

OPINION

ΦΒΜ 577 # 19
10. 07. 20

on materials regarding competition for the academic rank “Professor”, in higher education field 6.0 „Agrarian Sciences and Veterinary Medicine“, professional field 6.4 „Veterinary Medicine“, scientific specialty “Animal Pathology”, discipline “Pathology/Special Pathological Anatomy” announced with decision by the Academic Council of the University of Forestry, promulgated in State Gazette issue 32 of 3 April 2020, procedure code **VM-P-0320-35**.

Candidate for participation in the competition: Assoc. Prof. Vasil Kostadinov Manov, DVM, PhD.

Person prepared the opinion: Professor Ivan Stoyanov Zarkov, DVM, DSc, “Veterinary Microbiology, Infectious and Parasitic Diseases” Department, Veterinary Medicine Faculty, Trakia University – Stara Zagora. Professional field 6.4 „Veterinary Medicine“, retiree, scientific specialties "Epizootology, infectious diseases and prevention of infectious animal diseases" and "Virology".

Member of the academic jury by Order No. ZPS-173 from 11.05.2020 by the Rector of the University of Forestry and decision of the Faculty Council of the Faculty of Veterinary Medicine (Protocol 46/14.01.2020). At the first meeting of the Academic jury (12.05.2020) I was designated to prepare an opinion.

I declare no conflict of interest between me and the candidate in the competition.

1. Short biography of the candidate.

Associate Professor Vasil Kostadinov Manov, DVM, was born on 5 December 1962. He graduated from the Veterinary College in Lovech in 1981 and the Faculty of Veterinary Medicine at Trakia University in 1993 with a qualification "Master of Veterinary Medicine". He is a lecturer at the Faculty of Veterinary Medicine at Trakia University – Stara Zagora and later at the Faculty of Veterinary Medicine (FVM) at University of Forestry – Sofia (UF). He held the positions of Assistant Professor, Senior Assistant Professor and Chief Assistant Professor from 1989 to 2010 and teaches in the disciplines "General Pathological Morphology and Special Pathological Anatomy". In 2009 he obtained the educational and academic degree **PhD** on the topic "Comparative pathomorphological studies in animals infected with Bulgarian isolates of Aujeszky's disease virus". Since 2010 he was acquired the scientific title "**Associate Professor**" and teaches students in "Veterinary Medicine" – lectures, seminars and conducts practical and theoretical exams in the disciplines "General Pathological Morphology" and "Special Pathological Anatomy". Fluent in Russian and English, both written and spoken. He has professional skills for work with histological and microscopic equipment necessary for scientific and diagnostic activities. The applicant is a member of the Faculty Council of the FVM, of the Academic Council of the UF, of the Commission for selection of students under the “Erasmus program” of UF-Sofia, of the Ethics Committee of the Bulgarian Veterinary Union (BVU), of the Bulgarian Association of Small Animal Veterinarians and the Union of Veterinarians in Bulgaria. Participates in Committees at the UF, at the Ministry of Agriculture and the Bulgarian Food Safety Agency, in the establishment and management (from 2012 to 2016) of the University Clinic for Small Animals.

2. Compliance of applicant’s materials and documents to the requirements listed in the Statute for Development of the Academic Staff of the University of Forestry.

All copies of documents and materials related to the application of Associate Professor Manov were presented in a good form. One part of them is associated to the procedure itself,

and another part – to the research activities of the applicant. All of them are compliant to the the Statute for Development of the Academic Staff of the University of Forestry.

