Review

of Associate professor PhD Dimitar Stefanov Kozuharov, Department of General and Applied Hydrobiology, Biological Faculty, University of Sofia "St. Kliment Ohridski".

Member of the Scientific Jury by Order No. RD-38 - 548 /19.09.2023 of the Rector of SU "St. Kliment Ohridski" - About competition for the academic position "associate professor" in the professional direction 4.3 Biological Science (Hydrobiology - Management of the waters) for the needs of department "General and applied Hydrobiology" announced in State Newspaper N 67/04.08.2023.

In present competition the only candidate is Assist. prof. PhD Ivajlo Iotinov. The candidate has submitted all the necessary documents in accordance with the requirements of the Act on Development of the Academic Staff in Republic of Bulgaria – (ADASRB), the Rules for the Implementation of the ACT, and the Rules for the Terms and Procedures for Acquiring Scientific Degrees and Holding Academic Positions in Sofia University "St. Kliment Ohridski". The submitted documentation of the candidate in the competition is given in way that summarize candidate's academic activity and achievements.

Biographical remarks about the candidate

Assistant prof. Iotinov graduated in the Faculty of Biology of the Sofia University "St. Kliment Ohridski" as bachelor degree in specialty "Biology" in 2010 year. He graduate as a master degree in "Ecological Biotechnology" in 2012 year. In 2016 he obtained the scientific and educational degree - "Doctor of Philosophy" (PhD). Assist. prof. Iotinov has 8 years of teaching experience at Sofia University. Most of them were at academic position – assistant and chief assistant professor. He took in 2016 position of assistant in the Department of "General and Applied Hydrobiology". From 2017 he was elected at the position of chief assistant. The academic development and professional qualifications of the candidate correspond to the subject of the announced competition of the associate professorship in the Department of "General and Applied Hydrobiology".

Teaching activity.

The teaching activity of Assist. prof. Iotinov is significant. It includes lecturing and giving practical exercises and seminars in several courses for students from various specialties in bachelor and master degree at the Faculty of Biology of the University of Sofia. Assist. prof. Iotinov leads the lecturing courses in Water management for specialty of Biomanagement and Total quality management for bachelor degree students of specialties Biomanagement and Ecology and Environment protection. He also gives lecturing and practical exercises in different courses for the bachelor and master degree students. He takes participation also in the Summer field practices in Hydrobiology for bachelor degree students and Summer field course for the master degree students in Ecological Biotechnology . His average year teaching load is calculated as 534 hours. For the last academic year his academic horrarium was 803 hours.

Assist. prof. Iotinov was the scientific supervisor of 18 graduate students who successfully defended their diploma thesis.

I can say that teaching and learning activity of assistant prof. Ivajlo Iotinov, is more then needed for such type of competitions.

Research activities of the candidate.

The main research topic of assist. prof. Iotinov is in the field of water management. He took participation in 32 scientific and applied scientific projects. It is very high number of projects – for 8 years of experience as a university teacher he has average of 4 projects per year. Assist. prof. Iotinov participates in this competition with total of 37 publications such as 28 scientific articles, and 9 published in full text conference reports. Assist. prof. Iotinov is the first author in 10 of the 37 scientific articles submitted for the competition. The work with co-authors in scientific publications shows abilities for cooperation with colleagues, specialists in the team of the laboratory of Ecological Biotechnology and these from the department of General and applied Hydrobiology. The candidate gives list of 63 citations of his scientific papers. This is proof of the interest about his publications from the scientific community.

The scientific researches of the candidate are contemporary and have high scientific and appliedscience values. The studies used different modern methods of analysis. They are pointed on biological elements of the water technologies and biotechnological elements of the control and management of the waters. Some original and some upgrading contributions in the papers of the candidate were shown. The focus in the numerous of his papers is about the control and management of the waste water treatment processes. A system of the control of the Hydrobiological, microbiological, hydro chemical, enzimological and molecular-genetic indexes what more then standard ones in the WWTP were used. Here may comment is that I disagree with used from the candidate term – natural seflfpurificational process, this process can be only natural – that's why it is call "self-purification".

