

OPINION

SUBJECT: competition for the academic position "Professor" in the field of higher education 4. Natural sciences, mathematics and informatics; professional field 4.3. Biological sciences (scientific specialty 01.06.23 "Immunology")

BY Prof. Nina Dimitrova Ivanovska, the Stephan Angeloff Institute of Microbiology, BAS, member of the scientific jury according to the Order of the Director of ImikB Б № I-82/01.07.2020

LEGALITY OF THE ANNOUNCED COMPETITION

The competition was announced in the State Gazette, issue 47 / 22.05. 2020, as well as on the website of the Institute of Microbiology, BAS (IMiKB) for one place in the scientific specialty "Immunology". During the two-month period from the date of announcing the competition in the State Gazette, documents were submitted by one candidate - Associate Professor Andrey Ivanov Chorbanov. The candidate has been admitted to participate in the competition by the Commission, determined by order of the Director of IMiKB. After reviewing the submitted set of documents, the Commission has confirmed the regularity and availability of all required documents and the procedure for selection of Associate Professor Andrey Chorbanov for the academic position of "Professor" can be initiated. Based on these data, I believe that the competition meets the requirements for holding the academic position "PROFESSOR" under the Law on the Development of Academic Staff in the Republic of Bulgaria, the Regulations thereto, as well as the Regulations on the terms and conditions for obtaining degrees and holding academic positions at ImikB, BAS.

BIOGRAPHICAL INFORMATION ABOUT THE APPLICANT

Assoc. Prof. Andrey Chorbanov graduated from Sofia University "St. Kliment Ohridski", in the field of Genetic and Cell Engineering. From 1992 to 1998 he worked at NCIPD, Section "Applied Immunology and Biotechnology", and from 1999 at the Institute of Microbiology BAS, since 2009 as an associate professor. He has been a visiting researcher at the University of Utrecht, Netherlands National University of Taiwan, Taipei, University of Nis, Serbia. He is a Chairman of the Immunology Section of the Union of Scientists in Bulgaria, the European Federation of Immunological Associations (EFIS) and the European Association for the Study of Diabetes (EASD). He is a winner of several awards for scientific activity, including twice winner of the Pythagorean Prize.

CHARACTERISTICS OF RESEARCH AND APPLIED ACTIVITIES

The presented publications are in the field of fundamental theoretical and applied problems, which can be thematically belong to three directions: (1) development of fundamentally new types of engineered protein and DNA vaccines and adjuvants, (2) application of newly synthesized cyan dyes for studying molecular and cellular mechanisms in model systems, (3)

selective suppression of the pathological immune response through the administration of chimeric molecules in a variety of murine and human models of autoimmune diseases.

1. Engineering protein and DNA vaccines and adjuvants

Antiviral DNA vaccines in which a vector encoding a viral antigen is bound to a specific antibody fragment is an innovative approach to induce protective immune responses. A new genetic vaccine has been developed that is a chimeric molecule encoding haemagglutinin from influenza A virus, comprising a T- and B-cell epitope linked to a sequence of a 7G6 antibody specific for complement co-stimulating B-cell receptors 1 and 2. Intramuscular injection of DNA construct results in *in vivo* synthesis of the immunogenic chimeric protein, which cross-links surface cell receptors located on influenza virus-specific B cells. The modulation of the immune response through the application of engineered chimeric molecules was studied in a humanized system in the NOD SCID Gamma transfer model. The resulting chimeric molecules are characterized by small size and molecular homogeneity and can be used to construct other similar recombinant vaccines.

Despite their high efficacy and lack of side effects, the new vaccines have the disadvantage of showing low immunogenicity. To enhance the immune response, highly immunogenic non-specific immunomodulation molecules are added, such as adjuvants or protein carriers covalently bound to a specific antigen to elicit active specific immunostimulation. The adjuvant properties of hemocyanins isolated from the marine gastropods *Rapana thomasiana* and *Megathura crenulata* are presented. Immunization of mice with influenza vaccine or tetanus toxoid in combination with hemocyanins elicits a relatively prolonged anti-influenza cytotoxic response as well as a humoral response to viral proteins. The results obtained show that they are suitable as potential bio-adjuvants and can be used for peptide transport. Highly significant are the results proving the potential anti-tumor effect of hemocyanins in a mouse model of colon cancer, suggesting their use in the treatment of various types of cancer.

2. Application of newly synthesized cyan dyes in model systems

Fluorescence techniques, including fluorescence microscopy, are one of the most modern and promising areas in biomedical research. Some of the presented results are related to studies in this direction. The potential possibilities for the application of three newly synthesized asymmetric dication monomethine cyanine dyes, chlorine-containing analogues of Thiazole Orange, as fluorescent markers for nucleic acid binding, have been investigated. They were found to be particularly useful in assessing apoptosis processes. Experiments using murine embryonic fibroblast cells have shown that the dyes do not penetrate a healthy cytoplasmic membrane and are suitable for evaluation of late apoptosis / necrosis.

