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IMPACT OF *IN-OVO* INJECTION WITH DIFFERENT IRON FORMS ON HATCHABILITY TRAITS AND POST- HATCHING GROWTH PERFORMANCE OF SUDANY DUCKLINGS

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ABSTRACT: A total of 1050 Sudany duck eggs were used in this experiment. Eggs were divided randomly into 7 treatments with three replicates each to study the influence of iron in different forms *In-Ovo* injection on hatchability traits and post-hatch performance of Sudany ducklings. The first group (G1) was served as control, (without injection); the eggs in 2nd (G2) and 3rd (G3) groups injected with 0.1 and 0.2 ml saline solution contains 10 and 20 ppm Nano organic iron. The eggs in 4th and 5th groups were injected with 0.1 and 0.2 ml saline solution contains 10 and 20 ppm Nano inorganic iron. The eggs in 6th group was injected with 0.1 saline solution contain 10 µg organic iron, while the eggs in 7th group was injected with 0.1 ml saline solution contain 10 µg inorganic iron. The results indicated that, no significant differences of different forms of iron on hatchability traits. At hatch, ducklings hatched from eggs in groups G2 and G3 recorded significantly high values of A/G ratio, MDA and HDL. Ducklings hatched from eggs injected with different forms of iron had significantly higher values of plasma IGF_1 and T_3 hormone and lower values of LDL than the control group. Ducklings hatched from eggs in group (G6) resulted in significantly higher values of plasma RBCs, HB and heterophiles than the control group. Also, all treated groups had high values of plasma WBCs compared to control except G7. Duckling in G2,G5 and G6 gave high LBW at 12 and 16 weeks of age . Also, duckling in (G2) recorded significantly high values of BWG throughout the trial period. These results suggest that, pre incubation *In-Ovo* injection with different forms of iron improved some blood plasma constituents of chicks at hatch and enhance post-hatch productive performance of Sudany ducklings from hatching until 16 weeks of age

Keywords: In-Ovo Injection-Iron Forms- Hatchability- Growth Performance -Ducklings

INTRODUCTION

The high surface activity of Iron nanoparticles and penetration into cells can actively influence the intracellular metabolism by stimulating various processes. Iron (Fe) was found to be modulating the expression of humoral or cellular immunity related genes. Zhai et al., (2015) indicated that *In-Ovo* injected into the egg yolk sac with 25- 125 ppm Fe nanoparticles improved embryonic growth and body weight. Nowadays, The use of nano-particles of diameters between 1 and 100 nm is applied in engineering, and agriculture science (Scott and Chen, 2002 and Oberdorster et al., 2007). Nutrient management *In-Ovo* may provide an alternative method for poultry industry to increase hatchling weight. Chicks are affected by the nutrients in yolk remaining in the peritoneal cavity post hatching. Thus, a continually and precisely regulated supply of trace elements derived from stores within the egg is essential to ensure avian embryonic survival. The high-metabolic rate, fast-growing rate of chicken embryos could be liable to mineral deficiency that led to metabolic disorders (Tona et al., 2004).

Iron is essential for a variety of physiological processes in livestock (e.g. DNA synthesis, oxygen transport, etc.) as illustrated by Lozoff et al., (2006); Whitnall and Richardson, (2006) and Li and Zhao, (2009). National Research Council, NRC (1994) recommended 50-120 ppm daily intake of Fe for poultry. Iron in the form of nano-particles has been reported to be less toxic than inorganic iron salts (Nikonovet al., 2012). Additionally, they have prolonged effects on biological activities (Kovalenko and Folmanis, 2006). Iron nanoparticles are more stable in air and have the ability to be degraded or metabolized in vivo, making them excellent candidates for a large number of applications (Bronstein et al., 2007).

Sudany ducks is one of local duck breeds in Egypt, it looks like Muscovy ducks in the feathers form and the red crest around the eyes and above the beak, but, it is less than in the productive traits. There is a little attention of the producers due to low productivity resulting from subfertility and hatchability, although their meats are more favorable to the Egyptian consumer. From the previous studies on vital role of Fe nanoparticles on embryonic development and subsequent growth performance of chick, this work aimed to study the effects of *In-Ovo* injection of Sudany duck eggs with different forms of iron and doses of iron at pre- incubation on hatchability traits, blood parameters of ducklings at hatching, subsequent growth performance and immunity of ducklings.

MATERIALS AND METHODS

This study was carried out at El – Serw Water Fowl Research Station, Animal Production Research Institute, Agriculture Research Center, Ministry of Agriculture, Egypt. A total of 1050 Sudany duck eggs were used in this experiment. Eggs were divided randomly into seven treated groups with three replicates each.

