prof. Iwona Ewa Zawieja

Częstochowa, 01.06.2026

REVIEW

of the doctoral dissertation of Mr. Bhautik Dave, M.Sc. Eng.

entitled

"Degradation of natural and anthropogenic pollutant on surface water in aspect of self-cleaning phenomenon"

prepared at the Faculty of Environmental Engineering and Energy of the Silesian University of Technology in Gliwice,

carried out under the supervision of the Supervisor PhD hab. eng. Ewa Łobos-Moysa,

The review was prepared at the request of the Chairman of the Council of the Scientific Discipline of Environmental Engineering, Mining and Energy of the Silesian University of Technology in Gliwice

prof. Krzysztof Labus, dated 23.03.2026

1. Formal basis for the review

The review was made based on a contract for work No. UMC/1337/2026 of March 23, 20256 concluded between the Silesian University of Technology in Gliwice and its author.

2. Information on the education and course of scientific and professional work of M.Sc. Eng. Bhautik Dave

The Doctoral Candidate obtained the degree of engineer in 2018 from Sardar Patel University in Anand (India) and his master's degree in 2021 from Marwadi University in Gujarat (India). To my knowledge, **M.Sc. Eng. Bhautik Dave** has not previously applied for a PhD degree in the discipline of Environmental Engineering, Mining, and Energy. The Doctoral Candidate was a participant of the Doctoral School of the Silesian University of Technology in the discipline of Environmental Engineering, Mining and Energy.

Positions held:

✓ From October 2021 to March 2026. The Doctoral Candidate worked as a PhD Researcher/Project Investigator at the Silesian University of Technology in Gliwice, participating in 3 research projects as the main contractor.

✓ From January 2024 to April 2024, the Doctoral Candidate worked as a Research Intern (full-time) at Gujarat University, Ahmedabad, Gujarat, India.

✓ From April 2019 to July 2019, the PhD student held the position of Plant Supervisor and R&D Engineer (full-time) at Sheetal Cool Products Limited, Amreli, Gujarat, India.

✓ In 2018-2019, the Doctoral Candidate student took up a full-time job at Ingress Bio-solutions, Ahmedabad, Gujarat, India (startup), gaining expertise in microbiology and managerial experience.

M.Sc. Eng. Bhautik Dave is the author and co-author of several scientific publications in international journals, including Sustainability, Molecules, International Biodeterioration & Biodegradation, Desalination and Water Treatment, and the International Journal of Environmental Science and Technology. In some of these works, he appears as the first author, confirming his participation in the preparation of the research concept, conducting experiments, analyzing results, and preparing manuscripts.

In addition, **M.Sc. Eng. Bhautik Dave** is a co-author of two chapters in scientific monographs and of papers presented at national and international conferences, both oral and poster formats.

The Doctoral Candidate obtained funding for scientific research carried out as part of a subsidy for young scientists at the Department of Water and Wastewater Engineering of the Faculty of Environmental Engineering and Energy of the Silesian University of Technology, as well as for research under the IDUB (Excellence Initiative – Research University) program. He also participated in the implementation of three international research projects, such as:

1. "Enhancing biodegradation with microorganisms via a media replacement strategy", BKM grant 2024, no. 08/040/BKM24/0212
2. "Biodegradation of caffeine and nicotine, by microorganism and microbial consortia", BKM No. 08/040/BKM22/0177,
3. "Biodegradation of ingredients of cosmetics" Pro-quality grant No 32/014/SDU/10-22-3.

The Doctoral Candidate student received a distinction for the best presentation of an article on "Biodegradation and self-cleaning phenomenon in wastewater" during Young Science Beyond Borders 2023 2nd Edition, 1-2 June 2023, Krakow.

M.Sc. Eng. Bhautik Dave conducted or assisted with laboratory classes in Environmental Engineering and Biotechnology. The classes included the following subjects: chemistry, environmental chemistry, and computer science.

3. Subject matter of the doctoral dissertation

The subject of the doctoral dissertation of **M.Sc. Eng. Bhautik Dave** concerns the degradation of natural and anthropogenic pollutants in surface waters, with the particular emphasis on self-cleaning. The paper assumes that new pollutants (ECs) such as caffeine, nicotine, methylparaben (MeP) and trichlorocarbanilide (TCC) are commonly detected in the aquatic environment, exhibit diverse physicochemical properties and environmental behaviour, and pose an ecological hazard. Natural attenuation and microbial self-cleaning processes contribute to the degradation of pollutants. However, it is important to stress that these mechanisms are not sufficient to cope with the increasing load and complexity of pollutants entering water systems. Persistent dilution effects, ecological competition, and environmental variability limit the effectiveness of natural microbial degradation, leading to incomplete removal, and long-term persistence of biologically active compounds. The paper raises the problem of the natural ability of surface waters to self-clean. It was noted that, under current environmental conditions, this capacity is often insufficient. Microbial biodegradation is the primary biological mechanism for transforming organic pollutants and, under favorable conditions, can lead to significant degradation. In natural aquatic environments, however, the process of microbiological self-cleaning is passive and highly variable, resulting from dilution effects, limited nutrient availability, ecological competition, and changing physicochemical conditions. As a result, many biologically active compounds persist in surface waters despite their potential biodegradability under controlled laboratory conditions. Bacteria and fungi are complementary biological systems responsible for the degradation of pollutants. The Doctoral Candidate's research on the microbiological self-cleaning of surface waters from

