Contents lists available at ScienceDirect

Process Safety and Environmental Protection

journal homepage: www.elsevier.com/locate/psep

# Investigation of the impact of immobilized cells and the nitrification process using a coupled moving bed biofilm reactor and activated sludge bioreactor for biodegradation of high concentrations of dimethyl formamide

*Seyed Ali Rahmaninezhad*[a,*], *Hamideh Fathi*[b], *Ali Reza Pendashteh*[c], *Naz Chaibakhsh*[d], *Babak Tavakoli*[e]

[a] *Department of Environmental Engineering, University of Tehran, Tehran, Iran*
[b] *Environmental Research Institute, Iranian Academic Center of Education, Culture and Research (ACECR), Rasht, Iran*
[c] *The Caspian Sea Basin Research Center, University of Guilan, Rasht, Iran*
[d] *Department of Chemistry, Faculty of Science, University of Guilan, Rasht, Iran*
[e] *Environmental Science Department, Faculty of Natural Resources, University of Guilan, Rasht, Iran*

ARTICLE INFO

*Article history:*
Received 29 August 2015
Received in revised form 7 March 2016
Accepted 24 April 2016
Available online 10 May 2016

*Keywords:*
Dimethyl formamide
Immobilized cell
Moving-bed biofilm reactor
Activated sludge
Thickness of biofilm

ABSTRACT

In this study investigating the effect of immobilized cells on dimethyl formamide (DMF) biodegradation, the performance of a mixed-bed biofilm reactor (MBBR) was evaluated with an activated-sludge reactor (AS). MBBR and AS bioreactors could biodegrade 9000 mg/l COD of feed DMF with DMF removal efficiencies of 55.6% and 43.6%, respectively, after 7 days. In the next step, when nitrifying microorganisms were fed to the AS bioreactor, then coupled with MBBR, 50% of the treated wastewater of this coupled bioreactor was recycled to the feed point, with the DMF removal efficiency of the coupled bioreactor for 12,000 mg/l COD reaching 94.17% after 10 days. In contrast to MBBR and AS bioreactors being used by themselves (as opposed to together), which tends to increase pH due to the ammonia produced during DMF biodegradation, in a coupled bioreactor the completed nitrification process caused reduction of more than 98.80% of the ammonia, with the pH remaining neutral as a result. Furthermore, the concentration of microorganisms stayed stable for all organic loading rates. During this experiment, the thickness of biofilm increased gradually at the rate of 1.2, 0.13, and 0.36 μm per day in batch phase, continuous phase in MBBR, and coupled bioreactor, respectively.



## 1. Introduction

Dimethyl formamide (DMF: $(CH_3)_2NCHO$) is a water-miscible polar industrial solvent that has several applications in chemical industries, including the production of flat-plate membranes, synthetic polymers, textiles, organic chemicals; and in the pharmaceutical and agrochemical industries (Swaroop et al., 2009; Kamimoto et al., 2009). Annually, a considerable amount of this material is released into the environment as the effluent of industrial factories due to its various uses (Birnboim and Doly, 1979; Hasegawa et al., 1999; Dziewit et al., 2010;

* *Corresponding author*. Tel.: +98 9125734326; fax: +98 1333364028.
E-mail address: ali.rahmaninezhad@ut.ac.ir (S.A. Rahmaninezhad).
http://dx.doi.org/10.1016/j.psep.2016.04.022
0957-5820/

**Table 1 – Composition of sugar-manufacturing synthetic wastewater.**

| Chemical | Amount (mg/l) | Chemical | Amount (mg/l) |
|---|---|---|---|
| $CaCl_2 \cdot 2H_3O$ | 20.5 | $MgSO_4 \cdot 7H_2O$ | 10.0 |
| $NaHCO_3$ | 9.20 | $NiSO_4 \cdot 7H_2O$ | 0.083 |
| $KH_2PO_4$ | 24.0 | Ammonia-nitrogen | 15.25 |
| $ZnCl_2$ | 0.25 | Ammonium phosphate | 17.35 |
| $FeSO_4 \cdot 7H_2O$ | 0.208 | Urea | 80.15 |
| $MnCl_2 \cdot 4H_2O$ | 0.048 | COD | 750 |

Bromley-Challenor et al., 2000). It is estimated that approximately $2.7 \times 10^5$ tons of DMF wastewater are produced annually worldwide (Johnson and Yagi, 2002; Marsella, 1994), and demand for DMF continues to increase (Dziewit et al., 2010; Lu, 2004; Yang et al., 2014). Therefore, because of its widespread use, DMF is commonly found in different concentrations at industrial effluents, leading to environmental pollution (Hasegawa et al., 1999; Bromley-Challenor et al., 2000).

Waste DMF is treated primarily by using physicochemical processes such as distillation, absorption, and adsorption (Kamimoto et al., 2009). Since DMF is water miscible, it is difficult to remove it effectively through these physicochemical processes (Kamimoto et al., 2009). Due to the slow rate of DMF's chemical degradation (Swaroop et al., 2009), seeking biological treatment methods might represent a viable alternative for the treatment of DMF from industrial effluents and DMF-polluted sites (Swaroop et al., 2009; Ghisalba et al., 1985).

