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# Structural Organization of Renal Medulla in Domestic Swine

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The morphological and physiological features of kidney in humans and different animals have been determined over the years. However, the detailed data about the structural organization of the porcine renal medulla are absent. In the present study it was established that in the outer stripe of outer medulla, the diameter and the number of proximal straight tubules were larger than in distal straight tubules. The number and the diameter of collecting ducts (CD) in the outer and in the inner stripe of the outer medulla were the same. The height and number of epithelial cells of CD increased towards the inner medulla. The diameter of *Ductus papillaris* was the largest. The diameter of thin limbs of Henle in the inner stripe and in the inner medulla unchanged, but in the tip of papilla it increased.

The pattern of the tubular and vascular organization of medulla that was established by the current study is evidence that porcine renal medulla belongs to the simple type of renal medulla.

Key words: renal medulla, morphometry, swine.

# Introduction

Many studies have paid attention to the structural organization of the mammalian kidney [1, 2, 3]. The morphological and physiological features of this organ is studied in detail in rat, mouse, rabbit, guinea pig, minipig, cat, dog, horse and human [2, 8, 3, 13].

According to the urine concentrating ability the authors divide the kidneys into three types: the first type has a high urine concentrating ability (rat, mouse, golden hamster, Pammomys, Meriones), but the second one has a low concentrating ability (mountain beaver, muskrat). In comparison with other species, cats and dogs have an average urine concentrating ability. According to Kokubo et al. [8] the concentration ability of the urine is the highest in hamster, followed in order by dog, man and swine. Swine share a number of anatomic and physiologic characteristics with humans that make swine potentially a better model for some procedures and studies compared with other large animal species [16]. Pig has a true multirenculate, multipapillate kidney with true calices as in humans [17]. Systems that are most commonly cited as being suitable models include the cardiovascular, urinary, integumentary, and digestive systems. These features have led to the increasing use of swine as a major species in preclinical toxicology testing [16]. Histologically, both the proximal and distal tubules in the pig appear dilated when the lumen diameter is compared with other experimental species such as the dog or the rodent, but this is a normal finding in pigs and minipigs. Similarly, glomeruli tend to have a greater degree of apparent dilation of the capillary [17].

The aim of the current work was to study the light microscopical tubular organization of the renal medulla in domestic swine.

# Materials and Methods

#### Animals

The material was obtained from the kidneys of 10 castrated male pigs (Landras  $\times$  Bulgarian White), aged 6 months, slaughtered for meat consumption in a slaughterhouse.

#### Hematoxylin and eosin staining of tissue samples

Immediately after death, the samples were immersed in 10% neutral formalin for 48 h. Then they were soaked in tap water, dehydrated and embedded in paraffin. Serial longitudinal sections through the cortex and medulla of the renal lobus and cross-sections at different level of renal medulla (from the tip of papilla to the corticomedullary border) were prepared. The paraffin sections were stained with hematoxylin and eosin for light microscopy measurements.

#### Micromorphometrical investigation

In the outer stripe of the outer medulla, the diameter and the number of proximal straight tubules (PST, S3 segment - thick descending limb), distal straight tubules (DST, thick ascending limb), vessel bundles (VB), outer medullary collecting ducts (OMCD) per 1 mm<sup>2</sup> were estimated. In the inner stripe of the outer medulla, the diameter and the number of DST, TLH, VB and OMCD were determined. In the inner medulla at the border to the outer medulla, the diameter and the number of the TLH, VB and the inner medullary collecting ducts (IMCD) were measured. In the renal papilla it was established the diameter and the number of TLH and the papillary ducts (DP).

#### **Statistics**

Data for number and dimensions are given as mean  $\pm$  SD (Standard deviation). For that purpose was used a light microscope (ZEISS Primo Star, Germany), camera (Progres, Capture 2.6 - JENOPTIK) and software analysis programme (Soft Imaging Sistem GmbH). Statistical data processing was done using Data Analysis tool and Student's t-test by means of the StatMost for Windows software and the difference was considered significant when P-values were less than 0.05.

