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THE DISTRIBUTION OF LEPTOSPIRES IN THE KIDNEY TUBULES OF SOME BRITISH WILD MAMMALS

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Abstract: Histological examination of 69 pairs of infected kidneys from 12 species of Rodentia, Lagomorpha, Carnivora and Insectivora revealed that leptospires were confined mainly to the proximal and distal convoluted tubule, were less often found in the thick loop of Henle and only rarely in the collecting duct. On no occasion were the organisms present in the thin loop of Henle. Preliminary observations on the relationship of leptospires to tubule epithelium indicate some degree of physical attachment. It is suggested that the avoidance of the thin loop of Henle might be a reflection of its structural properties.

INTRODUCTION

Many leptospiral serotypes have been isolated from the kidneys of wild carrier mammals. Despite the fact that many authors e.g. Abdulla *et al.*,¹ McGowan and Karstad,² have drawn attention to the interstitial nephritis which is a common feature of the kidneys in these animals, there is a lack of information on the actual location of leptospires within the renal tubule. Most studies fail to reinforce isolation techniques with histologic examination and in studies relying on kidney sections as the main evidence in epidemiological studies, e.g. Broom,³ the precise tubule location is never stated.

In a discussion of the role of glucose in stimulating the growth of serotype *pomona* and nine other serotypes, Ellinghausen⁴ states that the reason leptospires reside in the renal proximal tubule might be because in that section glucose is first excreted and then resorbed. This is one of the few occasions when the actual kidney site is mentioned and since Ellinghausen was referring to comments by Fulton and Spooner⁵ on the presence of glucose in the rat renal tubule he probably had the rat in mind.

MATERIAL AND METHODS

During our studies on leptospires in wild mammals in the British Isles (Twigg, Cuerden and Hughes⁶ and unpublished

data) we routinely sectioned all kidneys even if leptospires were not isolated. In nearly every case the kidneys were removed within 10 min of death, cut into 2 mm slices and fixed in 10% buffered formalin. They were sectioned at 5 μ m and silver-stained by the Levaditi or Warthin-Starry technique. The observations presented here are the result of an analysis of leptospire distribution in 69 pairs of infected kidneys from 12 species of Rodentia, Lagomorpha, Carnivora and Insectivora.

RESULTS

Localisation of leptospires

The localisation data are presented in Table 1 but some additional comments are needed to amplify the summary. Where both proximal and distal convoluted tubule have colonies of leptospires, all sections of these two regions show the organisms, indicating that the bacteria utilise all of that part of the tubule. The one exception to this is the rabbit where leptospires are found mainly in the proximal tubule and more rarely in the distal.

Without exception, in these species, there is no evidence for colonies in the thin loop of Henle and only two examples of colonies in the collecting duct region. Colonies have been recorded in the latter zone only when it was considered there was sufficient evidence for

TABLE 1. Distribution of leptospires in the renal tubule.

Species	Proximal convoluted tubule	Thin loop of Henle	Thick loop of Henle	Distal convoluted tubule	Collecting duct	Lesions
RODENTIA						
<i>Apodemus sylvaticus</i> (L.) (Long-tailed Field Mouse)	++	—	—	++	—	
<i>Clethrionomys glareolus</i> Schr. (Bank vole)	++	—	—	++	+	
<i>Microtus agrestis</i> (L.) (Short-tailed vole)	++	—	—	—	—	
<i>Mus musculus</i> (L.) (House mouse)	++	—	++	—	—	
<i>Rattus norvegicus</i> Berkenhout (Brown rat)	++	—	++	++	—	
LAGOMORPHA						
<i>Lepus europaeus</i> Pallas (Brown hare)	++	—	—	—	—	
<i>Oryctolagus cuniculus</i> (L.) (Rabbit)	++	—	—	++	—	
CARNIVORA						
<i>Vulpes vulpes</i> (L.) (Red fox)	++	—	—	—	—	+
<i>Mustela nivalis</i> (L.) (Weasel)	++	—	+	++	—	
<i>Mustela erminea</i> (L.) (Stoat)	—	—	—	++	—	
INSECTIVORA						
<i>Erinaceus europaeus</i> (L.) (Hedgehog)	++	—	++	++	+	
<i>Sorex araneus</i> (L.) (Common shrew)	++	—	++	++	—	

(++) denotes presence and (—) absence of leptospires and lesions

considering them to be resident rather than transitory organisms in the urine flow.