Original and upgrading contribution of the candidate is with the accent on the relationships between the micro and metafauna with the bacteria in the activated sludge in the biobasins and the sequencing-batch reactors in waste water treatment plants. These relationships were investigated as a basic indicator for control and management of waste water treatment processes in the model conditions for biodetoxication of xenobiotics like amaranth, phenol and waters from hard wastes depot. For the micro and metafauna a significant role in the adaptive response of the activated sludge and specially this of new bacterial complexes were found and registered. The candidate clearly shows that his investigations in this direction upgrade previous one of the scientific team part of what is now. It is right from my point of view that relationship between the bacterial complexes and micro and metafauna is one of the mechanisms what can be use effectively for the detoxication in the real waste water treatment processes. Second important role, what he confirm for the fauna complexes is that they contribute for the compensative increasing of the bacterial quantities. These questions were considered in six of the papers in the competition.

Another accent with upgrading of the contributions, is application of the enzimological and molecular – genetics indexes in the management of the waste water treatment processes. The application of the specific oxygenize and dehydrogenase abilities of the microorganisms in the activated sludge give detail information about their biodegradable abilities. The investigations were conduct on the oxygenize activity/catechol – 1,2 – deoxygenize, catechol- 2,3 – deoxygenize, protocatechat – 3,4, deoxygenize etc. together with other indexes like COD, BOD and volume index, philament and biotic index of AS. The were applied for the assessment from the effects of the shock load with heavy fractions of the petrol products in the waste water treatment plant Kubratovo. In connection of this is the specialization of the candidate – increasing of the information on the connection between different trophic levels and the dynamics in these

connections to the increasing effectiveness of the waste water treatment processes. These questions were discussed in 12 of the papers in the competition.

The candidate have been worked on the assessment and the control of the biological management of the river ecosystems, what are used as sources for energy production by water power plants. Such system was build up as a cascade of 5 water power plants in the region of Iskar river gorge. In this antropogenically influenced system many problems what need technological decisions exists. The goal of the completed 12 years monitoring program was to assess changes in the water quality and the ecological status of the river sediments, as well the status of the hydroecosystem in the region of the build and exploitation of the cascade of the middle Iskar river stream.

In connection of these studies I have a question – How, when and from what sampling points was assessed the changes in the subterranean waters in the region of the cascade of these micro reservoirs? The whole region is karstic and several big karstic springs exists there. Most of them are used as water sources for domestic water supply.

During the study was established that most risky are the reservoir sediments. They contain high organic content, xenobiotic contaminants and heavy metal pollution. The candidate was work on the microbial complexes and search for some indicators for express control.

The experiments for the application of the newly found factors of the biodetoxication processes such as nano diamantes and nano tubs was carried out. Also he constructs algorithms and strategies for the control and management of risk events in the sediments of the cascade "Middle Iskar". That is new tendency in the investigations and the education practices. It can be accept as a contribution to the management of the water resources. In the laboratory conditions were prepared model for treatment of the reservoir sediments, contaminate with both domestic organic wastes and xenobiotics. In these conditions the run off the adaptation and self-purification process was followed.

The candidate was carried out experiments connected with functional approaches for microbial complexes in anaerobic facilities of waste water treatment plants.

In this interdisciplinary study different specialists such as specialists of management, analogizes, partners who give samples, technologies from different fields were involved. The study is in direct connection to the field of the competition – **water management.**

Some molecular methods were described what can be effectively used for the biological control of the biogas production from the sludge's of waste water treatment plants by the building up of

bioindicative system. This system for the management of the methanogen processes can be unique and could give high plasticity of the control in the anaerobic tanks of different purification plants.

In the part of the included papers the application of the elements for the functional control and management of the key processes of the water cycle were discussed.

Correlation between fundamental parameters of the control of the selfpurificational potential in waters and sediments was found. This was achieved during the in situ monitoring strategy of the Middle Iskar cascade of reservoirs. The aim of these correlations was to make easier path for understanding the processes in the sediments of the lotic and lentic ecosystems.

Often the study of these elements takes difficult to use methodological apparatus, what is very expensive and takes long periods f time for having propriety results. In this connection the establishment of the functional indicators, what give possibility for express diagnosis for the assessment and management of the risk processes, have great ecological and financial importance.

Paralley in the laboratory model were build up the conditions of the processes of self-purification in the reservoir sediments what were highly contaminated by organic meter and different xenobiotics. These xenobiotics are most probable contaminant of the ecosystem of Lakatnik reservoir as a result of the anthropogenic influence.

