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Case Report

Spontaneous Renal Tumors Suspected of Being Familial in Sprague-Dawley Rats

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Abstract: Spontaneous renal tubule tumors (RTTs), with a distinctive morphological phenotype, were present in three Sprague-Dawley rats, 1 male and 2 females, out a total of 120 animals of each sex from untreated and placebo control groups in a 2-year carcinogenicity study. One female had one carcinoma, adenoma and hyperplasia, and the other female had five adenomas and many hyperplastic lesions; the male case had one carcinoma. From these cases, a biological continuum of hyperplasia, adenoma and carcinoma could be recognized. The tumors were present in the renal cortex and appeared as solid lobulated growths with occasional central necrosis. The lobules were divided by a small amount of fibrovascular tissue. Occasionally the larger tumors contained a cystic area. Tumor cells appeared distinctive and exhibited variable amounts of eosinophilic/amphophilic and vacuolated cytoplasm. Nuclei were round to oval with a prominent nucleolus. Mitotic figures were uncommon, and no distant metastasis was noted. The tumors were seen as multiple and bilateral lesions in two animals and had no apparent relationship to chronic progressive nephropathy (CPN). Foci of tubule hyperplasia were also noted to contain the same type of cellular morphology. The morphological and biological features of these 3 cases resembled the amphophilic-vacuolar (AV) variant of RTT that has been posited to be of familial origin. This is a report of spontaneous familial renal tumors in Sprague-Dawley rats from Japan. (DOI: 10.1293/tox.25.277; J Toxicol Pathol 2012; 25: 277–280)

Key words: adenoma, carcinoma, familial, kidney, rats, spontaneous

The identification and interpretation of neoplasms noted in 2-year carcinogenicity studies is one of the main goals of toxicological pathology examination. Pathologists must use all information including previous experience and reported literature to make sound decisions concerning the carcinogenic potential of a test article. The rat kidney is often a target organ in carcinogenicity studies, and spontaneous kidney tumors in the rat have an overall low incidence[1]. Therefore, any increase in tumors of the rat kidney within treated groups must be viewed as a potentially serious occurrence. Presently, two familial RTT syndromes have been described in the laboratory rat[2], one in the Long-Evans-derived Eker rats[3, 4] and the other in the Sprague-Dawley rat from Japan, termed the Nihon rat[5–7]. Both of these RTT models have been reported to be associated with germline insertional mutations. In addition, there are several case reports of suspected familial tumors in F344 and Sprague-Dawley rats[8, 9]. In the present report, we describe three cases of suspected familial RTTs that had a distinctive morphological phenotype, namely amphophilic-vacuolated (AV) cytoplasm with H&E staining, from a 2-year carcinogenicity study in Sprague-Dawley rats.

Sprague-Dawley rats (Crl:CD(SD)) were purchased from Charles River Laboratories Japan, Inc. (Kanagawa, Japan) at 5 weeks of age and used for a 2-year carcinogenicity study after a 1-week quarantine period. The animals were housed individually in wire mesh cages under controlled conditions (temperature of 23 ± 3°C, relative humidity of 50 ± 20% and a 12-h light/dark cycle) and fed CRF-1 diet (Oriental Yeast Co., Ltd., Tokyo, Japan) and tap water *ad libitum*. They were treated in accordance with the recommendations of the Guide for the Care and Use of Laboratory Animals of Bozo Research Center Inc.

Three cases of grossly visible kidney masses were observed in untreated and placebo control animals from a total of 120 animals of each sex. They were either found dead at 65 weeks during the test, sacrificed in a moribund state at 74 weeks during the test or sacrificed at 100 weeks during the test for scheduled necropsy. At necropsy, all tissues including the kidney were fixed in 10% phosphate-buffered formalin (pH 7.2), embedded in paraffin and sectioned at a thickness of 4 μm. The sections were stained with hematoxylin and eosin (HE) for histological examination.

Summary information regarding the 3 cases is presented in Table 1.

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Case 1: A male that was sacrificed at terminal kill (week 100).

Grossly, a single nodule, 40×35×35 mm in diameter, was observed in the left kidney.

Microscopically, a large cortical mass was present that had invaded through the capsule and was attached to the pancreas. This lesion was diagnosed as a renal tubule carcinoma. The tumor was divided by fibrous connective bands. Areas of hemorrhage and cystic spaces were present. The tumor was encapsulated by a slight amount of fibrous tissue, although in some areas, the tumor was encapsulated by only compressed renal parenchyma. The tumor was divided into variably sized lobules separated by a thin amount of fibrovascular tissue (Fig. 1A). Each lobule contained solid to papillary growth of pleomorphic eosinophilic/amphophilic to vacuolated cells with central necrosis. Cytoplasmic borders were indistinct. Tumor cell nuclei were round to oval in shape and contained 1 nucleolus. Mitotic figures were un-

Table 1. Summary of Proliferative Lesion in Renal Tubules in the 3 Cases

Case	Sex	Age (week)	Fate	Number of proliferative lesion in renal tubules			Location	CPN [a)]
				Adenoma	Carcinoma	Hyperplasia		
1	Male	100	TS	0	1	0	Unilateral	Minimal
2	Female	65	MS	5	0	11	Bilateral	Minimal
3	Female	74	FD	1	1	1	Bilateral	None

Case 1 and 2: untreated control group (60 rats). Case 3: placebo control group (60 rats). [a)] CPN was graded for severity on a scale of 5 grades: none, minimal, mild, moderate, and severe. Minimal grade: minimal number of the earliest lesions. TS: terminal sacrificed. MS: moribund sacrificed. FD: found dead.

