Research Article

Open Access

Journal of Veterinary Sciences and Dairy & Poultry Research

jvsd@scientificeminence.com

Migratory Japanese Quail (*Coturnix coturnix japonica*) as a Host and Carrier for Coccidiosis and Ascaridiasis

Faizullah*[1], Jan S[1], Taj K[1], Ud Din Z[1], Akbar Khan M[2], Sattar A[2] and Farooq M[2]

[1]Center for Advanced Studies in Vaccinology and Biotechnology, University of Balochistan, Pakistan

[2]Livestock and Dairy Development Department, Govt. of Balochistan, Pakistan

*** Corresponding Author**

Faizullah, Department of Parasitology, University of Balochistan Quetta, Pakistan, Tel: 03354612236, E-mail: Faizullah_68@yahoo.com

Citation

Faizullah, Jan S, Taj K, Ud Din Z, Akbar Khan M, et al. (2021) Migratory Japanese Quail (*Coturnix coturnix japonica*) as a Host and Carrier for Coccidiosis and Ascaridiasis. J Vet Sci Dairy Poult Res 1:1-10

Publication Dates

Received date: October 10, 2021

Accepted date: November 10, 2021

Published date: November 11, 2021

Abstract

This study was conducted to detect *Eimeria tenella* and *Ascaridia galli* in migratory Japanese quail (*Coturnix japonica*). To idientify the protozoan and intestinal prasite of wild birds Out of 300 fecal samples of birds processed through microscopy, 218 (72.6%) were positive for Eimeria oocysts. Length, width and shape index of oocysts of *E. tenella* were 21.7 ± 1.29 µm, 19.5 ± 1.9 µm and 1.27± 0.12, respectively. The oocysts of *E. tenella* were further confirmed by PCR targeting mitochondrial fragment CO-1 gene (727bp). BLAST results of the sequence showed similarity (99%) to *E. tenella* (JX853830.1). Furthermore, 230 birds were slaughtered; gastro-intestinal tracts (GIT) were collected and examined for the presence of parasitic infestation, out of which 120 quails were found positive (52.17%) for *Ascaridia galli*. In molecular study the targeted gene, CO-1 (Cytochrome oxide-1), was used for the identification of *Ascaridia galli*. The result of sequence revealed 99% similarity of *Ascaridia galli on BLAST with Ascaridia galli of chicken. In conclusion,* molecular methods are best for species identification of *Eimeria. tenella* and *Ascaridia galli.* Finally, migratory Quails are potential risk factors for dispersal of protozoans and nematodes which then may infect and infest local and commercial chicken, respectively.

Keywords: *Ascaridia galli; Eimeria tenella;* Migratory Japanese quail (*Coturnix japonica*); Oocysts; PCR Sequence

Introduction

Quail is a medium-sized bird that belongs to various genera of family Phasianidae. Two species, the Japanese quails (*Coturnix japonica*) and Bobwhites quails (*Colinus virginianus*) are considered as domestic birds since 14th century (El-Ghany, 2019) [1] Quail production is a branch of the modern poultry industry (Teixeira et al., 2004) [2]. The breeding of Japanese quails has excelled in aviculture due to increased consumption of exotic meats and eggs. Japanese quails can be a substitute for chicken production (Berto et al., 2011) [3] and is used for commercial breeding (Rahman & Manap, 2014) [4]. The natural habitat of Japanese quail is East Asia. It is migratory in nature. The wild Japanese quail are also available in South Eastern Siberia Manchuria and Korean Peninsula. It is also found in some parts of the North Eastern regions of India. Quails live in grasslands and cultivated field (Anonymous, 2014) [5].

Geographically located in the middle of Asian flying route and having smart wetlands, each year Pakistan gets a large number of migratory birds (Umar et al.2018) [6]. Different studies have shown that bird's migration is due to seasonal changes, availability of food and to avoid threat of predation (Scott1991; Lank *et al.*2003) [7,8]. The species of birds that migrate from Siberia to Pakistan are Houbara bustard, cranes, teals, pintails, mallards, geese, spoon bills, waders, quails and pelicans. Migration of birds takes place mainly from Northern arctic region towards Southern plains. These birds spend winter season in tropical areas and breed where they stay for 2 to 3 months (Umar et al. 2018) [6]. The presence of migratory birds in the particular areas indicates that the site is favorable for feeding, nesting and breeding. Birds are believed to spread a variety of pathogens, including viruses, bacteria, protozoa and helminths (Mihaela & Marina 2014) [9].

Parasitic diseases in poultry like coccidiosis are major issues to the poor farming community both in tropical and subtropical regions in Pakistan. Species of *Eimeria* like *E. tenella* and *E. maxima* cause coccidiosis in chicken, quails and pigeon (Latif et al., 2016) [10]. *Eimeria tenella* is a well-known host-specific protozoan parasite that causes coccidiosis in chicken. However, experimental studies have shown that this host specific coccidia can sexually develop in the intestine of Japanese quail (Mathis & McDougald, 1987) [11]. The predilection site of *Eimeria tenella* is caeca of intestine, causes bloody diarrhea which results in severe morbidity and motility with major economic losses. Birds infected with *Eimeria* are considered potential source of infection for other birds as they release oocysts in their feces (Baillie & Peach, 1992) [12]. In Pakistan, due to poor farming system, the disease becomes more severe causing heavy economic losses. Although the exact losses caused by coccidiosis in poultry industry are not known, however, such losses may be in millions of rupees in Pakistan.

