Opinion

from

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Internal member of the Scientific Jury for awarding scientific and educational
degree**"Doctor"**

Subject: Dissertation for awarding the **scientific and educational** degree "Doctor"

Field of higher education: 7. "Health and sport"

Professional field: 7.1. "Medicine "

Doctoral program : "Hematology and blood transfusion" 03.01.39

Author: Dr. Vanya Slavcheva

Form of doctoral studies: independent doctoral studies

Department: Second Department of Internal Medicine , Board of Hematology

Topic : "Clinical application of prognostic factors and their integration into a scale for risk assessment and time to treatment , in untreated patients with B-chronic lymphocytic leukemia"

Scientific adviser: Prof. Dr. Liana Gercheva-Kyuchukova, Ph.D.

Opinion prepared by: Assoc. Prof. Dr. Ilina Dimitrova Micheva, MD, Head of Clinical Hematology Clinic, University Hospital "St. Marina "EAD, Varna, Head of the Board of Hematology at the Second Department of Internal Medicine, Medical University " Prof. Dr. Paraskev Stoyanov ", Varna.

By order № 109- P- 377 / 06 . 10 .20 20 of the Rector of MU, Varna, on the basis of a decision of the Faculty of Medicine Council № 29 / 28.09.2020. I have been selected a member of the scientific jury to discuss the dissertation work of Dr. Vanya Slavcheva, on the basis of protocol № 1 / 19.10.2020. I have been appointed to prepare an opinion on the procedure for awarding the educational and scientific degree "Doctor".

Brief data on the professional development and qualification of the doctoral student:

Dr. Vanya Slavcheva graduated with a master's degree in medicine in 1990 at the Medical Institute - Pleven, Bulgaria. In 1997 she acquired a BC/BE degree in internal medicine at the Medical University – Sofia; in 1999 she acquired the second degree in clinical hematology at the Medical Institute - Pleven. Since 1996 she has been working as an assistant at the University Hospital "Dr. Georgi Stranski" / Clinic of Hematology, Pleven (in the period 2000-2014 as a senior assistant). In 2019, V. Slavcheva was enrolled as a PhD student in an independent form of study at the Second Department of Internal Medicine, Board of Hematology at MU-Varna (order of the Rector of MU Varna – P-109-126\01.04.2019). Topic of the dissertation "Clinical application of prognostic factors and integrating them into scale for assessment of risk and time to treatment in previously untreated patients with B-chronic lymphocytic leukemia". She was expelled with the right to defense by order of the Rector of MU-Varna № R-109 / 06.10.2020. The doctoral student's research interest is in the field of CLL. She has participated in two scientific research projects on CLL funded by Medical University- Pleven . In this regard, the current dissertation has been developed.

Relevance of the topic

Chronic lymphocytic leukemia (CLL) is one of the most common leukemias in adults. About 80-85% of patients do not meet the criteria for initiating treatment and are subject to periodic monitoring in order to determine a strategy for behavior and choice of therapeutic option. The risk stratification of the patients is of great importance both for the follow-up and determination of the time until the initiation of treatment, as well as for the first line of therapy choice.

In recent years, thanks to the development and improvement of immunology, immunogenetics and molecular biology, a number of prognostic models, indices and scales for risk stratification in patients with newly diagnosed B- CLL have been developed and proposed . In 2016, an International Prognostic Index was proposed - CLL-IPI, which serves not only , for predicting overall survival but also to assess the time to first treatment .

Nowdays, there are no reported studies in our country based on evidence related comprehensive assessment of available molecular genetic and classical prognostic factors, although the need for a more accurate assessment of the risk of progression is becoming increasingly important to introduce new drugs for the treatment of B- CLL.

Characteristics and evaluation of the dissertation.

The presented for review dissertation is structured according to the standards adopted in our country for a dissertation for obtaining the scientific degree "Doctor" in medicine. The dissertation is written on 123 standard pages and includes: content (1 page), abbreviations (3 pages), literature review (34 pages), purpose and tasks (1 page), materials and methods (10 pages) , results (30 pages), discussion (17 pages), conclusion (1 page), conclusions (2 pages), list of publications related to the dissertation (1 page), bibliography (18 pages). The bibliographic list contains a total of 199 literature sources, 5 in Cyrillic and 194 in Latin. The work is illustrated with 30 tables and 45 figures.

