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**Chapter**

# Prevention and Treatment of Diabetic Nephropathy with Vitamin D

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## Abstract

The number of people suffering from diabetes mellitus, especially Type 2 diabetes mellitus, is increasing every year. Approximately one-third of the patients with diabetes mellitus will develop diabetic nephropathy and chronic kidney disease. Diabetic nephropathy represents the main cause of end-stage renal disease. Vitamin D deficiency is often present in patients with diabetes mellitus and could present a risk factor for a higher incidence of cardiovascular events. Vitamin D supplementation could have a renoprotective effect and the potential to delay occurrence and slow down the progression of diabetic nephropathy. The renoprotective effect is reflected in better glycoregulation, reduction of proteinuria and proinflammatory cytokines, and improved lipid regulation. New research shed the light on the important role of vitamin D in reducing renal fibrosis and stabilization of podocyte function. If we take into consideration the cost of end-stage renal disease treatment and the quality of life of patients on dialysis, any delay in end-stage renal disease is significant.

**Keywords:** vitamin D, diabetes mellitus, diabetes nephropathy, podocyte, urinary biomarker

## 1. Introduction

Diabetes mellitus (DM) is a chronic metabolic disease and one of the leading health problems in the world. The incidence is constantly increasing, not only in the developed countries of western Europe and America but also in the developing countries due to abrupt changes in lifestyle [1, 2]. According to the 2019 Global Burden of Disease project, DM is the fifth leading cause of death and the eighth leading cause of disability worldwide, with an increasing prevalence in low-middle income countries (LMICs) [1]. In addition, LMICs have a higher percentage of undiagnosed diabetes than in high-income countries (80–90% in sub-Saharan Africa versus 20–30% in western Europe and North America) [1].

According to the International Diabetes Federation (IDF) data for 2021, there are 537 million adults (aged 20–79) living with DM in the world. This number is projected to rise to 643 million by 2030 and 783 million by 2045. More than 3 out of 4 adults with DM live in LMICs. In 2021, DM is responsible for 6.7 million deaths in adults

aged 20–79 years, excluding the risk of mortality associated with the COVID-19 pandemic. This corresponds to 12.2% of the global deaths of all causes in this age group. Of course, the percentage of mortality varies across different regions. Approximately one-third (32.6%) of all diabetic deaths are people of working age (under 60). This corresponds to 11.8% of the total number of global deaths in people under the age of 60. It is estimated that one person dies of DM every 5 seconds [3].

The cost of treating DM patients and its complications is very high. During 2021, at least $966 billion was spent on DM therapy, which represents an increase of 316% in the last 15 years [3].

About 541 million adults have impaired glucose tolerance (IGT) and therefore a high risk of developing Type 2 diabetes mellitus (T2DM). Life expectancy prolongation as well as the global increase in obesity strongly affects the increase in the prevalence of T2DM [3–5].

The prevalence of DM is similar between the sexes. Although according to IDF data, the estimated prevalence of DM in men, aged 20–79 years, is slightly higher than in women (10.8% vs 10.2%). Only at the age above 70 years, the percentage of female patients is higher, presumably due to the longer women's lifespan [3].

About 87–91% of DM patients have T2DM, which is also caused by the growing epidemic of obesity worldwide. The frequency depends on genetic predisposition and environmental factors. Risk factors for the development of T2DM are obesity, a positive family history, belonging to certain ethnic groups, female sex, low body weight at birth, excessive weight in the adolescent period, and gestational diabetes of the mother during pregnancy increases the risk of the newborn developing DM in the later period. An increase in the incidence of T2DM is found in an increasingly young population [1, 3, 6].

The incidence of type 1 DM (T1DM) is also on the rise, and the lower age limit is constantly lowering, most commonly occurring in Europe and North America [3]. Risk factors for T1DM are genetic predisposition, viral infections, especially enterovirus infections, immunization (only in genetically predisposed), diet, exposure to cow's milk in early life, higher socioeconomic status, obesity, vitamin D deficiency, and perinatal factors, such as maternal age, preeclampsia history, and neonatal jaundice. Contrary to T2DM, low birth weight reduces the risk of developing T1DM [3, 7].

A lot of complications develop during DM. Besides the changes in large blood vessels, small blood vessels are also affected called microvascular complications. Microvascular complications arise from changes in the arterioles, capillaries, and venules, which are responsible for the control of the permeability of blood vessels and the muscle tone that adjusts blood flow to the metabolic needs of tissues. DM causes pathognomonic changes in small blood vessels, affecting the basal membrane of arterioles in the glomerulus, retina, myocardium, skin, and muscles, leading to its thickening [8]. One of the most common microvascular complications is diabetic nephropathy (DN) or diabetic kidney disease, which develops in just over a third of DM patients. Diabetic nephropathy is the leading cause of end-stage renal disease (ESRD) in the world [9, 10].

## 2. Diabetic nephropathy

DN is a chronic microvascular complication of DM, manifested as a clinical syndrome characterized by persistent albuminuria (urine albumin-to-creatinine ratio (UACR) > 300 mg/g), progressive reduction in glomerular filtration rate (GFR),

hypertension, and an increase in the incidence of cardiovascular events and cardiovascular fatal events [11]. In addition to the term diabetic nephropathy, Dr. Krolewski introduced the term diabetic kidney disease in 1995 to indicate clinically diagnosed kidney disease caused by DM, and the term DN should be reserved for pathohistologically confirmed kidney damage by DM; the same attitude is expressed by the Japanese Society of Pathology and the Japanese Society of Nephrology [12]. Both terms are still used in the literature. Based on previous research, we distinguish two forms of diabetic nephropathy: proteinuric diabetic nephropathy (P-DN) and non-proteinuric diabetic nephropathy (NP-DN) [13].

## 2.1 Pathophysiological mechanisms in diabetic nephropathy

The two main risk factors for developing DN are poor glyco-regulation and duration of DM. The mechanisms by which hyperglycaemia causes DN are complex and numerous, with renal haemodynamics disorder, impaired glucose metabolism, ischaemia, increased oxidative stress, inflammation, and increased activity of the renin-angiotensin-aldosterone system (RAAS) at the kidney level being the most important [14].

