

Comparative performance assessment of polymer based bio-carriers for enhanced biological wastewater treatment

Manish Kumar¹, Dayanand Sharma^{1*}, Vishwesh Mishra²

¹ Department of Civil Engineering, Sharda University, Greater Noida, 201310, India

² Department of Mechanical Engineering, Institute of Engineering and Technology, GLA University, Mathura, Uttar Pradesh, 281406, India

* Corresponding author's e-mail: dayanandsvnit@gmail.com

ABSTRACT

The study aims to comparatively evaluate the performance of five bio-carrier media biochip, white 1, white 2, black and m-media 1 used in biological wastewater treatment systems. The objective is to determine the influence of media geometry and surface characteristics on biofilm formation, organic load removal, and overall treatment efficiency. A modified activated sludge system was used to assess the performance of each bio-media type. Key performance indicators such as chemical oxygen demand (COD), biochemical oxygen demand (BOD), total suspended solids (TSS), and biofilm biomass accumulation were measured. The analysis also incorporated literature based data to compare the tested media with conventional bio-carriers used in moving bed bioreactors (MBBR) and fixed film reactors. Among all tested media, white 1 media of size 12×25 mm demonstrated the highest biofilm biomass density (10.13 g/m²) and superior organic load handling, achieving the lowest COD (334.5 mg/L) and BOD (110.2 mg/L) values, with a maximum COD loading rate of 3.19 g/m². Biochip media exhibited the largest specific surface area (5500 m²/m³) and the highest biomass concentration per volume (6.10 kg/m³), though with comparatively lower specific loading rates. The M-media 1 of size 20×20 mm displayed moderate pollutant removal and limited biofilm accumulation despite a surface area of 950 m²/m³. The findings highlight that media geometry and surface characteristics significantly affect treatment performance. White 1 media 12×25 mm media proved most efficient for organic load removal. Optimized bio-media selection can enhance wastewater treatment efficiency and promote sustainable water reuse in agricultural and landscape irrigation applications.

Keywords: bio-media, fixed film media, trickling filters, rotating biological contractors, suspended carrier media, moving bed bio reactors.

INTRODUCTION

Treatment of wastewater is an important environmental safeguard that protects human health and helps preserve waterways. It includes successive physical, chemical, and biological operations designed to reduce contaminant loads before discharge or reuse. The first treatment stage is dominated by mechanical unit processes such as screening, grit and oil removal, and primary sedimentation to eliminate suspended solids and floating debris (Morgan-Sagastume, 2018). Dissolved and colloidal organic matter and BOD is targeted in the subsequent

biological or secondary treatment stage, which, however, involves high aeration energy demand and is susceptible to organic and hydraulic shock loads. One of the earliest and widely used biological process is activated sludge process (ASP), wherein the aerobic or anaerobic active biomass suspended in mixed liquor degrades pollutants to carbon-di-oxide, water and new microbial cells (Solon et al., 2019). Nevertheless, the stability and performance of ASP systems in long-term periods are usually limited by operational problems dealing with low sludge settling velocity and high aeration energy requirements, and also organic or hydraulic shocks.

In order to overcome these drawbacks, biofilm-based or attached-growth systems have received significant attention in the last decades. At such treatment, microorganisms develop as a thin layer (or film) on the surface of solids which are used for growth called bio-carriers, while wastewater passes through or flows over them. The biofilm growth regime contributes to the stability of such a process and further to versatile microbial complexes, which can remove combined carbon and nutrients (Nhut et al., 2020). The cyclic events of formation and sloughing of biofilm mechanism provide the possibility of continuous operation, and efficient way to pollute degradation (Nema et al., 2023). The profile performance of the biofilm reactors is strongly dependent on the shape and media properties of carrier. High-surface-area carriers with higher porosity and more hydrophilicity are beneficial to the adhesion of microbes attachment and pollutant removal rate. Conventional materials like PE, PP and PU, ceramics or composite polymers have been studied by Ødegaard (2006) and Andreottola et al. (2000). The hydrodynamic regime and oxygen mass transfer are directly related to the biofilm thickness and efficiency in ring, spherical or fibrous carriers.

A number of biofilm-based processes have been developed to combine the advantages of attached and suspended growth systems. The moving bed biofilm reactor (MBBR) and the integrated fixed-film activated sludge (IFAS) are two well established technologies that enhance organic and nutrient removal in compact reactor volumes (Wanner et al., 2004; Rusten et al., 2006; Tianzhi et al., 2021). Although these systems are widely used, issues such as uneven biofilm growth, carrier clogging, and the cost or durability of synthetic media continue to present challenges. It is well documented that the type and morphology of carrier material have a critical influence on pollutant removal. Yang et al. (2022) demonstrated that PE and PU carriers perform better in nitrogen and COD removal in hybrid reactors. Zeolite-embedded carriers provide ion-exchange sites for ammonium adsorption, promoting nitrification under different load conditions (Morgan-Sagastume, 2018). Wang et al. (2021) observed that polyester fiber carriers achieved more than 95% removal of COD, TN, and $\text{NH}_4\text{-N}$ due to their microporous and hydrophilic characteristics. Similarly, Metcalf and Eddy (2013) and Nhut et al. (2020) reported that ceramic and iron-based carriers not

only supported microbial growth but also aided in phosphorus precipitation and removal. Yu et al. (2024) used rice husk-based biochar as a carrier, achieving significant COD removal efficiencies, demonstrating its potential as a sustainable and low-cost material. These findings indicate that reactor kinetics, microbial adhesion, and pollutant removal efficiency are strongly governed by both material composition and geometry.

Hybrid biofilm processes are now being explored to further improve performance. Al Hosani et al. (2022) and Boavida-Dias et al. (2022) reported that modified configurations such as the anoxic-oxic MBBR, sequential batch biofilm reactor (SBBR), and anaerobic-aerobic integrated biofilm systems have achieved simultaneous nitrification, denitrification, and phosphorus removal. According to Jagaba et al. (2024) rough, irregular carriers promote turbulence and prevent excessive biofilm thickness, thereby maintaining consistent oxygen and nutrient transfer. However, various limitations of bio-carriers such as low microbial compatibility at temperatures below 4 °C, high cost, and poor surface characteristics have motivated the pursuit of experimental and sustainable substitutes. While individual carrier types have been well studied, there is a lack of comparative studies between commercial and novel bio-carrier media under identical operating conditions. Most previous investigations have focused mainly on pollutant removal efficiencies without exploring the correlation between carrier surface properties, microbial morphology, and reactor performance. Furthermore, limited efforts have been made to identify cost-effective polymeric materials that offer comparable performance to engineered synthetic carriers, especially under variable influent conditions.

The originality of the current investigation lies in a systematic comparative assessment of commercial and laboratory-fabricated bio-carriers specifically polypropylene (PP), high-density polyethylene (HDPE), and virgin HDPE operated under identical conditions. The study highlights biomass characteristics, microbial attachment patterns, and performance indicators such as BOD, COD, and TSS removal. It aims to identify performance–property correlations that can guide the future design of new carrier media based on material properties and treatment efficiency. Additionally, the research incorporates sustainability aspects by evaluating cost and energy considerations, offering insights that can inform the

development of efficient, low-cost, and scalable biofilm-based wastewater treatment technologies suitable for both on-site and industrial applications. By comparing these aspects, this study contributes to a better understanding of how media properties influence biofilm behavior and pollutant removal efficiency. The findings are expected to provide a strong foundation for optimizing hybrid bioreactors and advancing sustainable practices in wastewater treatment.

