Research Article

Effect of spraying some plant extracts on hatching eggs of broiler chickens on the hatching rate and embryo mortality and the weight of hatched chicken

Salah Mahdi GATEA^{*1}, Haider Qassim BAQER¹, Salam Merza Suhail ALTAIE¹, Thamer Kareem ALJANABI¹, Mohammed Abdulkadhim HUSSAIN², Baha Abdel Hussein MUSA¹

¹Department of Animal Production, College of Agriculture, University of Kerbala, Kerbala, Iraq.

²Department of Animal Physiology, College of Veterinary Medicine, University of Kerbala, Kerbala,

Iraq.

^{*}Email: salah.katia@uokerbala.edu.iq

Abstract: This study was conducted to investigate the effect of spraying the hatching eggs of layer eggs (Lohman) with water extracts of pomegranate, clove, and anise on the hatching characteristics and the weight of the hatched chicks. The eggs were distributed randomly into five treatments, each containing 60 eggs per treatment as three replicates. The experimental treatments were divided into T1: control treatment, T2: spraving distilled water, T3: spraying aqueous clove extract at a level of 5 ml / 100 ml water, T4: spraying the aqueous pomegranate extract in 5 ml / 100 ml water, and T5: spraying the aqueous anise extract with 5 ml / 100 ml water. Then, the eggs were transferred to the incubator to start the hatching process for 21 days. The results indicated a significant improvement in the hatching percentage of fertilized eggs with a significant decrease in the number of deaths of total embryos from fertilized eggs of T2, T4, and T5 and a significant decrease in the percentage of live clicker chicks and deformed chicks in (T2, T4, and T5) during the total incubation period for all egg spraying treatments compared to T1. All spraying treatments had the highest significance in the relative weight of hatched chicks, with a highly significant improvement for all spraying treatments in the phenotypic characteristics of hatched chicks compared to control treatment T1.

Keywords: Fertility, Egg traits, Hatchability, Spraying, Chicks.

Citation: Gatea, S.M.; Baqer, H.Q.; Altaie, S.M.S.; Aljanabi, T.K.; Hussain, M.A. & MUSA, B.A.H. 2023. Effect of spraying some plant extracts on hatching eggs of broiler chickens on the hatching rate and embryo mortality and the weight of hatched chicken. Iranian Journal of Ichthyology (Special Issue 1): 280-285.

Introduction

Plant extracts have many compounds that contain wide and varied chemical and therapeutic properties. Therefore, they have been used in the treatment of many infections and bacterial, fungal, and other diseases. Among the most important of these plants distinguished by their antibacterial properties are mint leaves, anise powder, and pomegranate peels (Sacco et al. 1989; Mona 2011). Additionally, the use of some plant extracts led to the stimulation of the digestive system of animals and domestic birds, which improved the function of the organs, especially the liver. This led to an increase in digestive enzymes, which increase the utilization of food intake and provide the body's need for nutritional components (Jamroz & Kamel 2002).

One of the parts of the pomegranate fruit is its peel, which has many therapeutic properties, including the antioxidant action (Singh et al. 2002), the stimulating effect on the immune system (Gracious et al. 2001), and the antimicrobial effect because it contains flavonoids, glycosides, polyphenols, tannin, and alkaloids. The tannins such as tannin, which is about 25% in the peels, inhibit the growth of bacteria through its association with proteins and the formation of a complex with the cell wall that destroys the bacterial cell (Prashanth et al. 2001). The aqueous extract of pomegranate peels is known to inhibit the growth of pathological microorganisms in the gastrointestinal tract as well as support beneficial microorganisms, which to stimulates the improvement of the secretion of digestive enzymes. Therefore, it improves the efficiency of digestion to benefit from the food intake, and the food conversion factor (Saleh et al. 2017). Using pomegranate peel powder as a nutritional supplement at different levels improved broiler chickens' final body weight, feed intake, and feed conversion ratio (Saleh et al. 2018). Therefore, this study aimed to find out the effect of spraying the hatching eggs of broiler chickens with plant extracts of pomegranate peels, anise, and cloves on the hatching rate, the percentage of embryonic mortality, and the weight of the hatched chick at one day of age.

