**SCIENTIFIC OPINION**

by professor Bistra Tsaneva Kaltcheva, PhD, professor of Biochemistry
at the Medical University "Prof. Dr. Paraskev Stoyanov" of Varna

**Re:** a competition for acquiring the academic position "**associate professor**", scientific specialty „**Medical genetics**", field of higher education 4. **Natural sciences, mathematics and informatics**, professional field 4.3. **Biological sciences**

**Competition details**

The competition for the academic position "Associate Professor" was announced in State Gazette No 83/03.10.2023 and is for the needs of the Department of medical genetics, Faculty of Medicine, Medical University - Varna.
Based on the decision of the Faculty Council at the Faculty of Medicine (Protocol No 12/13.11.2023) and an order of the Rector of the Medical University of Varna (No. R-109-515 /November 30, 2023), I have been appointed as an internal member of the Scientific Jury. At the first meeting of the Scientific Jury, I was assigned to prepare a scientific opinion (Protocol No. 1/12.12.2023). The only candidate in the competition is assistant-in-chief Maria Kostadinova Levkova, MD, PhD from the Department of Medical genetics, Faculty of Medicine, Medical University of Varna.

**Brief professional details of the applicant**

Assistant-in-chief Maria Kostadinova levkova, Md, PhD e began his professional carreer in 2017 as a specialist in Medical Genetics at the Department of Medical Genetics, Medical University "Prof. Dr. Paraskev Stoyanov"- Varna. In 2016 she was appointed as a Visiting Assistant Professor and in 2017 as an Assistant Professor at the same Department. In 2021 Dr. Levkova acquired the specialty "Medical Genetics" at the Ministry of Health. Since 2018 the candidate is a full-time PhD student at the Department of Medical Genetics. In 2021, she successfully defended her doctoral thesis on "Molecular genetic and immunological biomarkers in medical genetic counseling of families with infertility" and in the same year she was selected by competition as an assistant-in-chief at the Department of Medical Genetics at the Medical University "Prof. Dr. Paraskev Stoyanov" - Varna.

**Evaluation of the candidate's scientific production**

For participation in the competition assistant-in-chief Maria Levkova has submitted all the required documents, according to the requirements of the Law for development of the academic staff in the Republic of Bulgaria and of the Regulations for development of the academic staff at the Medical University of Varna for tenure the academic position "associate progfessor". All the documents together with the evidentiary materials are neatly arranged.
From the attached ACADEMIC REFERENCE on the scientometric indicators for holding the academic position "Associate Professor" and from the evidentiary material, it is established that

assistant-in-chief Maria Levkova covers the required **minimum of 400 points** for acquiring the academic position "Associate Professor" (Table 1).

Table 1. Minimum national requirements for acquiring the academic position "associate professor" in the field of higher education 4. Natural sciences, mathematics and informatics, professional field and the relevant scientometric data of assistant-in-chief Maria Levkova.

| Group of indicators | Content | Minimum national requirements for acquiring the academic position "Associate Professor" | Scientometric data of assistant-in-chief Maria Levkova |
|---|---|---|---|
| A1 | **Indicator 1** - Dissertation thesis for acquiring the educational and scientific degree "PhD" | 50 | 50 |
| B | **Indicator 2** - Dissertation thesis for acquiring the scientific degree "DSci" | - | - |
| C | **Indicator 3** – habilitation work, a monograph | - | - |
| C | **Indicator 4** – scientific publications equivalent to a habilitation thesis published in the editions referenced and indexed in worldwide scientific databases (Web of Science и Scopus)<br>25 for publications in Q1;<br>20 for publications in Q2;<br>15 for publications in Q3 ;<br>12 for publications in Q4;<br>10 for publications in journals with SJR without IF | 100 | 107<br><br>3 publications in Q1 -> 3x25 = 75<br>1 publications in Q2 -> 1x20 = 20<br>1 publications in Q4 -> 1x12 = 12 |
| D | **Indicator 7** - Scientific publications published in journals, referenced and indexed in worldwide scientific databases (Web of Science и Scopus)<br>25 for publications in Q1;<br>20 for publications in Q2;<br>15 for publications in Q3;<br>12 for publications in Q4;<br>10 for publications in journals with SJR without IF | 200 | 216<br><br>3 publications in Q2 -> 3x20 = 60<br>4 publications in Q3 -> 4x15 = 60<br>8 publications in Q4 -> 8x12 = 96 |
| E | **Indicator 11** - Citations in scientific publications, monographs and collective volumes and patents, referenced and indexed in worldwide scientific databases (Web of Science и Scopus) | 50 | 50<br><br>based on 25 citations |
| | **Total scores:** | **400** | **423** |

Assistant-in-chief Maria Levkova participated in the competition with **20 scientific publications**, referenced and indexed in worldwide scientific databases (Web of Science and Scopus), in 8 (40%) of which she is the first author. Of these, **3** fall into quartile 1 (Q1), **4** into quartile 2 (Q2), **4** into quartile 3 (Q3), and **9** into quartile 4 (Q4). Eight (40%) of the presented full-text publications are in journals with an impact factor, with a **total impact factor** of **28,743**.
The candidate has presented a list of 27 participations in International scientific forums, of which she is the first author in 5 forums. There are 15 participations in National scientific forums, of which she is the first author in 4.