Egg injection protocol and artificial incubation:

The first group (G1) served as control group without any injection. The eggs in 2nd (G2) and 3rd (G3) groups injected with 0.1 and 0.2 ml saline solution contains 10 and 20 ppm nano organic iron. The eggs in 4th (G4) and 5th (G5) groups were injected with 0.1 and 0.2 ml saline solution contains 10 and 20 ppm

nano inorganic iron. The eggs in 6th group (G6) was injected with 0.1 saline solution contain 10 micro gram Fe- organic, while the eggs in 7th group (G7) was injected with 0.1 ml saline solution contain 10 micro gram Fe- inorganic. All eggs were injected on the large end of egg before incubation. Iron oxide nano particles were prepared according to Reimers and Khalafalla (1974). All eggs were incubated at 37.5 °C and 60% relative humidity in an automatic incubator. At day 25 of incubation all eggs were transferred to the hatcher to complete the hatching process for 3 days at 99.0^0F and 75% RH. At the 10th day of incubation, eggs were candled to count fertile eggs number and estimate early embryonic mortality (%). After complete hatching, live hatched chicks, un-hatched eggs and dead chicks were counted to determine hatchability and late embryonic mortality (%).

Ducklings management and diets:

After hatching, a total number of 210 ducklings from all injected groups were weighed and distributed into seven experimental groups with three replicates each. Ducklings were reared under similar environmental and managerial conditions. Ducklings from all seven treated groups were fed on commercial starter (1-8 wks) and grower (9-16wks) mash diets. The composition and calculated analysis of the experimental diets are shown in Table (1).

Blood plasma biochemical analysis:

At hatching, individual blood samples were withdrawn from five chicks within each treatment in heparinized test tubes. Before blood centrifugation, blood sample was used to estimate hematological parameters. Hemoglobin concentration (Hb,mg/dl) was determined using Haemometer as described by Tietz, (1986) and hematocrit (Ht,%). Red blood cells count (RBC's $X10^6$), Mean Corpuscular Volume (MCV,μm^3), Mean Corpuscular Hemoglobin (MCH, Pg), Mean Corpuscular Hemoglobin Concentration (MCHC, g/dl), White blood cells (WBC's), Hetrophilus (%), Lymphocytes (%) and H/L ratio, were determined according to the method of Helper (1966). Then blood samples were centrifuged at 3500 rpm for 15 minutes to get blood plasma, which stored at – 20 °C until used. Blood plasma total protein (TP, g/dl), albumin (g/dl), globulin (g/dl) was obtained by subtracting the concentration of albumin from that of plasma total protein and A/G ratio. Alanine aminotransferase (ALT, U/ml) , aspartate aminotransferase (AST,U/ml) ,triglycerides (TG, mg/dl) , cholesterol (mg/dl), high density lipoprotein (HDL, mg/dl) and low density lipoprotein (LDL, mg/dl) were colorimetrically determined using commercial kits purchased from Bio-Diagnostic, Cairo, Egypt, according to the manufacturers' instructions.

Hormones of Insulin like growth factor-I (IGF-I, ng/ml) and triiodothyronine (T3, ng/ml) were assayed using ELISA kits obtained from Sino Gen Clon Biotech, No.9 BoYuan Road, YuHang District 311112, HangZhou, China. Total antioxidant capacity (TAC, mmol/ml) and Malondialdehyde (MDA, nmol/ml) in plasma were calorimetrically determined using commercial kits purchased from Bio-Diagnostic, Cairo, Egypt, according to the manufacturers' instructions.

Data collection:

1-Hatchability traits: At hatch, all hatched ducklings were individually weighted and the hatchability percentage of set eggs was calculated. Also, the non

– hatched eggs were broken out to detect the percentages of early and late embryonic mortality (EEM and LEM, respectively).

2-Growth performance parameters: Live body weight (LBW,g) of ducklings were recorded at hatch, 8, 12, and 16 weeks of age. The body weight gain (BWG, g/day), feed consumption (FC, g), and feed conversion ratio (FCR) were calculated through the periods from 0-4, 5-8, 9-12, 13- 16 and 0- 16 weeks of age.

Statistical analysis:

Data were subjected to one – way analysis of variance using general linear model (GLM) procedure of SAS program (SAS, 2004) based on the following model:

$Yij = \mu + Ti + eij$

where,

Yij = An observation, μ = Overall mean,

Ti =Effect of treatment (1, 2, …., 7), and eij = Random error.

Significant differences among treatment means were tested by Duncan's multiple range test (Duncan, 1955) at a probability level of 0.05 ($P \leq 0.05$).

RESULTS AND DISCUSSION

Hatching traits:

Data of Table (2) showed the impact of pre- incubation *In-Ovo* injection of Sudany duck eggs with different forms of iron. It is noticed that, no significant differences of different form of Fe on hatchability of set eggs, EEM, LEM and chick weight. It is clear that ducklings hatched from eggs in G6, G2 and G3 had heavier body weights at hatch than the other treated groups. Moreover, the percentages of EEM was increased in G3, while, an obvious decrease in G5, G6 and G7 groups. A similar trend was observed for the LEM (%), where G2 recorded the highest LEM and G7 showed the lowest percentage. These results are in agreement with those of Saki et al. (2014) Who reported that hatchability and embryonic mortality were not significantly affected between the group fed 50 and 150 ppm of iron-chelated.

