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Original article

Occurrence of cestodes and comparative efficacy of *Typha angustata* and sulphadimidine against cestodes in *Columba livia domestica* (Domestic Pigeon)

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ABSTRACT

The occurrence of intestinal parasites of Columba livia domestica has been on the increase, leading to high economic and production losses with more fatal cases. This study was designed to investigate the prevalence of cestodes in pigeons and determine the efficacy of Typha angustata extract and sulphadimidine against these cestodes in the domestic pigeon. A total of 30 pigeons were examined. 18 (60%) pigeons were found infected with only one type of cestode species (Raillietina spp.). The difference in prevalence between males and females was statistically significant (χ^2 = 8.167, p = 0.004). The mean EPG count in group A (treated with T. angustata extract) before treatment and after treatment was 176 ± 4.33 and 155 ± 4.24, respectively. In group B (treated with sulphadimidine), the mean EPG calculated before treatment and after treatment was 184 ± 6.74 and 35 ± 3.53, respectively. The efficacy at day 28 of T. angustata and Sulphadimidine was 11.93% and 80.97%, respectively. It was concluded on the basis of the EPG and efficacy data that T. angustata extract had low efficacy against raillietiniasis, while as sulphadimidine, which is also used before to treat different intestinal parasites, had a good efficacy against raillietiniasis. Further studies are required to know the prevalence of other gastrointestinal parasites in pigeons and efficacy of different medicinal plants against such parasites.

1. Introduction

Pigeons are present almost in every part of the globe (Marques et al., 2007). It is estimated that 170–340 million pigeons reside in cities worldwide (Haag-Wackernagel and Bircher, 2010). Many health issues can affect pigeons, but infestations with parasites play an essential part. Many ecto- and endoparasites, associated with several pigeon diseases, are found on the skin and various internal organs (Parsani et al., 2014). Helminths have been involved in causing infection and higher mortality in domestic pigeons. They are considered as one of the critical weaknesses in the production of commercial pigeons in Pakistan (Tanveer et al., 2011). Roundworms, stomach wall worms, hairworms, gapeworms, strongyloids, and tapeworms are the common worms reported in pigeons. Among cestodes, *Raillietina* spp. is the most prevalent parasite reported in pigeons. Its extreme infestations causes droopiness and weight loss (Kamal et al., 2020).

Various anthelmintics was used for the treatment of helminths infections in birds. The most widely used anthelmintic is albendazole and fenbendazole (Bowman et al., 1995; Tucker et al 2007)). Ssenyonga (1982) reported that fenbendazole efficacy is 100% against helminths parasites of birds. Albendazole has very adverse side effects and induce liver injury, even in small doses (Piloiu and Dumitrascu, 2021). Sulphadimidine is used regularly in chicken

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husbandry for its wide-spectrum activities toward bacterial and coccidian infections (Khan et al., 2021). However, the therapeutic efficacy of sulphadimidine on helminths infection in pigeons was not yet investigated. Therefore, sulphadimidine was used is this study to find out its efficacy against cestodes.

Recently, there has been a growing interest in ethno-veterinary medicine worldwide. This change responds to the development of animals free from synthetic chemical additives in the developing world and the need to find new beneficial substances of natural origin with relatively low toxicity to humans and animals (Guarrera, 1999, Gasbarre et al., 2001). In traditional literature in India and China, *T. angustata* is the most common medicinal herb used for various medicinal properties. It is a member of the Typhaceae family. It is an annual plant growing in a pond or on the side of a river in shallow water. It is 3–6 feet in height and growing straight with its leaves and stems. The thickness of its leaf is 5–12 mm. The leaves are used for diuretic purposes (Duke and Ayensu, 1985). It is used for hematemesis, nosebleeding, hematuria, dysmenorrhea, uterine bleeding, abdominal postpartum pain, scrofula, gastralgia and abscesses. The root material is diuretic and astringent (Chopra et al., 1986). It contains various ingredients, i.e., flavonoids, tannins, sterols, and triterpenes. In the stems and flowering tips, flavonoids are present, i.e., quercetin, naringenin, isorhamnetin-3–0-neohesperido sine, isorhamnetin, kaempferol-3–0-neohesperidosine, isorhamnetin-3–0-rutinoside, β-sitosterol, lanosterols, and cholesterol are contained in the sterols. (Kumar et al., 2013). In Pakistan very less work has been done with regard to the anthelmintic properties of indigenous medicinal plants in poultry. Therefore, this plant was used in this study to know its efficacy against cestodes in pigeons.