The applicant meets the minimum national requirements for the groups of indicators required to hold the academic position "Professor". Out of a total of **550 points** needed, he has **1570.43 points**. He has a dissertation for the award of educational and academic degree PhD; **Habilitation** – monograph: Manov, V. Special Veterinary Pathology, Panev Publishing, Sofia, 2020, ISBN 978-619-90789-4-5; Published monograph, which is not presented as a main habilitation: Manov, V. Morphological characteristics of some neoplasms in animals, 2019, Panev Publishing, Sofia, ISBN 978-619-90789-3-8; Articles published in scientific journals – referenced and indexed in world-famous databases with scientific information (15 articles) and in non-refereed journals with scientific review or published in edited collective volumes (22 articles). He has a doctoral student with educational and academic degree PhD. Associate Professor Manov participated in two national **research projects**. He has published two university textbooks (Manov, V. General Veterinary Pathology. Textbook for veterinary medicine students. Panev Publishing, Sofia, 2018; ISBN 978-619-90789-2-1), (Manov, V. Pathoanatomical characteristics of diseases in domestic animals. Textbook for veterinary medicine students. Panev Publishing, Sofia, 2020; ISBN 978-619-90789-5-2) and **five manuals** (Stoykov, D., I. Nikiforov, S. Stoev, I. Dinev, V. Manov, N. Grozeva, R. Simeonov, R. Todorov, Y. Yordanov. Veterinary Autopsy Technic and Incineration, Tutorial Manual Stara Zagora, 2007, ISBN 978-954-9383-24-9; Stoev, D., I. Dinev, V. Manov, R. Simeonov, N. Grozeva. Veterinary Autopsy Technic and Incineration Tutorial Manua, Stara Zagora, 2016, ISBN 945-9887-24-3; Dinev, I., I. Nikiforov, S. Stoev, V. Manov, N. Grozeva, D. Pavlov, R. Todorov. Veterinary Histopathology. Tutorial Manual, Bogomilovo, 2012, ISBN 978-934-9443-14-1; Dinev, I., S. Stoev, V. Manov, R. Simeonov, N. Grozeva, I. Kalkanov, K. Dimitrov, G. Popov. Veterinary Histopathology. Tutorial Manual, Bogomilovo, 2016, ISBN 978-954-9443-38-3; Dinev, I., S. Stoev, V. Manov, R. Simeonov, N. Grozeva, I. Kalkanov, K. Dimitrov, G. Popov.), which is used in the school network.

3. Evaluation of teaching & learning activities.

Associate Professor Manov has a total pedagogical experience of over 30 years both at Trakia University – Stara Zagora, and at University of Forestry. He gives lectures in courses with teaching in Bulgarian in 2 disciplines – General Pathomorphology – 60 hours, and Special Pathological Anatomy – 120 hours. He conducts practical training in 3 disciplines – General Pathomorphology – 60 hours, Special Pathological Anatomy – 30 hours and Diseases of Bees, fish and game - 6 hours. The total course load of Associate Professor Manov for the academic year 2018-2019 is 276 hours, of which 180 hours of lectures and 96 hours of practical training.

The applicant is the holder of the two academic disciplines – General Pathomorphology and Special Pathological Anatomy. After acquiring the academic title "Associate Professor" to improve his education, the applicant has participated in the development of curricula (2 in Bulgarian and 2 in English), in the creation of three training laboratories and centers. The educational process was helped by publishing 5 textbooks on electronic media in a virtual library and 2 textbooks in paper copy for veterinary medicine students.

The activities outside lectures and practical training include conducting semester exams for students, participation in examination committees of PhD students, in committees for protection of clinical internships and student reports.

As an **expert** and erudite scientist he has carried out consulting activities of organizations, public and private companies (15); has reviewed curricula (3) and manuals (5), articles and reports (15), projects (3). The applicant is in the editorial board of a non-refereed scientific journal, has been in the management of international scientific forums (7 times), has managed a scientific laboratory. Associate Professor Manov has participated in an academic jury for obtaining the educational and academic degree PhD (6 times), academic degree

"Doctor of Science" (1 time), academic titles "Chief Assistant" (2 times), "Associate Professor" (5 times), and "Professor" (2 times).

The whole educational and pedagogical, and expert activity shows that Associate Professor Manov is a perfectly prepared and highly experienced teacher.