Helminthes infestation is more common in chickens due to their free-ranging mode of life (Ondwassy et al., 2000) [13]. It is estimated that parasitic disease is among the most important challenge, which hinder poultry farming (Kaufman et al. 2007) [14]. The most common disease caused by helminthes parasite in poultry is Ascaridiosis (Fatihu et al., 1991). *Ascaridia galli* is one of the most pathogenic and economically important parasites of poultry industry. Studies on *Ascaridia galli* in wild and migratory birds are important while determining the risk that this parasite may be transmitted to local bird populations during migration period. *Ascaridia galli role* was also reported in the transmission of *salmonella* infection resulting in diseases and economic losses (Kaufmann 1996) [15].The current study was conducted to determine pathogens of coccidiosis and ascaridiosis in migratory Japanese Quail *(Coturnix coturnix japonica)* from the migratory-route regions of Balochistan, Pakistan.

Materials and Methods

Sampling method

Quails were trapped by using Ball Chatri method (Ali, A & Khan, 2020) [16] from Quetta, Pishin, Zhob and Sibi districts of Balochistan. Identification of the Japanese Quail was performed with the help of experts from Zoology Department of University of Balochistan. Fecal samples were collected from November 2017 to April 2018 in sterile polythene bags and stored at 4°C till further laboratory procedures.

Processing of oocysts

Fresh fecal samples were examined by direct smear method (Zajac. A et.al 1994) [17] and fecal flotation technique (Tsuji *et al.*, 1999);(O, Horo, 2007) [18,19]. The oocysts were purified by salt flotation technique (Mohammad, 2012) [20] and sporulated in 2.5% potassium dichromate solution. The sporulated oocysts were washed with distilled water and stored at 4°C for molecular studies.

Morphology of oocysts

Microscope (Olympus BX 51) fixed with Olympus DP71 camera and software Image-Pro Express 6.0 were used for oocysts measurement (Carvalho et al., 2011) [21]. Oocyst shape index

was determined by measuring its length and width by using a calibrated ocular micrometer at 20X magnification.

Collection and identification of Ascaridia galli

All nematodes were collected from each GIT of the Quails and identified. For achieving the actual worm burden, each sample was counted separately. The birds were slaughtered and GIT were collected and transported safely to laboratory in polythene bags. The GIT samples were grossly examined for the presence of parasitic infestation. Adult worms were collected and washed in 1x Phosphate Buffer Saline (PBS). The washed worm samples were preserved in 95% ethanol for further morphological identification. Taxonomic keys were used to identify the nematode as previously described (Soulsby 1983, Rahman at al. 2009 Rahman and Manap2014) [4,22,23].

DNA extraction

The DNA was extracted using tissue kit following the manufacturer's instruction (Gene All Exgene mini cat No.104-101 Korea). The DNA was purified using kit following the manufacturer's instruction (Gene All Exgene Tissue Kit mini cat No.104-101 Korea). Quality and concentration of DNA was measured by Nano drop using thermo fisher spectrophotometer.

PCR and sequencing

PCR were performed for identification of *E. tenella* using *Eimeria* specific cocci mitochondrial fragment gene primers. The set of the primers used to amplify a product of 727 bp consisted of forward primer 5'GGTTCAGGTGTTGGTTGGAC'3 and reverse primer.5 'A TTC C A ATA AC CGCACCAAG'3. (Folmer et al., 1994) [24]. On the other hand, the primers for 533pbCO-1(Cytochromeoxide-1) gene of *Ascaridia galli* were F(5′ATTATTACTGCTCATGCTATTTGATG-3′)R(5 CAAAACAAAGTGTTAAATCAAAGG-3′). (Katakam et al., 2010) [25] Thermal cycler (Model GS 5482) was used for DNA amplification. The amplified products were electrophoresed in 1.5 % agarose gel stained with ethidium bromide, visualized through trans-illumination device and photographed. The PCR amplicons of CO-1 were sent for partial sequencing to Advanced Bioscience International Singapore. Multiple sequence alignment was performed using BioEdit version 7.2.5. Mega software (Mega 6) was used to reconstruct the phylogenetic tree using Maximum Likelihood (Tamura et al., 2013) [26].

Animal Ethics

This experiment was approved by Wild life and Forest Department Balochistan Quetta, license No. 127, dated 03.03.2017 and ethical committee of Center for Advanced Studies in Vaccinology and Biotechnology (CASVAB), University of Balochistan, Quetta.

Statistics Analysis

Data was entered in Microsoft Excel (version 13.0) and SPSS (version 20.0) was used for data analysis. Descriptive data was presented as frequency tables and percentages. Sizes were presented as mean and standard deviation.

Results

Eimeria tenella

Results of the current study showed that Oocyst of *Eimeria* spp were present in 72.6% (218/300) of fecal samples from migratory Japanese Quails. Microscopic examination revealed that oocysts of *E. tenella* were ovoid, smooth, colorless, without micropyle or residuum but with a polar granule. The sporocyst had a stieda body, (Figure 1a and b). Average size of oocysts of *E. tenella* was 21.7 ± 1.29 µm in length, 19.5 ± 1.9 µm in width and 1.27± 0.12 as shape index (Figure 2).

