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Design and Implementation of an Intelligent Sphingan Bioreactor Architecture Based on Cyber-Physical Fusion

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Abstract: Aiming at the problems of low degrading bacteria activity, difficult demulsification of emulsified oil, extensive process regulation, and weak working condition adaptability in traditional bioreactors for ship engine room oily wastewater, an intelligent bioreactor scheme integrating Cyber-Physical System (CPS), *Sphingomonas*-produced sphingan, and Microbial Electro-Fenton (MEF) coupled degradation is proposed. This reactor takes *Sphingomonas* as the core high-efficiency degrading strain, and uses sphingan biosurfactant produced by its metabolism to realize in-situ demulsification, solubilization and high-efficiency degradation of emulsified oil. A four-layer CPS closed-loop architecture of perception-transmission-decision-execution is adopted to regulate strain growth, gum production efficiency and MEF degradation working conditions in real time, coupling the first-stage Microbial Fuel Cell (MFC) biodegradation and the second-stage Electro-Fenton (EF) advanced oxidation process. At the hardware level, the integration of *Sphingomonas* culture module, MEF reaction unit and CPS perception control hardware is completed; at the software level, modules for strain activity monitoring, gum production optimization and intelligent regulation of degradation process are developed. Performance test results show that the sphingan production of *Sphingomonas* in the reactor is stable, the demulsification rate of emulsified oil reaches 91.8%, the total oil pollution degradation rate reaches 93.42%, and the COD removal rate reaches 90.8%. The reactor can adapt to water quality fluctuations and achieve stable discharge up to standard.

Keywords: Cyber-Physical Fusion; *Sphingomonas*; Sphingan; Intelligent Bioreactor; Microbial Electro-Fenton; Oily Wastewater Treatment.

1. Introduction

The rapid development of the shipping industry has led to a continuous increase in the discharge of oily wastewater from ship engine rooms. Such wastewater is mainly contaminated with emulsified oil, characterized by high oil-water interfacial tension, stable emulsified systems, poor biodegradability, and great difficulty in treatment [1]. Traditional physical separation methods exhibit limited removal efficiency for emulsified oil; chemical demulsification tends to cause secondary pollution; and conventional biological methods struggle to achieve rapid degradation of emulsified oil due to the lack of efficient demulsification approaches [4].

Sphingomonas is a functional microorganism that can efficiently degrade petroleum hydrocarbons while metabolically producing sphingan gum, an acidic exopolysaccharide. As a natural biosurfactant, sphingan gum enables in-situ demulsification, solubilization and dispersion of emulsified oil, significantly improving the bioavailability of emulsified oil and solving the core problem of its recalcitrance to degradation [6]. Furthermore, Gao et al. found that sphingan gum maintains relatively stable viscoelasticity under the action of sodium ions, calcium ions, high temperature and long-term heating [8]. Cyber-Physical Systems (CPS) enable real-time perception, intelligent decision-making and precise regulation of bioreaction processes, addressing the issue that strain activity and gum production cannot be dynamically optimized in traditional bioreactors [10]. Microbial Electro-Fenton (MEF) technology achieves synergistic enhancement of biodegradation and advanced oxidation, further mineralizing refractory organic matter [12].

Against the backdrop of Industry 4.0, foreign countries have initiated explorations into the integration of biomanufacturing and intelligent manufacturing. Siemens of Germany proposed the concept of "digital twin for biomanufacturing" and realized mechanism-model-based parameter monitoring in penicillin fermentation, yet without integrating in-situ physiological sensing (e.g., EET), still relying on off-line sampling to analyze cell status [14]. A 2025 study in Science Advances showed that in-situ wireless sensing of 15 parameters including metabolites and cell viability has been achieved, with the localization rate of high-end sensors exceeding 80%. However, the model adaptability is poor under extreme operating conditions, and gaps in mechanistic understanding result in a prediction accuracy of only 70–80% for complex systems [16]. In a review, Petzold J C et al. pointed out that AI-MPC hybrid control combined with CFD digital twin has raised the success rate of bioreactor scale-up from 500 L to 5000 L to 92%, and self-driving laboratories (SDLs) have realized autonomous operation through human-machine collaboration [18].

