EFFECT OF GARLIC OIL EXTRACT ON BLOOD PARAMETERS IN CHICKENS INFECTED WITH ECTOPARASITES

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ABSTRACT

The study was conducted from September 2018 to April 2019, A total of 350 chicken samples were examined from different areas of the Basrah province for the diagnosis of ectoparasites and 154 chickens were infected with ectoparasites. Four species of lice were isolated, which were *Menacanthus cornutus*, *Menacanthus stramineus*, *Menacanthus pallidulus*, *Menapon gallinae* and one species of mite which was *Dermanyssus gallinae*. The experimental study was carried out to eliminate the ectoparasites where 24 chickens were purchased and placed in the field of the faculty of agriculture. The chickens were divided into 3 groups. First group was the control group and the second group was infected with ectoparasites and the third group was infected with ectoparasites which was sprayed with garlic extract at a concentration of 10%. Ten days later, the blood parameters were measured for the three groups where the blood parameters of the group treated with garlic extract were close to the blood parameters of the control groupand show significant different (P<0.05).

INTRODUCTION

In developing countries, animal production in general and chicken production in particular play an essential socio-economic role. Poultry production is one area of livestock production with significant contribution to human food production (1).

Poultry is an economic and effective source of animal protein within the shortest possible time, playing a vital role in narrowing down the animal protein supply gap. Poultry can easily be infected with several types of bacterial, viral, fungal and parasitic Diseases, Ectoparasites infestation are of great significance. Ectoparasites live on under the feathers or penetrate within the skin or even in to the air sacs and some lives under the feathers (2). Lice is the common name for members of the order Phthiraptera which contains nearly 5,000 species of wingless insect. The impacts of louse parasitism on birds are often severe, including retarded growth, low egg production and susceptibility to other infections (3). Lice is one of the most important blood sucking ectoparasites of poultry and it is causing irritation to decreased body weight, which adversely affects the economical production of poultry (4).

Poultry Red Mite (PRM), *Dermanyssus gallinae* is a blood-feeding ectoparasites with a severe impact on laying hens (5). It can survive in poultry equipments for long periods without host and is easily spread directly by the host or indirectly by multiple fomites (6).

A number of reproductive inhibitors and repellents of arthropods used to be derived from certain plants. These extracts affect the feeding behavior and life cycle of parasites (7). As pesticides of synthetic origin may have a negative impact on the environment, and pest resistance to toxic chemicals which can be developed after repeated applications, the use of natural substances has become more popular (8). Garlic has been widely studied for its immune boosting properties. Many studies have shown that the compound allicin, found in fresh garlic has antibiotic, antiparasitic and antifungal properties (9). The compound is a colorless liquid with a distinctly pungent odor that shows anti-bacterial and anti-fungal characteristics. Allicin in garlic imparts defense reaction against attacks by insect pests (10). Therefore, The aim of this study was to evaluate effect of garlic oil extract on ectoparasites on chickens.

MATERIALS AND METHODS

The present study is conducted from September 2018 to April 2019, a total of 350 samples of chicken (*Gallus gallus domesticus*) were examined from different areas of the Basrah province to investigate the existence of ectoparasites.

Ectoparasites collection:

Out of 350 chickens examined, 154 chickens were infected with ectoparasites. The ectoparasites were isolated randomly from each chicken by using eye lenses 4X and fine forceps with good light source from vent, head, neck, breast, back, comb, and wing. Any suspected ectoparasites or substance are kept in small Petri-dish contains 70% alcohol and collected for further examination.

Ectoparasites diagnosis:

Glass slides of ectoparasites were prepared according to the method of (11). The samples were placed in 70 % alcohol and then transferred to 10% NaOH for 24 hours. The samples were washed with distilled water for 15 minutes then transferred to ascending grades of alcohols starting from 50%, 70%, 90% and absolute alcohol. In each grade of alcohol the specimen was kept for 15 minutes. The samples were transferred to the Xylole solution for 2 minutes then ectoparasites were placed in the middle of glass slide and covered with a drop of Canada balsam and covered with the lid of the slide. The glass slides were placed above the hot plate for 3 minutes and at temperatures 40 °C. Glass slides were examined under a dissecting microscope and the ectoparasites were classified Depending on: (12), (13) and (14).