When glycoregulation is normal, 90% of the glucose filtered in the glomeruli is reabsorbed in the S1 segment of the proximal tubule via the sodium-glucose cotransporter-2 (SGLT2) receptors on the brush epithelium, and only 10% via the sodium-glucose cotransporter-1 (SGLT1) receptors located in the distal parts of the proximal tubules. In hyperglycaemia, a large amount of glucose is filtered in the glomeruli resulting in hypertrophy of the proximal tubule due to an attempt of the proximal tubule to reabsorb this glucose. The absorption of glucose is associated with sodium chloride absorption via the SGLT2 receptor. Due to the hypertrophy of the proximal tubule glucose and sodium chloride, reabsorption are increased, resulting in a smaller amount of sodium chloride arriving in the distal tubule, which is recognized by macula densa as hypovolemia and consequently reduces tubuloglomerular feedback (TGF). TGF is the mechanism by which the macula densa controls the vasoconstriction of the afferent arteriole by releasing adenosine, and other signalling molecules. By blocking TGF, vasodilatation of the afferent arteriole occurs via vasoactive mediators, such as insulin-like growth factor-1 (IGF-1), nitric oxide (NO), prostaglandins, and glucagon. At the same time, due to the increased local effect of angiotensin II in hyperglycaemia, increased vasoconstriction of the efferent arteriole and the consequent disruption of autoregulation occur, resulting in the development of intraglomerular hypertension. Thus, the "sodium paradox" is expressed in DN, the higher the sodium chloride restriction in the diet, the smaller the amount of sodium chloride that arrives in the distal tubule and the GFR increases. GFR may be increased up to 40%. In normoglycemia, changes in systemic perfusion pressure are not transferred to intraglomerular pressure due to autoregulation, but since autoregulation is compromised in DM, systemic perfusion pressure is freely transferred to the capillary glomerular network [14, 15].

Functional changes in blood flow and glomerular pressure provoke structural changes. Increased intraglomerular pressure damages the endothelial surface of glomerular capillaries, disrupts the normal structure of the glomerular barrier, and induces the expression of cytokines and growth factors that cause the increased synthesis of collagen, fibronectin, and laminin. Hyperfiltration is associated with glomerulomegaly and renal enlargement. Glomerular enlargement is due to an increase in the number of capillary loops with a consequent increase in the filtration area and enlargement of the mesangial matrix [16].

Changes also affect tubular cells, besides glomeruli, such as proliferation and increase in cellular size. Mechanisms by which increased blood glucose levels induce hypertrophy to include stimulation and increased expression of multiple renal growth factors, including IGF-1, epidermal growth factor, platelet-derived growth factor (PDGF), vascular endothelial growth factor (VEGF), and transforming growth factor-Beta 1 (TGF-β1) [16–18].

The following section will briefly describe the main pathogenetic mechanisms in DN.

### 2.1.1 The role of protein kinase C

One of the main side effects of hyperglycaemia is the increased activation of protein kinase C (PKC). PKC is an enzyme, a member of the serine-threonine-protein kinase family that regulates various vascular functions, including contractility, blood flow, cellular proliferation, and vascular permeability. PKC activity in DM patients is particularly increased on the surface of the retina, aorta, heart, and glomerular cells including podocytes, presumably due to increased de novo synthesis of diacylglycerol, being the main endogenous PKC activator [19, 20].

Several isoforms of PKC exist, however, PKCα is considered to be the most important for DN development. The main role of PKCα is in increased endocytosis of nephrin, which is one of the main components of the slit diaphragm (SD). Increased expression of PKCα on podocytes in DM patients increases nephrin endocytosis leading to SD instability [20, 21]. The PKCβ isoform is responsible for hyperglycaemia-induced renal fibrosis via the upstream regulation of TGF-β1, which contributes to the development of glomerular fibrosis. The PKC is form activated by advanced glycation end products (AGEs) leads to increased expression of VEGF164 and VEGF165 isoforms at the podocytes, which leads to structural and functional renal changes seen in animal models of DN, including proteinuria, glomerular hypertrophy, thickening of the glomerular basal membrane, mesangium proliferation, and loss of SD and foot extensions of podocytes [18–20].

### 2.1.2 Effect of advanced glycation end products

Chronic hyperglycaemia leads to non-enzymatic glycosylation of amino acids and proteins known as AGEs. AGEs mediate various cellular activities, including molecule expression and adhesion, cellular hypertrophy, extracellular matrix synthesis, epithelial-mesenchymal transformation of tubular cells, and inhibition of nitrogen monoxide synthesis. They can bind to a variety of cell types, including macrophages and mesangial cells. Binding to macrophages leads to increased production and release of proinflammatory mediators: tumor necrosis factor alpha (TNF-α), interleukins, PDGF, and IGF-1. Released mediators stimulate mesangial cells to increase mesangial matrix synthesis, which contributes to glomerulosclerosis. AGEs stimulate proximal tubule epithelial cells to increase TGF-β1 synthesis and its release, leading to tubulointerstitial fibrosis [18, 20, 22].

### 2.1.3 Effect of sorbitol

Hyperglycaemia stimulates the polyol pathway of glucose metabolism and increased sorbitol production. Chronic hyperglycaemia leads to increased glucose uptake in

tissues that do not require insulin for glucose uptake, such as retina, eye lens and kidney. Increased glucose concentration in the tissue leads to sorbitol accumulation. The deposition of sorbitol in the epithelial cells of the proximal tubules leads to decreased free myo-inositol and decreased activity of sodium potassium adenosine triphosphatase (Na+/K+-ATP-ase), an increase in the consumption of the nicotinamide dinucleotide phosphate (NADPH) and nicotinamide dinucleotide (NAD+), which results in a change in cell oxidative potential and disruption of cell function [18, 20, 22].

### 2.1.4 Renin-angiotensin-aldosterone system in diabetic nephropathy

Local RAAS activity is increased in DM patients, that is, activity is increased in the glomerulus and renal vessels. Prorenin production is increased and prorenin binds to specific prorenin-renin receptors, followed by non-enzymatic activation of prorenin into renin, then renin induces local production of angiotensin II. The expression of angiotensin II type 1 receptor (AT1R) is increased, through which angiotensin II, as a potent vasoconstrictor, induces vasoconstriction of the efferent arteriole, which, in addition to dilatation of the afferent arteriole, leads to increased hydrostatic pressure in the glomerular capillaries and increased glomerular filtration. Increased activity of angiotensin II also leads to an increase in systemic blood pressure. Angiotensin II also directly stimulates renal production of TGF-β1 in the mesangium and epithelial tubular cells and stimulates the production of other cytokines and growth factors in renal cells, such as endothelin-1, monocyte chemoattractant protein-1 (MCP-1), interleukin-6 (IL-6), regulated upon activation, normal T cell expressed and presumably secreted (RANTES), and osteopontin. In addition to lowering intraglomerular pressure, angiotensin-converting enzyme inhibitors (ACEi) reduce renal expression of TGF-β1. Angiotensin II increases the release of VEGF from podocytes, which contributes to the disruption of the filtration barrier and the progression of proteinuria. All released mediators contribute to the development of glomerulo-sclerosis and tubulointerstitium fibrosis. The aldosterone release is increased, which also accelerates the progression of renal damage independent of angiotensin II. Several clinical studies have shown that blocking the mineral corticosteroid receptor lowers proteinuria [14, 18, 22].