## Results

Light microscopic observation showed that the tubules in outer and in inner medulla were located around vascular bundles (**Fig. 1** and **Fig. 2**). The diameter of PST (61.75  $\pm$  11.92 µm) was greater than those of DST (49.63  $\pm$  3.12 µm) with statistically significant difference P < 0.01. The number of PST per mm<sup>2</sup> (26.80  $\pm$  0.79) was more than those (26.80  $\pm$  0.79) of DST, P < 0.001 but the number of the cells (11.1  $\pm$  2.64) of PST per 1 tubule was less than those of the cells of DST (15.30  $\pm$  1.70), P < 0.01.





Fig. 1. Proximal (PST) and distal (DST) straight Fig. 2. Collecting ducts and thin limbs of Henle tubules situated around vascular bundles (VB) near the arterioles and venules of vascular bundles consist of arterioles and venules in the outer me- in the inner medulla. Bar =  $25 \,\mu m$ dulla. Bar = 25  $\mu$ m

The number and the diameter of OMCD in the outer  $(16.40 \pm 0.69 \text{ and } 62.06 \pm 7.12)$ um, respectively) and in the inner stripe  $(16.4 \pm 0.70 \text{ µm} \text{ and } 62.01 \pm 7.29 \text{ µm}, \text{ re-}$ spectively) of the outer medulla were the same. The height and number of simple columnar epithelial cells of collecting ducts increased towards the inner medulla (from  $11.92 \pm 0.45 \ \mu m$  and  $16.2 \pm 2.15 \ \mu m$  in the outer stripe to  $14.16 \pm 1.10 \ \mu m$  and 48.10 $\pm$  7.75 µm, respectively in the inner medulla). However, the width of this epithelium decreased to the inner medulla. In the tip of the papilla the columnar epithelial cells of IMCD is replaced by higher ( $25.82 \pm 2.19 \mu m$ ) transitional epithelium of *Ductus papillaris*. In the papillary duct, the number of epithelial cells increased to  $54.20 \pm$ 5.14. The diameter of DP was the largest  $(136.56 \pm 13.07 \ \mu m)$ . The distance between OMCD in the outer stripe  $(40.90 \pm 21.11 \ \mu\text{m})$  and in the inner stripe  $(40.45 \pm 21.11 \ \mu\text{m})$  $20.03 \mu m$ ) remained the same but the distance between the IMCD decreased in the inner medulla at the border to the outer medulla. In the papilla, the distance between the DP was  $106.55 \pm 28.58 \,\mu\text{m}$  which is longer than those between IMCD, on the one hand, and those between OMCD, on the other hand. In the inner stripe the number of DST increased to  $49.30 \pm 1.16$  but the number of DST cells remained the same. The OMCD cells are almost as high as in the outer stripe. The diameter of TLH in the inner stripe  $(19.01 \pm 1.30 \text{ }\mu\text{m})$  and in the inner medulla at the border to the outer medulla  $(19.23 \pm 0.67 \,\mu\text{m})$  unchanged, but in the tip of papilla it increased to  $32.18 \pm$ 32.15 µm. The number of TLH increased from the inner stripe  $(11.50 \pm 1.35)$  to the inner medulla  $(16.6 \pm 1.26)$  at the border to the outer medulla but decreased in the tip of papilla to  $8.70 \pm 0.20$ . The ratio between TLH and CD increased from the inner stripe  $(2.40 \pm 1.08 \text{ to } 1, \text{ respectively})$  to the inner medulla  $(3.40 \pm 0.52 \text{ to } 1, \text{ respec-})$ tively) at the border with outer medulla. In the tip of papilla this ratio was the smallest  $(1.80 \pm 0.79 \text{ to1}, \text{respectively}).$ 

The diameter and the number of VB per 1 mm<sup>2</sup> in the outer (292.64  $\pm$  12.63  $\mu$ m and  $1.00\pm2.34$ , respectively) and in the inner stripe ( $287.99\pm35.63 \mu m$  and  $1.00\pm2.34$ , respectively) of the outer medulla were the same. In the inner medulla at the border with the outer medulla their diameter decreased to  $225.77 \pm 51.89 \,\mu\text{m}$  and their number increased to  $3.40 \pm 0.52$ . In the papilla only single arterioles were found. In this part of medulla, the capillaries dominated.