The literature review is set out on 34 pages. The biology of CLL in historical and modern aspect, the pathogenesis and the role of BCR signaling, the clinical course are presented. In a separate section, the published scientific data on the classic prognostic factors in CLL related to the patient (age, sex, comorbidities, ECOG status) related to the disease (Rai / Binet stage , beta 2-microglobulin , serum thymidine kinase) are reviewed, IGVH mutational status, CD 38, CD49d, ZAP70, genetic instability) associated with treatment. The importance of new prognostic factors such as cytogenetic and molecular markers, IGVH mutational status, flow cytometric markers (CD38, CD49d, ZAP70, serum CD 23), as well as their incorporation into modern prognostic scoring systems are discussed in detail .

At the end of the literature review the topicality of the problem as well as the grounds for the development of the problem in Bulgarian conditions are considered, which leads to the formulation of a clear scientific hypothesis on which the dissertation is based.

The aim of the study is clearly and precisely formulated, namely to study and analyze the impact of available clinical,laboratory,molecular and genetic factors related to the specific

characteristics of both the patient and his disease, and to assess their impact on time to the first treatment in untreated B-CLL patients.

The set 6 main tasks are clearly formulated and follow the logically set goal.

The Materials and Methods section is presented on 10 standard pages. The study covers a period of three years and the design includes two types of observation: prospective and retrospective. 97 patients diagnosed with B-CLL were analyzed. Routine methods for diagnosing and staging CLL are described. The specific methods used to assess the risk factors studied in the dissertation are presented in detail: Flucytometry, Fluorescence *insitu* hybridization (FISH), multiplex polymerase chain reaction after reverse transcription was used to study the expression of LPL and ADA29 genes. (RT) Polymerase Chain Reaction (PCR) in complementary DNA (cDNA).

The data from the study were processed with statistical software product SPSS 19. The statistical methods used and the software product for data analysis and presentation are described in detail.

The section "Results" is presented in 30 standard pages. The results of the set 6 tasks are analyzed, systematized, presented concisely and consistently and illustrated with the help of tables, figures, graphs and boxing tables. The correlation of each of the studied risk factors with the time to first therapy - demographic, clinical and laboratory (stage, spleen size, markers of tumor load-absolute lymphocyte count, β2-M), molecular genetic markers, mutation status of IGVH**.** The importance of well-known and accessible risk factors for the time to first therapy and survival, has been confirmed.

In the Discussion part, which is presented in 17 pages, the own results are analyzed and discussed in the context of the data published so far from the scientific literature. The considered prognostic factors in the diagnosis of the disease can be applied in clinical practice to assess the time to treatment. The assessment of mutational status and chromosomal aberrations is of particular importance due to not only their prognostic but also predictive role.

The conclusions logically follow the goal and the set tasks.

The bibliography *contains* 199 literary sources, of which 4 are by Bulgarian authors, one by Russian author. The sources are selected in a targeted manner, which shows the doctoral student's ability to select important scientific information with high research awareness in the chosen field.

Publications related to the dissertation

Four full-text publications are presented. In three of the presented publications the dissertation is the first author, which testifies to its leading role in conducting research and preparation of scientific publications.

The abstract makes a brief summary of the dissertation and is set out on 62 pages. The most important researches, results and discussions on the scientific problem are summarized.

Conclusion

The presented project of dissertation work on topic " Clinical application of prognostic factors and their integration into the scale for assessing risk and time to treatment in previously untreated patients with B-chronic lymphocytic leukemia", by Dr. Vanya Slavcheva is updated and contains important scientific and scientific and applied results . The dissertation work corresponds to all requirements of the development of academic staff in the Republic of Bulgaria (LDASRB) and its implementing Rules of the Medical University - Varna . The goal has been achieved, the set tasks have been fulfilled. The contributions have not only scientific but also practical value. For the first time, a method is used to study the mutational status of IGHV and its significance for the time to the start of therapy is assessed. The presented publications and scientific communications meet the requirements. In these circumstances, I believe that the dissertation of Dr. Vanya Slavcheva meets all the requirements for awarding the scientific and educational degree "Doctor" and I strongly recommend the esteemed jury to vote positively.

Date: 02.11.2020   Opinion prepared by: Assoc Prof.: Dr Ilina Micheva MD

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