MATERIALS AND METHODS

The experimental study was carried out at the Delhi Jal Board, sewage treatment plant 45 MGD phase four, Delhi, India, where pilot-scale systems were established to examine the relative performance of various bio-carrier media for enhanced biological wastewater treatment. The objective of the investigation was to evaluate the influence of carrier geometry, material type, and surface properties on microbial colonization, biofilm formation, and pollutant removal efficiency under controlled environmental and hydraulic conditions. Two identical pilot-plant facilities were constructed and installed adjacent to the full scale treatment works to enable continuous operation with real wastewater under process controlled conditions. Each pilot unit consisted of three rectangular aerobic bio-reactors arranged in series, followed by a settling tank. The first and

second reactors had internal dimensions of $150 \times 150 \times 500$ mm, while the third reactor measured $200 \times 200 \times 500$ mm, designed to process a daily flow rate of approximately 100 liters of settled wastewater. All tanks were stainless steel sheets to allow visual observation of biofilm growth, flow patterns, and aeration performance. A simplified schematic of the internal arrangement and flow sequence is shown in Figure 1 and Figure 2.

Aeration was provided through medium bubble diffusers placed at the bottom of each reactor to ensure uniform oxygen transfer and adequate mixing. The airflow rate was adjusted to maintain the dissolved oxygen (DO) concentration between 1.5 and 2.0 $\text{mg} \cdot \text{L}^{-1}$, monitored in situ using a portable DO meter (HACH, USA). This DO range represented a compromise between efficient aerobic biofilm development and minimizing shear stress that could detach the attached biomass. The pilot system was designed to operate under fluctuating DO levels, simulating the natural variability in oxygen availability typical of full scale systems. Both air and wastewater flow rates were normalized based on the protected surface area of the bio-carrier media to achieve uniform organic and nutrient loading across all trials.

The influent wastewater introduced into the first aeration stage was the effluent drawn from the primary sedimentation clarifier of the existing sewage treatment plant (STP) operated by the Delhi Jal Board. To facilitate the establishment of an active microbial consortium and ensure stable

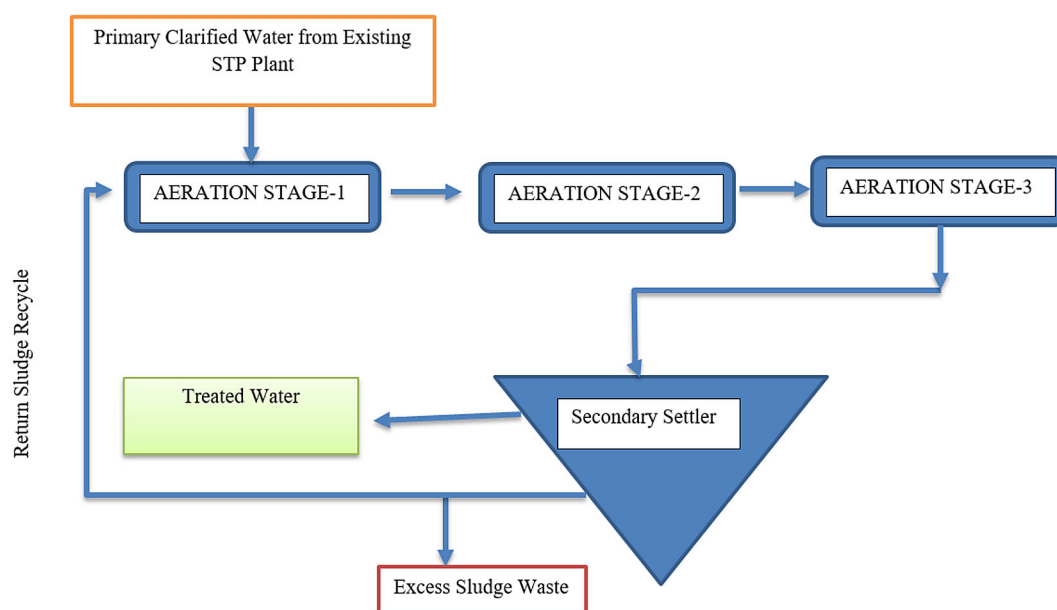


Figure 1. Process flow adopted for wastewater treatment

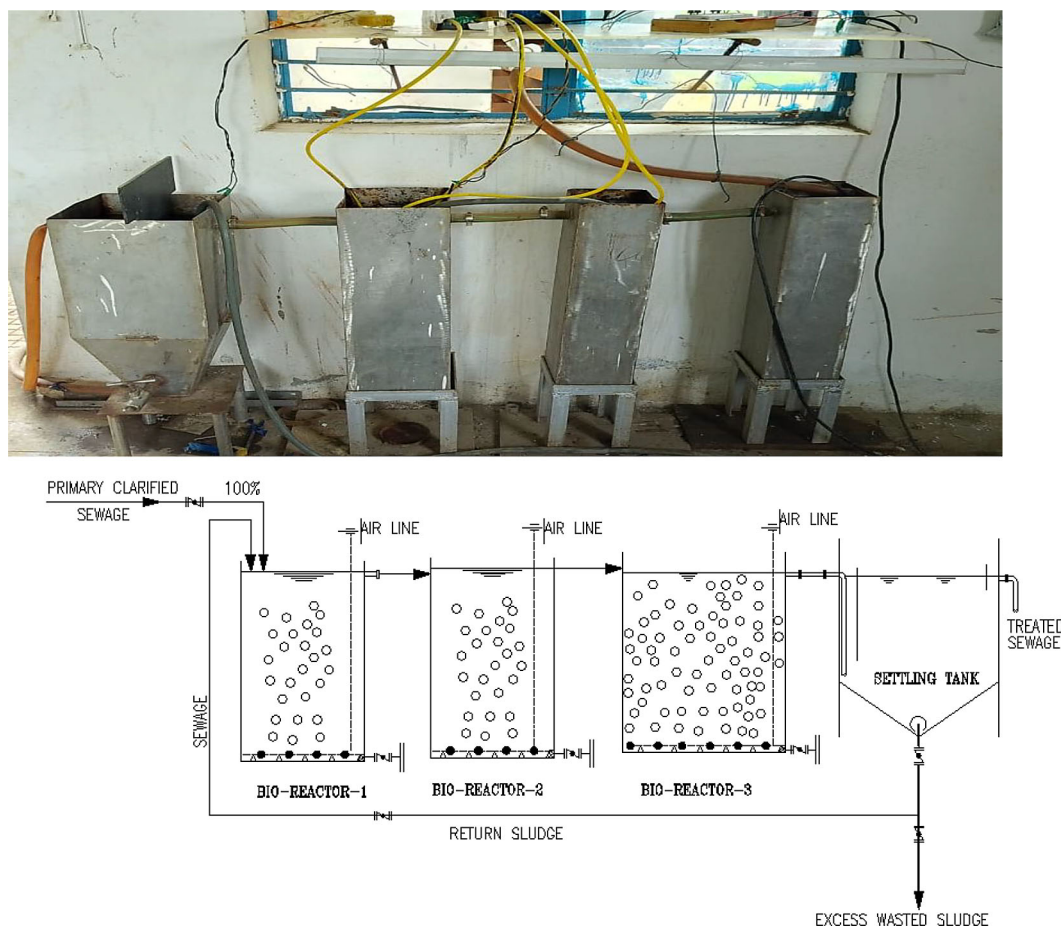


Figure 2. Schematic flow diagram of the pilot-scale wastewater treatment process

biofilm development, the reactors were inoculated with return activated sludge (RAS) sourced from the final clarifier of the same treatment facility. The mixed liquor suspended solids (MLSS) concentration was maintained at approximately 3000 mg/L, similar to that of conventional activated sludge systems. The influent to the pilot reactors was drawn from the primary clarifier effluent of the same plant, and dilution water was added as needed to obtain wastewater characteristics representative of typical domestic sewage. A recycle pump was installed upstream of the first bio-reactor to return 25–50% of the clarifier effluent back to the aeration basin at settling tank.