## Discussion

The results of our research on the structural features of the porcine renal medulla using light microscopy correlate with studies on renal medulla in species which possess both long- and short-looped nephrons by several authors. Most species have both shortand long-looped nephrons. However, cats, dogs, and many species native to arid climates have only long-looped nephrons, which conserve water more efficiently than short-looped nephrons. Conversely, beavers, which live in fresh water, have only shortlooped nephrons [3].

The inner medulla of porcine kidney was located deep to the outer medulla. It contains no thick ascending limb segments, only collecting ducts and descending and ascending thin limbs of Henle's loop in addition to capillaries. As it can be seen in the reports by Dellman and Eurell [3], the inner medulla can be subdivided into the base and the papilla. The base is adjacent to the outer medulla. The papilla, is the terminal portion of the inner medulla, which extends into the renal calices.

Compared to the medullary architecture of other mammalian species (rat [9], mouse [10] and Psammomys [6]) investigated in detail the rabbit renal medulla is the most simply organized. In the rabbit kidney all the vascular bundles are of the same small size and are regularly distributed throughout a cross-section of the inner stripe. They do not fuse, reflecting in the whole medulla the state of the bundles as originally developed in the outer stripe. In the current study, the same pattern of vascular bundles organization was observed in swine. This fact is evidence that the vascular architecture of porcine renal medulla is much closer to those of rabbit than the other rodents.

The inner stripe of the outer medulla is the most constant part of the renal medulla [4, 5, 9]. The tubules are arranged around these bundles in a pattern similar to that found in the outer stripe. This pattern of the tubular and vascular organization of medulla was confirmed by the current study, which means that porcine renal medulla belongs to the simple type of renal medulla.

The inner medulla is very differently developed among species. Species (mountain beaver, muskrat) with only short loops of Henle do not have an inner medulla and their urine-concentrating ability is poor [11, 14]. All species with high urine-concentrating ability have a well developed inner medulla [1, 13, 15].

Regarding the ratio between Henle's loops and collecting ducts along the inner medulla, considerable differences are found when comparing the base with the tip of the inner medulla; there are interspecies differences [7]. Our findings are closer to those in rat than those in rabbit.

The final sodium concentration of the interstitial tissue of the inner medulla is the result of the interaction of sodium and water transport from both Henle's loops and collecting tubules. According to Kokubo et al. [8], the rabbit kidney is structurally similar to those of man and swine. Sasaki and Suwa [12] calculated that this structural feature counteracts the effect of producing a gradient of sodium concentration towards the medullary apex on the basis of the countercurrent multiplier system. They regarded the Henle's thin segment as an exchanger, sodium depriving system. However, in contrast to man and swine, thin loops of Henle was less than collecting ducts along the entire length of the inner medulla in dog and hamster. According to Kokubo et al. [8], this finding indicates that in the pile of Henle's loop, short loops predominate over long ones. The concentration ability of the urine is the highest in hamster, followed in order by dog, man and swine.

# Conclusion

In this study, the tubular and vascular architectural pattern of porcine renal medulla showed that it belongs to simple type of renal medulla and is structurally similar to that of man and rabbit, as a whole. However, it was established that some differences from this pattern exist, such as the ratio between Henle's loops and collecting ducts along the inner medulla is closer to that in rat than in rabbit. It was observed that in the outer and inner medulla of swine, the collecting ducts were situated almost regularly and did not tend to form groups of four ducts as they do in rabbit and man. So we can assume that the domestic swine renal medulla has specific tubular organization which defines the differences in concentration ability in comparison with other mammals.

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