### 2.1.5 Inflammation and diabetic nephropathy

DN was previously defined as a non-inflammatory disease. However, the results of numerous clinical studies show that activation of the immune system and chronic inflammatory processes play a very important role in the development of DN [20, 23].

The most significant inflammatory cells involved in the development of DN are macrophages (M1 and M2 macrophages) and T lymphocytes. M1 macrophages activated by Th1 cells increase the inflammatory response by increased expression of cytokines (interleukins, TNF-α, and interferon γ), unlike M2 macrophages (activated by Th2 cells), which lead to increased expression of anti-inflammatory cytokines. M1 macrophages increase the inflammatory response also by increasing the production of free oxygen radicals [14, 18, 23].

Numerous chemokines are involved in the inflammatory response in DN, the most significant and first described is MCP-1, which also plays an important role in the early development of atherosclerosis. MCP-1 induces the transformation of

monocytes into macrophages, which then increasingly produce IL-6 and TNF-α, leading to initial atherosclerotic changes in the blood vessel wall, resulting in disease progression [14, 18, 20, 23].

In DM patients, the secretion of proinflammatory cytokines, interleukin-1 (IL-1), interleukin-6 (IL-6), and interleukin-18 (IL-18), is increased. IL-1 induces increased expression of adhesion molecules in glomerular endothelial cells and other renal structures, leading to changes in glomerular haemodynamics, vascular permeability, and increased chemokine expression, resulting in proliferation and synthesis of extra-cellular matrix in the mesangium. IL-6 in DN has pleiotropic effects, the proliferation of the extracellular matrix, thickening of the glomerular membrane, and is considered important for the progression of DN. Local IL-18 in the kidney DM patients is secreted by T lymphocytes, macrophages, monocytes, and proximal tubule cells. It increases the expression of intracellular adhesion molecule 1 (ICAM-1), as well as the production of other inflammatory cytokines in mesangial cells, and is responsible for endothelial apoptosis. A direct correlation between IL-18 and increased urinary excretion of albumin has been confirmed, some authors even consider it as an early indicator of DN [14, 18, 20, 23].

TNF-α has multiple roles in the inflammatory response, and it is synthesized by monocytes, macrophages, and T lymphocytes, as well as by some renal cells. It leads to the activation of secondary messengers, transcriptional factors, growth factors, adhesion molecules, and the expression or synthesis of other cytokines. TNF-α as well as the before mentioned interleukins in renal structures changes the haemodynamics within glomeruli, increases vascular permeability, increases infiltration by inflammatory cells, and increases extracellular matrix production and production of reactive oxygen species (ROS) [14, 18, 20, 23].

Adhesion molecules ICAM-1 and vascular cell adhesion molecule-1 (VCAM-1) allow the leukocytes to bind to the vascular wall and their infiltration of tunica intima, where they secrete proteolytic enzymes responsible for the tissue damage and the development of atherosclerosis [18].

### 2.1.6 Oxidative stress in diabetic nephropathy

Oxidative stress plays a significant role in the development of microvascular complications of DM. Metabolic abnormalities in DM, and above all hyperglycaemia leads to increased production of ROS in mitochondria. Increased production of ROS at the mitochondrial level causes activation of the five main pathways involved in the development of microvascular complications of DM: polyol pathway, increased AGEs production, increased AGEs receptor expression and ligand activation, PKC isoform activation, and increased hexosamine pathway activity. Through these pathways, the increased concentration of intracellular ROS causes a change in angiogenesis in response to ischemia, activates numerous proinflammatory pathways, and causes long-acting epigenetic changes. These epigenetic changes lead to persistent expression of proinflammatory genes also after normalization of blood glucose (so-called hyperglycaemic memory) [14, 18, 20].

Persistent expression of proinflammatory genes is achieved through multiple signalling pathways; however, the importance of the JAK/STAT pathway is particularly emphasized [20, 24].

## 2.2 Diagnosis and clinical manifestation of diabetic nephropathy

### 2.2.1 Diagnosis and clinical manifestation of proteinuric diabetic nephropathy

Diagnostic criteria for proteinuric diabetic nephropathy (P-DN) include UACR of 30 mg/g or greater [11]. According to the recommendations of the American Diabetes Association (ADA) and the National Kidney Foundation Kidney Disease Outcomes Quality Initiative (NKF KDOQI), a positive finding in at least two out of three urine samples, 3–6 months apart, is required for the diagnosis of persistent microalbuminuria [25]. Albuminuria can be determined in a spot sample of morning urine from which the UACR is calculated. UACR for normal albuminuria is < 30 mg/g, for microalbuminuria UACR is between 30 and 300 mg/g, for macroalbuminuria UACR is > 300 mg/g. Albuminuria can be calculated also from a 24-hour urine sample, where albuminuria up to 150 mg/24h is considered normoalbuminuria, microalbuminuria from 150 to 300 mg/24h, and macroalbuminuria over 300 mg/24h [11].

DN develops through five different phases: 1st phase glomerular hyperfiltration, 2nd phase normoalbuminuria, 3rd phase microalbuminuria, 4th phase macroalbuminuria, and 5th phase renal insufficiency (GFR less than 60 mL/minute/1.73 m$^2$). These five phases are clearly separated in T1DM, while in T2DM when diagnosed, microalbuminuria may be present, due to the long silent lasting of hyperglycaemia [11]. Changes in the first three phases are reversible, especially in patients with T2DM. Japanese authors reported regression of microalbuminuria in almost 50% of the patients followed [26]. Japanese and other authors published that regression was positively correlated with glycated haemoglobin A1c (HbA1c) values, systolic blood pressure, and duration of microalbuminuria [26, 27].

The occurrence of microalbuminuria and macroalbuminuria in addition to kidney damage is an independent risk factor for cardiovascular diseases. P-DN is developed in patients with T1DM and T2DM. In patients with T1DM, DN occurs after a longer disease evolution, over 10 years, and is usually associated with diabetic retinopathy (DR). In T2DM, DN can be diagnosed immediately after DM diagnosis, even without DR [11, 13].

