LOCALIZATION OF NEPHRONEOGENESIS IN ADULT LITTLE SKATE; Leucoraja erinacea

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During growth the kidney mass increases and in little skate the number of renal corpuscles rises significantly (Hentschel, H. Am.J.Anat. 183:130-147,1988). By light- and electronmicroscopy it has been shown that adolescent spotted dogfish, *Scyliorhinus caniculus* (Hentschel, H. Am.J.Anat. 190:309-333, 1991) as well as adult spiny dogfish, *Squalus acanthias* (Hentschel, H. et al., this bulletin) display all developmental stages of nephrons in defined zones of their opisthonephric kidneys. The present study was intended to reveal nephrogenetic stages in little skate, *Leucoraja erinacea*. The hypothesis was tested that addition of new nephrons to a functioning kidney (nephroneogenesis) is not only a regular feature of growing elasmobranch kidney, but is also involved in regeneration of damaged renal tissue.

Ten female little skate (disc size 28 to 32 cm width) were anaesthetized with tricaine. The right kidney was exposed through an incision of the ventral body wall. One third to one half of the organ was destroyed by heat coagulation and the tissue was surgically removed. The body cavity was closed by a double-suture. The animals were kept for one month in running seawater and fed frozen shrimps. After two months, seven skates were perfusion-fixed and processed for immunocytochemistry, *in situ* hybridisation, and light- and electronmicroscopical morphology. The epidermis on the ventral side was not grown together, the inside of the wound was closed by a continous layer of tissue, which sealed off the body cavity against the ambient seawater.

Upon histological inspection of the operated kidney, the space where the tissue had been removed was filled by a loose connective tissue consisting for the major part of fibroblasts and fibrocytes. In the direction of the remnant kidney tissue this fibrocyte-rich tissue was bordered by a zone, where nephric tubules had been damaged by the nephrectomy. The lumens of these transsected and interrupted nephrons were filled by nonpolarized cells and cellular debris. However, the epithelial cells of most tubules still showed the organization and morphology of cells of the respective segment. The glomeruli and the tubules of the intact nephrons were enlarged and the tubular lumens appeared dilated. In the vicinity of collecting ducts aggregates of mesenchymal cells were situated (fig. 1). These mesenchymal masses resembled the condensed mesenchym of the first developmental stage of nephrons in dogfish.



Fig. 1. Histological section of right, operated, kidney. Arrow points to condensed mesenchyme. Many tubular profiles are filled by cellular debris. However, dilated collecting tubules on this figure appear empty. Calibration bar equals 40 μ m. Fig. 2. Histological section of left, intact, kidney. Early nephron developmental stage II (s-shaped body) is in contact with the end of a collecting tubule. Calibration bar equals 25 μ m. Fig. 3. Epon section (0.5 μ m) of left, intact, kidney. Nephron developmental stage I. Calibration bar equals 15 μ m. Fig. 4. Cross section through island of lymphomyeloid tissue between dorsolateral bundle zone and kidney capsule. Calibration bar equals 40 μ m.

The intact contralateral (left) kidney was essentially similar by architecture and complicated nephron organization to control kidneys from previous histological studies (Hentschel, H. et al. Anat. Embryol. 198:73-89, 1998). In addition, serial sections revealed a large number of developmental stages of nephrons. Usually two to three nephrogenetic stages could be encountered on a kidney cross-section. Small but distinct islands of lymphomyeloid cells were also observed (fig. 2). All stages of nephron development were revealed (fig. 3 and 4): condensed mesenchym (stage I), s-shaped bodies (stage II), developing nephrons (stage III), and young nephrons (stage IV). These stages were regularly seen in a thin ventro-lateral strap of tissue at the outer, convex curvature of the kidney.

These experiments have uncovered that the kidneys of partially nephrectomized, adult little skate can exhibit all stages of nephron development as well as islands of lymphomyeloid tissue. These observations with batoid elasmobranchs corroborate the new finding of nephroneogenesis in adult, sexually mature shark, the dogfish *Squalus acanthias* (Hentschel, H. et al., this bulletin). The large amount of developmental stages in nephrectomized skate strongly suggests that loss of nephrons can be compensated by repair of the tissue involving nephroneogenesis. With financial support by DFG.