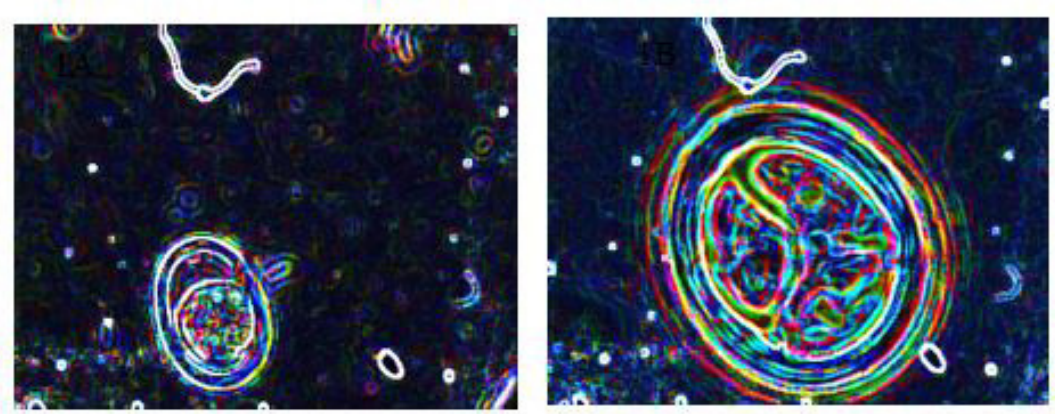

Figure 1: (A) showed unsporulated and **(B)** sporulted oocyst of *Emeria tenella*

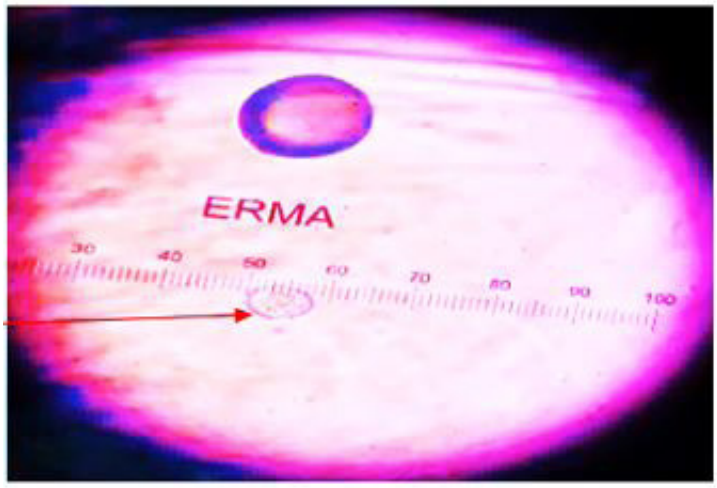

Figure 2: Micrometry of Oocyst of *Eimeria tenella* (Average size 21.7 µm)

PCR based detection and phylogenetic analysis of Eimeria tenella

The oocysts of *E. tenella* were further confirmed by PCR targeting mitochondrial fragment CO-1 gene (727bp). The amplified products were electrophoresed in 1.5 % agarose gel stained with ethidium bromide, visualized through trans-illumination device and photographed (Figure 3). BLAST results of the sequence showed similarity (99%) to *E. tenella* available publicly in NCBI GenBank. The BLAST result indicated that the sequence obtained during the current study had high similarity with the sequence of *E. tenella* isolated from chicken, and the Phylogenetic analysis (Figure 4) also showed that sequence of *E. tenella* had closely related to *E. tenella* isolates.

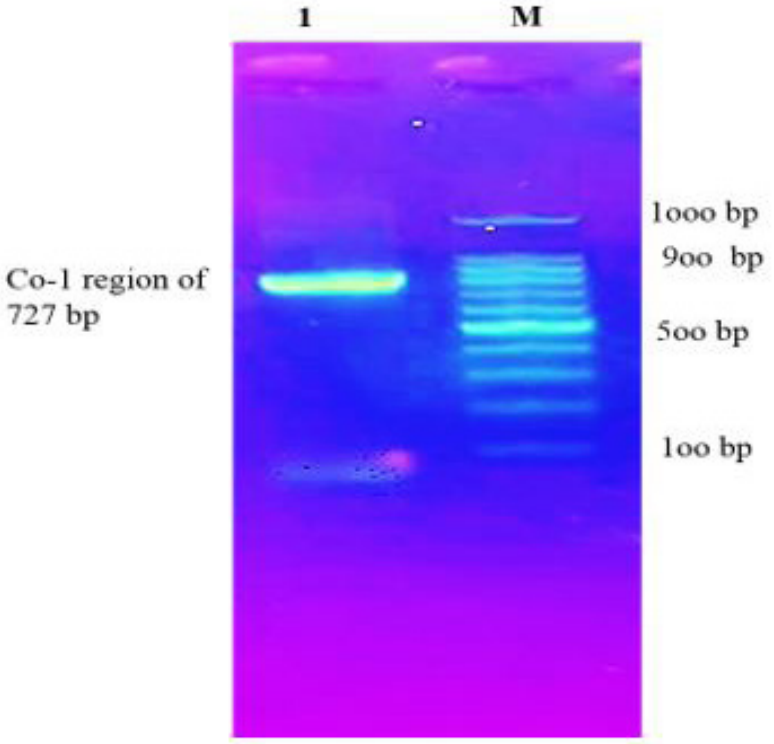

Figure 3: PCR image of E. tenella Lane 1: Amplification of CO-1 (727 bp). Lane M: DNA marker 1000 bp