pollutants through degradation aligns with current research directions, which primarily aim to protect the environment and safeguard the quality of life of future generations. In this context, this research is a development of the concept of an engineering approach to microbiological self-cleaning of surface waters, the essence of which is to intentionally enhance biologically controlled processes for degrading pollutants beyond the level of passive natural attenuation. Specialized microorganisms capable of degrading pollutants are adapted to grow in sewage matrices, enabling the degradation of pollutants using the natural nutrient content of synthetic wastewater as the sole source of growth. Microbial systems adapted in this way are systematically evaluated under increasingly complex conditions, from biodegradation experiments on a laboratory scale to integration into artificial wetland systems. This paper redefines the self-cleaning paradigm by shifting the focus from uncontrolled natural processes to intentionally designed, biologically driven strategies enabling increased, systemic removal of emergent pollutants from surface water matrices.

The quest to combine laboratory optimization with pilot-scale validation in artificial wetland systems indicates that this work sets a new research direction, grounded in a scalable, sustainable strategy for the active intensification of microbial self-cleaning, going beyond passive natural attenuation in polluted aquatic ecosystems.

The tests were carried out on two consecutive scales: a laboratory scale and a system of artificial wetlands. The Doctoral Candidate noted that the work's originality lies in the use of synthetic wastewater designed to map polluted surface water, which at the same time served as a model water system and the only source of nutrients for microorganisms growth during the research. It should be noted that the introduction of nutrient restrictions forced a direct coupling of microbial growth with pollutant metabolism, thereby increasing environmental relevance and scaling potential compared to laboratory media optimised for microbial growth. As the Doctoral Candidate emphasizes, this strategy was deliberately designed to move away from traditional laboratory media and to increase

pollutant degradation forcing microorganisms to use wastewater nutrients while metabolizing the target compounds.

The choice of the subject of the doctoral dissertation is very interesting and up-to-date. The author noted that targeted adaptation of microorganisms to synthetic wastewater enhances pollutant degradation by increasing metabolic activity using nutrients derived from wastewater.

The Doctoral Candidate redefined the concept of self-cleaning of surface waters as a deliberately designed and biologically enhanced process rather than a natural phenomenon. Such an innovative approach to the degradation of natural and anthropogenic pollutants in surface waters, grounded in terms of the phenomenon of self-cleaning, is a significant extension of current knowledge on this subject.

It is worth emphasizing not only the scientific nature of the work but also its future application, i.e., the implementation of the proposed strategy of microbiological support for the self-cleaning process.

The work concerns extremely current research areas, i.e. the improvement of self-cleaning of surface waters through the deliberate selection, adaptation, and integration of microorganisms capable of degrading caffeine, nicotine, methylparaben, and trichlorocarbanilidide, while accounting for the system's scaling from laboratory experiments to wetland configuration. About the raised research topics, it should be noted that in natural aquatic ecosystems, self-cleaning processes occur passively and are strongly limited by environmental variability, whereas this research treats self-cleaning as a deliberately designed, biologically intensified process.

4. Characteristics of the doctoral dissertation

The doctoral dissertation of **M.Sc. Eng. Bhautik Dave** presented for review, including 202 pages of typescript that contain references to 59 figures and 25 tables. The dissertation consists of a summary (in English and Polish), a table of contents, a list of

co-authored 4 publications of the Doctoral Candidate included in the dissertation, a list of other scientific activities, other scientific publications, a list of abbreviations and designations used, a theoretical part in the form of Chapter 1 entitled "Introduction", Chapter 2 entitled "Literature Review", Chapter 3 devoted to motivation, scientific justification for undertaking the topic, goals, scope and hypotheses of the work, the experimental part divided into chapters such as: Chapter 4 entitled "Research Methodology", Chapter 5 entitled "Analytical Methods and Data Validation", Chapter 6 entitled "Results and Discussion" summary in the form of Chapter 7 entitled "General Conclusions and Future Perspectives" and Chapter 8 entitled "Conclusions", a list of literature, a list of tables and figures.