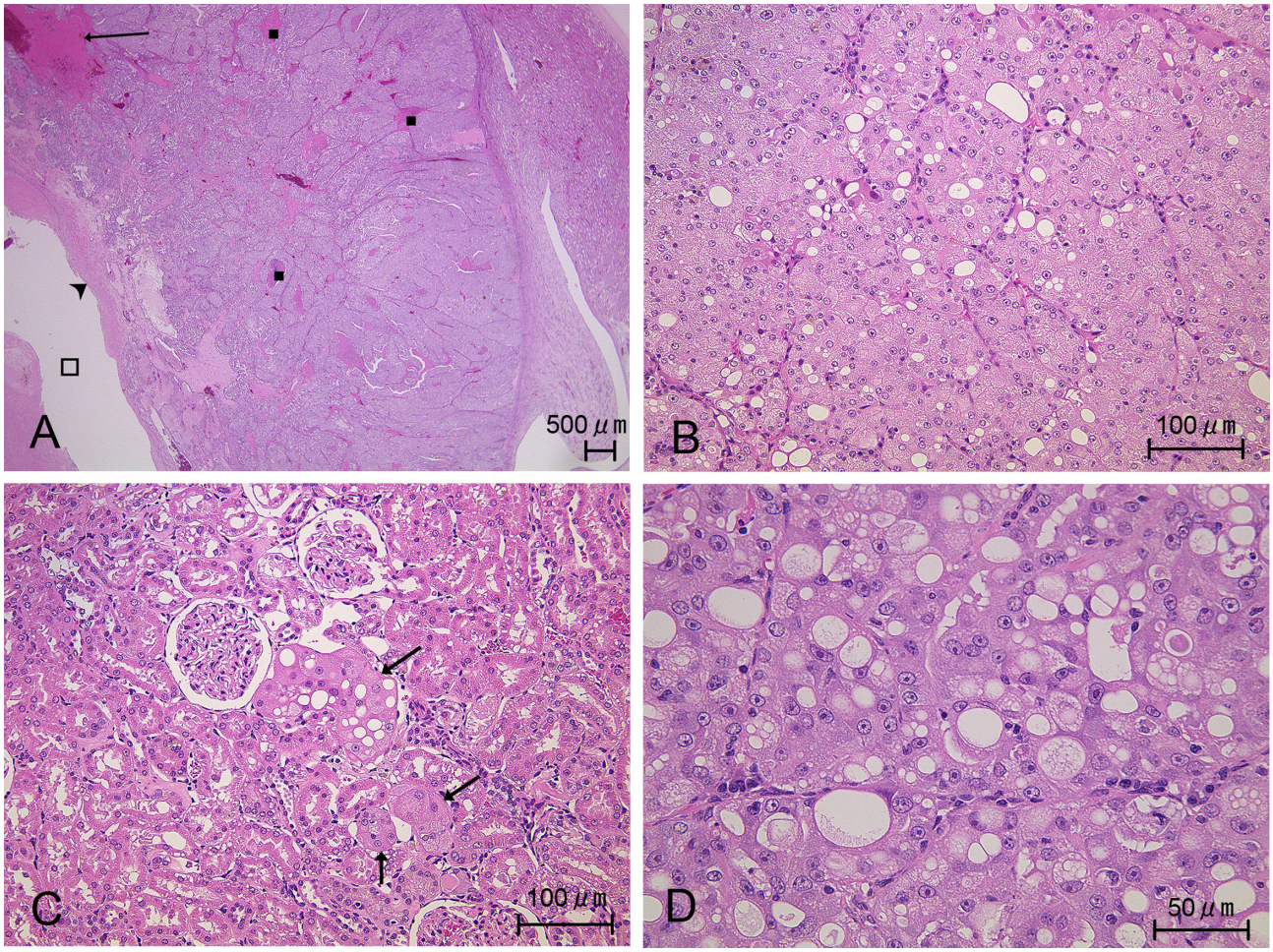

Fig. 1. Photomicrographs A) Renal tubule carcinoma from case 1. The tumor consists of multiple solid lobules with central necrosis (■), hemorrhage (arrow) and cystic space (□), and is divided by fibrovascular tissue bands (arrow head). B) Renal tubule adenoma from the case 2. The tumor consists of eosinophilic/amphophilic to vacuolated cells and is separated by thin fibrovascular tissue. C) Multiple tubule hyperplasia with eosinophilic/amphophilic and vacuolated cytoplasm (arrow) from case 2. Hyperplasia represents growth within 1 tubule with little to no compression. H.E. Stain. D) Higher magnification of the renal tubule carcinoma from case 3. Cells are variable in size and contain multiple vacuoles. Nuclei are round to oval and contain 1 prominent nucleolus.

common. No distant metastases were observed.

