

MICROBIAL AND ENZYMATIC MECHANISMS IN INDUSTRIAL SLUDGE DEWATERING: EPS MODIFICATION AND KINETIC PERSPECTIVES

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ABSTRACT

Industrial sludge dewatering is challenging because of high moisture content and extracellular polymeric substances (EPS), which form hydrated matrices that retain water and hinder solid - liquid separation. Biological dewatering offers an alternative conditioning strategy by modifying these water-retaining structures through microbial activity and enzymatic hydrolysis. This review critically examines microbial processes and enzymatic strategies for improving industrial sludge dewaterability, with emphasis on microbial EPS degradation, microbial consortia, bio-drying, proteases, cellulases, amylases, lysozyme, and combined enzyme treatments. Microbial activity can progressively modify EPS through endogenous hydrolytic processes, whereas exogenous enzymes provide targeted degradation of protein- and carbohydrate-rich matrix components. These processes alter EPS composition, floc structure and water distribution, potentially increasing the fraction of water accessible to mechanical separation. Kinetic approaches are considered for describing EPS modification, enzymatic hydrolysis and biological moisture removal and for relating treatment time to measurable dewaterability. The review further examines reactor configurations, operational constraints, scale-up challenges and techno-economic and life-cycle considerations. Biological approaches remain constrained by slow reaction rates, mass-transfer resistance, enzyme instability, sludge heterogeneity and inhibition under high-salinity or toxic conditions. Integrated biological-mechanical systems and sludge-specific kinetic models are therefore needed for practical industrial application.

KEYWORDS: Biological dewatering; Industrial sludge; Microbial processes; Enzymatic strategies; Extracellular polymeric substances; Kinetic modelling

1. INTRODUCTION

Industrial wastewater treatment generates substantial quantities of sludge with variable physicochemical characteristics depending on the source industry and treatment process. Sludges from pulp and paper, petrochemical, textile, tannery, and food-processing industries commonly contain high moisture, organic matter, inorganic constituents, and extracellular polymeric substances (EPS), making dewatering essential for reducing sludge volume, transportation and disposal requirements. (Wu et al., 2020; Yuan & Zhu, 2024) Sludge dewaterability is strongly influenced by water distribution within the sludge matrix and EPS characteristics. EPS consists mainly of proteins, polysaccharides, humic-like substances, lipids and nucleic acids and forms a hydrated matrix around sludge particles and microbial cells. Its interaction with water contributes to surface-associated and bound-water retention. (Liu & Fang, 2003; Yu, 2020). Tightly bound EPS, particularly protein-rich fractions, is associated with greater water retention and poorer dewaterability. (Janiga-MacNelly & Lavado, 2024; Lin et al., 2019). Thus, EPS composition and structure are important determinants of sludge dewaterability.

Conventional dewatering relies mainly on mechanical separation and chemical conditioning. Biological conditioning provides an alternative by exploiting microbial activity and enzymatic reactions to modify water-retaining structures. Microorganisms can transform EPS through extracellular hydrolytic activity, while microbial consortia may provide complementary degradation of complex organic matrices. (Li et al., 2024; Shi et al., 2017). Microbially mediated bio-drying can additionally promote moisture removal through biological degradation, heat generation and mass transfer. (Cai et al., 2016; Zhang et al., 2024)). Enzymatic strategies provide more targeted modification; proteases, cellulases, amylases and lysozymes can act on different protein-, carbohydrate- and cellular components of the sludge matrix. (Allam et al., 2022; Kang et al., 2023)

Because microbial transformation, enzymatic hydrolysis and moisture removal are time-dependent processes, kinetic modelling can help describe treatment rates and relate biological transformation to dewatering performance. (Nicell, 2003). However, the transfer of such kinetic descriptions across different sludge matrices remains limited by variations in substrate composition and operating conditions. (Shao et al., 2020).