### 2.2.2 Diagnosis and clinical manifestation of non-proteinuric diabetic nephropathy

Some DM patients have a reduction in GFR of less than 60 mL/minute/1.73 m$^2$ without microalbuminuria. This type of DN is called non-proteinuric diabetic nephropathy (NP-DN). NP-DN is more commonly associated with female sex, hypertension, poor glyco-regulation, smoking, and use of RAAS blockers. It occurs more often in patients with T2DM. NP-DN shows a weak correlation with retinopathy but shows a linear correlation with the percentage of cardiovascular complications, that is, with macroangiopathy [12, 13, 28].

According to the results of the DEMAND study in the population of patients with T2DM and creatinine clearance lower than 60 ml/min/1.73m2, about 40% of patients had normal albumin secretion. These individuals were shown to have a higher degree of insulin resistance, a higher level of serum triglycerides and total cholesterol, and low-density lipoprotein (LDL), that is, a more pronounced metabolic syndrome compared to those with preserved GFR [29]. In his work, Krolewski proposes the theory that NP-DN occurs primarily as part of inflammation and tubular lesions, while

Reutens believes that repeated and untreated episodes of acute renal impairment play a key role in NP-DN [30, 31]. Authors published that endothelial dysfunction and increased vascular resistance in interlobar renal arteries are important for the pathogenesis of NP-DN, which can be monitored by measuring the renal resistance index if elevated in patients with DM a more intensive treatment of DM should be started even before decreasing GFR [20, 32, 33].

Based on several studies, it is concluded that patients with NP-DN compared to P-DN still have a slower GFR decrease, better blood pressure control, and a lower risk of developing ESRD [13, 27, 28].

## 2.3 Pathohistological changes in diabetic nephropathy

Pathohistological changes of DN are thickening of the glomerular basal membrane, mesangium proliferation, glomerulomegaly, nodular glomerular sclerosis (Kimmelstiel-Wilson nodule), hyalinosis lesions characterized by exudative/insudative lesion and fibrin cap, arteriolar hyalinosis, and reduction in podocyte count with loss of foot extensions. Nodular glomerular sclerosis is characteristic of PN-DN. Tubulointerstitial basal membrane thickening, tubular atrophy, interstitial peritubular fibrosis, and atherosclerosis occur at the tubulointerstitium level.

Tervaret et al. proposed a classification of morphological changes in DN and divided into IV classes:

Class I: Thickening of the glomerular basal membrane—changes in GBM are present with the absence of mesangium expansion, as well as nodular changes in the mesangial matrix.

Class II: Mesangium proliferation—defined as the increase of the mesangium extracellular matrix so that the width of the interspace reaches two nuclei of mesangium cells in at least two glomeruli. Mesangium proliferation may be mild (IIa) and severe (IIb).

Class III: Nodular sclerosis (Kimmelstiel-Wilson nodes)—focal, oval, or round mesangium lesions composed of an acellular hyalinized nucleus that is surrounded by rare crescent mesangial nuclei. It is sufficient to see one clear Kimmelstiel-Wilson change in the preparation and if there is no more than 50% global glomerulosclerosis, the changes are classified as Type III.

Class IV: Global (diffuse) diabetic glomerulosclerosis—the final stage characterized by the accumulation of extracellular matrix proteins in the mesangium space with the formation of numerous Kimmelstiel-Wilson changes [34].

These changes can be seen in PN-DN and NP-DN. However, in NP-DN, changes in glomerulus are less pronounced, smaller number of patients have changes characteristic of class III and class IV, while changes in tubules have similar prevalence in both forms of DN [13, 28].

## 3. Vitamin D and diabetic nephropathy

Considering the burden of constantly increasing DM, with more than a third of the patients developing DN, and the fact that DN is the leading cause of ESRD, it is important to find other possible mechanisms and risk factors for the development of DN. Since the costs of ESRD treatment by haemodialysis or kidney transplantation are

extremely high, constant research is being conducted to prevent the development of DN or slow its progression, and to find biomarkers for early diagnosis of DN.

Vitamin D deficiency is becoming pandemic and is found in different age populations. Vitamin D deficiency is not recognized and not adequately treated in many countries. A vitamin D deficiency is present in more than one billion people worldwide. The prevalence of vitamin D deficiency is high in countries around the world. In Italy, the prevalence in the general population is 17%, in Spain 33.9%, in Germany 50%, while in the UK, it is 87.1%. Vitamin D deficiency is associated with an increase in morbidity for cardiovascular, carcinogenic, metabolic—DM, immunological, psychological, and other chronic diseases [35].

For the normal function of vitamin D, the kidney plays an important role. Vitamin D is transported bound to vitamin D binding protein (VDBP). After the first hydroxylation is carried out in the liver, the resulting 25-hydroxyvitamin D3 (25(OH)D3) is transported to the kidney bound to VDBP, where it is first filtered in the glomerulus and then reabsorbed through the receptors on the brush border of the proximal tubule. In the kidney, in the presence of 1-alpha-hydroxylase (CIP27B1), the second hydroxylation is carried out to form a more polar compound 1,25-hydroxyvitamin D3 (1,25(OH)2D3), that is, calcitriol, being a physiologically active form of vitamin D, and based on its physiological effects, vitamin D is classified as a vitamin and as a hormone [36, 37].

Multiple studies have shown that the prevalence of 25(OH)D3 deficiency is significantly higher in patients with T1DM and T2DM compared to the healthy population. Complementary, a significant negative correlation has been found between the concentration of 25(OH)D3 and HBA1c (r = -0.277, p < 0.0001) [34]. Also, several studies have shown a significantly higher risk of developing T2DM in individuals with a concentration of 25(OH)D3 of $\leq$ 20 ng/mL ($\leq$50 nmol/L). Studies also showed a significantly strong positive correlation between 25(OH)D3 concentration and GFR. Patients with DM and concentration of 25(OH)D3<15ng/mL (<37,5 nmol/L) have a faster GFR reduction compared to patients with a concentration of 25(OH)D3>15 ng/mL (>37,5 nmol/L) [38, 39].

Patients with microalbuminuria and macroalbuminuria have significantly lower serum concentrations of 25(OH)D3 than patients with normoalbuminuria [40].

In lean DM patients, the risk of vitamin D deficiency is 25% higher than in the general population, while in obese DM patients, the risk is 35% higher. Females with DM more often have vitamin D deficiency, which is explained by the higher fat content in women with consequent formation of vitamin D depot in the body [35, 38].