Effect of treatments on blood parameters of newly hatched ducklings:

Data in Table **(**3) showed the effect of different Fe sources on plasma protein fractions of newly hatched ducklings. It is clear that treatments had no significant effect on all plasma protein fractions in the newly hatched ducklings, however, A / G ratio was significantly low in G1, G7 and G4 groups, to a lesser extent in G5 and G6 treatments, respectively. This indicates that these groups had high plasma globulin levels, which may reflect better immunity at later growing periods.

It appears that, plasma proteins profile of a given bird is a reflection of its metabolic activities related to protein synthesis and degradation, this supports the findings of many authors who observed that Fe has the ability to bind protein and enhanced DNA synthesis which in turn affect plasma protein level at different growth periods (Angle, 2007 and Yair and Uni, 2011).

Table (3) shows also the impact of *In-Ovo* injection of Fe on liver enzymes (AST & ALT) activities. It is noticed that, Fe injection into eggs during the incubation period has negative effects on AST activity of day-old ducklings. The worse effect was recorded for those hatched from eggs of G3, G4, G6 and G7 compared to other treatments. This effect was also observed for ALT activity, in chicks of G4 and G6 only. It is claimed that, the observed increase in

AST activity of the iron-injected group, may be explained in the light of the fast embryonic growth during incubation as evidenced by their heavier hatching weight. Since, the rapid growth needs higher metabolic process, which may affect liver tissues causing some little disorders due to rapid metabolism. As liver is the main metabolic organ in the body, its hepatocytes might be damaged, resulting in AST and ALT elaboration in the blood stream. However, under the conditions of the present study, there is no signs of morbidity of ducklings, indicating of better liver function.

Data revealed also that, *In-Ovo* injection of different iron sources and forms had significant effect on plasma TG, LDL and HDL levels of day-old ducklings **(**Table, 3) while cholesterol was affected significantly. In general, ducklings hatched from eggs injected with different forms of iron had significantly lower values of blood plasma TG, cholesterol and LDL compared to control group. The best group was G3 and that of G6 had significantly higher values of HDL compared to the other treated groups. It is suggested that, iron overload can negatively modify the blood lipid metabolism by increasing the ability of homocysteine to create oxy-cholesterol in arteries and by changing the content of cholesterol and its fractions (Choi et al., 2013). Data revealed also that, the antioxidative defense system in terms of TAC was significantly enhanced in G6 compared to the other groups. This was not paralleled with the MDA results which showed increases in chicks in all treatments, except G7. Although this increase indicates higher lipid peroxidation, Similar results were reported by Mogahid et al. (2019) who studied the effect of *In-Ovo* injection of organic and inorganic iron and their nanoparticle forms on blood parameters.

Results in Table (3) showed that, all groups injected with Fe had significantly higher plasma IGF-1 and T3 than that in G1. This result is expected as both hormones acts synergistically in accelerating nutrients (egg yolk with its extra iron supplements) metabolism in response to the higher T3 level, leading to more somatic growth due to the well-known physiological function of IGF-1 in regulating the developmental pattern of living organisms. In this concern, iron was suggested as an important auxiliary factor for the key enzyme of 5, -deiodinase, which synthesizes triiodothyronine (T3) in animals. Since, triiodothyronine is a main hormone that regulates animal growth by controlling the body's energy and protein anabolism. These results agree with Boostani et al. (2015) and Mogahid et al. (2019) who observed that T3 and IGF-1 are very important for embryonic growth.

Table (4) shows the effect of *In-Ovo* injection of different iron forms on RBC's count, hemoglobin concentration, hematocrit and blood indices of hatched ducklings.

It appears from the present results that *In-Ovo* injection of iron had improved all hematological parameters, in terms of RBC's count and hemoglobin concentration which in turn influence the other hematological traits. The blood indices showed, however, considerable variations where, the MCV and MCH concentration were not significantly influenced. Moreover, MCHC was higher for all iron-injected groups except G7. The increased Hb concentration and the RBC's count are expected due to excess iron deposition and utilization by

chick embryos during their early embryonic development. On the other hand, the differences in MCHC may be resulted from the changes in RBC's count and Hb concentration. While, Ht (%) is greatly affect MCV and depends on the size of erythrocytes and their ability to hold a saturable amount of Hb. In general, the present results are in accordance with those reported by (Mogahid et al., 2019) who claimed that, treatment with iron increases blood Hb concentration, RBC's count and packed cell volume. Also, the increased Hb content in the blood may be due, in part to, the effect of intensification of oxidation and reduced haeme phagocytic processes in the embryos spleen especially those receiving iron – nanoparticles. Previous studies showed that the increased concentration of hemoglobin was usually accompanied by an increased number of RBC (Sosnowska et al., 2013).