Despite increasing research interest, biological dewatering of industrial sludge remains constrained by sludge heterogeneity, limited substrate accessibility, slow reaction rates, enzyme instability, and inhibition under adverse

conditions. Although recent reviews have discussed microbial-assisted sludge dewatering and EPS-targeted conditioning, the relationship between microbial EPS transformation, enzyme-specific hydrolysis, water redistribution, and treatment kinetics remains insufficiently integrated. Therefore, this review develops a mechanism-to-kinetics framework integrating microbial processes, enzymatic strategies and kinetic modelling for industrial sludge dewatering, with emphasis on EPS modification, water redistribution, treatment-rate behaviour and implications for process optimization and scale-up.

2. Bibliometric Analysis

A systematic literature search was conducted using the Scopus database with the query: (sludge) AND (dewatering OR conditioning OR thickening) AND (biological OR microbial OR enzymatic OR bioleaching), without restriction on publication year. The search retrieved 977 articles spanning 1976 - 2025. The dataset was examined to characterize publication trends and the thematic landscape of biological sludge conditioning and dewatering. Because the present review focuses on microbial processes, enzymatic strategies, and kinetic modelling, bioleaching- and bioflocculant-specific studies were excluded from the subsequent mechanistic synthesis.

2.1. Annual publication trends

The literature shows a gradual development of research on biological sludge dewatering over the past five decades, followed by a marked increase in publication activity during the last decade. The existing dataset records the first publication in 1976, while annual output increased substantially after 2005. Publications increased from 41 articles in 2016 to 104 articles in 2025, with 83–92 articles reported annually during 2021–2023. The recent increase indicates growing scientific interest in biological approaches to sludge conditioning and dewatering, although the 2025 value should be interpreted cautiously because of possible database-indexing delays. (Figure 1) says about Annual scientific production on biological sludge dewatering research (2025 partial year may reflect indexing lag), which have Source from Scopus, March 2026

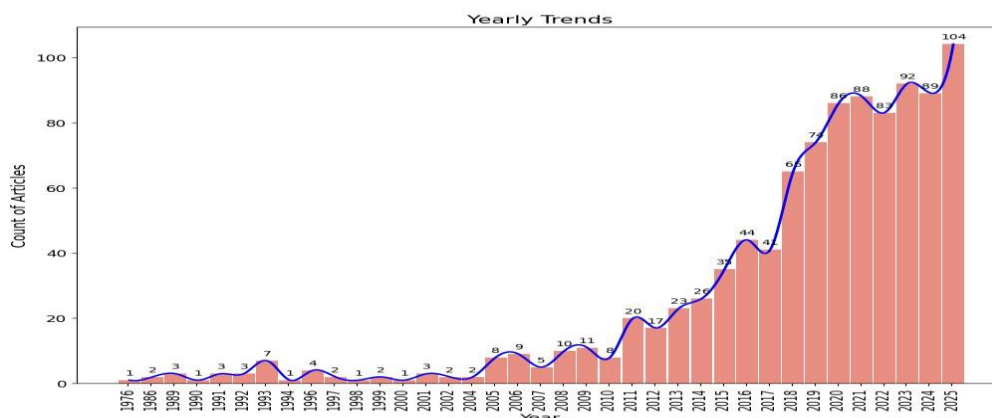


Figure 1. Annual scientific production on biological sludge dewatering research (2025 partial year may reflect indexing lag). Source: Scopus, March 2026

2.2. Research themes and keyword structure

Keyword analysis of the complete dataset indicates that the literature encompasses several biological conditioning themes. The initial network is heavily affected by terminology associated with bioleaching, such as *Acidithiobacillus ferrooxidans*, leaching, copper, chalcopyrite, and metal recovery. (Figure 2) illustrates the keyword co-occurrence network of the literature retrieved on biological sludge dewatering.