In the work, the Doctoral Candidate included his key research in the form of 4 co-authored, monothematic scientific publications adequate to its topic, in the form of a review - one publication, and a research one, i.e.:

1. **Dave Bhautik***, Łobos-Moysa Ewa; *A comprehensive review of caffeine and nicotine as emerging environmental contaminants in water systems*; Architecture Civil Engineering Environment; 2025; vol. 18, no. 2; pp. 171–190; (**Ministry of Science and Higher Education: 70 points, IF: 0.4**)

2. **Dave, B.***, Łobos-Moysa, E., Kuźnik, A.; Enhancing fungal adaptation for efficient Caffeine degradation in wastewater: biomimetic approach and environmental optimization; *Desalination and Water Treatment*; 2024; 321; 1–16; (**MNiSW:100 points, IF:1.0**)

3. **Dave Bhautik***, Łobos-Moysa Ewa, Kuźnik Anna; *Nicotine degradation by Trametes versicolor: insights from diverse environmental stressors and wastewater medium*; Molecules; 2025; vol. 30, no. 12; pp. 1-20 (Art. no. 2658); (**MNiSW: 140 points, IF: 4.6**)

4. **Dave Bhautik***, Łobos-Moysa Ewa, Kuźnik Anna, Maqsood A., Appiah A. N. S., Krzeszowski S., Joshi R.; *Enhancing microbial biodegradation of PPCPs in wastewater*

via natural self-purification in a novel constructed wetland system; Sustainability; 2026; vol. 18, no. 1; pp. 1-18 (Art. no. 548); (**MNiSW: 100 points, IF: 3.9**)

Chapter 1, entitled "Introduction", provides the scientific background for the work. In this chapter, the Doctoral Candidate synthetically presented the problem of the occurrence of new-generation pollutants (ECs) in the aquatic environment, which, despite their low concentrations, can have a significant ecological and toxicological impact. The Doctoral Candidate noted that a promising process contributing to the degradation of pollutants in surface waters is microbiological self-cleaning, which, however, in natural conditions shows limited effectiveness, resulting, among other things, from environmental variability, nutrient deficiency, and ecological competition. It should be noted that in this chapter, based on the literature review, the Doctoral Candidate identified significant scientific problems and thus indicated the main research directions of the dissertation, which were confirmed in the following chapters of the thesis. The subject of this dissertation was to solve an important research problem by developing an innovative microbial self-cleaning strategy that intensifies biologically induced degradation through the deliberate use of specialized fungal and bacterial degraders adapted to the growing conditions in wastewater.

In Chapter 2, entitled "Literature Review" in subsection 2.1 The Doctoral Candidate characterised new pollutants in surface waters, accounting for their occurrence intensity and environmental significance. Then, in subchapter 2.2, the classification of target pollutants in water systems is presented, including natural and anthropogenic pollutants, and then, in the following sub-points of the dissertation, **M.Sc. Eng. Bhautik Dave** provided a detailed characterization of the pollutants analyzed in the experimental part of the work, including caffeine, nicotine, methylparaben (MeP), and trichlorocarbanilidide (TCC). In the next subsection 2.3 The Doctoral Candidate raised an important research problem concerning the limitation of natural weakening and uncontrolled microbial degradation in water systems, which resulted in a thorough

assessment of the microbiological self-cleaning process in the degradation of pollutants in terms of the mechanisms controlling it, i.e., bacterial degradation (subsection 2.4.1) and fungal degradation (subsection 2.4.2). Section 2.4.3 assesses the effectiveness of the microbial self-cleaning process, accounting for the complementarity and synergistic interactions among microbial systems. The subsection is supplemented by subsection 2.4.4, the content of which refers to the identification of limitations in the aspect of degradation involving a single micro-organism. In subsection 2.5, the Doctoral Candidate characterized artificial wetlands, assigning them the potential role as designed self-cleaning systems driven by microbiological remediation.

In subsection 2.6, the Doctoral Candidate pointed out significant challenges related to scaling microbiological biodegradation, i.e., transferring research conducted on a laboratory scale to real-world conditions and implementing it effectively while maintaining the effectiveness achieved at the experimental stage.

In subsection 2.7, summarising the literature review, a research gap concerning the discussed issue, i.e., the process of microbiological purification of surface waters from selected pollutants, is identified. Against the background of the presented issues, the significance and purposefulness of the conducted research were justified. As emphasized by the Doctoral Candidate, contaminants (EC) such as caffeine, nicotine, methylparaben (MeP), and trichlorocarbanilide (TCC) are commonly detected in aquatic environments and exhibit diverse physicochemical properties, environmental behaviour and ecological hazards. At the same time, natural attenuation and microbial self-cleaning processes show limited effectiveness in removing contaminants. The Doctoral Candidate pointed out that this phenomenon is conditioned by the increasing load of pollutants, their complexity with industrialization, the dilution effect, ecological competition, and environmental variability.

