To: The Chairman of the Scientific Jury,
determined by an order of the Rector of MU-Plovdiv

OPINION

by Professor Mariana Stoycheva Vertigova, MD, PhD, DMSc

external member of the Scientific Jury, specialist in infectious diseases

Regarding the dissertation of **Stanislav Vasilev Kotsev , MD "Prevalence and clinical characteristics of hepatitis E virus infection in persons from risk groups for the period 2018. – 2022"**
presented for awarding the educational and scientific degree **"DOCTOR"**
PhD program: "Infectious Diseases", field of higher education 7 - "Health and Sports" in the professional field 7.1 - "Medicine" and scientific specialty "Infectious Diseases"

Supervisors: Prof. Oliana Boykinova, MD, PhD, Faculty of Medicine, and Prof. Nikolay Vatev, MD, PhD, Faculty of Public Health, MU-Plovdiv

Form of doctoral studies: independent preparation

The dissertation was discussed and sent for public defense by the Department of Infectious Diseases, Parasitology and Tropical Medicine, MU - Plovdiv (protocol No. 5 /19.05.2026) and by decision of the Faculty Council of the Faculty of Medicine, MU-Plovdiv (protocol No. 5/03.06.2026).

The set of materials has been submitted on digital carrier for the awarding of the Scientific degree DOCTOR in accordance with the Law on the Development of the Academic Staff in the Republic of Bulgaria, the Regulations for its implementation and the Regulations of the Medical University - Plovdiv.
The documents are correct and precise, which facilitates their analysis and evaluation.

Career profile of the PhD student

Stanislav Vasilev Kotsev, MD graduated from the Bertolt Brecht Language School, Pazardzhik, with English and German. He is a graduate of the Faculty of Medicine, Sofia University St. Kliment Ohridski, where, after a six-year academic course, in 2017 he graduated as a medical doctor. Since April 2018 he has been working as a doctor at the Pazardzhik Hospital AD. In 2022 he acquired a specialty in infectious diseases and works as a specialist with the same medical institution at present. He works on his PhD (own initiative) in Infectious Diseases at the Department of Infectious Diseases, Parasitology and Tropical Medicine, MU-Plovdiv, by order of the

Rector of MU-Plovdiv, R-643/22.02.2024. His academic profile is complemented by the position of a part-time assistant professor at the Department of Zoology, Faculty of Biology, Plovdiv University Paisii Hilendarski, in the discipline *Vectors in Transmissible Infections* for 3rd year students, specialty: "Microbiology and Virology", which testifies to a sustained interest in scientific and teaching activities.

At present, Stanislav Vasilev Kotsev MD combines intensive clinical work, teaching and research, and his professional development is aimed at improving diagnostic and therapeutic approaches in the field of Hepatitis E viral infection, tick-borne infectious diseases and casuistic infectious diseases.

He is the author and co-author of 41 publications, 20 of which are indexed in Scopus and Web of Science.

He has participated in five projects, three of which are international.

He speaks English and German.

Topicality

The problem considered in the dissertation has indisputable relevance and high clinical significance. Viral hepatitis E (VHE) is another etiologically deciphered variant of viral hepatitis. Intensive scientific research has allowed the improvement of diagnostic methods of hepatitis E virus (HEV) infection. It was found that the prevalence of HEV infection is far higher than expected. In Europe, about 15% of acute viral hepatitis is etiologically related to HEV. The risk factors for severe and even fatal course are less studied, which makes the choice of topic for the dissertation especially appropriate. The work is a kind of continuation of the research of the team in which Dr. St Kotsev works, under the guidance of Associate Professor M. Pishmisheva, MD, PhD and her dissertation "Clinical and epidemiological aspects of acute hepatitis E viral infection in Pazardzhik region". The emphasis in the present dissertation is on HEV infection in people with previous hepatopathies, CKD and chroniodialysis, immunosuppression and alcohol abuse, patients with a real risk of severe and fatal course.

I think that the topic of the dissertation is extremely relevant.

The dissertation is written on 138 pages and structured according to the accepted rules: literary review, purpose, tasks, materials and methods, results, discussion, conclusions, contributions and bibliography.

In terms of structure, a balance has been achieved in the voluminous presentation of the individual components of the dissertation.

The literature review is prepared on the basis of 403 literature sources, of which 366 are in Latin and 36 in Cyrillic and show very good information of the author on the problems under consideration. It is commendable that the PhD student has cited a significant number of Bulgarian authors (55) who have worked on the problem and contributed to the clarification of the characteristics of HEV infection in the Bulgarian population.

The literature review presents a comprehensive and in-depth review of the etiology, epidemiological characteristics of VHE, with a definition of risk groups for infection and for severe course. The factors determining the severity of the disease, pathogenesis and clinical characteristics in different risk groups, as well as in different geographical regions are presented.

The intelligently presented literature review testifies to the good theoretical preparation of the PhD student and the desire to systematize the literature data and their comparison.

A good impression is made that the review does not exceed 30 pages and ends with **a Summary,** which formulates the insufficiently studied aspects regarding VHE infection, and argues the need for the current scientific development.

The goal and tasks are clearly and precisely defined. The goal fully corresponds to the topic, and the set 5 specific tasks allow its implementation.