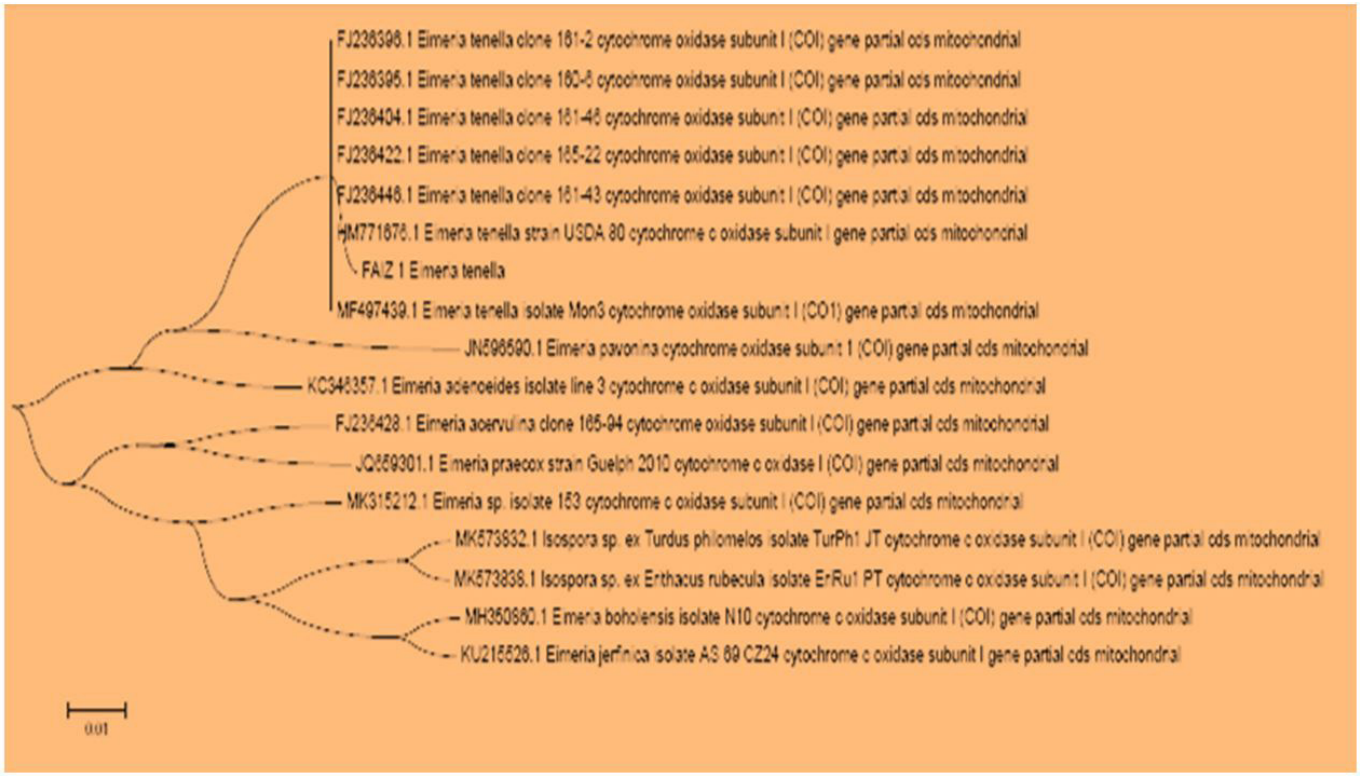

Figure 4: Phylogenetic analysis of *E. tenella*

Ascaridia galli

Ascaridia galli was found in the gastrointestinal tract of 52.17% (120/230) samples of Japanese Quail. Morphological points such as mouth, 3 strong lips (a single dorsal and two sub-ventral), club shaped esophagus without posterior bulb and lateral alae were observed. In male's Caudal alae was either poorly developed or absent. Spicules were almost equal in size, caudal papillae were relatively large and Pre-anal sucker was also present. The females were larger and thicker than males with characteristic dark interlacing ovary lying above the intestine. As this study was concerned with male specie, that's why we focused only on male worm for further morphological identification and comparison (Table 1).

Particulars	Balde Govind Hanmantrao (2019)	Sofi TA, F, A (2016)	Present Study
Body length	27-29	15-17	45-48
Max. Width	----------	0.7-0.9	0.4-0.6
Esophagus length	2.1-7.2	2.4	1.95-4.1
Esophagus width	----------	----------	0.29-0.42
Tail length	1.56-1.8	1.5	0.81-1.13
Spicule length	0.54-1.25	2.7 and above	2.56-2.71

Table 1: Comparative Characteristics (Measurements in mm) of *Ascaridia galli*

The parasites were found elongated, round, semitransparent and creamy white in color. Mouth was surrounded by three lips. Esophagus was found with no posterior bulb. These parasites were isolated from small intestines of Quail. Pictorial details of morphological identification of *Ascaridia galli* are shown in Figures 5a, 5b, 5c, 6a, 6b, and 6c.

Figure 5 *Ascaridia galli* adult worm 5a Mouth parts with three lips, one lips behind. 5b, Mid portion 5c, Tail region of female *Ascaridia galli* with pre-anal suckers. Figure 6, Morphological identification of *Ascaridia galli*. A Mouth parts with three lips, one lip behind, B Mid portion and C Tail region of female *Ascaridia galli* with pre-anal suckers. D indicates tail region of female worm where posterior region of interlacing ovary could be seen. E tail region of male worm showing spicule and prenatal sucker.