As the Doctoral Candidate pointed out, the research so far has focused mainly on isolated microbial degradation under laboratory conditions or on the use of artificial wetlands as passive treatment systems based on undefined communities of microorganisms.

An additional under-recognized issue is the lack of a controlled scaling framework that would facilitate the transfer of microbial degradation strategies from laboratory conditions to systems that maintain ecological complexity while controlling process efficiency.

The Doctoral Candidate emphasized the environmental significance of the research, noting that the experiments contributed to the development of biologically controlled self-cleaning strategies, thereby filling the gap between laboratory research on biodegradation and designed ecological treatment systems, a novelty of the dissertation.

In my opinion, the theoretical part of the dissertation is a concise and well-thought-out elaboration of issues closely related to the work's aim, the hypotheses put forward, and the proposed scope of research. By conducting a critical review of the literature, the Doctoral Candidate identified research problems that have not yet been recognized and noted that the state of knowledge concerning them needs to be expanded. The theoretical part of the dissertation is supported by 133 literature items, indicating that the PhD student has a good understanding of the dissertation's subject.

In Chapter 3 of the dissertation entitled "Research Aim, Objectives, and Scope of the Study", the Doctoral Candidate presented the premises constituting the basis for the implementation of the assumed topic of the thesis with justification. In addition, he specified the main and detailed objectives of the dissertation, research theses, and scope of work. In subsection 3.1, the motivation is that "microbial biodegradation emerges as an environmentally sustainable alternative, offering the potential for low-energy, in situ transformation of organic contaminants". Considering the above assumption, it was stated that "the effective application of microbial processes beyond laboratory conditions remains a key scientific and technological challenge, particularly for achieving sustained

and reliable contaminant degradation in nutrient-limited, dynamically changing aquatic environments".

In subsection 3.2, the Doctoral Candidate indicated that the main objective of this doctoral dissertation is to develop and experimentally validate a microbial self-purification strategy designed to enhance the biodegradation of emerging anthropogenic contaminants and surface water environments. However, in subsection 3.3, the Doctoral Candidate specified detailed research goals resulting from the adopted overarching goal, i.e.:

1. Selection and functional evaluation of microbial degraders
2. Investigation of microbial adaptation to wastewater-based growth conditions
3. Kinetic and process-level evaluation of biodegradation
4. Design and operation of an engineered constructed wetland system
5. Assessment of microbial synergy and system-level performance
6. Validation of system-scale performance and reproducibility

The author of the dissertation adopted the research hypothesis that the natural microbiological ability to self-clean polluted surface water systems is generally insufficient to effectively remove emerging pollutants under real environmental conditions, especially when nutrient resources are limited. Adequate research was conducted to confirm the hypothesis.

In addition, the Doctoral Candidate formulated the scope of work, indicating the chronology of research stages and demonstrating the interconnections and the resulting dependencies, including research at the laboratory scale and in artificial wetlands, using synthetic wastewater as a model matrix for polluted water. In the paper, special emphasis is placed on the functional performance of biodegradation and results at the system level. The experimental program was implemented in phases to reflect the increasing complexity of the system and the availability of representative microbial systems. Laboratory tests were carried out using fungal systems for contaminants such as caffeine

and nicotine, while pollutants such as methylparaben and trichlorocarbanilide were tested using bacterial consortia. Then, to evaluate microbial synergies and system-level performance under mixed-pollutant conditions, artificial wetland-scale experiments were conducted using three representative pollutants: caffeine, methylparaben, and trichlorocarbanilide.

The research methods described by the Doctoral Candidate in chapter 4, entitled "Research Methodology", allowed for the verification of the research hypothesis put forward in the thesis and are adequate to the assumed primary goal of the doctoral dissertation.

In subsection 4.1, the author has included information on the preparation and characterization of the target impurities, presenting the general characteristics of the extraction process of natural impurities (caffeine and nicotine) (subsection 4.1.1); extracted compounds (point 4.1.2), and the procedure for the preparation of solutions of anthropogenic impurities - methylparaben and trichlorocarbanilide (point 4.1.3) is also described. In subsection 4.2 The Doctoral Candidate included information on the course of experiments conducted on a laboratory scale concerning the process of biodegradation and adaptation, describing in detail the tasks concerning the adaptation and growth of fungi in the environment of natural pollutants (subsection 4.2.1); degradation of caffeine and nicotine by fungi (point 4.2.2); degradation of methylparaben (MeP) and trichlorocarbanilide (TCC) by bacteria (point 4.2.3). In subsection 4.2.4, the Doctoral Candidate defined the basic assumptions of the microbiological inoculation procedure, while in subsection 4.2.5, the principles of sampling, extraction, and purification were discussed.

