Clinical approaches to identify *Trichomonas gallinae* in local pigeons (Columba livia domestica) in Al-Qadisiyah Province, Iraq

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Abstract

Trichomonas gallinae is a unicellular protozoan parasite that is responsible for causing trichomonosis. Trichomonosis is a globally prevalent illness that has lately emerged as a pandemic danger to several species of wild birds. The purpose of this study is to look at the frequency of Trichomonas gallinae in the Iraqi province of Al-Diwaniya. A total of 150 oral swab samples were taken from birds and analysed for T. gallinae infection using clinical swabs and direct smear; the direct swab indicated that 98 (65.33%) were positive. Adult pigeons had a greater infection rate (76%), whereas juvenile pigeons (44%) demonstrated significant variance (P0.05) in overall prevalence. When the infection rate was compared by sex, it was found to be 64.10% in male and 66.66% in females. The highest infection rate was recorded in February and March (68%), (72%), respectively, while the lowest infection rate was reported in November and December (23%), (28%), respectively, with significant (P0.05) variations detected in the frequency of infections according to months. In conclusions, these findings emphasize the essential requirement for undertaking rigorous preventative measures to effectively reduce avian trichomoniasis.

Keywords: Trichomoniasis, Pigeon, direct smear, age and sex.

الاساليب السريرية للتعرف على *Trichomonas gallinae* في الحمام المحلي (Columba livia domestica) في محافظة القادسية ، العراق

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الخلاصة

Trichomonas *gallinae* هو طفيلي وحيد الخلية مسؤول عن التسبب في داء المشعرات . داء المشعرات هو مرض منتشر عالميا وقد ظهر مؤخرا باعتباره خطرا وبائيا على عدة انواع من الطيور البرية . الغرض من هذه الدراسة هو دراسة مدى انتشار طفيلي *Trichomonas gallinae* في محافظة الديوانية. تم اخذ 150 عينة فموية من الطيور وتحليلها بحثا عن العدوى باستخدام المسحات السريرية والمسحة المباشرة واظهرت المسحة المباشرة، واظهرت المسحة المباشرة ان 98 (65.33 %) كانت ايجابية . كان لدى الحمام البالغ معدل اصابة اكبر(76 %) ، في حين اظهر الحمام اليافع (44%) تباينا كبيرا (P0.05) في معدل الانتشار الجمالي. وعند مقارنة نسبة الاصابة حسب الجنس ، فقد بلغت 64.10% عند الذكور و 66.66% عند الناث . تم تسجيل اعلى معدل اصابة في فبراير ومارس (68%)،(72%) على التوالي ، في حين تم تسجيل ادنى اصابة في شهري نوفمبر وديسمبر (23%)،(28%) على التوالي ، وبنسبة كبيرة (P0.1) التباينات المكتشفة في تكرار الاصابة باختلاف

الاشهر .في الاستنتاجات ،توكد هذه النتائج على الحاجة الاساسية لاتخاذ تدابير وقائية صارمة للحد بشكل فعال من داء المشعرات في الطيور .

كلمات مفتاحية : داء المشعرات ، حمام ،المسحة المباشرة ، العمر ، الجنس .

