Role of CD64, P53 and P21 Proteins in the Pathogenesis of the Tubulo-Interstitial Syndrome in Masugi Chronic Nephritis

Yu. E. Rogovyi, L. G. Arkhipova, M. V. Dikal, L. O. Filipova, and E. N. Mil'

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Experiments of 42 outbred male albino rats (160-180 g) showed an increase in the content of CD64, fibroblast growth factor, and proapoptotic proteins (p53 and p21) in the renal cortical matter on day 45 of experimental chronic Masugi nephritis. A positive correlation between CD64 and fibroblast growth factor characterizes the macrophage-fibroblast interactions, the key point in the formation of tubulo-interstitial fibrosis.

Key Words: *Masugi nephritis; tubulo-interstitial syndrome; CD64; p53; p21*

The tubulo-interstitial syndrome (combined degenerative injury to the renal tubules and renal interstitium with edema, infiltration of the stroma with monocytes/macrophages, and connective tissue growth [1]) is now acknowledged as the main factor determining rapid progress of chronic pathological processes in the kidneys. In addition, it is known that monocytes/macrophages express CD64 on their surfaces [4], and atrophy of nephrons is associated with apoptosis, which is activated via proapoptotic proteins p53 and p21 [8]. On the other hand, the role of CD64 and p53 and p21 proapoptotic proteins in the pathogenesis of the tubulo-interstitial syndrome in chronic Masugi nephritis is little studied.

We studied the role of CD64 and p53 and p21 proapoptotic proteins in the pathogenesis of the tubulo-interstitial syndrome in chronic Masugi nephritis.

MATERIALS AND METHODS

Experiments were carried out on 42 outbred male albino rats (160-180 g). Masugi nephritis was induced by two intraperitoneal injections of rabbit nephrotoxic serum (titer of antirenal antibodies in the complement fixation test not lower than 1:1024). The study was carried out on day 45, which corresponded to the development of chronic Masugi nephritis with tubulo-interstitial syndrome [6]. The animals were decapitated under ether narcosis. Chronic nephritis and the tubulo-interstitial syndrome were morphologically verified by histological studies of the renal cortical matter with staining of deparaffinated sections with hematoxylin and eosin and after Slinchenko.

Specimens of the rat renal cortical matter were evaluated by the IMEDIS TEST+ vegetative resonance test. The samples (50-100 mg) in tubes of thin organic glass were placed into a container of IMEDIS BRT PK device (Registration certificate No. FS 022a3066/0414-04 for a medical engineering article, granted on July 8, 2004 by Federal Service for Surveillance in Public Health and Social Development of the Russian Federation). The study was carried out using the software and bioindex scale (the content of CD64, p53 and p21 proteins, fibroblast growth factor, angiotensin II (AT II), and hydroxyproline were measured) [5]. The bioindex scale values from 1 to 21 were taken for arbitrary units. The

Department of Physiology, Bukovina State Medical University, Ministry of Health of the Ukraine, Chernovtsy

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data were processed by methods of parametrical statistics using Statgraphics and Excel 7.0 software.

RESULTS

Histological findings confirmed the development of chronic Masugi nephritis and the formation of the tubulo-interstitial syndrome, which manifested in increased growth of the connective tissue stroma, infiltration of the interstitium with monocytes/macrophages and lymphocytes, and degeneration of tubules due to apoptosis activation. The results indicated increased levels of CD64, p53 and p21 proteins, fibroblast growth factor, AT II, and hydroxyproline in the renal cortical matter on day 45 of chronic Masugi nephritis (Fig. 1).

The expression of CD64 in chronic Masugi nephritis positively correlated with the levels of fibroblast growth factor and p53 proapoptotic protein. The level of protein p53, in turn, directly correlated with the content of AT II and hydroxyproline (collagen marker) under these conditions. Regression analysis of these relationships is presented in Fig. 2.

The increase in CD64 content in the renal cortical matter in chronic Masugi nephritis is caused by the development of the tubulo-interstitial syndrome with infiltration of the stroma with phagocytosing monocytes/macrophages expressing CD64. The realization of the macrophageal-fibroblastic interactions promotes the growth of connective tissue in the interstitium, which is confirmed by positive correlation between CD64 and fibroblast growth factor. The increase in connective tissue growth in the renal cortical matter is confirmed by increased level of hydroxyproline (collagen marker). The increased level of AT II in the renal cortical matter in chronic Masugi nephritis is due to activation of the renin-angiotensin system as a result of injury to the nephron proximal compartment and triggering of the tubuloglomerular feedback mechanism [2]. Angiotensin II, in turn, promotes the growth of the connective tissue and formation of the tubulo-interstitial syndrome. The increase in the levels of p53 and p21 proteins in the renal cortical matter in chronic Masugi nephritis is caused by activation of apoptosis, induces the nephron tubule and glomerule