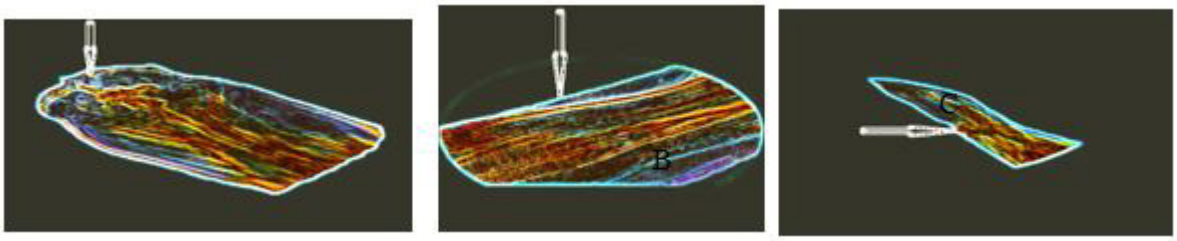

Figure 5: A, B and C showing the morphological feature of *A. galli*

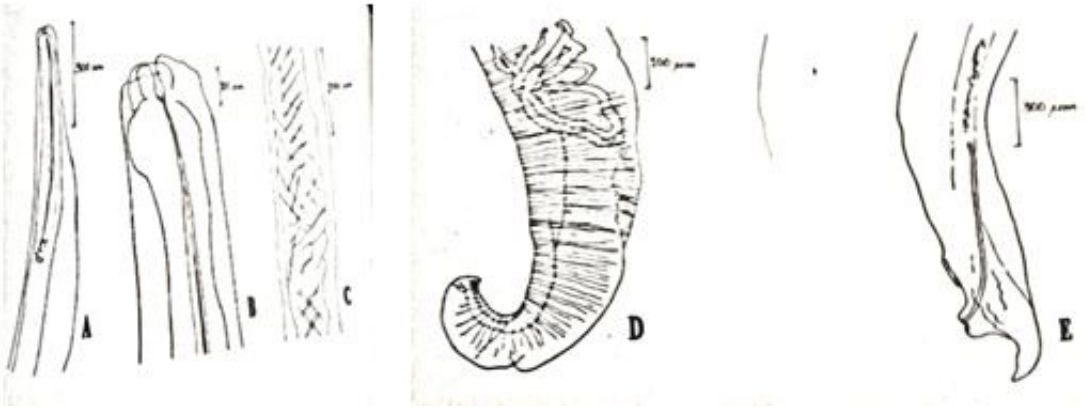

Figure 6: A, B, C, D and E showing morphological identification by *Camera lucida* drawing of *A. galli*

PCR based detection and phylogenetic analysis of Ascaridia galli

The small piece of *A galli* were further confirmed by PCR targeting mitochondrial fragment CO-1 gene (533bp**:** Figure 7 shows the result of amplified product of CO-1 gene of *Ascaridia galli* in 1.5 % agarose gel stained with ethidium bromide, visualized through trans-illumination device. The BLAST analysis revealed 90% genetic identity with *Anisakidae* and *Ascarididae* families that also fall in the order *Ascaridida*. In the current study the sequence obtained depicted 99% matching with that of *Ascaridia galli* of poultry. The phylogenetic tree was generated through Maximum Likelihood (ML) algorithm. Intra-specific and inter-specific pair wise distances of *Ascaridia galli* was analyzed by using complete deletion in the ML. Phylogenetic relationship of selected native *Ascaridia galli* was based on the maximum likely tree method (Figure 8). The nearest strain to our *Ascaridia galli* was *Ascaridia galli* GU 138670.1. Over all negligible distance was found between currently isolated sequence and other sequences of *Ascaridia galli* when analyzed.

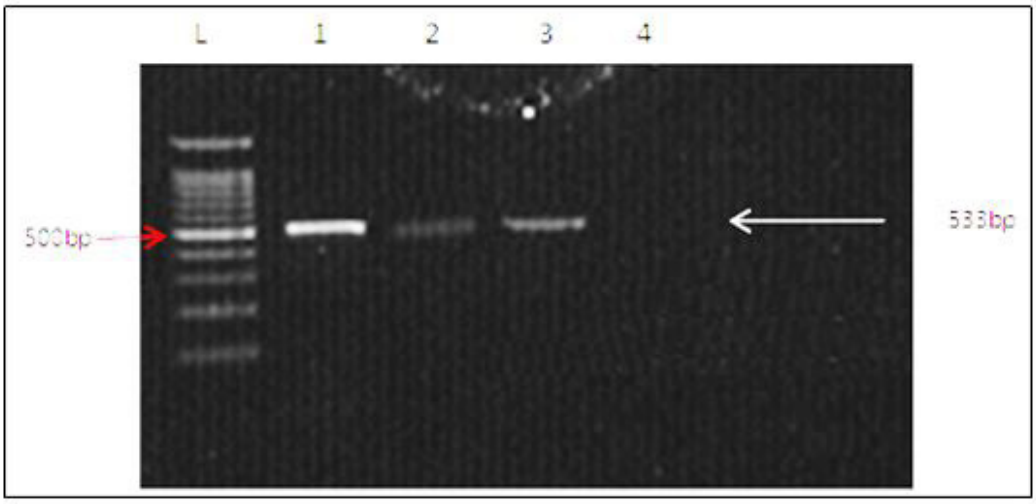

Figure 7: PCR (CO-1) product of *Ascaridia galli*. L: 100 bp apart molecular DNA marker; Line-1: Positive control; Lines-2 and 3: Positive sample; Line-4: Negative control