**Citations**

A total of 25 citations in scientific editions are presented.

**Research activity and scientific contributions**

1. **Scientific publications equivalent to a habilitation thesis (C4)**

For indicator C4 the candidate has presented scientific works in 3 main scientific areas in the field of medical genetics, which fall within the scientific priorities of Medical University - Varna: **1. Reproductive health - "Genetic aspects in the etiology of male infertility"; 2. Medical Oncology - "Small Circular RNAs in the Etiology of Cancer"; 3. Cardiology - "Genetic aspects in the etiology of ischemic strokes"**.

The research in the field "**Genetic aspects in the etiology of male infertility" (C4.3 and C4.5)** are review articles (systematic review and meta-analysis) aimed at assessing the role of key genes relevant to infertility in men. A critical analysis of the literature has differentiated a total of 15 genes responsible for the production of dynein structural components, as well as those associated with the production of dynein-assembling factors relevant to male infertility. The results of this meta-analysis suggest that a number of genes involved in dynein biosynthesis and assembly are relevant to male infertility due to impaired sperm motility and are potential targets for investigation in men with reproductive problems.

In the analysis of genetic testing registry data (National Center for Biotechnology Information, NCBI) pointing out the current target gene panels used in medical genetics associated with male infertility due to impaired spermatogenesis, the gene encoding the transmembrane conductance regulator in cystic fibrosis was found to be (CFTR) is stated in all panels. Based on the analysis of data from the literature, and due to the lack of established protocols specifying genes to include in the different panels for testing, recommendations are made to clinicians for staged testing that includes target gene sequencing at a later stage of the study.

The research in the fiels "**Small Circular RNAs in Cancer Etiology**" (**C4.1 and C4.2**) are review articles aimed at evaluating the role of circular RNAs and various SNPs as potential prognostic and diagnostic biomarkers in patients with colorectal cancer (CRC).

After a critical analysis of the literature on the role of circular RNAs, the following conclusions were drawn: 1. Depending on their localization, circular RNAs may be involved in the regulation of gene expression as well as in transcription and modulation of alternative splicing; 2.

Overactivated circular RNAs have oncogenic functions such as stimulation of cell proliferation, invasion/migration, metastasis, and suppression of apoptosis; 3. Circular RNAs are involved in the pathogenesis of CRC as three types miRNA-SNPs (miR-146a rs2910164, miR-27a rs895819 and miR-608 rs4919510) are promising prognostic, diagnostic and predictive biomarkers for CRC; 4. circular RNAs have the potential to be used as therapeutic targets in the treatment of patients with CRC.

Regarding the third research field "**Genetic aspects in the etiology of ischemic strokes**", mentioned in the habilitation reference, a scientific paper is presented, which is in its essence a meta-analysis of the literature on the problem and aims to clarify the role of hereditary predisposition to thrombophilia in the etiology of ischemic strokes in young people.

After a critical and thorough analysis of the literature, genetic markers factor V Leiden (FVL), prothrombin (PT), protein C (PC), protein S (PS) and antithrombin (AT) were found to be risk factors for ischemic stroke in young adults. Research in this area would have extremely important health and social implications regarding the prevention and control of risk factors for ischemic stroke, especially in young people.

## 2. Scientific papers, published in journals, refereed and indexed in worldwide scientific databases (Web of Science и Scopus) (D7)

According to **indicator D7**, the candidate has submitted **15 scientific publications** that meet the minimum scientific-metric requirements for the position "Associate Professor", which fall into **4 subfields** of medical genetics.

### 2.1. Reproductive genetics (D7.4, D7.11, D7.12)

#### *2.1.1 Molecular-genetic causes of impaired spermatogenesis*

For the first time, the IVS8-5T variant of the CFTR gene has been studied in Bulgarian men with infertility and has been shown to be a risk factor for impaired spermatogenesis. The importance of microdeletions in the AZFa, AZFb and AZFc regions of the Y chromosome as a cause of reduced sperm concentration in ejaculate in carriers and the need for molecular genetic analysis was confirmed. Data were obtained on the presence and significance of different chromosomal disorders and variants of chromosomal polymorphisms among Bulgarian patients with reproductive disorders, with twice as many polymorphisms observed in males as in the studied females.

