

Prevalence and Pathology of *Heterakis gallinarum* in Free Roaming Domestic Chicken of Kashmir Valley

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Context of Research

- Free ranging domestic fowl are very susceptible to parasitic infections owing to their exposure to parasites during their roaming in the backyards in search of food.
- *Heterakis gallinarum* infection is usually subclinical but it may function as a vector for *Histomonas meleagridis* (black head) which induces severe pathological lesions in the gut and liver leading to high mortality rates in susceptible hosts (Gibbs, 1962, Springer et al., 1969; Lund & Chute, 1972).
- The present study was designed to have an idea about the prevalence of the nematode *Heterakis gallinarum* in free ranging chicken of Kashmir Valley and to study the extent of pathology caused by it to the caecum and liver in both monoinfection and coinfection with *Histomonas meleagridis* to design some strategy to curb this fatal association of these parasites which causes great economic losses to our backyard poultry industry.

Methodology

- **Prevalence:**

Sample size - 478 domestic chicken (*Gallus g. domesticus* L., 1758), 243 males and 235 females, weighing between 1 - 2.5 kg each collected from the different localities covering almost entire kashmir valley.

Individual clinical evaluation and euthanization according to Zander et al.(1997).

Nematodes collected, rinsed in normal saline (0.85%) , fixed in hot 70% alcohol and then counted using a stereoscopic microscope.

The nematodes cleared in lactophenol, mounted in glycerine jelly, photographed and identified following Vicente et al. (1995).

Prevalence was calculated as a percentage of the host population infected at a point in time (Thrusfield, 1995).

Mean intensity was calculated as number of parasites per infested bird.

- **Faecal Examination:**

- The flotation technique (Mc Master)
- Worm eggs were identified using the keys described by Thienpont *et al.* (1986).
- Faecal egg counts (FECs) were undertaken within 24 hours by a modification of the McMaster technique with a sensitivity of 50 eggs per gram of faeces (Thienpont *et al.*, 1986).

- **Impression smears:**

Impression smears from the liver and scrapings from the caeca and intestine were stained with Giemsa and Gram's stain.

- **Pathological studies:**

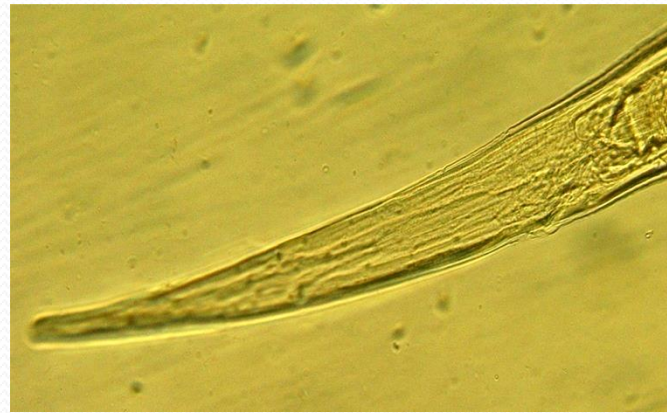
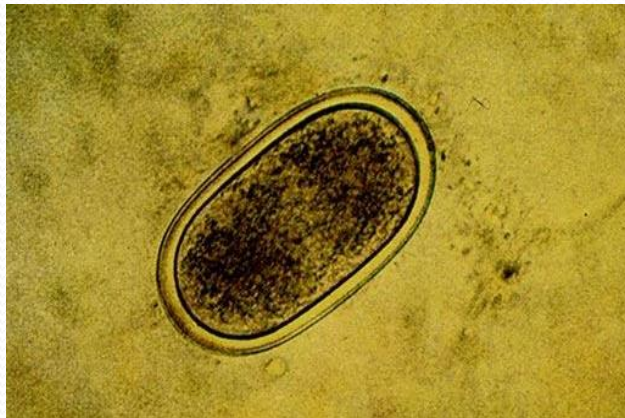
For histopathological studies, fragments of the parasitized caecae and liver fixed in formalin and then routinely processed (Bancroft and Gamble, 2002) for paraffin embedding. 5µm sections were cut and stained with hematoxylin and eosin (H&E).