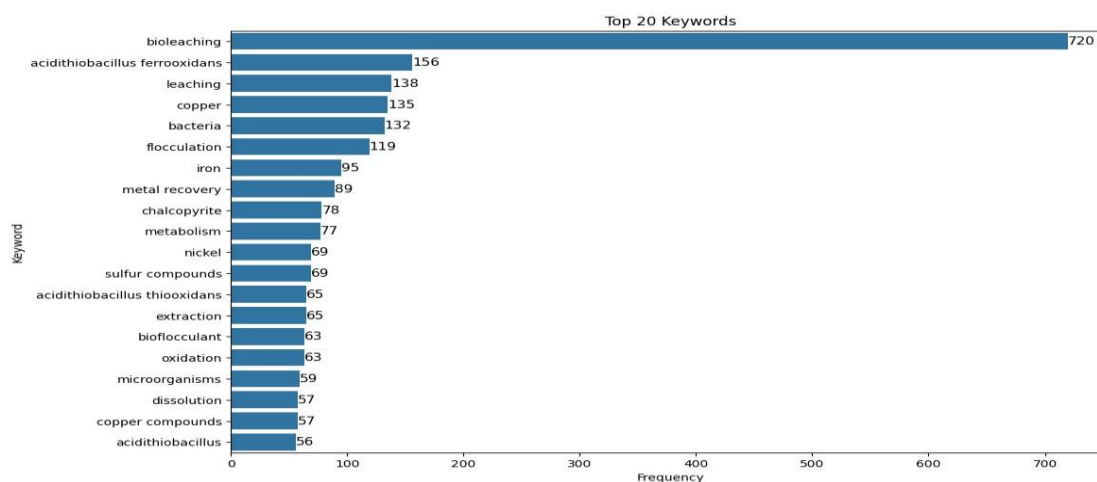


Figure 2. Keyword co-occurrence network of the retrieved literature on biological sludge dewatering.

Because these themes fall outside the defined scope of the present review, the keyword network should not be interpreted as representing the microbial–enzymatic dewatering field alone. Instead, the bibliometric analysis is used to demonstrate the broad interdisciplinary nature of biological sludge-conditioning research, while the subsequent review sections focus specifically on microbial EPS degradation, microbial consortia, enzymatic hydrolysis, bio-drying and kinetic modelling.

2.3. Research direction and emerging themes

The retrieved literature indicates increasing interest in biological modification of sludge properties through microbial activity, EPS transformation and enzymatic processes. Within the scope of the present review, recent research is particularly relevant to microbial EPS degradation, microbial consortia, bio-drying, enzyme-assisted conditioning and the use of kinetic approaches to describe biological transformation and moisture removal. These themes provide the basis for the subsequent discussion of microbial processes, enzymatic strategies and kinetic modelling in industrial sludge dewatering.

3. Physicochemical Basis Of Industrial Sludge Dewaterability

Industrial sludge dewaterability is governed by the interactions among sludge particle characteristics, floc structure, surface charge, extracellular polymeric substances (EPS), and water distribution within the sludge matrix. These properties determine the mobility of water during mechanical separation and also influence the accessibility of microbial cells and enzymes during biological conditioning.

3.1. Water distribution and sludge structure

Water in sludge can be broadly classified into free, interstitial, surface-associated, and bound fractions. Free and interstitial water are relatively accessible to mechanical separation, whereas surface-associated and bound water are retained more strongly by interactions with sludge particles and EPS (Sam et al., 2022). Consequently, improving dewaterability requires modification of the structures that restrict the mobility of less readily removable water.

Particle size and floc architecture influence these water fractions. Fine particles provide greater surface area for water and EPS interactions, while compact flocs restrict drainage pathways (AbuSeif et al., 2026). Surface charge also affects floc stability because negatively charged sludge particles and EPS can generate electrostatic repulsion that limits aggregation (Liu & Fang, 2003). Multivalent cations, including Fe, Al, Ca and Mn, can modify these interactions through charge neutralization and bridging, thereby influencing floc structure and permeability (Liu & Fang, 2003; Zhang et al., 2019). However, surface charge alone does not adequately predict dewaterability and should be considered together with particle characteristics, EPS composition, and direct dewatering indicators such as capillary suction time (CST) and specific resistance to filtration (SRF). (Tunçal & Mujumdar, 2023)

