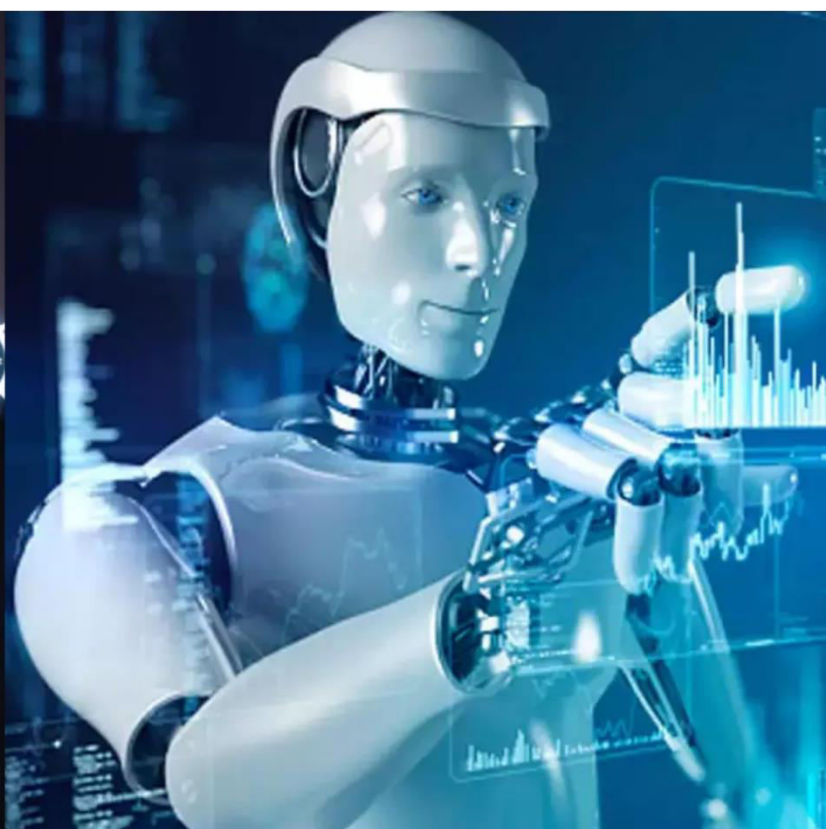




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Experimental Investigation of Operational Parameters Affecting Pathogen Removal in Sequencing Batch Reactor (SBR)

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ABSTRACT: Increasing wastewater generation, microbial contamination, and the growing demand for safe water reuse have created a critical need for advanced and sustainable wastewater treatment technologies. Conventional biological treatment systems are primarily designed for organic pollutant removal and often exhibit inadequate pathogen removal efficiency under fluctuating operational conditions. Sequencing Batch Reactor (SBR) technology has emerged as a promising biological treatment system due to its operational flexibility, compact configuration, and enhanced treatment performance. The present study experimentally investigates the influence of key operational parameters on pathogen removal efficiency in a lab-scale Sequencing Batch Reactor treating municipal wastewater. The effects of Hydraulic Retention Time (HRT), Dissolved Oxygen (DO), Mixed Liquor Suspended Solids (MLSS), and cycle duration were systematically evaluated under controlled laboratory conditions. Microbial indicators including *Escherichia coli* and total coliforms were analyzed along with physicochemical parameters such as Chemical Oxygen Demand (COD), pH, and dissolved oxygen.

Experimental results demonstrated significant enhancement in microbial reduction under optimized operational conditions. Maximum *E. coli* and total coliform removal efficiencies of 96.4% and 95.8%, respectively, were achieved at optimum conditions of 18 h HRT, 3 mg/L DO, 3500 mg/L MLSS, and 8 h cycle duration. COD removal efficiency reached 88.5%, indicating stable biological degradation and efficient reactor performance. Statistical optimization using Response Surface Methodology (RSM) and ANOVA confirmed the significant influence of HRT and DO on pathogen removal efficiency. Furthermore, first-order kinetic modelling successfully described microbial decay behavior within the SBR system.

