

## REVIEW

### **In a competition for the academic position "Professor"**

**In the field of higher education 4. "Natural sciences, mathematics and informatics", professional field 4.3. "Biological Sciences" and scientific specialty "Biochemistry", announced in SG no. 63/30. 07. 2021 (correction in SG No. 65 of 06.08.2021)**

**One candidate has submitted documents for participation in the competition - Dr. Albena Georgieva Yordanova, Associate Professor in Biochemistry at the Department of "Chemistry and Biochemistry, Physiology and Pathophysiology" of the Medical Faculty of Sofia University "St. Kliment Ohridski"**

**Reviewer: Professor Dr. Antoaneta Vidolova Popova at the Institute of Biophysics and Biomedical Engineering, Bulgarian Academy of Sciences.**

Albena Yordanova graduated the National High School of Natural Sciences and Mathematics "Acad. Lyubomir Chakalov" in the city of Sofia in 1986, and in 1992 she received a Master degree in "Biochemistry and Microbiology" from the Faculty of Biology of the Sofia University "St. Kl. Ohridski". From 1995 to 1997 Albena Yordanova occupied a position of biologist-specialist in the Department of Biochemistry at the Faculty of Biology at Sofia University (SU), and from 1997 to 2002 - a biologist-specialist at the Institute of Biophysics of the Bulgarian Academy of Sciences (BAS). From 2002 to 2012 she was a senior assistant at the Institute of Biophysics and Biomedical Engineering, BAS and a part-time lecturer in "Biochemistry" at the Department of Biochemistry at the Faculty of Biology at Sofia University and of Bachelor and Master students from the Faculty of Physics at SU.

In 2007 Albena Yordanova received the educational and scientific degree "Doctor" after defending a dissertation on "Surface properties and behavior of lipid liquid-crystalline phases" with supervisors Acad. Boris Tenchov (Institute of Biophysics, BAS) and Assoc. Prof. Zdravko Lalchev (SU). Since 2012 she has been an Associate Professor and Head of the Department of "Chemistry and Biochemistry, Physiology and Pathophysiology" at the Medical Faculty of SU. In addition to being the Head of the Department, Assoc. Prof. Dr. Albena Yordanova is the holder of the mandatory course "Biochemistry" for students in specialty "Medicine", "Medicine in English" and

"Nurse", of the mandatory course "Clinical Laboratory, Biochemistry and Immunology" for students in the specialty "Nurse" as well as of the elective courses "Peculiarities of normal metabolism and pathology" and "Phytoproducts and their application in medicine" for students in specialty "Medicine" and "Medicine in English" at the Medical Faculty of Sofia University.

Assoc. Prof. Yordanova was a reviewer and author of Stand Points in 10 Scientific Juries for the educational and scientific degree "Doctor" for the Medical University of Sofia and Plovdiv, SU "St. Kl. Ohridski", different institutes of BAS and National Sport Academy (NSA), member of Scientific Juries for selection of chief assistant for SU (5) and for other universities and BAS (9), as well as in competitions for "Associate Professor" (BAS) and Diploma thesis reviewer. For the period 2016-2018, Assoc. Prof. Yordanova was an academic mentor in the practical training of students in a real work environment "Student Internships". She has participated in 28 scientific and educational projects funded by the Ministry of Education and Science, Sofia University and the Medical University in Sofia, and has been the leader of one project funded by Bulgarian National Fund and of 10 projects funded by SU. Assoc. Prof. Yordanova has participated in 158 International and National Scientific Forums through oral and poster presentations. Since 2005 Dr. Yordanova has been a member of the Union of Scientists in Bulgaria, and since 2019 - a member of the Association of Biochemists in Bulgaria. Since 2013 she has been an evaluator in the competitions for funding of projects for fundamental research of young scientists and post-doctoral students for the National Science Fund. In the period 2012-2017 Dr. Yordanova was a research supervisor of a part-time doctoral student, specialty Molecular Biology.

In addition to her scientific and teaching activities at the Medical Faculty of SU, Dr. Yordanova is a member of the National Commission for Organizing and Conducting the National Olympiad in Biology and Health Education since 2008 and since 2011 is the Chairman of the National Commission. Since 2010 Dr. Yordanova is the Head of the National Biology Team, a member of the International Jury of the International Biology Olympiad and a member of the Board of the Association of Olympic Teams in Natural Sciences (since 2010). The assets of the National Biology Team include 3 gold, 7 silver, 24 bronze medals and 8 diplomas for worthy performance. Dr. Yordanova is the initiator and organizer of Winter and Summer Schools of Biology (2014-2019). She is the organizer of the First and Second National Olympic Conferences on Natural Sciences and Innovations (2014 and 2016). For the period 2015 - 2019 Assoc. Prof. Yordanova is a member of the National Commission for organizing and conducting the National Competition "Young Talents" by the Ministry of Education and Science. Since 2021, she has been

a member of the National Commission for conducting and evaluating the participants in the “Science on Stage” festival.