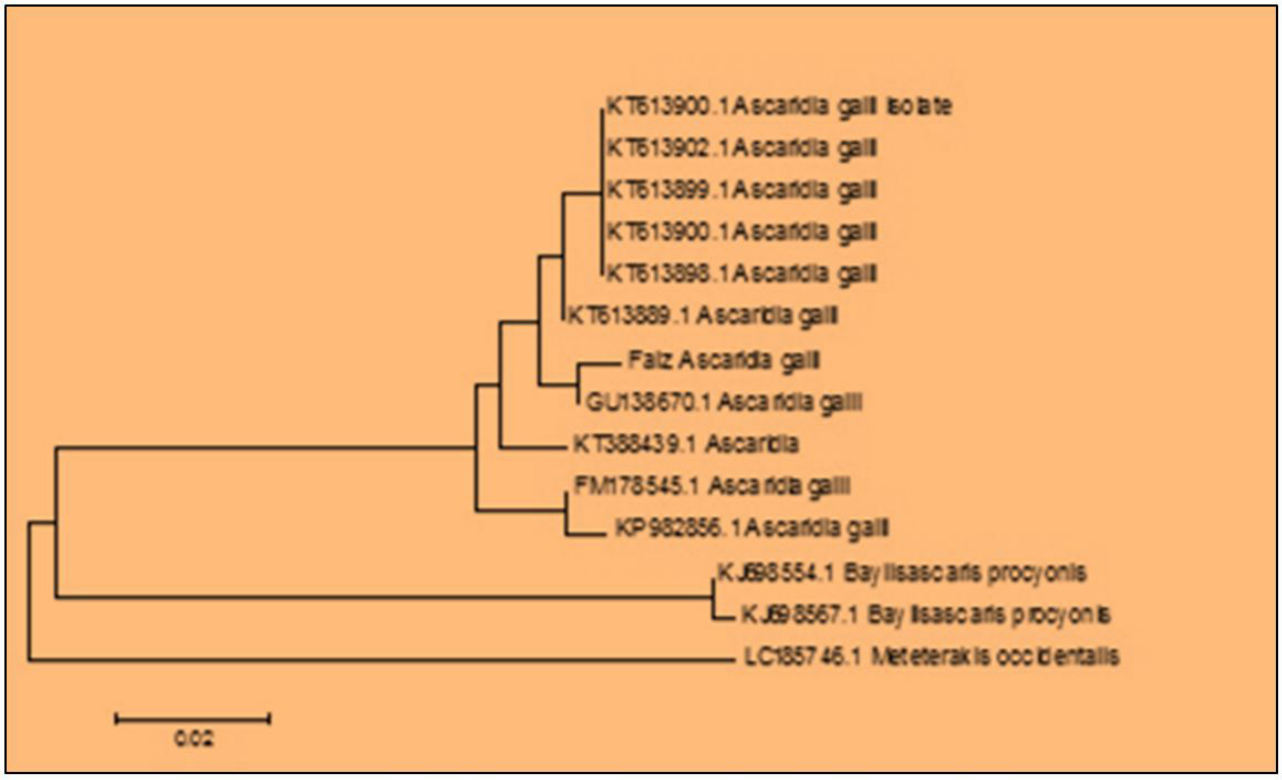

Figure 8: Phylogenetic Tree of *Ascaridia galli*

Discussion

Coccidiosis caused by *Eimeria* species is a global issue in poultry industry and commonly transmitted mechanically through movements of infected birds, personnel moving between pens, houses, or farms (Williams, 1999) [27]. The role of migratory birds in the spread of pathogens (virus, bacteria and parasites) is well established (Altizer et al., 2003) [28]. However, migratory strategies and behaviors potentially affecting transmission of infections are too much difficult to study (El-Ghany., 2019) [1]; (Pulgarín-R et al., 2019) [29]. The current study examined the dynamics of infection by protozoan parasite *Eimeria* (coccidian) through the annual cycle of a long distance migratory wild quail dispersing pathogens (Williams, 1999); Moller et al., 2011; Chapman et al., 2013) [27,30,31]. Results of the current study showed that Oocyst of *Eimeria* spp were present in 72.2% of fecal samples from migratory Japanese Quails. Finding of this study are in line with previous study (Mohammad, 2012) [20], which reported *E. tenella* in 49.9% Japanese quails in Mosul, Iraq. Another study also reported *E. tenella* in 52% in Japanese quails in Azerbaijan. The difference in the percentages might be due to management conditions, seasonal fluctuations in biotic factors and exposure of the migratory birds to anti-coccidial drugs (Nematollahi et al., 2009) [32]. The findings of the current study show that migratory quails may play a potential role in the dispersion of *Eimeria* spp. This study is the first study reporting *E. tenella* in quails in the study area.

Coccidia and *Isopora* are host specific parasites to chicken (Boughton, 1937) [33]. However, current study was successful to isolate *E. tenella* from migratory quails. Isolation of *E. tenella* from quails is in line with previous studies. (Nakai et al. 1992) [34] reported that *E. tenella* can sexually develop in the Embryo of Japanese quail. They cultivated *E. tenella* in Japanese quail embryos and complete oocysts were seen in the quail embryo after seven (7) days. Oocysts harvested from embryos were also sporulated. Furthermore, studied *E. tenella* isolated from infected chickens and *E. kofidi* isolated from young quails. Both *Eimeria* spp are host specific to chicken and quails, respectively. The authors reported that sporulated oocysts of both *Eimeria* spp developed to shizogone in nonspecific hosts. In another study Mathis & Mcdougald (1987) [11] reported that *E. tenella* equally infect chicken and quails. Chicken and quail hybrids were orally inoculated with chicken coccidia like *E. tenella*, *E. acervulina and E. maxima* and quail coccidian *E. bateri*. The chicken and quail hybrid were infected with chicken and quail coccidia passing oocysts in feces during normal latent period and control chicken and quails were severely infected.

The DNA extraction from oocysts of *E. tenella* is a challenge because of its thick resistant wall. Several methods like sonication , phenol chloroform method, enzyme digestion by a high pressure cell (Abrahamsen et al., 1995) [35], freezing and thawing, grinding with liquid nitrogen (Tsuji et al., 1999) [18] and grinding by glass beads (Fernandez et al., 2003) [36] have

shown no satisfactory results. In the current study, glass bead beating method was modified by replacing glass beads with steal beads which showed satisfactory results to get PCR quality DNA. It may be due to weight of the steal beads which successfully ruptured the resistant and thick oocyst wall to get PCR quality DNA. However, further evaluation of this DNA extraction method may be required.