The findings establish that optimized SBR operation can achieve high pathogen removal efficiency and support the application of SBR technology for sustainable wastewater treatment, decentralized sanitation systems, and safe wastewater reuse practices..

KEYWORDS: Sequencing Batch Reactor; Pathogen Removal; Wastewater Treatment; Hydraulic Retention Time; Dissolved Oxygen; MLSS; SBR Optimization; Microbial Kinetics; Water Reuse.

I. INTRODUCTION

Water pollution and microbial contamination resulting from rapid urbanization, industrialization, and population growth have become significant environmental challenges worldwide. Municipal wastewater contains high concentrations of organic pollutants, suspended solids, nutrients, and pathogenic microorganisms including *Escherichia coli*, total coliforms, viruses, and protozoa. The discharge of untreated or partially treated wastewater into natural water bodies poses severe risks to public health and aquatic ecosystems.

Conventional wastewater treatment systems are primarily designed for the removal of organic pollutants and suspended solids; however, their efficiency in pathogen removal often remains inadequate under fluctuating operational conditions. Therefore, advanced biological treatment technologies capable of achieving enhanced microbial reduction are required for sustainable wastewater management and safe water reuse applications. Sequencing Batch Reactor (SBR)

technology has emerged as an advanced biological treatment process due to its operational flexibility, compact footprint, reduced sludge generation, and ability to integrate multiple treatment stages within a single reactor system. The SBR process operates sequentially through fill, react, settle, decant, and idle phases, enabling effective treatment control and improved microbial activity. Despite its advantages, pathogen removal mechanisms in SBR systems are influenced by several operational parameters such as Hydraulic Retention Time (HRT), Dissolved Oxygen (DO), Mixed Liquor Suspended Solids (MLSS), cycle duration, pH, and temperature. Variations in these parameters significantly affect microbial growth, sludge characteristics, oxygen transfer efficiency, and pathogen inactivation mechanisms. Although extensive studies have focused on organic matter and nutrient removal in SBR systems, limited research has systematically investigated the relationship between operational parameters and pathogen removal efficiency. Therefore, the present postgraduate research focuses on the experimental investigation and optimization of operational parameters affecting pathogen removal in a laboratory-scale SBR system for sustainable wastewater treatment applications

II. RESEARCH OBJECTIVES

2.1. Aim

To experimentally investigate and optimize operational parameters affecting pathogen removal efficiency in a Sequencing Batch Reactor (SBR) system.

2.2. Objectives

- To design and operate a laboratory-scale SBR reactor for municipal wastewater treatment.
- To evaluate the effect of Hydraulic Retention Time (HRT) on pathogen removal.
- To investigate the influence of Dissolved Oxygen (DO) concentration on microbial reduction.
- To analyze the impact of MLSS concentration on pathogen adsorption and settling.
- To optimize cycle duration for enhanced treatment efficiency.
- To evaluate microbial reduction using *E. coli* and total coliform indicators.
- To develop first-order kinetic models for pathogen decay.
- To apply Response Surface Methodology (RSM) and ANOVA for optimization of operational parameters.

III. MATERIALS AND METHODOLOGY

3.1 Experimental Setup

A laboratory-scale acrylic Sequencing Batch Reactor with a working capacity of 15 L was fabricated and operated under controlled laboratory conditions.

