Influence of spraying different solutions at different incubation periods on hatchability parameters of local Iraqi's eggs.

S M Gatea¹, S M S Altaie ¹, S S Khafajij, T K ALjanabii, D H Shattii ², M A Hussain ³ and

¹ Department of Animal production, College of Agriculture, University of Kerbala, Iraq.
² Microbiology, Faculty of Dentistry, University of Al – Ameed.
³ Animal physiology, Veterinary medicine, University of Kerbala, Iraq.

Email: salah.katia@uokerbala.edu.iq.

Abstract This study was carried out on 26/5/2019 until 17/6/2019 At local hatchery (College of Agriculture / University of Karbala), to investigate of different spraying protocols on the eggs of local chicken with the eggs of local chicken with different solutions at variant periods on embryonic mortality during incubation period and on the hatchability of setting eggs. 600 eggs (55 g weight) used in this study from local chicken divided into three groups each group divided into 5 groups spraying by distilled water, olive oil, ethanal alcohol 70%, egg albumen and not treated groups that represented as GW, GO,GE,GC. first group G1 (n =120) was sprayed at the first day of incubation period, the second group G2 was spraying at beginning of the 2nd week of incubation at aged 7 days, the third group G3 in which the eggs was sprayed at the beginning of 3rd week of incubation at aged 14 days. The results recorded significant reduction of early embryonic mortality in G1 and GC. The statistical analysis of the in all groups spraying by ethanal alcohol GE, water group GW and control groups GC. At 14 days of the incubation period, there were no significant differences. At 15-21 days of incubation period, late embryonic mortality was not recorded in groups treated olive oil, water and albumen when compared with groups treated with alcohol or control. Conclusion, that spraying Iraqi local eggs with water and olive oil at 14 days of incubation period caused high percentages of hatchability of setting and fertilized eggs recorded 100% compared with different materials that used in current research.

1. Introduction
The standard conditions for chicken embryonic development and growth need some assistance for supporting embryonic development and hatchability percentage during the stages that given a high productive rate of local chickens, but there was a lowering in hatchability percentage during the last phase of Incubation to the small weight of the egg [1]. Several researchers attempted to improve the Hatchability percentage by applying different materials on surface eggs during the incubation period [2], therefore, preventing egg shell from contamination could be elevated embryo survivability rate [3]. Besides, the researchers used water and several types of oils extracted from natural compounds among these active compounds is olive oil [4 ; 5 ; 6] which contains docophorals that are natural antioxidants, as well as, it contained important phenolic compounds such as hydroxytyrosol, vanillin, vanillic acid, tyrosol and caffeic acid which possessed the inhibitory effect for microbes through the...
complex formation of cells with the outer walls of bacteria that prevented their spread [7]. Olive oil was consisted of mainly of the triglyceride esters and palmitic acid and of other fatty acids[8] . Because high embryonic mortality during incubation period of local chicken eggs that negatively reflected on hatchability percentage, this study was designed to improve this character by using various spraying solutions.

2. Materials and Methods
This study was conducted at local hatchery belong to the college of Agriculture /Karbala University from 26/5/2019 until 17/6/2019, where the local chicken eggs were obtained from the field of raising local chicken immediately after collecting and eggs storage 15°C the second day. breeder flock was fed production diet that containing 16.88% crude protein and metabolic energy 2726 Kcal/ kg.

2.1. Spraying hatching eggs
600 eggs with average weight 55 g were used in current study and collected from local chicken flock, it divided into five treatments and each treatments in divided into three replicates per treatment and spraying by distilled water, olive oil, ethanol alcohol, egg albumen and not treated 'control' groups that represented as GW, GO, GE and GC, respectively. first group G1 (n=120) is spraying at the first day of incubation period by these solutions. And the second group G2 is spraying at beginning of the 2nd week of incubation at aged 7 days by these solutions, the third group G3 in which the eggs was spraying at the beginning of the third week of incubation at aged 14 days by these solutions.

2.2. Hatching process
After disinfection, egg groups were transported to the Hatchery machine. Each replicate of 120 eggs were placed in egg trays. After cleaning the egg surface with 100% ethanol, the eggs shell was sprayed with water, olive oil (olea europaea), ethanol alcohol 70% and egg's albumen. Standard procedure of incubation process were followed; dry temperature were 37.5 C and 37 C, RH were 55-60% and 70-80% in setter and hatcher section respectively. Candling test were carried at 1, 7 and 14 days of incubation period. Un fertile eggs were removed immediately, and eggs were not containing viable embryos were removed and broken to determine fertility status and if fertile the approximate day of embryonic mortality was noted.

