EARLY LABORATORY DIAGNOSIS AND PROGNOSIS OF DIABETIC NEPHROPATHY

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РАННЯЯ ЛАБОРАТОРНАЯ ДИАГНОСТИКА И ПРОГНОЗ ДИАБЕТИЧЕСКОЙ НЕФРОПАТИИ

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Annotation. Currently, diabetes mellitus and its macro- and microvascular complications, in particular diabetic nephropathy (DN), are considered chronic non-infectious diseases, which, due to the rate of increase in prevalence, have acquired the character of a non-infectious epidemic [3, 11 Glomerulosclerosis, which develops in diabetic nephropathy (DN), causes impairment of the filtration function of the kidneys, the terminal stage of which is chronic renal failure [1, 5]. In this regard, early diagnosis of DN at the preclinical stage is a task, the importance of which is due to its decisive role in the prevention, effectiveness of treatment and prognosis of the course of DM.

Key words: diabetes mellitus, diabetic nephropathy, laboratory diagnostics, prognosis.

Аннотация. В настоящее время, сахарный диабет и его макро- и микрососудистые осложнения, в частности, диабетическая нефропатия (ДН), считаются хроническими неинфекционными заболеваниями, которые по скорости роста распространенности приобрели характер неинфекционной эпидемии [3, 11]. Гломерулосклероз, развивающийся при диабетической нефропатии (ДН), вызывает нарушение фильтрационной функции почек, терминальной стадией развития которой является хроническая почечная недостаточность [1, 5]. В связи с этим ранняя диагностика ДН на доклинической стадии представляет собой задачу, важность которой обусловлена определяющей ролью для профилактики, эффективности лечения и прогноза течения СД.

Ключевые слова: сахарный диабет, диабетическая нефропатия, лабораторнгая диагностика, прогнозирование.

Introduction. Diabetes mellitus (DM) is a widespread disease characterized by multiple complications, in particular diabetic nephropathy (DN). There are several theories explaining the pathogenesis of DN: metabolic, hemodynamic and genetic. However, most researchers agree that permanent hyperglycemia plays an important role in its occurrence and progression. It triggers a “vicious circle” of events that are a key link in the occurrence and development of DN and include disorders caused by the processes of protein glycosylation, oxidative stress reactions, as well as subclinical inflammation. Currently, DN is not a fatal complication of diabetes, since its development can be prevented. However, just a few decades ago it was detected in every second patient with diabetes. The development of proteinuria was the first laboratory indicator of DN and indicated the irreversibility of the pathological process in the kidneys. However, there were no methods for its early diagnosis [1, 6].

To date, a sufficient number of laboratory methods for assessing the excretory and filtration capacity of the kidneys have been developed and standardized. However, early diagnosis of DN is an urgent problem in modern medicine. Currently, a set of highly sensitive and specific biomarkers has been obtained that allows one to assess renal function and predict the development of DN [3, 9].

Currently, the possibilities for diagnosing DN at the preclinical stage have significantly expanded. And, although MAU is still the main clinical indicator and the “gold standard” for laboratory diagnosis of the onset and progression of DN in diabetes (especially type 1 diabetes), however, its diagnostic significance is very limited. Increasingly, information appears in the scientific literature about biomarkers that are more specific than MAU and predictors of kidney damage in diabetes [3]. The limited

prognostic value of MAU is primarily due to the fact that its high level is not always specific for kidney damage, since protein concentration is influenced by many non-renal factors (intense exercise, high protein diet, circulatory failure, arterial hypertension, infections urinary tract, etc.). Thus, the interpretation of the MAU indicator in type 2 diabetes has undergone significant changes.

The authors propose various classifications of DN biomarkers [8,9], according to which we can distinguish: 1) markers of tubular damage; 2) glomerular markers and metabolic products of the extracellular matrix; 3) immunoinflammatory factors; 4) markers of oxidative stress; 5) other biomarkers. For early diagnosis of renal dysfunction, it is very promising to determine the concentration of these substances in human biological fluids (blood and urine).

Markers of tubular damage. Determining the concentration of markers of tubular damage in blood and urine can be very useful in assessing kidney damage at the preclinical stage of development of DN, since these markers make it possible to diagnose dysfunction of the renal tubular apparatus due to the development of DN at the preclinical stage, i.e. before the appearance of UIA, and also worked well in patients with normoalbuminuric DN.