3.2. EPS Structure and Water Retention

Extracellular polymeric substances (EPS) form a hydrated matrix surrounding sludge particles and microbial cells and consist mainly of proteins, polysaccharides, humic-like substances, lipids, and nucleic acids. (Huang et al., 2022; Yu, 2020). Their hydrophilic functional groups promote water retention through hydrogen bonding and other molecular interactions, contributing to the persistence of surface-associated and bound water. (Peng et al., 2021; Sam et al., 2022). (Figure 3) illustrates the interactions of EPS that influence water retention and the ability to dewater sludge. EPS is generally categorized into three types: soluble EPS (S-EPS), loosely bound EPS (LB-EPS), and tightly bound EPS (TB-EPS). Among these categories, TB-EPS, especially its protein-dense portion, is closely linked to water retention and difficulties in dewatering, while the polysaccharide-rich fractions help enhance matrix cohesion and viscosity. (Lin et al., 2019; Yu et al., 2021) The molecular organization of EPS can further influence water-holding behaviour. (He et al., 2024; Zhang et al., 2019)

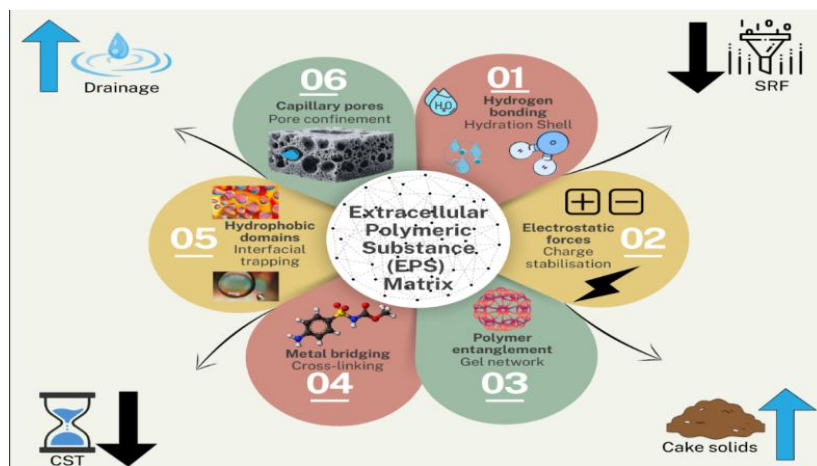


Figure 3. EPS interactions controlling water retention and sludge dewaterability.

As illustrated in Figure 3, interactions between EPS and water contribute to the formation of a hydrated sludge matrix that restricts water mobility. Therefore, modification of EPS composition and structure is an important target for biological dewatering. However, excessive EPS disruption may destabilize flocs or increase soluble and colloidal material, potentially reducing filtration performance. Controlled EPS modification, rather than complete degradation, is therefore desirable for improving sludge dewaterability. (Dai et al., 2018). The accessibility of EPS to microorganisms and enzymes is therefore governed by sludge structure, while controlled EPS modification is essential because excessive disruption may impair floc integrity and dewaterability. (He et al., 2024; Wu et al., 2020)

4. Microbial Processes For Biological Dewatering

4.1. Microbial EPS degradation and consortia

Microorganisms can improve sludge dewaterability by transforming EPS and modifying floc structure. EPS-degrading microbial communities contribute to the hydrolysis of protein- and polysaccharide-rich matrix components, altering EPS stratification and water retention (Li et al., 2021; Yu et al., 2021). Because industrial sludges contain heterogeneous EPS, microbial consortia may provide complementary metabolic functions and sustain transformation of different substrates (Huang et al., 2022; Shi et al., 2017). The effectiveness of microbial treatment depends on the extent of EPS transformation and the resulting changes in sludge structure and water release. Therefore, microbial performance should be evaluated using both EPS characteristics and direct dewaterability indicators.