In connection of this I have a question – Is the Lakatnik reservoir separated ecosystem, heaving in mind its volume, depth and surface area and the exchange of the water quantities, or it is a part of the larger ecosystem of Iskar river.

The candidate take participation in the study of the adaptation mechanisms of the micro- and metafauna and bacterial complexes in the activated sludge during the shock loading with xenobiotic – amaranth, with or with out adding of the specialized bacterial strain *Pseudomonas aureofaciens AP-9*. To achieve the goal changes in the quantities of different organisms from the groups of Ciliophora, Amoeba, Flagellata, Rotifera and Nematoda were followed during the model biodegradation process in the presence of real activated sludge from waste water treatment plant with toxic pollutant amaranth in concentration of 200mg/l. Paralley the effect of the added microbial culture of *Pseudomonas aureofaciens AP-9* on the micro- and metafauna complexes was analyzed.

It was established that during such shock load a fast transformation in the micro- and metafauna complexes becomes. A correlation between the quantities of the micro- and meta fauna and azo-degradable bacterial complex and aerobic heterotrophic bacteria was found. Connection between the decreased quantities of the cultivated microorganisms and change to the lower trophic levels of the micro and meta fauna complexes. Except this a positive effect from the added strain of *Pseudomonas aureofaciens AP-9* for the micro and meta fauna complexes was registered.

In connection with these papers and published in them results I have two questions: What organisms from the micro fauna s.str. were positively influenced from the adding of *Pseudomonas aureofaciens AP-9*? Are there similar data what have been found during some previous experiments with this particular strain?

It was found out that this helps surviving the fauna complexes and keeps alive fundamental trophic levels of activated sludge. This gives bigger chance for the effective flow in of the purification processes. These questions were discussed in seven papers of the candidate.

The effect of the adding of the nanodiamants on the biodegradation processes of the microbial dominant complexes in the reservoir sediments was studied too.

It was confirmed that reservoir sediments from Middle Iskar cascade were inhabit by functioning adapted to the risk contamination bacterial communities.

The idea was supported also by the results from the enzimological indexes of the bacterial complexes from the sediments, what prove their high biodegradation activity, connected with intensive self-purification processes. In the laboratory conditions with poor microbial cultures and model xenobiotic – phenol was investigated the effect of the nanodiamants on the bacterial activity focused on the biodegradation of phenol.

The pictures taken by scanning electron microscope proof that exists consortium of nanodiamants and bacterial cells. The formed consortiums possibly were zones for intensive phenol biodegradation. This can shows new way for increasing the speed and effectiveness of biodegradation. On the base of the modeling of the effect of the nanodiamants on the phenol biodegradation in laboratory conditions for first time were proposed hypothesis about the mechanisms for improving effects of the nanodiamants on the biodegradation of the toxic arilcontaining xenobiotics. That was discussed in five of the papers.

Questions – How economically effective will be the application of such nanodiamants in industrial scale treatment processes?

What effects will have such application of nanodiamants on the other organisms in the ecosystems what inhabits pelagial and sediments?

The nanodiamants were applied in the several experiments in conditions close to the real one during the model bioremediation of the detoxication process of the contaminated by the phenol reservoir sediments from the Lakatnik reservoir. The model bioremediation of the reservoir sediments what were contaminate simultaneously by domestic pollutants and phenol in high quantities was provided too. Before these experiments with nanodiamants, there were no information about their application in the processes of the purification of contaminate waters and sediments in the accessible literature.

The candidate established that during the shock load by phenol in the concentrations near to the critic nanodiamants decrease phenol concentrations because of physics chemical processes and helps increasing of the biomass of the inhabiting organisms. The nanodiamants accelerate the process of the detoxication and their effect is higher in the risky situations during the shock loads with extremely high concentrations of xenobiotics. The applied results can be used in the close conditions in the reservoir and river sediments.

Chef assistant Iotinov participates in the studies of the effects of treatment of water by the so call plasma torch, with surface waves at 2,45GHz. As result deactivation of the Gram positive and Gram negative bacterial cultures in suspension was registered. That effect depends on the time of treatment, power of the wave and the volume of the treated liquid. It can be presume that in next studies the effects of plasma on the liquid can be use for the water treatment in different facilities, like propriet method for bacterial inactivation.

From my point of view this technology is more applicable during medical treatment of the waters then during the treatment of waste waters.

