

OPINION

by Prof. Fedya Nikolov, MD, PhD

Head of the First Department of Internal Medicine at the Medical University of Plovdiv

Regarding: dissertation of Assoc. Prof. Maria Negreva, MD, PhD. Associate Professor of Cardiology at the First Department of Internal Medicine, Education Sector of Cardiology, Medical University of Varna, for the award of the degree "Doctor of Science" with title: **Early deviations in the coagulation and fibrinolytic system in paroxysmal atrial fibrillation.**

By decree of the Rector of the Medical University of Varna, №P-109-216 dated 20.05.2022, I have been appointed to prepare an opinion that complies with the requirements of the Law for the Development of Academic Staff in Bulgaria and the Regulations of the Ministry of Education and Science and Medical University of Varna for its implementation.

1. Brief biographical data:

Assoc. Prof. Maria Negreva earned her Master's degree in Medicine in 2006 from the Medical University of Varna. In the same year she was appointed as resident at the First Cardiology Clinic, where she still works. In 2008 she became an Assistant Professor of Cardiology, and in 2016 an Associate Professor of Cardiology. In 2013 she acquired the specialty Cardiology, and in 2015 she was awarded the degree "Doctor" in the scientific specialty Cardiology. Assoc. Prof. Negreva is a co-author of 5 textbooks for students and graduates. She is supervisor to two doctoral students in Cardiology.

2. Structure of the work: It contains the obligatory elements for a dissertation work and they are balanced in volume and logically arranged:

- Contents: 2 pages.
- Introduction: 2 pages.
- Literary Review: 70 pages.
- Aim and Tasks: 2 page.
- Materials and Methods: 15 pages.
- Results and Discussion: 80 pages.
- Conclusions: 2 pages.
- Contributions: 3 pages.
- Bibliography: 28 pages. There are 577 sources, of which 572 in Latin and 5 in Cyrillic. Almost half of them (242 of the cited sources) have been published in the last 10 years.
- It is illustrated with 24 tables and 69 figures.

3. Evaluation of the relevance of the topic

The dissertation considers a particularly relevant topic, presenting early deviations in the coagulation and fibrinolytic system associated with clinical manifestation of short episodes of paroxysmal atrial fibrillation lasting ≤ 24 hours. Its significance is determined by the following facts:

- Paroxysmal atrial fibrillation has a high thromboembolic potential and remains one of the leading causes of acute ischemic stroke, despite its short duration and often mild course;
- Adequate anticoagulant therapy remains key to its effective prevention;
- The periprocedural thromboembolic potential of brief (≤ 24 hours) episodes of paroxysmal atrial fibrillation is still debatable and postprocedural anticoagulation remains an unresolved issue, even in recent European guidelines for the treatment of atrial fibrillation;
- In the light of modern notions of coagulation, the exact mechanisms of thrombosis in atrial fibrillation remain unclear.

4. Literary review

The literature review is detailed and logically arranged, it clearly outlines the need for research and solution of presented tasks.

Assoc. Prof. Negreva discusses the anticoagulant approach in paroxysmal atrial fibrillation in the long term, outlining the indisputable role of the CHA₂DS₂-VASc score, as well as current trends in the perception of the time characteristic of the episode as a factor relevant to the thromboembolic potential of the disease. She presents the unresolved aspects of anticoagulant prophylaxis after acute recovery of sinus rhythm. Data from clinical and experimental studies on the coagulation and fibrinolytic system, main components of hemostatic imbalance in atrial fibrillation, are analyzed in the light of the topic. Data, describing the possibilities of hemostatic indicators as prognostic factors for the manifestation of the disease and related thromboembolic incidents are derived from the literature.

5. Aim and tasks

The aim of the dissertation is clearly stated. It is fully achieved by solving the set tasks. The refinement of the tasks comes after thorough knowledge of classical and modern ideas about the coagulation balance by the candidate, which is evident from presented data on the structure and normal functioning of both systems in the literary review.

6. Material and methods

The following facts are impressive:

- precise selection of patient and control group for maximum clinical reliability of results;

- simultaneous study of a large number of indicators, a total of twenty (fourteen coagulation indicators, six indicators of fibrinolysis), which complement each other;
- multicomponent nature of the statistical analysis.