Mc Manus periodic acid Schiff (PAS) stain was used to demonstrate protozoan inclusions in the sections.

Grocott's stain was used to differentiate with fungal elements. Micrographs were obtained using digital microscope model BX60F-3, Olympus Optical Co. Ltd. (Tokyo, Japan), fitted with the Olympus camera model DP12.

Results

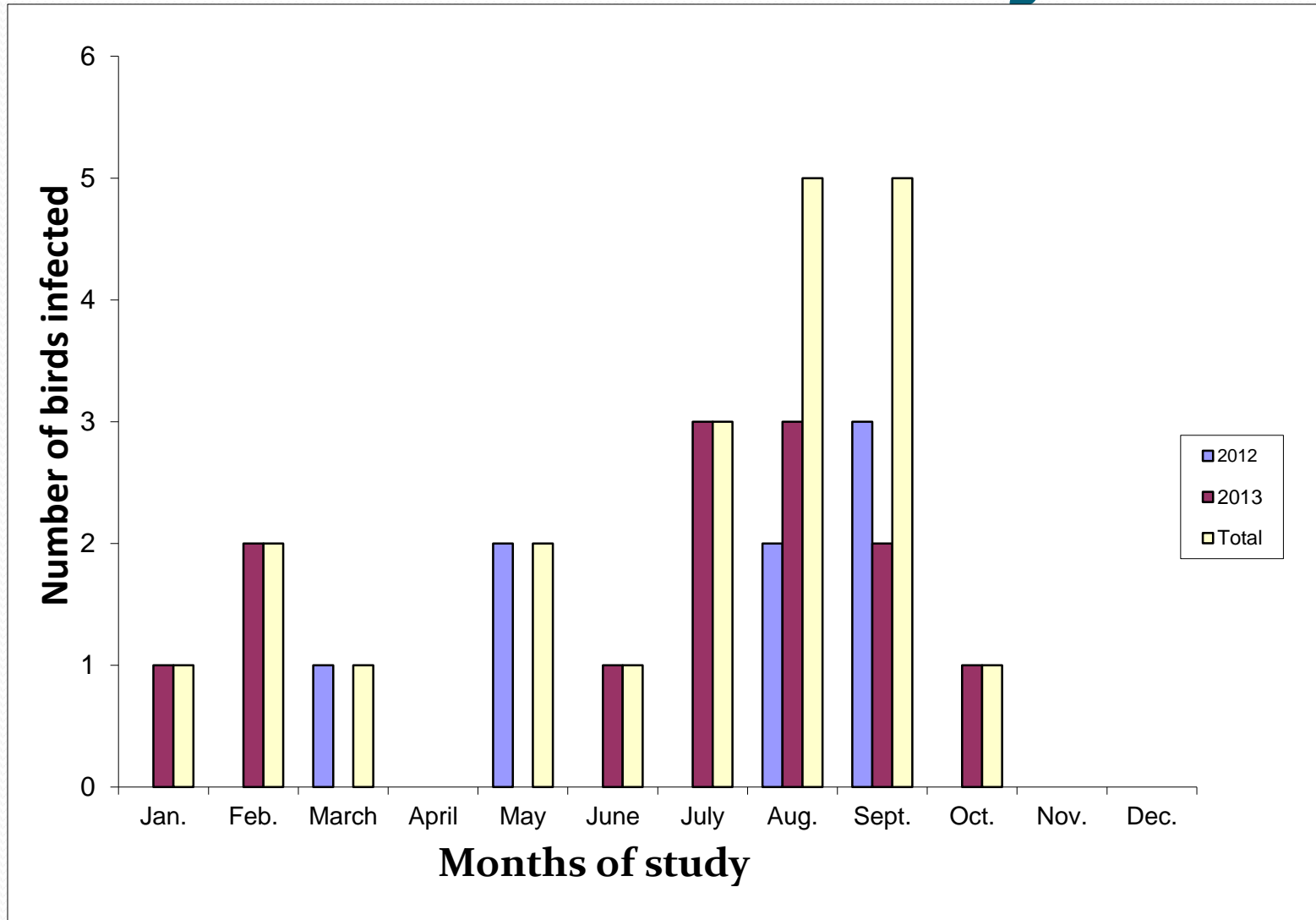
Identification



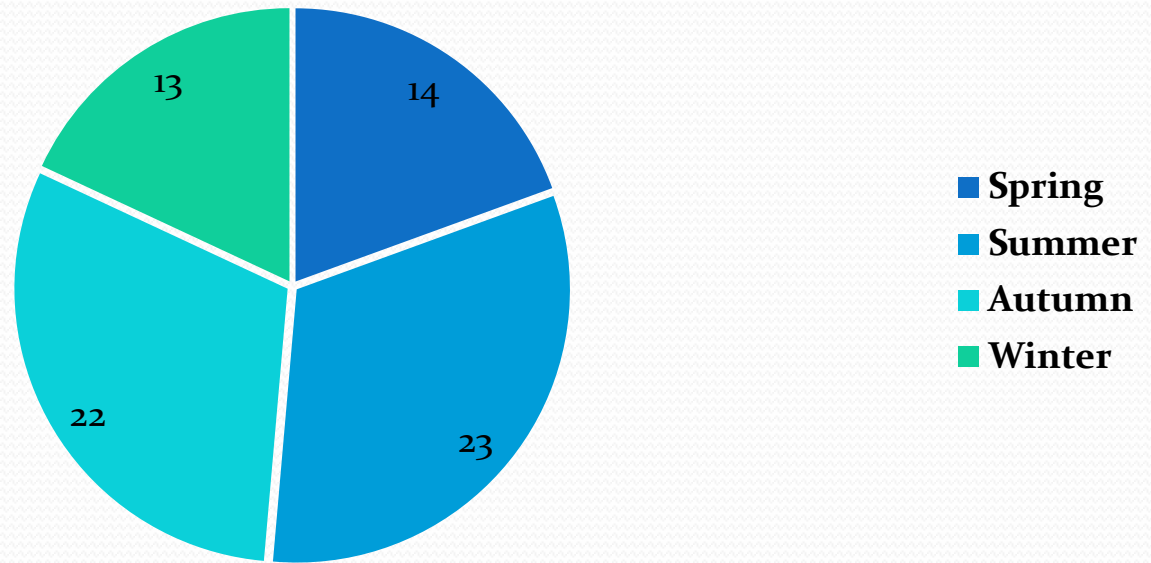
Prevalence

- Prevalence rate for 1st Year of study: **3.43%(8/233)**
- Prevalence rate for 2nd Year of study: **5.3%(13/245)**
- Overall prevalence rate for two year study (Jan 2012 to Dec 2013): **4.39% (21/478)**
- Range of intensity of worms: **02 – 55**
- Mean intensity of infection: **20 ± 2**
- Faecal Egg Count: **<40 eggs per gram of faeces**
- Only two cases of *Heterakis gallinarum* co-infection with *Histomonas meleagridis* were reported, one in September 2012 and other in August 2013 giving overall prevalence of coinfection only **0.42%**.

Month wise Occurrence of *Heterakis gallinarum*



Season wise Mean intensity of infection



Reason: Decreased resistance to infection and increased availability of intermediate host (Earthworms) due to high temperature and more rainfall in Summer and autumn (Especially August – September)

Pathology

- **Monoinfection:**

Histological findings in the ceca during monoinfection of *Heterakis gallinarum* has shown:

- Presence of adults in the lumen along with cellular debris.
- larvae penetrating the epithelium of cecum has been reported (Sang-Ik Park¹ and Sung-Shik Shin,2010) but could not get those sections in this study.
- Intense chronic diffuse inflammatory processes with mononuclear and polymorphonuclear (heterophils) leucocyte infiltrations extending upto submucosa.
- Mucosal erosion with parasites and cellular debris in the transverse section.
- no gross lesions seen in the caeca and liver.