DM patients have multiple risk factors for vitamin D deficiency. In DN, vitamin D metabolism may be disrupted due to major loss of VDBP as part of proteinuria, which may also be of nephrotic rank. Also, 25(OH)D3 filtered in glomerulus bound to VDBP can be reabsorbed to a significantly lesser extent at the level of proximal tubules due to their damage, which contributes to vitamin D deficiency [38].

Previous studies showed that vitamin D may have an anti-inflammatory, immunomodulatory, and hypoglycaemic effect and may affect later occurrence and slower progression of DN. These are renoprotective effects of vitamin D that are assumed that can delay the occurrence and slow down the development of DN

## 3.1 Effect of vitamin D on glycoregulation

The vitamin D prevents the occurrence of DM by increasing insulin sensitivity by stimulating and increasing the expression of insulin receptors in the skeletal muscles

with the activation of the peroxisome proliferator activator receptor δ (PPAR δ), and by inhibiting RAAS (inhibitor of the insulin effect on peripheral tissues) [36]. Also, vitamin D affects increased insulin release by increasing intracellular calcium in pancreatic beta cells. Vitamin D can also have a direct effect on beta cell function, which is done by binding its active form in the blood to the vitamin D receptor (VDR), which is present in the beta cells of the pancreas. Mice without functional VDR have been shown to have impaired insulin secretion after glucose load [38, 41, 42]. Vitamin D also increases the level of osteocalcin synthesized in the osteoblasts, which in turn increases insulin synthesis in the pancreas and reduces the inflammation responsible for insulin resistance [38, 42]. It is considered that an optimal blood level between 80 and 119 nmol/L is required for vitamin D to achieve its effect on lowering insulin resistance [35, 43].

The use of vitamin D can reduce the risk of diabetes to a certain extent [39, 42, 44]. Thus, the observational study in Denmark "Copenhagen City Heart Study", conducted on 9841 non-DM patients followed for 29 years, showed that patients who had an average value of 25(OH)D <12.5 nmol/L (< 5 ng/ml) had a 22% higher risk of developing T2DM compared to patients with serum concentration of 25(OH)D ≥50 nmol/L (≥20 ng/ml), patients who had 25(OH)D ≥75 nmol/L (≥30ng/ml) had an 8% lower risk of morbidity compared to patients whose 25(OH)D level was between 50 and 75 nmol/L (20–30 ng/ml) [45].

A double-blind, randomized, placebo-controlled study of 118 patients with T2DM and vitamin D deficiency showed that 8-week combined administration of vitamin D at a dose of 50000 IU/week and 1000 mg/day of calcium was significantly more effective in reducing the levels of morning glycaemia and HbA1c compared to the effect of administration of individual components or placebo [46].

Petrovic et al. conducted a prospective cohort study on 90 patients with T2DM and vitamin D deficiency in whom vitamin D supplementation was conducted for six months (at the beginning, cholecalciferol drops supplementation was started at the dose of 20,000 IU twice a week). After two months in patients who normalized vitamin D levels, the supplementation was continued with 5000 IU of cholecalciferol twice a week for the next four months, and in patients whose vitamin D levels remained reduced, the substitution was continued with 20,000 IU twice a week. The study showed that vitamin D supplementation for six months led to a statistically significant reduction of HbA1c in all patients regardless of the degree of albuminuria [40].

## 3.2 Anti-inflammatory effect of vitamin D

Chronic inflammation is considered to play a central role in the development of DM and DN, and vitamin D can directly or indirectly reduce its effect [23]. The anti-inflammatory effect is reflected in the reduced release of numerous proinflammatory cytokines, such as TNFα, IL-6, IL-12, IL-18, IL-1β, interferon-gamma (INF-γ), dendritic cell differentiation blockade, inhibition of lymphocytic proliferation, inhibition of foam cell formation, reduced cholesterol uptake into macrophages, and improved regulatory development of T lymphocytes (Th1 inhibition and Th2 lymphocyte activation) or in increased release of anti-inflammatory cytokines, such as IL-10. Vitamin D also induces CD4+ and CD25+ regulatory lymphocytes that inhibit inflammation and inhibit the effects of TNF-α, ICAM, and VCAM-1 [20, 38, 47].

All of the above mechanisms affect the slower development of atherosclerotic changes and the lower incidence of adverse cardiovascular events.

### 3.3 Effect of vitamin D on proteinuria

Several authors reported the anti-proteinuric effect of correcting vitamin D deficiency in DM [40, 48–50]. Since proteinuria is the main treatment target in terms of slowing DN progression, this effect should be highlighted. The anti-proteinuric effect of vitamin D is done by the inhibition of RAAS in the juxtaglomerular kidney apparatus by downstream regulation of gene transcription, due to binding to the transcription factor cyclic adenosine monophosphate-response element (CRE)-binding protein and disabling renal transcription. In addition to the inhibition of renal RAAS, vitamin D lowers the expression of renin at the heart level, thus decreasing blood pressure. De Zeeuw confirmed the anti-proteinuric effect of vitamin D in the VITAL study, similar results were published by other authors [40, 49, 50]. In addition to the effect on renin, the hypotensive effect is also due to the direct effect on vascular cells and calcium metabolism. With these mechanisms, vitamin D leads to better control of blood pressure and intraglomerular pressure [38]. According to the literature, a negative correlation was found between serum vitamin D concentration and blood pressure level [51]. The antiproteinuric effect is also achieved by improving glyco-regulation, anti-fibrotic effect—lowering the activity of TGF-$\beta$1 in the urine, increasing the expression of nephrin at the kidney level, reducing levels and other growth factors that interfere with SD and podocyte function, such as VEGF-A and MCP-1 [38]. In addition, slowing fibrosis prevents and slows the progression of left ventricular hypertrophy and the development of cardiac weakness and reduces the expression of genes involved in atherosclerosis as well as vascular growth factors [38, 52].

### 3.4 Effect of vitamin D on lipid status

Correction of vitamin D deficiencies in patients with T2DM improves lipid status, and thus, can prevent cardiovascular complications [40, 53]. Xiao Fei Qin et al. have demonstrated that the administration of vitamin D in patients with reduced vitamin D levels and hyperlipidaemia, after use of statins and vitamin D (2000IU/day) together for six months, leads to a significant reduction in both triglycerides and cholesterol levels compared to patients who used placebo with statins [54]. PalomaMuñoz-Aguirre also achieved a significant reduction in triglyceride levels after administration of vitamin D at a dose of 4000 IU/day in postmenopausal women with T2DM [55]. Other authors have also confirmed a significant decrease in cholesterol and triglyceride levels after vitamin D supplementation in patients with T2DM and vitamin D deficiency [40]. Some authors have not confirmed any positive effect of vitamin D on lipid status [56].