7. Results and Discussion

The results are significant, informative enough and allow to draw the conclusions. They fully reflect the coagulation status of patients in brief (≤ 24 hours) episodes of paroxysmal atrial fibrillation. Undoubtedly, they show presence of hypercoagulability with significant activation of the coagulation and fibrinolysis systems. They also present specific features of the established coagulation imbalance, namely participation of FXII and FXI, present endothelial dysfunction and damage, as well as decreased activity of the main inhibitors of the fibrinolytic system PAI-1 and $\alpha 2$ -antiplasmin. For the first time, data on the influence of the thromboembolic risk characteristic of the patient, defined by the established scale CHA_2DS_2 -VASc score, on the coagulation status by directly comparing the values of the studied hemostatic indicators among patients with low thromboembolic risk (CHA_2DS_2 -VASc score = 0 in men/ 1 in women) and those at increased risk (CHA_2DS_2 -VASc score ≥ 1 in men/ ≥ 2 in women). The dependence of the coagulation and fibrinolytic activity on the time characteristic of the episode is objectively presented through logistic regression models. With the help of ROC analysis and Cox regression, hemostatic indicators are presented in terms of their predictive ability to manifest atrial fibrillation and related thromboembolic events.

8. Evaluation of contributions

This is the first study of its kind, which simultaneously and in detail presents the systems of coagulation and fibrinolysis in short (duration ≤ 24 hours) episodes of paroxysmal atrial fibrillation in a clear clinical model with patient and control groups.

The well-developed methodological study allows for the formation of both fundamental contributions to the pathophysiological mechanisms of impaired coagulation balance and as well as one of applied nature, related to optimizing anticoagulant prophylaxis in PAF and providing opportunities for new studies to predict the disease and its thrombotic incidents.

9. Critical notes on the dissertation

Assoc. Prof. Negreva critically notes the main limitation of the dissertation: one-time study of hemostasis indicators, predetermined by the goal. This does not allow to specify the duration of post-procedural anticoagulation from own results.

The ideal version of the study would be to examine the clinical benefits of the conclusions made, but this goes far beyond the purpose of the dissertation.

10. Scientometric analysis of the dissertation

All indicators for the minimum requirements of the Medical University of Varna for obtaining the scientific degree "Doctor of Science" are exceeded. The candidate presents 10 full texts scientific publications related to the dissertation. 5 articles are indexed in Web of

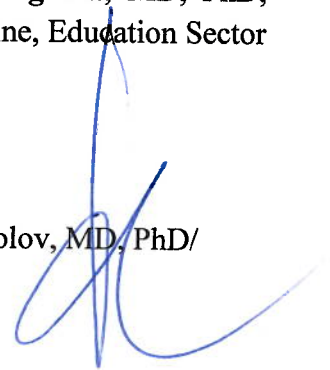
Science/Scopus, two from them in impact factor journals. She is the first author in 7 publications, and second in 3. She presents also 3 abstracts from international congresses, two of them published in impact factor journals. 8 positive citations of presented publications are found in international scientific databases. The total impact factor in connection with the scientific publications related to the dissertation is 51.465.

11. Conclusion

The above facts, related to the present dissertation **Early deviations in the coagulation and fibrinolytic system in paroxysmal atrial fibrillations** show that it has an original character, fully corresponds to the aim and tasks. This allows me to give a positive assessment of the dissertation and recommend to the esteemed members of the Scientific Jury to award the scientific degree "Doctor of Science" in the scientific specialty Cardiology, professional field 7.1 Medicine, field of higher education 7. Healthcare and Sports to **Assoc. Prof. Mariya Negreva, MD, PhD**, Associate Professor of Cardiology at the First Department of Internal Medicine, Education Sector of Cardiology at the Medical University of Varna,

05/30/2022

Sincerely,
/Prof. Fedya Nikolov, MD, PhD/

A handwritten signature in blue ink, appearing to be 'F. Nikolov', is written over the typed name of Prof. Fedya Nikolov. The signature is fluid and cursive, with a large loop at the end.