Keeping in view the background information the current research was designed to study the occurrence of cestode infections in pigeons of District Dir Lower, Khyber Pakhtunkhwa, Pakistan and determine the anticestodal activity of *T. angustata* and sulphadimidine in *Columba livia domestica* (domestic pigeon).

2. Materials and methods

2.1. Study area

This study was performed following the international guiding principles for biomedical research involving animals, and approval was taken from the animal ethics committee of the Abdul Wali Khan University Mardan.

2.2. Study area

The present study was carried out in District Dir Lower, Khyber Pakhtunkhwa, Pakistan. Its geographical coordinates are 34° 33′ 56″ North, 71° 55′ 52″ East. This area is characterized by heavy rainfall in winter and hot weather in summer. Temperature ranges from 12 °C to 40 °C. The map of the study area was prepared using ArcMap 10.5 software (Fig. 1).

2.3. Pigeons'

Thirty (30) domestic pigeons (*Columba livia domestica*) of both sexes were purchased from local markets of District Dir lower (Fig. 1). The history, sex, and age of the pigeons were recorded. They were kept in wide and well-ventilated animal house chambers at the University of Malakand, Khyber Pakhtunkhwa, Pakistan. The pigeons were fed on freshwater with wheat and grains. The specimens were placed in this environment for acclimatization for two weeks before the beginning of the experiment.

2.4. Preparation of plant extract

The herb *T. angustata* were collected from the side of the river Panjkora, Timergara, Dir lower. The plant was botanically identified in the Department of Botany, University of Malakand, Khyber Pakhtunkhwa, Pakistan. Whole plant of *T. angustata* were cut off, rinsed with fresh water, and dried under a shed for two weeks.

