

TO  
THE CHAIRMAN OF  
THE SCIENTIFIC JURY  
*ORDER NO P-19/08-01.2021*

### STANDPOINT

From **Assoc. Prof. Dr. Stanislav Minchev Filipov, MD, PhD**

Department of Anatomy, Histology, General and Clinical Pathology and Forensic  
Medicine "

Faculty of Medicine, Sofia University "St. Kliment Ohridski"

On the topic:

### "IMMUNOMORPHOLOGICAL ASPECTS OF UROTELIAL CARCINOMAS OF THE BLADDER"

for acquiring the educational and scientific degree 'DOCTOR' in Direction 7. Healthcare  
and sports, Professional Direction 7.1. Medicine and

Scientific specialty "Pathology and Cytopathology" with  
code 03.01.03

Supervisor - Assoc. Prof. Dr. Dorian Ivanchev Dikov, MD, PhD

**Milena Gulinac, MD**

Doctoral student from

Department of General and Clinical Pathology,

Medical Faculty, Medical University of Plovdiv

DEAR MR. CHAIRMAN OF THE SCIENTIFIC JURY,

I present to your attention an opinion on the dissertation study for acquiring the  
educational and scientific degree "Doctor" from a doctoral student

Milena Gulinac, MD according to the requirements of the Regulations on the terms  
and conditions for obtaining scientific degrees and holding academic positions at MU -  
Plovdiv in connection with an order from the Rector of MU - Plovdiv.

## I. Education and qualifications

2002 -2005 - High school with medical profile "Goce Delchev "- Kumanovo, Republic of North Macedonia

2005 – 2011 - MU Plovdiv, specialty "Medicine"

2016y - Acquisition of a specialty

## Career development

2011 - 2015 - doctor in the Department of Clinical Pathology of University Hospital "KASPELA" - Plovdiv,

2013 - 2020 - pathologist in the Department of Clinical Pathology of University Hospital "St. George"- Plovdiv,

2015 - Assistant professor in the Department of General and Clinical Pathology, MU-Plovdiv

2016 - pathologist in the Department of Clinical Pathology of Hospital "ASENOVGRAD", Asenovgrad,

2016 - Pathologist at Medicus Alpha Clinic - Plovdiv, Department of General and Clinical Pathology

Her interests are focused in the field of pathology of thyroid gland and pathology of the urinary system.

She is a member of the Bulgarian and European Societies of Pathology.

## II. General evaluation of the dissertation study:

The presented dissertation is dedicated to the **insufficiently researched scientific and medical problem** - bladder carcinomas, which still occupies one of the leading places as a cause of death due to a small disease and is a test for medical teams. The treatment of bladder cancers is a multidisciplinary problem. Decisions about therapeutic behavior must be made jointly by a large number of medical professionals: urologists, medical oncologists, interventional radiologists, and last but not least, pathologists and laboratory specialists.

Based on the latest GLOBOCAN data, bladder cancer accounts for 3% of diagnoses worldwide and is particularly prevalent in developed countries. In the United States, bladder cancer is the seventh most common neoplasm. A total of 90% of bladder cancers are diagnosed over 55 years of age and older, and the disease is four times more common in men than in women. While the median 5-year survival rate in the United States is 77%, the 5-year survival rate for patients with metastatic disease is a promising 5%.

The most severe risk factor for bladder cancer is smoking, which accounts for 50-65% of all cases. Occupational and environmental toxins also contribute significantly to the severity of the disease (representing approximately 20% of all cases). Although the exact proportion may be obscured by the fact that bladder cancer develops decades after toxic exposure, even if exposure has lasted only a few years. Schistosomiasis infection is a common cause of bladder cancer in regions of Africa and The Middle East is considered the second largest tropical pathogen after malaria. With 81% of cases due to known risk factors (and only 7% due to hereditary mutations), bladder cancer is the main candidate for preventive strategies. Smoking cessation, workplace safety practices, weight loss, exercise and schistosomiasis prevention (through water disinfection and mass drug use) have been shown to significantly reduce the risk of bladder cancer, which is a growing burden Worldwide.

As many as 7 out of 10 cases of bladder cancer are detected in the early stages, thus allowing for resection and improved survival. Non-muscular invasive bladder cancer (NMIBC) is usually removed by transurethral resection.

Alternatively, a cystoscopy plus biopsy procedure for certain resections may be used. For high-risk cases, intravesical cytostatic therapy may be added.

For 30% of patients with muscle-invasive bladder cancer (MIBC), neoadjuvant or adjuvant chemotherapy is considered the standard for reducing the risk of recurrence, and radical cystectomy is the primary surgical treatment. It is also possible to use external beam radiation.

For the treatment of metastatic disease (which currently has only a 5% 5-year survival), platinum chemotherapy remains the standard, although newer immunotherapies, namely checkpoint inhibitors, are increasing as first-line and post-treatment options.

Tumor immunotherapy is emerging as a promising component for the treatment of neoplastic diseases.

The most promising is immunotherapy which shows good results includes antibodies targeted to inhibitory immune control points.

In this regard, the development of optimized structural algorithms for evaluation and selection of patients with primary urothelial carcinomas subject to surgical and combined treatment is an acoustic medical problem.