Since 2013, Assoc. Prof. Yordanova is a member of the National Commission for Assessment of the State Matriculation Exam in Biology, and for 2017 and 2018 - is a Super Assessor in the National Commission. Since 2018 she has been a member of the commission for verification and evaluation of written works of students from the module “Talented Biologist” for admission to National High School of Natural Sciences and Mathematics. Since 2015 Dr. Yordanova has been a member of the Commission for examination and evaluation of the candidate-student works in biology for the specialty "Medicine" for the Medical Faculty of SU, and since 2021 - the Chairman of the Commission. Since 2018 she has also been the Chairman of the Commission for Verification and Evaluation of Candidate Student Works in Chemistry for the Faculty of Medicine. Since 2020 Dr. Yordanova has been the Deputy Chairman of the Examination Commission for conducting a competitive exam (test) in Biology and Chemistry in English for foreign students for ranking in the specialty "Medicine in English". For the period 2006-2009 she has been the author of 48 popular science articles in the magazine for innovative technologies HiComm.

In the research carrier Assoc. Prof. Dr. Yordanova has published 95 papers, 81 of which are in scientific journals. 32 publications are in journals with IF, and 38 in scientific journals which are ranked with quartiles (Q1 - 8 publications, Q2 - 12, Q3 - 7 and Q4 - 11). Dr. Yordanova is the author of a monograph entitled: "Alveolar surfactant - normal metabolism and pathology. *In vitro* methods for assessment of its functional state” and is a co-author of 13 collections and manuals for pupils and students.

The scientific papers of Dr. Yordanova have been noticed and cited at least 266 times, which is an indication of the relevance and high quality of the published articles. The H factor of Assoc. Prof. Yordanova according to SCOPUS after excluding of auto-citations is 7.

In the current competition for "Professor" in the field of higher education 4. "Natural Sciences, Mathematics and Informatics", professional field 4.3. “Biological Sciences” and specialty “Biochemistry”, Assoc. Prof. Dr. Albena Yordanova participates with 41 publications, including one monograph, 12 articles in journals with IF, 12 collections and manuals for pupils and students and 16 publications in journals without IF and quartiles, mainly in the materials of scientific forums.

The publications in scientific journals with IF and quartiles are grouped as follows (Q1-3 papers, Q2-4, Q3-3 and Q4-2). Dr. Yordanova is the first author in one and second in 5 articles. In

the materials from scientific forums Dr. Yordanova is the first author in 4, and last in 4 materials, 9 being in Bulgarian and 7 in English. In the manuals, 2 of which are in English, Dr. Yordanova is the first author in 7 and the last in 4.

A completed reference for the implementation of the Minimum National Requirements under Art. 2b of the Law for Development of the Academic Staff of the Republic of Bulgaria (ZRASRB) for scientific field 4. "Natural sciences, mathematics and informatics", professional field 4.3. "Biological Sciences", scientific specialty "Biochemistry" for the academic position "Professor". By the group of indicators A and B Dr. Yordanova presents the required 50 and 100 points, respectively. By group of indicators D and E the presented points exceed the Minimum National Requirements as follows: by indicator D - 224 points at required 200 and by indicator D - 404 points at required 100. By indicator E are presented 184.67 points at required 150.

An **academic report** is presented, which summarizes the main scientific, scientific-applied and methodological contributions of Assoc. Prof. Dr. Albena Yordanova, grouped in 5 sections. The contributions in the second section - "II. Contributions of scientific papers under indicator D 7: Scientific publications in journals that are referenced and indexed in a world-famous database" are discussed as contributions to fundamental research and as such with potential for practical application.

### **I. Contributions of the scientific works according to indicator B 3.: Habilitation work - Monograph**

The monographic work presents detailed information about the composition, structure, properties and molecular mechanisms of interaction of the components of the alveolar surfactant for the implementation of physiological processes in the lung in health and pathology. The mechanisms of development, symptoms, classical and modern therapeutic approaches in the treatment of various lung diseases, which occur as a result of lack of "mature" surfactant or its inactivation, under the influence of various factors are discussed in details. New modern biochemical and biophysical methods for diagnosis of the condition and activity of the alveolar surfactant are described which supplement and enrich the classical diagnostic methods used so far. The described *in vitro* methods are convenient, fast and highly informative and can be applied to a variety of clinical trials - tracheal and gastric aspirates, amniotic and lavage fluids. The alternative possibility for wider use of gastric aspirates as reliable, non-invasive and highly informative clinical trials for diagnosis, which will help neonatologists for rapid diagnosis and therapeutic behavior has been shown. The monograph is a systematic practical guide for modern laboratory

methods for diagnosis the effectiveness of alveolar surfactant and would find application as a guide to biochemistry, neonatology and pediatrics and would be useful for the training of medical students and in the clinical practice of neonatologists, physiologists and pulmonologists.

