

REVIEW

BY PROF. DR. BORYANA DELIJSKA, MD, DSc

**REGARDING DISSERTATION FOR THE EDUCATIONAL AND
SCIENTIFIC DEGREE “DOCTOR”**

**ON THE TOPIC: “ROLE OF SOME PATHOGENETIC AND GENETIC
FACTORS FOR THE DEVELOPMENT OF DIABETIC
NEPHROPATHY IN PATIENTS WITH TYPE 2 DIABETES
MELLITUS”**

OF DR. STANISLAVA LYUBOMIROVA ILIEVA, MD

Doctoral program “Internal Medicine”

ACIBADEM CITY CLINIC UMHAT “TOKUDA” EAD

CLINIC OF INTERNAL DISEASES

SCIENTIFIC SUPERVISOR: ASSOC. PROF. DR. LACHEZAR BOYANOV
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According to the order №15-05-147/20.11.2024 of the Executive Director of “Acibadem City Clinic UMHAT Tokuda”EAD, as well as on the basis of Art. 4 of the ZRASRB and the Regulations of the development of the scientific community of Acibadem City Clinic UMHAT Tokuda”EAD and the resolution of the Scientific Council (Protocol 54/6.11.24), I have been appointed as a member of the scientific jury for adjudging of Educational Scientific Degree “Doctor” to Dr. Stanislava Ilieva, MD, in the area of higher education 7. Healthcare and sports field 7.1. Medicine, doctoral program “Internal Diseases”.

Brief biographical data:

Dr. Ilieva graduated from the Medical University Sofia in 1997. From the same year she started working as a physician at First Nephrology Clinic, SBALENG “Acad. Ivan Penchev”, after that – at Nephrology Clinic, UMHAT

“Queen Joanna”- ISUL. She has two specialties – Internal Medicine (2004) and Nephrology (2013). Dr. Ilieva has participated in 8 educational courses, 7 of which abroad and one – in Bulgaria, as a part of continuous medical education. She has been certified in biopsy methods in nephrology and conventional abdominal ultrasound. She is fluent in English, certified by Cambridge First Certificate in English (2000г.); Cambridge Certificate in Advanced English (2001г.). Also she speaks Russian.

Analysis of the dissertation work:

The dissertation work, represented to myself for a review, covers a current topic, since Diabetes Mellitus type 2 is a disease of social importance and Diabetic nephropathy leads to end stage renal disease and to a permanent disability of the patients. Still there are a lot of obscurity regarding pathogenetic and genetic mechanisms of this nephropathy. Elaborating this dissertational work will contribute for clarifying some of them and possibly will give an opportunity for an individual approach to each diabetic patient.

The literature review presents a detailed discussion of historical and epidemiologic data of Diabetes mellitus, its complications, one of which is diabetic nephropathy. In the part of the review, concerning pathogenesis of the disease, there is an analysis of the importance of hemodynamic, metabolic, inflammatory mechanisms, growth and genetic factors, the role of homocysteine, folic acid and methylenetetrahydrofolate reductase gene. Literature data have been overviewed regarding the development and progression of diabetic nephropathy, its clinical and morphological characteristics, as well as differentiation between diabetic and non-diabetic renal injury. Histological findings in diabetic nephropathy have been discussed. The literature review is based on 148 literary sources, 14 of which are in Bulgarian.

Based on a thorough analysis of the data from the literature sources, Dr. Ilieva gives a formulation of the aim of the study: to establish a relationship between specific clinical and laboratory biomarkers and the manifestation of

diabetic nephropathy in patients with type 2 Diabetes mellitus in order to identify predictors for its development and optimize the diagnostics of diabetic kidney injury.

To achieve this aim, the dissertation candidate has formulated the following tasks, which include: analysis of histological changes from puncture kidney biopsies, considering their incidence in diabetic patients; analysis of certain clinical and laboratory markers in biopsied patients and comparing the obtained results with a control group of patients with histologically proven nephropathies, but without diabetes; to make an attempt to determine a relationship between diabetic nephropathy and the remaining micro- and macroangiopathic complications of diabetes and to make a possible correlation between particular biomarkers (clinical, laboratory, genetic) in diabetic patients with and without diabetic nephropathy; to try to establish specific predictors of diabetic renal complications and to propose a diagnostic approach and recommendations for the clinical practice in order to optimize the diagnosis of diabetic kidney complications.