This recirculation minimized short-circuiting, improved solids retention, and supported continuous microbial activity and stable operation. To reduce hydraulic short-circuiting and enhance flow distribution, two vertical baffles were installed in the settling tank one near the inlet and the other near the outlet ensuring efficient sedimentation and uniform flow across the tank section. The hydraulic retention time (HRT) in the reactors

was maintained at approximately 7–8 hours, depending on daily fluctuations in inflow. The pilot plants operated under ambient temperature conditions typical of Delhi, with liquid temperatures ranging between 20.7 °C and 23.2 °C throughout the experimental period.

The current study utilized five types of bio-media: Media 1 (Biochip), consisting of circular polypropylene (PP) disks with an average diameter of 32 mm and thickness of 1.25 mm, offering a high specific surface area of 5500 m²/m³; Media 2 (White 12 × 25 mm), a cylindrical PP carrier with a specific surface area of 350 m²/m³; Media 3 (White 16 × 22 mm), a cylindrical PP carrier providing a surface area of 400 m²/m³; Media 4 (Black 16 × 22 mm), a cylindrical HDPE carrier of identical dimensions to Media 3 with a surface area of 400 m²/m³; and Media 5 (M-Media-1), a cubical carrier (20 × 20 × 20 mm) made from virgin HDPE, offering a specific surface area of 950 m²/m³ (Table 1).

These media included both commercially available and experimentally developed

Table 1. Characteristics of five typed of media used for the biological treatment of wastewater

S.No.	Media	Total surface (m ² /m ³)	Shape	Dimensions		Material	Density(g/cm ³)
				Length (mm)	Diameter (mm)		
1.	Biochip	5500	Circular	1.25	32	PP	0.92
2.	White 1	350	Cylindrical	25	12	PP	0.92
3.	White 2	400	Cylindrical	22	16	PP	0.92
4.	Black	400	Cylindrical	22	16	HDPE	0.97
5.	M-Media-1	950	Cube	20	20	Virgin HDPE	0.94

alternatives, selected to provide a broad comparison of performance under identical operating conditions. Prior to use, all carriers were thoroughly rinsed with tap water to remove any surface contaminants or loosely attached particles. The media filling ratio in each reactor was maintained at 30% of the working volume, as recommended by Metcalf & Eddy (2013), to promote uniform biofilm growth and efficient mass transfer. All of the five media were run for 45 days each, and the total experimental time was roughly 130 days. The clean media were filled directly into the reactors at the beginning of each experiment and aerated stepwise for natural microbial colonization. No foam or scum was formed, suggesting the successful acclimatization and stable biofilm development. pH, temperature, DO and flow rate were measured daily for all the experimental runs to control the process in a uniform manner. DO was controlled by altering the aeration rate and hydraulic load. Periodic backwashing and diffuser cleaning were conducted to minimize plugging and maintain uniform oxygen transfer during operation.

Sampling and analytical procedures

Influent and effluent samples were taken daily in both the startup and steady state periods to evaluate the efficiency of each bio-carrier system. Analyses were performed in accordance with the Standard Methods for Examination of Water and Wastewater (Gzar et al., 2021). The essential parameters evaluated were BOD₅, COD, and TSS as the key parameters for determining effluent treatment performances in removal of organic compounds and suspended solids. Moreover, in-site pH, temperature and DO were recorded with the aid of calibrated portable meters (HACH UK). All samples were analyzed in duplicate so that the results are reliable, and mean values were included in result interpretation. Influent wastewater

demonstrated the typical variability expected with domestic sewage characterized by COD levels of 294–540 mg/L, BOD levels were determined as 98–193 mg/L, and TSS levels varied between 103–376 mg/L. The bio-carrier media were assessed by pollutant removal performance (COD, BOD and TSS) biofilm growth rate and operational long-term aspect. The relationship between media shape and microporous structure in terms of specific surface area, and therefore the potential for microbial attachment were considered to be important determinants of reactor performance. The study observed the time to start, steady-state stability, oxygen-transfer efficiency and biofilm morphology of membranes in the five media. In summary, this systematic optimizing established a relationship between the kinetics in reactors and the carrier design but also its material properties paving the way for identifying optimum configurations for large scale application. The results of the present pilot-scale study further enhance the development of low-cost and energy-effective biofilm technology for decentralized/industrial wastewater treatment.

RESULTS AND DISCUSSIONS

Operational environment and influent characteristics

Wastewater temperature remained under natural atmospheric conditions during the operation of the pilot-scale biofilm reactor, with liquid temperatures ranging from 20.7 °C to 23.2 °C for all five tested media. These temperature fluctuations, due to seasonal changes, also affected microbial kinetics and biochemical reaction rates. Mohammed et al. (2021) reported a similar trend in pilot-scale biofilm reactors, with optimal microbial activity observed at 20–25 °C for sustainable organic removal. The pH values

of the medium, which remained near neutral (7.07–7.43), provided favorable conditions for microbial growth and enzymatic activity. This agrees with Nhut et al. (2020), who stated that most heterotrophic bacteria are efficient within a pH range of 6.5–8.0. The characteristics of the influent wastewater (Table 2) showed COD values ranging from 241 to 540 mg/L, BOD values from 98 to 193 mg/L, and influent TSS ranging between 103 and 376 mg/L, indicating moderately high-strength domestic sewage. Changes in the influent quality resulted from variations in domestic discharge patterns and irregular organic loading. A similar degree of variability has been reported by Bassin et al. (2016) and Di Capua et al. (2022), who emphasized the impact of population density and water usage behavior on influent characteristics in small-scale treatment units.

The temporal behavior of influent COD and BOD is shown in Figures 3(a) and 3(b) for the reactor start-up period. The average influent COD for Media 1 to Media 5 was 419, 334, 337, 419, and 416 mg/L, respectively, whereas the corresponding average BOD values were found to be 142 mg/L and 141 mg/L. These variations corresponded to natural fluctuations in feed composition rather than process upsets. Similar patterns of influent variation were also recorded by McQuarrie et al. (2011) during the adaptation period of a hybrid biofilm process, where daily variations were correlated with differences in organic loads and diurnal flow cycles. The type and shape of the carrier media were found to be important parameters that significantly influenced system stability and microbial acclimatization. The variation in COD and BOD for Media 1 (Biochip) and Media 5 (M-Media-1) was smoother due to their higher specific surface areas ($5500 \text{ m}^2/\text{m}^3$ and $950 \text{ m}^2/\text{m}^3$), which enhanced microbial colonization and provided strong buffering capacity. High-surface-area carriers are known to promote rapid biofilm development and greater operational resilience against organic shock loads. Yu et al. (2024) and Prasad et al. (2021) found, that media with

enhanced surface morphology allow higher microbial retention, greater metabolic diversity, and more stable start-up performance.

Overall, the results of influent monitoring indicated a stable operational environment within the pilot reactor. Stabilization of temperature, pH, and organic load in the combined system created favorable conditions for biofilm growth and acclimatization. Comparable start-up stabilization periods were observed by Prasad et al. (2023) and Falletti et al. (2014) in MBBRs operated as packed-bed reactors, highlighting that a well-balanced influent composition is a key factor in maintaining sustainable treatment efficiency.