Case 2: A female that was sacrificed moribund in week 65.

Grossly, multiple nodules, 1×1×1 mm – 2×2×1 mm in diameter, and cysts, 1×1×1 mm in diameter, were observed in both kidneys.

Microscopically, both kidneys contained cortical lesions that were expansive but not encapsulated and were diagnosed as renal tubule adenomas. Occasionally, cystic areas were present in areas adjacent to the adenomas. These cysts were lined by cells similar to those within the adenomas. Tumor cells were divided by a small amount of fibrovascular tissue into lobules of solid growth with occasional central necrosis. The cells were slightly pleomorphic, cytoplasmic borders were indistinct, and there was eosinophilic/amphophilic staining with vacuolization. Nuclei contained prominent nucleoli. Mitotic figures were rare (Fig. 1B). Areas of small tubule hyperplasia were also observed as lesions contained within 1 tubule profile and were slightly expansive (Fig. 1D). Distinct lobular areas or areas of central necrosis were not present in the hyperplastic lesions. The cell morphology of the hyperplasia was similar to that of the adenomas. No mitotic figures were noted.

Case 3: A female that was found dead in week 74.

Grossly, a single nodule, 20×15×15 mm in diameter, was observed in the left kidney. No gross lesion was observed in the right kidney.

Microscopically, all lesions appeared histologically similar to those described previously in cases 1 and 2 (Fig. 1C). Lesions were present in both kidneys. One carcinoma was diagnosed in the left kidney, and an adenoma and area of hyperplasia were diagnosed in the right kidney. No metastases were observed.

All renal tubule proliferative lesions within the 3 cases were remarkably similar in histological appearance. In these cases, these proliferative lesions, which differed in their progressive stage, may have developed from hyperplasia to adenoma and than to carcinoma, because there are similarities in their morphological features. The incidence of RTT in this study was higher (0.83% in males:, 1.67% in females:) than in the historical control incidence data for all types of RTT (0.14% in males:, 0.08% in females:) in our laboratory. Furthermore, the incidence of renal tubule hyperplasia was slightly higher (1.82%) than in the historical control data (0.73%) in females. Also, the hyperplasia in the control data was composed of a different cell type. In this study, there were multiple RTTs and foci of tubule hyperplasia in 2 female rats, whereas, previously, only a single proliferative lesion was present in an animal in our background data. In the kidneys in all 3 cases, CPN was graded as minimal or not present. There was no apparent relationship of the proliferative lesions with CPN-affected tissue.

The hyperplasia and RTTs described in this report are similar to those of the amphophilic-vacuolar (AV) variant of the rat RTT detailed in a large survey of rats conducted by the National Toxicology Program, National Institutes of Environmental Health Sciences, National Institutes of Health, USA[2]. This resemblance is greatly enhanced by the agreement in both the morphological features (eosinophilic/amphophilic staining, lobular appearance, variably sized cytoplasmic vacuoles, frequent cystic area and low mitotic rate) and biological features (no association with CPN, spontaneous in origin and no metastasis). This AV renal tubule variant appears similar to the morphological phenotype (eosinophilic type) of the Eker rat tumor. Other case reports have also described this characteristic tumor phenotype. The AV variant of RTT, which can arise spontaneously in several different rat strains, is currently considered to be a familial neoplasm, although the genetic background of the AV variant is not known. Based on these facts, the RTTs described in this report were suggested to be of familial origin as found in the report mentioned above[2]. The AV variant has never been reported to be induced by chemical administration. Accordingly, it differs from conventional rat RTTs, which are often observed in male rats and are rarely observed in female rats. Typically, conventional RTTs appear as solid, non-lobulated growths of basophilic tumor cells often noticed in rats with an increased severity of CPN[10]. Spontaneous renal tubule carcinomas are uncommonly observed in 2-year carcinogenicity studies. Because of the apparent familial nature and the lack of any reported relationship to chemical administration, it has been suggested that the AV variant of RTT be recorded as a separate category from conventional (basophilic) RTT when assessing the numbers of compound-associated renal tumors in 2-year carcinogenicity bioassays[2]. In addition, it is recommended that laboratories maintain a separate category for the AV variant in their historical control database. This is the first report of spontaneous familial renal tumors in Sprague-Dawley rats from Japan observed in a 2-year carcinogenicity study.

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