Cystatin C is a protein synthesized by all cells of the body. Its low molecular weight allows it to easily pass through the kidney filter and be completely excreted by the kidneys. The level of cystatin C in biological fluids is not affected by age, gender and muscle mass. Its level in the blood correlates with the severity of renal pathology. It has been proven that cystatin C is a more sensitive marker, compared to creatinine, reflecting impaired renal filtration function, especially in the case of a moderate decrease in GFR, when there is no proportionality between creatinine and GFR. In this case, formulas are used that allow one to estimate GFR by the concentration of cystatin C in the blood, and not by the level of creatinine [1, 17].

In case of damage to the tubular apparatus, it is very informative to determine the level of cystatin C in the blood serum, while an increase in its concentration in the urine indicates a violation of reabsorption in the proximal tubules.

Neutrophil gelatinase-associated lipocalin (NGAL) is a 24 kDa lipocalin family protein associated with neutrophil gelatinase. Under physiological conditions, NGAL is synthesized by various cells of the body. It passes through the renal filter and is reabsorbed in the proximal tubules. NGAL has a stimulating effect on the renal epithelium when it is damaged, and also has a protective effect against bacterial infections. Damage to kidney tissue causes an increase in NGAL levels in the blood, which correlates with the severity of the pathology. NGAL, synthesized in the distal parts of the nephron, is excreted in the urine. Kidney damage of ischemic and toxic origin leads to a multiple increase in the expression and excretion of NGAL in the urine, 24-48 hours ahead of the increase in serum creatinine concentration. This gave

rise to the authors to propose NGAL as a biomarker of acute kidney injury and a predictor of its possible transformation into a chronic form [2, 16].

Kidney Injury Molecule-1 (KIM-1) is a transmembrane protein of the proximal tubular epithelium, first described in 1998. A characteristic feature of KIM-1 is a low level of expression in the intact kidney and an increase in expression in the first 12 hours of ischemic damage. However, its increase is not always associated with an increase in the concentration of urea nitrogen and creatinine in the blood serum [2, 13]. Thus, KIM-1 is a marker of acute kidney injury, reflecting the degree of tubular damage. KIM-1 is currently being investigated as a marker of kidney damage in various chronic diseases.

Alpha-1-microglobulin (A1M) is a 27 kDa protein found in various body fluids. It passes freely through the glomerular barrier and most of it is reabsorbed in the proximal tubule. Therefore, elevated urinary A1M levels are an early sign of renal proximal tubular damage. In particular, an increase in the urinary level of A1M was shown in diabetes against the background of normoalbuminuria compared with healthy individuals, which indicates the possibility of using urine A1M as a predictor marker for the development of DN to the stage of albuminuria. It has also been shown that high levels of A1M in urine in patients with diabetes correlate positively with the concentration of urine albumin and serum cystatin C and negatively with GFR. Urinary A1M levels are influenced by duration of diabetes and glycemic control. An increase in the concentration of A1M in urine precedes the onset of albuminuria, which indicates that A1M can be used in the diagnosis of the onset and development of DN in patients with type 2 diabetes [2, 10].

Retinol-binding protein (RBP, retinol binding protein) and B-2-microglobulin (b2-microglobulin) is a protein with a folded structure that can specifically bind retinoids, in particular vitamin A. B2-microglobulin is a surface protein-antigen of cell nuclei. B2-microglobulin takes part in the regulation of lymphocyte proliferation. Due to the fact that both peptides are reabsorbed in the proximal tubules, under normal conditions their content in urine is minimal [26]. An increase in urinary excretion of these peptides is observed when the tubular apparatus of the kidneys is damaged. In particular, a direct relationship has been shown between the level of B2-microglobulin and the degree of DN in type 1 and type 2 diabetes [1, 9].

Liver-type fatty acid-binding protein (LFABP) is a transport protein with a molecular weight of about 15 kDa. It is expressed by hepatocytes and the epithelium of the proximal and distal tubules of the kidney and is involved in intracellular metabolism and transport of fatty acids. It is assumed that LFABP in the kidneys plays the role of an endogenous antioxidant that suppresses tubulointerstitial disorders. Damage to proximal tubule cells causes activation of the LFABP gene, which leads to

an increase in its expression, resulting in increased excretion of LFABP in the urine [8, 9].

Glomerular biomarkers and metabolic products of the extracellular matrix. Permanent hyperglycemia underlies the disruption of the transformation of extracellular matrix components, which ultimately leads to the development of glomerulosclerosis. Also, impaired degradation of these components is the pathogenetic basis for the development of fibrosis. In this regard, close attention of researchers is focused on metabolic products of the extracellular matrix and they are actively studied as markers of DN.

Urinary transferrin is a protein with a molecular weight of 76.5 kDa, which is easily filtered through the glomerular barrier of the kidneys. A relationship has been established between urinary excretion of transferrin and the progression of DN, as well as a positive correlation with the albumin-creatinine ratio and a negative correlation with GFR. An increased concentration of this marker is observed with insulin resistance, poor glycemic control and elevated triglyceride levels [1, 7].

Immunoglobulin G (IgG) in urine. IgG antibodies are anionic plasma proteins with a molecular weight of about 150 kDa, which are difficult to filter by the glomerular apparatus of the kidney.

Immunoglobulin M and fibronectin in urine. Fibronectin is a high molecular weight protein that is a component of the extracellular matrix of the glomerular apparatus of the kidneys. IgM is secreted by plasma cells and is the heaviest antibody (molecular weight about 990 kDa) in human blood. Because of its large molecular weight, the appearance of IgM in urine indicates a serious defect in size selectivity in the glomerular capillary wall.

Urine ceruloplasmin. Ceruloplasmin is a negatively charged copper-transporting serum protein that is difficult to filter through the kidney filter. Therefore, the increased content of cerruloplasmin in the urine in patients with type 2 diabetes with normoalbuminuria gives grounds to consider it, along with urinary transferrin and IgG, as a predictive marker of DN.

Hyperglycemia triggers a cascade of metabolic, hemodynamic, immunoinflammatory and profibrogenic processes that promote excessive accumulation of type IV collagen, which causes sclerosis of the glomerular apparatus of the kidneys. At the same time, a close relationship is shown between the level of urinary excretion of collagen and the filtration function of the kidneys, as well as the degree of morphological changes in the tubular, glomerular apparatus of the kidneys and interstitium. Histological examination of kidney biopsy samples demonstrated that the accumulation of type IV collagen is more typical for diabetic glomerulosclerosis than for glomerulopathies of other etiologies. It has been shown that the accumulation of type IV collagen precedes the development of glomerular hypertrophy and

hyperfiltration in the early stages of DN. An increase in its urinary excretion in diabetes has also been demonstrated and a direct relationship has been shown between the level of urinary excretion of type IV collagen and the clinical stage of DN [2, 11].

Laminin in urine. Laminin is a component of the glomerular basement membrane. Its urinary excretion increases in type 2 diabetes even at the stage of normoalbuminuria and correlates with urinary levels of NAG, A1M and type IV collagen.

Glycosaminoglycans in urine. Glycosaminoglycans are components of the glomerular basement membrane. Heparan sulfate is of greatest interest from the glycaminoglycan family in the development of renal pathology. Negatively charged heparan sulfate molecules prevent the diffusion of similarly charged albumin through the kidney filter. Damage to the glomerular apparatus of the kidneys and a decrease in the level of heparan sulfate leads to the loss of negative charge in the membrane and, as a result, it becomes permeable to albumin and other proteins. In patients with diabetes, even with normoalbuminuria, the content of glycosaminoglycans in the urine increases [1, 12].

Lipocalin-type prostaglandin synthase (L-PGDS) is a biomarker reflecting increased capillary permeability of the glomerular wall. Many authors consider it as a predictor marker of kidney damage, and not as an early marker of DN.

Matrix metalloproteinases (MMP, Matrix metalloproteinases) are enzymes of the family of zinc-dependent endopeptidases involved in the processes of degradation of the extracellular matrix. Currently, more than 30 MMPs have been studied, which are characterized by a specific molecular structure, tissue and substrate specificity. The most studied are MMP-2 and MMP-9. They are involved in the destruction of extracellular matrix components and type IV collagen in the kidneys. MMP enzymatic activity is regulated by tissue inhibitors of metalloproteinases (TIMPs). An imbalance in the system of synthesis and destruction of MMPs serves as the pathogenetic basis for the emergence and development of a number of acute and chronic renal pathologies. Long-term diabetes and inadequate glycemic control cause a decrease in the activity and rate of MMP synthesis. This, in turn, provokes a decrease in the rate of catabolism of extracellular matrix products and creates the basis for fibrous degeneration of the glomerular apparatus and interstitium of the kidneys [1, 15].

Immunoinflammatory factors. In recent years, it has been shown that immunoinflammatory reactions play an important role in the pathogenesis of DN. Modern understanding of the formation and course of the inflammatory process is based on the study of the pathogenetic role of cellular mediators of inflammation in it, namely cytokines, which play a key role in the processes of inflammation and tissue regeneration, cell proliferation and apoptosis. Thus, in recent years, the close relationship between the level of proinflammatory cytokines in the blood and urine and

the severity of changes in the kidneys in diabetes has attracted the close attention of researchers [2, 13]. Cytokines are small peptide molecules that carry out short-range regulation of intercellular and intersystem interactions. Cytokines include various types of interleukins (IL, interleukins).

TNF-a is a cytokine that is produced by monocytes, macrophages, T cells, as well as mesangial, glomerular, endothelial, tubular and dendritic cells. TNF-a plays a key role in the occurrence and development of the inflammatory response. Activation of the nuclear transcription factor under the influence of TNF-a causes an increase in the excretion of endothelin-1 by mesangial cells of the kidneys, which in turn leads to activation of the processes of proliferation and sclerosis in the kidney tissue. In mesangial cells, TNF-a induces reactive oxygen species, which leads to a change in the barrier function of the glomerular capillary wall and increases its permeability to albumin [3, 9].

Chemokines. Monocyte chemo-attractant protein-1 (MCP-1, monocyte chemo-attractant protein-1): ischemia of the renal parenchyma in DN triggers a subclinical inflammatory process with a possible outcome in nephrosclerosis. MCP-1 ensures the migration of mononuclear cells and leukocytes to the area of inflammation, and is also involved in the initiation and maintenance of inflammation by stimulating the production of pro-inflammatory cytokines. Thus, increased expression of MCP-1 has been shown in both types of diabetes. In type 1 diabetes against the background of MAU, an increased level of MCP-1 in the blood is observed, compared with healthy individuals and diabetic patients at the stage of normoalbuminuria. Also shown is increased urinary excretion of MCP-1 in type 1 diabetes, even against the background of normalbuminuria, and its correlation with the clinical stage of AC. Thus, monitoring of urinary excretion of MCP-1 can be used to assess the process of fibrogenesis in the interstitium of the kidneys both in chronic nephropathies and in their traumatic lesions [3, 6].

Orosomucoid (or alpha 1-acid glycoprotein, UOER) is a glycoprotein involved in inflammatory responses. Urinary levels of orosomucoid are significantly increased in patients with DN compared to healthy individuals, and there is a direct correlation between its urinary excretion and the clinical stage of DN. In addition, a direct relationship has been shown between urinary excretion of orosomucoid and albumin, as well as serum levels of creatinine and C-reactive protein [1, 7].

Biomarkers of oxidative stress. Oxidative stress (OS) is a state of imbalance between pro- and antioxidants in biological systems (cells, tissues and organs) towards the predominance of oxidants. It contributes to the emergence and development of various pathological conditions. It is now generally accepted that OS initiated by hyperglycemia is an important pathogenetic link in the occurrence and development of

diabetic vascular complications. In particular, it has been proven that OS is a key link in the pathogenesis and progression of DN [2, 8].

MicroRNAs are short (21-25 nucleotides) non-coding RNA molecules. They function to regulate post-transcriptional gene expression and block protein translation and/or promote the destruction of messenger RNA (mRNA). Currently, five microRNAs specific for renal pathology are known, which are classified according to the place of synthesis - mainly in the cortical layer and mainly in the medulla. Their quantitative determination in urine and blood can be used for early diagnosis of DN. The molecular mechanisms that initiate the development of DN are different in different patients, so it is of particular interest for specialists to study the individual microRNA profile of each patient, since this can contribute to the development of a personalized approach to the diagnosis and monitoring of treatment of DN [2, 5].

Conclusions. Currently, the possibilities for diagnosing and predicting the development of diabetic kidney damage have expanded significantly and make it possible to diagnose them at the preclinical stage. Now, using molecular diagnostic methods, it is possible to predict the risk of developing nephropathy before the appearance of MAU (the reference marker of DN). At the same time, to increase the accuracy of early diagnosis, personalized therapy and prevention of DN, clinical examination of patients should include an optimal combination of biomarkers that reflects pathological processes in the glomerular and/or tubular parts of the kidneys, or indicates damage to the interstitium. However, the clinical significance of some potential markers of DN needs clarification. Efforts to diagnose DN at the preclinical stage are necessary to identify groups at high risk of developing nephropathy among patients with diabetes in order to be able to choose effective preventive personalized therapy using modern glucose-lowering drugs with a nephroprotective effect.

