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Histopathological studies of leptospiral isolates from Andhra Pradesh

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Abstract
Leptospirosis is a global important re-emerging disease of both humans and animals. The disease is a common zoonosis, acquired by exposure to infected animals or contaminated soil and water. Though the leptospira are potentially lethal, often undiagnosed. In the present study, histopathological studies were conducted in guinea pigs inoculated with leptospiral isolates recovered from sheep and pigs. Histopathological studies of liver and kidneys revealed congestion, hemorrhages, vacculation and infiltration of mono nuclear cells. The changes observed were due to Leptospirosis in liver and kidneys of guinea pigs. Re-isolation of leptospira could be made from cortical part of kidney and liver of guinea pig.

Keywords: Histopathology, leptospira and leptospirosis, hemorrhage, glomerular infiltration

Introduction
Leptospirosis is a bacterial zoonosis that is common worldwide. Organisms are shed in the urine of infected animals, including rodents and domesticated animals which may not show signs of disease. Humans also usually become ill after contact with infected urine or through contact with water, soil or food that has been contaminated. In animals, the clinical signs of Leptospirosis are often related to kidney disease, liver disease or reproductive dysfunction. In humans, many cases are mild or asymptomatic and go unrecognized. In some patients, the illness may progress to kidney or liver failure, aseptic meningitis, life threatening pulmonary hemorrhage and other syndromes.
Leptospirosis is considered as one of the major diseases that affect the reproductive systems, primarily causing abortions, infertility, still births in animals. It is one of the main causes of low live stock productivity both nationally and globally [1]. The lesions caused by leptospira species are observed mainly in the kidneys where the organism arrives via the blood stream. In the kidneys, the species multiplies, causing tubulo interstitial lesions [2, 3]. In the leptospireamic phase, the liver is the first site to be reacted by the organism where it causes necrosis liver cells, intra hepatic cholestasis resulting in decreased liver bilirubin excretion [4, 5].

Materials and Methods:
Pathogenic studies were carried out using guinea pigs weighing 150-250 grams in the laboratory. Cultures of leptospiral isolates were inoculated intra peritoneally into guinea pigs. The guinea pigs were kept under observation. Blood samples (0.5ml) were collected from heart after 12hrs of post inoculation and inoculated into the fresh EMJH medium for re-isolation. Guinea pigs were sacrificed, liver and kidney tissues were collected in 10 percent formalin for histopathological studies.

Histopathology
Liver and kindly tissue sections were prepared by fixing in 10 percent neutral buffered formalin for 12hrs followed by dehydration by immersing in increasing concentration of ethanol 70 percentage, 90 percentage and 1,000 percentage and then xylene before embedding in paraffin wax. The sections were then stained with haemotoxylin and eosin for examination.

Results
On necropsy, gross pathologically liver was haemorrhagic, frazile and enlarged. Histopathological examination of liver collected from infected guinea pigs revealed congestion, degeneration vacuolation, sinusoidal hemorrhages, perivascular mono nuclear infiltration in hepatic cells.

In majority of the cases bi-nucleated hepatic cells were prominent (fig.1).

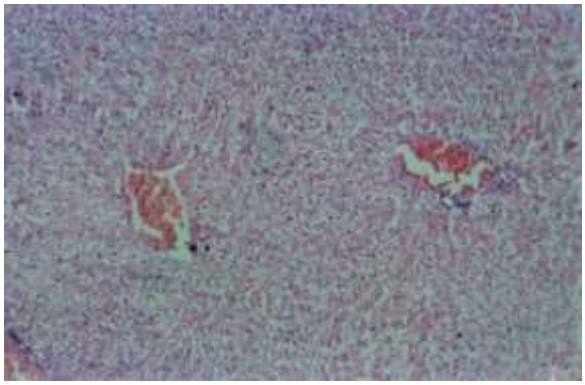
On gross pathology, the kidneys are hyperemic with petechial hemorrhages. Histopathological examination of kidneys collected from infected guinea pigs revealed focal areas of congestion, glomerular congestion, inter tubular hemorrhages, peri glomerular infiltration of neutrophils and monocytes, inter tubular infiltration of mononuclear cells i.e. interstitial nephritis, vacculation and inter tubular fibrosis were observed during the study (fig.2).