4. Evaluation of research, research & development and teaching activities of the candidate.

Associate Professor Manov presents 84 materials. 7 of them are presented for the obtain of main academic degree PhD with included dissertation and abstract, 29 are presented in the competition for the academic title "Associate Professor" (27 publications and 2 teaching aids). After a dissertation defence for educational and academic degree PhD and obtaining the academic title "Associate Professor", the academic activity of Associate Professor Manov includes **48 materials** with which he participates in the competition for the academic title "Professor". These are monographs (2), publications in scientific journals (37, of which 12 with IF, 1 in a refereed journal abroad, 2 in Bulgarian refereed and 17 in Bulgarian and foreign non-refereed journals), collections from national scientific forums (5), participation in 2 national research projects. The applicant's publications are included in 2 textbooks and 5 teaching guides. In addition, the candidate has participated in 24 scientific forums (23 national and 1 international) with published abstracts.

4.1. Participation in research, research & development and educational projects.

In the enclosed materials, Associate Professor Manov presents 2 research projects to the Research Fund of the Ministry of Education and Science. The first in the competition "Stimulation of research in public universities" – 2010 and concluded Contract No. ДДВУ 02/62, dated 20 December 2010, Sofia. Project title "Studies on the epizootiology of current infestations in domestic and wild animals in Bulgaria, creating opportunities for early diagnosis and effective prevention ". The second project is in the National Scientific Program "Healthy Foods for a Strong Bioeconomy and Quality of Life" of the Ministry of Education and Science, approved by Decision of the Council of Ministers No. 577/17.08.2018 and Contract ДМ 01/1/216, co-financing contract with COST Action CM1704, ДКОСТ 01/11/2016. The first started in 2010 is at the Research Department of the UF, and the second in 2016 at the Bulgarian Academy of Sciences from the Institute of Biophysics and Biomedical Engineering with the participation of scientists from the UF.

The results of the research projects have been reflected in the published research work.

4.2. Characteristics of the published research results.

From all 48 works subject to review, with research results are 39 (81.25%) published in the period from 2007 until 2020. These include 2 monographs, 34 publications in scientific journals, including from national research projects and 5 in collections of national scientific forums. Out of 32 publications in scientific journals 12 are with IF 37.5% with **total IF = 23.659**, one in non-refereed journal (3.1%), 2 in Bulgarian refereed journals (6.3%) and 17 in Bulgarian and foreign non-refereed journals (53.1 %). 50% of them are in foreign journals.

The monographs are an independent work of the applicant (100%), and of the publications (37) in 5 of them he is an author (13.5%) and in the others he is a co-author.

The main part of the conducted researches is in the field of pathology caused by infectious agents, neoplasms and toxic substances. Current and modern are the researches for confirmation of the pharmacological activity of medicinal plants by using pathohistological methods. In addition, researches of scientific and applied nature were conducted. They are related to the creation of a model system for experimental cancer research, development of an *in silico* protocol for prediction of pharmacological activity, differential diagnostic scheme of diseases related to reproductive disorders in pigs and others.

4.3. Impact of applicant's research activities (citations).

Cited research works are 14 with total of 86 citations. Out of them, 6 are in scientific journals referenced and indexed in world-famous databases of scientific information (Scopus, Web of Science) with 54 citations and 8 in non-refereed journals with scientific review with 32 citations.

4.4. Contributions in research works of the candidate.

Scientific contributions.

Much of the research of Associate Professor Manov is related to studies of extracts, saponin fractions and secondary metabolites with hepatoprotective, antioxidant, antidiabetic and neuroprotective activity. The studies were conducted on **rats, isolated rat hepatocytes** (No.No. 1-3, 10), **brain synaptosomes** (No. 8) or **mice** (No.10). The effects were monitored by biochemical and pathomorphological studies.