Materials and Methods

This study was conducted in the College of Agriculture, Karbala University incubator from 20/12/2022 until 11/1/2023, where broiler chicken eggs were obtained from the field of raising broiler chickens. Immediately after collecting the eggs, they were put in the refrigerator until the second day. The chicken was fed a production diet that contained 16.88% protein and energy (2726kg).

Preparation of aqueous extract of pomegranate, anise, and clove peel powder: We obtained pomegranate peels, anise, and cloves from the markets of the Karbala governorate and dried at room temperature for seven days with daily stirring, and the materials were ground separately by a small electric mill to be in the form of powder. Then the extract was prepared according to Hernandez et al. (1994). This was done by mixing 1 gram of dry powder for each substance with 2ml of distilled water using an electric mixer, then placing the solution in a water bath at a temperature of 60°C for one hour. The solution was left at room temperature for 24 hours, and filtered through some layers of sterile medical gauze to be ready for use in the experiment.

Spraying hatching eggs: 500 eggs with an average weight of 56.45±3 g were used in the current study, collected from broiler chickens. They were divided into five groups, with each group subdivided into 3 replicates: Treatments were those sprayed with distilled water, anise extract, pomegranate peel extract, clove extract, and control groups (T1 to T5, respectively).

Hatching process: Egg groups were transferred to the hatchery unit. Each replicate of 60 eggs (three per replicate) was placed in egg trays. After cleaning the egg surface with 100% ethanol, the eggs were sprayed with distilled water, anise extract, pomegranate peel extract, clove extract groups, and control one. Setter temperature and relative humidity were kept at 37.5°C and 55-60%, respectively, and 37.0°C and 70-80% in the hatcher, respectively. Eggs not containing viable embryos were removed and broken open to determine fertility status, and if fertile, the approximate day of embryonic death was noted.

Post-hatching processes: After incubation of eggs, non-hatched eggs were counted to determine fertility rate (%) and hatchability rates (%) and broken to classify embryo mortality as early (0-7) days, midterm (8 to 15 days), and late (16 to 21 days); fully grown, with and piped eggs) mortality. The following formulas were used to calculate parameters (Abuoghaba, 2017):

Hatchability of fertile eggs (%) = (number of hatched chicks / number of fertilized eggs set) x100. Early embryo mortality (%) = (number of dead embryos on days 0-7 of incubation /number of fertilized eggs) x100.

Mid-dead embryo mortality (%) = (number of dead embryos on days 8-15 of incubation/number of fertilized eggs) x 100.

Late dead embryo mortality (%) = (number of dead embryos on 16-21 days of incubation/number of fertilized eggs) x 100.

Statistical analysis: The present data were tested by

Table 1. The effect of spraying eggs of hatching w	vith some aqueous extracts on th	he percentage of embryonic deaths during the
embryonic development period.		

Treatment Traits	Death in the first week (0-7) Early embryonic mortality %	Death at second weeks (8-15) Intermediates embryonic mortality %	Death at third weeks (16-21) Late embryonic mortality %
T1	5.1	0.0	6.9
T2	2.6	0.0	5.2
Т3	2.5	0.0	5.1
T4	2.2	0.0	2.0
T5	2.9	0.0	5.0

T1: control treatment, T2: sprayed with distilled water, T3: sprayed with aqueous clove extract at a level of 5 ml / 100 ml water, T4: sprayed with aqueous pomegranate extract 5 ml / 100 ml water, and T5: sprayed with aqueous anise extract 5 ml / 100 ml water. * The different letters within one column indicate that there are significant differences between the totals at the probability level of 0.05.

Table 2. Effect of spraying hatching eggs of broiler chickens with some aqueous extracts on hatching. percentage ± standard error.

Treatment	Hatchability %
T1	89.66±0.102C
Τ2	89.64±0.352C
Τ3	91.61±0.587BC
T4	94.64±0.402AB
T5	92.54±0.502 BC
Significant	0.01

T1: control treatment, T2: sprayed with distilled water, T3: sprayed with aqueous clove extract at a level of 5 ml / 100 ml water, T4: sprayed with aqueous pomegranate extract 5 ml / 100 ml water, and T5: sprayed with aqueous anise extract 5 ml / 100 ml water. * The different letters within one column indicate that there are significant differences between the totals at the probability level of 0.05.

analyzing variance (SAS, 2001). Significant changes among groups were verified via Duncan's test at the level of 0.05.