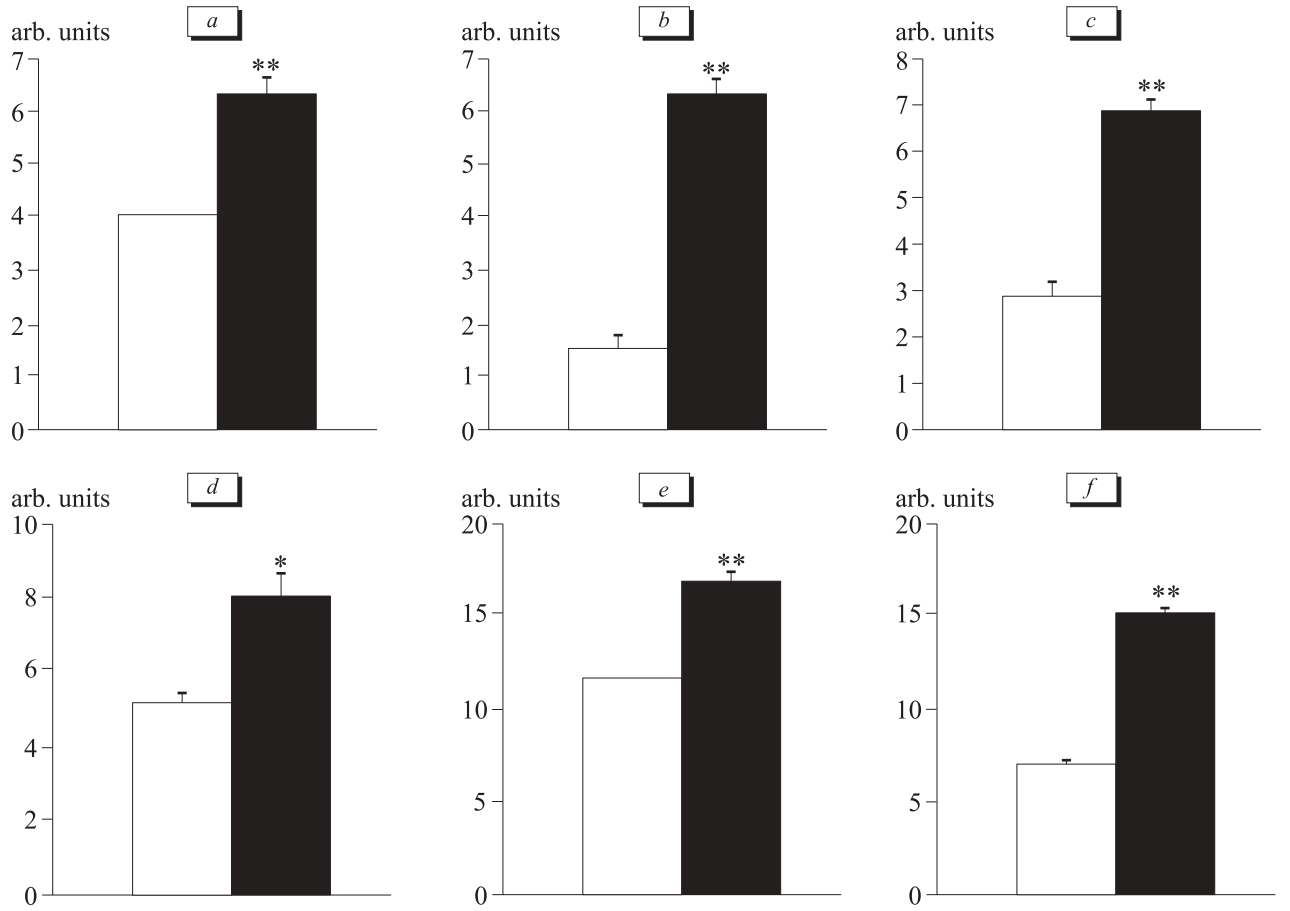

Fig. 1. The content of CD64 (*a*), p53 (*b*) and p21 proteins (*c*), fibroblast growth factor (*d*), AT II (*e*), and hydroxyproline (*f*) on day 45 of experimental chronic Masugi nephritis ($X \pm SX$). Light bars: control; dark bars: chronic Masugi nephritis. *$p<0.01$, **p0.001 compared to the control.