In subsection 4.3, the Doctoral Candidate has defined a framework for experiments conducted with the use of pilot biomimetic artificial wetlands, taking into account such issues as: design and configuration of the artificial wetland system (subsection 4.3.1); the adaptation of micro-organisms and their growth dynamics in an

environment with mixed pollutants (point 4.3.2); inoculation of micro-organisms and operation of the system (point 4.3.3), monitoring of operational parameters (point 4.3.4); sampling and treatment of wetlands (point 4.3.5).

It should be emphasized that the presented description of the research methodology is comprehensive, reflects the appropriate phased nature of the conducted research, adheres to the accepted scope of work, and sheds light on the research issues discussed in the final part.

The next subsection 5 of the dissertation describes the analytical methods used and data validation, taking into account the review of the analytical strategy and the selection of an adequate method (subsection 5.1); Fourier transform infrared spectroscopy (FT-IR) (subsection 5.2); nuclear magnetic resonance (NMR) spectroscopy (subsection 5.3); high performance thin film chromatography (HPTLC) (subsection 5.4) and scanning electron microscopy (SEM) (subsection 5.5).

To assess the course of the research, the doctoral student used analytical methods that ensure chemical identification, accurate concentration determination, and the interpretability of results across different experimental scales and varying matrix complexity. An integrated analytical system was used to validate the effectiveness of microbial self-cleaning from laboratory systems to a pilot biomimetic wetland system. The Doctoral Candidate used complementary spectroscopic, chromatographic, and structural analyses, providing independent and mutually complementary evidence. Spectroscopic techniques were used as basic tools for confirming chemical identity and analyzing the transformation of contaminants of natural origin, i.e., caffeine and nicotine. Nuclear magnetic resonance (NMR) spectroscopy, on the other hand, was used as the main analytical technique for quantitative and qualitative validation of biodegradation processes. Qualitative NMR analysis was used to monitor changes in concentrations associated with microbial degradation in both laboratory and wetland systems. Chromatographic analysis was used to reliably determine levels of anthropogenic

contaminants in complex biological liquid systems. High-performance thin-layer chromatography (HPTLC) was selected as the primary method for monitoring methylparaben and trichlorocarbanilidide during bacterial biodegradation experiments. To support interpretation of the system's functioning at the scale of the entire system, scanning electron microscopy (SEM) was also used as a complementary structural tool. However, SEM observations were not used to infer chemical degradation mechanisms, but only to confirm the physical and biological integrity of the treatment system.

In chapter 6 of the dissertation entitled "Results and discussion" divided into three subchapters, the PhD student in subsection 6.1 analysed research issues concerning: biodegradation on a laboratory scale and microbiological adaptation, taking into account quantification and validation of extracted caffeine and nicotine using the qNMR method (subsection 6.1.1); adaptation and growth of fungi to natural contaminants (caffeine and nicotine) (point 6.1.2); the effectiveness of biodegradation of natural pollutants by fungi (point 6.1.3); the effectiveness of biodegradation of anthropogenic pollutants by bacteria (point 6.1.4); comparative degradation efficiency and interpretation at system level of fungal and bacterial systems (point 6.1.5). In the next subchapter of the dissertation 6.2, the performance of the biomimetic hydrophyte system on a pilot scale in terms of microbiological adaptation and operational stability was assessed (point 6.2.1); the efficiency of the hydrophytic system to remove contaminants (point 6.2.2); structural and biological observations on porous media, evaluation of the performance of hydrophytic self-cleaning on a pilot scale (point 6.2.4). In subsection 6.3 The Doctoral Candidate has made a thorough analysis of issues concerning: general synthesis and implications for scaling, taking into account the functional distinction between natural and anthropogenic pollutants (subsection 6.3.1); microbiological adaptation as the primary self-cleaning performance (point 6.3.2); synergistic microbial interactions and accelerated degradation kinetics (point 6.3.3); the conversion of laboratory systems to artificial wetlands (point

6.3.4); implications for the design of artificial wetlands and their future applications (point 6.3.5).

As emphasized by the Doctoral Candidate, based on the designed laboratory experiments, the mechanistic foundations of the engineering approach to the self-cleaning process were developed, identifying biological mechanisms of degradation in relation to hydraulic effects, plant interactions, and physicochemical factors. Studies were conducted using functionally matched microbial systems in relation to two different classes of natural pollutants, such as caffeine and nicotine, using fungal degradation, and anthropogenic pollutants, such as methylparaben and trichlorocarbanilidide, using a bacterial consortium. According to **M.Sc. Eng. Bhautik Dave's** research on the possibility of scaling microbiological systems by integrating them with a biomimetic system of an artificial wetland operating in a periodic mode was carried out using three biological configurations: wetland A (bacterial system), wetland B (fungal system), and wetland C (fungal-bacterial consortium), as well as a control system without inoculation. All bioaugmented wetlands have achieved stable operation after the initial acclimatization phase. Stability indicators included sustained growth of microorganisms under mixture-related stress, stable plant development (Phragmites australis), and stable pH values in the near-neutral range. According to the Doctoral Candidate, the populations of fungi and bacteria did not collapse with simultaneous exposure to caffeine, methylparaben, and trichlorocarbanilidide. The results of the research indicate that the wetland environment was conducive to maintaining the long-term viability of microorganisms. The wetland matrix has been shown to provide a stable scaffold for biofilm development and biomass retention, confirming the assumption that contaminant removal is a biologically driven self-cleaning process rather than a dominant adsorption mechanism.