Biofilm development and attachment dynamics on different carrier media

Formation and development of biofilms on carriers are key aspects in knowing how microorganisms attach and system stability in attached-growth waste water treatment processes. As observed in the results reported from this study, biofilm was accumulated at a time-dependent manner and even among the carriers different carrier media were distinguished like Biochip, White 12×25 , White 16×22 , Black 16×22 and M-Media1 (Figure 4). The findings of comparative analysis indicate that the physicochemical properties such as surface texture, porosity, hydrophilicity etc., have an attack being indispensable to microbial adhesion and growth stabilization. In this context, the results are in harmony with a widely acknowledged reality that biofilm formation proceeds generally in three phases: lag phase (the earliest response to adherence surface), slower accumulation or colonization phase of bacterial cells and maturation or stabilization phase.

In this study, Biochip media had relatively low attachment rate and affinity in the early operation. A clear absence of attachment was evidenced in the first 20 d with the attached biomass being low (ca. $0.072 \text{ g}/\text{m}^2$). On the contrary, accumulation () was greater at d 25 and then

Table 2. Characterization of the influent wastewater fed to the pilot plant operated with different media during start up

Parameter	Unit	Media 1	Media 2	Media 3	Media 4	Media 5
pH range		7.18–7.28	7.2–7.31	7.07–7.18	7.08–7.18	7.08–7.43
COD range	mg/l	308–540	294–406	241–390	296–508	294–414
BOD range	mg/l	99.4–193	98–133	99–135	112–191	110–187
TSS range	mg/l	103–308	156–212	159–376	103–299	120–296

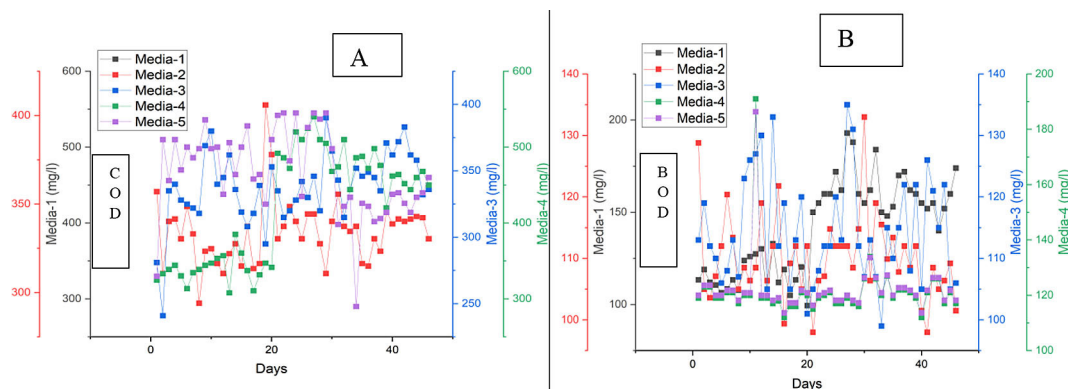


Figure 3. (a) COD and (b) BOD profile of sewage influent in pilot reactor

accelerated toward a plateau of 1.085 g/m^2 at d 30 that reached to 1.70 g/m^2 by d 36 when the experiment ended. The delayed colonization of the polymer may be ascribed to the smooth surface and low surface energy of biochip, which offers less adhesive sites. Here, similar trends were found as well by Xu et al. (2025) and Abdelfattah et al. (2020), which described slower microbial adhesion and global biomass accumulation for smoother polyethylene-based carriers leading to a delayed biofilm appearance at the surface. This indicates that although the Biochip medium is suitable for steady biofilm formation, the low surface roughness of the medium contributes to slower start-up kinetics in operating bioreactor systems by retarding microorganisms to adhere.

In marked contrast, White 12×25 demonstrated a steep increase in biofilm formation when compared to the non-porous, geometrically simple material. This carrier further supported a fast stage of attached growth which was observed during the first 20 days, allowing the biomass to reach about 2.324 g/m^2 . This was followed by a robust exponential phase with Day 30 being the time to a sharp rise (6.97 g/m^2) and accumulation after this period reached an average of $\sim 14.048 \text{ g/m}^2$. The high colonization rate of White 12×25 might relate to its texture (housing micro-environments that could enhance bacterial adhesion, hells and EPS production). These observation was in line with report by Xu et al. (2025) and Ødegaard (2006), who reported that media with more surface area and microspore density resulted in greater microbial attachment and nitrification performance. Also, there were reports that polymeric supports grafted to generate rougher surface lead to shear protected biological niches with the development of thick biofilm and

reduction in detachment 4, 5 – a possible reason for the better performance shown by White 12×25 in this study.

The White 16×22 and Black 16×22 carriers exhibited similar temporal growth patterns but differed in total biomass accumulation. Initially, biofilm formation on White 16×22 was moderate, with biomass reaching 3.454 g/m^2 at Day 20. A sharp increase followed, reaching 8.940 g/m^2 at day 30, before leveling off at 9.570 g/m^2 . The results for Black 16×22 were slightly lower, with 3.295 g/m^2 at day 20, 5.539 g/m^2 at day 30, and a final plateau at 7.405 g/m^2 . Although the differences between the carriers were modest, they indicate that polymer composition and color influence microbial attachment behavior. The darker carrier likely absorbed more heat, influencing localized temperature gradients and enhancing microbial metabolic rates, but its smoother surface may have reduced adhesion strength.

Similar observations were made by Madan et al. (2022) and Mohammed et al. (2021), who reported that surface chemistry and optical properties of biofilm carriers control the spatial distribution and metabolic activity of attached microorganisms. Additionally, black carbon-infused polymers are more hydrophobic, which can deter initial bacterial adhesion, especially for hydrophilic microbial communities commonly found in domestic wastewater systems. A particularly noteworthy observation emerged for M-Media-1, a carrier developed specifically for this study. Unlike conventional media, M-Media-1 demonstrated a balanced biofilm growth pattern, characterized by moderate and consistent biomass accumulation. The attached biomass increased steadily from 1.424 g/m^2 at Day 10 to 1.523 g/m^2 at Day 20, indicating active microbial colonization.