Based on the above data, we conclude that correction of vitamin D deficiency in DM patients with DN has a significant renoprotective effect, which is reflected in the improvement of glyco-regulation, lowering of proteinuria, improvement of lipid status, anti-inflammatory effect, anti-fibrotic effect, inhibition of RAAS.

Since albuminuria is not always an indicator of initial changes within DN, identification of new renal biomarkers is crucial to establish early diagnosis and start early treatment, aiming to slow the progression of DN. Several urinary biomarkers have been distinguished from previous studies. Biomarkers of glomerular injury are nephrin, VEGF-A, collagen type IV, transferrin, podocalyxin, immunoglobulins,

ceruloplasmin, laminin, fibronectin, and glycosaminoglycans. Biomarkers of the tubular injury are neutrophil gelatinase-associated lipocalin (NGAL), nephrin, VEGF-A, alpha 1-microglobulin, kidney injury molecule-1 (KIM-1), MCP-1, N-acetyl-β-D glucosaminidase, cystatin C, liver-type fatty acid binding protein (L-FABP), and others. Data in the literature related to these biomarkers still refer to studies conducted on a small number of patients, so it is necessary to plan studies on a larger number of patients. The following text will describe the additional renoprotective effect of vitamin D based on the effect on some of these biomarkers.

## 3.5 Effect of vitamin D on renal fibrosis in diabetic nephropathy

The renoprotective effect of vitamin D is also reflected in the anti-fibrotic effect because it has been proven that in DM patients, the use of vitamin D leads to a lowering of the level of TGF-β1 in the urine, and TGF-β1 is considered the main culprit for the development of renal fibrosis. Also, the administration of vitamin D lowers the increased synthesis of type 1 collagen and fibronectin, which are components of the extracellular matrix [38, 52].

TGF-β1 is a multifunctional cytokine considered to play one of the key roles in DN pathophysiology. In conditions of hyperglycaemia, TGF-β1 formation is increased in almost all renal cells, its expression is increased in both glomerulus and tubulointerstitium. Increased mRNA expression for TGF-β1 has been demonstrated in all renal tissue structures. In DM patients, in addition to hyperglycaemia, increased TGF-β1 synthesis is influenced by: AGEs, oxidative stress, intraglomerular hypertension, PKC activation, de novo diacylglycerol synthesis, and elevated levels of vasoactive substances, such as angiotensin II and endothelin. TGF-β1 stimulates the synthesis of the extracellular matrix. Receptors for TGF-β1 can be observed on all glomerular structures. In response to the action of TGF-β1, mesangium and epithelial cells increase the synthesis of proteoglycans, fibronectin, collagen, and laminin. TGF-β1, on the other hand, inhibits the synthesis of collagenases and stimulates the tissue production of metalloproteinase inhibitors, thereby reducing the activity of matrix metalloproteinases that are responsible for the degradation of the extracellular matrix. Matrix metalloproteinases consist of interstitial collagenase, stromelysin, and type IV collagenase, the most important being matrix metalloproteinase-2 (MMP-2) – produced by mesangium cells. In human renal bioptic material in patients with DN, downstream gene regulation for MMP-2 has been demonstrated. It has also been confirmed that the glycosylated matrix components are resistant to degradation. Angiotensin II directly stimulates renal production of TGF-β1 in the mesangium and epithelial tubular cells, also leads to the upstream regulation of TGF-β1 receptor expression, and increases their production and sensitivity of mesangium cells to this growth factor [38, 52, 57].

In addition to the increased synthesis of the extracellular matrix and its reduced degradation, TGF-β1 induces the transformation of tubular epithelial cells into fibroblasts, and this process is responsible for renal fibrosis as a result of persistent inflammation. Inhibition of TGF-β1 at the level of the proximal tubules is considered to have a greater effect on the reduction of albuminuria and renal fibrosis, while inhibition at the level of the glomeruli has a greater effect on the preservation of GFR [57, 58].

Inflammation-related effects of TGF-β1 are done mainly through SMADs. SMADs are a family of structurally similar intracellular proteins, some of which are signalling molecules, and some transcriptional factors that carry the extracellular signal after binding TGF-β1 to its receptor in the nucleus, and subsequently inhibit or stimulate the expression of certain genes [38].

Based on the above, we can conclude that urinary TGF-β1 can be considered as a biomarker of renal impairment in DN. The level of TGF-β1 according to some authors correlates with albuminuria and GFR decrease. Min Joeng Kim et al. and Yanian Tian et al. have demonstrated that the administration of vitamin D preparations can reduce the urine level of TGF-β1 in DM type 2 patients with consequent albuminuria reduction [59, 60]. The use of 1,25(OH)2D3 leads to downstream regulation of the TGF-β1 signalling pathway, lowering the expression of SMAD3 in the renal tissue of DM patients [41]. Also, the use of vitamin D in DM patients lowers hyperglycaemia-induced increased synthesis of fibronectin, type 1 collagen, the main components of the proliferated extracellular matrix. It is assumed that only TGF-β1 suppression at the renal level is significant for slowing the progression of DN. TGF-β1 suppression at the systemic level affects the metabolism of water and sodium leading to primary hyperaldosteronism [57].

### 3.6 Vitamin D and podocyte

Podocyte dysfunction is known to play one of the key roles in DN pathophysiology. It is assumed that in the future, the stabilization of podocyte function will play a key role in slowing the progression of DN.

Nephrin, podocin, and podocalyxin are structural proteins that determine the structure of SD and are responsible for its selective permeability. They can be considered as biomarkers of podocyte damage, nephrin also being a marker of proximal tubule damage. Within DN, the expression of nephrin, podocin, and podocalyxin at the podocyte level is reduced, but their urinary secretion is increased and is considered to correlate with proteinuria [38, 61].

Nephrin is a structural and signalling protein, member of the immunoglobulin family, responsible for the control of cytoskeletal architecture, the shape, and viability of podocytes. For adequate glomerular function, a certain level of nephrin expression is necessary. In patients with DM, mutations occur in the nephrin gene NPHS1 and the podocin gene NPHS2 leading to their reduced expression at the podocyte level. As a consequence, permeability dysfunction of the glomerular membrane and albuminuria occur. One of the main mechanisms in the development of proteinuria in diabetic rats is increased nephrin endocytosis, mediated by a complex consisting of PKCα, protein interacting with C kinase 1 (PICK1), and beta-arrestin 2 [21, 38, 62].