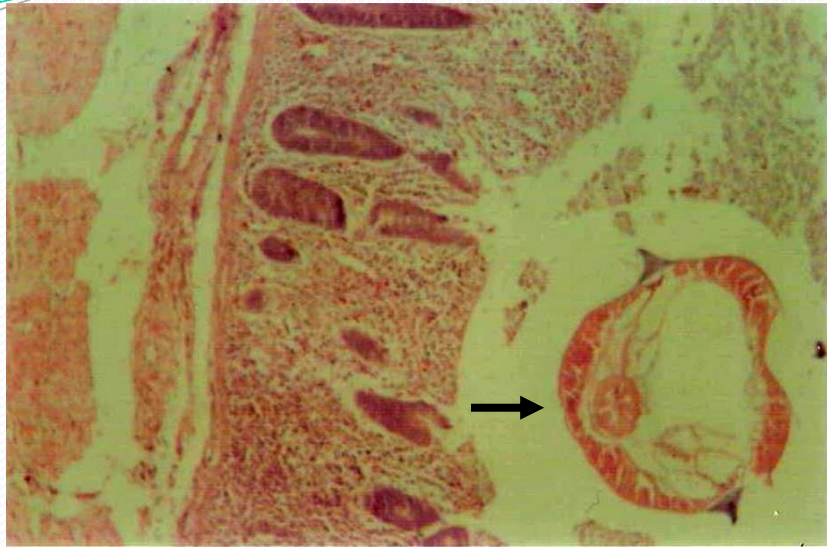


Figure 1: Photomicrograph of caecum of domestic fowl revealing adult *Heterakis gallinarum* in the lumen, Note the mucosal denudation and cellular debris in lumen H&E 40X

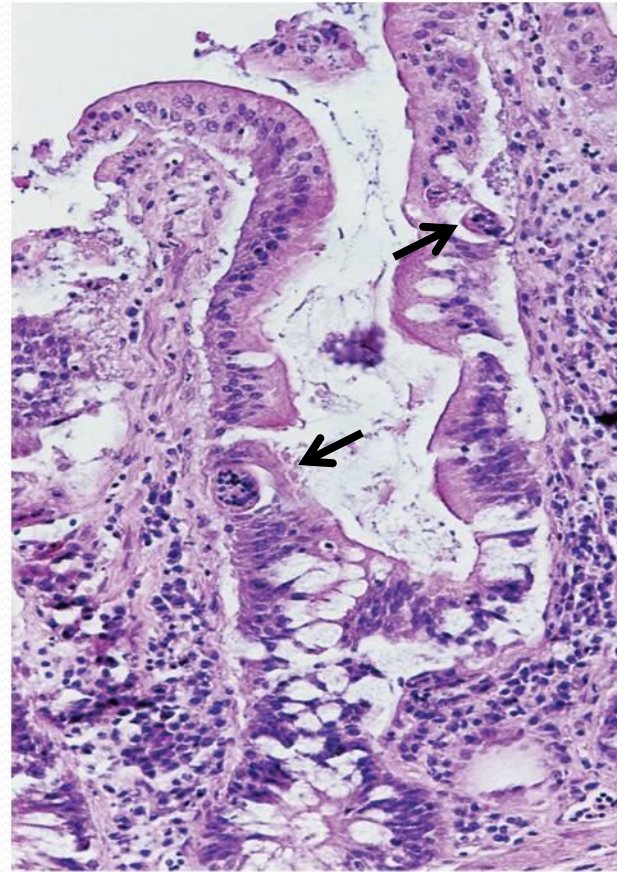


Figure 2: Photomicrograph revealing larvae (Arrows) of *Heterakis gallinarum* in the epithelium of caecum from domestic fowl. H&E 40X. (Courtesy - Sang-Ik Park¹ and Sung-Shik Shin, 2010)