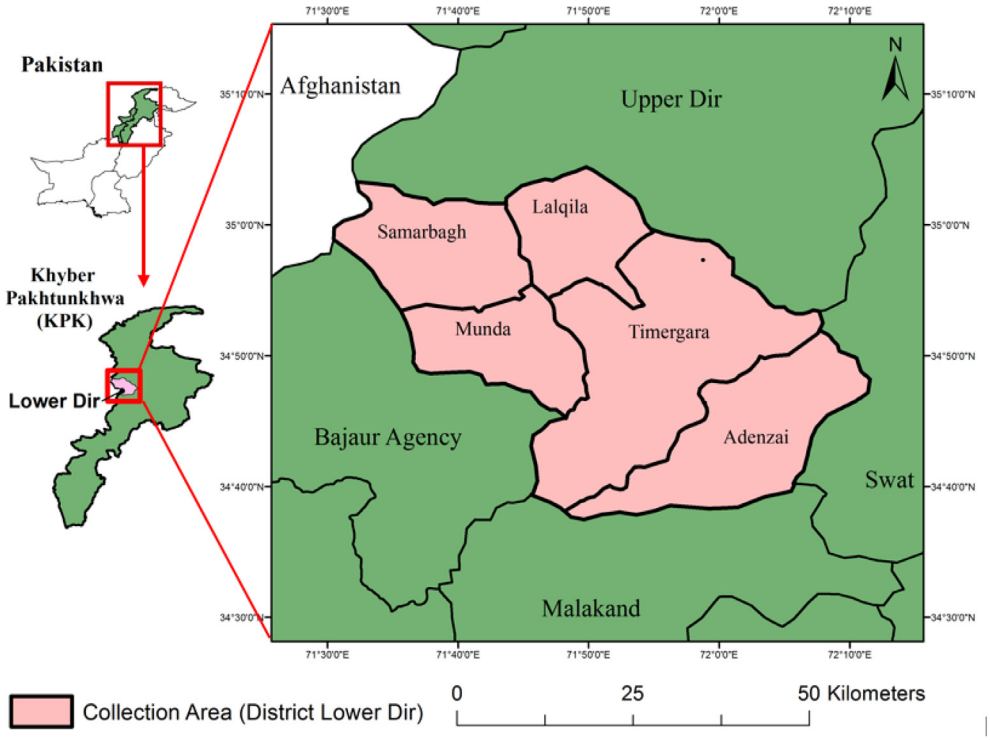

Fig 1. Map showing study area i.e. District Dir Lower.

Table 1
Experimental details of birds grouping and dosage per kg body weight.

Group	No of Pigeons	Drug	Dosage per Kg body weight			
			Day 1	Day 8	Day 15	Day 22
A	6 (Infected)	Typha Angustata	30 ml	45 ml	60 ml	75 ml
B	6 (Infected)	Sulphadimidine	30 mg	45 mg	60 mg	75 mg
Cp	6 (Infected)	No treatment	–	–	–	–
Cn	12 (Uninfected)	No treatment	–	–	–	–

The shed dried plant was mechanically ground in an electric chopper to get fine powder forms.

During extract preparation, 100 g of powdered *T. angustata* was soaked in 500 ml de-ionized water. The soaking continues for three days with occasional shaking. The soaked powdered plant was filtered through muslin cloth for removing debris. The filtrate was then filtered through Whatman No.1 filter paper, and a clear aqueous extract was obtained. The filtrate was stored at 4 °C in the refrigerator to avoid the fungal attack.

2.5. Sulphadimidine (Commercially available drug)

This drug was purchased from a veterinary shop at Lower Dir. This drug was in tablet form. Each tablet was 350 mg.

2.6. Grouping of birds

Firstly, 30 pigeons' droppings were collected and examined for cestodes eggs. 18 pigeons were found infected. These pigeons were then divided into four groups (A, B, Cp and Cn). For each individual's identity in a group, different coloured foot rings were used and then birds were weighted by manual weight balance and placed in separate cages. The experimental details are given in Table 1.

2.7. Collection and laboratory examination of droppings samples

Dropping samples were collected (5–50 g each) at day 0 (before treatment), day 7, 14, 21 and 28 (after treatment). The samples were collected in a plastic container, preserved in 10 per cent formalin solution, and transferred to the laboratory of parasitology, University of Malakand Chakdara, for qualitative and quantitative examination.

Direct microscopy and flotation techniques were used for qualitative analysis of dropping samples and for quantitative analysis, the McMaster methodology was performed as per the standard protocols. The protocols laid down by William (2001), Dranzoa et al. (1999) and (Soulsby, 1982) for Direct microscopy, Flotation method and McMaster method, respectively were followed.

2.8. Collection and examination of gastrointestinal tracts

Each pigeon was placed in a vacuum chamber containing cotton wool soaked with 10 ml of chloroform for 6 to 7 min after the end of the trail. The anaesthetized pigeons were then dissected. Each of the pigeon was slit open from the cloaca to the neck region. The visceral organs like gizzard, small and large intestines, and liver were removed from the body.

