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REVIEW

for the competition for the academic position of "professor" in the professional field 4.3 Biological Sciences, scientific domain **"Parasitology and Helminthology", for the needs of the Section Biodiversity and Ecology of Parasites, Department Animal Diversity and Resources of IBEI-BAS**

Candidate: Assoc. Prof. Dr Yasen Zhelyazkov Mutafchiev

Reviewer: Prof. Dr Aneta Kostadinova, IBEI-BAS

In the announced competition (SG, issue 45/18.07.2025) for Professor in Parasitology and Helminthology at the Section Biodiversity and Ecology of Parasites, Department Animal Diversity and Resources (IBER-BAS), Dr Yasen Zhelyazkov Mutafchiev, currently an Associate Professor in the same section, is the only candidate.

1. Overview of the candidate's career development

Yassen Mutafchiev graduated from the Faculty of Biology of Sofia University "St. Kliment Ohridski" with an MSc degree in the field "Zoology of Invertebrates" in 2005. In 2009, he defended his PhD dissertation entitled "*Species diversity of nematodes of the superfamily Acuarioidea (Spirurida) in Bulgaria*". He worked as a biologist in the Central Laboratory of General Ecology (2005–2010) at the Bulgarian Academy of Sciences (BAS), and as a Senior Assistant Professor (2010) and Associate Professor in the Department Animal Diversity and Resources of the Institute of Biodiversity and Ecosystem Research (IBER-BAS) (2015-present). Dr Mutafchiev's scientific and career development is associated with the implementation of a short-term specialization (2010: Muséum National d'Histoire Naturelle, Paris) and a series of research visits for joint studies *via* invitations by leading scientists (2009: Muséum d'Histoire Naturelle, Ville de Genève - invitation by Dr Jean Mariaux; 2011: Muséum National d'Histoire Naturelle, Paris - two visits; invitations by Dr. Odile Bain; 2013: University of Bari, Italy - invitation by Prof. Domenico Otranto; 2013: Muséum National d'Histoire Naturelle, Paris - invitation by Dr. Coralie Martin).

2. Publications submitted in connection with the competition

For participation in the current competition, Dr Mutafchiev has submitted a list of 56 publications: 1 chapter in a book published by an international academic publishing house and 55 articles in

predominantly international journals, of which 52 are in journals with an impact factor (IF) or impact rank (SJR). All publications are written in English.

The scientific output after the last habilitation of Dr Mutafchiev comprises 22 publications, of which 18 are in international journals with an IF, and 4 are in a Bulgarian journal with an IF. The candidate is the sole author of one publication, and as a co-author, he is the first and corresponding author of 6 publications, the second author of 7 publications, and the third- and subsequent author of the remaining 8 publications. I should note that most of the articles were published in prestigious international specialised scientific journals such as *Systematic Parasitology*, *International Journal for Parasitology*, *PLoS Neglected Tropical Diseases*, *Parasites & Vectors*, *Current Research in Parasitology & Vector-Borne Diseases*, *Parasitology*, *Zoologica Scripta*, *Diversity*, *Parasitology Research*, and *Journal of Helminthology*, including journals with high IF and SJR rank (Q1 and Q2: publications 35, 37, 38, 44, 48, 49, 50, 52, 53, 55, 56).

The submitted report on meeting the minimum indicators according to the Law on Academic Achievement and Research of the Republic of Bulgaria and the Regulations of IBER-BAS shows that these minimum requirements are significantly exceeded: the total number of points for groups of indicators A, B, D, E and E of Dr Mutafchiev is 1057, and the minimum number of points is 640.

3. Main areas of the candidate's research and most important scientific contributions

I fully accept the report on the scientific contributions presented by Dr Mutafchiev, in connection with the competition. As his habilitation work, he has chosen 7 publications in the field of morphology, taxonomy, phylogeny and life cycles of nematodes parasitic in vertebrates, for which he has a leading role as a member of the author team. Dr Mutafchiev grouped his most important scientific contributions into 13 thematic areas, closely related to the scope of the competition, and can be briefly summarised in 4 main areas as follows:

Contributions to the taxonomy, systematics, phylogeny and life-cycles of nematodes of the family Acuariidae (Spirurida)