Extraction Garlic oil:

Dried *Allium sativum* was obtained from the market of Basrah city, then grind with an electric mill and then stored in glass bottles. 70 gm of dried garlic was placed in paper container (thimble) in the extraction apparatus (succulet) using 500 ml of ethanol solvent for 6 hours. The solution was then placed in rotary evaporate the solvent and leave the remaining material at room temperature to dry and then the garlic oil was put in screw tube (15).

Experience design:

The experiment was carried out in the animal field of the Faculty of Agriculture. Twenty four chickens (*Gallus gallus domesticus*) were purchased and placed in the field and all chickens were two week of age. The blood and faeces of the chickens were examined before the experiment to make sure that they have no parasites. The chickens were divided into three groups, Each group consisting of 8 chickens: The first group was control group which was free from ectoparasites and the second group

was infected with ectoparasites and the third group was infected with ectoparasites which was sprayed with garlic extract at a concentration of 10%.

The concentration of garlic oil that used in this experiment was prepared by adding 90 ml of Diomethyl Sulfoxide to 10 ml of garlic oil. Third group of chicken infected with ectoparasites was sprayed with 10% of garlic oil every five days, After 10 days of processing. Blood samples were collected aseptically with sterile syringe from the jugular vein of all groups of chickens and were placed in a vacuum tube (EDTA K3) to evaluate the hematological parameters.

Hematological Parameters.

Hemoglobin concentration (Hb) (g/dl).

The concentration of Hb was measured by using Sahli apparatus technique according to (16)

Packed cell volume (PCV) (%).

Packed cell volume was measured by use The microhematocrit method according to (16).

Red blood cells count (RBC) (cell/mm³).

Red blood cells were counted using hematocymeter technique according to (17).

White blood cell count (Cell/mm3).

Total leukocyte cells were counted using hemocytometer technique according to (16).

Statistical analysis

Statistical analysis is performed by using statistical analysis software (Statistical Package for the Social Science) (SPSS) version 22. Therefore, the means, standard error (Std. error), and significant variances performed with One-way ANOVA by using Least significant difference (LSD) test at (P<0.05).

RESULTS

Identification of chickens ectoparasites

In this study, the results of the microscopic examination of chicken lice shows presence of four species of lice, they are:

Menacanthus cornutus, Menacanthus stramineus, Menacanthus pallidulus, Menapon gallinae and one species of mite which is Dermanyssus gallinae (Figure 1,2,3,4 and 5).

(Table. 1) shows the infestation rate for *Menacanthus cornutus*, *Menacanthus stramineus*, *Menapon gallinae ,Menacanthus pallidulus* and *Dermanyssus gallinae* are 41.7 %, 32.4 % ,13.9 % ,0.8% and 11.2 % respectively.



Figure 1: A.Male of *Menacanthus cornutus* 4x B.Female of *Menacanthus cornutus* 4x



Figure 2: Female of *Menacanthus stramineus* 4x





Figure 3: A.Female of *Menacanthus pallidulus* 4X B. Male of *Menacanthus pallidulus*4X

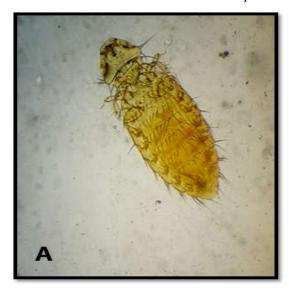




Figure 4: A.Male of *Menapon gallinae* 4x B.Female of *Menapon gallinae* 4x

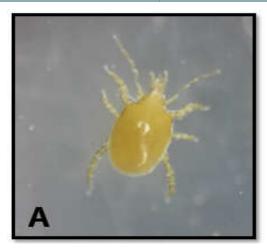




Figure 5: A.Dorsal surface of *Dermanyssusgallinae*10x B.Ventral surface of *Dermanyssus gallinae*10x

Table.1Infestation rate of ectoparasites species that collected from infected chickens.