In contrast, research on bioreactors in China still focuses on "automation upgrading" and lags behind in intelligence. The Tianjin Institute of Industrial Biotechnology, Chinese Academy of Sciences, proposed that digital twin technology integrating CFD and machine learning can achieve precise scale-up of bioreactors from 250 mL to 2000 L, yet insufficient understanding of metabolic mechanisms limits the model prediction accuracy to only 75–85% [19].Universities including Hebei University of Science and Technology and Jiangnan University have realized constant

PID parameter control (e.g., pH, dissolved oxygen DO) during fermentation, but fail to adaptively adjust according to cellular physiological states such as cell concentration and specific growth rate [20].

Based on the core concept of bacteria-gum synergistic degradation, this paper takes *Sphingomonas* as the core functional strain and sphingan (its metabolite) as the demulsification synergist, integrates CPS intelligent regulation and MEF coupled degradation technology, designs and implements an intelligent *Sphingomonas* bioreactor. Through CPS closed-loop regulation of strain growth, gum production efficiency and degradation process, it realizes efficient, intelligent and green treatment of ship engine room oily wastewater.

2. Architecture Design and Implementation of Intelligent Bioreactor

2.1. Related Technical Basis

2.1.1. Core Architecture of Cyber-Physical Fusion (CPS)

Cyber-Physical System (CPS) is an intelligent system deeply integrating computing, communication and physical processes, realizing intelligent control of physical objects through a four-layer closed-loop architecture[22]. The perception layer collects physical entity state data in real time, the transmission layer ensures reliable transmission of perception data and control instructions, the decision layer completes data analysis and optimal decision-making based on models and algorithms, and the execution layer accurately executes regulation instructions and feeds back to the physical process. This architecture can realize autonomous perception, autonomous decision-making and autonomous regulation of wastewater treatment processes, replacing traditional manual experience-based operation and maintenance, and promoting the operation of wastewater treatment systems towards autonomy and intelligence.

2.1.2. Gum Production Characteristics of Sphingomonas and Bacteria-Gum Synergistic Degradation Mechanism

Sphingomonas is a Gram-negative bacterium with salt tolerance and broad-spectrum petroleum hydrocarbon degradation ability, adapting to the high-salt complex environment of ship oily wastewater [23]. Sphingan produced by its metabolism is polymerized from monosaccharides such as glucose and glucuronic acid, with both hydrophilic and hydrophobic groups, making it a typical biosurfactant. Its mechanism of action includes: reducing oil-water interfacial tension to destroy the stable system of emulsified oil; solubilizing and dispersing hydrophobic petroleum hydrocarbons; forming a protective layer on the bacterial surface to improve strain tolerance [24].

The strain can realize a synergistic cycle of self-gum production self-demulsification self-degradation: growing with petroleum hydrocarbons as carbon source to secreting sphingan for in-situ demulsification → improving pollutant bioavailability → efficient degradation and continuous gum production, fundamentally solving the bottleneck of difficult degradation of emulsified oil.

2.1.3. Microbial Electro-Fenton (MEF) Coupled Degradation Mechanism

The MEF system is coupled by first-stage MFC biodegradation and second-stage EF advanced oxidation[25]. The MFC unit catalyzes the oxidation of organic matter and recovers electric energy with electroactive microorganisms; the EF unit deeply mineralizes refractory organic matter through hydroxyl radicals (·OH), combining the advantages of green and low consumption of biological methods and thorough degradation of advanced oxidation methods, and can form functional complementarity with the *Sphingomonas* gum production system.

2.2. Design of CPS-Based Intelligent Sphingan Bioreactor Architecture

The reactor takes the four-layer closed-loop architecture of CPS as the core, *Sphingomonas* gum production-demulsification-degradation as the biological core, and MEF as the advanced treatment process to construct an integrated intelligent treatment system, as shown in Figure 1.

