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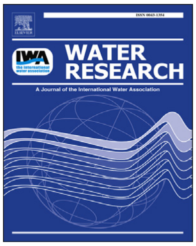

The important implications of particulate substrate in determining the physicochemical characteristics of extracellular polymeric substances (EPS) in activated sludge

Bin-Bin Wang [a,b,*], Dang-Cong Peng [a,b], Yin-Ping Hou [a,b], Hui-Juan Li [a,b], Li-Ying Pei [a,b], Li-Fang Yu [a,b]

[a] School of Environmental and Municipal Engineering, Xi'an University of Architecture and Technology, Xi'an 710055, China

[b] Key Laboratory of Northwest Water Resource, Environment and Ecology, Ministry of Education, Xi'an 710055, China

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ABSTRACT

Since the notable amount of particulate substrate in wastewater, the implications of particulate substrate on treatment efficiency have been a topic of major interest in the field of biological wastewater treatment. The particulate substrate has to be hydrolyzed by the extracellular enzymes, which are mainly embedded in extracellular polymeric substances (EPS) matrix of microbial aggregates, prior to consumption. Therefore, the important relevance between the particulate substrate and the characteristics of EPS can be expected. In this study, two lab-scale sequencing batch reactors were performed in parallel to investigate the effects of particulate and soluble substrate on the physicochemical characteristics of EPS in activated sludge. The results showed that the particulate substrate in the influent could significantly change the properties of activated sludge and the characteristics of EPS. More open and fluffy flocs with poorer settleability and dewaterability were formed with particulate substrate. More protein and humic compounds were introduced into the EPS matrix due to the deep involvement of protein and humic compounds in hydrolysis process of particulate substrate. The increments of protein and humic compounds then caused the slight higher molecular weight, higher hydrophobicity and lower zeta potential of EPS in particulate substrate system. The results in this study permitted for obtaining answers to understand the significant implications of particulate substrate in determining the physicochemical characteristics of EPS in biological wastewater treatment systems.

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* *Corresponding author*. School of Environmental and Municipal Engineering, Xi'an University of Architecture and Technology, $13^{\#}$ Yanta Road, Xi'an 710055, China. Tel.: +86 (0)29 82202729; fax: +86 (0)29 82202729.
E-mail address: bbwangchn@gmail.com (B.-B. Wang).

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1. Introduction

Particulate substrate has been reported to be an important fraction of total COD present in wastewater (Martins et al., 2011). Levine et al. (1985) stated that non-soluble organic matter can account for 75% of the organic load in wastewater treatment systems. In Netherlands, 50% of the total COD in municipal wastewater is contributed by particulate substrate (Roeleveld and van Loosdrecht, 2002). Even higher values were reported, 60% in northern Poland (Czerwionka et al., 2008), 70–90% in South Africa (Casey et al., 1999) and about 90% in Switzerland (Kappeler and Gujer, 1994).

Since the notable amount of particulate substrate in wastewater, the implications of particulate substrate on treatment efficiency has been a topic of major interest in the field of biological wastewater treatment. Martins et al. (2011) reported that particulate substrate has positive effect on the sludge settleability. Drewnowski and Makinia (2011) stated that the removal of colloidal and particulate COD in wastewater leads to the reduction of denitrification, phosphate release and phosphate uptake rates. Gori et al. (2011) investigated the effects of soluble and particulate substrate on the carbon and energy footprint of wastewater treatment processes and found that the increase of soluble COD in wastewater increases the energy demand on the aeration reactors. de Kreuk et al. (2010) studied the behavior of particulate substrate in an aerobic granular sludge system and demonstrated that the substrate gradients formed by the hydrolysis of particulate substrate cause irregular filamentous outgrowth on the granules. The effects of particulate substrate on microfauna populations were even investigated (Puigagut et al., 2007). However, few studies were reported to compare the effects of particulate and soluble substrate on extracellular polymeric substances (EPS), an important high molecular weight mixture of polymers, of microbial aggregates in biological wastewater treatment systems. The EPS have a significant influence on the physicochemical properties of microbial aggregates, such as floc structure (Dignac et al., 1998; Adav et al., 2008), surface charge (Wilén et al., 2003; Sponza, 2003), flocculation, settling and dewatering properties (Liu and Fang, 2003; Li and Yang, 2007), and adsorption ability (Sheng et al., 2008; Wang et al., 2012). Therefore, the implications of particulate substrate on the physicochemical characteristics of EPS in microbial aggregates are pressingly needed to be evaluated.