The study was conducted on 91 patients hospitalized at the Department of Nephrology, Clinic of Internal Medicine of "ACC UMHAT Tokuda" EAD, for the period 01.2018 - 02.2024. Patients are selected according strict exclusion criteria. All patients had chronic kidney diseases and histologically proven nephropathies. Fifty five of them had Diabetes mellitus type 2, found before or during the diagnosis of the kidney disease and the rest 36 were without diabetes. Patients are divided into three groups depending on the presence of Diabetes mellitus and diabetic nephropathy: patients with type 2 DM and histologically proven diabetic nephropathy, patients with type 2 DM and non-diabetic kidney injury in the biopsy and a control group of patients with histologically proven nephropathies, but without DM.

Except the routine clinical and laboratory tests, there has been investigated the polymorphism of methylenetetrahydrofolate reductase (MTHFR) gene,

specifically the variants A1289C and C677T, thyroid function, markers of inflammation and vascular injury. Modern statistical methods have been used, which assure the reliability of the obtained results.

In different chapters of the dissertation work, there is a discussion of the results, related to the frequency of morphological changes from puncture kidney biopsies, demographic findings and clinical data, indices of metabolic control, including glycated hemoglobin, serum LDL, HDL, triglycerides and uric acid in the three groups of patients. Protein and albumin urinary excretion have been measured as well. Other investigated parameters include CRP and IL-6 as inflammatory markers and D-dimer and fibrinogen as indices for coagulation disorders and vascular damage. The following chapters of the dissertation work focus on studying polymorphism of MTHFR gene, serum levels of homocysteine and folic acid, thyroid gland function.

Each part of the results has been analyzed in details and very well illustrated with 33 tables and 15 figures.

The dissertation ends with the discussion of the results, obtained by the PhD candidate and their comparison with the data from previous studies in the literature. This allows her to make 9 basic conclusions: in the majority of diabetic patients there is a prevalence of tubulointerstitial and vascular and hypertensive renal damage; male sex could be considered as a risk factor for development of diabetic nephropathy; the appearance of diabetic nephropathy is not connected with the age of the patients, even though age over 60 years and duration of Diabetes mellitus, longer than 10 years are associated with a higher incidence of CKD of any nature; hypertension, obesity, glycemic control, dyslipidemia and hyperuricemia in diabetic patients are not pathogenetic factors for the development of diabetic nephropathy, but they are connected with an accelerated progression of CKD; the onset of diabetic nephropathy is not associated with an elevated inflammatory (CRP, IL-6) and coagulation (D-dimer, fibrinogen) markers, but their high levels might be considered as possible predictors of CKD.

A matter of interest is the conclusion that carriage of mutant alleles of MTHFR gene, homocysteine and folic acid levels have no predictive value for the manifestation of diabetic nephropathy, as well as stating a correlation of the changes in thyroid function in diabetic patients.

I would define the contributions of the dissertation work as ones of original nature, because for the first time in Bulgaria there has been conducted a study and have been analyzed a multiple pathogenetic and genetic factors, leading to the onset of diabetic nephropathy; for the first time in Bulgaria there has been investigated the connection between diabetic nephropathy and the polymorphism of methylenetetrahydrofolate reductase (MTHFR) gene, variant A1289C, as well as for the first time in the country there has been studied the relationship between diabetic nephropathy and an impaired thyroid function. The rest of conclusions that have an essential practical importance include determination of the role of puncture kidney biopsy in diabetic patients, which allows to make a differentiation between diabetic and non-diabetic renal injuries and reports the dominance of tubulointerstitial and vascular-hypertensive lesions. Also the male sex is determined as a risk factor for diabetic nephropathy.

The dissertation ends with some practical recommendations regarding clinical approach to patients with DM type 2.

The Abstract is structured clearly and correctly, with an adequate scientific terminology and it is fully represents the essence of the dissertation work.

Dr. Ilieva has presented a total of six publications and participations in scientific events, in relation with the topic of the dissertation.

Based on everything stated above, the dissertation work of Dr. Stanislava Lyubomirova Ilieva meets all the requirements of the relevant laws in the Republic of Bulgaria and the Regulations for their implementations for the acquisition of the educational and scientific degree "Doctor" (PhD) in the area higher education 7. Healthcare and sports field 7.1. Medicine, doctoral program

“Internal Diseases”. I would like to recommend to the other reviewers and the members of the scientific jury to vote positively.

Prof. Dr. Boryana Delijska, MD, DSc

A handwritten signature in black ink, appearing to be 'BD' or similar initials, written in a cursive style.