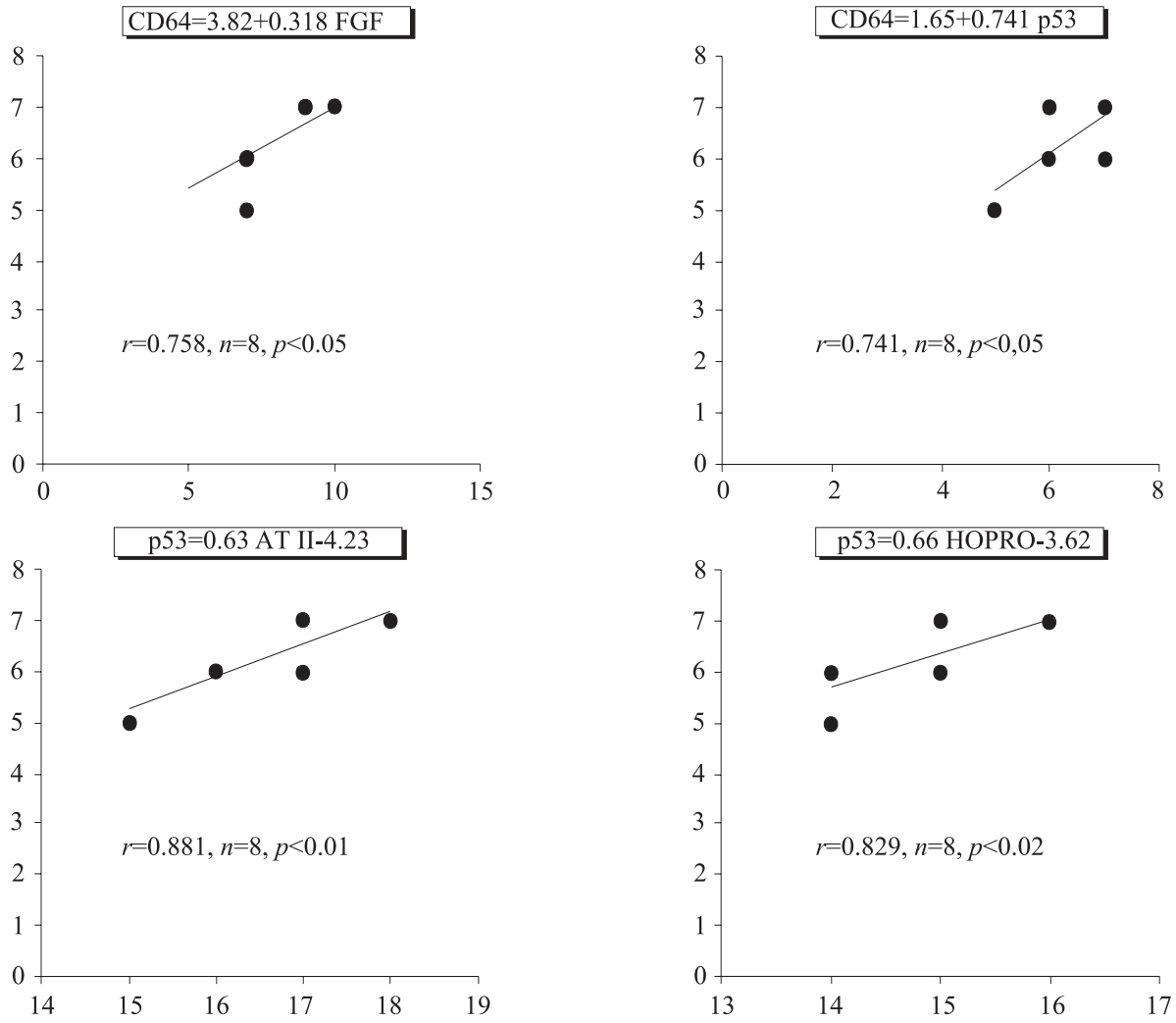

Fig. 2. Regression analysis of relationships between CD64, p53 protein, AT II, fibroblast growth factor (FGF), and hydroxyproline (HOPRO) in the rat renal cortical matter on day 45 of experimental chronic Masugi nephritis. *r*: coefficient of correlations; *n*: number of observations; *p*: significance of correlation.

atrophy, and promotes the decrease in the bulk of functioning nephrons, which is characteristic of chronic nephritis. Positive correlation between CD64 and p53 protein is explained by the fact that the realization of macrophage-fibroblast interactions with the expression of CD64 on monocytes/macrophages leads to connective tissue growth in the interstitium and to nephron atrophy due to apoptosis activation by increased level of p53 proapoptotic protein in the renal cortical matter. Positive correlation between p53 and AT II is due to the fact that activation of the renin-angiotensin system in chronic Masugi nephritis with increase of the AT II vasoconstrictor collagen-stimulating potential [6,7] leads to an increase in the connective tissue growth with subsequent nephrocyte atrophy at the expense of apoptosis activation with increase in the level of p53 proapoptotic protein in the renal cortical matter. Positive correlation between hydroxyproline and p53 protein is explained similarly (because hydroxyproline is a collagen marker).

Hence, the levels of CD64, fibroblast growth factor, and p53 and p21 proapoptotic proteins in the renal cortical matter increased on day 45 of experimental chronic Masugi nephritis. A positive correlation between CD64 and fibroblast growth factor detected in this study characterizes the process of macrophage-fibroblast interactions, the key point in the formation of tubulo-interstitial fibrosis.

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