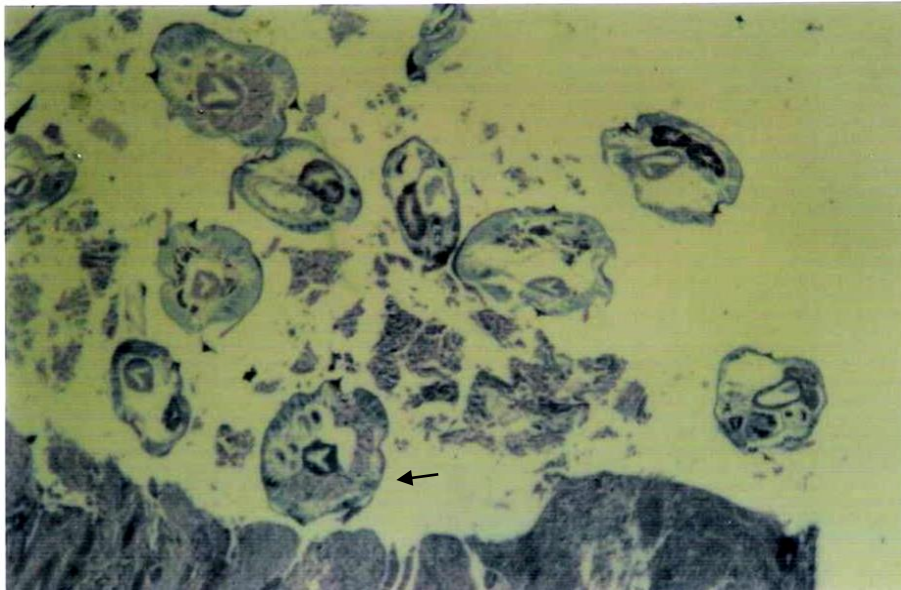


Figure 3: Photomicrograph of caecum from domestic fowl revealing multiple sections of *Heterakis gallinarum* in the lumen. Mucosal denudation is also evident. T.B. 10X

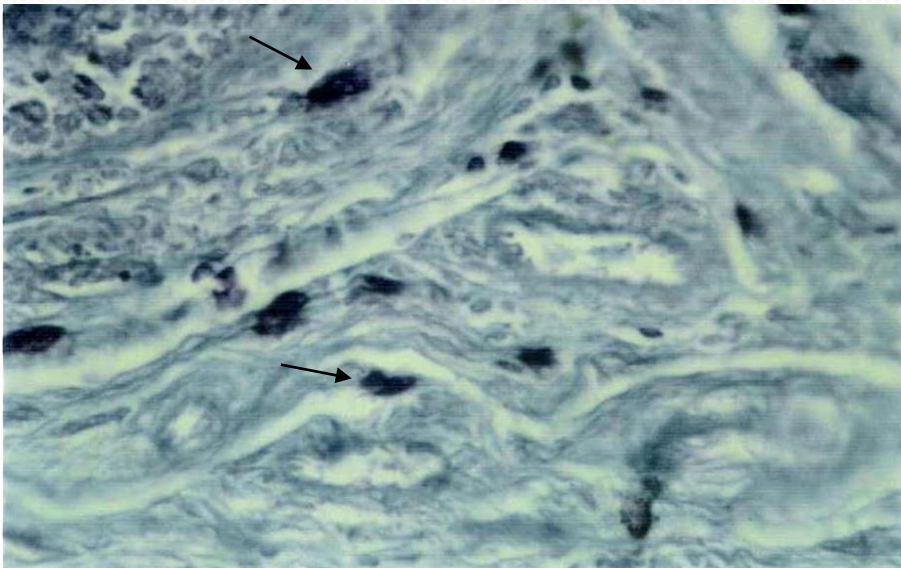


Figure 4: Photomicrograph of section of caecum of domestic fowl infected with *Heterakis gallinarum* revealing presence of mast cells. T.B. 100X.

- **Co infection:**

Histological studies of the liver and ceca of domestic fowl during co-infection of *Heterakis gallinarum* and *Histomonas meleagridis* has revealed:

Liver:

- Enlarged brilliant greenish livers with numerous round to oval, circumscribed lesions ranging from 0.5 to 1.5 cm in diameter and extending deep into the parenchyma.
- Extensive infiltration of lymphocytes, mononuclear cells and scarce heterophils.
- Hepatocytes towards the periphery of the lesions contained punched out ovoid bodies which stained positive and became visible with PAS (Periodic Acid Schiff) stain.

Cecum:

- Microscopic examination of cecal lesions showed hyperemia, ulceration and infiltration of heterophils, lymphocytes and macrophages extending from mucosa to the muscular layer .
- Sloughing off of the epithelium and lumen packed with fibrin, red blood cells and tissue debris.