The dissertation presented by PhD student Milena Gulinac,MD corresponds to the **basic requirements of Art. 27 of the Regulations for application of the law for the development of the academic staff in the Republic of Bulgaria (Promulgated SG No. 75 / 24.09.2010 „amended SG No. 19 / 8.03.2011):**

The dissertation study is presented in the form and volume of a total of 153 pages, containing: content (a total of 2 pages); exposition structured in the following sections.

literature review (28 pages); II. purpose and tasks (1 countries); III. material and methods (13 pages); IV. results (52 pages); V. discussion (25 pages); main conclusions from the conducted research and contributions (6 pages); bibliographic reference. The dissertation is illustrated with 25 figures and 37 microscopic photographs. The references includes 157 printed sources - 8 in Cyrillic and 149 in Latin. The text also includes 10 tables, which are numbered.

List of publications in connection with the dissertation is attached that includes two publications in English in foreign scientific journals with an impact factor literature review in a collection and two posters.

There is 64-page summary attached that reflects the main points of the dissertation study divided into eight chapters.

The dissertation study shows that the doctoral student has theoretical knowledge in his specialty and ability to conduct research. The presented results are indication of an independent scientific contribution.

The dissertation study contains a "**Literary Review**", which shows the necessary information of the doctoral student on the problems of urothelial carcinomas. In a volume of 28 pages, the doctoral student presents an overview of the cellular and functional characteristics of urothelial carcinomas and main nosological categories. The analysis of the data on surgical, medical methods of treatment and associated categories allow the doctoral student to formulate unresolved and controversial problems, which are included in the present dissertation.

The doctoral student demonstrates skills for analysis and summarization of the cited scientific facts, based on which he formulates an ambitious aim of the dissertation study - **to study epidemiological, morphological and immunochemical / IHC / features of urothelial carcinoma / UC /, with attention to the expression of the program death-1 (PD-1) and ligand - programmed death ligand 1 (PD-L1) and the immunomorphology of giant cells in the tumor stroma, as well as to determine the diagnostic and prognostic value of these indicators compared with the histological differentiation and tumor stage.**

1.1 The doctoral student has presented the "**Materials and Methods**" used in the research, which have been selected in accordance with the aim and main tasks.

A set of laboratory methods is proposed, described in detail and explained in the section in a way that allows a clear understanding of the nature of the research performed.

Used:

- set of histopathological methods (light microscopic, histochemical, immunohistochemical) and statistical methods for analysis.

1.2. The dissertation study presents "**Results**", described correctly, illustrated with figures, photos and tables, as follows:

**Study of the main epidemiological features** of bladder UC in populations of Bulgarian and French patients by frequency, age, gender and comparison of the results in these groups to determine elements of geographical pathology.

**Study of morphological parameters** of bladder UC: histological variant and tumor stages.

**Study of IHC expression of programmed death receptor-1 (PD-1) and its ligand, programmed cell death ligand 1 (PD-L1) in the bladder UC** parenchyma and correlation of the obtained results for gender and age of patients with stage, histological appearance and degree of tumor cell differentiation.

**Histo-epidemiological, histological and IHC examination of giant cells** in the stroma of the bladder, pathological and prognostic significance compared with tumor stage, histological type and degree of differentiation of tumor cells.

**Development of a differential algorithm for diagnostics of multinucleated giant cells** in the stroma of the bladder UC.

1.3 In the presented "**Discussion**" the doctoral student analyzes the data from her own research and those of other research groups she has found, which allows her to formulate summaries and "**Conclusions and Contributions**" regarding the possibilities for assessment of clinico-pathological parameters.

Therapies with ICI (immune checkpoint inhibitors) have been shown to cause persistent reactions and long-term remission in several cancers, with many patients failing to respond, developing resistance over time, or showing immune-dependent adverse effects.

The lack of response or toxicity of ICI is a strong justification for the combination of ICI with other treatments to increase the response rate of non-immunological tumors. Therapeutic approaches that induce the release and presentation of tumor antigens could stimulate a new (de novo) antitumor T-cell response.

The proposed study may support the clinical decision-making process for therapeutic behavior and monitoring.

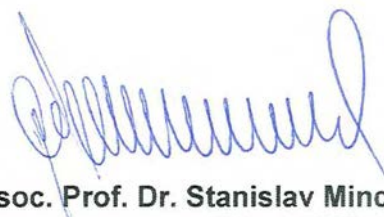
### III. Conclusion

In conclusion, the presented dissertation of Milena Gulinac, MD, PhD student in self-study at the Department of General and Clinical Pathology, Medical Faculty, MU - Plovdiv, on the topic "IMMUNOMORPHOLOGICAL ASPECTS OF UROTHELIAL CARCINOMAS OF THE URINARY BLADDER" with a current scientific and medical problem, structured, shaped and illustrated according to the requirements.

The dissertation study, with the scientific publications attached to it, meets the criteria of Article 6 of the Law on the Development of Academic Staff in the Republic of Bulgaria (SG No. 38 of 21 May 2010, last amended SG No. 68 of 2). August 201Z.) And the regulations for the conditions and the order for acquiring scientific degrees and holding academic positions in MU – Plovdiv.

Based on the above, I give a **positive assessment** of the dissertation study and I suggest to the esteemed Scientific Jury to award the educational and scientific degree "DOCTOR" in Direction 7. Health and Sports. Professional Direction 7.1. Medicine and Scientific specialty "Pathology and Cytopathology" with code 03.01.03. of doctoral student **Milena Gulinac, MD**

2021, January 15



**Assoc. Prof. Dr. Stanislav Minchev  
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