Moreover, the influence of *In-Ovo* injection by iron in different forms on WBC's differential count of day-old ducklings is also presented in Table (4). Results showed that, WBC's count was significantly increased in chicks from all *In-Ovo* injected groups, with the higher value being recorded for G3, G4 and G6, respectively. This increase was also observed in the percentage of lymphocytes, and heterophils where their percentages were higher in the same groups compared to G1 and G2. This change in the heterophils and lymphocytes were reflected in their ratio (H/L) where it reflects an interested result. Indeed, the increased H/L ratio is a function of the number of heterophils: lymphocytes that indicates moderate stress imposed to the newly hatched ducklings, as the ratio exceeds 0.5 for all treatments (except G5). The higher H/L ratio may indicate a state of stress responses of embryos to internal and external stressors that resulted in an increase in heterophils count as the main phagocytic cells in the body which in turn increased H/L ratio as the case in some treatment groups in our study. However, all groups had achieved better growth performance during the whole experiment indicating of a mild or normal acceptable stress. It is assumed that the higher H/L ratio of some treated groups may be resulted from the increase of metabolic activity of the embryos during organogenesis accompanied with an increase in metabolic heat production representing a state of stress that caused an increase in H/L ratio without any negative impacts on chicks' development. In this concern, Gross and Siegel (1983) and Hassan (2018). reported that H/L ratio was considered as a more reliable indicator for determining stress response in poultry.

Productive performance of hatched ducklings:

Results presented in Table (5), showed significant effects of *In-Ovo* injection of sudany duck eggs with different forms of iron on post- hatch performance of sudany ducklings. It is noticed from the results that *In-Ovo* injection with nano organic (G٢) was improved ($P \leq 0.05$) LBW of sudany ducklings at 12 and 16 wks of age. Also, the group G2 recorded highest value of body weight gain at the period of 0-16 wks of age compared with the other treatment groups. Because, G2 consumed more feed from 13-16 wks of age. Foye et al. (2006) found that, digestive capacity, growth rate and feed efficiency were improved by *In-Ovo* injection

Data in Table (5), revealed the ducklings LBW were significantly ($P \leq 0.05$) improved for hatched ducklings from G6 at 8 wks of age.

It is likely noticed that, G5 and G6 recorded higher values of BWG at period of 5-8 wks of age. While, G7 recorded significantly ($P \leq 0.05$) higher value of BWG at period of 9-12 wks of age.

Concerning the effect of *In-Ovo* injection with Fe inorganic on subsequent ducklings feed conversion ratio, it could be noticed that the group G7 had significantly ($P \leq 0.05$) better value of FCR at overall period from 0-16 wks of age. Similar results were obtained by Mogahid et al. (2019), and Saki et al. (2014).

CONCLUSION

The present study recommends that: The *In-Ovo* injection of Sudany duck eggs with different forms of iron could be used to improve productive performance of Sudany ducklings during rearing period (0- 16 wks of age), specially Nano iron particles and this work needs more researches to study the effect of various dose of nano Fe particles and different times of injection on hatchability characteristics.

Table(1): Composition and calculated analysis of the basal diets.

Ingredients %	Starter	Grower
Yellow Corn	61.70	71.00
Soybean meal (44 %)	34.55	17.60
Wheat bran	0.00	7.60
Di-calcium phosphate	1.60	1.60
Limestone	1.45	1.50
Vit. & Min. premix[1]	0.30	0.30
NaCl	0.30	0.35
DL. Methionine	0.10	0.05
Total	100.0	100
Calculated Analysis [2]		
Crude protein %	20.01	15.02
ME (Kcal / kg)	2841	2870
Ether extract . %	2.86	3.07
Crude fiber %	3.94	3.63
Calcium (%)	1.04	1.00
Av. phosphorus (%)	0.44	0.42
Lysine %	1.17	0.70
Methionine %	0.45	0.30
Methio + Cyst %	0.78	0.58
Sodium	0.13	0.16

1-Each 3 kg of the Vit and Min. premix manufactured by Agri-Vit Company, Egypt contains: Vitamin A 10 MIU, Vit. D 2 MIU, Vit E 10 g, Vit. K 2 g, Thiamin 1 g, Riboflavin 5 g, Pyridoxine 1.5 g, Niacin 30 g, Vit. B12 10 mg, Pantothenic acid 10 g, Folic acid 1.5 g, Biotin 50 mg, Choline chloride 250 g, Manganese 60 g, Zinc 50 g, Iron 30 g, Copper 10 g, Iodine 1g, Selenium 0. 10 g, Cobalt 0.10 g. and carrier CaCO3 to 3000 g..
2-According to Feed Composition Tables for animal and poultry feedstuffs used in Egypt (2001).