Because only monomers and oligomers are able to cross the bacterial membrane via specific active transports, particulate substrate cannot be directly assimilated by microorganisms. To be taken up by the bacteria, these high molecular weight polymers must be hydrolyzed by the extracellular enzymes (Frølund et al., 1995; Nybroe et al., 1992). Many studies have found that hydrolytic enzymes are associated with EPS in microbial aggregates, rather than release into the bulk water (Frølund et al., 1995; Guellil et al., 2001; Cadoret et al., 2002). Cadoret et al. (2002) reported that the higher the molecular weight of a substrate, the more hydrolytic activity is associated with the EPS. Frølund et al. (1995) said that the exoenzymes should be an integrated part of the EPS. Therefore, the amount of particulate substrate in the influent is more likely to affect the exoenzyme content (protein) in EPS and change the composition of EPS in microbial aggregates. This assessment points out the important relevance between the particulate substrate and the characteristics of EPS.

Accordingly, the present work aims to investigate the influence of particulate and soluble substrate on the physicochemical characteristics including composition, excitation and emission matrix (EEM), molecular weight (MW), hydrophilic/hydrophobic distribution and zeta potential of EPS in activated sludge. This type of study can permit for obtaining answers to understand the important implications of particulate substrate in determining the physicochemical characteristics of EPS in activated sludge.

2. Material and methods

2.1. *Experimental setup*

The experiment was performed in parallel in two sequencing batch reactors (SBR) with different form of substrates. The reactors were controlled at a temperature of 25 ± 1 °C and pH of 6.8–7.3. Liquor mixing was achieved using a vertical mixer with standard three blade impeller. Oxygen was provided by means of aquarium air pumps. The operating conditions, such as feed rate, mixing time, settling time and withdraw rate, were controlled automatically by programmable logic controller. The dissolved oxygen and pH were monitored online by the probes (Mettler-Toledo, Switzerland) which connected to a data acquisition program. The reactors were operated with 2 min of influent filling, 268 min of reacting, 60 min of settling, 5 min of effluent withdrawing, and 25 min of idling. The applied organic loading rate was 0.8 g COD L^{-1} d^{-1}. The exchange ratio was controlled in 0.5 which corresponded to a 12 h of hydraulic retention time. The sludge retention time was kept in 10 d through discharging 500 mL mixed liquor daily.

2.2. *Feed medium*

Depending on the SBR system, the feed wastewater contained sweet potato starch (SPS) or soluble glucose (SolG) as model substrate to simulate particulate and soluble organic matter, respectively. All other components in the feed medium were identical. The synthetic wastewater used as influent was stored in a refrigerator at 4 °C to prevent the hydrolysis of SPS. The detailed compositions of synthetic wastewater were shown as the following in mg L^{-1} (except for trace element solution): COD (SPS/SolG), 400; NH_4Cl, 114.6; KH_2PO_4, 26.3; $NaHCO_3$, 50; trace element solution, 50 μL $L^{-1}{}_{\text{influent}}$. The trace element solution contained in g L^{-1}: $FeSO_4{\bullet}7H_2O$, 8; $MnCl_2{\bullet}4H_2O$, 0.5; $CoCl_2{\bullet}6H_2O$, 0.88; $CuCl_2{\bullet}2H_2O$, 0.035; $ZnSO_4{\bullet}7H_2O$, 0.1; H_3BO_3, 0.1; EDTA, 0.05; $NiCl_2{\bullet}6H_2O$, 0.036; $(NH_4)_6Mo_7O_{24}{\bullet}4H_2O$, 0.64; $MgSO_4{\bullet}7H_2O$, 5. All the solutions were prepared by the pure water.

2.3. *EPS extraction*

Cation exchange resin (CER) method was used for the EPS extraction in this experiment. The detailed procedure of EPS