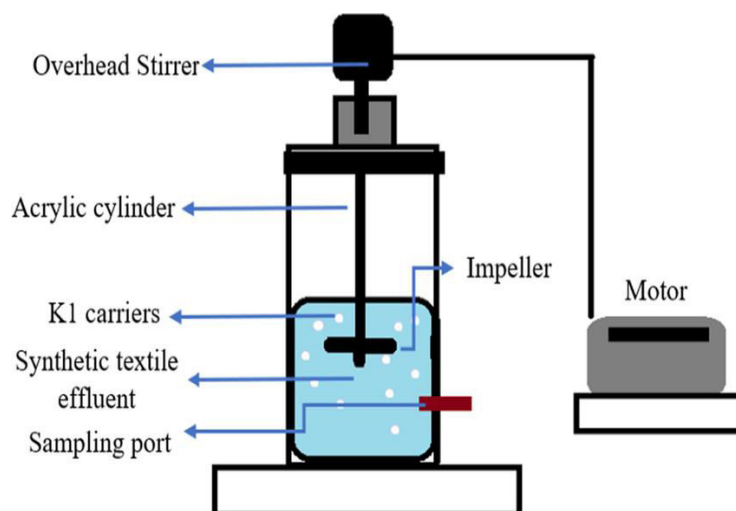


Figure 3: Laboratory-Scale Sequencing Batch Reactor Setup

3.2. Reactor Components

- Acrylic cylindrical reactor
- Air diffuser and compressor
- Sampling ports
- Decanting arrangement
- Mechanical mixing system

3.3. Wastewater Collection and Characterization

Municipal wastewater samples were collected and characterized according to APHA standard methods

Table 3: Initial Wastewater Characteristics

Parameter	Unit	Value
pH	—	7.3
COD	mg/L	540
DO	mg/L	1.2
Total Coliform	MPN/100 mL	2.8×10^6
E. coli	CFU/100 mL	1.9×10^6
Temperature	°C	28

IV. EXPERIMENTAL RESULTS & DISCUSSION

4.1 Effect of Hydraulic Retention Time (HRT) on Pathogen Removal Efficiency

Hydraulic Retention Time (HRT) is one of the most significant operational parameters influencing biological treatment efficiency and microbial reduction in Sequencing Batch Reactor (SBR) systems. HRT directly affects the contact duration between wastewater contaminants and active microbial biomass, thereby controlling substrate degradation, microbial predation, sludge floc formation, and pathogen inactivation mechanisms.

In the present study, HRT was varied from 8 h to 24 h to evaluate its influence on the removal efficiency of *Escherichia coli* and total coliforms. Experimental observations indicated a substantial increase in pathogen removal efficiency with increasing HRT up to an optimum retention period of 18 h. The enhanced removal efficiency at higher HRT can be attributed to improved microbial interaction, prolonged aeration exposure, higher adsorption of pathogens onto sludge flocs, and increased settling efficiency.

At lower HRT (8 h), insufficient contact time between microorganisms and wastewater resulted in incomplete biological stabilization and lower microbial reduction. As the HRT increased to 18 h, microbial activity became more stable, leading to enhanced biodegradation and higher pathogen removal efficiency. However, a marginal decline in removal efficiency was observed at 24 h HRT, which may be associated with reduced substrate availability and endogenous microbial respiration under prolonged retention conditions.

The results demonstrate that optimized HRT significantly improves pathogen removal performance in SBR systems and contributes to stable biological treatment conditions suitable for wastewater reuse applications.

4.1.1. Discussion of Findings

The maximum *E. coli* removal efficiency of 96.4% and total coliform removal efficiency of 95.8% were achieved at 18 h HRT. Similar findings have been reported in previous studies where extended retention periods improved microbial reduction through enhanced biological oxidation and sludge settling characteristics. The observed trend confirms that HRT is a critical operational parameter governing pathogen removal kinetics in SBR systems

Table 4.1: Effect of HRT on Pathogen Removal Efficiency

HRT (h)	E. coli Removal (%)	Total Coliform Removal (%)
8	72.5	70.8
12	82.4	80.6
18	96.4	95.8
24	95.2	94.7