4.2. Microbially mediated bio-drying

Bio-drying provides a distinct route for moisture reduction. Microbial degradation of readily biodegradable organic matter generates heat, while controlled aeration promotes evaporation and moisture transfer. (Xu et al., 2024). Bio-drying performance is influenced by microbial activity, biodegradable solids, aeration, and initial moisture conditions. (Cai et al., 2016; Li et al., 2022; Li et al., 2019). (Zhang et al., 2024) Further demonstrated that ventilation conditions influence biological moisture removal from dewatered sludge. Unlike EPS-focused conditioning, bio-drying primarily achieves moisture reduction through biological heat generation and mass transfer.

5. Enzymatic Strategies For Biological Dewatering

Enzymatic conditioning provides a targeted approach for modifying EPS components responsible for water retention. Proteases primarily hydrolyse protein-rich EPS, whereas cellulases and amylases act on carbohydrate-containing components; lysozyme can disrupt cellular structures and facilitate the release of matrix-associated material. (Allam et al., 2022; Cabeza et al., 2023). The principal enzymes and their target components are summarized in Table 1. The effectiveness of enzyme treatment depends on substrate accessibility and operating conditions, particularly pH, temperature, enzyme dosage, and sludge solids concentration (Cabeza et al., 2023; Kang et al., 2023). Consequently, enzyme selection should be matched to the dominant biochemical composition of the target sludge rather than applying a universal treatment.

Table 1. Enzymes relevant to biological sludge conditioning

Enzyme/st category	Principal target	Mechanistic role	Evidence relevant to dewatering	Key reference(s)
Protease	Protein-rich EPS	Hydrolysis of proteinaceous matrix components and EPS modification	Can alter sludge water release and dewaterability; response is condition-dependent, and CST may increase despite reduction in water content	(Pei et al., 2010)
Cellulase	Cellulose / carbohydrate-rich polymers	Hydrolysis of carbohydrate-containing matrix components	Can promote matrix hydrolysis and modify water retention; dewatering effects are condition-dependent	(Allam et al., 2022)
α-Amylase	Starch / α -linked polysaccharides	Hydrolysis of starch-containing polymers	Can modify soluble polymer fractions and sludge water content; dewatering response depends on operating conditions.	(Kang et al., 2023)
Lysozyme	Cell-wall / cellular structural components	Cellular disruption and exposure of intracellular or matrix-associated substrates	Can facilitate structural disruption and subsequent hydrolysis; effects on water release depend on treatment conditions.	(Allam et al., 2022; Zou et al., 2023)
Protease + lysozyme	Proteinaceous and cellular components	Sequential/complementary hydrolysis	Enhances sludge hydrolysis and solubilization under optimized dosing; dewatering benefit should be evaluated against appropriate controls.	(Zou et al., 2023)

Cellulase + protease + α-amylase	Multiple protein- and carbohydrate-rich components	Multi-substrate hydrolysis	Promotes solubilization and accessibility; response remains dependent	EPS/biomass and substrate dewatering condition- (Cabeza et al., 2023; Kang et al., 2023)
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Combined enzyme treatments can address different EPS components simultaneously or sequentially. Protease-lysozyme combinations, for example, have been investigated for enhanced sludge solubilization, while enzyme-assisted hydrolysis has also been examined for improving substrate accessibility and resource recovery(Waseem et al.).(Zou et al., 2022). However, improved performance should not automatically be described as synergistic unless the combined treatment demonstrates an advantage over appropriately optimized individual enzymes(Dai et al., 2018). Microorganisms can contribute to EPS disruption through extracellular enzymes with different substrate specificities. Proteases act on structural proteins, DNases on extracellular DNA (eDNA), and glycoside hydrolases on polysaccharide components(Al-Madboly et al., 2024). Their combined action can disrupt the EPS network, reduce matrix stability, and facilitate the release of previously retained water, thereby improving sludge dewaterability (Figure 4).