The Doctoral Candidate noted that self-cleaning is not a single process, but an emergent property resulting from the interactions among the adaptation of microorganisms, the chemical properties of pollutants, and the structure of the

environment. It was found that the mechanisms of microbial self-cleaning can be scaled, i.e., it is possible to move from laboratory systems to wetlands, provided that the proper spatial structure, biomass retention, and limited nutrient availability are ensured. Effective biodegradation of the studied pollutants, supported by synthetic wastewater, does not require supplementation with external nutrient.

It is worth emphasizing that both the research methodology and the research stages are consistent and have been correctly defined by the Doctoral Candidate, in accordance with the dissertation's assumed goal. The results of the research were presented logically, and the tables and figures, which are an integral part of the dissertation, facilitated their proper interpretation and evaluation. It is particularly important that the research results confirmed the thesis of the doctoral dissertation, indicating that the processes of self-cleaning of surface waters can be effectively enhanced by designing microbiological systems adapted to the characteristics of pollutants, rather than relying solely on passive natural attenuation.

In Chapter 7 of the dissertation, the Doctoral Candidate consolidated the general conclusions and the resulting prospects for further development by formulating experimental conclusions (subsection 7.1), taking into account the assessment of microbiological biodegradation on a laboratory scale (subsection 7.1.1) and the assessment of the efficiency of microbial biodegradation on a pilot scale (subsection 7.1.2). In subsection 7.2, the Doctoral Candidate defined the key conclusions, while in subsection 7.3, he emphasized the importance of the research and its contribution to current knowledge.

On a laboratory scale, the research has unequivocally demonstrated the diverse functional roles of fungal and bacterial systems. The great usefulness of fungi in the long-term degradation of biologically active natural pollutants has been demonstrated.

Bacterial experiments, on the other hand, showed high kinetic efficiency and metabolic resistance of bacterial consortia to anthropogenic pollution. Synthetic wastewater, on the

other hand, provided degradation efficiency comparable to or higher than nutrient-rich media after a period of microbial adaptation. The doctoral student showed that nutrient restriction can stimulate the use of pollutants as metabolic substrates rather than inhibit them. An important contribution of the Doctoral Candidate to the development of science is the formulation of a conclusion that undermines the assumption that clean laboratory media are an optimal environment for biodegradation. The Doctoral Candidate emphasized the importance of systems based on sewage matrices.

The Doctoral Candidate showed that biomimetic artificial wetlands can be effective systems for self-cleaning surface waters. The engineering configuration used ensured biomass retention, spatial organization, and stabilization of environmental conditions, enabling long-term microorganisms activity.

Contaminated surface water can serve as active, biodegradable matrices rather than merely passive carriers of pollutants.

As indicated by the Doctoral Candidate, the precise principles of designing microbiological systems and the concept of bioaugmented wetlands form the basis for implementing the studied technology, which is a sustainable tool for protecting and reclaiming surface waters.

Chapter 8, "Conclusions," contains two pages of text. Based on the research results, the Doctoral Candidate demonstrated the validity of the research hypothesis and formulated 9 conclusions with a high degree of detail.

In terms of editing, the dissertation does not raise any objections. When evaluating the style of work, it is linguistically appropriate and consistent; I consider any errors and inaccuracies minor and do not affect its overall assessment.

5. General substantive evaluation of the doctoral dissertation

There is a close correlation between the analyses carried out that penetrate the essence of the process of microbiological self-purification of surface waters, leading to

an increase in the biodegradation of anthropogenic pollutants in the surface water environment, and their practical use, which, in my opinion, is a significant contribution of the Doctoral Candidate to the development of science. **M.Sc. Eng. Bhautik Dave** is a researcher who can plan an experiment, conduct the research, and correctly analyze the results, presenting them in a highly academic manner. It is worth emphasizing that the Doctoral Candidate achieved the thesis goal by confirming the research hypothesis.