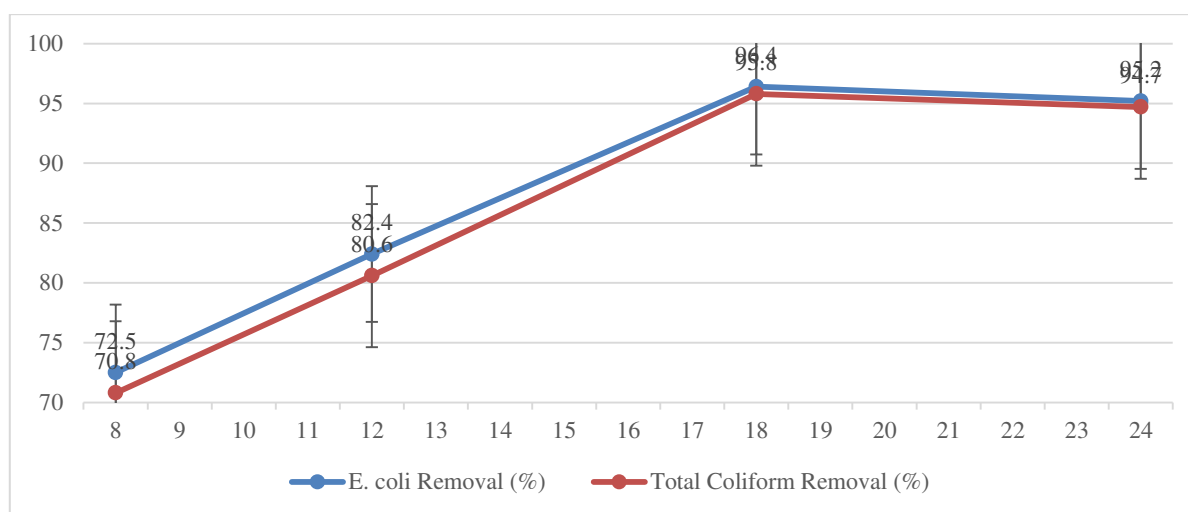


Figure 4.2 Effect of Hydraulic Retention Time on Pathogen Removal Efficiency in SBR System

4.2 Effect of Dissolved Oxygen (DO) on Pathogen Removal

Dissolved Oxygen (DO) concentration plays a crucial role in maintaining aerobic microbial activity and regulating biological oxidation processes within SBR systems. Adequate oxygen availability enhances microbial metabolism, promotes sludge floc formation, and improves biodegradation efficiency, thereby influencing pathogen inactivation.

In the present investigation, DO concentration was varied between 1 and 4 mg/L to evaluate its effect on microbial reduction performance. Experimental results revealed that pathogen removal efficiency increased significantly with increasing DO concentration up to an optimum value of 3 mg/L. Enhanced oxygen transfer improved aerobic microbial growth, resulting in greater microbial competition, predation, and oxidation of organic contaminants.

At lower DO concentration (1 mg/L), limited oxygen availability suppressed microbial activity and reduced biodegradation efficiency, leading to lower pathogen removal. Increasing the DO concentration to 3 mg/L substantially enhanced microbial reduction due to improved oxygen transfer efficiency and stable aerobic conditions. However, a slight decrease in removal efficiency was observed at 4 mg/L DO, which may be attributed to excessive aeration causing floc disruption and turbulence within the reactor.

The results indicate that maintaining optimum DO concentration is essential for achieving stable biological treatment performance and maximum pathogen removal efficiency in SBR systems.

4.2.1. Discussion of Findings

The highest microbial reduction efficiency was achieved at 3 mg/L DO concentration, indicating optimum aerobic conditions for microbial stabilization and pathogen decay. Adequate oxygen availability promoted biological oxidation, sludge flocculation, and microbial predation mechanisms responsible for pathogen removal.

Table 4.2 Effect of DO Concentration on Pathogen Removal

DO (mg/L)	E. coli Removal (%)
1	68.2
2	82.5
3	96.4
4	95.1

The findings confirm that DO concentration is a critical operational parameter affecting microbial kinetics and treatment efficiency in SBR systems.