Figure 5: Liver of Chicken infected with *Histomonas meleagridis* showing numerous round to oval circumscribed lesions resembling a bull's eye target

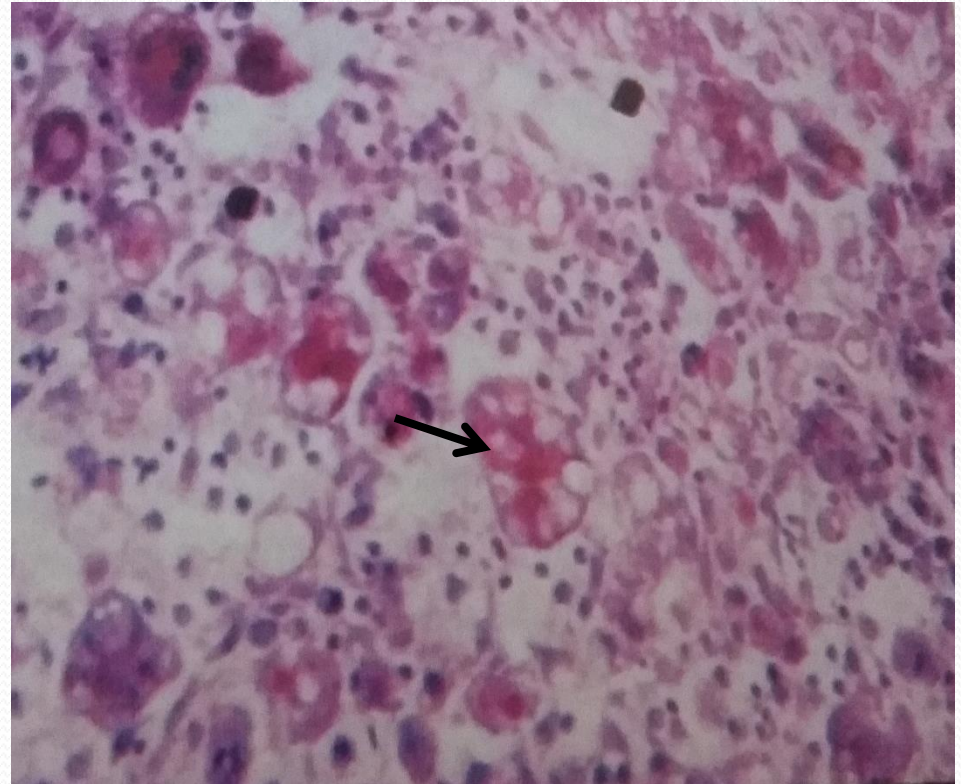


Figure 6: T.S of Liver of Chicken infected with *Histomonas meleagridis* showing punched out vacuolar bodies within hepatocytes (HE x 400)