The conducted research and the results obtained contributed to a better understanding of the mechanisms of transformation of selected micropollutants of natural and anthropogenic origin in the aquatic environment, in the context of natural processes of self-cleaning of surface waters. The Doctoral Candidate evaluated the effectiveness of removing various groups of micropollutants, characterized by different persistence in the aquatic environment, using selected and adapted fungi and bacteria. He achieved the assumed goal of the dissertation, which was to determine the possibility of biodegradation of four organic compounds occurring in surface waters, such as caffeine, nicotine, methylparaben, and t trichlorocarbanilidide in the process of microbiological self-purification of surface waters.

The Doctoral Candidate demonstrated that there is a possibility of dedicated strengthening of microbial communities involved in the self-purification of surface waters, through the selection, adaptation, and integration of fungal and bacterial-specific degraders, thereby enhancing the effectiveness of biodegradation of anthropogenic pollutants beyond natural suppression.

This goal was achieved by integrating the adapted microbial degraders into an artificial wetland platform and systematically evaluating their degradation performance across scales, from laboratory studies to a pilot treatment system.

Such adaptation enables the degradation of pollutants by using the internal nutrient content of wastewater as the sole source of growth, eliminating the need for external nutrient supplementation.

In addition, the Doctoral Candidate demonstrated that:

1. The targeted enhancement of microbial self-purification processes leads to more efficient removal of selected contaminants compared to natural attenuation processes.

2. The white-rot fungus *Trametes versicolor* demonstrated high efficiency in the biodegradation of natural alkaloids under nutrient-limited conditions, achieving more than 97–98% removal of caffeine and nicotine under optimal environmental conditions.

3. Bacterial consortia (*Alcaligenes–Rhodococcus* and *Pseudomonas aeruginosa*) exhibited high tolerance to anthropogenic contaminants and the ability to effectively degrade compounds such as trichlorocarbanilidide and methylparaben.

4. The integration of fungal and bacterial systems within a single treatment system resulted in complementary metabolic activity, enabling more effective removal of contaminant mixtures with different chemical structures.

5. The composition and availability of nutrients in synthetic wastewater can stimulate biodegradation processes, as the limitation of easily assimilable carbon sources promotes the utilization of contaminants as metabolic substrates.

6. Temperature and other environmental parameters significantly influence biodegradation efficiency, with fungal activity being particularly sensitive to increases in temperature above the mesophilic range.

7. Biomimetic constructed wetland systems planted with Phragmites australis enable stable functioning of contaminant-degrading microorganisms and effective removal of the studied compounds under environmentally relevant conditions.

8. Scaling the system from laboratory experiments to a wetland configuration confirmed the feasibility of practical implementation of the proposed strategy for microbial enhancement of self-purification processes.

9. The developed concept of bioaugmented biomimetic wetlands represents a promising, low-energy and scalable solution for supporting the treatment of

contaminated surface waters and wastewater containing mixtures of anthropogenic pollutants.

The doctoral dissertation provides systematic, multi-scale evidence that functional complementarity among groups of microorganisms increases the efficiency of purifying mixed contaminants.

The Doctoral Candidate demonstrated that polluted wastewater can simultaneously serve as a nutrient source and a driver of degradation. By using the internal nutrient content of wastewater, microbial systems can be designed to support both microbial growth and contaminant removal without relying on external nutrient supplementation. This approach directly supports the development of cost-effective, energy-efficient, and environmentally sustainable strategies for treating high-concentration wastewater streams.

The successful transition from laboratory experiments to a hydrophytic system demonstrates that increasing the effectiveness of microbial self-cleaning is not limited to controlled laboratory systems but can be implemented in porous media, plant-related environments, and mixed contaminant conditions. This is an important aspect of the research, combining the biodegradation mechanism of the investigated pollutants with the practical application of treatment systems based on hydrophyte technologies.

To sum up, the dissertation by **M.Sc. Eng. Bhautik Dave** makes a significant contribution to both the development of mechanistic knowledge and its application, indicating that it is possible to use microbiological self-treatment tailored to wastewater specificity as a scalable, low-energy, and biologically controlled strategy for cleaning the aquatic environment.

The dissertation presents an original solution to a scientific problem, and the Doctoral Candidate has acquired the theoretical knowledge necessary to carry out the planned research task, which is of significant cognitive and utilitarian importance. It is worth emphasizing the fact that **M.Sc. Eng. Bhautik Dave** conducted a wide range of

research that required the implementation of various methodological and analytical tools, demonstrating the Doctoral Candidate's high research activity and scientific independence.

6. Critical remarks

Based on the dissertation, several polemical questions and remarks were formulated. However, they do not affect the final assessment of the dissertation's scientific value.

The main criticisms are directed to subsection 3.2, entitled: "Aim of the Study". In my opinion, it would be advisable for the Doctoral Candidate to consider a clear formulation of both the scientific and utilitarian goals, which come to mind after reading the point. Formulating the scientific and utilitarian goal would allow the establishment of adequate research hypotheses.