#### *2.1.2. Molecular genetic causes of recurrent miscarriages*

For the first time in Bulgarian women with recurrent miscarriages, polymorphic variants in genes involved in the regulation of the immune system and with a potential role in the successful outcome of pregnancy (the 14 bp insertion/deletion variant in the HLA-G gene and the 308 GA polymorphic variant in the TNF-alpha gene) were studied. The variant in the HLA-G gene was found to be associated with a significantly higher risk of miscarriage in carriers.

### 2.2. Paediatric genetics and dysmorphology (D7.2, D7.5, D7.6, D7.7, D7.8, D7.9, D7.10, D7,15)

**Eight scientific papers** are presented in this scientific field.

In a molecular genetic selective screening study for fragile X chromosome syndrome among 52 high-risk children with intellectual disability or developmental delay or autistic behavior, three boys (5.7%) were found to be affected.

The results of cytogenetic analyses of 1781 children with suspected chromosomal pathology were summarized, and 275 of them were found to have a pathological karyotype. The results clearly demonstrate the important role of cytogenetic analysis in the diagnostic process.

The candidate has also contributed to the field of rare diseases. Clinical cases of patients with Kabuki syndrome, Bardet-Biedl syndrome, Cornelia de Lange syndrome are presented and analysed.

In this research area, the candidate has prepared a systematic literature review on the role of the gut microbiome in the etiology of autistic spectrum disorders. Based on data from the literature, the ten bacterial genera that show statistically significant differences between patients with autism and healthy controls were identified.

### 2.3. Neurogenetics (D7.1, D7.14)

In this line of research, the role of polymorphic variants for factor V (F V) Leiden G1691A, factor II (F II) G20210A, plasminogen activator inhibitor (PAI) 4G/5G and methylenetetrahydrofolate reductase (MTHFR) C677T and their association with hereditary predisposition to thrombophilia among two groups of patients, a group with vascular disorders and patients with recurrent miscarriages, was investigated. Patients with vascular disorders were found to have a significantly higher incidence of the F V Leiden variant compared to patients with recurrent miscarriages.

### 2.4. Медико-генетично консултиране (D7.3, D7.13)

Under this scientific direction, information has been selected and developed for GPs on the nature of medical genetic counselling and molecular genetic analyses used in clinical practice.

A questionnaire survey was conducted among 500 women of childbearing age regarding their awareness of screening and diagnostic tests during pregnancy to detect chromosomal diseases in the fetus. Based on the results of the survey, recommendations were made to the antenatal clinics to improve the education of pregnant women during the consultations.

**Educational and teaching activities**

From the submitted reference for teaching workload it is evident that for the period 2017-2023 the average teaching workload of assistant-in-chief Maria Levkova, MD, PhD amounts to **1009 hours of exercises**, with a **norm of 220 hours**. Of these, **399 hours** are teaching in Bulgarian and **610 hours** are teaching in English.

The information given above clearly demonstrates candidate's active participation in the educational and teaching process at the Department of Medical genetics.

**Others**

Assistant-in-chief Levkova has presented certificates for **9 completed professional trainings**, of which **7 abroad** and **2 in Bulgaria** in: medical genetics, clinical genomics, genetic diagnostics, European certificate in medical genetics and genomics, molecular andrology and others.

The applicant has a recognized specialty in Medical genetics (2021) at the Ministry of Health.

**Critical remarks and recommendations**

I have no substantive criticisms of the candidate scientific work. It is evident from the materials presented and the science metrics that she is a promising young scientist. With a view to her future growth, I have the following **recommendations**:

1. To focus her research activities in a particular area of medical genetics, which will support her future development as a scientist and the establishment of a school of young scientists trained and mentored by her.
2. To continue to maintain a high publication activity by increasing the number of original articles at the expense of review articles.

**Conclusion**

The materials submitted by the candidate for the competition clearly demonstrate that assistant-in-chief Maria Kostadinova Levkova, MD, PhD covers all scientometric indicators and meets the requirements for acquiring the academic position "Associate Professor", according to the Law for the development of the academic staff in the Republic of Bulgaria and of the Regulations for the development of the academic staff in the Medical University of Varna. The presented scientific works and evidentiary material for his scientific activity meet the quantitative and qualitative criteria for tenure the academic position "associate professor".

I give my **positive assessment** to **assistant-in-chief Maria Kostadinova Levkova, MD, PhD**, as a participant in this competition, and I reccomend to the respected members of the Scientific Jury to award **assistant-in-chief Maria Kostadinova Levkova, MD, PhD** the academic position "**associate professor**" in the scientific specialty „**Medical genetics**", area of higher education **4. Natural sciences, mathematics and informatics**, professional field **4.3. Biological Sciences** for the needs of the Department „Medical genetics" at the Faculty of Medicine, Medical University of Varna.



January 30, 2024

Varna

Opinion prepared by

/prof. Bistra Kaltcheva, PhD/