Species	NO. of ectoparasites	Percentage %
Menacanthus cornutus	283	41.7%
Menacanthus stramineus	220	32.4%
Menapon gallinae	94	13.9%
Menacanthus pallidulus	6	0.8%
Dermanyssus gallinae	76	11.2%
Total	679	

The effect of garlic oil on ectoparasites

The results of the blood measurement revealed that the treatment group showed clear elevation in blood parameters as compared with the infestation group and close to the results of blood parameters of the control group. And it has been observed that six chickens of treated group has become completely free from ectoparasites (Table 2).

Table .2 The blood parameters of experimental groups after treated with 10% garlic oil .

Blood	Chickens groups									
Parameters	Control group			Infestation group			Treatment group			
	Value	Rate	*Mean	Value	Rate	*Mean	Value	Rate	*Mean	
			St.D			St.D			St.D	
HB(gm/d L)	9.8-	11	3.35	7.3-	8.8	2.5	9.2-	10.3	3.18	
	12.2		±0.24a	10.2		±0.24b	11.4		±0.25a	
PCV(%)	30-36	33.5	21.4	24-30	25.1	30.17	28-35	32.4	22.5	
			±1.92a			±4.26b			±1.29a	
RBC	3.1-	3.4	19.98	2.2-	2.5	8.8	2.9-	3.2	10.3	
(c/mm)X10*6	3.7		±0.94a	2.9		±0.51b	3.6		±0.64a	
WBC	19.4-	21.4	33.4	26-39	30.1	25.3	21.5-	22.5	32.4	
(c/mm)X10*3	25		±1.92a			±2.05b	25.7		±2.39a	

^{*}Means with different letters in each row are significantly different from each other at the 5% level (P<0.05).

DISCUSSION

Ectoparasites play critical roles in veterinary medicine and remain a persistent problem in the modern world. For many years, A number of pesticides, including organophosphates, organochlorides and synthetic pyrethroids, has given an efficient control of these parasites; but the extensive use of these drugs has led to the development of resistance in many target species (18). The results of the current study recorded four species of lice: *Menacanthus cornutus, Menacanthus stramineus, Menacanthus pallidulus* and *Menapon gallinae* in addition to one species of mite which is *Dermanyssus gallinae*.

On the other side, the species *Menacanthus cornutus* recorded the highest rate which was 41.7 %. In Iraq this species were first recorded by (19) and also detected in Basrah by (20) and (21) atpercentage rate 26.9% and 33.2% respectively while (22) recorded the same species in Al-Diwaniya with a prevalence rate 10.8 %. However the prevalence rate of infestation with *Menacanthus stramineus* was 32.4 % and it is considered a very common species in poultry. In Iraq, it was first isolated by (23) and

it has been detectedin Basrah with a rate 26.4% by (21) while (24) recorded it with a rate 66% in Al-Diwaniya. In our study, the species *Menapon gallinae*was also recorded in a percentage rate 13.9%, The first one which isolated it in Iraq was (23), While (21) recorded the same species with a rate 12% in Basrahand (22) recorded it in Al-Diwaniya with a rate 6.25%. Similar to the current study this species was recorded by (25) with a rate 7.5% in Spain while both (26 and 27) recorded the same species with a rate 88% and 8.1% respectively. Although the *Menacanthus pallidulus* was recorded in the current study at a percentage rate 0.8%. This species has a limited spread and recorded for the first time in Iraq by (21) with a rate 28.4% and has been recorded globally by (28) with infestation rate 13.7%. Our results also revealed that *Dermanyssus gallinae*was infested chicken a percentage rate 11.2%. Furthermore (29) was recorded the same species at a rate 13.3% in Kenya. These variations could be attributed to the season, time, and the study location and environmental factors favor their propagation and life cycle progress of the diverse ectoparasites species.