Furthermore, mitochondrial fragment CO-I genes were targeted for identification of *Eimeria* species in the current study. Mitochondrial CO-I gene has most widely used genetic target in animal barcoding and found useful in species identification (Teletchea, 2010) [37]. The results of the current study suggest that the mitochondrial CO-I is useful for species identification and phylogenetic investigations in coccidian protozoan parasite. Studies on avian helminth parasites are important both from economic, zoonotic and parasitic point of view. A total 230 migratory quails were investigated in this study for the presence of nematodes, *Ascaridia galli.* The overall prevalence was 52.17%. Similar kind of study was also conducted by Mauricio Silva Rosa et al (2017) [38] in Brazil in which the prevalence of *Ascaridia galli* in quail was 20%. Gersonval (2018) [39] observed that 16.1% had mixed infection of *Ascaridia galli* Schrank (1778) in Japanese quails, *Coturnix coturnix japonica,* in Amazon region among 31 quails. The result (52.17%) of our study is near identical with that of Movessia et al (1994) [40] who reported prevalence of *A. galli* as 64.7 % in quails in Hungary.

Molecular characterization of *Ascaridia galli* including DNA quantification, sequencing and phylogenetic studies were performed in the present study of which were also conducted by cohorts (Katakam et al., 2010) [25]. They extracted DNA from larva of *Ascaridia galli* collected from chicken and performed the polymerase chain reaction-linked restriction fragment length polymorphism (PCR-RFLP). A 533-bp long region of the cytochrome C oxidase subunit 1 gene of the mitochondrial DNA was targeted and *Ascaridia galli* females were allocated to three different haplotypes. In the present study male *Ascaridia galli* was obtained from the GIT of quail, extracted the DNA, the 533 bp fragment exploited was same as that in the above study [41,42].

Conclusion

This study concludes that *E. tenella* may not be more host specific. It can parasitize other species of the birds like Japanes quail (*Coturnix coturnix japonica*). Molecular methods are best used for species identification of *Eimeria tenella* and *Ascaridia galli.* Furthermore, migratory Quails are potential risk factor for the spread of protozoal and nematodal infestation in local and commercial chicken.

Acknowledgement

We acknowledge molecular parasitology laboratory, department of parasitology, University of Veterinary and Animal Sciences (UVAS), Lahore, Pakistan for providing laboratory facilities and technical support during the current research.

Competing Interests

The authors declare that they don't have competing interests.

References

1. El-Ghany WAA (2019) A Comprehensive Review on the CommonEmerging Diseases in Quails. J World Poult Res 9: 160-74.

2. Teixeira M, Teixeira Filho W, Lopes C (2004) Coccidiosis in japanese quails (Coturnix japonica): characterization of a naturally occurring infection in a commercial rearing farm. Revista Brasileira de Ciência Avícola 6: 129-34.

3. Berto BP, Luz HR, Flausino W, Teixeira-Filho WL, Ferreira I, et al. (2011) Isosporoid coccidia (Apicomplexa: Eimeriidae) parasites of tanagers (Passeriformes: Thraupidae) from the Marambaia Island, Brazil. Pesquisa Veterinaria Brasileira 31: 798-805.

4. Rahman WA, Manap NH (2014) Descriptions on the Morphology of Some Nematodes of the Malaysian Domestics Chicken (Gallus domesticus) using Scanning Electron Microscopy. Malaysian Journal of Veterinary Research 5: 35-42.

5. Anonymous (2014) The Japanese quail.

6. Umar M, Hussain M, Murtaza G, Shaheen FA, Zafar F (2018) Ecological concerns of migratory birds in Pakistan: A review. Punjab University Journal of Zoology 33: 69-76.

7. Scott DM (1991) The time of day of egg laying by the brown-headed cowbird and other icterines. Canadian Journal of Zoology 69: 2093-9.

8. Lank DB, Butler RW, Ireland J, Ydenberg RC (2003) Effects of predation danger on migration strategies of sandpipers. Oikos 103: 303-19.

9. Mihaela N, Marina S (2014) Wild birds as potential vectors for pathogen dissemination on migration routes in the Danube Delta Wetlands. Int J Curr Microbiol App Sci 3: 890-7.

10. Latif AA, Fazal, Manzoor S, Maqbool F, Asghar A, et al. (2016) A Comparative Study on Prevalence of Coccidian Parasites in Broiler Chicken (Gallus gallus domesticus), Japanese Quail (Coturnix coturnix japonica) and Wild Pigeon (Columba livia). Pakistan J Zoo 48: 297.

11. Mathis GF, McDougald LR (1987) Evaluation of interspecific hybrids of the chicken, guinea fowl, and Japanese quail for innate resistance to coccidia. Avian Diseases 31: 740-5.

12. Baillie SR, Peach WJ (1992) Population limitation in Palaearctic-African migrant passerines. Ibis 134: 120-32.

13. Ondwassy HO, Okitoi LO, Obali MP, Simwa S, Wakhusama SW (2000) Epidemiology of helminth in indigenous poultry In: Western Kenya in de Jong and Mukisira (Editors), Testing of livestock technologies on smallholder mixed farms in Kenya, Research-extension-f, Encyclopedia of Volcanoes.

14. Kaufman PE (2007) External Parasites of Poultry 1.

15. Kaufmann J (1996) Parasitic infections of domestic animals: A diagnostic manual. Parasitology Today 12: 496.

16. Ali A, Khan AA (2019) Trapping techniques and lure (Bait) used in capturing of falcons in Punjab, Pakistan. Pakistan Journal of Zoology 52: 381-4.

