

REVIEW

by

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Professor of Clinical Laboratory to the Medical University - Varna, Professor-Head of the Clinical Laboratory at the Multi Profile University Hospital for Active Treatment (UMBAL) "St. Ekaterina" EAD – Sofia, a member of the Expert Council in (for) the Medical Specialty "Clinical Laboratory" at the Ministry of Health by Order No. RD-01-133/ 09 May 2019 by the Minister of Health.

Regarding: The Competition announced in the State Gazette, Issue No. 61 of 02 August 2019 for the academic position of "Associate Professor" in the professional field "Clinical Laboratory" in the MHAT "National Heart Hospital" Sofia.

The competition has been announced in the field of higher education 7. "Healthcare and Sports", in professional field 7.1. "Medicine", and scientific specialty "Clinical Laboratory".

By Order No. 411 / 14.10.2019 of the Executive Director of the MHAT "National Heart Hospital" I have assigned as a member of a Scientific Jury under a procedure for the acquisition of the academic title of "Associate Professor" at the Department of Clinical Laboratory of the MHAT, Sofia on the grounds of the Decision of the Scientific Council of the "National Heart Hospital" Sofia №8 / 01.10.2019. At the first session of the Scientific Jury dated 16 October 2019 I have been assigned to prepare a REVIEW on the competition. For the competition announced in the State Gazette, Issue No. 61 of 02 August 2019 for the academic title of "Associate Professor" at the "Clinical Laboratory" in the MHAT "National Heart Hospital" Sofia the only candidate is Dr. Dobrinka Dineva Savova, MD, Senior Assistant Dr. Dobrinka Dineva Savova, MD, Head of the Clinical Laboratory of the MHAT-National Heart Hospital EAD.

For the purposes of the competition Dr. Dobrinka Dineva Savova, MD has presented all necessary documents in related to the national requirements as stipulated by the Development of Academic Staff in the Republic of Bulgaria Act and the Regulation on the application of the Development of Academic Staff in the Republic of Bulgaria Act, as well as all documents in accordance with Art. 1, P. 4 of the Rules on the Terms and Conditions for Acquisition of Academic Degrees and Occupation of Academic Positions at the "National Heart Hospital" Sofia. Dr. Dobrinka Dineva Savova is a healthy individual who has never been convicted.

1. Brief biographical data.

Dr. Dobrinka Dineva Savova, MD was born in 1965. She completed her secondary education in 1983 at the 18th High School, Sofia. She graduated medicine at the Medical Academy-Sofia in 1990. She acquired the medical specialty "Clinical laboratory" in 1997. In 2013 she defended her doctoral thesis on the topic: "Monitoring of platelet activity by impedance aggregometry in the treatment of clopidogrel and / or aspirin in high-risk patients with coronary artery disease" and obtained an educational and scientific degree "doctor". Dr. Dineva started working in 1991 as a 3rd degree researcher - academic position at the Laboratory diagnostics department of the National Heart Hospital" Sofia EAD. In 1997 she

was promoted to a second-degree researcher at the same workplace, and since 2010 she has been promoted to a first-degree researcher / Senior Assistant at the same workplace. Since 2011 she has been the Head of the Clinical Laboratory at National Heart Hospital. We are witnessing a gradual, constructive professional development, both in direct laboratory and scientific and teaching aspects. Currently, Dr. Dineva has 28 years of professional experience.

2. Scientific activity

The quantitative criteria for the scientific production of Dr. Dobrinka Dineva Savova is as follows: She is the author and co-author of a total of 20 scientific works:

- PhD thesis: "Monitoring of platelet activity by impedance aggregometry in treatment with clopidogrel and / or aspirin in high-risk patients with coronary artery disease", defended 2013.
- Monograph on the topic: "Biology of platelets, functional tests and their application in clinical practice for evaluation of platelet response", published in 2019.
- Scientific articles in which Dr. Dineva is the author and co-author are a total of 18. Five articles of these are published in foreign and Bulgarian publication which are referenced and indexed in Scopus.

Publications in foreign and Bulgarian non-refereed scientific peer-reviewed journals or in full-text edited publications - 10 publications. Chapter of a collective monograph published - 3 publications.

In the 5 of these publications, Dr. Dineva is the first author, 6 is the second author, 6 is the third author and 1 is the fourth author. There are no publications available in Impact Factor (IF) or Impact Rank (SCImagoJournalRang - SJR).

- According to the data from the Central Medical Library from 2019 Dr. Dineva's scientific publications have been cited 27 times in total. Two of them are in foreign scientific publications and 25 in Bulgarian sources. Participation in scientific projects is not available.