## **II. Contributions of scientific papers under indicator D 7: Scientific publications in journals that are referenced and indexed in a world-famous database**

### **I. Contributions to fundamental science**

#### **I.A. Biochemical and biophysical studies of clinical samples from patients with different diseases to assess the functional status of the lungs**

- Based on a comparative analysis of protein and lipid concentrations in gastric aspirates (GA) immediately after the birth from preterm infants with Neonatal Respiratory Distress Syndrome (NRDS) and healthy term infants, has been shown that phospholipid and protein concentrations in GA from premature infants are lower compared to samples from full-term infants. The dynamic surface characteristics of GA show significantly higher mean values of the minimum surface tension ( $\gamma_{\min}$ ) in premature infants than in full-term infants. The values of parameter  $\gamma_{\min}$  have been shown to be important for assessing the maturity of the surfactant in the lung.
- For the first time the combination of modern innovative techniques Axisymmetric Analysis of the Shape of a Hanging Drop and Brewster-angle microscopy (BAM) was used for fast and reliable analysis of gastric aspirates from healthy full-term and premature infants to quickly assess the functional status of alveolar surfactant and pulmonary maturity.
- Gastric aspirates from preterm infants with primary surfactant deficiency and NRDS, a control group of healthy neonates and at-risk neonates after an *in vitro* procedure, and maternal corticosteroid therapy were studied. The maturity of the alveolar surfactant in high-risk infants was tested by the combined application of the methods of thin liquid films and Brewster-angle microscopy and it was shown that the probability of formation of a thin black film and the morphology of monolayers from gastric aspirates differ significantly between the tested groups of children, which is important for undertaking the correct strategy in the neonatology units.
- Using polyacrylamide gel electrophoresis and Western blot analysis, different isoforms of the specific surfactant proteins SP-A, SP-B and SP-C were detected for the first time in gastric aspirates depending on the stage of maturation of the alveolar surfactant.
- By studying the surface properties of gastric aspirates, it was found that corticosteroid therapy with betamethasone, routinely used in high-risk pregnant women, after *in vitro*

fertilization and multiple pregnancies, improves the surface characteristics of aspirates and leads to visible changes in the morphology of their formed monolayer films. The results of the biophysical analysis confirm that the administration of betamethasone activates the biosynthesis of the components of the alveolar surfactant.

- Laboratory analysis of lavage fluids in the treatment of pulmonary alveolar proteinosis to eliminate intraalveolar accumulation of lipoproteins showed that at each subsequent stage of the procedure the concentrations of proteins and phospholipids decrease and the values of equilibrium surface tension increase, which increases the efficiency of treatment.

- In the study of the biochemical and biophysical characteristics of the alveolar surfactant in the non-ventilated (operated) and ventilated lobe of the lung in patients with non-small cell lung cancer, it was found that in patients with lung cancer changes in the composition and properties of the alveolus hypoxia and inhalation anesthesia alter its biochemical and biophysical characteristics.

### **I.B. Investigation of intermolecular interactions in model biological membranes through modern highly informative laboratory methods**

- The presence of hydrophilic polymers has been found to prevent the inactivation of alveolar surfactant by plasma proteins via creating an attractive osmotic pressure in the surface film, leading to the displacement of unwanted inhibitors from the surface and improving the quality and effectiveness of the surfactant.

- The interaction between synthetic methionine-enkephalin and its amidated analogue with the characteristic for membrane rafts POPC, sphingomyelin and cholesterol, as well as with their mixtures was confirmed by applying a combination of two methods, the Langmuir monolayer technique and the method of Wilhelm registration of surface pressure. Brewster-angle microscopy showed an increase in the surface density of the formed mixed films, especially in two- and three-component lipid mixtures, the effect being more pronounced in enkephalinamide, which suggests differences in the folding of the two enkephalins. Similar differences have been found in the interaction of synthetic leucine-enkephalin and leucine-enkephalinamide with the lipid phase formed by POPC, sphingomyelin and cholesterol.

- Newly synthesized benzantrone (quaternary ammonium benzantrone) has been found to penetrate monolayers composed of phospholipids (cholines, glycerols and ethanol amines) that resemble bacterial membranes. The strongest interaction was found with phosphatidylethanolamines and negatively charged phosphatidylglycerols. It has been suggested that electrostatic interactions with

polar lipid heads, the formation of hydrogen bonds between the amino group of ethanolamine and keto groups in the structure of benzoquinone, as well as the occurrence of hydrophobic interactions with the group of ethanolamine are important for the antimicrobial action of quaternary ammonium benzantrone.