2.3. Post-hatching processes
After incubation eggs Non-hatched eggs were counted to determine fertility (%) and hatchability (%) and broken to classify embryonic mortality as early (0-7 days), mid-term (8 to 14 days) and late (15 to 21 days; fully grown, with and piped eggs) mortality. The following formulas were used:
• Hatchability of fertile eggs (%): (number of hatched chicks / number of fertilized eggs set) x100.
• Early embryonic mortality: (number of dead embryos on days 0-7 of incubation / number of fertilized eggs) x100.
• Mid-term embryonic mortality: (number of dead embryos on 8-14 days of incubation / number of fertilized eggs) x100.
• Late embryo mortality: (number of dead embryos on 15-21 days of incubation / number of fertilized eggs) x100. [9].

2.4. Statistical Analysis
Statistical analysis of the experimental data was performed using CRD and Duncan (1955) multiple comparison test was applied to compare differences among treatments [10].

3. Results and Discussion
The results of experimental treatments on early embryonic mortality percentage were illustrated in table (1). The results showed that early embryonic mortality was significant decreased (P≤0.05) in G1 at first days of incubation day of incubation that spraying by alcohol and in control groups that
recorded (26.66% ) when compared with other groups. the results of second phase (G1) showed significantly decreasing (P≤0.05) in early embryonic mortality in groups treated with water (13.33% ) in comparison with the other treatments with other treatment groups.

Table 1. Effect of different solutions sprayed the local Iraqi eggs on early embryonic mortality at 1st week of incubation.

| Incubation period | Beginning of incubation period | After 7 day of incubation period | After 14 day of incubation period |
|-------------------|--------------------------------|----------------------------------|----------------------------------|
| Groups            | G1                             | G2                               | G3                               |
| Control           | 26.60 ±0.00 d                  | 26.60 ±0.00 d                    | 26.60 ±0.57 d                    |
| Water Spraying    | 40.00± 1.15 e                  | 13.33± 0.57 f                    | 0.00±0.00 H                      |
| Olive oil Spraying| 66.60±0.00 b                   | 100.00±0.00 a                    | 0.00±0.00 H                      |
| Albumin Spraying  | 66.60±0.57 b                   | 66.60 ±0.57 b                    | 20.00 ±0.57 E                    |
| Ethanol alcohol Spraying | 26.66±0.57 d | 26.66 ±0.57 d | 6.66 ±1.15 G |

Different small letters refers to significant differences at P≤0.05 in same column.

This changes might be attributed to the importance of water spraying on hatching eggs to decline the temperature inside the incubator that affected on embryonic development and growth because elevated incubator temperature could be caused raising egg's shell temperature leading to increase embryonic cells division subsequently causing embryonic defects during development causing to elevate early embryonic mortality [11] . On the other hand, the results of G3 in which eggs were be sprayed at the beginning of 3rd week of incubation (14 day) didn’t improved early embryonic mortality in eggs sprayed by water and olive oil that recorded (0.00%) while it increased significantly (P≤0.05) in control group GC that recorded (26.66%) when compared with other groups.

Table 2. Effect of spraying of local Iraqi eggs by different solutions on Middle embryonic mortality at 2nd week of incubation period

| Incubation period | Beginning of incubation period | After 7 day of incubation period | After 14 day of incubation period |
|-------------------|--------------------------------|----------------------------------|----------------------------------|
| Groups            | G1                             | G2                               | G3                               |
| Control           | 13.33 ±0.00 e                  | 13.33 ±0.57 e                   | 13.33 ±0.57 e                    |
| Water Spraying    | 0.00± 0.00 g                   | 33.33 ±1.15 d                   | 0.00±0.00 g                      |
| Olive oil Spraying| 0.00± 0.00 g                   | 0.00±0.00 g                     | 0.00±0.00 g                      |
| Albumin Spraying  | 66.60 ±0.57 a                  | 0.00±0.00 g                     | 53.33 ±0.00 b                    |
| Ethanol alcohol Spraying | 6.40 ±1.15 f   | 40.00 ±1.15 a                   | 13.33 ±0.57 e                    |

Different small letters refers to significant differences at P≤0.05 in same column.
All this significant improvement might be due to antioxidant role of olive oil because it contained tocopherol considered as natural active antioxidant, as well as, that oil contained phenolic compounds like hydroxytyrosol, vanillin, vanillic acid, tyrosol and caffeic acid that inhibited microbes action and stopped distribution, as well as, the oily matrix of olive oil could occlude egg shell’s pores which supported the cuticle layer and didn’t breakdown it [12 ; 13 ; 14] that will be positively reflected on embryonic growth by preventing any microbes invasion inside eggs subsequently supported embryonic development and declined embryonic mortality [15], and beneficial effect of olive oil was good source of protein, fat, calcium, copper, and cobalt and digestible protein, and mineral content, and high lignin content [16 ; 17]. which increased of blood in body embryo [18], and digestive enzyme increase that could be released into the intestine and help digestive tract to normal embryonic development[ 19] to improve embryonic development.