Density of leptospires in tubules

The number of infected tubules in an animal may vary from one or two to almost every tubule. In the former case the density of organisms within a tubule section is usually low with the bacteria forming a thin lining to the lumen, closely applied to the epithelium: the same condition may also be seen, however, in some tubules of a kidney with a high degree of tubule involvement. In other cases the leptospires may form a denser mass in the form of a thick rope around the lumen but with the centre of the lumen clear (Figure 1). In the most extreme case the entire lumen is occluded

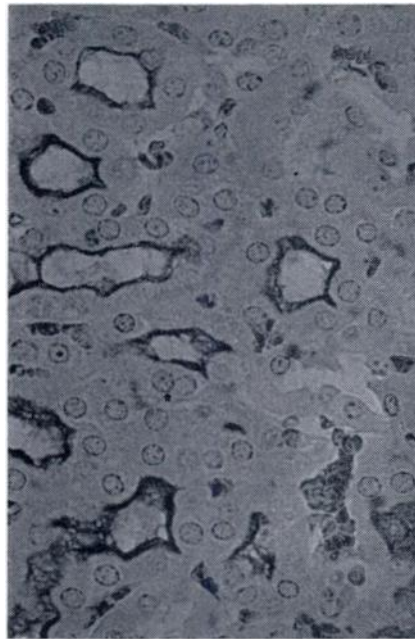


FIGURE 1. Leptospires in the renal tubules of the Long-tailed Field mouse, *Apodemus sylvaticus* (L.) stained by the Warthin-Starry silver method. Points of possible attachment to the renal epithelium may be seen.

by a tangled mass of leptospires. All stages may be seen in a kidney from one animal.

Relationship of leptospires to tubule epithelium

There are indications in this material that leptospires are embedded in the wall of the tubule in positions closely related to the cell walls separating adjacent epithelial cells. Lightly infected tubules are usually the best material for illustrating this but it may be seen in tubules with dense colonies. In a tubule section the leptospire rope or thin lining appears to have part of some organisms projecting at right angles into the epithelium (Figure 1). In a section three to five such points of possible attachment may be seen. Occasionally the attachment may penetrate as far as the base of the epithelial cells at one point or even at two points. In a few cases the relationship of such possible attachments to an intercellular position is not apparent and many smaller attachments can be seen. These observations are based on the light microscope and may be modified when our current studies using the electron microscope are completed.

DISCUSSION

In this material the proximal convoluted tubule is colonised in 11 of the 12 species and the distal in eight species with the thick segment of the loop of Henle having colonies on five occasions. The thin loop of Henle has no trace of leptospires in any of these species.

In mammals all the glucose, most salts, and some water is absorbed in the proximal tubule and the remaining water is absorbed in the loop of Henle and the distal convoluted tubule. Physiologically there is considerable difference in the environment between the two convoluted tubules, yet in an animal which is carrying only one serotype both parts may be colonised equally. Colonies in both zones appear to be permanent, as do those in the thick loop of Henle, but where groups

of leptospire are seen in the collecting ducts these sometimes lack points of attachment and are presumably moving in the urine flow.

In experimental situations animals experience acute leptospirosis with evidence of various kidney lesions before survivors proceed to a chronic state and eventual recovery.^{4,7} In the wild species studied in the British Isles and elsewhere many species function as carriers for varying periods of time. In none of these, except the fox, have any lesions been seen. In none of the many kidneys examined have intracellular leptospire been positively observed, such as can be a feature of experimental leptospirosis. In laboratory infec-

tions of hamsters with serotype *pomona* Miller and Wilson⁷ pointed out that during acute leptospirosis there was some actual penetration of proximal and distal tubule cells.

Despite the variety of mammals studied and the range of serotypes isolated from these animals there is still considerable similarity in the distribution pattern and for some reason the thin loop of Henle is never utilised. Since the bacteria use such physiologically different zones as the proximal and distal tubule as well as the thick loop of Henle, their avoidance of the thin segment might be a reflection of its structural properties rather than the properties of the fluid passing through it.

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