Many researchers have investigated the effects of essential oils, plant extracts and oriental medicinal plant extracts, against ectoparasites. They can be used as killing agents or repellents. Garlic oil was used in this study and the result showed great improvements in chicken treated with garlic oil as compared to untreated chickens in blood parameters and chicken behavior and this due to the ability of garlic oil to reduce ectoparasites. There are several studies where garlic oil has been used to eliminate ectoparasites such as (30) and (31). As well as (32) reported that essential oils of *Allium sativum* have neurotoxic effect on insects, causing rapid paralysis and killing effects

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تأثير مستخلص زيت الثوم ضد الطفيليات الخارجية التي تصيب الدجاج في محافظة البصرة

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الخلاصة

أجريت الدراسة الحالية للفترة من ايلول ٢٠١٨ إلى نيسان ٢٠١٩ ، وتم جمع ٣٥٠ عينة دجاج من مناطق مختلفة من محافظة البصرة لتشخيص الطفيليات الخارجية للدجاج. حيث كانت ١٥٤ عينه دجاجه مصابه بالطفيليات الخارجية الدجاج المصاب، وهي Menacanthus وهي Menapon gallinae، الخارجية الخارجية الواع من الدجاج المصاب، وهي Menapon gallinae، Menacanthus pallidulus، Menacanthus stramineus ونوع واحد من الحلم Dermanyssus gallinae، وتم أجراء دراسة تجريبية للقضاء على الطفيليات الخارجية حيث تم شراء ٢٤ دجاجة ووضعها في حقل دواجن تابع لكلية الزراعة في جامعة البصرة. تم تقسيم الدجاج إلى ٣ مجموعات. المجموعة الأولى كانت مجموعة السيطرة وكانت خاليه من الطفيليات الخارجية التي تم رشها بمستخلص كانت مصابة بالطفيليات الخارجية والمجموعة الثالثة كانت مصابة بالطفيليات الخارجية التي تم رشها بمستخلص الثوم بتركيز ١٠٪. بعد عشرة أيام تم قياس المعايير الدموية للمجموعات الثلاث حيث كانت المعايير الدموية المجموعة التي تعامل مع مستخلص الثوم قريبة من المعايير الدموية من مجموعة السيطرة وتظهر بمستوى احتمالية (٥٠٥٥) (٩).

REFERENCES

- **1- Thornton, P. K. (2010).** Livestock production: recent trends, future prospects. *Philosophical Transactions of the Royal Society B: Biological Sciences*, *365*(1554), 2853-2867.
- 2- Nadeem, M., Khan, M. N., Iqbal, Z., Sajid, M. S., Arshad, M., & Yaseen, M. (2007). Determinants influencing prevalence of louse infestations on layers of district Faisalabad (Pakistan). *British poultry science*, 48(5), 546-550.
- **3- Byford, R. L., & Maurice, E. C. (2007).** Biology of arthropods. *Flynn's Parasites of Laboratory Animals*, 51-68.
- **4- Durden, L. A. (2019).** Lice (Phthiraptera). In *Medical and veterinary entomology* (pp. 79-106). Academic Press.

- 5- Lesna, I., Sabelis, M. W., Van Niekerk, T. G., & Komdeur, J. (2012). Laboratory tests for controlling poultry red mites (Dermanyssus gallinae) with predatory mites in small 'laying hen' cages. *Experimental and Applied Acarology*, 58(4), 371-383.
- 6- Gunnarsson, E. (2017). Poultry red mites in Swedish laying hen flocks.
- 7- Smith, C. M. (2005). Plant resistance to arthropods: molecular and conventional approaches. Springer Science & Business Media.
- **8- Rattan, R. S. (2010).** Mechanism of action of insecticidal secondary metabolites of plant origin. *Crop protection*, *29*(9), 913-920.
- **9- Lee, J. Y., & Gao, Y. (2012).** Review of the application of garlic, Allium sativum, in aquaculture. *Journal of the World Aquaculture Society*, 43(4), 447-458.
- 10- Debra, K. R., & Misheck, D. (2014). Onion (Allium cepa) and garlic (Allium sativum) as pest control intercrops in cabbage based intercrop systems in Zimbabwe. IOSR Journal of Agriculture and Veterinary Science, 7(2), 13-7.
- 11- Price,R.D. ,Hellenthal, R.A. , Palma, R.L. , Johnson, K.P. and Clayton, D.H.(2003) the chewing lice : world checklist and biological overview. Illinois natural history survey species puplication 24 , x+501 pp.
- **12- Mani, M. S. (1974).** Modern classification of insects. *Modern classification of insects*.
- **13- Soulsby, E.J.L.(1982).** Helminthes, Arthropods and Protozoa of Domestic Animal. 7th ed. Balliere, Tindall& Cassell, London. P. 973.
- 14- Wall, R., & Shearer, D. (1997). Veterinary entomology: Arthropod ectoparasites of veterinary importance. Springer Science & Business Media.
- **15- Alam, P. (2013).** Densitometric HPTLC analysis of 8-gingerol in Zingiber officinale extract and ginger-containing dietary supplements, teas and commercial creams. Asian Pacific journal of tropical biomedicine, 3(8), 634-638.
- **16- Coles, E.H. (1986).** Veterinary Clinical pathology .4thedition. W.B.Saunder Company. Pp: 12-20.
- 17- Sood, R. (1996). Haematology for students and practioners, Jaypee Brothers Medical Publishers (P) Ltd., New Delhi.