Nephrin is also a signal protein located on the surface of β cells of the pancreas. Nephrin phosphorylation mediates the release of insulin from pancreatic β cells. At the experimental level, Uchida K. et al. demonstrated that nephrin dephosphorylation occurring in the kidney in DM plays a significant role in the development of albuminuria [63].

Zhang et al. argue that the use of vitamin D preparations in DM and DN patients increases nephrin expression in podocyte culture. It has been confirmed that the administration of vitamin D also enhances podocin and podocalyxin expression on podocytes, thus stabilizing their function [38, 41].

VEGF-A belongs to the group of pro-angiogenic glycoproteins. It is essential for the survival, proliferation and differentiation of endothelial cells, podocytes, and mesangial cells, and is also considered responsible for normal glomerulogenesis. It is produced at the renal level by podocytes and epithelial cells of the proximal tubule. VEGF-A is a part of the SD and controls its permeability. In hyperglycaemia, its synthesis and release are increased. The histopathological finding of early DN stage renal biopsy shows increased expression of mRNA VEGF-A. Increased VEGF

synthesis lowers the level of nephrin expression in renal tissue via the VEGFR2 receptor, inhibiting phosphorylation of nephrin and accelerating its endocytosis, causing SD permeability dysfunction and albuminuria. VEGF-A stimulates TGF-β1 activity under hyperglycaemic conditions and contributes to mesangial proliferation, fibrosis, and later glomerulosclerosis. VEGF-A also lowers nitrogen monoxide levels with consequent endothelial dysfunction. High VEGF concentrations are responsible for pathological angiogenesis in both the kidney and other tissues [38, 52, 62, 64].

The isoform of PKCs activated by AGEs in hyperglycaemia leads to increased expression in the podocytes of VEGF164 and VEGF165 isoforms that in animal models lead to structural and functional renal changes seen in DN, including proteinuria, glomerular hypertrophy, thickening of the glomerular basement membrane, mesangium proliferation, and loss of SD function and foot extensions [65].

In the period of initial and moderate renal tissue damage, the level of VEGF-A is increased, however, over time, as podocytes and proximal tubular cells damage progresses, as fibrosis is more pronounced, the level of VEGF-A decreases because the cells that secrete it are definitely damaged. It is assumed that the stabilization of podocyte function and control of the effect of VEGF-A will play a key role in slowing the progression of DN. Several authors have already presented the positive effect of vitamin D on VEGF-A in patients with DN, in terms of lowering its value after the administration of vitamin D [38, 64].

Some authors have not confirmed the positive effect of vitamin D in DM. A meta-analysis, including 35 studies, showed no positive effect of vitamin D on glucose homeostasis and DM prevention [66]. Different results on the effect of vitamin D in DM type 2 patients are assumed to be due to incoherent data, a small number of patients and a short follow-up period.

## 4. Administration of vitamin D in patients with diabetes mellitus and diabetic nephropathy

Based on the results of numerous researches and multicentric studies, several scientific associations such as the European Society for Clinical and Economic Aspects of Osteoporosis and Osteoarthritis, European Menopause and Andropause Society, Kidney Disease: Improving Global Outcomes Clinical Practice, or American Geriatrics Society, and American Academy of Developmental Medicine and Dentistry published a recommendation for the use of vitamin D. The individual approach to the patient was considered, that is, life in different climatic conditions in terms of exposure to the sun, followed by diet, physical activity, age, obesity and associated diseases [35, 67].

Serum concentration of calcidiol—25(OH)D is the most reliable indicator of vitamin D status in the humans. The recommended optimal blood level of 25(OH)D should be between 30–50 ng/mL or 75–125 nmol/L or even 40–60 ng/mL (100–150 nmol/L). It is necessary for vitamin D to reach a serum level of at least 32 ng/mL (80 nmol/L) in DM patients to express a positive effect [35, 42, 67–69].

Screening of 25(OH)D level should be performed in all DM patients, especially in patients who use chronic therapy that affects vitamin D metabolism and with associated diseases, such as osteoporosis, osteomalacia, hyperparathyroidism, cardiovascular diseases, malabsorption syndrome, hepatic insufficiency, recurrent respiratory tract infections, cancer, neurological diseases, autoimmune diseases, over 65 years of age, obese (Body mass index -(BMI)>30 kg/m2), people with non-traumatic fractures, musculoskeletal pain, and persons with black skin colour [35, 67–69].

In order to maintain optimal vitamin D level, a healthy lifestyle is recommended for all persons as a diet with sufficient vitamin D and calcium intake, safe controlled sun exposure, physical activity, weight control, and smoking cessation. When using vitamin D preparations, adequate calcium intake through diet is also necessary. If the diet cannot provide adequate calcium intake or there is a malabsorption problem, calcium supplements (1000 mg calcium /day) should be used [35, 67, 69].

A detailed overview of vitamin D administration is given in **Tables 1** and **2** [35, 67, 69].

For the treatment of vitamin D deficiency in adults, oral cholecalciferol (vitamin D3) is recommended. Calcifediol can be used instead of vitamin D in certain conditions, such as obesity, malabsorption, liver disease, chronic kidney disease (stage 3 or 4), and in all conditions where rapid correction of vitamin D deficiency is required. The use of calcifediol may also be helpful in patients taking drugs that interfere with the hepatic cytochrome P-450 enzyme system, including those taking glucocorticoids, anticonvulsants, anticancer drugs, or antiretroviral drugs. In certain risk groups (e.g., patients with severe malabsorption), vitamin D may also be administered parenterally [67, 70, 71].