Table 3 explained effect of spraying of local Iraqi eggs by different solutions on Late embryonic mortality percentages at (15-21 days of incubation period). The results of G1 didn’t showed late embryonic mortality in eggs spraying by olive oil, while it recorded significantly increase (P≤0.05) eggs spraying by albumen and alcohol that recorded (20.00%) for each treatment. But G2 recoded significantly increase (P≤0.05) in late embryonic mortality in eggs spraying by albumen that recorded (66.60%) when compared with other groups. In G3, the results showed significantly increase (P≤0.05) in late embryonic mortality in control group that recorded (13.33%), while other continuous paragraph didn’t recorded any late embryonic mortality. These changes might be attributed to the effective role of olive oil for supplying the early developing embryo by all necessary requirements for growth during incubation, on other hand, the negative role of alcohol on embryonic growth might be due to dryness action of alcohol leading to break down the cuticle layer that coat whole egg leading to facilitate microbes penetration egg’s shell subsequently causing increased embryonic mortality in eggs spraying by ethanol alcohol, this agreement with [20] who reported that using any sanitizers could be caused changing and/or removing of cuticle coat that influencing on hatchability

| Incubation period | Beginning of incubation period | After 7 day of incubation period | After 14 day of incubation period |
|-------------------|--------------------------------|---------------------------------|---------------------------------|
| Groups            |                                |                                 |                                 |
| Control           | 13.33 ± 0.00                   | 13.33 ± 0.57                    | 13.33 ± 0.57                    |
| Water Spraying    | 6.25 ± 0.00                    | 20.00 ± 1.15                    | 0.00 ± 0.00                    |
| Olive oil Spraying| 0.00 ± 0.00                    | 0.00 ± 0.00                     | 0.00 ± 0.00                    |
| Albumin Spraying  | 20.00 ± 1.15                   | 66.60 ± 0.57                    | 0.00 ± 0.00                    |
| Ethanol alcohol Spraying | 20.00 ± 1.15 | 13.33 ± 0.00                  | 6.66 ± 0.00                    |

- Different small letters refers to significant differences at P≤0.05 in same column.

The results of different solutions spraying effects on hatchability of local Iraqi eggs were illustrated in Table 4 In G1, the hatchability increase significantly (P≤0.05) in control and eggs spraying by albumen that recorded (73.33 and 66.00%) respectively. While in G2, the hatchability in control group significantly increased (P≤0.05) when compared with that spraying by water that recorded (73.33% and 36.30%) respectively. But results of G3 showed significantly increase (P≤0.05) in hatchability percentage in eggs spraying with water and olive oil in comparison with other treated groups, that recorded (100.00%) for water and olive oil eggs groups. This significant changing in hatchability due percentage to results of embryonic survival during incubation period, because eggs
spraying by olive oil and/or water may increase embryo protection from external contaminations by making barrier between external and internal eggs environment that prevent bacteria penetration inside eggs that reflected positively by decreasing embryonic mortality percentage subsequently increased hatchability [21; 22].

### Table 4. Effect of spraying of local Iraqi eggs by different solutions on hatchability percentage.

| Incubation period | Beginning of incubation period | After 7 day of incubation period | After 14 day of incubation period |
|-------------------|--------------------------------|---------------------------------|----------------------------------|
| Groups            | G1                             | G2                              | G3                               |
| **Control**       | 73.33 ±0.00                    | b                               | 73.33 ±0.00                      |
| **Water Spraying**| 52.00±0.57                     | d                               | 100.00±0.00                      |
| **Olive oil Spraying**| 37.50±0.57                  | e                               | 100.00±0.00                      |
| **Albumin Spraying**| 66.60 ±0.57                  | g                               | 100.00±0.00                      |
| **Ethanol alcohol Spraying**| 0.00±0.00                      | g                               | 73.33±0.00                       |

- Different small letters refers to significant differences at P≤0.05 in same column.

