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Features of Clinical and Immunological Parameters in Chronic Tubulointerstitial Nephritis in Children

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Abstract.

In childhood, a chronic (latent or undulating) course of tubulointerstitial nephritis (TIN) is more often observed, due to a genetic predisposition, the development of which is determined by endogenous (congenital and hereditary) factors. This type of non-destructive inflammation, predominantly of the medulla, ends with interstitial fibrosis with the involvement of all structures of the nephron in the pathological process.

Purpose of the study. Determine the value of cytokines in the blood and urine against the background of tubular dysfunctions in CTIN in children.

Research material. We observed 132 patients with chronic TIN (CTIN). Of these, 72 children (54.5%) with relapsing and 60 (45.5%) patients with a latent course of the disease. The average age was 8.85 years (85% of girls from 7.22 to 10.47 years).

Results. The study of the serum concentrations of the studied cytokines in CTIN patients revealed a statistically significant excess of the control values of pro-inflammatory TNF-α. almost six times. Anti-inflammatory IL-4 was almost three times higher than the control values, while the concentration of INF-$^{\gamma}$ was only doubled. In this case, the patients were characterized by the absence of systemic signs of the activity of the chronic process.

Conclusions. Disturbances of renal function in all manifestations of tubulointerstitial renal lesions in children, regardless of their polyetiology and clinical polymorphism, occur mainly due to its tubular component (reabsorption, osmotic concentration, acido-ammoniogenesis). The severity of tubular disorders is determined by the type of tubulointerstitial renal lesions ($p < 0.001$). The most significant decrease in acidogenesis (p

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<0.05) was found in the a ctive phase of recurrent tubulointerstitial nephritis.

Key words: chronic tubulointerstitial nephritis, cytokines, TNF-α, functional state of the kidneys.

In childhood, a chronic (latent or undulating) course of tubulointerstitial nephritis (TIN) is more often observed, due to a genetic predisposition, the development of which is determined by endogenous (congenital and hereditary) factors [8,9,11]. This type of non-destructive inflammation, predominantly of the medulla, ends with interstitial fibrosis with the involvement of all structures of the nephron in the pathological process [2,10].

The formation of CTIN is promoted by the persistence of respiratory viruses against the background of hypoimmune conditions, radionucleide contamination and contamination of areas with heavy metal salts [3,4,12].

Damage by various etiological agents to epithelial cells of tubular cells as a key event in the formation of tubulointerstitial changes leads to a change in their phenotype (transdifferentiation) [5,13]. Tubular cells acquire the ability to express the main proinflammatory cytokines, chemokines and growth factors, which act as local mediators formed directly in the renal tissue [6,7,14].

An imbalance in the balance of pro- and anti-inflammatory systems of cytokines, pro- and antifibrogenic molecules determines the severity of inflammatory and proliferative processes in the renal tissue, which allows us to consider them as markers of dysfunction of the regulatory mechanisms of the local immune response of the body [1].

Purpose of the study. Determine the value of cytokines in the blood and urine against the background of violations of tubular functions in CTIN in children.

Material and research methods.

Material. We observed 132 patients with chronic TIN (CTIN). Of these, 72 children (54.5%) with relapsing and 60 (45.5%) patients with a latent course of the disease. The average age was 8.85 years (85% of girls from 7.22 to 10.47 years).

During the study, the following analyzes were carried out: biochemical analysis of blood (total protein, proteinogram, CRP); general urine analysis; cumulative samples (according to Nechiporenko, Addis-Kakovsky); sowing urine for flora with a quantitative assessment of the degree of bacteriuria, nitrite test; morphology of urine sediment; uroleukogram.

To assess the nature of renal dysfunctions, we used:

1 - methods to identify the state of different parts of the nephron: glomerular apparatus (Reberg's test); proximal tubules (excretion with urine (daily proteinuria); distal tubules (ability to osmotic concentration (Zimnitsky's test, modification of Volhard's test), determination of daily excretion of titratable acids and ammonia excretion, urine pH));

2 - methods that detect violations of the total work of the nephron (determination of serum levels of creatinine, urea, electrolytes, acid-base state of the blood).

To assess the content of the studied cytokines - interleukin-4 (IL-4), interferon-γ (INF-γ), tumor necrosis factor-α (TNF-α) in serum and urine, we used diagnostic kits of commercial test systems IL-4, INF -γ, FNO-α (company CJSC "Vector-Best", Novosibirsk). The studies were carried out on the basis of the ODMMC of the Samarkand Medical Institute (headed by M.K. Azizov, Doctor of Medical Sciences).