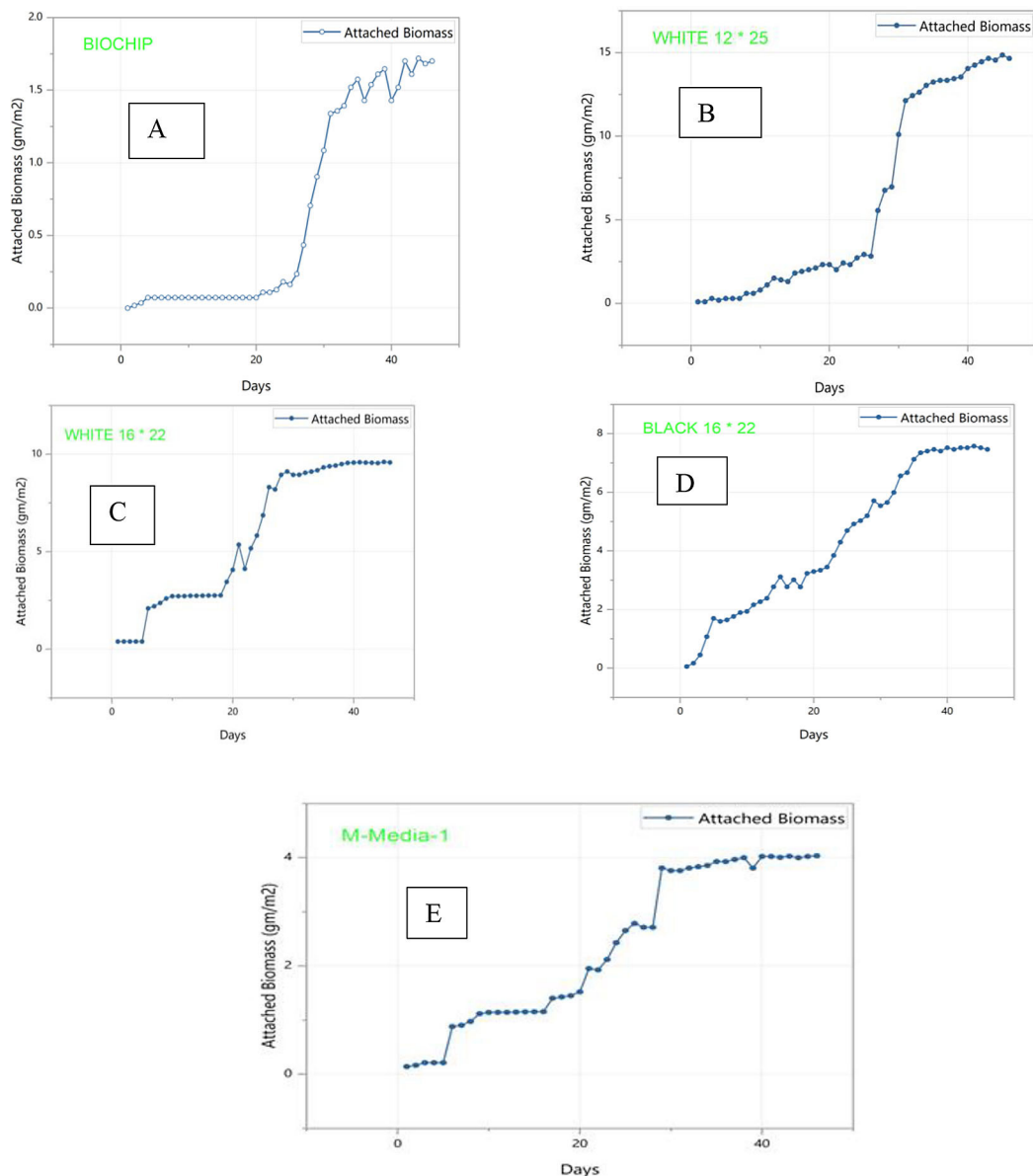


Figure 4. Biomass attachment profile of (a) Media-1 (Biochip) (b) Media-2 (White 12 × 25 mm) (c) Media-3 (White- 16 × 22 mm) (d) Media-4 (Black 16 × 22 mm) (e) Media-5 (M-Media-1)

When the system entered the stabilization phase after Day 30, the biofilm mass rose sharply to 3.761 g/m² and eventually stabilized at 4.034 g/m². These growth trends suggest that M-Media-1 provides favorable physicochemical conditions for effective microbial adhesion while preventing excessive overgrowth. Such controlled biofilm formation is beneficial for maintaining stable reactor performance, as excessively thick biofilms can lead to clogging, limited oxygen diffusion, and sloughing under high shear stress. Similar advantages of regulated biofilm formation have been reported by (Zhuang et al., 2019), who emphasized that optimized carrier design should balance microbial retention with hydraulic stability.

The ranking order of the final attached biomass across all tested media was: White 12 × 25 > White 16 × 22 > Black 16 × 22 > M-Media-1 > Biochip. This hierarchy shows that both geometric arrangement and surface properties have significant effects on the rate of microbial colonization. Better performance of White 12 × 25 could be a proof of the statement made by [5] that increasing ESS/v volume leads to larger surface area for biofilm growth on carriers and enhanced reactor efficiency in MBBRs. But we should emphasize that such a large surface area also has its disadvantages as, for example, the potential to favour formation of biofilm of rather excessive thickness can amplify detachment on poor system design.

In contrast, M-Media-1 showed a lower maximum biofilm density but an easily regulated and more sustainable accumulation that is favourable for the stability of the reactor over time. Such balanced growth kinetics is particularly important for the aerobic biofilm reactor design treating high strength organic wastewater, where shear stress and nutritional limitation are encountered to be significantly relevant operating difficulties. Results of this study demonstrate that the choice of carrier material is a key to control biofilm growing kinetics and reactor performance. Rougher surfaces, like those described in the XPS results in addition to our own XPS and AFM data, as well as hydrophilic substrates induce faster microbial attachment and thicker biofilms (Katsikogianni et al., 2007) due predominantly to modes of action at the cellular level (fluctuations in surface energy due to roughness at dimensions similar to that of cells). Fifteen engineered surfaces, such as M-Media-1 appear to yield mediation of microbial adhesion evident through more predictable and consistent development also stabilization. These findings play a key role in further improving biofilm-based reactors, stressing that carrier design need to meet particular process goals and operation modes.

Evaluation of pollutant removal efficiencies across biofilm media

Five bio-carrier media had the performances compared in terms of identical operation (Figure 5), where treatment efficiency and colony growth showed significant variation. Values listed in Table 3 indicate that both the structural and morphological properties of the media greatly affected COD (a), BOD (b) and TSS (c) removal efficiencies. For Biochip (Media-1), the average COD, BOD and TSS concentrations in effluent over 15 days were 26 mg/L, 6.7 mg/L and 10.3 mg/L. The continuous biomass load was low (1.4–1.6 g/m²) and the microorganisms were loosely attached and poorly colonized to the surface of tiles. These results corroborate Jagaba et al. (2024) and Dias et al. (2018), who also observed poor biofilm formation on biochips with smooth surfaces, as these do not tend to confine microorganisms efficiently. Lower efficiency of Media-1 (Biochip) could be due to its low surface area and less availability of nutrients, which resulted in poor development of biofilm and lesser organic matter degradation.

- Media-2 (White 1) recorded the best treatment performance of the tested media, resulting in effluent concentrations of COD = 25 mg/L, BOD = 5.6 mg/L and TSS = 8.7 mg/L, with biomass formation of between 10.4 g/m² and 14.5 g/m² which reflects excellent microbial