introduction

Avian trichomoniasis is a potentially lethal parasite illness mostly caused by the pathogen *Trichomonas gallinae*, which infiltrates birds via the upper respiratory system. This parasite exerts a worldwide influence on a diverse array of avian species, such as pigeons, chickens, turkeys, and other domesticated birds (1,2). Infection with Trichomonas gallinae can present itself as either asymptomatic or lethal. Common symptoms encompass reduced hunger, emesis, unkempt plumage, diarrhoea, respiratory distress, dysphagia, weight reduction, and heightened thirst (3,4,5). The localpigeon, scientifically known as Columba livia domestica, is considered the primary carrier of T. gallinae, which therefore aids in the transmission of the disease. The parasite mostly infests juvenile pigeons. Trichomoniasis can potentially lead to death, however there is also a condition known as asymptomatic trichomoniasis, when there are no visible signs of illness. This scenario frequently arises (6). T. gallinae is very virulent and can cause excessive growth of abnormal tissue in the oropharynx, resulting in mortality in birds that have not developed immunity (7). Adult pigeons get trichomoniasis when they engage in nursing their young or when they utilise feeders and water sources (8). To identify T. gallinae, the typical method entails employing microscopic and morphological investigation. Nevertheless, this approach is time-consuming and requires the use of new samples and a specific procedure (9,10). The objective of this study is to track the occurrence of avian trichomoniasis in pigeons in Al-Qadisiyah Province, Iraq. This will be done by the clinical symptoms of trichomoniasis and visualizing the causative organism using a light microscope as well as employing a molecular along with observing.

Materials and methods:

Ethical clearance: Approval granted based on ethical considerations The authors of this study affirm that all methods in our experiment were conducted in accordance with the authorized Ethical Norms by the scientific board of the College of Veterinary Medicine, University of Al-Qadisiyah (committee permission number 1422 on 18/10/2023).

Birds and study area

The current investigation comprised a total of 150 localpigeons, technically known as Columba livia domestica. The study was conducted in Al-Qadisiyah Province, Iraq, with a sample length of about one year, from September 2022 to September 2023. The process of collecting samples is crucial in diagnosing Trichomonas gallinae infection. The local markets in the Al-Diwaniya

Governorate served as a primary source for Columba livia domestica pigeons. Sampling was conducted on the oral cavities of birds as conducted by Qiu et al. (11). By using sterile cotton-tipped applicators to mix swabs with phosphate buffered saline (PBS). The swabs were then studied under a light microscope at 400x magnification to observe T gallinae in a wet mount.

Swabs preparation:

Following the addition of a tiny quantity of physiological saline solution (also known as normal saline solution) to the pit, moist cotton swabs were extracted from it using a thin rubber tube connected to a syringe. Subsequently, the slide was secured using fire and treated with Giemsa stain. To conduct a thorough analysis of the parasite, it is necessary to see a pristine glass slide under a light microscope and place another slide on top for direct viewing Fig 1.

Figure 1: collection swab sample from infected pigeon.

Statistical analysis:

The total prevalence rates of parasitic illnesses in doves and local pigeons, as well as their sexes, were compared using a Chi-square test with the SPSS computer. The statistical differences were deemed significant at a significance level of $P<0.05$ (12).

Results and discussion

Clinical study

The current study observed that the macroscopic lesions in infected pigeons were characterized by white to yellow caseous masses, together with inflammatory ulcerated oral cavities and digestive tracts Fig 2.

Fig 2: A pigeon that was naturally infected with Trichomonas gallinae exhibited the presence of yellowish caseous material in its oral cavity.

Microscopic study

1-Direct Examination

The parasite was discovered based on physical characteristics, the direct swab revealed the parasite's typical vibratory movement caused by the anterior flagella and the undulating membrane, as well as a pear shape if the anterior half was wider than the posterior. From 150 samples, 98 (65.33%) were positive.

2- Stanning method

The microscopic features of T.gallinae were clearly visible when smears were produced and stained with Giemsa. The cytoplasm is stained with Giemsa, appearing light purple, while the nucleus is stained dark purple. The flagella, nucleus, and cytoplasm are clearly visible Fig (3).