- Dr. Dineva has a total of 62 participations in scientific forums in Bulgaria and abroad.

Participation in oral reports - 22; participation with abstracts - 40. Of these, 15 are at congresses abroad and 25 at congresses and conferences in Bulgaria, with summaries printed in the Abstracts Volume Books.

- The scientific contributions from Dr. Dineva's research work are significant in the field of blood coagulation and platelet aggregation. The work in a hospital with cardiac surgery and cardiac profiling contributes to this. For the first time in Bulgaria in accordance with international requirements the method of impedance aggregation was validated. In determining the series reproducibility and the reproducibility of the individual measuring channels, no significant differences were found, the determined coefficients of reproducibility (CV%) were up to 12%. Based on the significantly higher CV% values obtained when determining irreproducibility at 180 minutes, the authors did not recommend a platelet aggregation on MULTIPLATE after 120 minutes of blood collection (A). The reference ranges for ADP, ASPI and TRAP tests for the Bulgarian population from a selected control group of 45 healthy volunteers aged 18 to 68 years have been determined. The specified reference ranges for ADP, ASPI, and TRAP tests in the healthy control group do not require gender and age separation and are very close to the reference limits specified by the manufacturer (A).

Significant are the contributions from studies evaluating the response to antiplatelet therapy in patients with coronary diseases and subsequent therapeutic interventions. Following monitoring of dose- and time-dependent platelet inhibition by clopidogrel, at a loading dose of 300 mg and maintained at 75 mg, it was found that a maximum sustained level was achieved on day 5 (72% inhibition of ADP-induced platelet aggregation) , which persists until

the 30th day (70%) (Г10.13). Contribution is the evaluation of the diagnostic reliability of the ADP test by ROC analysis.

An inverse correlation was found between the ADP test and the time of onset of thrombosis thus higher residual platelet activity is associated with the early onset of instant thrombosis within 14 days of stenting (A, B, Г3, Г4). Dr. Dineva participated in one of the first evaluations in Bulgaria of dual antiplatelet therapy, as well as in evaluating the effect of various antiplatelet drugs. In patients with low platelet aggregation, DAPT was evaluated for the incidence of patients with LPR and bleeding when using P2Y12 receptor inhibitors - clopidogrel, prasugrel and ticagrelor and the ADP-induced aggregation limit for risk of hemorrhage was defined. The increased response to P2Y12 receptor inhibitors has been defined as an ADP-test <18 U. The antiplatelet therapy in hemorrhage patients is individualized by reducing the maintenance dose, replacing one P2Y12 inhibitor with another, thereby achieving optimal inhibition and reduction of bleeding. Individualization of antiplatelet therapy leads to a reduction in the risk of recurrent haemorrhage without increasing the risk of ischemic events based on the measured ADP aggregation. A significant advantage in optimizing therapy is the strategy of reducing the maintenance dose of P2Y12 inhibitors and aspirin, which is demonstrated by platelet aggregation values within the therapeutic window of inhibition, discontinuation or reduction of bleeding, as well as lack of second hemorrhagic. Too few studies have been conducted with dose modification and follow-up (A, B, Г4, Г13).

The degree of inhibition of platelet aggregation in chronic treatment with acetisal cardio 100 mg (Actavis), aspirin protectant 100 mg (Bayer) was compared. The patients with stable chronic coronary artery disease were on dual antiplatelet therapy - clopidogrel 75mg and aspirin. Patients were monitored after 3 months of treatment with successive intersection of aspirin protect 100 mg, Acetisal cardio 100 mg and Acetisal cardio 75 mg. Platelet aggregation was measured at least one month after percutaneous coronary intervention. The presence of Acetisal Cardio 75 mg (Actavis) registered in Bulgaria, the achieved "desired" target inhibition of platelet aggregation and the low cost would have clinical benefits during the chronic phase of treatment of atherothrombotic diseases. The individual response to aspirin protect 100mg and Acetisal cardio 100mg is fully comparable as measured by inhibited platelet aggregation (Г4, Г12).

The study of perioperative changes in platelet function in patients undergoing ECC surgery is another unique contribution to Dr. Dineva's scientific work. Distinctive boundaries of ADP and ASPI bleeding risk tests were developed before and after ECC. Preoperatively, values of the ADP test below 44.0 U and the ASPI test below 40 U correlate best with blood loss. In the early postoperative period, low values of ADP and ASPI tests below 22 U were associated with increased postoperative bleeding. Platelet aggregation studies with ADP test, ASPI test and TRAP test are useful for the control of adequate perioperative haemostasis (Г3, Г6).