1. As a result of a detailed study of the morphology, taxonomy and systematics of nematodes of the family Acuariidae, 10 species new to science were described (*Acuaria europaea*, *Acuaria paraguayensis*, *Cosmocephalus pelecani*, *Cosmocephalus podicepsi*, *Proyseria petterae*, *Quazithelazia alata*, *Quasithelazia minuta*, *Quasithelazia pearsoni*, *Quazithelazia rostrata*, and

Syncuaria mackoi) and 3 new genera were erected (*Chabaudacuaria Parachordatortilis* and *Pelecanema*). Keys were prepared for the identification of the species of the genera *Cosmocephalus* (7 species) and *Streptocara* (6 species), as well as of the species of the genus *Acuaria* from the New World (16 species). Changes and amendments to the generic diagnoses of 3 genera (*Proyseria*, *Quasithelazia* and *Syncuaria*) were proposed, 19 species were redescribed (*Acuaria anthuris*, *A. mamillaris*, *A. attenuata*, *A. muscicapae*, *Alinema sturni*, *Chabaudacuaria multispinosa*, *Cosmocephalus capellae*, *C. jaenschi*, *C. obvelatus*, *Decorataria magnilabiata*, *Parachordatortilis mathevossianae*, *Pelecanema pelecani*, *P. sirry*, *Proyseria decora*, *Quasithelazia tenuis*, *Streptocara crassicauda*, *S. incognita*, *S. recta*, and *Stammerinema hyalinum*) and 5 new synonyms at the species level and 10 new combinations were justified [2, 6, 7, 8, 9, 11, 13, 23, 24, 27, **36**, **45**, **48**, **52**, **55**].

2. A phylogenetic hypothesis for the family Acuariidae was proposed, based on newly generated 28S rDNA sequences for 18 species from 15 genera, which confirms the monophyly of the family Acuariidae and supports the relationship of this family to the family Cystidicolidae. Two large groups were identified in the family; one corresponds to the subfamily Schistorophinae, and the other includes representatives of the subfamilies Acuariinae and Seuratiinae. It is proposed to synonymise the Seuratiinae and include its genera within the subfamily Acuariinae. The distribution of morphological characters and host spectra in the different evolutionary lineages of the group was analysed [**48**].

3. The-life cycle of *Streptocara incognita* (Spirurida: Acuariidae) was elucidated based on an integrative (morphological and molecular) approach. Comparative analysis of mitochondrial 12S rRNA gene sequences generated from infective third-stage nematode larvae found in the amphipod *Gammarus pulex pulex* and from adults morphologically identified as *S. incognita* demonstrated their conspecificity. Characteristic morphological features of the third-stage larva, as well as of the fourth-stage larva and young and mature adults, were documented by light and scanning electron microscopy. The brief original description of the species was supplemented. The first published 12S rRNA and *cox*1 sequences associated with morphologically characterised larval and adult stages of *S. incognita* will serve as reference sequences for the species [**55**].

Contributions to the morphology, systematics, biology and phylogeny of filarial nematodes of the family Onchocercidae (Spirurida)

4. The first comprehensive morphological and morphometric description of *O. lupi* was prepared based on material from dogs, studied by light and scanning electron microscopy, as well as histological observations [16].

5. A phylogenetic analysis of the family Onchocercidae was performed, based on seven nuclear and mitochondrial loci from 48 species representing seven of the eight subfamilies. The results support the monophyletic origin of the family and the hypothesis of early differentiation of the subfamilies Oswaldofilariinae, Waltonellinae and Icosiellinae, but do not support the monophyly of the subfamilies Onchocercinae, Dirofilariinae and Splendidofilariinae, suggesting that the boundaries between these three subfamilies need to be reassessed [**35**].

6. The phylogenetic relationships of 13 species of the genus *Onchocerca* were studied based on 7 genes. Three strongly supported groups were identified. The study showed that in the group of *O. lupi* (parasite of canids – dogs and wolves), *O. gutturosa*, *O. lienalis*, *O. ochengi* (parasites of domesticated bovines) and *O. volvulus* (parasite of humans), speciation is associated with adaptation to new hosts (host-switching). The results indicate that domestication and close contact of domesticated animals (cows and dogs) and humans contributed to this. Co-phylogenetic analyses between *Onchocerca* and their endosymbiont bacteria of the genus *Wolbachia* showed a strong co-evolutionary relationship [**44**].