### 4. Conclusions

It can be determined from current study that the spraying local eggs with olive oil and/or water could improve the hatchability percentage by decreasing embryonic mortality percentage during various incubation period that will enhance using local chicken eggs for at hatcheries.

### 5. Acknowledgments

Deepest thankfulness to the chairman of animal production department and to the dean of agriculture college / Karbala university for providing all requirements for current experiment.

### References

[1] Al-Rawi B A 1969 Effect of crossbreeding on productive and reproductive characters in Iraqi chicken . M.Sc. Thesis , College of Agric. Baghdad Univ. Iraq.

[2] Altaie SMS et al 2019 Evaluation the influence of dwarf gene on some fecundity features in dwarf hens. *Biological and cellular archives*, 19, 2.

[3] Ryan D et al 2002 Biotransformations of phenolic compounds in Olea europaea L. *Scientia Horticulturae*, 92:147-176.

[4] Abo Omar J M 2000 The effect of different levels of olive pulp on the feed intake and digestibility of broilers. *Bethlehem University Journal*, 19:9

[5] Afsari M et al 2013 Effects of phytase supplementation of low phosphorous diets included olive pulp and date pits on productive performance of laying hens, egg quality traits and some blood. Parameters Annual Review & *Research in Biology*;

[6] Botsoglou E N et al 2013 Olive leaves (Olea europaea L.) versus α-tocopheryl acetate as dietary supplements for enhancing the oxidative stability of eggs enriched with very-long-chain n-3 fatty acids. *J. Sci. Food Agric.* 93:8

[7] Khalil H A 2009 Productive and physiological response of Japanese quail embryos to light regime during incubation period. *Slovak J. Anim. Sci.*, 42:2

[8] Benavente-Garcia O et al 2000 Antioxidant activity of phenolics extracted from Olea europaea L. leaves. *Food Chemistry*

[9] Abuoghaba A A 2017 Impact of spraying incubated eggs submitted to high temperature with ascorbic acid on embryonic development, hatchability, and some physiological responses of
hatched chicks. *Canadian Journal of Animal Science*, 32:172-1

[10] SAS 2001 SAS User's Guide : statistics Version 6.12. SAS Institute , Inc., Car

[11] Kuo F et al 1997 UV irradiation of shell eggs: Effect on populations of aerobes, molds, and muculated Salmonella typhimurium. *J. Food Prot.*, 60:

[12] Gikas E et al 2007 Conformation of oleuropein, the major bioactive compound of Oleaeuropea. *J. Mol. Struct.: Theocem.* 821

[13] Silversides FG and Scott TA 2001 Effect of storage and layer age on quality of eggs from two lines of hens. *Poultry Sci.*, 80:

[14] Wenk C 2002 Herbs, botanicals and other related substances . WPSA – Bremen.

[15] Brake J and Sheldon BW 1990 Effect of a quaternary ammonium sanitizer for hatching eggs on their contamination, permeability, water loss and hatchability. *Poultry Sci.*

[16] Baylan MI et al 2018 The Effects of Using Garlic Extract for Quail Hatching Egg Disinfection on Hatching Results and Performance. *Brazilian Journal of Poultry Science*. v.20

[17] Molenaar R et al 2011 High eggshell temperatures during incubation decrease growth performance and increase the incidence of ascites in broiler chickens. *Poul. Sci.*, 90: 624–632. doi:10.3382/ps.2010-00970.

[18] Erener G et al 2009 The effects of olive leaf extract on performance, some blood parameters and cecal microflora of broilers. *The Scientific and Technological Research Council of Turkey, Agriculture, Forestry and Veterinary Research Group, AFVRG - Project No : 10708*

[19] Sayehban P I et al 2016 Effects of Different Levels of Two Types of Olive Pulp with or without Exogenous Enzyme Supplementation on Broiler Performance and Economic Parameters. *Brazilian Journal of Poultry Science*

[20] Yassein D M M et al 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. *Egyptian Poultry Science Journal*.

[21] Kafi LA 2014 A comparative study between olive oil and Nigella Sativa oil in treatment of hyperlipidemia induced in male albino mice. *The Iraqi Journal of Veterinary Medicine*,

[22] Yıldırım I et al 2003 The use of oregano (*Origanum vulgare L.*) essential oil as alternative hatching egg disinfectant versus formaldehyde fumigation in quails (Coturnix coturnix japonica) eggs. *Food Control*, 15: 169-172.