Materials and methods describe in detail the impressive number of patients observed (616), with 371 from risk groups, and the control group including 245 individuals, with no data on risk factors (from the general population and blood donors). The rich clinical material allows for reliable interpretation and correct conclusions.

All methods used for data collection are described: clinical, laboratory, serological, molecular-biological (performed in the NRL "Hepatitis Viruses" NCIPD), imaging diagnostics and a set of statistical methods for processing the results obtained. I would like to note that the methods used are described accurately, clearly, without unnecessary, burdensome details, since the dissertation is a clinical development.

The chosen research methodology allows achieving the goal and obtaining an adequate response to the specific tasks.

The results are presented sequentially, in order of the tasks set and convincingly illustrated with 39 author's figures of high quality, attractive and diverse design and 28 tables in the main text, and another 31 tables in the Appendices. The Results present the author's data on the studied demographic, epidemiological and clinical characteristics; laboratory, immunological imaging and diagnostic findings in the patients in the groups under consideration.

The PhD student presents data on the prevalence of HEV infection in the region of Pazardzhik among people from risk groups, such as from the general population and blood donors. 34% of all examined, mostly men of Bulgarian origin, aged >55 years, are seropositive, and most often *anti-HEV IgG(+) is established.*

The frequency of seropositivity and the type of specific anti-HEV antibodies in different groups are compared. Seropositive are more often people from risk groups, most often with concomitant liver disease (CHD) ($p<0.05$). Specific *anti-HEV is* found in 51% of individuals with CHD, significantly more often in those with hepatic cirrhosis and steatosis. Anti-HEV IgG are most common, with a higher incidence in people with risk factors (excluding those on dialysis treatment).

Clinical manifestations of hepatitis were detected in 22% of 210 anti-HEV positive individuals. The patients were mainly male, Bulgarian ethnicity and age > 55 years. It

has been found that clinically manifested HEV infection develops significantly more often in persons with risk factors, especially those with CHD (liver cirrhosis, chronic viral hepatitis, hepatic steatosis). A very detailed clinical analysis of subjective, objective signs and laboratory abnormalities, monitored in dynamics, is presented. In patients with risk factors, the course is more severe, with complications and extrahepatic manifestations.

A mortality rate of 6.8% was found, most often associated with acute or acute on chronic liver failure.

I would especially like to note the numerous comparisons made regarding the epidemiological, clinical characteristics and laboratory parameters of HEV infection in patients from different subgroups.

I admire the prepared algorithm for testing for HEV of donor blood and persons from risk groups, which increases the clinical significance of the dissertation. Recommendations to various medical specialists would help diagnostic thinking and increase the likelihood of verification of HEV infection in suspicious clinical scenarios.

The discussion follows the logic of the presented results, which are thoroughly analyzed and compared with those existing in the modern medical literature. The discussion is in-depth and includes logical judgments and hypotheses. The PhD student reasonably compares his own observations with the data from the literature, analyzes both statistically significant and insignificant results and seeks a pathophysiological explanation of the established dependencies.

5 conclusions have been formulated, entirely arising from the results obtained. They are convincing, scientifically reasoned and correspond to the goals and objectives set.

3 contributions are presented with original scientific, 3 with scientifically applied and 1 with a confirmatory nature, with which I agree.

Evaluation of the publications on the dissertation.

4 publications are attached, and Dr. Stanislav Kotsev is the first author in two of them and actively participates in the generation of the rest. Three of the publications are in refereed international journals and two are in English. 5 participations in scientific forums are also presented, and Dr. Kotsev is the first author of the scientific works.

The dissertation of Dr. Stanislav Vasilev Kotsev is his personal work, and the formulation of the conclusions and contributions are the merit of the doctoral student and his supervisors.

The abstract is prepared according to the requirements and reflects the main results achieved in the dissertation.

Critical remarks and recommendations

In the process of preliminary review, a number of recommendations were made aimed at improving individual sections of the dissertation. In the final version, the author has taken into account to a large extent the recommendations made.

Personal impressions

I know Dr. Stanislav Kotsev as an excellently trained and responsible doctor, an ambitious and intelligent young colleague with a taste for scientific work. He strives for new knowledge, new diagnostic and therapeutic models that can be implemented in infectology

Conclusion

The dissertation is an independent, in-depth and scientifically grounded study, which ***contains scientific and applied results, representing an original contribution to science*** and meets **all the** requirements of the Law on the Development of the Academic Staff in the Republic of Bulgaria, the Regulations for the Implementation of the Academic Staff Act and the Regulations of the Medical University - Plovdiv. The dissertation shows that the PhD student Stanislav Vasilev Kotsev has in-depth theoretical knowledge and professional skills in the scientific specialty Infectious Diseases by **demonstrating** qualities and skills for independent scientific research.

With pleasure and firm conviction, I give my ***positive assessment*** of the conducted research, presented by the above-reviewed dissertation, abstract, achieved results and contributions, and ***propose to the honorable scientific jury to award the educational and scientific degree of 'Doctor'*** to Dr. Stanislav Vasilev Kotsev in the doctoral program in Infectious Diseases

Professor Mariyana Stoycheva, MD, PhD, DMSc
Plovdiv, 23.10.2026

Заличено на основание
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