- It has been confirmed that Langmuir monolayers are an elegant, convenient and informative model system for studying the molecular interactions and properties of biological membranes.

### **III. Contributions with potential for practical application**

#### **II.A. Biochemical and biophysical studies of clinical samples from patients to assess the functional status of the lungs in various diseases**

This section describes the practical applications of the results described in the articles included in indicator D 7. The possibility of using gastric aspirates as a rapid, non-invasive and informative clinical trial for determining the surfactant maturity of the lung in premature infants is indicated. It has been shown that the minimum value of surface tension, determined by Axially Symmetrical Analysis (ADSA) of the Shape of a Hanging Drop is a reliable parameter by which it is possible to quickly and unequivocally to determine the lung maturity of newborns. The effectiveness and potential of this method for rapid diagnosis of lung function using a minimal amount of pulmonary lavage has been demonstrated. A rapid, convenient, and reliable approach to assessing lung function in patients with respiratory disease is offered through the combined application of ADSA, Brewster-angle microscopy, and the thin liquid film method. The applied procedure for conducting complete pulmonary lavage has been improved, with the main contribution being the reduction of the volume of the used physiological solution and the acceleration of the process. This technique is actually already applied in clinical practice.

#### **II.B. Investigation of intermolecular interactions in model biological membranes through modern highly informative laboratory methods**

The results obtained in the study of effects of hydrophilic polymers on the restoration of the surface properties of albumin-inhibited exogenous surfactants may find application in clinical practice in the treatment of Acute Respiratory Distress Syndrome in adults, which may occur as a result of albumin inhibitory action in the lungs. The combined application of the Langmuir monolayer model with Brewster-angle microscopy can be used successfully in preclinical studies to test the biological activity and membrane penetration of newly developed analogs of natural analgesics. Establishing specific interactions between newly synthesized benzantrone and bacterial membrane-like model membranes and suggesting that these interactions underlie the antibacterial properties of benzantrone

would be a good starting point for investigating the potential antimicrobial activity of newly synthesized antibacterial agents.

#### **IV. Contributions under indicator E 19: Publication of a university textbook or textbook used in the school network**

The published textbooks, in Bulgarian and English, are prepared in accordance with the approved training program in Biochemistry for students at the Faculty of Biology and Medicine of SU "St. Kl. Ohridski" and contain tasks for preliminary training of students, a set of practical exercises and practical tasks to facilitate students in learning the material and applying basic methods used in biochemical and clinical practice.

The published collections of theoretical tests and practical tasks from the Regional and National rounds of the Olympiad in Biology and Health Education are aimed mainly at the formation of basic concepts from various branches of biological science, but also provide new, unknown to the student information and are especially useful for preparing the teams for National and International Olympiads.

A textbook has been published, containing richly illustrated theoretical material on human anatomy and physiology and various tests for candidate-student exams in biology for the Medical Universities in Bulgaria. The test questions and theoretical explanations are in accordance with the curriculum in biology and health education for 8<sup>th</sup> grade, approved by the Ministry of Education and Science.

#### **V. Other scientific contributions**

This section systematizes the contributions of Assoc. Prof. Dr. Yordanova in the papers published in the materials of scientific forums, both in terms of contributions of a fundamental science, as well as in terms of surveys and analysis of every day work related to the National Olympics.

The scientific problems that are in the focus of Assoc. Prof. Dr. Yordanova's research are extremely important not only from the point of view of fundamental science, but also provides an opportunity for application in the education of medical students, and has the potential for application in the diagnostic and therapeutic work of neonatologists, therapists and pulmonologists. Some of the results are new to science and already used in medical practice.



**Summary:**

**The submitted documents and references by Assoc. Prof. Dr. Albena Yordanova for participation in the current competition for the academic position "Professor" clearly demonstrate that Assoc. Prof. Dr. Yordanova is an excellent researcher and lecturer in the field of Biochemistry, which performs perfectly in scientific, teaching and organizational work and that significantly exceeds the Minimum National Requirements set out in the Law for the Development of Academic Staff in the Republic of Bulgaria for the position of "Professor" of "Biochemistry" at the Faculty of Medicine of Sofia University.**

**Based on everything listed above I strongly recommend to the members of the Scientific Jury to award the academic position "Professor" in the field of higher education 4. "Natural sciences, mathematics and informatics", professional field 4.3. "Biological Sciences" and scientific specialty "Biochemistry", for the needs of the Medical Faculty of Sofia University "St. Kl. Ohriski", to Associate Professor Dr. Albena Georgieva Yordanova.**

22. 11. 2021

Sofia

Signature:

/Prof. Antoaneta Popova/