Calcitriol (1,25(OH)2D) and its analogues are used at much lower doses compared to vitamin D3, have a relatively high risk of hypercalcaemia, and have a relatively

| Sufficient 25(OH)D 30–50 ng/mL or 75–125 nmol/L |
| --- |
| • Moderate sunlight exposure is recommended, with adequate dietary intake of vitamin D. The use of vitamin D preparations is recommended depending on the season, BMI, age, skin colour, and risk factors. |
| • During the winter months from November to April, the use of 800–2000 IU/day of vitamin D is recommended in people without risk factors |
| • Administration throughout the year of 800–2000 IU/day of vitamin D is recommended in:<br> • women planning pregnancy, during prolonged hospitalizations, in patients over 65 years of age, in patients with osteoporosis or the risk of falling and fractures, as maintenance dose after treatment of vitamin D deficiency |
| • 2–3 times higher doses of vitamin D are recommended in people with malabsorption syndrome, obese (BMI >30 kg/m2), and persons with black skin colour. |

**Table 1.**
*Preventive use of vitamin D.*

| Insufficient $\geq 20 - < 30\ ng/ml$ ($\geq 50$- $< 75$ nmol/L) or deficient $< 20$ ng/mL($< 50$ nmol/L) |
| --- |
| • If clinically indicated rapid 25(OH)D correction in patients with vitamin D deficiency without significant risk factors, it is recommended to use 6000 IU D3/day. In certain persons, high doses of vitamin D-10,000 IU D3/day are applied: patients with malabsorption syndrome, patients using chronic therapy with drugs that disrupt vitamin D metabolism and obese persons (BMI > 30 kg/m$^2$) |
| • The treatment lasts 4–12 weeks depending on the degree of vitamin D deficiency. |
| • If the recommended vitamin D level is reached, 30–50 ng/mL or 75–125 nmol/L, the administration of 800–2000 UI/day of the vitamin D is continued. |
| • Approximately 6–12 weeks after starting treatment, the level of 25(OH)D is checked. |

**Table 2.**
*Therapeutic doses of vitamin D.*

narrow therapeutic window. Active treatment with calcitriol is indicated only in certain diseases, for example, chronic hypoparathyroidism and chronic kidney disease–mineral and bone disorder (CKD-MBD) [67, 72, 73].

## 4.1 Side effects of vitamin D

Vitamin D intoxication occurs at a level of 25(OH)D > 375 nmol/l (150 ng/ml) and is a very rare complication. The first manifestation of vitamin D intoxication is calciuria, caused by reducing the reabsorption of calcium in the proximal tubules. When the compensatory mechanisms can no longer increase calciuria, its concentration in the blood increases. Hypercalcaemia decreases the parathyroid hormone (PTH) level, which consequently reduces renal phosphorus excretion. A high concentration of 25 (OH)D directly through VDR in the intestine further increases intestinal absorption of calcium and phosphorus, which further increases their blood level, and as a result calcium/phosphorus solubility product increases, leading to calcifications in soft tissues and renal tissue (nephrocalcinosis), and also in the blood vessels. Hypercalcaemia also causes constipation, vasoconstriction with subsequent arterial hypertension, cardiac arrhythmias, depression, confusion, polyuria, and polydipsia [35, 74].

Therefore, in patients with advanced DN and significantly reduced GFR, care should be taken when supplementing vitamin D, that is, monitor the level of calcium and phosphorus in the blood. If the phosphorus level is elevated, stop the administration of vitamin D, until the phosphorus in the blood normalizes and include one of the phosphorus-binding preparations in the therapy. Also in case of hypercalcemia, discontinue the administration of vitamin D.

## 5. Conclusion

We can conclude that the use of vitamin D in patients with DM and DN has a significant renoprotective effect, both in patients with albuminuria and in patients without albuminuria. The particularly significant renoprotective effect of vitamin D is reflected in the stabilization of podocyte function. Vitamin D has also cardiovascular protection by decreasing albuminuria, correcting lipid status, and improving parameters of glycoregulation and inflammation.

In order to obtain more data on the renoprotective effect of vitamin D, additional studies are needed, including more patients and longer follow-up.

## Conflict of interest

The authors declare no conflict of interest.

## Acronyms and abbreviations

| | |
|---|---|
| 1,25(OH)2D3 | 1,25- hydroxyvitamin D3 |
| 25(OH)D3 | 25-hydroxyvitamin D3 |
| ACEi | Angiotensin-converting enzyme inhibitors |
| ADA | American Diabetes Association |
| AGEs | Advanced glycation end products |

| AT1R | Angiotensin II type 1 receptor |
| BMI | Body mass index |
| CIP27B1 | 1-alpha-hydroxylase |
| CKD-MBD | Chronic kidney disease–mineral and bone disorder |
| CRE | Cyclic adenosine monophosphate-response element |
| DM | Diabetes mellitus |
| DN | Diabetic nephropathy |
| DR | Diabetic retinopathy |
| ESRD | End-stage renal disease |
| GFR | Glomerular filtration rate |
| HbA1c | Glycated haemoglobin A1c |
| ICAM-1 | Intracellular adhesion molecule 1 |
| IDF | International Diabetes Federation |
| IGF-1 | Insulin-like growth factor 1 |
| IGT | Impaired Glucose Tolerance |
| INF-γ | Interferon-gamma |
| IL | Interleukin |
| KIM-1 | Kidney injury molecule-1 |
| L-FABP | Liver-type fatty acid binding protein |
| LDL | Low-density lipoprotein |
| LMICs | Low- and middle-income countries |
| M | Macrophages |
| MCP-1 | Monocyte chemoattractant protein-1 |
| MMP-2 | Matrix metalloproteinase-2 |
| Na+/K+-ATP-ase | Sodium potassium adenosine triphosphatase |
| NAD+ | Nicotinamide dinucleotide |
| NADPH | Nicotinamide dinucleotide phosphate |
| NGAL | Neutrophil gelatinase-associated lipocalin |
| NKF KDOQI | National Kidney Foundation Kidney Disease Outcomes Quality Initiative |
| NP-DN | Non-proteinuric diabetic nephropathy |
| P-DN | Proteinuric diabetic nephropathy |
| PDGF | Platelet-derived growth factor |
| PICK1 | Protein interacting with C kinase 1 |
| PKC | Protein kinase C |
| PPAR δ | Peroxisome proliferator activator receptor δ |
| PTH | Parathyroid hormone |
| RAAS | Renin-angiotensin-aldosterone system |
| RANTES | Regulated upon activation, Normal T cell expressed and presumably Secreted |
| ROS | Reactive oxygen species |
| SD | Slit diaphragm |
| SGLT1 | Sodium-glucose cotransporter-1 |
| SGLT2 | Sodium-glucose cotransporter-2 |
| T1DM | Type 1 diabetes mellitus |
| T2DM | Type 2 diabetes mellitus |
| TGF | Tubuloglomerular feedback |
| TGF-β1 | Transforming growth factor Beta 1 |
| Th | T lymphocytes |
| TNF-α | Tumor necrosis factor-alpha |

| UACR | Urine albumin-to-creatinine ratio |
|------|-----------------------------------|
| VCAM-1 | Vascular cell adhesion molecule-1 |
| VDBP | Vitamin D binding protein |
| VEGF | Vascular endothelial growth factor |

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