Simulation of Electrolyte Nephropathy in Rats

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Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny*, Vol. 138, No. 8, pp. 238-240, August, 2004
Original article submitted October 16, 2003

Stages of modeling of polyetiological electrolyte nephropathy in rats are described. Morphological study confirmed the presence of interstitial and tubular disorders in rat kidneys. The model is intended for studies of the mechanisms underlying the formation, development, and correction of nephrological diseases.

Key Words: *kidneys; electrolyte disorders; lipids; rats*

Experimental methods allow modeling of pathological states and investigation of mechanisms underlying the effects of physical factors at the organ, tissue, and cellular levels using analytical methods inapplicable for clinical studies. The onset and correction of renal diseases are usually studied on experimental models involving changes in water, electrolyte, nitrous, and lipid metabolism. Pathological processes in the kidneys are induced by injections of some inorganic and organic substances (mercuric chloride and other mercuric preparations, uranium, mercury, chromium, and lead preparations, *etc.*). Some models of metabolic nephropathies (nephrocalcinosis) are induced by parathyroid hormone, tachystine-dihydrotachysterone, ethylene glycol, vitamin D_2 [3]. Steroid nephropathies are induced by castration, uni- and bilateral nephrectomies [2]. However, these pathological states lead to selective involvement of different nephron compartment [1,3,4], are technologically difficult, and are associated with animal mortality during the intervention, this making the experiments more expensive. We found no specific recommendations on simulation of polygenic disorders in rat renal tissue, induced by the above-listed pathogenic factors or their combinations. Research and applied problems often require the creation of an experimental model with metabolic changes in biological media of the body and in animal kidneys, involving minimum mortality and allowing investigation of the mechanisms underlying the development and correction of pathological processes.

The aim of this study was to create an experimental model, reflecting morphological changes in rat kidneys, for the studies of the mechanisms of formation, development, and regression of nephropathy at the cell, tissue, and organ levels.

MATERIALS AND METHODS

The study was carried out on 20 male Wistar rats from Stolbovaya Breeding Center, Russian Academy of Medical Sciences. The experimental model was reproduced in adult animals (180.5±10.6 g). The animals were kept under standard vivarium conditions and were taken into experiment after at least 14-day quarantine. Experiments were carried out in accordance with Regulations on Handling Laboratory Animals (1977).

The rats were decapitated under light ether narcosis, the kidneys were removed and fixed in 10% neutral formalin. Tissue fragments were dehydrated in ascending alcohols and embedded in paraffin routinely. Deparaffinized sections were stained with hematoxylin and eosin. The preparations were microphotographed using Svema FN film (64 U).

RESULTS

Disorders in the electrolyte, nitrous, and lipid metabolism play an important role in the pathogenesis of

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0007-4888/04/1382-0210

metabolic and inflammatory mechanisms in nephropathies. Dyselectrolytemic nephropathy is characterized by dysfunctions of individual compartments of the nephron and is mainly caused by K^+, Mg^{2+} deficiency. These metabolic disorders can be caused by secondary nephropathies, when the underlying disease involves the development of electrolyte imbalance paralleled by renal involvement. Deficit of K^+ is associated with epitheliocyte degeneration in the ascending compartments of Henle loops and pronounced changes in acid mucopolysaccharide status in the interstitium. Mg^{2+} deficiency causes severe disorders in the renal tissue with deposition of Ca^{2+} salts and impairment of K^+ and Na^+ transport into the cell [5]. It was experimentally shown that high-lipid diet caused only moderate changes in the glomeruli, while the same diet in the presence of initial disorders in animals with experimental nephritis and electrolyte imbalance led to rapid development of glomerulosclerosis [3]. Polyorgan disorders and concomitant disorders in lipid metabolism aggravating sclerosis of the renal tissue determine the interest of scientists to simulation of nephropathy caused by several pathogenic factors. It is expected that renal tissue damage is more severe under the effect of several pathogenic factors.

Electrolyte nephropathy was induced in animals receiving a semisynthetic diet containing $Na_2HPO_4 \times 12H_2O$ and deficient for K^+ and Mg^{2+} (Tables 1, 2) for 12 days. During this diet the animals received 1% NaCl solution instead of water and in parallel were daily injected (intramuscularly) with hydrocortisone acetate (1.5 mg/100 g). The duration of exposure to pathogenic factors and the course of pathological process were determined from the results of morphological studies of the kidneys.

Sensitization of the rat renal tissue was induced by special nutrition (semisynthetic nephrovasopathogenic Na^+ electrolyte-rich and K^+ and Mg^{2+}-deficient diet). Intramuscular injections of hydrocortisone acetate, augmenting Na^+ loading, served as the causative factor. K^+ deficit developing in the presence of hormone excess in the blood causes disorders in the concentration capacity of the kidneys, decreases the sensitivity of the distal tubules to antidiuretic hormone, impairs the functions of cell pumps, as a result of which Na^+ reabsorption and its content in the renal medulla decrease. The most significant of the pathogenetically oriented factors was electrolyte imbalance caused by saline loading. If this factor was absent, the percentage of nephropathogenic changes decreased. Pathological K^+/Na^+ ratio (1:18 *vs.* the normal 1.0:0.9) played an important role in these changes. Cholesterol was excluded from the standard semisynthetic diet as a factor activating lipid disorders, vascular sclerosis, and significant for animal mortality.

TABLE 1. Semisynthetic Diet for Rats, g/kg

Ingredient	Diet	
	normal	pathogenic
Casein	180	300
Cod liver oil	10	10
Margarine	90	45
Lard		45
Dry yeast	30	20
Salt mixture I	40	
Starch	650	510
Vitamin D_2, mg		15
Salt mixture 2		50
Dry bile		10

TABLE 2. Salt Mixture Composition, g/kg

Ingredient	Salt mixture 1 (normal)	Salt mixture 2 (pathogenic)
$Na_2HPO_4 \times 12H_2O$		300
$NaH_2PO_4 \times 2H_2O$	170	170
$K_2HPO_4 \times 3H_2O$	342	40
NaCl	168	270
$CaCO_3$	150	150
$MgSO_4 \times 7H_2O$	170	35
$FeCl_3 \times 7H_2O$	4.34	6.04
$MnCl_2 \times 4H_2O$	1.9	1.6
$CuSO_4 \times 5H_2O$	0.20	0.16
KI	0.04	0.08

Characteristic morphological changes in the kidneys proved the development of experimental nephropathy in animals. The kidneys in animals with induced nephropathy were enlarged (by 1.2 times), mainly at the expense of the cortical and medullary layers. Compared to kidneys from intact rats, these kidneys were lighter colored, with gray surface incorporations, adhesions between the capillaries and capsules were found in some glomeruli. Histological study showed proliferative infiltration in the convoluted tubule epithelium and basal membranes between the renal cortex and medulla. Degenerative changes were seen along the entire length of the renal tubules; the productive reaction in the stroma resembled interstitial medullary nephritis. Microliths appeared in the lumen of convoluted tubules, which was paralleled by necrosis of the epithelium; some tubules were lined with cubic epithelium, and vacuolation of the cell cytoplasm was observed in the thin portions of the tubules and in collecting tubules.

Hence, an experimental model of renal disease based on electrolyte nephropathy was created in rats.

Histomorphological findings indicate that this method of simulation of electrolyte nephropathy completely reflects pathological changes in the kidneys typical of nephrological diseases. Reproduction of the experimental model in rats as the most available biological species, the absence of animal mortality, and the possibility of using the animals within 2 months for further investigations suggest that this method is economically beneficial for the study of mechanisms of nephropathy formation, development, and regression.

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