Discussion

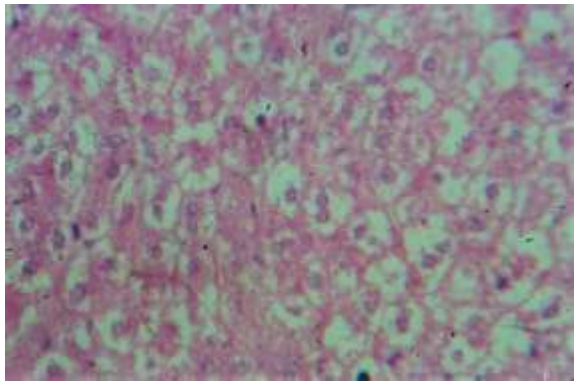
The present findings of gross pathological changes in the liver were in agreement with the observation of earlier workers (Semenovich *et al*, 1987; Everard *et al*, 1983; Venugopal and Ratnam, 1989) [6-8]. This type of histopathological changes in the liver was reported earlier by Brito de *et al*, 1967 [9] and Zaki and Spiegal, 1998 [10].

Similar type of histopathological lesions in kidneys of present study were also recorded earlier by Penna *et al*, 1963 [11], Arean, 1962 [12] and Zaki and Spiegal, 1998 [10].

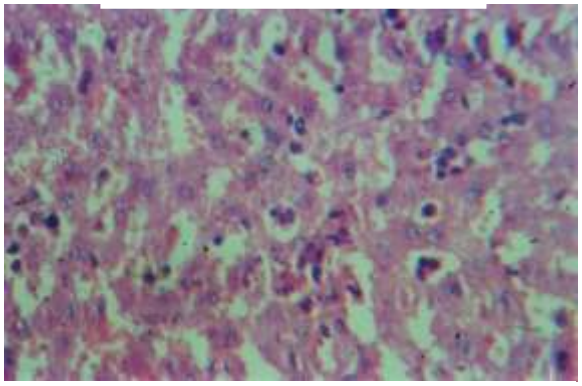
The lesions observed in the liver and kidneys were probably due to multiplication of leptospires in the leptospiraemic phase, impairing productive and reproductive performance of the animals.



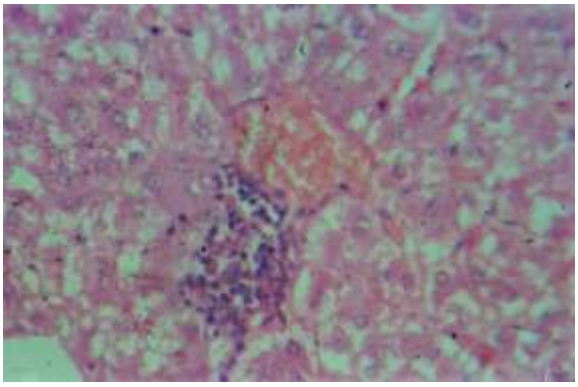
Severe congestion H & E X 70



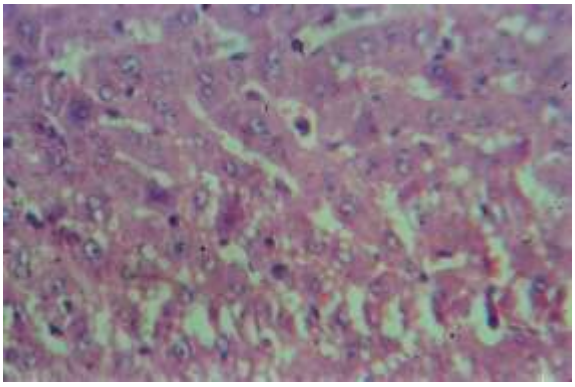
Severe degenerative changes in hepatic cells with vacuolation H & E X 280