The data shows the based on plasma technologies can be used in purification of waters contaminated by aril, as a method for next purification and for the increasing of the efficiency of the biological elimination. The bactericide effect of the plasma can positively influenced on the treatment of the waters when these technology is in use in combination with biological method for elimination of contaminants.

With leading role of the laboratory of "Ecological Biotechnology" two applied indexes were applied – philament index of the activated sludge and the biotic index of the activated sludge from the waste water treatment plants.

When think about such application of these indexes I have two questions:

Is the candidate applied the biotic index in general or just in the laboratory of Ecological biotechnology?

There are many indexes what are prop pried for assessment of the activated sludge state such like index of individual species diversity after Shannon – Weaver, or common species diversity after Margaleff etc. Why the candidate thinks that this index is more appropriate then the many other indexes what exists and are in use in the Hydrobiological practice and what concern communities in the natural and artificial water bodies and waste water treatment plants?

The applied filament index of the activated sludge what was found in cooperation with Veolia group – Sofia Water LTD, and what is in use in their laboratory for waste waters in Kubratovo is highly usable. This index helps assessment and control of the critical technological problems – such as enlarging of the volume of activated sludge. Dr. Iotinov took participation in application of method for determination of the common organic carbon in waters and sediments.

The candidate participate in the development of the methods for investigation of bacteria and micro- and metafauna from waters and sediments by use of confocal electron laser microscope. These methods were developed by use of apparatus in the Centre for competence – Clean technologies for sustainable environment – waters, wastes and energy where in addition candidate works.

Except in these scientific investigations the candidate works on development of the methods what are use for education of the bachelor and master degree students.

My personal opinion is that this is important contribution, what is in direct connection with teaching activity of Dr. Iotinov. That is link between scientific and teaching processes.

As a chief of master degree program "Bio business and biocontractor" the candidate take active participation in the teaching process in development of projects aimed to the bio contracting and development of the new idea in the field of the clean technologies and water management.

The candidate participates too, within the team of the laboratory of Ecological Biotechnology in the establishing and developing of the "business incubator" in partnership with several partners like Sofia water LTD and City factory for waste treatment of hard wastes and Center of competence Clean and Circle master degree program Ecological Biotechnology.

All the students had aiming education in the Faculty of Biology, as a played party membership in the incubator and now they work by managing some green technologies such as production of biogas, compost, waste water treatment and using of standards about quality and management of risks. They can be call innovative green technologies on the territory of Sofia municipality. The candidate was participate as a mentor and adviser in different lectures on bio contracting for the students organized by the different companies like AmGen, Coca Cola Hellenic, Junior Achievement Bulgaria and Cleantech Bulgaria. Dr. Iotinov have participate in organizing of every year practical exercises on Water management for the specialties Biomanagement and sustainable development and Ecology and environmental protection. The practicum is conducting in the laboratories of Sofia waste water treatment plant and the laboratory for the control of the quality of the drinking water in the treatment plant Bistrica. Except participation in these exercises the candidate is actively involved in the Summer field courses o General Hydrobiology for the bachelor degree students for specialties Biology and Biomanagement and sustainable development. He is providing the Summer field courses for the master degree student in Ecological biotechnology too.

The marked basic aspects of the water management find place in the biotechnological contract acting, in the leaded by the candidate master degree program Bio business and bio contracting in Sofia University during the work with master degree students about establishing and development of the projects for innovative circle solving for the waters. Very important part of the candidates activities is the control and management of the risk factors by the standards ISO 14001, ISO 31000, total management of the quality – ESG - Environmental, Social, and Governance.

Conclusion

On the base of research and teaching activities of Assist. prof. Dr. Ivajlo Iotinov, I thing that he positively covers the requirements of the academic position "associate professor" according to the Law of Development of the Academic Staff in the Republic of Bulgaria. The Regulations for the realization of ADASRB, and the Regulations for the terms and conditions for acquiring scientific degrees and academic positions at SU "St. Kliment Ohridski". This gives me the reason, as a member of the Scientific Jury and reviewer for the announced competition, to give a positive assessment and to recommend to the members of the Faculty Council of the Biological Faculty to vote positively for the election of assist. prof. Dr. Ivajlo Iotinov for the position - " associate professor" in professional direction 4.3. Biological Sciences (Hydrobiology – Water management).

17.11.2023

(assoc. prof. PhD Dimitar Kozuharov)