ملحوظ في نسبة الفقس وصفات الجودة للكتكوت. ومن ثم فإن رش البيض بهذه العناصر الغذائية يمكن أن يستخدم كوسيلة من الوسائل الناجحة للحد من نفوق الأجنة وتحسين نسبة الفقس و النمو بعد الفقس.