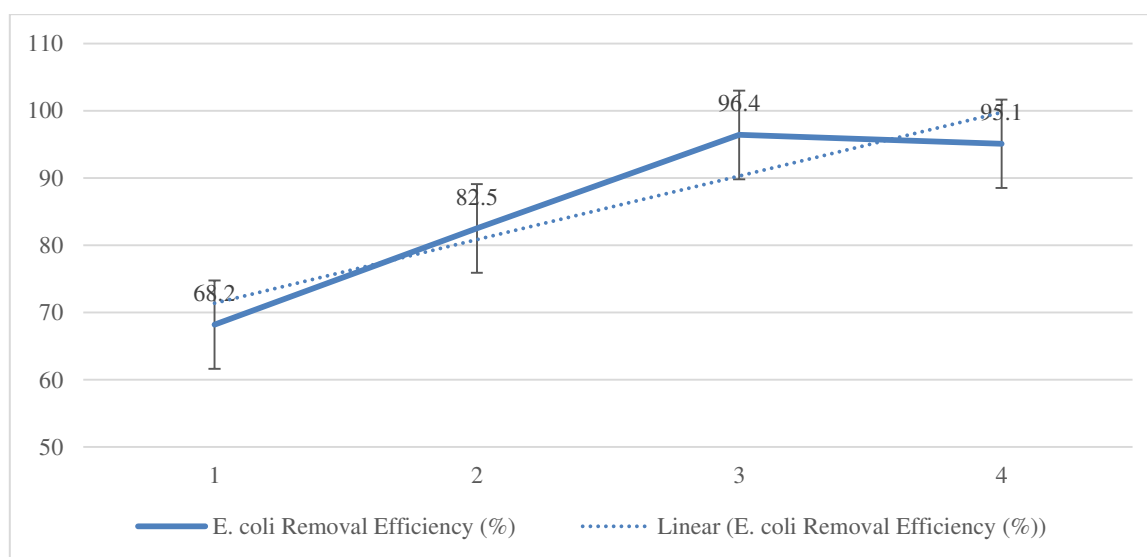


Figure 4.2.1. Influence of Dissolved Oxygen Concentration on Pathogen Removal Efficiency

4.3 COD Removal Performance

Chemical Oxygen Demand (COD) removal efficiency is an important indicator of biological treatment performance and organic pollutant degradation efficiency in wastewater treatment systems. COD reduction reflects the stabilization of biodegradable organic matter through microbial oxidation and metabolic activity.

The SBR system demonstrated excellent organic pollutant removal performance under optimized operating conditions. Influent COD concentration of 540 mg/L was reduced to 62 mg/L after treatment, corresponding to a COD removal efficiency of 88.5%.

The high COD reduction observed in the present study can be attributed to:

- Enhanced aerobic biodegradation
- Effective microbial oxidation
- Improved sludge settling characteristics
- Stable reactor operation
- Optimum aeration conditions

The sequential operational phases of the SBR system provided sufficient contact time between wastewater contaminants and active microbial biomass, leading to efficient degradation of biodegradable organic matter.

Table 4.3 COD Removal Efficiency

Parameter	Influent	Effluent	Removal (%)
COD	540 mg/L	62 mg/L	88.5

4.3.1. Discussion of Findings

The observed COD removal efficiency confirms the effectiveness of the SBR system in achieving stable biological treatment performance. High COD reduction also contributed indirectly to enhanced pathogen removal by improving overall reactor conditions and reducing microbial competition for substrate availability.

The treated effluent COD concentration satisfied permissible discharge standards for secondary biological treatment systems and demonstrated the suitability of SBR technology for sustainable wastewater treatment applications.



Figure 4.3.1 Comparative Analysis of Influent and Effluent COD Concentration

4.4 Kinetic Modelling of Pathogen Removal

Understanding microbial decay kinetics is essential for evaluating biological treatment efficiency and predicting pathogen removal behavior in wastewater treatment systems. In the present study, pathogen removal data obtained from the SBR reactor were analyzed using first-order kinetic modelling.