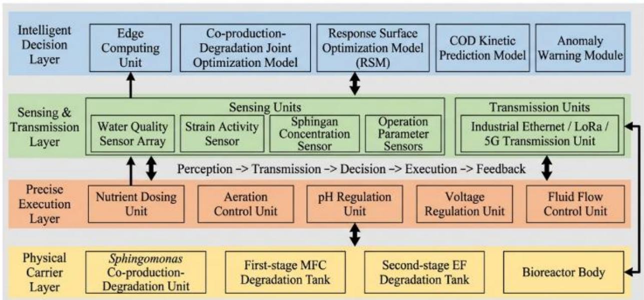


Figure 1. Four-layer closed-loop architecture of CPS-based intelligent *Sphingomonas* bioreactor

2.2.1. Physical Carrier Layer: Core Reaction and Execution Units

The physical carrier layer is the physical entity foundation of the intelligent bioreactor, undertaking core functions such as microbial degradation, electro-Fenton oxidation and water flow transmission, forming a direct carrier for the interaction between digital space and physical space.

The immobilized flora is *Sphingomonas*, fixed through multiple mechanisms such as physical adsorption and spatial embedding, providing space for strain growth, gum production and degradation, equipped with temperature control, dissolved oxygen and nutrient salt regulation devices.

The MEF coupled degradation reaction module consists of two parts. The first-stage MFC degradation tank adopts a double-chamber continuous flow structure, with an effective volume of 50 mL for each anode and cathode chamber, separated by a Nafion-117 proton exchange membrane (10 cm×10 cm). The anode is a WC-CC composite electrode loaded with sphingan (tungsten carbide modified carbon cloth + biofilm), and the cathode is a carbon brush (3 cm×3 cm). The catholyte is a mixed solution of potassium ferricyanide-potassium chloride (90 mM / 60 g/L), with a maximum output voltage of 662 mV. The emulsified oil degradation rate of this unit is ≥80.95% and COD removal rate is ≥79.1% within 7 days, while supplying part of electric energy for the second-stage EF reaction. The second-stage EF degradation tank is a single-chamber continuous flow structure with an effective volume of 1 L (10 cm×10 cm×15 cm), equipped with a magnetic stirring device at the bottom. The anode is a carbon rod, and the cathode is a foam iron-nickel modified electrode prepared by electrodeposition, enhancing in-situ generation of H_2O_2 and ·OH yield. Relying on MFC power generation supplemented by an external power supply, it deeply mineralizes refractory organic matter.

The main cavity of the reactor is made of polycarbonate

(PC), and the pipeline is made of PVC anti-corrosion material. Upflow water inlet design is adopted, integrating precipitation zone, diversion zone and reaction zone. The basic effective volume is 0.3 m^3, with a daily treatment capacity of 0.4 m^3; it can be expanded to a combination of 4 groups of MFC + 1 group of EF, with the daily treatment capacity increased to 1.6 m^3.

2.2.2. Perception and Transmission Layer: Data Collection and Communication

As the nerve ending of the CPS architecture, the perception and transmission layer collects full-dimensional parameters in real time and realizes low-latency and high-reliability data transmission. The perception unit adopts a distributed sensor network, with all water quality sensors having accuracy ≤±2% and response time ≤10 s.

The transmission unit adopts local and remote dual-mode transmission: local adopts industrial Ethernet (Modbus-RTU) and RS485 bus, with distance ≤1 km and delay ≤50 ms; remote adopts LoRa (≤5 km) and 5G, supporting cloud monitoring. Encryption, redundant transmission and breakpoint resume technology are adopted, with data transmission reliability ≥99.9%.

2.2.3. Intelligent Decision Layer: Core Algorithms and Control

The intelligent decision layer takes the edge computing module as the core, equipped with three models of process parameter optimization, dynamic prediction and anomaly diagnosis, realizing autonomous analysis and precise regulation.

The core algorithm models are divided into three parts:

(1) Process Parameter Response Surface Optimization Model

Based on Box-Behnken design and Response Surface Methodology (RSM), with real-time COD, oil content, pH and temperature as inputs, and maximizing emulsified oil and COD removal rates and minimizing energy consumption as objectives, the optimal parameters are obtained: electrolysis voltage 8.47 V, pH 3.12, predicted degradation rate 94.58%. The model fitting degree $R^2 \geq 0.9251$, prediction error ≤1.8%.

(2) COD Degradation Kinetic Prediction Model

Based on the first-order reaction kinetic equation:

$$lnC = -kt + lnC_0 \quad (1)$$

where k is the reaction rate constant, the model determination coefficient $R^2=0.9908$. It can predict effluent COD in real time and adjust operating parameters in advance. The prediction error of three groups of test water samples is ≤1.8%.