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Results. The peculiarity of the chronic course of the studied nephropathy is that the clinic is asymptomatic. The manifest onset of CTIN was detected in 51 patients (38.6%) out of 132, which subsequently took on a wave-like character. The active period of this variant of the CTIN course was characterized by the absence of a well-defined beginning and the predominance of the minimum degree of activity (30/51). Only in 21 patients did the clinic correspond to the II degree. Symptoms of endogenous intoxication (including weight loss, anorexia, fatigue) and arterial hypotension dominated in 100% of cases. Abdominal pain and pain in the lumbar region occurred in 15.9% (21). Taking into account the nonspecificity and low symptoms of exacerbation of the chronic process, all children were admitted to the clinic only in the fourth week from the moment of manifestation of activity, but most often (111/132) it was difficult to find out when collecting anamnesis the date of exacerbation. At the time of examination, the patients had no complaints, but urinary syndrome remained in the accompanying test results.

The latent variant of the chronic course occurred in 60 out of 132 children (45.5%). All patients with this variant of CTIN were characterized by the absence of complaints and accidental detection of urinary syndrome during clinical examination or examination in connection with the disease of any of the family members. However, an objective examination in 88.3% of patients (53/60) revealed signs of endogenous intoxication, a tendency to hypotension in 61.6% of cases (37/60). The constant persistence of urinary syndrome in these patients did not allow to clearly delineate the stages of this variant of nephropathy.

The duration of the disease averaged 48.0 months (min 12.0, max 132.0 months in the sample). In children with latent CTIN was determined from the moment of detection of urinary syndrome according to form No. 112.

Non-specific markers of inflammation (n = 132) indicated the absence of systemic signs of the process activity. The average ESR values were 6.91 ± 3.03 mm / h; the number of leukocytes - 7.90 ± 1.98x 109 / l. Proteinogram in all children was normal. Mild normochromic anemia was found in more children (125/132).

Urinary syndrome was characterized by leukocyturia (up to 5-10 cells in the field of vision), microhematuria (up to 25 or more erythrocytes in the field of vision). An insignificant number of leukocytes in the urocytogram did not allow us to establish the predominant type of leukocyturia. Cylindruria was less typical for urinary syndrome (25/132) and was represented by hyaline casts (no more than 2-3 in the FOV). Proteinuria did not exceed 1.05 g / L in a single serving. The maximum value of daily proteinuria in the sample was 1235.0 mg / 24 hours, while the minimum was 0 mg / 24 hours.

In all examined children, bacteriuria in single portions and three-fold bacteriological culture of urine were negative.

The study of glomerular function indices in patients with CTIN showed the absence of glomerular filtration disorders (Table 1)

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Table 1

Renal function indices in children with CTIN upon admission in the acute phase (M ± m)

Indicators	Healthy children (n = 25)	RTIN (n = 72)	LTIN (n = 60)
Glomerular filtration rates			
GFR, ml / min.	98,6±7,8	74,61±1,0 P<0,05	69,11±1,16 P<0,05
Indicators of the functional state of the distal tubules			
Osmolarity, mmol / day	1000±200	705,68±20,5 P<0,001	610,46±20,05 P<0,001
Ammonia, mmol / day	46,8±1,2	34,13±1,0 P<0,001	30,7±0,81 P<0,001 P_1<0,02
Titre acid urine mmol / day	51,0±2,8	30,18±0,72 P<0,001	23,85±0,84 P<0,001
AK	0,478	0,530 P<0,001	0,571 P<0,001
Indices of the functional state of the proximal tubules			
Daily urine output, l / day	1,7±0,036	1,185±0,08 P<0,001	1,06±0,028 P<0,001
Minute diuresis, ml / min	1,2±0,037	0,82±0,017 P<0,001	0,74±0,083 P<0,001

Note: P - significance of the difference between the indicators of healthy people and in children with CTIN

There were no statistically significant differences between the studied parameters in patients with an active phase of CTIN.

In the study of serum concentrations of the studied cytokines in patients with CTIN, a statistically significant excess of the control values of pro-inflammatory TNF-α (Table 2) was revealed by almost six times. Anti-inflammatory IL-4 was almost three times higher than the control values, while the concentration of INF-Y was only doubled. In this case, the patients were characterized by the absence of systemic signs of the activity of the chronic process.

It should be noted that when compared with the same parameters in patients with LTIN and the active phase of RTIN, significant differences were found for TNF-α and INF-Y (Table 2). Higher levels of TNF-a (tenfold over control) and INF-Y (fourfold) corresponded to active inflammation with RTIN. The level of IL-4 was characterized by a statistically similar increase (almost threefold in patients with both LTIN and RTIN).

Differences in serum cytokine concentrations in LTIN patients from those in RTIN patients were established only in terms of TNF-α. IL-4 values in patients with LTIN did not differ significantly from the concentrations of this cytokine in patients with RTIN.

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Table 2

Serum cytokine concentration in CTIN patients (pg / ml)

Indicators	Healthy (n = 25)	RTIN (n = 72)	LTIN (n = 60)
IL-4, pg / ml	5,73 ±2,48	36,05 ±2,3 P<0,001	34,8 ±2,1 P<0,001
TNF-α, pg / ml	7,26±1,8	56,54±2,1 P<0,001	48,15±2,1 P<0,001
INF-, pg / ml	27,48±2,55	110,95±2,14 P<0,001	108,6±3,4 P<0,001

Note: P - significance of the difference between the indicators of healthy people and in children with CTIN

Concentrations of the studied cytokines in urine in patients with CTIN are presented in Table 3.