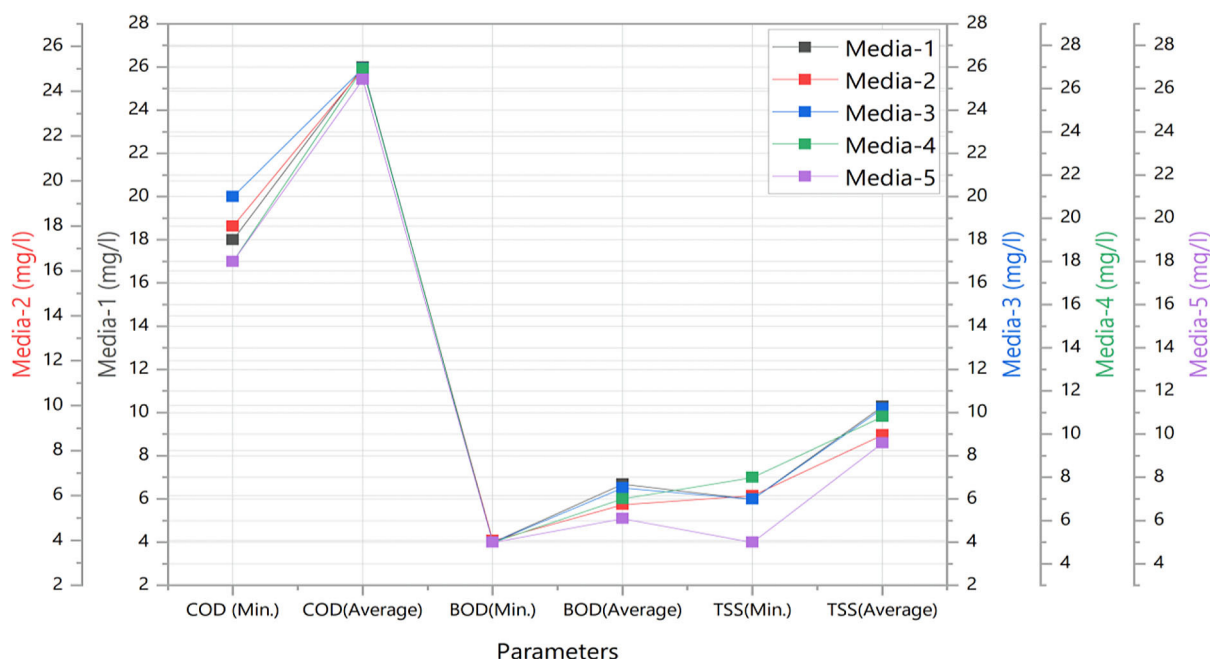


Figure 5. Performance based analysis of treated effluent using media 1 to 5

adaptability and a strong growing biofilm. This is indicative of the surface structure of Media-2 providing more points for microbial attachment and enough voids spaces for oxygen transfer both important to aerobic biodegradation. Comparable results were found as given in Regmi et al. (2011) and Dereli et al. (2021) that media with an irregular, porous and hydrophilic structure have high microbial biomass density which will further increase the COD and BOD removal by microorganisms. Therefore, Media-2 was suitable to biofilm development and the oxidation of pollutants.

- Media-3 (White 2) showed stable and effective pollutant removal performance, with 15-day average effluent concentrations of 25.95 mg/L COD, 6.52 mg/L BOD, and 10.2 mg/L TSS. Biomass stabilization ranged from 8.6–9.5 g/m², slightly lower than Media-2 but still demonstrating high microbial retention. The consistent results confirm Media-3's ability to maintain biological equilibrium under continuous loading. Boavida-Dias et al. (2022) and Prasad et al. (2023) reported that uniform-pore bio-carriers foster stable microbial floc structures, enhancing process resilience under varying hydraulic and organic loads. The moderate but stable pollutant removal by Media-3 suggests that oxygen diffusion and microbial stratification were adequate for combined aerobic and facultative biodegradation processes.
- Media-4 (Black) exhibited moderate performance, with higher effluent concentrations (COD = 26.93 mg/L, BOD = 7.02 mg/L, TSS = 10.82 mg/L) and biomass stabilization between 6.27–7.57 g/m². This reflects limited microbial colonization, likely due to suboptimal surface heterogeneity or reduced pore accessibility. Although COD and BOD residuals were slightly higher, Media-4 demonstrated

consistent results, indicating adaptability to fluctuating operational conditions. According to (Pratap et al., 2024), bio-carriers with moderate surface roughness and hydrophilicity often sustain active microbial populations but restrict deeper biofilm penetration. Consequently, Media-4 appears suitable for treating moderate organic loads but less effective for high-strength wastewater.

- Media-5 (M-Media 1) showed comparatively lower performance, with average effluent concentrations of 26.43 mg/L COD, 6.1 mg/L BOD, and 9.6 mg/L TSS, and biomass stabilization ranging from 3.63–5.71 g/m². Although the effluent range was narrower (COD: 18–26 mg/L; BOD: 5–7 mg/L; TSS: 5–9 mg/L), overall treatment efficiency was inferior to Media-2 and Media-3. The reduced biomass accumulation suggests limited microbial affinity, possibly due to insufficient surface area or low wettability. Carriers with low surface energy inhibit microbial adhesion, resulting in thin and unstable biofilms consistent with the lower biomass retention and higher effluent concentrations observed.

The combined interpretation of Figure 5 supports these trends. Media-2 consistently achieved the lowest COD, BOD, and TSS levels, confirming its superior pollutant removal capacity. In contrast, Media-1 and Media-5 exhibited higher effluent values, reflecting weaker microbial support and limited mass transfer. The relationship between biomass stabilization and effluent quality aligns reported a positive correlation between biofilm thickness, stability, and organic load removal efficiency. Overall, Media-2 proved to be the most efficient bio-carrier, achieving an optimal balance between surface morphology, porosity, and microbial colonization. Media-3 followed

Table 3. Analytical summary of media based treatment performance

Media type	COD range (mg/L)	BOD range (mg/L)	TSS range (mg/L)	Last day Avg COD	Last day Avg BOD	Last day Avg TSS	Biomass stabilization (g/m ²)
Media-1 (Biochip)	18–43	4–8	6–13	26.00	6.70	10.30	1.4–1.6
Media-2 (White 1)	18–30	4–9	6–14	25.00	5.60	8.70	10.4–14.5
Media-3 (White 2)	20–32	4–8	6–12	25.95	6.52	10.20	8.6–9.5
Media-4 (Black)	18–38	5–12	8–19	26.93	7.02	10.82	6.27–7.57
Media-5 (M-Media 1)	18–26	5–7	5–9	26.43	6.10	9.60	3.63–5.71

closely, providing stable and reproducible treatment performance. Media-1, Media-4, and Media-5, having lower surface area availability for biofilm support, demonstrated only moderate pollutant removal. These results emphasize the crucial role of carrier morphology and surface chemistry in determining biological treatment efficiency. Future studies should focus on improving long-term biofilm stability, hydraulic performance, and resistance to detachment to ensure reliable operational outcomes.

Comparative evaluation of bio-media performance in biological wastewater treatment

The comparative study of the five bio-carrier media (Table 4) also shows the effects of geometry, surface area, and material on pollutant removal and biofilm growth. Among the tested media, White 1 – 12×25 mm performed best overall, producing lower effluent COD (25 mg/L) and BOD (5.6 mg/L) compared to other materials, with a biomass accumulation of 10.4–14.5 g/m². Such high efficiency can be attributed to its well-proportioned surface area (350 m²/m³) and cylindrical shape, which provided homogeneous biofilm growth and optimal oxygen transfer. The results are consistent with those of the Kaldnes K1-type MBBR process, which generally achieves COD removals in the range of 85–95% and demonstrates robust nitrification due to favorable hydrodynamic characteristics and biofilm retention (Andreottola et al., 2000; McQuarrie et al., 2011).

Biochip media exhibited moderate performance (COD: 26 mg/L, BOD: 6.7 mg/L) despite

its high specific surface area (5500 m²/m³). The lower specific loading rates obtained for Biochip are consistent with data reported for trickling filter systems, where high surface area promotes biomass retention but may restrict substrate penetration through thick biofilms (Di Capua et al., 2022; Bassin et al., 2016). This behavior further emphasizes that increased surface area does not necessarily guarantee better treatment, particularly when mass transfer resistance increases in mature biofilms.

The White 2 – 16 × 22 media (400 m²/m³) demonstrated steady COD (25.9 mg/L) and BOD (6.5 mg/L) removal, indicating effective organic matter decomposition. Its behavior is comparable to that of IFAS systems, where attached and suspended microbial growths collectively contribute to both COD and nitrogen removal (Ødegaard et al., 1999; Boavida-Dias et al., 2022). This indicates that moderate media porosity and geometry provided sufficient surface for microbial attachment while maintaining adequate hydrodynamic mixing for substrate diffusion. The Black 16 × 22 mm media, made of HDPE, showed slightly higher effluent COD (26.9 mg/L) and BOD (7.0 mg/L) compared to the polypropylene media. Similar results have been reported for sponge-reinforced or polymeric MBBR carriers, which exhibit good structural stability and maintain treatment performance under fluctuating loadings but support slower biofilm growth compared to micro-carriers (Nhut et al., 2020). The greater density of HDPE could affect carrier buoyancy and mixing behavior, which may, in turn, influence total biofilm thickness and oxygen penetration.