The parasites were only present in the large intestines and were gently removed in Petri dishes containing 70% alcohol. The specimens were then processed and permanent mounts of the parasites were made by following the standard protocol of Al Quraishy et al. (2019). The slides were then examined under the microscope and the parasites were identified using standard morphological keys (Cheng, 1973; Soulsby, 1982; Ruprah et al., 1986). The photomicrographs were prepared using an automatic photomicrographic camera mounted on a trinocular microscope (Labomed Labo America, Inc USA) in the laboratory of Zoology, University of Malakand, KPK, Pakistan. The silent morphological features of the identified cestode are: Total body measurement varies from 17.65 mm to 60.80 mm in length and 0.4 mm to 1.26 mm in width. Scolex is simple, wider than long. Rostellum with a row of minute hammer shaped hooks. Sucker is simple. Immature, mature and gravid segments are wider than long. Gravid segments are also much wider than mature segments. The uterus is broken into egg capsules, each containing several eggs (Fig. 2). The prepared slides were labelled and deposited in Department of Zoology, University of Malakand, for future reference.

2.9. Statistical analysis

Microsoft Excel 2013 was used for raw data entry and calculation of prevalence, parasitic burden, EPG and efficacy. Chi-square analysis was performed for the assessment of risk factors in EpiInfoTM Center for Disease Control. Below 0.05 P value was considered significant. Prevalence and parasitic burden were calculated by the following formulas:

Prevalence = number of pigeons infected /Total number of pigeons examined*100

Parasitic burden per pigeon = Total number of parasites recovered/infected pigeons.

The % efficacies of the drugs were measured by the formula given below (Thrusfield, 2005):

$$Efficacy\ (\%) : (Mean\ EPG\ before\ treatment\ --\ Mean\ EPG\ after\ treatment)\ X\ 100\ Mean\ EPG\ before\ treatment$$

3. Results

3.1. Prevalence

A total of 30 domestic pigeons were examined. 60% of pigeons were found infected with *Raillietina* spp. tapeworm (Table 2). In the present study 82.35% males and 30.77% females were found positive for *Raillietina* spp. The difference in prevalence between males and females was statistically significant (χ^2 = 8.167, p = 0.004) (Table 2). On gastrointestinal tract examination, a total of 151 *Raillietina* spp. cestodes were recovered from all the birds. The mean parasitic burden among infected pigeons was 8.38 ± 2.43.

3.2. Dropping egg count

The mean egg per gram (EPG) of droppings was calculated before and after treatment in groups A, B, Cp, and Cn. Group A, which was treated with *T. angustata* exhibited the mean EPG before treatment on day 0 as 176 ± 4.33. EPG after treatment at day 7, 14, 21, and 28 was 167 ± 8.13, 165 ± 5.44, 159 ± 6.32, and 155 ± 4.24, respectively. Before treatment and after treatment, the mean EPG count was 176 ± 4.33 and 155 ± 4.24, respectively (Table 3).