(3) Anomaly Diagnosis and Early Warning Module

A dual early warning mechanism of threshold and trend is established. When COD/oil content exceeds the threshold, electrode short circuit, biofilm shedding or MFC power generation abnormality occurs, sound and light alarm and cloud push are triggered, and emergency strategies are automatically implemented: increasing voltage, supplementing flora or adjusting HRT.

2.2.4. Precise Execution Layer: Closed-Loop Regulation Execution Unit

The precise execution layer receives instructions from the decision layer, and performs millisecond-level precise execution of electrical, fluid and physical and chemical parameters to realize closed-loop regulation.

This layer executes four types of regulation operations: microbial metabolism regulation (nutrient salt, dissolved oxygen, temperature); electrical parameter regulation (electrolysis voltage); fluid parameter regulation (flow rate); physical and chemical environment regulation (pH), with instruction response time ≤100 ms.

2.2.5. Overall Closed-Loop Synergy Mechanism

The four-layer CPS-based architecture forms a closed loop of "perception → transmission → decision → xecution → feedback": parameters collected by the perception unit are sent to the edge computing module through the transmission layer; the decision layer generates instructions through optimization and dynamic models; the execution layer quickly adjusts voltage, pH and other parameters; the changes after execution are perceived again to realize iterative optimization.

2.3. System Implementation

2.3.1. Hardware Implementation

Sphingomonas culture includes constant temperature fermenter, aseptic operating table, temperature control module and nutrient salt dosing pump, realizing strain domestication, expansion and gum production regulation; the perception unit integrates core detection equipment such as ultraviolet spectrophotometer, multi-parameter water quality detector and conductivity meter to ensure the reliability and accuracy of monitoring data; the control module adopts an integrated architecture of embedded controller and edge computing module, realizing local real-time regulation and data preprocessing, and the supporting cloud management platform completes remote monitoring, data storage and instruction issuance, forming a "local-cloud" collaborative control mode.

2.3.2. Software Implementation

The data collection system is designed based on modularization, collecting monitoring data of various sensors in real time, purifying data through preprocessing processes such as outlier elimination and data normalization, and completing local storage and visual display synchronously, providing high-quality data support for subsequent analysis and decision-making. The intelligent algorithm system integrates response surface optimization algorithm, pollutant degradation kinetic prediction model and adaptive regulation algorithm, realizing dynamic optimization of key parameters of degradation process and precise control of reaction efficiency. The human-computer interaction system has functions such as custom parameter setting, real-time operation status monitoring, historical data export and abnormal working condition alarm, with a simple and intuitive operation interface, improving system usability and operation and maintenance convenience.

2.3.3. Process Flow Implementation

The system adopts a stepped treatment process, with the specific flow as follows: pretreatment → bacteria-gum synergistic demulsification degradation → MEF coupled oxidation → advanced precipitation → discharge up to standard. In the pretreatment stage, mechanical impurities in wastewater are removed by gravity sedimentation technology to reduce the load of subsequent treatment units; the first-stage degradation unit adopts the synergistic effect of bacteria-gum synergistic demulsification degradation and Microbial Fuel Cell (MFC) to efficiently degrade emulsified oil and various organic pollutants; the second-stage degradation unit uses the strong oxidation technology of Electro-Fenton (MEF) to deeply remove residual refractory pollutants; the final effluent after advanced precipitation treatment meets the requirements of Ship Pollutant Discharge Standard (GB 3552-2018) and Class III standard of Urban Sewage Treatment Plant Pollutant Discharge Standard (GB 18918-2002), realizing stable discharge up to standard, as shown in Figure 2.

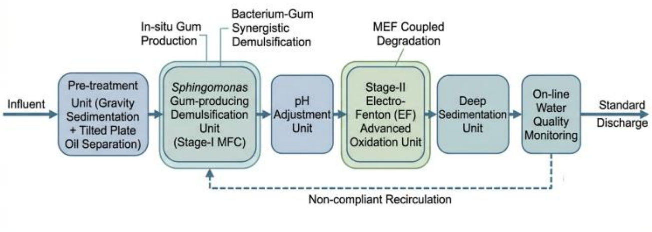


Figure 2. Schematic diagram of the process flow of the intelligent *Sphingomonas* bioreactor

2.4. Performance Test and Analysis

The experimental water is oily wastewater from the engine room of a supply ship in Nanshan Port, with oil content 102±10 mg/L and COD 540±50 mg/L; functional strain: *Sphingomonas*; detection methods: oil content (HJ 637-2018), COD (HJ 828-2017), sphingan (GB/T 35818-2018), demulsification rate (GB/T 7305-2018).