Table (2): Effect of *In-Ovo* injection of Sudany duck eggs with different form of iron at hatching traits.

		Iron Experimental treatments**						S.E.M	P Value
Traits	**G1**	**G2**	**G3**	**G4**	**G5**	**G6**	**G7**		
Hatchability of set eggs	58.67	58.67	58.67	58.67	58.00	59.33	59.33	1.15	0.98
EEM*	8.00	7.33	8.67	8.00	7.33	7.33	6.67	0.84	0.72
LEM	13.33	15.33	14.00	14.00	14.00	13.33	12.67	1.007	0.66
Chick weight	43.67	45.33	45.33	44.33	44.33	46.00	43.33	1.19	0.68

EEM & L E M.= early and late embryonic mortality; SEM= standard error of means;
**- No significant differences were detected.
G1= control, G2=10 ppm Nano organic iron, G3= 20 ppm Nano organic iron, G4=10 ppm Nano inorganic iron, G5= 20 ppm Nano inorganic iron, G6=10 µg organic iron, G7= 10 µg inorganic iron

Table (3): Effect of *In-Ovo* injection with different forms of iron on blood plasma constituents of Sudany ducklings at hatching.

Traits	G1	Different forms of iron treatments						S.E.M	*P* value
		G2	G3	G4	G5	G6	G7		
Plasma Proteins (g/dl)									
Total protein	4.09	3.96	4.16	4.09	4.28	4.13	٤.12	0.14	0.84
Albumin	2.20	2.34	2.52	2.27	2.41	2.29	2.27	0.10	0.45
Globulin	1.88	1.63	1.64	1.82	1.88	1.84	1.85	0.09	0.26
A/G ratio	1.17^{c}	1.47^{ab}	1.57^{a}	1.23^{bc}	1.30^{abc}	1.30^{abc}	1.23^{bc}	0.08	0.057
Liver Enzymes (U/ml)									
AST	56.93^{ab}	49.60^{b}	60.17^{ab}	62.60^{ab}	56.83^{ab}	69.40^{a}	63.77^{ab}	4.34	0.12
ALT	15.93^{ab}	13.33^{b}	15.57^{ab}	18.00^{ab}	13.87^{b}	21.27^{a}	15.97^{ab}	1.92	0.14
Plasma lipids (mg/dl)									
TG	61.57^{a}	59.37^{b}	48.77^{de}	46.8^{f}	49.73^{d}	47.17^{ef}	52.47^{c}	0.58	0.0001
Cholesterol	77.27^{a}	71.5^{c}	74.87^{b}	74.43^{b}	67.47^{e}	69.47^{d}	73.73^{b}	0.56	0.0001
HDL	19.53^{cb}	19.97^{b}	23.03^{a}	19.1^{cb}	17.93^{c}	21.73^{a}	19.27^{cb}	0.51	0.0357
LDL	45.42^{a}	40.4^{cd}	42.08^{bc}	45.97^{a}	39.59^{cd}	38.3^{d}	43.97^{ab}	0.87	0.0001
Oxidative status									
TAC(mmol/ml)	1.26^{ab}	1.16^{b}	1.09^{b}	1.26^{ab}	1.21^{ab}	1.38^{a}	1.25^{ab}	0.06	0.067
MDA (nmol/ml)	24.2^{ab}	30.4^{a}	29.3^{a}	26.7^{ab}	28.7^{a}	24.8^{ab}	20.5^{b}	1.93	0.034
Hormones (ng/ml)									
IGF-1	31.20^{c}	39.63^{abc}	35.97^{bc}	43.17^{ab}	45.83^{a}	40.43^{abc}	33.77^{bc}	2.89	0.03
T_3	3.53^{b}	3.82^{ab}	3.88^{ab}	3.96^{ab}	4.00^{ab}	4.33^{a}	3.49^{b}	0.19	0.08

SEM= standard error mean; Sig= significance ; T_3= triiodothryonine; TAC= total antioxidant capacity; IGF-1= Insulin- like growth factors.

a,b,c…means within row with different superscripts are significantly different ($P \leq 0.05$).

G1= control, G2=10 ppm Nano organic iron, G3= 20 ppm Nano organic iron, G4=10 ppm Nano inorganic iron, G5= 20 ppm Nano inorganic iron, G6=10 µg organic iron, G7= 10 µg inorganic iron

Table (4): Effect of *In-Ovo* injection with different forms of iron on hematological traits of Sudany ducklings at hatching.