7. Two cases of human filariasis caused by *Onchocerca lupi*, a poorly studied parasite of dogs and wolves, were documented in Turkey and Tunisia, proving that *O. lupi* is the causative agent of ocular nematodiasis in humans along with *Dirofilaria repens*. For the first time, a detailed morphological description of *O. lupi* was made based on material isolated from a dog in Portugal. The morphological features documented by light and scanning electron microscopy are important for the correct identification of the parasite and the identification of potential agents of dangerous zoonoses in humans [12].

8. A study was conducted on the distribution and diurnal activity of *O. lupi* microfilariae in dogs. Microfilariae were concentrated in certain regions of the host's body and showed higher numbers at certain times of day [14].

9. *Cercopithifilaria rugosicauda* was found in the subcutaneous connective tissue of a roe deer in Italy. The report of the species, previously known from temperate regions in the Palearctic, is the first in subtropical climates. The morphological description of the species was accompanied by the publication of the first molecular data (sequences of *cox*1 and 12S rRNA genes). A sample of *C.*

rugosicauda from France was morphologically and molecularly characterised. Immunological and molecular tests of the species were performed, which did not detect bacterial symbionts of the genus *Wolbachia*, characteristic of some filariae. The study systematised the main morphological features of the species of the genus *Cercopithifilaria* and complemented the generic diagnosis [21, 22].

10. A clinical case of rare equine onchocerciasis caused by *Onchocerca boehmi* was documented. This is only the second report of the species in Europe since its description in Austria in 1956. Morphological and molecular data (fragment of the *cox*1 gene) obtained as a result of the study support the systematic position of the species in the genus *Onchocerca*, rather than in *Elaeophora*, where it was originally described [**37**].

Contributions to the morphology, taxonomy, systematics, and phylogeny of other nematode groups of the orders Ascaridida, Camallanida, and Spirurida

11. Family Spirocercidae (Spirurida): Three new species of parasitic nematodes of the genus *Cylicospirura* (*C. crocutae*, *C. pardalis*, and *C. phiri*) were described. The generic diagnoses of *Cylicospirura*, *Gastronodus*, and *Skrjabinocercina* were amended, and the validity of the genera *Gastronodus* and *Skrjabinocercina*, considered by previous authors to be synonyms of *Cylicospirura*, was restored. Three new combinations were proposed. One species (*C. felinea*) was redescribed, and the morphological descriptions of 2 species (*C. crocutae* and *C. pardalis*) were supplemented [17, **53**].

12. Family Physalopteridae (Spirurida): One species (*Thubunaea schukurovi*) was redescribed, and a critical analysis of the species of the genus *Thubunaea* from the Palearctic and Indo-Malaysian regions was carried out. As a result, *Thubunaea dessetae* was transferred to the genus *Pseudabbreviata*, and 5 species (*Thubunaea singhi*, *T. brooki*, *T. aurangabadensis*, *T. syedi*, and *T. hemidactylae*) were transferred to the genus *Physalopteroides* [**50**].

13. Family Aproctidae (Spirurida): One new species of the genus *Aprocta* (*A. bainae*) was described. In addition to the morphological description made using light and scanning microscopy, sequences of the nuclear 18S rRNA gene and the mitochondrial 12S rRNA gene were published. Phylogenetic analysis based on the 18S rRNA gene placed the new species in a monophyletic relationship with another North American species, published as *Aprocta* sp. [56].

14. Family Heterocheilidae (Ascaridida): Two new species (*Ingwenascaris sprenti* and *Typhlophoros kwenae*) were described, and a new genus *Ingwenascaris* was erected with two species: the newly described *I. sprenti* and *Porrocaecum assymmetricum*, for which a new combination is proposed. A detailed redescription of *Multicaecum agile*, a species with insufficiently studied morphology, was provided, with illustrations and scanning electron microscopy [**41**, **42**, **47**].

15. Family Kiwinematidae (Ascaridida): One new species, *Mammalakis zambiensis*, a parasite in the colon and cecum of *Fukomys anselli* (Rodentia: Bathyergidae), was described. [17].