17. Zajac A, Conboy G (1994) Fecal examination for the diagnosis of parasitism. Veterinary clinical parasitology 3: 164.

18. Tsuji N, Ohta M, Kawazu SI, Kamio T, Isobe T, et al. (1999) DNA Polymorphism of srRNA Gene among Eimeria tenella Strains Isolated in Japan. Journal of Veterinary Medical Science 61: 1331-3.

19. O' Horo M, Comstock A, Hoffmaster L, Hunter A, Jones R, ET AL. (2007) Fecal Examination Techniques for the Recovery of Parasite Ova in Large Animal. Vet Tech 442-3.

20. Mohammad NHE (2012) A Study on the Pathological and Diagnosis of Eimeria Species Infection in Japanese Quail. Basrah Journal of Veterinary Research 11: 318-33.

21. Carvalho FS, Wenceslau AA, Teixeira M, Matos Carneiro JA, Melo ADB, Albuquerque GR (2011) Diagnosis of Eimeria species using traditional and molecular methods in field studies. Veterinary Parasitology 176: 95-100.

22. Soulsby EJL (1983) Helminths, Arthropods and Protozoa of Domesticated Animals. The American Journal of Tropical Medicine and Hygiene 32: 906-7.

23. Rahman AW, Salim H, Ghause MS (2009) Helminthic parasites of scavenging chickens (Gallus domesticus) from villages in Penang Island, Malaysia. Tropical Life Sciences

Research 20: 1-6.

24. Folmer O, Black M, Hoeh W, Lutz R, Vrijenhoek R (1994) DNA primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates. Molecular Marine Biology and Biotechnology, 3: 294-9.

25. Katakam KK, Nejsum P, Kyvsgaard NC, Jørgensen CB, Thamsborg SM (2010) Molecular and parasitological tools for the study of Ascaridia galli population dynamics in chickens. Avian Pathology 39: 81-5.

26. Tamura K, Stecher G, Peterson D, Filipski A, Kumar S (2013) MEGA6: Molecular evolutionary genetics analysis version 6.0. Molecular Biology and Evolution 30: 2725-9.

27. Williams RB (1999) A compartmentalised model for the estimation of the cost of coccidiosis to the world's chicken production industry. International Journal for Parasitology 29: 1209-29.

28. Altizer S, Nunn CL, Thrall PH, Gittleman JL, Antonovics J, et al. (2003) Social Organization and Parasite Risk in Mammals: Integrating Theory and Empirical Studies. Annual Review of Ecology, Evolution and Systematics 34: 517-47.

29. Pulgarín-R PC, Gómez C, Bayly NJ, Bensch S, FitzGerald AM, et al. (2019) Migratory birds as vehicles for parasite dispersal? Infection by avian haemosporidians over the year and throughout the range of a long-distance migrant. Journal of Biogeography 46: 83-96.

30. Moller AP, Szép T (2011) The role of parasites in ecology and evolution of migration and migratory connectivity. Journal of Ornithology 152: S141-S150.

31. Chapman HD, Barta JR, Blake D, Gruber A, Jenkins M, et al. (2013) A selective review of advances in coccidiosis research. Adv Parasitol 83: 93-171.

32. Ahmad N (2009) Prevalence of Eimeria species among broiler chicks in Tabriz (northwest of Iran). Europe 4: 53-8.

33. Boughton C (2013) Notes on Arian Coccidiosis.

34. Nakai Y, Tsuchiya H, Takahashi S (1992) Cultivation of Eimeria tenella in Japanese quail embryos (Coturnix coturnix japonica). Journal of Parasitology 78: 1024-6.

35. Abrahamsen MS, Clark TG, White MW (1995) An improved method for isolating RNA from coccidian oocysts. Journal of Parasitology 81: 107-9.

36. Fernandez S, Pagotto AH, Furtado MM, Katsuyama AM, Madeira AMBN, et al. (2003) A multiplex PCR assay for the simultaneous detection and discrimination of the seven Eimeria species that infect domestic fowl. Parasitology 127: 317-25.

37. Teletchea F (2010) After 7 years and 1000 citations: Comparative assessment of the DNA barcoding and the DNA taxonomy proposals for taxonomists and non-taxonomists. Mitochondrial DNA 21: 206-26.

38. Mauricio (2017) Enteroparasites of the families Eimeriidae and Ascarididae in Japanese quail (Coturnix japonica) in the metropolitan region of the Cuiabá River Valley, State of Mato Grosso, Brazil.

39. Gersonval LSM, Cavalcante DG, Oliveira JBS, (2018) Parasitic profiling of Japanese quails (Coturnix japonica) on two farms with conventional production system in the Amazon region. Pesquisa Veterinaria Brasileira 38: 847-51.

40. Movsessian SO, Pkhrikian LV (1994) Reciprocal infection of quails and hens with the nematodes Ascaridia galli (Schrank, 1788) and Heterakis gallinae (Gmelin, 1790): single and mixed infections parasite hung; 27; 83-85. Endocrinology and Metabolism Clinics of North America, 23 (Physiology of the hypothalamic-pituitary-adrenal axis in health and dysregulation in psychiatric and autoimmune disorders.) 451-66.

41. Luz HR, Ferreira I, Ventura PE (2005) Study of the prevalence of ecto parasite seized in Rio de Janeiro In: Brazilian Congress of Ornithology, 12.Belém - PA. Annals. Belém, Brazil.

42. Sofi TA, FA (2016) Morphology and Prevalence of Some Helminth Parasites in Gallus domesticus from Gurez Valley of Jammu and Kashmir, India. J Fisheries Livestock Production 4: 1982.