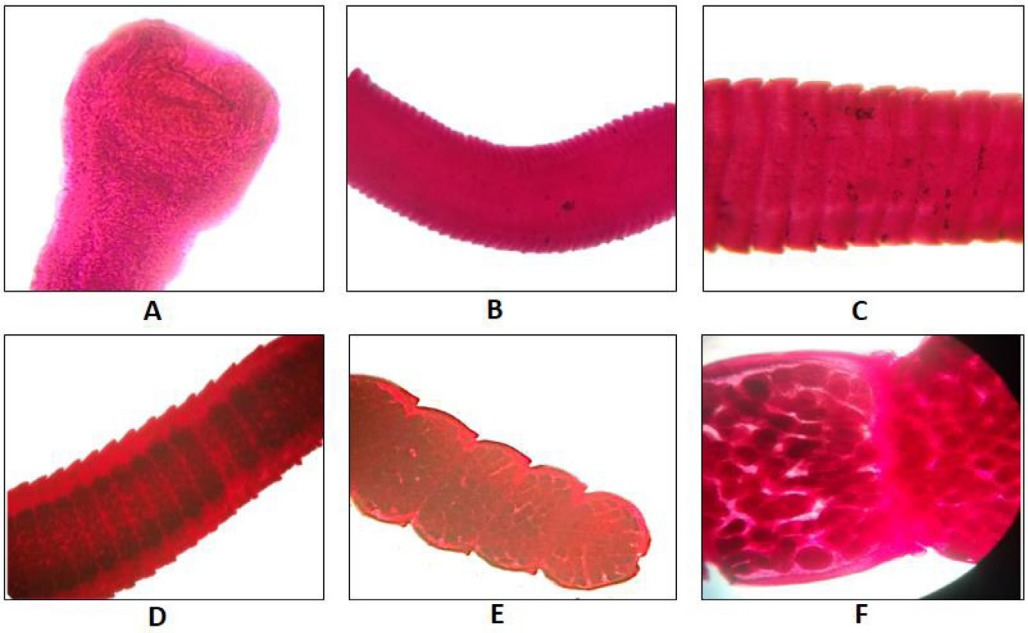

Fig 2. *Raillietina* spp.: A = Scolex; B-C = Immature segments; D = Mature Segments; E-F = Gravid segments occupied by uterine eggs.

Table 2
Prevalence of *Raillietina* spp. parasites in male and female pigeons.

Sex	Pigeons Examined	Pigeons Infected	Prevalence %	Chi-Square	P-value
Male	17	14	82.35	8.167	0.004*
Female	13	4	30.77		
Total	30	18	60.00		

Table 3
Mean EPG in different groups observed on different days.

Groups	Treatment	Mean egg per gram of droppings (Mean EPG)				
		Day 0	Day 7	Day 14	Day 21	Day 28
A (n = 6)	*Typha angustata*	176 ± 4.33	167 ± 8.13	165 ± 5.44	159 ± 6.32	155 ± 4.24
B (n = 6)	Sulphadimidine	184 ± 6.74	155 ± 3.37	126 ± 3.55	84 ± 4.55	35 ± 3.53
Cp (n = 6)	Nil	162 ± 8.87	175 ± 4.44	179 ± 7.22	168 ± 5.36	187 ± 1.33
Cn (n = 12)	Nil	0	0	0	0	0

Cp = Control Positive; Cn = Control Negative.

Group B was treated with sulphadimidine and the mean EPG calculated before treatment on day 0 was 184 ± 6.74. EPG after treatment at day 7, 14, 21, and 28 was 155 ± 3.37, 126 ± 3.55, 84 ± 4.55, and 35 ± 3.53, respectively. The mean EPG count before treatment and after treatment was 184 ± 6.74 and 35 ± 3.53, respectively (Table 3).

Group Cp was untreated positive control, and the mean EPG was 162 ± 8.87 at day 0. EPG at day 7, 14, 21, and 28 was 175 ± 4.44, 179 ± 7.22, 168 ± 5.36, and 187 ± 1.33, respectively. EPG count on day 28 was slightly increased, while in the Cn group, no cestode egg was noted from day 0 to day 28 (Table 3).

3.3. Efficacy

The efficacy of *T. angustata* was calculated against *Raillietina* spp. for four weeks. The per cent efficacy calculated on day 7, 14, 21, and 28 were 5.11%, 6.25%, 9.65%, and 11.93%. The lowest efficacy was noted on day 7, while the highest efficacy was noted on day 28. When the quantity of dose increased in the next week, the efficacy slightly increased (Table 4).

The efficacy of Sulphadimidine was also calculated against *Raillietina* spp. for four weeks. The percent efficacy calculated at day 7, 14, 21, and 28 were 15.76%, 31.52%, 54.34% and 80.97%. The lowest

Table 4
Efficacy of T. angustata and Sulphadimidine against raillietiniasis in pigeons.

Groups	Treatment	Efficacy (%)			
		Day 7	Day 14	Day 21	Day 28
A (n = 6)	*Typha angustata*	5.11%	6.25%	9.65%	11.93%
B (n = 6)	Sulphadimidine	15.76%	31.52%	54.34%	80.97%

efficacy was noted on day 7, while the highest efficacy was noted on day 28. When the quantity of dose increased in the next week, the efficacy increased significantly (Table 4).