Table 4. Comparative performance of bio-media with literature based trickling filter and activated sludge systems

Media type	Material / geometry	Surface area (m ² /m ³)	Observed COD/BOD removal (this study)	Comparable literature system	Literature performance	Reference
Media 1-Biochip	PP Circular Disc	5500	COD: 26 mg/L, BOD: 6.7 mg/L	Trickling Filter with PVC/HDPE rings	Moderate COD and NH ₄ ⁺ -N removal; stable biofilm formation	(H Ødegaard et al., 2000)
Media 2-White 1-12 × 25 mm	PP Cylindrical	350	COD: 25 mg/L, BOD: 5.6 mg/L (Highest removal)	MBBR using Kaldnes K1 (HDPE)	COD removal 85–95%; effective nitrification	(Rusten et al., 2006)
Media 3-White 2-16 × 22 mm	PP Cylindrical	400	COD: 25.9 mg/L, BOD: 6.5 mg/L	IFAS (Integrated Fixed-Film Activated Sludge)	Enhanced biomass retention; improved COD & TN removal	(H Ødegaard et al., 2000)
Media 4-Black 16 × 22 mm	HDPE Cylindrical	400	COD: 26.9 mg/L, BOD: 7.0 mg/L	Sponge-Based MBBR / SBBR	COD removal ~80–90%; stable under shock loads	(Nhut et al., 2020)
Media 5-Box 20 × 20 × 20 mm (M-Media-1)	Virgin HDPE Cubical	950	COD: 26.4 mg/L, BOD: 6.1 mg/L	Hybrid MBBR (ceramic/zeolite carrier)	COD removal 82–93%; moderate biofilm accumulation	(Di Capua et al., 2022)

Finally, the $20 \times 20 \times 20$ mm Box shape material (M-Media-1) with cubic structure and moderate surface area ($950 \text{ m}^2/\text{m}^3$) also showed moderate performance (COD: 26.4 mg/L ; BOD: 6.1 mg/L). The flat sheets had a slower response than the cylindrical PP media but still performed well and consistently, with controlled biofilm accumulation ($3.63\text{--}5.71 \text{ g/m}^2$). This is advantageous for avoiding congestion and securing long-term stability of the reactor. This behaviour is consistent with the hybrid and zeolite biofilm reactors where surface roughness and porosity were moderate enough to allow optimal microbial adhesion without a significant growth of the biofilm (Di Capua et al., 2022; Arabgol et al., 2020). Overall, the comparative analysis suggests that media structure/chemistry is a key determinant in biofilm development and pollutant removal performance. Tubular polypropylene media (White 1- 12×25 and White 2- 16×22) were superior to media of cubical geometry and manufactured with HDPE which means that some curvature or sphericity is necessary for high mass transfer rates (effective colonization by microorganisms). These findings are in line with available literature, on MBBR and IFAS systems and demonstrate the potential of tailored bio-media design to improve treatment efficiency for sustainable water reuse.

CONCLUSIONS

The effluent quality evaluation across Media 1 to 5 highlights distinct variations in pollutant removal efficacy, particularly in terms of average concentrations of COD, BOD, and TSS. Among the tested configurations, Media 2 consistently achieved the lowest mean values (COD: 25.00 mg/L , BOD: 5.60 mg/L , TSS: 8.70 mg/L), indicating superior treatment performance driven by effective biodegradation and particulate retention mechanisms. Conversely, Media 4 recorded the highest pollutant levels (COD: 26.93 mg/L , BOD: 7.02 mg/L , TSS: 10.82 mg/L), reflecting diminished treatment efficiency and underscoring the importance of optimal media selection for enhanced reactor performance. Media 5 demonstrated intermediate performance, with average values of COD (26.43 mg/L), BOD (6.10 mg/L), and TSS (9.60 mg/L). This outcome aligns with its physical characteristics an active surface area of $950 \text{ m}^2/\text{m}^3$ supporting moderate biofilm attachment, measured between 3.63 g/m^2 and 5.71 g/m^2 .

Notably, the progression toward optimum biofilm development contributes to reduced oxygen consumption, offering a potential decrease in energy demand and operational costs.

REFERENCES

1. Abdelfattah, A., Hossain, M. I., Cheng, L. (2020). High-strength wastewater treatment using microbial biofilm reactor: a critical review. In *World Journal of Microbiology and Biotechnology* 36(5). Springer. <https://doi.org/10.1007/s11274-020-02853-y>
2. Al Hosani, N., Fathelrahman, E., Ahmed, H., Rikabi, E. (2022). Moving Bed Biofilm Reactor (MBBR) for decentralized grey water treatment: Technical, ecological and cost efficiency comparison for domestic applications. *Emirates Journal of Food and Agriculture*, 34(9), 731–742. <https://doi.org/10.9755/ejfa.2022.v34.i9.2848>
3. Andreottola, G., Foladori, P., Ragazzi, M., Tatano, F. (2000). Experimental comparison between MBBR and activated sludge system for the treatment of municipal wastewater. *Water Science and Technology*, 41(4–5), 375–382. <https://doi.org/10.2166/wst.2000.0469>
4. Bassin, J. P., Dias, I. N., Cao, S. M. S., Senra, E., Laranjeira, Y., Dezotti, M. (2016). Effect of increasing organic loading rates on the performance of moving-bed biofilm reactors filled with different support media: Assessing the activity of suspended and attached biomass fractions. *Process Safety and Environmental Protection*, 100, 131–141. <https://doi.org/10.1016/j.psep.2016.01.007>
5. Boavida-Dias, R., Silva, J. R., Santos, A. D., Martins, R. C., Castro, L. M., Quinta-Ferreira, R. M. (2022). A Comparison of biosolids production and system efficiency between activated sludge, moving bed biofilm reactor, and sequencing batch moving bed biofilm reactor in the dairy wastewater treatment. *Sustainability (Switzerland)*, 14(5). <https://doi.org/10.3390/su14052702>
6. Dereli, R. K., Clifford, E., Casey, E. (2021). Co-treatment of leachate in municipal wastewater treatment plants: Critical issues and emerging technologies. *Critical Reviews in Environmental Science and Technology*, 51(11), 1079–1128. <https://doi.org/10.1080/10643389.2020.1745014>
7. Di Capua, F., Iannaccone, F., Sabba, F., Esposito, G. (2022). Simultaneous nitrification–denitrification in biofilm systems for wastewater treatment: Key factors, potential routes, and engineered applications. In *Bioresource Technology* 361. <https://doi.org/10.1016/j.biortech.2022.127702>
8. Dias, J., Bellingham, M., Hassan, J., Barrett, M., Stephenson, T., Soares, A. (2018). Impact of carrier