Fortunately, there are distinct soil-isolated microorganisms from areas polluted with DMF (Sanjeev Kumar et al., 2012; Urakami et al., 1990) and also in the sludge of municipal wastewater (Swaroop et al., 2009) that uses DMF as the sole source of carbon and nitrogen (Dziewit et al., 2010; Veeranagouda et al., 2006; Swaroop et al., 2009; Hasegawa et al., 1997; Bromley-Challenor et al., 2000) that could biodegrade DMF effectively (Okazaki et al., 1995; Bromley-Challenor et al., 2000; Kamimoto et al., 2009). Most of the available research indicates that sole use of aerobic microorganisms could effectively biodegrade DMF (Kamimoto et al., 2009; Bromley-Challenor et al., 2000). Moreover, the C/N ratio of DMF is 2.57, so it is important for biological DMF treatment processes to be able to effectively remove both nitrogen and organic carbon. Nitrogen could not be removed through use of a conventional aerobic activated sludge system, but was removed effectively by an anoxic–aerobic process (Kamimoto et al., 2009).

While the free cells (suspended growth bioreactors) fail to grow at higher concentrations of DMF due to toxic effects (Sanjeev Kumar et al., 2012; Yang et al., 2014; Veeranagouda et al., 2006), entrapped bacterial cells in a suitable matrix have improved tolerance to a variety of toxic and recalcitrant compounds (Sanjeev Kumar et al., 2012; Chen et al., 2008; Sarma and Pakshirajan, 2011; Tepe and Dursun, 2008; Zhao et al., 2009; Wang, 2002). Therefore, attached growth bioreactors such as moving-bed biofilm reactors (MBBRs) are suitable choices to biodegrade high DMF concentrations.

There are two possible pathways for aerobic decomposition of DMF (Kamimoto et al., 2009; Dziewit et al., 2010; Ghisalba et al., 1985; Swaroop et al., 2009). The first (used by some *Pseudomonas* strains) involves repeated oxidative demethylation to produce methylformamide and then formamide, which is further cleaved to ammonia and formate by formamidase. The second pathway depends on N,N-dimethylformamidase (DMFase), which splits DMF into formate and N,N-dimethylamine (DMA). DMA is further degraded by dimethylamine dehydrogenase to formaldehyde and methylamine, which is finally converted into ammonia and formaldehyde (Ghisalba et al., 1985; Hasegawa et al., 1997).

Several studies exist detailing the biodegradation of DMF as sole carbon source (Yang et al., 2014; Kamimoto et al., 2009; Dziewit et al., 2010; Swaroop et al., 2009; Sanjeev Kumar et al., 2012; Chou and Wu, 1998), but very few of them use a high-feed concentration of DMF (more than 1 g DMF/l). In the overwhelming majority of these studies, high biodegradability of DMF was reported. To the authors' knowledge, there is only one study in which high concentrations of DMF were used (more than 10 g DMF/l) (Sanjeev Kumar et al., 2012), and most of them had focused on low-feed DMF concentrations (under 5 g DMF/l); therefore, in this study the biodegradability of high feed concentrations of DMF (up to 12 g DMF/l) were investigated to evaluate the performance of bioreactors of free and immobilized cells and the impact of the completed nitrification process on DMF biodegradation.

## 2. Materials and methods

### 2.1. *Chemicals*

In this experiment, DMF was the sole carbon and nitrogen source (Bromley-Challenor et al., 2000) and potassium dihydrogen phosphate was added to the feed due to the need for phosphorus (Borghei et al., 2008) and because the nitrogen content of DMF is equal to C/N: 2.57 (Kamimoto et al., 2009). The ratio of COD/N/P was set to 100/50/10. All chemical compounds used were obtained from Merck (Darmstadt, Germany). In addition, in this experiment, beet sugar molasses (the composition of sugar-manufacturing synthetic wastewater) (Borghei et al., 2008) with a COD of 50 mg/l was added to the AS bioreactor as a carbon source for facilitating the nitrification process. The characteristics of molasses are shown in Table 1.

### 2.2. *Microorganisms and carriers conditions*

The sludge that was used in this experiment was obtained from the return sludge of the wastewater treatment plant of a DMF-producing factory in Isfahan, Iran. This sludge was used because it might consist of strains that use DMF as a source of carbon and energy (Dziewit et al., 2010). For the nitrification process, the sludge was obtained from a nitrification tank in the EKBATAN municipal wastewater treatment plant in Tehran, Iran.

The carriers used in the attached bioreactor were made by Lutz-JESCO (Rochester, New York, USA) and their characteristics are indicated in Table 2.

### 2.3. *Bioreactors*

During the experiment, two types of bioreactors made with polymethyl methacrylate were used. Suspended growth and

**Table 2 – The characteristics of carries.**

| Parameter | Amount | Unit |
|---|---|---|
| Material | HDPE | – |
| Shape | Cylindrical with cross in inside | – |
| Diameter | 10 | mm |
| Height | 7 | mm |
| Bulk specific surface area | 857 | $mm^2$ |
| Number in one liter | 604 | – |
| Net specific surface area | 517 | $m^2/m^3$ |
| Density | 0.96 | $g/cm^3$ |
| Average weight | 0.17 | g |