The experimental observations indicated that microbial decay followed first-order kinetics, suggesting that pathogen removal rate was directly proportional to microbial concentration within the reactor system.

The first-order kinetic model is represented as:

$$\ln\left(\frac{N_0}{N}\right) = kt$$

Where:

- N_0 = Initial microbial concentration
- N = Final microbial concentration
- k = First-order decay rate constant
- t = Reaction time

The linear relationship obtained between $\ln(N_0/N)$ and reaction time confirmed the applicability of first-order kinetics for describing pathogen decay in the SBR system. The kinetic analysis demonstrated that increased HRT and optimum DO concentration significantly enhanced microbial inactivation rates.

Table 4.4 First-Order Kinetic Plot for Pathogen Decay in SBR System:

Reaction Time (h)	Initial Concentration N_0 (CFU/100 mL)	Final Concentration N (CFU/100 mL)	$\ln(N_0/N)$
2	1.9×10^6	8.5×10^5	0.804
4	1.9×10^6	4.1×10^5	1.533
6	1.9×10^6	1.8×10^5	2.356
8	1.9×10^6	9.2×10^4	3.027
10	1.9×10^6	5.4×10^4	3.559

The first-order kinetic model provides an effective predictive framework for estimating pathogen removal performance under varying operational conditions and can be utilized for reactor design optimization and process control in advanced wastewater treatment systems.

4.4.1. Discussion of Findings

The observed kinetic behaviour confirms that biological pathogen removal in SBR systems is strongly influenced by operational conditions controlling microbial metabolism and sludge characteristics. The developed kinetic model can assist in predicting treatment efficiency and optimizing reactor operation for large-scale applications.

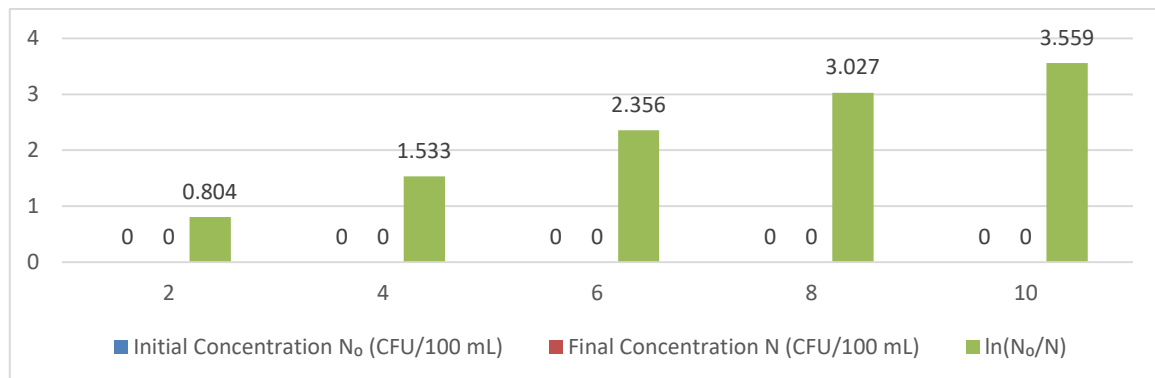


Figure 4.4.1 First-Order Kinetic Plot for Pathogen Decay in SBR System

V. CONCLUSION

Pathogen removal efficiency in a laboratory-scale Sequencing Batch Reactor treating municipal wastewater. The optimized operating conditions achieved:

- 96.4% E. coli removal
- 95.8% total coliform removal
- 88.5% COD removal

Hydraulic Retention Time and Dissolved Oxygen were identified as the most significant parameters influencing microbial reduction. Statistical optimization and kinetic modelling confirmed the effectiveness of SBR technology for sustainable wastewater treatment and safe reuse applications. The findings provide a scientific basis for future pilot-scale implementation and advanced optimization of SBR-based wastewater treatment systems suitable for urban and decentralized wastewater management applications.

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