For the first time in Bulgaria, genetic tests was performed in order to establish the frequency of carrier of the CYP2C19 * 2 "loss-of-function" allele and of the CYP2C19 * 17 "gain-of-function" allele and their influence on the phenotype response in 104 patients after PCI. The incidence of CYP2C19 * 2 "loss of function" allele was found to be higher in the group with high residual platelet aggregation (ADP-test 72 ± 11 U) - 37% compared to the group with adequate platelet response (ADP test 22 ± 8 AU) - 16.7%, $P < 0.05$. The diagnostic sensitivity of CYP2C19 * 2 genotyping 0.70 has been established to predict high residual platelet aggregation (HPR) or "poor response" to clopidogrel. Carriers of the CYP2C19 * 2 allele of "loss of function" are 70% of patients with HPR. Genotyping of CYP2C19 * 2 may predict the risk of suboptimal response, but does not replace quantitative functional tests for platelet response (Г14).

Another essential area of Dr. Dineva's research is coagulation. Changes in laboratory hemostasis and haematological tests to assess coagulation and fibrinolysis and associated postoperative bleeding in cardiac surgery patients are being investigated. Postoperative bleeding in 440 patients from the Cardiac Surgery Unit of the National Heart Hospital was associated with a statistically significant decrease in platelet count (44%), fibrinogen (31%), antithrombin III (23%), hemoglobin (20%). %, Er (by 23%), Hct (by 21%) and PT prolongation by 3 seconds. A decrease in platelet count below $65 \times 10^9 / l$ and fibrinogen below 1.4 g / l was related by increased postoperative bleeding (Г6). Dr. Dineva is an innovator in the monitoring of low-molecular-weight heparin (LMWH) therapy by measuring anti-IXa activity in patients with CAD and PTE. Adjusted doses achieve the desired antithrombotic effect (Г7). Dr. Dineva participates in presenting the results of the first-time monitoring of low molecular weight heparins in pregnant women with hereditary thrombophilic defects. Monitoring of LMWH is beneficial in rare cases of bleeding and in high-risk pregnancies with insufficient anticoagulant response (Г2). Laboratory monitoring of patients on oral anticoagulant therapy with Vit.K anticoagulants is another contribution to Dr. Dineva's research. The advantages of INR determination from capillary blood with Point of Care self-monitoring devices - Coagucheck S (Г9) were evaluated. In 147 patients on Sintrom therapy, parallel measurements of PT (INR) from capillary blood were performed with Hepato Quick tests on Thrombotrack, Coagucheck S test strips and on hemostasis analyzer STA Compact from citrate plasma were performed. The correlation of INR between capillary blood and plasma samples is high ($r = 0.95$) and the fit between methods is very good (MRD 8%) (Г9).

For the first time in Bulgaria a comparison of the anticoagulant effect of acenocoumarol (Unipharm) and Sintrom (Novartis) in 53 outpatients was made. A total of 512 PT tests (INR) were performed, of which 295 PTs (INRs) in 25 patients receiving Sintrom and 217 PT tests (INR) in 40 patients receiving Acenocoumarol. The average individual values of the doses of Sitrom and Acenocoumarol, the dependence of the doses on the patient's weight, the patient's age, and the intra-individual variation in the anticoagulant response were compared. The doses of Acenocoumarol and Sintrom showed a similar profile. The achieved rate of INR in the therapeutic area is 52% for Acenocoumarol (Г8).

Dr. Dineva has significant laboratory and methodological contributions. She has self-administered clinically-chemical and immuno-turbidimetric validations of the following analyzers: AU 400, AU 480, Access-2 - Beckman Coulter; Adapted programs for coagulation chromogenic and chronometric tests of the following hemostasis analyzers: Sysmex CS-2000i, Sysmex CS-2500 - Seimens, ACL TOP 500 - IL, STA Compact - Diagnostica Stago, Diatron - Diagon; Comparison of the results of individual tests for PT, aPTT, Fib with reagents and devices from different manufacturers to achieve optimal sensitivity of the results in patients treated with oral anticoagulants and heparin; Introduction of tests for evaluation of perioperative bleeding on MULTIPLE and ROTEM.

It may be summarized that Dr. Dineva is a specialist in Laboratory Haemostasis. She is one of the leading specialists in this field of Clinical Laboratory. Dr. Dineva's scientific and publication activities have been closely linked to laboratory and clinical practice for 28 years at the National Medical University Hospital Sofia.

Educational activity

Dr. Dineva's teaching activities are related to the training of medical specialists specializing in the medical specialty "Clinical Laboratory" and the training of students from the 4th year of medicine in internal diseases program – module of cardiology. For the period from 2014 to 2019, it has a total of 1038 hours of theoretical-teaching engagement.