Results and Discussion

There was a decrease in the percentage of periodic fetal mortality during the first week (0-7 days) of the embryo's life based on the fertilized eggs of treatments T2, T3, T4, and T5 compared to control one, while we did not find any deaths of embryos during the second week of the fetus's life (Table 1). A decrease in the percentage of fetal deaths in the third week of the fetus's life was found in the T4, while no differences were found between the other treatments, i.e. T2, T3, and T5 in the percentage of embryonic deaths in the third week. The highest value was found in T4 compared to T1, T2, T3, and T5 based on fertilized eggs, but no differences were found between T1, T2, T3, and T5 (Table 2). There was a higher hatching percentage in spray treatments with aqueous plant extracts, T3 and T5, compared to

control treatment, T1 and T2.

This improvement, in turn, improves the health of the embryos, which leads to an improvement in their growth and development and their ability to hatch. Many studies have proven that the antibacterial activity of some medicinal plants' components shows the same effects as antibiotics (Al-Juboory 2008). This reduces the bacterial load on the eggshell and thus reduces the chances of embryos being exposed to pathogens, and this explains the reason for the low percentage of total embryo mortality in egg spraying treatments in plant extracts. Therefore, early spraying with plant extracts of hatching eggs has improved the hatching characteristics associated with the vitality of the embryos by reducing the stress to which the embryo is exposed, especially in the last stages of hatching, resulting from the high temperature of the embryo and the hatched eggs during the process of turning the eggs when transferring from the incubator to the hatchery, which leads to the evolution of hatching traits (Al-Khafaji & AL-Jebory 2019; Gatea

Treatment	Chick weight at hatch (g)	
T1	38.22±0.702B	
T2	38.94±0.552B	
Т3	39.11±0.1q2B	
T4	42.66±0.862A	
T5	39.35±0.562B	
Significant	0.01	

Table 3. Effect of spraying broiler eggs with some aqueous extracts on the weight of the hatched chick (grams) at the age of one day \pm standard error.

T1: control treatment, T2: sprayed with distilled water, T3: sprayed with aqueous clove extract at a level of 5 ml / 100 ml water, T4: sprayed with aqueous pomegranate extract 5 ml / 100 ml water, and T5: sprayed with aqueous anise extract 5 ml / 100 ml water. * The different letters within one column indicate that there are significant differences between the totals at the probability level of 0.05.

et al. 2019). Also, the use of spraying plant extracts improves body weight, as it will activate the metabolic processes in the digestive system, which facilitate digestion and absorption, and its positive results are reflected in the weight of the chicks (Abdel Rahman et al. 2013). Therefore, pomegranate peel extract is a natural antibiotic, as it is effective against pathogenic bacteria and harmful microorganisms. Thus, the number of harmful microorganisms decreases compared to microorganisms and beneficial bacteria that improve the efficiency of digestion and facilitate the absorption process, as the tannins stimulate some enzymes secreted in the stomach to facilitate the digestion process (Ghazaleh et al. 2013).

In this context, Hamadani et al. (2013) confirmed that embryos that are fed in early embryonic stages will have increased activity of the gastrointestinal tract and enzymes that digest nutrients. Or it may be because the process of spraying eggs with these plant extracts is their action as biological antioxidants that interact with free radicals or their primary metabolites by pathological bacteria and transform them into less reactive molecules and prevent or delay the oxidation of biological molecules (Krishan & Narang 2014) of pomegranate peels contain antioxidants (Ghazaleh et al. 2013). They act as radical scavengers, inhibit lipid peroxidation, and protect animal cells from the oxidative damage they cause (Sahin et al. 2013) and this provides early protection for developing embryos inside the egg on

the one hand and acts as a barrier to prevent germs from penetrating the structural structure of the egg on the other hand. The results of this study are similar to those of Baylan et al. (2018), who noticed that spraying hatching eggs with garlic extract, had a significant role in reducing the percentage of embryonic mortality, increasing the hatching rate, and improving the weight of the hatched chicks. Olivera et al. (2021a) also found that spraying hatching eggs with clove extract significantly improved the percentage of early, medium, and late embryonic deaths during the hatching period and a significant increase in the hatching rate, as clove oil, in addition to its role as a sterilizer for the surface of the egg shell, is absorbed by the pores of the shell; the egg then improves embryonic growth and improves the hatching rate. Olivera et al. (2021b) did not find a significant improvement in the decrease in the percentage of embryonic deaths and the percentage of hatching when the hatching eggs were sprayed with anise extract with other extracts.

The highest significant increase ($P \ge 0.01$) in the average weight of the day-old hatching chick for T4 compared to T1, T2, T3, and T5, while we did not find any significant differences between T1, T2, T3, and T5, while higher results were found in spraying treatments with plant extracts water, T3 and T5 compared with the control of T1 and T2 (Table 3). The improvement in chick weight resulting from spraying the eggs with aqueous plant extracts may be due to the role of the extracts in improving the

efficiency of subsequent absorption of some of these extracts by the embryos, which play an important role in nutrition (Pramila 2012). The minerals available in plant extracts have a major role in skeletal growth and cellular metabolism by increasing the representation of mineral elements during the beginning of embryonic development, and this explains the significant increase in chick weight for spraying treatments with aqueous plant extract. The reason for the decrease in the percentage of deaths in the treatments of spraying eggs with an aqueous extract of pomegranate peels may be attributed to the fact that they contain tannins, which have an effective role in preventing infection through the destruction of the cell membrane of pathogenic bacteria, as well as blocking the effective sites of some enzymes inside the living cells of bacteria that are necessary for their growth and activity as well. This results in the deposition of proteins present in the cell membrane or within the cell when they permeate through the membrane, and the formation of hydrogen bonds between the free phenolic hydroxyl groups, which reflects positively on the vitality and health of the chicks, and a decrease in the mortality rate (Ghazaleh et al. 2013).

Reddy et al. (2007) indicated that pomegranate peel powder contains flavonoids and phenols that act as anti-pathological microorganisms as a result of the synergistic action of these compounds that lead to the inhibitory action of pathogenic bacteria. The use of pomegranate peel extract led to a significant increase in the length of the villi, the depth of the crypts, and the ratio of villus height to the depth of the crypts in the duodenum, jejunum, and ileum over the control treatment. Bacteria, which contribute to supporting the microbial balance of the intestinal flora, and increasing the beneficial microorganisms in the gastrointestinal tract, support the digestive system with some useful elements, and the production of some organic acids, which are a source of villiforming cells (Mourao 2013). Pomegranate peels are rich in tannins, which are considered an antibiotic for pathological microorganisms, and increase gastrointestinal secretions, especially liver and bile secretions, to increase the activity of cells and their

divisions in the small intestine, and thus increase the length of the villi (Ghazanfari et al. 2014). Olivera et al. (2021) found that spraying the hatching eggs with clove extract improved mathematically the weight of the one-day-old hatching chick compared to the control treatment.

Conclusion

It can be concluded from the current study that spraying water or natural extracts on broiler eggs could improve the hatchability percentage by decreasing the embryonic mortality percentage during various incubation periods, which will enhance the use of chicken eggs for hatcheries.

References

- Abdel Rahman, H.A.; Shawky, S.M.; Ouda, H.; Nafeaa, A.A. & Orabi, S.H. 2013. Effect of two probiotics and bioflavonoids supplementation to the broilers diet and drinking water on the growth performance and hepatic antioxidant parameters. Global Veterinarian 10(6): 734-741.
- Al-Juboory, M.S.R. 2008. Effect of some plant extracts on some pathogenic bacteria. Diala Journal 69: 413-419.
- Al-Khafaji, F.R. & AL-Jebory, H.H. 2019. Effect of injection in hatching eggs with different concentrations of nano-silver at 17.5 days age in some hatching traits and blood parameters for broiler chickens (ROSS 308). Plant Archives 19: 1234-1238.
- 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: 172-182.
- Baylan, M.; Akpınar, G.C.; Canogullari S.D. & Ayasan, T. 2018. The effects of using garlic extract for quail hatching egg disinfection on hatching results and performance. Brazilian Journal of Poultry Science 20(2): 343-350.
- Gatea, S.M.; Altaie, S.M.S.; Khafaji, S.S.; ALjanabi,
 T.K.; Shatti, D.H. & Hussain, M.A. 2019. Influence of spraying different solutions at different incubation periods on hatchability parameters of local Iraqi eggs.
 IOP Conference Series: Earth and Environmental Science 388(2019): 1-7.

Ghazaleh, M.; Sharifzadeh, M.; Hassanzadeh, G.;

Khanavi, M. & Hajimahmoodi, M. 2013. Anti-Ulcerogenic activity of the pomegranate peel (*Punica granatum*) methanol extract. Food and Nutrition Sciences 4(10): 6 -12.

- Ghazanfari, S.; Moradi M.A.; & Bardzardi, M.M. 2014. Intestinal morphology and microbiology of broiler chicken fed diets containing myrtle (*Myrtus communis*) essential oil supplementation. Iranian Journal of Applied Animal Science 4(3): 549-554.
- Gracious, R.R.; Selvasubramanian, S. & Jayasundar, S. 2001. Immunomodulatory activity of Punica granatum in rabbit a preliminary study. Journal of Ethnopharmacology 78: 85-89.
- Jamroz, D. & Kamel, C. 2002. Plant extracts enhance broiler performance. Journal of Animal Science 80(Suppl): 41-47.
- Krishan, G. & Narang, A. 2014. Use of essential oils in poultry nutrition; A new approach. Journal of Advanced Veterinary and Animal Research 1(4): 156-162.
- Mona, R.M. 2011. Effect of egg disinfection on embryonic development, hatchability characteristics and post-hatch productive performance in local chickens. Ph.D. Thesis, Faculty of Agriculture (Saba-Bacha) Animal and Fish Production Department, Alex University Egypt.
- Mourao, J.; Pereira, L.; Ermelinda, L.; Pinheiro, V.; Carla,
 D.; Eliana, F.; Vânia, P.; Saavedra S. & Maria, J.
 2013. Effects of olive leaf on gut microflora of broiler poultry. In: FABE 2013 International Conference on Food and Biosystems Engineering. Skiathos Island, Greece.
- Oliveria, G.S.; Nascimento, S.T.; Santos, V.M. & Dallago, B.S. 2021. Spraying hatching eggs with clove essential oil does not compromise the quality of embryos and one–day–old chicks or broiler performance. Animals 11: 2045.
- Oliveira, G.S.; Santos, V.M.; Nascimento, S.T. & Rodrigues, J.C. 2020b. Alternative sanitizers to paraformaldehyde for incubation of fertile eggs. Poultry Science 99: 2001-2006.
- Oliveira, G.S.; Sheila, T.; Nascimento, T.; Vinícius, M.; dos Santos Y. & Silva. M.G. 2020a. Clove essential oil in the sanitation of fertile eggs. Poultry Science 99: 5509-5516.

Pramila, D. 2012. Phytochemical analysis and

antimicrobial potential of methanolic leaf extract of peppermint (*Mentha piperita*; Lamiaceae). Journal Med Plants Restivia 6(2): 331-335.

- Prashanth, D.; Ash, M.K. & Amit, A. 2001. Antibacterial activity of *Punica granatum*. Fitoterapia 72(2): 171.
- Reddy, M.K.; Gupta, S.K.; Jacob, M.R.; Khan S.I. & Ferreira, D. 2007. Antioxidant antimalarial and . antimicrobial activities of tannin-rich fractions ellagitannins and phenolic acids from *Punica* granatum L. Planta Medica 73: 461-467.
- SAS. 2001. SAS User's Guide: Statistics Version 6.12. SAS Institute, Inc., Cary, NC.
- Sacco, R.E.; Renner, P.A.; Nestor, K.E.; Saif, Y.M. & Dearth, R.N. 1989. Effect of hatching egg sanitizers on embryonic survival and hatchability of turkey eggs from different lines on eggshell bacterial populations. Poultry Science 68:1179-1184.
- Sahin, K.; Orhan, C.; Smith M.M. & Sahin, N. 2013. Molecular targets of dietary phytochemicals for the alleviation of heat stress in poultry. World's Poultry Science Journal 69(2): 113-124.
- Saleh, H.; Golian, A.; Kermanshahi, H. & Mirakzehi, M.T. 2018. Antioxidant status and thigh meat quality of broiler chickens fed the diet supplemented with atocopherol, pomegranate pomace, and pomegranate pomace extract. Italian Journal of Animal Science 17(2): 386-395.
- Singh, R.P.; Chidambara, M. & Jayaprakasha, G.K. 2002. Studies on antioxidant activity of pomegranate (*Punica granatum*) peel and seeds extracts using in vitro models. Journal of Agricultural and Food Chemistry 50: 81-86.