16. Family Micropleuridae (Camallanida): One new species, *Micropleura huchzermeyeri*, a parasite in the body cavity of the Nile crocodile, was described. *Micropleura helicospicula* Dey Sarkar, 2003 was designated as a species of uncertain systematic position (*species incertae sedis*) [43].

Contributions to the fauna, biology and ecology of nematodes and helminths

17. In a study of the nematode fauna of the Nile crocodile in the Kruger National Park, 11 species of nematodes were identified: 5 species of the family Heterocheilidae (*Dujardinascaris madagascariensis*, *D. dujardini*, *Ingwenascaris sprenti*, *Typhlophoros kwenae* and *Multicaecum agile*); 1 species of the family Micropleuridae (*Micropleura huchzermeyeri*); 1 species of the family Capillariidae (*Crocodylocapillaria* sp.); 2 species of the family Camallanidae (*Camallanus kaapstaadi* and *Spirocamallanus* sp.); 1 species of the family Anisakidae (*Contracaecum* sp.); and 1 species of the family Cystidicolidae (*Ascarophis* sp.). Two species (*D. dujardini* and *D. madagascariensis*) were identified for the first time for the fauna of South Africa. During the course of the study, morphological descriptions of 2 species (*M. agile* and *C. kaapstaadi*) were also made [**47**].

18. The development of the nematodes *Aelurostrongylus abstrusus* and *Troglostrongylus brevior* (Angiostrongylidae), parasitising the lungs of cats, and the nematode *Crenosoma vulpis* (Crenosomatidae), parasitising the respiratory tracts of foxes and dogs, was studied in the common garden snail, *Cornu aspersum* (=*Helix aspersa*). It was found that the nematode larvae of *A. abstrusus* and *T. brevior* successfully reached the invasive stage in 11 days and were able to survive for at least 120 days after invasion of the snail. *Crenosoma vulpis* successfully reached the invasive third larval stage in 10 days and could survive in the snail for at least 180 days. *Cornu aspersum* was identified for the first time as a potential intermediate host for *T. brevior* and *C.*

vulpis. The invasion stages (first and third) for the intermediate and definitive hosts of all three species of metastrongyloid nematodes were morphologically described and documented [16, **38**].

19. The helminth fauna of four fish species in Atanasovsko Lake was studied (2012-2013). In the invasive species *Lepomis gibbosus*, 6 helminth species were found (metacercariae of the trematode *Posthodiplostomum centrarchi*; the monogeneans *Onchocleidus similis* and *O. dispar*); the nematode *Schulmanela petruschewskii*; as well as larvae of the nematodes *Spiroxys contortus* and *Contracaecum* sp.). The small number of specific parasites found (3 species), compared to their number in the natural range of the host, supports the enemy release hypothesis, which favours the successful invasion of the fish species. In *Knipowitschia caucasica*, 4 helminth species were identified (the trematodes *Aphalloides coelomicola* and *Paratimonia gobii*; the monogenean *Gyrodactylus bubyri*; and third-stage nematode larvae in the body cavity, identified as *Contracaecum* sp.). In *Gasterosteus aculeatus*, 5 helminth species were identified (metacercariae of the trematode *Posthodiplostomum brevicaudatum*; the monogenean *Gyrodactylus arcuatus*; plerocerci of the cestode *Progrillotia dasyatidis*; and third-stage nematode larvae of *Contracaecum* sp. and *Hysterothylacium* cf. *aduncum*). In *Syngnathus abaster*, 3 helminth species were identified (immature and mature individuals of the trematode *Timoniella imbutiformis*; metacercariae of the trematode *Cryptocotyle concava*; and third-stage nematode larvae of *Hysterothylacium* cf. *aduncum*). *Timoniella imbutiformis* and *G. arcuatus* are reported for the first time for the Bulgarian fauna. Morphological data are presented for the monogenean *G. arcuatus*, as well as for the larvae of the genera *Hysterothylacium* and *Contracaecum* sp. [**40**, **46**, **51**, **54**].