The antioxidant and hepatoprotective effect of saponarins (No.No. 1, 2) or n-butanol extract (No. 3) with induced damage with paracetamol (No. 1) or carbon tetrachloride (CCl₄) – No.No. 2 and 3 were reported in the study of rat hepatocytes.

Antioxidant effect is shown by defatted extract of *Astragalus spruneri* (EAS) after spontaneous hypertension (No. 5). Purified saponin mixture (PSM) after 6-OHDA induced stress in addition to antioxidant effect has a neuroprotective effect studied on isolated synaptosomes from rat brain (No. 8). *Tert*-butyl hydroperoxide (t-BuOOH) and after induced oxidative stress showed antioxidant activity and effect on cell viability of isolated rat hepatocytes (No. 13). In contrast, flavoalkaloids and flavonoids after 6-OH-dopamine-induced stress with a study on rat hepatocytes have three effects – antioxidant, hepatoprotective and neuroprotective (No. 11). The alcesefoliside after lipid peroxidation with ferrous sulphate/ascorbic acid and induced brain toxicity (No. 9) and hepatic damage to rat hepatocytes (No. 15) with carbon tetrachloride (CCl₄) has a neuroprotective (No. 9) and antioxidant effect. Acute oral toxicity was observed in mice on PMS and its beneficial effect on blood sugar, triglyceride and total cholesterol levels in spontaneously hypertensive rats with streptozotocin-induced diabetes (No. 10).

The effects of *Ruscus aculeatus extract* (ERA) as a source of steroidal saponins, which could mimic sex hormones and their association with osteoporosis in patients, have been evaluated *in vitro*. The effect on the proliferation of human osteoblast-like cell line (SaOS-2), on the bone structure of estrogen-deficient rats induced by bilateral ovariectomy with histopathological and radiological examinations, as well as on their hormones (No. 12) was followed-up.

Another study aimed at an *in silico* and *in vivo* study of saponins of metabolic syndrome modulators. Potential metabolites and nuclear peroxisome proliferator-activated receptor gamma (PPAR γ) were studied as a factor for modelling the basic saponin in purified saponin mixture (PSM), the toxicity of PSM and antihyperglycemic, hypolipidemic, antihypertensive and antioxidant effect (No. 10).

Another experimentation field were studies conducted with **avian embryos** (chicken embryos – No.No. 6, 22, Japanese quails – No. No. 23, 25, and guinea fowls – No. 29) after *in ovo* treatment with hepatocarcinogens (N-nitrosodimethylamine and/or N-nitrosodiethylamine) with monitoring of histopathological, hematological and biochemical effects. Preneoplastic or neoplastic liver lesions as well as hematological and biochemical changes have been identified.

The studies in **pigs** are related to examinations of pathological processes caused by infectious agents. Clinical, macroscopic and histological examinations have shown the characteristic changes in spontaneous infection with porcine circovirus 2 (PCV2) in one of its forms – porcine dermatitis and nephropathy syndrome (PDNS) – No. 16. In long-term studies of infection with the same causative agent, it has been found that in addition to the described clinical signs and pathological changes characteristic of post weaning multisystem wasting

syndrome (PMWS) and PDNS, other common manifestations are observed, such as fever, lymphadenitis, conjunctivitis with periocular edema, signs of respiratory disease, anorexia, apathy, anaemia, jaundice, hyperaemia and cyanosis, abortions and stillbirths, congenital tremor, weight loss, kyphosis and high mortality (No. 20).

A comparative morphological analysis of CNS changes in not nursed piglets administered with vaccine strain MK 35gE- and two intrauterine isolates of Aujeszky's disease were performed. There was a difference in the clinic, mortality and pathohistological changes (No. 31). The studies were continued with histochemical and electron microscopic examinations (No. 34) with new data on the infection. Publication with No. 37 describes the clinical and pathoanatomical changes in viral and bacterial diseases with reproductive disorders in pigs, which are systematized in a user-friendly differential diagnostic scheme.