3. Application of chimeric molecules in mouse and human models of autoimmune diseases

Systemic lupus erythematosus (SLE) is an autoimmune disease in which B-cell hyperactivity develops. It produces autoantibodies to nucleoprotein antigens, leading to the formation of immune complexes and causing inflammatory processes in a number of organs and tissues. Unfortunately, the applied methods of treatment of the disease is non-specific and is often

accompanied by many unwanted side effects, which determines the need for more specific therapies. Some of the presented results are dedicated to a new approach of clinical significance, which modulates the activity of B lymphocytes. The resulting DNA-like chimeric antibodies cross-link FcγIIB and immunoglobulin receptors to DNA-specific B lymphocytes. This results in inhibition of B and T cell proliferation and, accordingly, limits the production of anti-DNA IgG antibodies. Because experiments in patients with SLE are very limited, the developed model for the transfer of human cells in SCID mice depleted of T- and B-lymphocytes is particularly valuable. The tolerance of these humanized mice to native DNA results from the binding of native DNA-specific B cell receptor and inhibitory B cell receptor CR1 and the generation of a strong inhibitory signal.

Another model of autoimmune disease due to the presence of autoreactive T and B cells as well as autoantibodies against self-antigens is type 1 diabetes mellitus. The constructed chimeric molecules selectively modulate the activity of specific B cells by the GAD65 molecule (-cells). Selective binding of anti-GAD65-specific B cells and the immunoglobulin receptor CD35 generates an inhibitory antibody signal.

MATERIALS SUBMITTED FOR REVIEW AND COMPLIANCE WITH THE REQUIREMENTS

The scientific works of Assoc. Prof. Andrey Chorbanov can be distributed in accordance with the criteria for the minimum national requirements of ZRASRB and the Regulations to it, as well as the Regulations of IMIKB, BAS, as follows:

1. Criteria "A" - presented an abstract of a doctoral dissertation - 2002 on "Engineering and genetically engineered chimeric molecules as antigens" (50 points)
2. Criterion "B" - 5 articles are presented (equivalent to a monographic work), which do not repeat the ones presented for acquiring the educational and scientific degree "Doctor" and for holding the academic position "Associate Professor" (125 points). All of them are in specialized journals with impact factor, referenced and indexed in world databases and fall into category Q1.
3. Criterion "D" includes 11 publications in journals that are referenced and indexed in world-famous databases of scientific information (Web of Science and Scopus), which are in categories Q1 and Q2 (250 points).
4. Criterion "E" includes 466 citations in scientific journals, monographs, collective volumes and patents, referenced and indexed in world-famous databases of scientific information (Web of Science and Scopus) (922 points).
5. Criterion "E" includes 4 defended doctoral students (175 points), management of 5 international projects (250 points), as the attracted funds are 103 000 euros (40. 37 points), or a total of 465.37 points. A. Chorbanov has participated in 33 completed projects and is the leader or co-leader of 13 ongoing projects, and has more than 1050 teaching hours in universities and research institutes since 2012.

According to the additional requirements of IMikB, 33 articles with impact factor after associate professor are presented, in 23 of them A. Chorbanov is a leading author (required 20 with impact factor, 16 of them as a leader). A list of 117 participations with reports and posters at international and national forums (which are not included in the total number of papers) is also attached.

CRITICAL NOTES AND RECOMMENDATIONS

I have no critical remarks to the materials presented by Assoc. Prof. A. Chorbanov. They correspond to the theme of the competition, both as a number and quality. In addition, the documentation is presented precisely and allows to get a complete picture of all areas of the applicant's activity.

CONCLUSION

The documents and materials submitted by Assoc. Prof. Andrey Chorbanov, PhD meet all the requirements of ZRASRB, the Regulations for implementation of ZRASRB and the relevant Rules of IMikB. Assoc. Prof. Andrey Chorbanov is an eminent scientist in the field of experimental and applied immunology, has the ability to focus on current problems and to present innovative solutions for specific therapy of severe, socially significant diseases such as autoimmune, allergies and tumors. From the analysis it is clear that Assoc. Prof. A. Chorbanov participates in the competition with scientific production, which by scientometric indicators significantly exceeds the requirements for the academic position "Professor": publications in journals with high IF (almost all in category Q1 and Q2) and H factor 12, citations in renowned international journals, presentation of the results at a huge number of international and national forums. To these must be added the educational activity: defended PhD students and teaching. A. Chorbanov not only generates a lot of ideas, but very successfully ensures their implementation through the management of an impressive number of projects, including international ones and attracting significant funding.

Based on everything noted so far, I strongly recommend to the members of the esteemed scientific jury, formed by order № I-82/01.07.2020 of the Director of IMikB to propose to the Scientific Council to award Assoc. Prof. ANDREY IVANOV CHORBANOV the academic position "professor" in professional field 4. Natural sciences, mathematics and informatics, professional field 4.3 Biological sciences (Immunology).

26.08.2020

Prof. Nina Ivanovska, DSc