4. Discussion

Many studies have been conducted on the prevalence of parasitic infections in Pigeons worldwide (Diakou et al., 2013; Mehmood et al., 2019; El-Dakhly et al., 2019). In the current study, 60% of pigeons were found infected with only one type of cestode species: *Raillietina* spp. In Pakistan, Khan et al. (2018) also investigated the parasitic infestations in pigeons. They recorded an overall prevalence of 66.6% with presence of two cestodes (*Raillietina* spp. 60% and *Cotugnia* spp. 13.3%) and one nematode (*Ascaridia* spp 6.66%). The prevalence of *Raillietina* spp. was similar to the prevalence recorded in the current study.

Authors in different countries also worked on the prevalence of *Raillietina* tapeworm in pigeons and recorded lower or higher prevalence rate than reported in the present study. Mehmood et al. (2019) in India recorded 25% prevalence, Safi-Eldin et al. (2019) in Cairo, Egypt recorded 24.79% prevalence, Mohammed et al. (2019) in Kano State, Nigeria recorded 13.80% prevalence, El-Dakhly et al. (2019) in Aswan, Egypt recorded 43.33% prevalence, Sadeghi-Dehkordi et al. (2019) in Hamedan, Iran recorded 20% prevalence, Chaechi-Nosrati et al. (2018) in Lahijan, Guilan, Iran recorded 54.5% prevalence, Alkharigy et al. (2018) in Tripoli, Libya recorded 35% prevalence, Umaru et al. (2017) in Taraba State, Nigeria recorded 18.3% prevalence, Parsani et al. (2014) in Gujrat, India recorded 31% prevalence. In contrast, Al Quraishy et al. (2019) in Saudi Arabia recorded 77.78% prevalence, Diakou et al. (2013) in Thessaloniki, Nothern Greece recorded 70.58% prevalence. The highest (100%) prevalence was recorded by Begum and Sehrin (2012) in Dhaka, while the lowest (6%) prevalence was recorded by Ghosh et al. (2014) in CMA, Bangladesh.

The difference in prevalence between male and female was statistically significant (χ^2 = 8.167, p = 0.004). The results are similar to the observations of Mohammed et al. (2019) and Umaru et al. (2017). In contrast, Khan et al. (2018) reported non-significant difference in the prevalence rates in males and females'.

The per cent efficacy calculated at day 7 in group A and group B was 5.11% and 15.76%, respectively. On day 28, in group A and group B, per cent efficacies were 11.93% and 80.97%, respectively. After the analysis of EPG and efficacy percentage data, it is clear, that the *T. angustata* extract is not a suitable medication against raillietiniasis. In contrast, sulphadimidine, which is also used to treat different intestinal parasites, has also been found effective against raillietiniasis. Sulphadimidine has not been used before for the treatment of cestodes infection. The mechanism of action of sulphadimidine is unclear. Further studies are required to detect the toxic effects of sulphadimidine in animals and to find out its mode of action against cestodes.

5. Conclusion

Based on the present study, it is concluded that *Raillietina* spp. is the most prevalent parasite of domestic pigeons in Lower Dir, Khyber Pakhtunkhwa, Pakistan. *Raillietina* spp. Usually, parasitize the avian fauna of wide geographical regions.

The data suggested that *T. angustata* extract is less effective against raillietiniasis, while sulphadimidine is more effective to treat raillietiniasis. Further studies are warranted to know the prevalence of various parasitic diseases in pigeons along with the effective control strategies.

Declaration of Competing Interest

All authors declare no conflicts of interest. All authors listed in the manuscript contributed and attest to the validity and legitimacy of the data and its interpretation and agree to its submission to your journal.

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