A molecular biological study of clinical samples of **domestic and wild carnivores** with gastroenteritis in Bulgaria was performed to prove CPV-2 infection and differentiate the type of CPV2a, CPV2b and CPV2c viruses by MGB probe (No. 4). In a spontaneous case of a sick dog, ultrasound, hematological, cytological, histological and immunohistochemical examination were performed, followed by diagnostic laparotomy. Hepatomegaly and neoplastic lesions in the liver and pancreas were found (No. 19), and with the expansion of imaging methods, it was found that more various malformations of the portal vein can be found in dogs (No. 24). A case of foreign pneumonia in a dog caused by an aspirated awn has penetrated the lungs and created a gateway for the reproduction of microorganisms with the development of purulent-necrotic inflammation with lethal outcome (No. 18). The clinical manifestations and numerous pathomorphological changes caused by vaccine strain and field uterotrophic strains of Aujeszky's disease virus in dogs and cats have been studied. A difference in virulence and pathohistological changes of the strains was found (No. 32).

A case of **bovine** paratuberculosis after a history obtaining, monitoring of the clinical manifestations, the histological, microscopic examination and conventional PCR examination was reported. Emphasis is placed on the difficult detection in subclinical cases of the disease (No. 27). An atypical course of pneumonia was found in cows with a pathoanatomical findings characteristic of pasteurellosis without isolation of *Pasteurella*, probably due to antibiotic treatment of the studied animals (No. 35). Publications No. No. 14 and 26 are related to a study on the reproductive capacity of buffalo-calves and cows. In buffalo-calves of 11-14 months of age, some morphological characteristics of the ovaries were found in early post-puberty, with some of the follicles having normal structure and physiological activity, but no ovum, despite the endocrine maturity of the hypothalamic-pituitary-gonadal endocrine system (No. 14). In the cow's vagina and uterus, normal cell clusters have been studied, and the importance of neutrophil leukocytes count in inflammation has been elucidated. The increased number of neutrophil leukocytes is an indicator of inflammation, and in clinical endometritis it is higher than in subclinical. Emphasis is placed on the possibilities of cytological examination in the diagnosis of inflammation and impaired reproductive function (No. 26).

A publication based on disease history, X-ray and computed tomography (CT) imaging studies followed the development of secondary sinusitis in **horses**, in view of topographic and anatomical premises for unilateral involvement of the complex of all six sinuses. The established osteolytic changes open the possibility to explain the expansion and complications of sinusitis in horses, with an emphasis on the anatomical characteristics of their sinuses (No. 28). A publication with No. 36 described a case of poisoning of horses after feeding with you. The result was obtained based on anamnesis, clinical signs and macroscopic lesions after autopsy. Clinical evidence of anxiety, incoordination, muscle tremors, shortness of breath, weakness, and convulsions has been reported, and autopsy findings include partial blood clotting, pulmonary edema, cardiac dilatation, hyperaemia of the gastric mucosa, and small intestine.

Medical teachers and students are exposed to formaldehyde due to its widespread use, often causing contact allergies. The cross-sectional study showed different sensitization to formaldehyde and it was highest among veterinary students (94.4%) and veterinarians (85%) compared to students and specialists in dentistry (No. 7).

Imaging, diagnostic, pathomorphological and immunohistochemical examinations were performed on a dancing Eurasian brown **bear** suffering from pulmonary and gastrointestinal symptoms. Ultrasound examination of the liver revealed many inhomogeneous hyperechoic masses. The pathomorphological examination of the lungs, liver, thickened part of the ileal wall and the adjacent mesentery demonstrated atelectasis. Histopathologically, anthracosis, atelectasis, venous stasis, haemorrhage and mineral deposits were found in the lungs. In the lungs and liver, neoplastic cells characteristic of neuroendocrine tumors were present. Immunohistochemical examination confirmed the diagnosis (No. 21).