- media on oxygen transfer and wastewater hydrodynamics on a moving attached growth system. *Chemical Engineering Journal*, 351, 399–408. <https://doi.org/10.1016/j.cej.2018.06.028>
9. Falletti, L., Conte, L., Maestri, A. (2014). Upgrading of a wastewater treatment plant with a hybrid moving bed biofilm reactor (MBBR). *AIMS Environmental Science*, 1(2), 45–52. <https://doi.org/10.3934/environsci.2014.2.45>
10. Gzar, H. A., Al-Rekabi, W. S., Shuhaieb, Z. K. (2021). Application of Moving Bed Biofilm Reactor (MBBR) for Treatment of Industrial Wastewater: A mini Review. *Journal of Physics: Conference Series*, 1973(1). <https://doi.org/10.1088/1742-6596/1973/1/012024>
11. Jagaba, A. H., Abdulazeez, I., Lawal, D. U., Affam, A. C., Mu'azu, N. D., Soja, U. B., Usman, A. K., Noor, A., Lim, J. W., Aljundi, I. H. (2024). A review on the application of biochar as an innovative and sustainable biocarrier material in moving bed biofilm reactors for dye removal from environmental matrices. In *Environmental Geochemistry and Health* 46(9). Springer. doi: 10.1007/s10653-024-02122-z
12. Madan, S., Madan, R., Hussain, A. (2022). Advancement in biological wastewater treatment using hybrid moving bed biofilm reactor (MBBR): a review. In *Applied Water Science* 12(6), 141. Springer Science and Business Media Deutschland GmbH. <https://doi.org/10.1007/s13201-022-01662-y>
13. McQuarrie, J. P., Boltz, J. P. (2011). Moving bed biofilm reactor technology: process applications, design, and performance. *Water Environment Research*, 83(6), 560–575. <https://doi.org/10.2175/106143010x12851009156286>
14. Metcalf & Eddy. (2013). *Wastewater Engineering: Treatment and Resource Recovery*. Retrieved from <https://repositori.mypolycc.edu.my/handle/123456789/4586>
15. Mohammed, Z. B., Tarish, H. A., Jihad, H. A., Hasan, Y. R., Jaber, A. A. (2021). Municipal wastewater treatment using moving bed biofilm reactor (Mbbbr). *Journal of Green Engineering*, 11(1), 579–594. Retrieved from https://www.researchgate.net/profile/Zainab-Bahaa-2/publication/354722923_Municipal_Wastewater_Treatment_Using_Moving_Bed_Biofilm_Reactor_MBBR/links/6149adbc519a1a381f71c510/Municipal-Wastewater-Treatment-Using-Moving-Bed-Biofilm-Reactor-MBBR.pdf
16. Morgan-Sagastume, F. (2018). Biofilm development, activity and the modification of carrier material surface properties in moving-bed biofilm reactors (MBBRs) for wastewater treatment. *Critical Reviews in Environmental Science and Technology*, 48(5), 439–470. <https://doi.org/10.1080/10643389.2018.1465759>
17. Nema, A., Prasad, R., Sharma, D., Yadav, K. D., Christian, R. A., & Ibrahim, H. (2023). Performance evaluation of different macrophytes in small-scale vertical flow constructed wetlands for greywater treatment using principal component analysis. *Canadian Journal of Chemical Engineering*, 101(3), 1321–1334. <https://doi.org/10.1002/cjce.24527>
18. Nhut, H. T., Hung, N. T. Q., Sac, T. C., Bang, N. H. K., Tri, T. Q., Hiep, N. T., Ky, N. M. (2020). Removal of nutrients and organic pollutants from domestic wastewater treatment by sponge-based moving bed biofilm reactor. *Environmental Engineering Research*, 25(5), 652–658. <https://doi.org/10.4491/eer.2019.285>
19. Ødegaard, H., Gisvold, B., Strickland, J. (2000). The influence of carrier size and shape in the moving bed biofilm process. *Water Science and Technology*, 41(4–5), 383–391. <https://doi.org/10.2166/wst.2000.0470>
20. Ødegaard, Hallvard. (2006). Innovations in wastewater treatment: The moving bed biofilm process. *Water Science and Technology*, 53(9), 17–33. <https://doi.org/10.2166/wst.2006.284>
21. Prasad, R., Sharma, D., Yadav, K. D., Ibrahim, H. (2021). Preliminary study on greywater treatment using water hyacinth. *Applied Water Science*, 11(6), 88. <https://doi.org/10.1007/s13201-021-01422-4>
22. Prasad, R., Sharma, D., Yadav, K. D., Ibrahim, H. (2023). Sustainable ternary treatment system for greywater and potential reuse. *Canadian Journal of Chemical Engineering*, 101(3), 1298–1309. <https://doi.org/10.1002/cjce.24521>
23. Pratap, V., Kumar, R., Kumar, S., Yadav, B. R. (2024). Optimization of moving bed biofilm reactors for the treatment of municipal wastewater. *Environmental Research*, 241. <https://doi.org/10.1016/j.envres.2023.117560>
24. Regmi, P., Thomas, W., Schafran, G., Bott, C., Rutherford, B., Waltrip, D. (2011). Nitrogen removal assessment through nitrification rates and media biofilm accumulation in an IFAS process demonstration study. *Water Research*, 45(20), 6699–6708. <https://doi.org/10.1016/j.watres.2011.10.009>
25. Rusten, B., Eikebrokk, B., Ulgenes, Y., Lygren, E. (2006). Design and operations of the Kaldnes moving bed biofilm reactors. *Aquacultural Engineering*, 34(3), 322–331. <https://doi.org/10.1016/j.aquaeng.2005.04.002>
26. Solon, K., Volcke, E. I. P., Spérandio, M., Van Loosdrecht, M. C. M. (2019). Resource recovery and wastewater treatment modelling. In *Environmental Science: Water Research and Technology* 5(4), 631–642. <https://doi.org/10.1039/c8ew00765a>
27. Tianzhi, W., Weijie, W., Hongying, H., Khu, S. T. (2021). Effect of coagulation on bio-treatment of textile wastewater: Quantitative evaluation and application. *Journal of Cleaner Production*, 312. <https://doi.org/10.1016/j.jclepro.2021.127798>
28. Wang, R., Xu, Q., Chen, C., Li, X., Zhang, C., Zhang, D. (2021). Microbial nitrogen removal in

- synthetic aquaculture wastewater by fixed-bed baffled reactors packed with different biofilm carrier materials. *Bioresource Technology*, 331. <https://doi.org/10.1016/j.biortech.2021.125045>
29. Wanner, O., Morgenroth, E. (2004). Biofilm modeling with AQUASIM. *Water Science and Technology*, 49(11–12), 137–144. <https://doi.org/10.2166/wst.2004.0824>
30. Xu, J., Lai, Z., Zhang, W., Liu, T., Xiao, S., Yang, L., Yu, Z., Zhou, X. (2025). Moving bed biofilm reactor for blackwater treatment: insights into pollutant removal, microbial communities, and water quality prediction through machine learning. *Frontiers of Environmental Science and Engineering*, 19(8). <https://doi.org/10.1007/s11783-025-2022-7>
31. Yang, X., He, Q., Liu, T., Zheng, F., Mei, H., Chen, M., Liu, G., Vymazal, J., Chen, Y. (2022). Impact of microplastics on the treatment performance of constructed wetlands: Based on substrate characteristics and microbial activities. *Water Research*, 217. <https://doi.org/10.1016/j.watres.2022.118430>
32. Yu, M., Wang, L., Feng, P., Wang, Z., Zhu, S. (2024). Treatment of mixed wastewater by vertical rotating microalgae-bacteria symbiotic biofilm reactor. *Bioresource Technology*, 393. <https://doi.org/10.1016/j.biortech.2023.130057>
33. Zhuang, Y., Zhang, Z., Zhou, Z., Chen, M., Li, J., Chen, S. (2019). Co-treatment of shale-gas produced water and municipal wastewater: Removal of nitrogen in a moving-bed biofilm reactor. *Process Safety and Environmental Protection*, 126, 269–277. <https://doi.org/10.1016/j.psep.2019.04.010>