Table 3

Concentration of cytokines in urine of patients with CTIN (pg / ml)

Indicators	Healthy (n = 25)	RTIN (n = 72)	LTIN (n = 60)
IL-4, pg / ml	6,35 ±2,86	29,65 ±2,0	26,8 ±2,2
TNF-α, pg / ml	11,8±1,22	40,5±1,38	30,2±2,3
INF-, pg / ml	13,35±1,73	93,95±1,98	84,6±3,5

Note: P- the reliability of the difference between the indicators of healthy and in children with CTIN

Discussion. The condition of the tubular nephron of patients with CTIN was characterized by a partial decrease in the reabsorption function of the proximal tubules. At normal levels of amino nitrogen in the blood (6.78 ± 0.46 mg /%), the degree of aminoaciduria was minimal and approached negative values. Glucosuria in most cases had negative indicators (Mo-0 mmol / 24), only in seven patients it was recorded in the amount of 1.59 mmol / 24.

Daily proteinuria also exceeded normal values, and the maximum value was 1235.0 mg / 24. The condition of the distal tubules in patients with CTIN was characterized by a predominance of acidogenesis dysfunctions.

The concentration function of the distal tubules in patients with RTIN did not have statistically significant differences from the values in patients with LTIN. The urine reaction only clinically deviated towards the alkaline side.

The state of the total renal function in the chronic course of the disease remained intact.

Despite the absence of laboratory indicators of the severity of the process in LTIN

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patients, the local level of activity is minimal, manifested by urinary syndrome, impaired tubular functions and is accompanied by an increase in the concentration of the studied cytokines not only in serum, but also in urine (Table 3).

At the same time, the concentrations of TNF-α were almost four times (with RTIN eight), INF-Y five times (with RTIN ten) exceeded the control. The IL-4 values are only three times higher than in the control (almost seven times with RTIN). In contrast to the indices of patients with RTIN, all the studied cytokines in patients with LTIN had significantly lower values.

Among the local signs of RTIN activity, erythrocyturia (r = 0.606) has a strong relationship with INF-Y in urine as well as in serum, and leukocyturia (r = 0.466) has a weak one. The reverse position was found for urinary IL-4 levels. This cytokine is more strongly associated with leukocyturia (r = 0.550) and weaker with erythrocyturia (r = 0.400).

Renal function indices were cytokine-related with urine IFN-Y. Weakly urine INF-Y correlated with titratable acidity (r = - 0.471), ammonia (r = - 0.448), specific gravity (r = - 0.400).

For TNF-α, relationships were established with cytokines and daily proteinuria (r = 0.431). An increase in TNF-a in urine corresponds to an increase in the concentration of this cytokine in serum (r = 0.599).

Correlation analysis for TIN (n = 132) confirmed tubular type of proteinuria. Although the degree of LTIN activity was less than the degree of RTIN activity, the relationship between nonspecific signs of inflammation and the performance of the tubular section reflects the activity of the acute process: leukocytosis directly correlated with urine pH, inversely with the specific gravity, titrated acidity, ammonia.

Daily proteinuria was positively correlated with the duration of the disease. Perhaps, with an increase in the duration of the active process, an increase in the degree of damage to the proximal tubules occurs. It was erythrocyturia, as a sign of local activity of the process, that strongly correlated directly with the indices of tubular disorders (daily proteinuria), confirming its specificity for TIN. Although leukocyturia is less characteristic of TIN, it was inversely correlated with titratable acidity, reflecting deeper tubular damage with an increase in the degree of local inflammation activity.

Conclusion. Disorders of renal function in all manifestations of tubulointerstitial kidney lesions in children, regardless of their polyetiology and clinical polymorphism, occur mainly due to its tubular component (reabsorption, osmotic concentration, acido-ammoniogenesis). The severity of tubular disorders is determined by the type of tubulointerstitial renal lesions ($p < 0.001$). The most significant decrease in acidogenesis ($p < 0.05$) was found in the active phase of recurrent tubulointerstitial nephritis.

In all clinical forms of active inflammation, the concentrations of TNF-α and INF-Y increase significantly ($p < 0.001$) and the anti-inflammatory activity of IL-4 ($p < 0.003$) in serum and urine increases. The studied cytokines strongly correlate with nonspecific markers of inflammation and erythrocyturia, leukocyturia.

In tubulointerstitial lesions of the kidneys, strong and very strong correlations of the concentrations of cytokines in the blood serum and urine with indicators of impaired tubular functions were revealed. For patients with tubulointerstitial nephritis, an inverse strong correlation was found between urinary cytokine concentrations and the duration of the disease.

No relationship was found between the duration of the disease and the indices of renal functions, which makes it possible to determine TNF-α, INF-Y and IL-4 as local mediators of inflammation initiation in the renal tissue.

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