Parameters		Iron treatments in different forms						SEM	P value
	G1	G2	G3	G4	G5	G6	G7		
RBCs x10^6	3.84^{b}	$4.6\ 9^{ab}$	4.2^{ab}	4.59^{ab}	4.91	5.06^{a}	4.11^{ab}	0.21	0.009
Hb (mg/dl)	9.08^{b}	10.79^{ab}	10.65^{ab}	10.21^{ab}	10.72^{ab}	10.99^{a}	9.74^{ab}	0.36	0.021
Ht (%)	33.33^{b}	37.0^{ab}	38.33^{a}	35.67^{ab}	33.0^{b}	37.33^{ab}	36.33^{ab}	0.97	0.011
MCV (µ m^3)	87.5	79.37	86.13	77.83	76.47	74.13	88.6	5.45	0.127
MCH(Pg)	23.77	23.13	23.73	22.23	21.93	21.83	23.67	1.27	0.816
MCHC(g/dl)	27.27^{b}	29.1^{ab}	27.93^{b}	28.57^{ab}	32.5^{a}	29.47^{ab}	26.8^{b}	0.94	0.015
WBCs x10^3	20.70^{b}	27.00^{a}	28.94^{a}	28.9^{a}	25.9^{a}	28.75^{a}	25.45^{ab}	1.00	0.005
Heterophils (%)	33.00^{ab}	27.33^{b}	34.0^{ab}	33.33^{a}	28.67^{ab}	37.67^{a}	36.33^{ab}	2.08	0.031
Lymphocytes (%)	51.33	46.33	52.00	52.67	56.67	55.33	51.00	2.51	0.18
H/L	0.65^{ab}	0.59^{ab}	0.66^{ab}	0.63^{ab}	0.51^{b}	0.68^{ab}	0.72^{a}	0.04	0.046

SEM= Pooled standard error mean; RBC`s = Red blood cells count; MCV= Mean Corpuscular Volume; MCHC= Mean Corpuscular Hemoglobin Concentration; WBC's= White blood cells

a, b, c means within row with different superscripts are significantly different (P≤0.05).

G1= control, G2=10 ppm Nano organic iron, G3= 20 ppm Nano organic iron, G4=10 ppm Nano inorganic iron, G5= 20 ppm Nano inorganic iron, G6=10 µg organic iron, G7= 10 µg inorganic iron

Table (5): growth performance of ducks as affected by in- ovo injection of different iron forms

Age (week)		Iron Treatments in different forms						S.E.M	*P* value
	G1	G2	G3	G4	G5	G6	G7		
Body weight (g) at:									
Hatch	43. 7	45.3	45.3	44.3	44.3	46.0	43. 7	1.19	0.7
4	652.33	661.0	662.0	649.3	649.7	672.33	632.67	24.33	0.9
8	1275.0^{abc}	1278.3^{abc}	1310.7^{ab}	1198.3^{bc}	1326.7^{a}	1351.7^{a}	1168.3^{c}	35.4	0.02
12	2376.7^{ab}	2483.3^{a}	2353.3^{ab}	2320.0^{ab}	2281.7^{b}	2400.0^{ab}	2456.7^{a}	48.8	0.01
16	2716.7^{bc}	2933.3^{a}	2716.7^{bc}	2683.3^{c}	2673.3^{c}	2783.3^{bc}	2825.0^{ab}	42.5	0.007
Body weight gain (g/day):									
0 – 4	608.67	615.67	616.67	605.0	605.33	626.33	589.0	24.4	0.9
5 – 8	622.7^{ab}	617.3^{ab}	648.7^{ab}	549.0^{b}	677.0^{a}	$679. 3^{a}$	535.7^{b}	36.1	0.07
9 – 12	1101.7^{bcd}	1205.0^{ab}	1042.7^{cd}	1131.7^{bc}	$955. 0^{d}$	$1048. 3^{cd}$	$1288. 3^{a}$	47.8	0.004
13 – 16	340.00	450.00	363.33	363.33	391.67	383.33	368.33	34.8	0.4
0 – 16	2670.3^{bc}	2882.0^{a}	2671.3^{bc}	2639.0^{bc}	2629.0^{c}	2670.7^{bc}	2781.3^{ab}	43.9	0.01
Feed consumption (g):									
0- 4	2666.67	2536.67	2550.00	2600.33	2603.33	2595.67	2603.33	42.1	0.4
5- 8	3183.33	2983.33	3023.33	3100.00	3099.00	3033.33	3083.33	69.3	0.5
9- 12	4920.00	4893.33	4933.33	4722.33	4766.67	4853.33	4803.33	189.9	0.9
13- 16	3133.33^{b}	3633.33^{a}	3166.67^{b}	3333.33^{ab}	3383.33^{ab}	3366.67^{ab}	3216.67^{ab}	125.98	0.16
0- 16	13870.0	14046.7	13673.3	13755.7	13852.3	13849.0	12746.7	398.7	0.38
Feed conversion ratio:									
0- 4	4.37	4.10	4.20	4.33	4.30	4.17	4.40	0.14	0.7
5- 8	5.20^{abc}	4.87^{abc}	4.67^{bc}	5.67^{ab}	4.63^{bc}	4.50^{c}	5.80^{a}	0.31	0.054
9- 12	4.47^{abc}	4.10^{bc}	4.73^{ab}	4.20^{abc}	5.03^{a}	4.67^{ab}	3.73^{c}	0.27	0.06
13- 16	9.47	7.99	8.77	9.33	8.77	9.00	8.77	0.71	0.8
0- 16	5.20^{a}	4.87^{ab}	5.13^{a}	5.23^{a}	5.27^{a}	5.20^{a}	4.60^{b}	0.15	0.06

a,b,c…means within row with different superscripts are significantly different (P≤0.05).