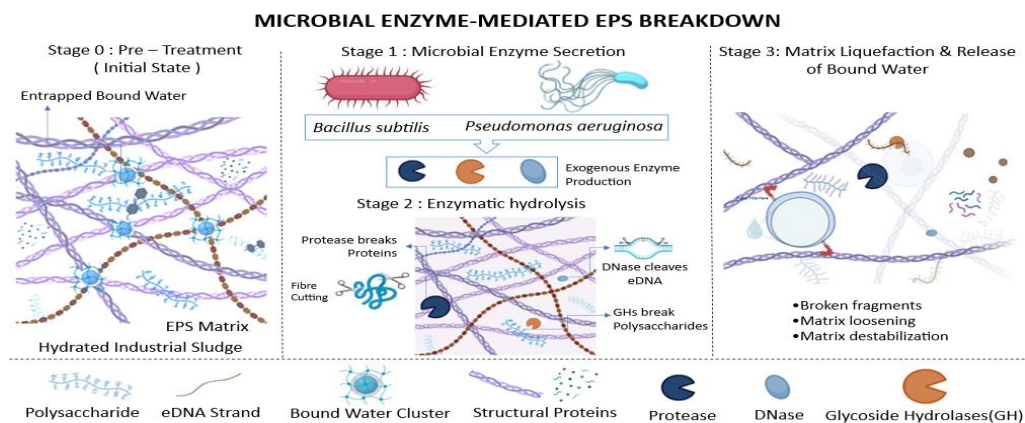


Figure 4. Microbial enzyme-mediated EPS degradation and bound-water release during biological sludge dewatering

6. Microbial - Enzymatic Integration

Microbial and enzymatic treatments can complement each other because they act at different stages of EPS modification. Microorganisms provide continuous biological activity and extracellular enzyme production, whereas externally added enzymes can act directly on accessible EPS components without requiring microbial growth. (Cabeza et al., 2023; Kang et al., 2023). This combination may be useful when the heterogeneous EPS matrix contains substrates that are not readily transformed by a single biological pathway.

However, the advantage of combined treatment should be demonstrated against appropriately optimized individual treatments. Enzyme accessibility, microbial activity, sludge composition, and operating conditions can influence the outcome, while excessive treatment may increase solubilization without improving dewaterability. Sequential application may also be preferable when the optimal conditions for microbial activity and enzyme stability differ. Therefore, integrated microbial - enzymatic systems should be designed according to the target EPS characteristics and evaluated using both biological transformation and direct dewaterability indicators. (Zou et al., 2023).

7. Kinetic Modelling Of Biological Sludge Dewatering

Kinetic modelling can help relate the rate of biological transformation to changes in moisture and dewaterability. (Jensen et al., 2014) For biological sludge treatment, the relevant processes include microbial degradation, EPS hydrolysis and moisture removal. The selected model should therefore reflect the dominant rate-limiting process rather than applying a single kinetic expression across all sludge systems. (Huong et al., 2024; Ling et al., 2025).

Bio-drying provides the clearest example of kinetic application(Li et al., 2019) used kinetic analysis to describe volatile-solids biodegradation and water mass balance during sludge bio-drying, while (Li et al., 2022) related microbial community changes to moisture-ratio dynamics. More recently, (Zhang et al., 2024) reported zero-order moisture-removal kinetics for biological drying of citric-acid-dewatered sludge, with 41.71% water removal over eight days. These studies show that biological activity, aeration, substrate availability, and moisture transfer jointly influence the apparent drying rate. (Table 2) highlighted that Kinetic approaches are relevant to biological sludge dewatering.