LITERATURE

1. Шестакова М.В. Сахарный диабет и хроническая болезнь почек: возможности прогнозирования, ранней диагностики и не-фропротекции в XXI веке. Терапевтический архив. 2016;6:84-8. doi:10.17116/terarkh201688684-88.
2. Маркова Т.Н., Садовская В.В., Беспятова М.Ю. Современные возможности диагностики хронической болезни почек при сахарном диабете. Сахарный диабет. 2017; 20(6): 454-60. doi: 10.14341/DM9268.
3. Захарова Н.Б., Пастушкова Л.Х., Ларина И.М., Каширина Д.Н., Лях Р.Н., Попков В.М. Значение протеомного состава мочи при заболеваниях мочевыводящих путей (обзор литературы). Экспериментальная и клиническая урология. 2017;(1):22-9.
4. Даминов Ф. А. и др. Диагностика и лечение интраабдоминальной гипертензии при ожоговом шоке //Журнал Неотложная хирургия им. ИИ Джанелидзе. – 2021. – №. S1. – С. 19-20.

5. Kudratova Z. E.Isomadinova L. K.Sirojeddinova S. F. Tursunova M. E.Current modern etiology of anemia. novateur publications international journal of innovations in engineering research and technology. № 10. 2023, P. 1-4.
6. Isomadinova L.K. Qudratova Z.E. Shamsiddinova D.K.Samarqand viloyatida urotiliaz kasalligi klinik-kechishining o'ziga xos xususiyatlari. Central asian journal of education and innovation №10. 2023 , P. 51-53
7. Ибрагимова Н. и др. ДИАГНОСТИКА, ЛЕЧЕНИЕ И ПРОФИЛАКТИКА САХАРНОГО ДИАБЕТА 2-го ТИПА //Центральноазиатский журнал академических исследований. – 2024. – Т. 2. – №. 1. – С. 9-13.
8. Ruziboev S. A., Daminov F. A. Our experience in treatment of stress ultrasons in severe burned //Euro-Asia Conferences. – 2021. – Т. 1. – №. 1. – С. 447-449.
9. Nabiyeva F. S., Ibragimova N. S., Diamatova D. N. 2-TIP QANDLI DIABET KECHISHINING O'ZIGA XOS XUSUSIYATLARI //TADQIQOTLAR. UZ. – 2024. – Т. 31. – №. 1. – С. 28-32.
10. Berdiyarova Sh.Sh., Ahadova M.M., Ochilov S.A. COMPLICATIONS OF TREATMENT OF ACUTE HEMATOGENOUS OSTEOMYELITIS, LITERATURE REVIEW, Galaxy International Interdisciplinary Research Journal 293-298
11. Бердиярова Ш.Ш., Юсупова Н.А., Ширинов Х.И. Клинико-лабораторная диагностика внебольничных пневмоний у детей, Вестник науки и образования, 80-83
12. Kudratova Zebo Erkinovna, Karimova Linara Alixanovna Age-related features of the respiratory system // ReFocus. 2023. №1. URL: https://cyberleninka.ru/article/n/age-related-features-of-the-respiratory-system.
13. Sabirovna I. N. et al. POSSIBILITIES OF RADIATION DIAGNOSTICS FOR CORONAVIRUS INFECTION //TADQIQOTLAR. UZ. – 2024. – Т. 31. – №. 2. – С. 90-94.
14. Nabiyeva F. S. et al. CREATION OF OPTIMUM CONDITIONS FOR PROPAGATION OF SACCHAROMYCES CEREVISIAE YEAST //Journal of new century innovations. – 2023. – Т. 23. – №. 1. – С. 85-91.
15. Sabirovna I. N., Raykhona K. CLINICAL AND LABORATORY CHANGES IN POST-TERM INFANTS //Web of Medicine: Journal of Medicine, Practice and Nursing. – 2024. – Т. 2. – №. 5. – С. 96-99.
16. Isomadinova L.K, Qudratova Z.E., Babaxanova F.Sh.clinico-laboratory features of the course of covid-19 with hepatitis b journal of new century innovations №-3. 2023 P. 60-65.
17. Daminov F. A., Tagaev K. R. Diagnosis, treatment and prevention of erosive-ulceral diseases of the gastrointestinal tract in heavy bears //Journal of Advanced Research in Dynamical and Control Systems. – 2020. – Т. 12. – №. 7 Special Issue. – С. 150-153.