2.4.1. Analysis of Gum Production and Demulsification Performance of *Sphingomonas*

The gum production capacity and demulsification effect of *Sphingomonas* are the core foundation for the efficient operation of the reactor. The gum production performance and demulsification capacity of the strain were tested by regulating the temperature, dissolved oxygen, nutrient ratio and other conditions in the reactor.

As shown in Figure 3, quantitative test results show that under the optimal operating environment of the reactor, the maximum gum production of *Sphingomonas* can reach 1.78 g/L, and the gum production during stable operation is maintained at 1.55~1.75 g/L; the demulsification rate of stable emulsified oil in ship oily wastewater is as high as 91.8%, which can quickly destroy the oil-water emulsification system, convert emulsified oil into free oil, and greatly improve the bioavailability of pollutants, laying a foundation for subsequent degradation.

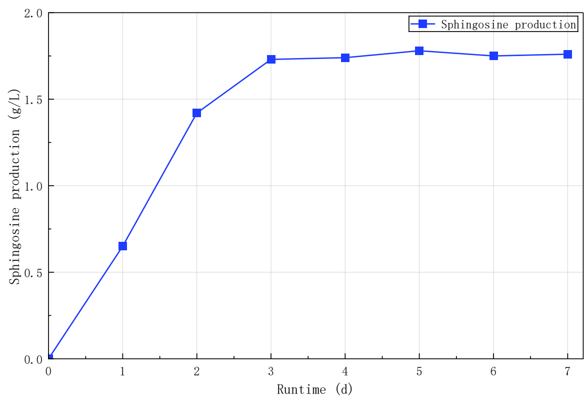


Figure 3. Curve of sphingan production in the reactor with operating time

2.4.2. Analysis of Degradation Performance of Reactor Units

The intelligent bioreactor adopts a two-stage series coupling design of bacteria-gum synergistic degradation + electro-Fenton advanced oxidation, and a joint degradation experiment was carried out with ship engine room oily wastewater as the treatment object. The first-stage unit takes *Sphingomonas* as the core, realizing primary degradation through in-situ gum production, demulsification and solubilization, with hydraulic retention time of 7 days, 7-day emulsified oil degradation rate of 80.95%, COD removal rate of 79.1%, and maximum power generation voltage of 662 mV, realizing synergistic degradation and power generation. The second-stage unit operates under the conditions of electrolysis voltage 8 V and pH 3.0, and deeply mineralizes residual organic matter through strong oxidizing hydroxyl radicals. As shown in Figure 4, after the two-stage units operate in series, the total emulsified oil degradation rate of the system reaches 93.42%, and the total COD removal rate reaches 90.8%, fully verifying the scientificity and high efficiency and feasibility of the two-stage coupling design of the reactor.

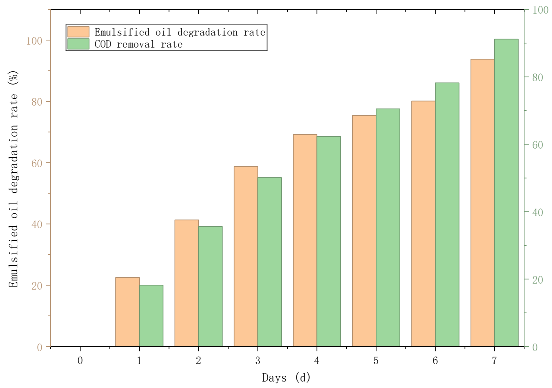
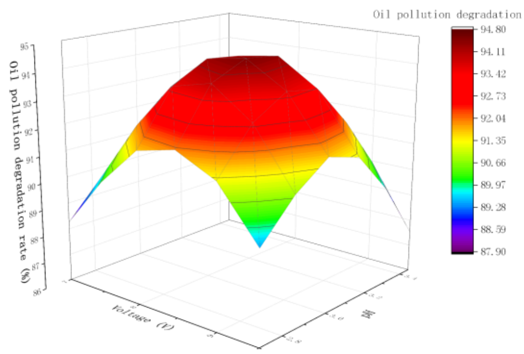
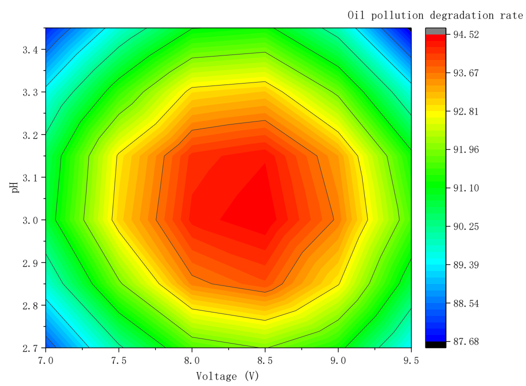