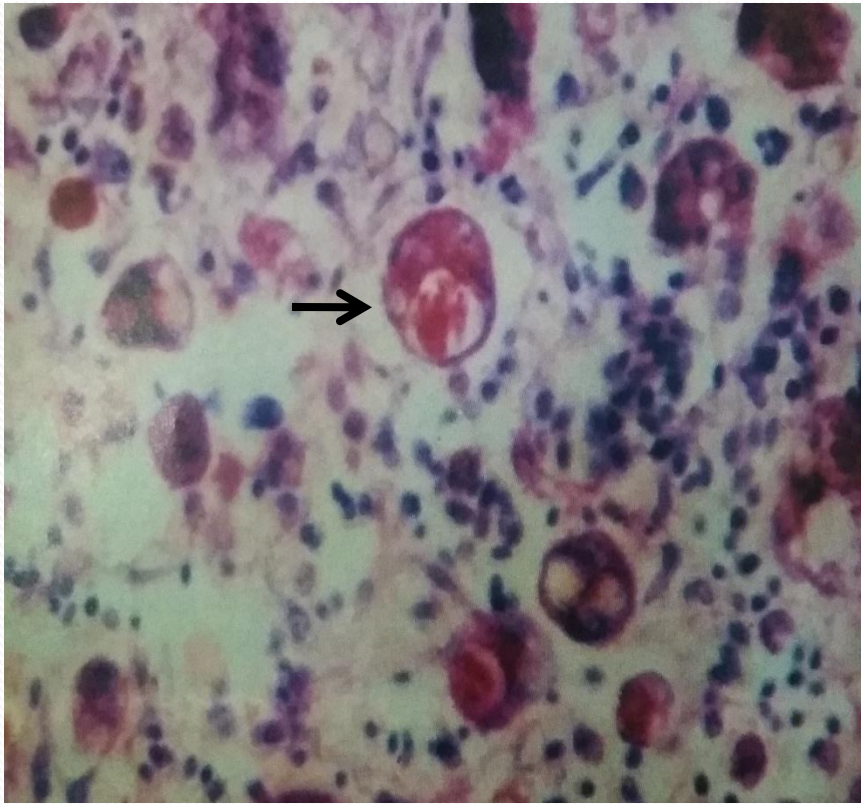


Figure 7: Ovoid bodies representing *Histomonas meleagridis* in the hepatocytes of chicken visible with PAS stain (x400)

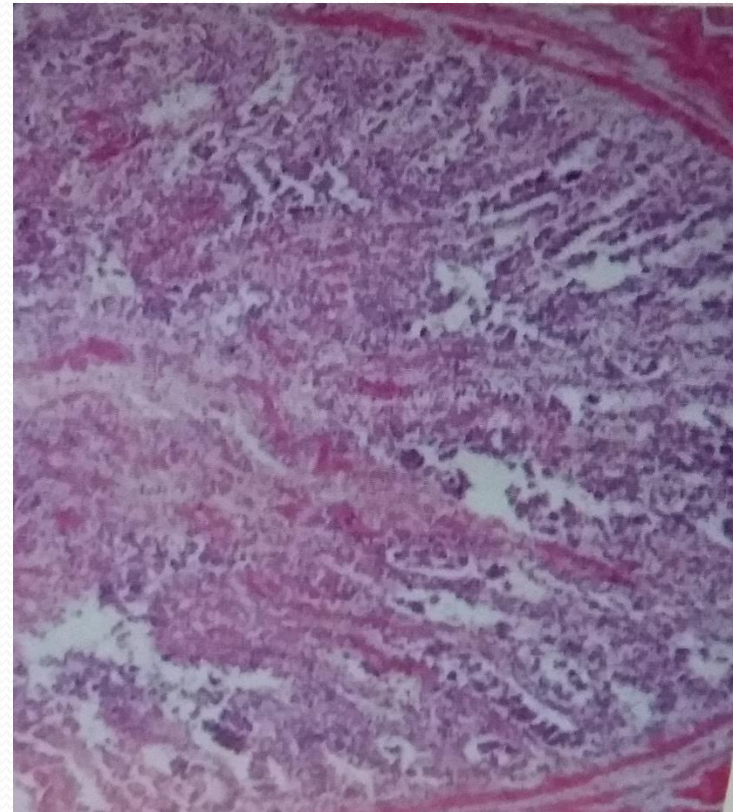


Figure 8: Caeca of chicken coinfecting with *Heterakis gallinarum* and *Histomonas meleagridis*. Note plugging of lumen with cellular debris, inflammatory cells and fibrin (HE x40)

Conclusion and Recommendation

- *Heterakis gallinarum* is comparatively harmless nematode but with great potential to cause severe pathological changes in the tissues during coinfection with *Histomonas meleagridis* especially in the warm and wet seasons.
- Investigations are thus advisable to know in detail immunological responses of the chicken intestine to the mono infection and co infection *Heterakis gallinarum* with *Histomonas meleagridis* to understand the extent to which this fatal association of the parasites may influence health status and productivity in free roaming backyard chicks which form the backbone of rural poultry industry of Kashmir valley .
- Possible existence of lateral transmission for *Histomonas meleagridis* without the possible role of *Heterakis gallinarum* ovum vectors needs to be investigated as during the study we come across one dead bird with severe infection of *Histomonas* without any *Heterakis* worm recovered from the cecum.



THANK YOU