Snakes at the age of 6 months of the species *Coluber caspius*, which died with neurological symptoms and allotriophagia, were studied. Microscopic examination of liver and peat litter revealed bacteria with a morphology typical of *Clostridium botulinum*. The clinical, pathological and microbiological results indicated that the most likely cause of these symptoms and death is botulinum intoxication (No. 30).

Contributions to learning/teaching activities.

The textbook on "General Veterinary Pathology" discusses the general pathomorphological processes. The morphological picture of the changes in these processes and states is recreated. The textbook is in accordance with the curriculum of the discipline General Pathomorphology of FVM at UF – Sofia (Textbook No. 1: Manov, 2018).

The textbook "Pathological characteristics of diseases in domestic animals" is intended for veterinary medicine students from UF – Sofia and for practicing veterinarians. Diseases significant for veterinary practice with data on etiology, general epizootological, pathogenetic and clinical aspects of manifestation are considered. Emphasis is placed on macroscopic and basic histological changes. Frequently used laboratory methods for definitive diagnosis and differential diagnostic options for comparison between diseases are presented (Textbook No. 2: Manov, 2020).

The tutorial manuals "Veterinary Autopsy Technic and Incineration" introduce the veterinary medicine students with the purpose of performing the pathoanatomical examination, its order and sequence, used tools, safety measures and more. The method of taking, processing and sending materials for additional laboratory analysis, preparation of permanent macroscopic preparations and preparation of autopsy protocols is described. The methods for carcass destruction are considered and the structure and technological processes in the incinerators (Technical manuals No. 3 and 4: Stoykov et al., 2007; Stoev et al., 2016).

The technical manuals (in Bulgarian and English) on "Veterinary histopathology" are used in the training of students in veterinary medicine in the study of microscopic changes occurring in the animal body in various pathological conditions. They allow the performance of individual work, the development of creative thinking and tracking of the studied material (Technical manuals No. No. 5-7: Dinev et al., 2012; Dinev et al., 2016).

5. Evaluation of the personal contribution of the applicant.

The review of the materials presented in the competition shows the in-depth approach and systematic study and summarization of the researched issues by the applicant. In addition to the detailed analysis, there is a clear desire to build on existing knowledge through personal contribution. This is evident both from the habilitation work of the applicant and from the scientific publications presented at the competition. I have reason to conclude that the achievements are the personal work of the applicant, combining critical and creative thinking.

6. Critical notes and recommendations.

None.

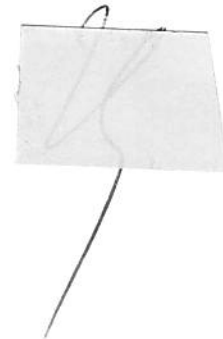
7. Personal impressions.

I know Associate Professor Manov from his work at the Trakia University. For the entire past period until now he has developed as an erudite and experienced teacher and researcher. He can work independently and in a team and has the qualities and skills to hold the academic position of "Professor".

8. Conclusion.

In summary, it can be stated that the volume and quality of the teaching and research activities of the candidate fully satisfies the requirements of the Law for the Development of Academic Staff in the Republic of Bulgaria (LDASRB), Regulations for the application of the LDASRB and Statute for Development of the Academic Staff of the Republic of the University of Forestry – Sofia. The quantitative requirements for holding the academic position "Professor" are met. In relation to the above, I **propose Assoc. Prof. Vasil Kostadinov Manov, DVM, PhD to be conferred as "Professor"** in the professional field 6.4 „Veterinary Medicine“, scientific specialty “Animal Pathology”, discipline “Pathology/Special Pathological Anatomy”.

Undersigned:

A handwritten signature is written on a rectangular piece of paper. The signature is in dark ink and appears to be a stylized name. The paper is slightly tilted and has a thin line extending downwards from its bottom edge.

The opinion report is presented on 10.07.2020