Subsection 3.3 entitled "Specific Research Objectives" formulates specific objectives which, in my opinion, should be reflected in the scope of work in the form of points or, which would make it much easier to interpret the dissertation, in graphic form.

In addition, the Doctoral Candidate indicated that in the dissertation he included his key research in the form of publications appropriate to the work's topic, i.e., 4 co-authored papers, including one in the form of a review, and the remaining research papers. It would be worth considering marking the graphic interpretation of the dissertation's scope, at what stage they were cited, and what issues they were concerned with. Such an approach at the beginning of the work would make it much easier to interpret, making it more transparent.

The author did not place the first scientific publication listed in the given series in the form of an unambiguous reference in the content of the dissertation. However, I conclude from the article that the information it contains was cited in the literature section of the dissertation.

It would be helpful in the evaluation process to provide the percentage of the Doctoral Candidate in the preparation of scientific articles that have been identified as crucial.

The Doctoral Candidate indicated that the efficiency of microbiological degradation observed on a laboratory scale can be transferred to the operation of a hydrophyte system using synthetic wastewater as a model matrix for polluted water, thereby enabling the creation of a repeatable framework for biological self-cleaning of polluted water systems. – whether verification of the system with the use of real wastewater was considered, e.g., for individual pollutants or their mixture.

Based on the conducted research, cognitive conclusions were formulated that significantly expanded the current state of knowledge regarding the discussed issue and demonstrated a significant application potential. Please specify the degree of readiness of the proposed solution for practical implementation, and whether there is potential to use the studied technology for self-cleaning, not only in standing water but also in flowing water. What challenges or technological barriers can occur during the implementation phase?

In the doctoral thesis, it is worth including a point on the legal conditions governing the occurrence of the studied pollutants in surface waters, even if such standards do not formally exist. The legal context would indicate that the research is embedded in a real environmental and legislative problem. The item on legislative considerations would highlight the regulatory gap, the topicality of the research problem, the significance of the results, and the conclusions formulated for future environmental regulations.

In my opinion, it is not correct to specify one sub-section for a subsection in the table of contents and adequately the content of the dissertation, which has been stated three times in the table of contents. I suggest incorporating the subsection's content into the parent subsection.

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In the case of Figures 1, 2, 4, 5, 7, and 8, there is no information regarding the information I have prepared myself, and no indication of possible literature sources on which the graphics were created, which is important in the case of copyright. Similarly, there are no literature references in the case of Table 1.

The dissertation did not use the numbering of the equations introduced on pages 57, 90, 113, 118, and 128, nor the relevant literature sources.

Other editorial and stylistic errors I noted in the work do not diminish its value, so I did not include them in the review.

7. Final conclusions

To sum up, I consider the doctoral dissertation presented for review by **M.Sc. Eng. Bhautik Dave** as very valuable. The Doctoral Candidate has demonstrated very good knowledge of research tools and innovation in the approach to the assumed research goal. The work contains elements of novelty and constitutes a significant extension of research on the degradation of natural and anthropogenic pollutants in surface waters, focusing on the phenomenon of self-cleaning. The dissertation presented for review is not devoid of errors and inaccuracies, but they are within the limits acceptable to the researcher's workshop and do not constitute cardinal errors, thus not affecting the general, very good evaluation of the work.

Summarizing the evaluation of the doctoral dissertation presented for review by **M.Sc. Eng. Bhautik Dave,** I state that it is an original achievement of a cognitive and utilitarian nature. The dissertation provides new insights into the potential for microbiological self-purification of surface waters by integratimg of functionally complementary fungal and bacterial degraders to degrade newly emerging pollutants.

The applicative nature of the work, the literature review, its implementation requiring research taking into account the transition from controlled experiments of microbial biodegradation on a laboratory scale to a biomimetic system of artificial wetlands on a

pilot scale, as well as the correct interpretation of the obtained research results, indicate very good scientific competence of the Doctoral Candidate and independence as a researcher. Therefore, I believe that the doctoral dissertation of **M.Sc. Eng. Bhautik Dave**, entitled "Degradation of natural and anthropogenic pollutant on surface water in aspect of self-cleaning phenomenon", presented for review, fully meets the requirements for doctoral dissertations set out in Article 187 of the Act of 20 July 2018 on the Law on Higher Education and Science (Journal of Laws of 2024, item 1571, as amended). Thus, I apply to the Council of the Scientific Discipline of Environmental Engineering, Mining and Energy of the Silesian University of Technology in Gliwice to allow it to take further steps in the procedure for awarding the scientific degree of Doctor of Engineering and Technical Sciences in the discipline of Environmental Engineering, Mining and Energy, and distinction.

Podpisała Iwona Zawieja

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