Figure 4. Curve of emulsified oil degradation rate/COD removal rate in the reactor with time

2.4.3. System Process Optimization and Degradation Kinetics Analysis



(a) 3D surface plot of the effect of voltage and pH on oil degradation rate



(b) Contour plot of the effect of voltage and pH on oil degradation rate

Figure 5. 3D surface and contour plots of the effect of voltage and pH on oil degradation rate

To determine the overall optimal operating process of the reactor, Response Surface Methodology was used to optimize the two key parameters of electrolysis voltage and pH. The influence of the interaction between voltage and pH on oil degradation rate is shown in Figure 5.

2.4.4. Overall System Performance and Effluent Compliance Verification

The core performance of the self-developed intelligent bioreactor was quantitatively compared with that of the traditional bioreactor, and the data are shown in Table 1.

Table 1. Comparison of treatment performance between intelligent reactor and traditional bioreactor

Detection Index	Traditional Bioreactor	Intelligent Reactor	Increase/Decrease Amplitude
Emulsified oil demulsification rate/%	36.2	91.8	Increased by 55.6%
Emulsified oil degradation rate/%	66.8	93.42	Increased by 26.62%
COD removal rate/%	45.73	90.8	Increased by 45.07%

The results show that the intelligent bioreactor developed in this paper is significantly superior to the traditional reactor in all performances: emulsified oil demulsification rate increased by 55.6%, emulsified oil degradation rate increased by 26.62%, COD removal rate increased by 45.07%, degradation cycle shortened by 50%, and the comprehensive treatment efficiency and operational stability are greatly improved.

The final effluent quality of the reactor was tested and compared with the Ship Water Pollutant Discharge Control Standard (GB 3552-2018), and the results are shown in Table 2.

Table 2. Comparison of reactor effluent quality and national standards

Detection Index	Reactor Effluent	GB 3552-2018	Compliance
Oil content/($mg \cdot L^{-1}$)	12.9	≤15	Compliant
COD/($mg \cdot L^{-1}$)	48.2	≤125	Compliant
pH	7.1	6~9	Compliant

The results show that all indicators of the reactor effluent meet the national discharge standards and can be directly discharged, fully adapting to the engineering application requirements of ship engine room oily wastewater treatment.

3. Conclusion

This paper designs and implements an intelligent *Sphingomonas* bioreactor based on cyber-physical fusion. The core innovation is that *Sphingomonas* produces sphingan in situ, and bacteria-gum synergistic demulsification and degradation completely change the traditional carrier fixation idea and solve the pain point of difficult degradation of ship emulsified oil:

(1)Taking *Sphingomonas* as the core functional strain, it can efficiently metabolize and produce sphingan, with a maximum gum production of 1.78 g/L and an emulsified oil demulsification rate of 91.8%;

(2)As a biosurfactant, sphingan breaks the emulsified oil system in situ, shortening the degradation cycle by 50% and greatly improving the degradation efficiency;

(3)The CPS four-layer closed-loop architecture realizes dynamic intelligent regulation of strain activity, gum production and degradation process, with strong system adaptability;

(4)Combined with MEF coupled degradation process, the total oil pollution degradation rate is 93.42%, COD removal rate is 90.8%, and the effluent is stably up to standard.

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