Dilatation of sinusoidal space with haemorrhages and infiltration of neutrophils and eosinophils H & E X 280

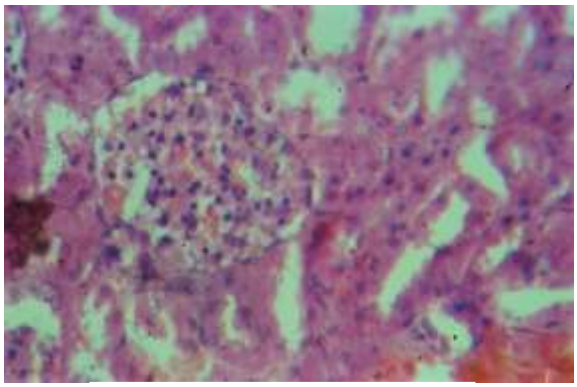


Perivascular, mononuclear aggregation H & E X 280

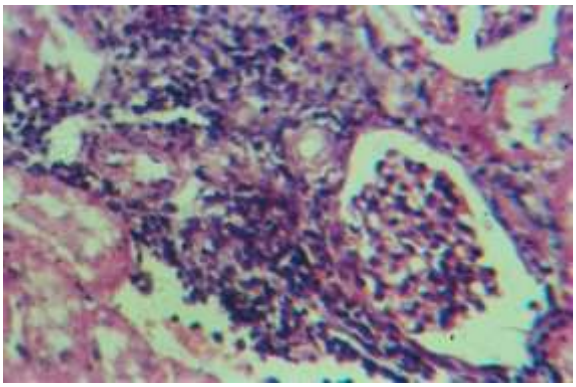


Several binucleated hepatic cells H & E x 280

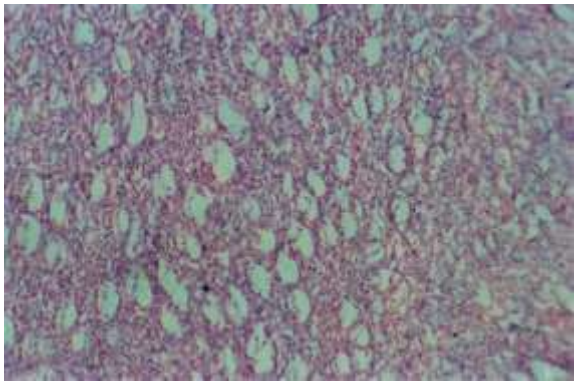
Fig 1: Histopathology - liver of Guinea pigs infected with leptospira



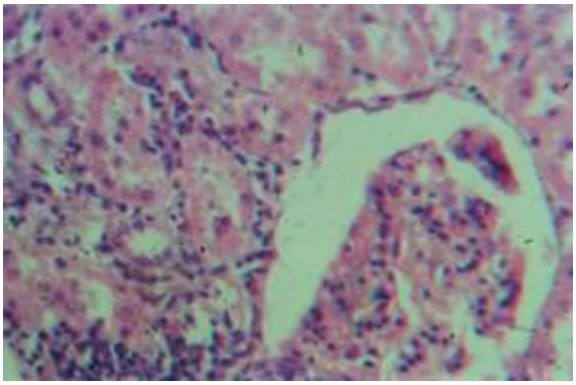
Glomerular H & E X 280



Periglomerular and intertubular infiltration H & E X 280



Cystic dilatation of tubules H & E X 70



Intetubular infiltration of mononuclear cells H & E X 280

Fig 2: Histopathology – kidney of guinea pig infected with leptospira

Conclusion

In conclusion, the present study deals with histopathological examination of liver and kidneys of guinea pigs infected with leptospiral isolates from sheep and pigs to know the changes in these organs indicative of Leptospirosis.

Acknowledgments

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