REVIEW

Regarding the dissertation work of
Vancho Donev, biologist from BulBio-NCZPB
entitled "Characterization of the potential immunoprophylactic and immunotherapeutic properties of human blood plasma" for a PhD degree
Higher education field: **4. Natural sciences, mathematics and computer science,**
Professional field: **4.3 Biological Sciences,**
Scientific specialty: **Immunology**

By Academician Dr. Bogdan Petrunov
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The dissertation presented by Vancho Donev concerns an undoubtedly very interesting issue, both from a theoretical and especially from a practical point of view, related to the great possibilities of human plasma and the immunoglobulins contained in it for the treatment, prevention and mechanism of their action in some bacterial and viral infections, parasitic invasions and autoimmune diseases. The relevance and importance of the studies conducted by Donev, under the expert guidance of Assoc. Prof. Georgi Nikolov, stand out even more clearly, considering that they are closely related to the intravenous immunoglobulin produced for many years by BulBio-NCZPB, a widely used commercial preparation, which is a source of pride for our country. I emphasize this because the in-depth characterization of this vital blood product, with the discovery and expansion of its possibilities for use for the purposes of immunotherapy and immunoprophylaxis, which led to the saving of hundreds of human lives, are of great importance for modern medicine.

The author has prepared a very thorough, informative review, based on 310 literary sources, well selected and showing his good knowledge of the problem under study. The historical data related to blood transfusion, the role of individual blood components in clinical medicine, the methods of their fractionation and purification, the mechanism of their action, the results of their application not only as replacement therapy for certain immunodeficiencies, but also as a very important immunomodulator are examined in detail. A special place is occupied by the data on the possibilities of human convalescent plasma and especially immunoglobulins

to be used in treatment during the Covid 19 pandemic and the results obtained in this regard. Very rightly, at the end of the literary review, Donev summarizes the unresolved or still insufficiently clarified questions related to the topic of his dissertation work, from which the goals and objectives of the study naturally arise. They are formulated clearly and precisely, which allows for the qualitative monitoring of their implementation. In this regard, it should be said that Donev's efforts are directed in the following directions: characterization of the contained antibodies and their isotypes; of the specific anti-Covid-19 antibodies; the cytokine profile of the plasma; of the specific anti-Candida albicans and anti-S. aureus antibodies and the presence of some autoantibodies in it. This is a fairly broad research program that required a lot of effort and which I must say was successfully implemented.

For the purposes of the study, a modern methodology was used, including various variants of ELISA and Immunoblot based on established commercial kits and high-quality equipment. The results obtained were processed very correctly with modern software programs, which guarantees their reliability.

I could summarize the main contributions of the material presented for review as follows:

For the first time, a study is being conducted on the possibilities of convalescent plasma from Covid-19 patients during the pandemic and the data show that the antibody titer is not important in the therapy of this infection, i.e. for the purposes of passive immunotherapy.

- It is very important to develop a model for the application of IVIG and its great therapeutic potential in various infections and autoimmune diseases, which is superior to that of convalescent plasma.
- The data on the classification of antibodies in IVIG are original, not only by their quantity, but also by their quality of binding/avidity to the various antigens/pathogens.
- An important theoretical and practical contribution is the information about the passive transfer of antibodies through IVIG and the risks associated with it in patients receiving it as regular therapy.
- An important theoretical and practical contribution is also the characterization of Immunovenin-intact in terms of the classes and subclasses of immunoglobulins it contains to various bacterial pathogens. Of particular interest in this regard are the data on the presence of specific antibodies to K. Albicans and Staphylococcus aureus.
- The data from determining the cytokine profile of human plasma used to produce Immunovenin intact with a wide range of labeled antibodies, which is being done for the

first time in Bulgaria, deserve high praise. The results show their low values and the lack of the possibility of unlocking various pathological immune conditions.

- An ELISA method for the determination of IgG antibodies to the hemolysin toxin of Staphylococcus aureus has been successfully developed and validated.

During the preliminary discussion of the dissertation and in my initial opinion, a number of remarks and suggestions were made to improve the dissertation. I must state that the author has taken them into account and made the necessary corrections.

The entire procedure for completing the dissertation has been completed in accordance with the law - the doctoral student's curriculum has been successfully implemented and the relevant exams have been taken, the discussion of the dissertation by the Collegium of the primary scientific unit - the Department of Immunology and Allergology... gave a positive assessment.

In connection with the dissertation work being developed, Donev has published 2 scientific papers, one of which in a journal with an impact factor, and participated in 2 international and 4 national scientific forums, thereby making his research available to the medical community.

In conclusion, I would like to state that based on the results obtained, their competent discussion, the correct and well-motivated conclusions, the work of Vancho Donev submitted for review is indeed a very well-planned and implemented scientific project with proven theoretical and especially practical contributions, which meet the requirements for a good dissertation work, which deserves a high rating. Considering all this, I recommend that the esteemed jury positively evaluate Vancho Donev's dissertation work and vote "YES", which I fully support, and award him the scientific degree " Philosophy Doctor ".

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