Table 2. Kinetic approaches relevant to biological sludge dewatering

Process	Kinetic approach	Main outcome	Major limitation
Microbial biodegradation during bio-drying	Biodegradation kinetics	VS degradation and water removal	Strong dependence on moisture and aeration (Li et al., 2019; Villegas & Huiliñir, 2014)

Bio-drying	Zero-order moisture-removal model	Moisture-removal rate	Sensitive to ventilation and biological activity (Zhang et al., 2024)
Moisture redistribution during bio-drying	Moisture-ratio modelling / water-distribution analysis	Moisture transformation during drying	Does not directly quantify biological reaction kinetics (Chen et al., 2026; Li et al., 2022)
Enzyme-assisted dewatering	Enzyme hydrolysis / treatment-response analysis	EPS modification and dewaterability response	Direct coupling between enzyme kinetics, EPS transformation and CST/SRF remains limited. (Yuan et al., 2025)

For EPS- or enzyme-based conditioning, kinetic modelling should ideally connect substrate degradation or EPS transformation with measurable dewatering responses such as CST, SRF, moisture content, or cake solids. (Yuan et al., 2025). However, direct kinetic data linking enzyme-mediated EPS hydrolysis to dewaterability remain limited. Water-fraction relationships may provide an additional bridge between biological treatment and subsequent mechanical separation. (Janssen et al., 2002). Thus, current models describe individual processes, but a validated framework linking biological reaction rates, EPS and water transformation, and dewaterability remains needed. Table 2 says about Kinetic approaches relevant to biological sludge dewatering

8. Engineering Applications And Scale-Up Considerations

Laboratory studies demonstrate the potential of microbial and enzymatic conditioning, but industrial application requires suitable reactor configuration and control of biological accessibility. Aerated batch, stirred-tank and bed-based systems can provide different levels of mixing, oxygen transfer and solids handling. Enzyme treatment generally requires controlled dosing and contact, whereas microbial processes require conditions that maintain biomass activity and mass transfer. (Verardi et al., 2012).

Scale-up is challenged by sludge heterogeneity. High solids concentration can restrict enzyme penetration and oxygen transfer, while salinity, heavy metals, hydrocarbons and other inhibitors may reduce microbial activity or enzyme stability. Temperature gradients, hydraulic dead zones and inadequate aeration may further reduce treatment efficiency. (Chen et al., 2021; Wu et al., 2020). Techno-economic assessment should consider enzyme or inoculum requirements, reactor and aeration energy, treatment time, downstream dewatering and disposal costs. (Zhou et al., 2025) However, comparable TEA/LCA data for microbial - enzymatic treatment of industrial sludge remain limited. Pilot-scale studies should therefore report both dewatering performance and material- and energy-balance data.

9. Challenges And Future Perspectives

Despite promising results, biological dewatering remains difficult to compare across studies because sludge characteristics, treatment conditions and evaluation methods vary widely. Industrial sludges may contain complex and poorly characterized EPS matrices, which can influence microbial accessibility and enzyme performance. The absence of consistent reporting of EPS fractions, treatment kinetics and dewaterability parameters further limits interpretation of biological treatment efficiency.

Another important limitation is the limited understanding of structure–function relationships within EPS during treatment. Changes in total EPS concentration alone may not explain changes in water distribution or filtration behaviour. Future studies should therefore examine specific EPS fractions and relate their transformation to CST, SRF and moisture content using time-resolved measurements. This would also improve the reliability of kinetic models.

Further research should focus on sludge-specific enzyme selection, microbial community–function relationships and experimentally validated kinetic models. Standardized experimental protocols would enable meaningful comparison among biological treatments and help identify conditions under which microbial or enzymatic conditioning provides a consistent dewatering benefit.

10. CONCLUSION

This review highlights microbial processes and enzymatic strategies as complementary approaches for modifying the structure and water-retention properties of industrial sludge. Microbial activity can transform EPS and support biological moisture removal, while targeted enzymes can act on specific protein- and polysaccharide-rich components of the sludge matrix. The effectiveness of both approaches is closely related to sludge characteristics, EPS composition and treatment conditions. Kinetic modelling provides an additional basis for understanding the rates of biological transformation and moisture removal. Integrating these kinetic descriptions with changes in EPS, water distribution and dewaterability can provide a clearer understanding of treatment performance. Overall, the combination of microbial processes, enzymatic strategies and kinetic modelling provides a coherent framework for evaluating biological dewatering of industrial sludges and for guiding its development towards more predictable and practical treatment systems.

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