- **18- McNair CM. (2015).** Ectoparasites of medical and veterinary importance: drug resistance and the need for alternative control methods. J. Pharmacol.; 67(3):351-63. doi: 10.1111 /jphp .123 68. Epub 2015 Feb 3.
- 19- Khalaf, K. T. (1959). A collection of insects from Iraq. Iraq Nat Hist Mus, 17, 17-26.)
- **20- Habeeb ,M.A. (2000).** Check listof Mallophaga of Basrah Province, Basrah J. Science,B., 18(1):55 60.
- **21- Kareem ,D.K.(2006).** Systematic study of sucking and chewing lice on some vertebrates with epidermiology of head lice in Basrah province, M.sc.Science, Basrah Univ.
- **22- Al-Shabani, M.S.A. (2013).** Epidemiological and diagnostic surveillance of the species of lice of some birds in the city of Diwaniya. Master Thesis, Faculty of Education, University of Qadisiyah: 011 pages.
- **23- Al-Hubaity, I. A.** (1976). Studies on the parasites of fowl Gallus gallus domesticus in Mosul district, Iraq (Doctoral dissertation, M. Sc. Thesis, University of Mosul).
- **24- Al-Jubouri, S.A.A. 2010.** endo and ectoparasitic infections in domestic fowl *Gallus gallusdomestcus* (Linnaeus, 1758). In: Al-Diwaniya city. M.Sc. Thesis, College of Education, University of Al- Qadisiya, Iraq (in Arabic).
- **25- Clavete, C., Estrada, R., Lucientes, J. and Estrada, A. (2003).** Ectoparasites ticks and chewing lice of red legged partridge, Alecorisrufa, in spain.Med. Vet. Entomol., 17(1): 33 37.
- **26- Gomez,S. Y. M. and Montano, J. A. B. (2007).** Parasitos en aves domesticas (Gallus domesticus) en el Noroccidente de Colombia. Vet. Zootec., 1(2): 43-51.
- 27- Bala, A.Y.; Anka, S.A.; Waziri, A. and Shehu, H.(2011). Preliminary Survey of Ectoparasites Infesting Chickens (*Gallus domesticus*) in Four Areas of Sokoto Metropolis. Nigerian J. of Basic and Applied Science, 19(2):173-180.
- **28- Manuel, M.F. (1981).** The ectoparasites (lice and mites) occurring on domestic chicken in Philippines . Philip. J. Vet. Med. ,20(1): 87–117.
- 29- Mungube, E. O., Bauni, S. M., Tenhagen, B. A., Wamae, L. W., Nzioka, S. M., Muhammed, L., &Nginyi, J. M. (2008). Prevalence of parasites of

- the local scavenging chickens in a selected semi-arid zone of Eastern Kenya. *Tropical Animal Health and Production*, 40(2), 101-109.
- 30- Birrenkott, G. P., Brockenfelt, G. E., Greer, J. A., & Owens, M. D. (2000). Topical application of garlic reduces northern fowl mite infestation in laying hens. *Poultry Science*, 79(11), 1575-1577.
- **31- Mägi, E., Järvis, T., & Miller, I. (2006).** Effects of different plant products against pig mange mites. *ActaVeterinaria Brno*, 75(2), 283-287.
- **32- Chaubey, M. K. (2017).** Study of insecticidal properties of garlic, Allium sativum (Alliaceae) and Bel, Aegle marmelos (Rutaceae) essential oils against Sitophilus zeamais L.(Coleoptera: Curculionidae). J. Entomol, 14, 191-198.