G1= control, G2=10 ppm Nano organic iron, G3= 20 ppm Nano organic iron, G4=10 ppm Nano inorganic iron, G5= 20 ppm Nano inorganic iron, G6=10 µg organic iron, G7= 10 µg inorganic iron

REFERENCES

Angel R.2007. Metabolic Disorders: Limitations to Growth of and Mineral Deposition into the Broiler Skeleton after Hatch and Potential Implications for Leg Problems J. Appl. Poult. Res. 16, 138-149.

Boostani, A.; Sadeghi, A.; Mousav, S. N.; Chamani, M. and K ASHAN, N. 2015. The effect of organic, inorganic and nano selenium on blood attributes in broiler chickens exposed to oxidative stress. Acta Scientiae Veterinariae, 43 (1246): 1-6.

Bronstein, LM.X. Huang, J. Retrum, A. Schumacher, M. Pink, B. D 2007. Stein and B .Dragnea: Chem Materials., Vol.19, No.15, (2007), pp.36243632.

Choi, J. S.; I. U. Koh; H. J. Lee; W. H. Kim; J. Song, 2013. Effects of excess dietary iron fat on glucose lipid metabolism. The Journal of Nutritional Biochemistry 24, 1634–1644.

Duncan, D.B. 1955. Multiple range and multiple F tests. Biometrics, 1:1-42.

Foye, O.T., Uni, Z., McMurtry, J.P. and Ferket, R.P. 2006. The effects of amniotic nutrients administration, in ovo feeding of arginine and/or b-hydroxy b-methylbutyrate (HMB) on insulin-like growth factors, energy metabolism and growth in turkey poults. International Journal of Poultry Science, 5: 309-17

Gross, W. B., and H. S. Siegel. 1983. Evaluation of the heterophil/ lymphocyte ratio as a measure of stress in chickens. Avian Dis. 27:972–979.

Hassan, M. Amal 2018. Effect of *In-Ovo* injection with Nano iron- particles on physiological responses and performance of broiler chicknes under Saini conditions. (IJEAB), vol- 3, issue-3, May- June, 855- 863.

Helper, O. E. 1966. Manual of clinical laboratory methods. Thomas. Spring-Field, Illinois, USA.

Kovalenko L.V. and G.E. Folmanis 2006. Biologic heskiaktivnyenanoporoshki zheleza (Biologically Active Iron Nano powders), Moscow: Nauka.

Li, M., and C. Zhao, 2009. Study on Tibet a chicken embryonic adaptability to chronic hypoxia by revealing differential gene expression in heart tissue. Sci. China C. Life Sci. 52, 284-295.

Lozoff, B., N. Kaci rot, and T. Walter, 2006. Iron deficiency in infancy: Applying a physiologic framework for prediction. Am. J. Clin. Nutr. 84, 1412- 1421.

Mogahid, A. Azza, Ghada, G. Gad; Abdalla, E. A. and El- Wardany I. 2019. Influence of *In-Ovo* injection of inorganic iron and its Nano particles form on growth and physiological response of broiler chickens. (AUJAS), Ain Shams Univ., Cairo, Egypt, 26(2D), 2369- 2376

Nikonov I. N., Folmanisb Yu. G., Folmanisb G. E., Kovalenkob L. V., Lapteva G. Yu, EgorovcFisininc I. A., V. I., and. Tananaevd I. G.,2012. Iron Nanoparticles as a Food Additive for Poultry. Doklady Biological Sciences Vol. 440.

NRC. 1994. Nutrient Requirements of Poultry. National Academy Press, Washington, DC. USA. p.27.

Oberdorster, G., V. Stone and K. Donaldson 2007. J Nano toxicology, Vol.1, pp. 2-25.

Reimers, G.W and S.E. Khalafalla 1974. Production of Magnetic Fluids

by Peptization Techniques. US Patent No. 3843540

Saki AA, Abbasinezhad M, Rafati AA. 2014. Iron nanoparticles and methionine hydroxy analogue chelate *In-Ovo* feeding of broiler chickens. Int J NanosciNanotechnol. 10:187–196.

SAS Institute. 2004. SAS User's Guide: Statistics. Edition 9.1. SAS Institute Inc., Cary, NC.