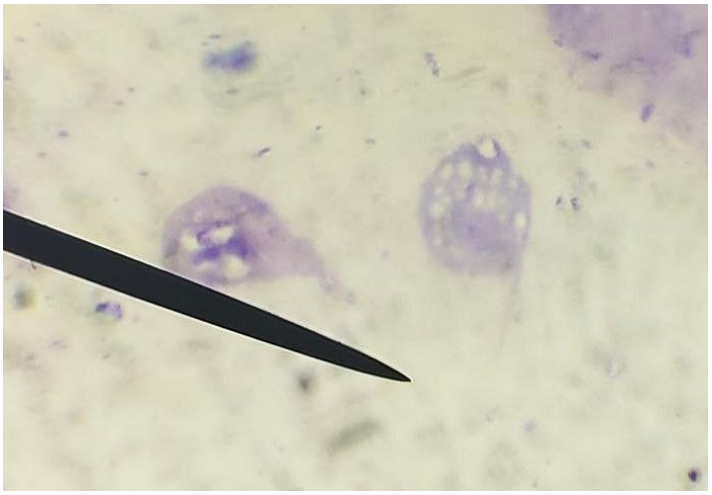

Figure 3: Microscopic examination of T.gallinae using a Giemsa stain at a magnification of 100x.

The prevalence of T.gallinae infection categorized by age.

The infection rate was higher in adult pigeons, with 76 out of 100 samples (76%) testing positive, compared to young pigeons, where 22 out of 50 samples (44%) were infected. Demonstrated noteworthy variance (P<0.05) in the overall prevalence of T.gallinae. table (1)

Table (1): The prevalence of T.gallinae infection in local pigeons varies according to age.

Age	Total number	Positive	%	X^2	P value
Adult	113	76	67.25	0.748	0.387*
Young	37	22	59.45		
Total	150	98	65.33		

*** No significant difference at P<0.05.**

The prevalence of T.gallinae infection according on gender:

A total of 150 samples were obtained from pigeons, consisting of 78 males and 72 females. These samples were analyzed for T.gallinea. The results revealed that 64.10% of males (50 individuals) and 66.66% of females (48 individuals) were verified to be infected, as indicated in Table (2). There was no statistically significant difference (p<0.05) in the total prevalence between males and females.

Table (2): The prevalence of T.gallinae infection in local pigeons categorized by gender.

Sex	Total number	Positive %	%	X2	P value
Male	78	50	64.10	0.109	0.742*
Female	72	48	66.66		
Total	150	98	65.33		

*** No significant difference at P<0.05.**

The monthly incidence of T.gallinae infection:

A total of 150 pigeons were participated in the study, with an average of 150 pigeons each month throughout a six-month period from October 2022 to April 2023. The findings indicated that the highest incidence of infection occurred in February and March, with rates of 68% and 72% respectively. Conversely, the lowest infection rates were observed in November and December, with rates of 23% and 28% respectively. Statistical analysis revealed significant differences (P<.05) in infection prevalence between different months, as presented in Table (3).

Table (3): The prevalence of T.gallinae infection in local pigeons varies according to the months.

Month	No. of collected samples	No. of positive infection	(%)	X^2	P value
November	25	9	36	20.08	0.001*
December	25	8	23		
January	25	7	28		
February	25	16	64		
March	25	17	68		
April	25	18	72		
Total	150	75	52		

*** No significant difference at P<0.05**

Discussion:

Upon conducting a physical examination of multiple local pigeons infected with *Trichomonas gallinae*, it was observed that there were lesions in the mouth and esophagus that bore a resemblance to a substance similar to cheese. These lesions varied in color, ranging from white to yellow. The outcome aligned with the one previously reported by (13, 14,15).

Many studies have demonstrated that the swab test utilised for diagnosing trichomoniasis is a fast and cost-effective method that involves sampling from local pigeons. This strategy yields an accuracy rate ranging from 50% to 70% (16, 17,18).

Bunbury and his team have stated that this method can be employed in practical settings to detect instances of trichomoniasis. The effectiveness of the technique may surpass that of the culture method, contingent upon various factors such as the examination's quality, precise application of intensity by the devices, and the severity of infection in the birds (19).