20. A four-year study was conducted on the dynamics of gastrointestinal helminths in translocated and resident European ground squirrels during and after population reinforcement. Eggs of acanthocephalans and nematodes were found in the studied coprological samples, the latter being morphologically identified as belonging to species of the families Capillariidae (Enoplida) and Trichostrongylidae (Strongylida), and the superfamily Spiruroidea (Spirurida). It was found that the overall prevalence of helminths and their diversity were higher in the donor colony (characterised by a high density of hosts) compared to the reinforced colony before the translocation. Levels of parasitism were significantly lower in spring than in summer, in both translocated and resident hosts. One year after the start of the translocation, the helminth prevalence and the number of species in the reinforced colony increased significantly. This is consistent with epidemiological models and other empirical studies that predict a positive relationship between host population density and parasite prevalence and species richness. However, no negative effects of parasites on the host were found [49].

4. Significance of the research conducted by the candidate

Dr Mutafchiev has presented a list of 339 citations of his scientific publications, the majority of which are in international and foreign journals indexed in Web of Science or Scopus. These data far exceed the criteria of the law and the additional requirements of IBER for holding the academic position of professor, thereby supporting the originality and relevance of Dr Mutafchiev's scientific research, which has received a wide response in the scientific community.

5. Most significant applied science contributions

The detailed studies of Dr Mutafchiev on the morphology, systematics, biology and phylogeny of filarial nematodes of the family Onchocercidae (contributions 4-10) hold significance for both human medicine and veterinary practice.

6. Project activities

After his last habilitation, Dr Mutafchiev has participated in 4 scientific projects and is currently leading 1 project funded by the Scientific Research Fund (2023-2027) (total participation/ leadership in 10 projects, including 3 international ones). I also believe that the five projects funded by the European programme Synthesys 1-3 (2007-2015), which are not included in the criteria compliance report, are particularly prestigious.

7. Candidate's research profile

The scientific output of Dr Mutafchiev after his previous habilitation, characterises him as a promising, productive scientist with a particularly clearly outlined research profile - studying the diversity, taxonomy, systematics, phylogeny, and life-cycles of zooparasitic nematodes. I believe that this profile of Dr Mutafchiev is unique on a global scale, and his significant contributions to the field make him a welcome leading participant in international research. One of these contributions is the establishment of modern standards of morphological study of zooparasitic nematodes and the integration of molecular data. Another aspect of the uniqueness of Dr Mutafchiev's expertise is the wide taxonomic extent of both the parasites and their hosts, as well as their geographical distribution. All contributions submitted under this competition are original in nature, and the number of significant taxonomic and systematic contributions as well as nomenclatural acts submitted within the competition is impressive: descriptions of 19 new species;

redescriptions of 32 species; erection of 3 new genera; taxonomic revision of 7 genera; elaborated keys for identification of 3 genera; 21 new combinations; and 6 new synonyms. These data support my confidence that the election of Dr Mutafchiev as a professor will have a positive impact on both the research conducted in the section Biodiversity and Ecology of Parasites and the staffing of the scientific speciality "Parasitology and Helminthology".

8. Role of the candidate for training young scientists

Dr Mutafchiev was a scientific advisor of a PhD dissertation (Maria Kachamakova, IBEI-BAS, PhD degree obtained in 2021), as well as a scientific supervisor of an undergraduate student (Krasimir Bachvarov, Faculty of Biology, Sofia University "St. Kliment Ohridski", MSc degree obtained in 2017).

9. Critical notes

There is no copy of scientific publication 55 in the documents – instead, a copy of publication 56 was given. In the reference for contributions, publication 55 was cited as 52.

Conclusion

The candidate Dr Yasen Mutafchiev participated in the competition with a significant in quantity and extremely valuable scientific works. The submitted documents show that his scientometric indicators conform and significantly exceed the requirements of the Act on the Development of the Academic Staff in the Republic of Bulgaria (ZRASRB), the Regulations of the Bulgarian Academy of Sciences for the implementation of this law and of the Scientific Council of IBER-BAS for holding the academic position of professor. The high professionalism and international expertise of the candidate give me reason to recommend with full confidence to the Scientific Council of IBER-BAS the election of Dr Yasen Mutafchiev as a professor in professional field 4.3. Biological Sciences, scientific speciality "Parasitology and Helminthology".

September 30, 2025

Reviewer:

(Prof. Dr. Aneta Kostadinova)