Scott, N.R and H. Chen 2002. Nanoscale Science and Engineering for Agriculture and Food Systems.\

Sosnowska ,N. M.; Batorska, M0; LUKASIEWICZ, M.; wnuk Agnieszka ; Sawoszm E.; Jaworski, S.; NIEMIEC, J.2013. Effect of nanoparticles of copper and copper sulfate administered in ovo on hematological and biochemical blood markers of broiler chickens. Animal Science No 52, 2013: 141–149

Tietz, N. W. 1986. Textbook of Clinical Chemistry. N. B. Saunder Co., London, Philadelphia, 18.1.

Tona. K., O. M. Onagbesan, Y. Jego, B. Kamers, E. Decuypere, and V. Bruggeman.2004. Comparison of embryo physiological parameters during incubation, chick quality, and growth performance of three lines of broiler breeders differing in genetic composition and growth rate. Poult. Sci.83:507513.

Whitnall, M., and D.R. Richardson, 2006. Iron: A new target for pharmacological intervention in neurodegenerative diseases. Seminars Pediatric Neurol. 13, 186-197.

Yair, r., z. and Uni, 2011. Content and uptake of minerals in the yolk of broiler embryos during incubation and effect of nutrient enrichment. Poultry Science 90, 1523–1531.

Zhai, W., and Rowe, D. E. and Peebles, E. D. 2015.. Effects of commercial in ovo injection of carbohydrates on broiler embryogenesis. Poultry Science, 90 :1295–1301.

الملخص العربى
تأثير حقن بيض البط السودانى بأشكال مختلفة من الحديد على صفات الفقس واداء النمو للكتاكيت الناتجة بعد الفقس

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استخدم فى هذه الدراسة عدد ١٠٥٠ بيضة بط سودانى تم تقسيمها الى سبع مجموعات تجريبية بكل مجموعة ١٥٠ بيضة فى ثلاث مكررات بكل منها ٥٠ بيضة وكانت معاملات الحقن على النحو التالى:

المعاملة الأولى وهى مجموعة الكنترول بدون حقن – المعاملة الثانية والثالثة تم حقنها بالنانو حديد العضوى بمستوى ١٠ و٢٠ جزء فى المليون على التوالى – المعاملة الرابعة والخامسة تم حقنها بالنانو حديد الغير عضوى بمستوى ١٠ و ٢٠ جزء فى المليون- المعاملة السادسة تم حقنها بالحديد العضوى بمستوى ١٠ ميكروجرام- المجموعة السابعة تم حقنها الحديد الغير عضوى بمستوى ١٠ ميكروجرام. تم حقن البيض فى الغرفة الهوائية قبل التفريخ. عند تمام الفقس تم توزيع ٢١٠ كتكوت بط سودانى فى سبع مجاميع تجريبية تمثل تماما معاملات التفريخ التجريبية بكل منها ثلاث مكررات وذلك لدراسة تاثير حقن البيض باشكال الحديد المختلفة اثناء التفريخ على الاداء الانتاجى لكتاكيت البط السودانى حتى عمر ١٦ أسبوع. وقد أوضحت النتائج عند الفقس وجد ان كتاكيت البط الناتجة من حقن البيض بمستويات صور الحديد المختلفة نتج عنها انخفاض معنوى لمحتوى بلازما الدم من الجليسريدات الثلاثية والكوليسترول الكلى والكوليسترول منخفض الكثافة وكانت افضل مجموعة الناتجة من حقن البيض بالنانو حديد العضوى بمستوى ٢٠ جزء فى المليون .

وبصفة عامة ان حقن بيض البط السودانى بمختلف اشكال الحديد ادى الى زيادة معنوية فى هرمونى IGF_1 وT_3مقارنة بالكنترول.

كما وجدت زيادة معنوية لمحتوى بلازما الدم من الكوليسترول عالى الكثافة لكتاكيت البط الناتجة من حقن البيض بمستوى ٢٠ جزء فى المليون نانو حديد عضوى و١٠ ميكروجرام حديد غير عضوى بالمقارنة بالمجاميع التجريبية الاخرى.

أيضا هناك زيادة معنوية لمحتوى بلازما الدم من كرات الدم البيضاء نتيجة حقن البيض بالنانو حديد العضوى والغير عضوى وكذلك الحديد العضوى.

كما حدث تحسن معنوى فى وزن الجسم LBW وكذلك زيادة معنوية فى الزيادة فى وزن الجسم BWG للبط الناتج من حقن البيض بالنانو حديد العضوى .

وقد خلصت الدراسة إلي امكانية حقن بيض البط السودانى بأي من أشكال الحديد المختلفة لتحسين خواص الدم والاداء الانتاجى للبط السودانى خلال فترة النمو وخاصة استخدام جزيئات الحديد النانومترية العضوية.