Contrary to the present study findings, many studies present conflicting evidence indicating that employing the wet mount preparation method results in a 50% decrease in diagnostic precision and requires a substantial concentration of protozoa to identify a positive case (20). The current work aligns with Mohammed et al., (21) findings, which observed that the nucleus and flagella were stained a light purple color with Giemsa, whereas the cytoplasm was stained a dark purple color. The current study's findings, which are consistent with other previous studies conducted in various locations of Iraq, namely in Sadia and Hazah in Kirkuk city, employed three distinct stains to characterize the morphology of T. gallinae. Among these stains, the Giemsa stain shown superior efficacy in staining T. gallinae compared to the other two stains (22). In addition, Fadhil et al. (18) reported that the Giemsa stain shown high efficacy when seen under a microscope at a magnification of x100.

The current study revealed a prevalence rate of 65% (98/150) for Trichomonas gallinae infection in local pigeons (Columba livia domestica) in Al-Diwaniya city. It is almost derived from the discoveries of Fadhil *et al.,* (18) When they documented that the prevalence of Trichomonas gallinae infection was 58% in Baghdad province. Also recorded by Jaafar (23) 61.32% in local pigeon and 63.4% in wild pigeon. As well as, the prevalence of trichomoniasis was consistent with Al-Barwari and Saeed (24) they reported 58% in Rock pigeon.

Our findings contradict those of Al-Bakry (25), who reported infection rates of 22% in birds, 17.5% in wild pigeons, and 10% in spilopelia senegalensis.

The incidence rate in local pigeons was established to be 50% in the kingdom of Saudi Arabia, whereas in wild pigeons it was found to be 68%. This result was closer to that of a neighboring country (26, 27). A study conducted in Iran found that wild pigeons had a 50% infection rate (28). The variation in infection rates observed across studies may be influenced by factors such as climate, geographical region (humidity and temperature), season, host resistance, feeding habits of birds, age of birds, number of examined pigeons, sample size, and the state of local pigeon breeding (29).

Hamza (30) found that the prevalence of T. gallinae infection in local pigeons varied according to age. In mature pigeons, the infection rate was 33.33%, while in young pigeons it was 20% in Diyala province. Quillfeldt et al. (31) observed a 50% infection incidence in adult columbi forms in Germany. The findings of the present study contradict the findings of Eman (32), who reported a prevalence of 59.50% in adult pigeons and 61.60% in young pigeons in Egypt.

One potential reason for variations in infection rates based on age may be the clustering of adult birds at the feeder and water source, as well as the stress caused by overcrowding. In addition, several pigeon species lack immunity to the particular strain of illness, and the practice of raising local pigeons in confined structures promotes the spread of parasites among the birds (33).

The infection rate of T.gallinae is similar across females and males, with 49.8% (49/98) and 51.09% (51/98) respectively. This suggests that both genders have an equal probability of becoming infected. This finding aligns with the research conducted by Begum et al (34), which reported an infection rate of 70.9% in females and 63.8% in males in Bangladesh.

This discrepancy may arise due to the clustering of mature avian species at the feeding and watering locations, as well as the strain resulting from excessive population density. Similarly, certain pigeons lack immunity to specific diseases, and the practice of breeding local pigeons in compact towers promotes the spread of parasites among the birds (35,18).

The prevalence of T. gallinae infection in local pigeons exhibited monthly variation, with February recording the highest infection rate of 22.4% (22 out of 98 pigeons), while April had the lowest incidence of 10.4% (10 out of 98 pigeons). In February, the incidence of infection is elevated due to the natural breeding season of pigeons. Subsequently, there is a surge in the population of juvenile pigeons, which are particularly vulnerable to the disease due to their consumption of milk contaminated with parasites from their mothers. The weather also influences the transmission of the illness (18).

Conclusions

The study revealed a notably high occurrence of T. gallinae in pigeons, surpassing previous studies conducted in various countries or regions within the country. This emphasizes the pressing need for owners to conscientiously adopt suitable practices for housing, disease control, and feeding. Implementing these procedures is essential for effectively decreasing the occurrence of trichomoniasis and perhaps eliminating the causal agent T. gallinae.

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