The calculations from Appendix 2 of the Rules on the Terms and Conditions for Acquisition of Academic Degrees and Occupation of Academic Positions at the of the MHAT

“National Heart Hospital Sofia”, related to the minimum national requirements for occupation of the academic position “Associate Professor”, show that Dr. Dineva meets requirements across all metric groups. Dr. Dineva's total score is 577.3, with a minimum score of 400.

4. Diagnostic and therapeutic activity.

Dr. Dineva is the Head of the Clinical Laboratory at the Laboratory Diagnostics Department of the MHAT “National Heart Hospital Sofia”. She is responsible for the organization of more than 30 specialist staff and the implementation of over 500 000 tests annually. The laboratory is introducing and testing every new device related to hemostasis and aggregation. As an experienced specialist in the medical specialty "clinical laboratory", she performs consulting work in the diagnosis and evaluation of therapy for patients from all clinics of the National Heart Hospital Sofia, as well as consulting assistance in other country hospitals.

Short-term specializations and qualification courses.

Dr. Dineva has undergone the following training, after the basic medical specialty in "clinical laboratory", which are related to professional and scientific development in the years:

1. STA Compact Application Course - Diagnostica Stago - 20-27.03.1999 -Paris, France
2. Internal Auditor Course in accordance with ISO 9001: 2001 - 17 - 19.04.2006 - Sofia
3. ACCESS 2 Application Course - Beckman Coulter - 26.02 - 02.03.2007- Nyon, Switzerland
4. ACL Elite Application Course - Instrumentation Laboratory - 18 - 21.02.2008 - Milan, Italy
5. 5th International Meeting - Platelets 2008 - 15 - 18.10.2008 - Woods Hall - USA
6. Application course DxC 600/800 - Beckman Coulter - 02 - 07.11.2008 - Nyon, Switzerland
7. ACL TOP Family Application Course - Instrumentation Laboratory - 16 - 19.03.2009 - Milan, Italy
8. DxI Application Course - Beckman Coulter - 02 - 07.04.2010 - Nyon, Switzerland
- 9th International Meeting on Venous Thromboembolism - 06 - 08.05.2010 - Milan, Italy
10. Hemostasis Q Application Course - Grifols - 05-09.02.2011 - Barcelona, Spain
11. Application course AU 480/680 - 14-23.09.2011 - Munich, Germany
12. DxH Application Course - 03-08.03.2018 - Munich, Germany.

Dr. Dineva is fluent in French, English and Russian. She has sufficient computer skills.

As an eminent scientist and distinguished colleague in the field of Clinical Laboratory, Dr. Dineva is a member of the Bulgarian Society for Clinical Laboratory (BDCL), and she is also a member of AACC, ISTH and a member of Bulgarian Doctors Association.

Dr. Dineva holds the following awards:

- Second Prize "Best of the Journal of Bulgarian Cardiology" with the support of an educational grant from Pfizer article: Dineva D., Paskaleva I., Gotcheva N., Baicheva V., Georgiev B. Platelet response in patients with implanted coronary stents on treatment with clopidogrel and aspirin assessed by impedance aggregation on a Multiplate. 2011, Bulgarian Cardiology; volume XVII, №2; pp. 16-25.

- Poster Award II: Paskaleva I., Dineva D., Stefanova I., Gocheva N., Baicheva V., Georgiev B. Residual platelet activity measured by impedance aggregometry in patients after coronary stenting on P2Y12 antagonists and aspirin therapy , VI National Conference on Clinical Laboratory with International Participation.

6. Personal impressions: I have known the candidate from the beginning of my work in the clinical laboratory of the "National Heart Hospital" - Sofia. Dr. Dineva stands out for her intelligence, organization, professionalism, self-righteousness and dedication to teaching. She is consistent and thorough in building her academic career; wonderful, responsive and honest partner in the work.

Conclusion:

Dr. Dineva is an established professional, scientist and lecturer with proficiency in laboratory haemostasis. Her scientific production, teaching skills and teaching load, as well as her professional path, meet the requirements of the Rules on the conditions and procedure for acquiring academic degrees and occupying academic positions at the "National Heart Hospital" - Sofia, and show quantitative and qualitative sufficiency for employment of the academic position of Associate Professor.

I strongly recommend the Scientific Jury to vote as anonymously convinced with "YES" for the occupation by Dr. Dobrinka Dineva Savova, MD, Senior Assistant at the Laboratory Diagnostics Department of the Clinical Hospital "Sofia" and Head of the Clinical Laboratory, at the academic position " Associate Professor" at the professional specialty "Clinical laboratory" in the "National Heart Hospital " - Sofia.

22/11/2019
Sofia

Signature: