

# Bacterial Flora in Intermittent Aerating Membrane Bioreactor

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## Introduction

Recently membrane separation processes have been applied for domestic and industrial wastewater treatment. Membranes, as a solid-liquid separation device combining with conventional biological treatment processes (e.g. activated sludge etc.) replace or modify the need of a secondary sedimentation tank. Minimum sludge wastage by maintaining low F/M ratio resulted in high biomass concentration within the system. Moreover, the process does not suffer from bulking problem in long sludge age condition since it does not allow any bacterial washout from the system. In this study, bacterial flora and sludge characteristics in long sludge age and starved condition were examined by respiratory quinone profile analysis and observation through microscope and scanning electron microscope.

## Materials and Methods

**Experimental system** The schematic of experimental system is shown in Fig.1. The system consists of two parts; main bioreactor and separation unit. The separation unit, with volume of 10 l, was immersed into the main bioreactor with volume of 62 l. Two hollow fiber microporous membrane of  $0.03\ \mu\text{m}$  pore size and  $0.3\ \text{m}^2$  surface area each were put in the separation unit. Permeate was extracted by a suction pump under intermittent operation. Aeration was intermittently supplied to the system in 90 minute cycle (90 minute for non-aerating period and 90 minute aerating period) to achieve nitrogen removal by simultaneous nitrification and denitrification. In aerating period DO was maintained at 1.5-2.0 mg/l. pH of mixed liquor was in the range of 6.5-7.5 and temperature was kept in the range of 25-29°C.

**Substrate** Synthetic wastewater was used in this study. Substrate composition of synthetic wastewater is shown in Table 1. Concentrated feed solution was stocked in the refrigerator and diluted with tap water to desired COD concentration prior to application. In addition to

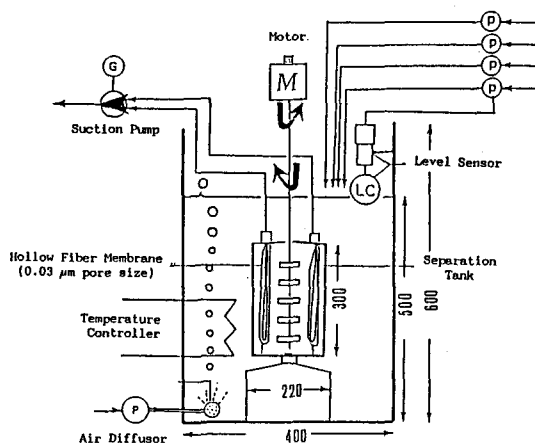


Fig 1. Schematic of experimental system

Table 1. Composition of synthetic substrate

$\text{CH}_3\text{COONa} \cdot 3\text{H}_2\text{O}$	22	g/l
Peptone	10	g/l
Yeast extract	1	g/l
$\text{K}_2\text{HPO}_4$	3.2	g/l
$(\text{NH}_4)_2\text{SO}_4$	16	g/l
$\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$	4	g/l
$\text{MnSO}_4 \cdot 4\text{H}_2\text{O}$	0.4	g/l
$\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$	0.02	g/l
$\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$	0.4	g/l
Toilet paper	1.5	m/d

the synthetic substrate, toilet paper (being shredded and mixed with water before application) was also applied to the system at the rate of 1.5 m/day.

**Analysis of respiratory quinone profile** Quinone profile of sludge was analysed according to the procedures described by Hiraishi (1988,1990,1991) with some modifications. Sludge was harvested by centrifugation and dried using freeze dryer (Eyela, FDU-830) for 24 hrs. Dried sludge was extracted twice by chloroform-methanol mixture (2:1,v/v) with rotary shaker (200 RPM) for 2 hrs and filtered. The filtrate was then evaporated and dissolved in small amount of acetone followed by purification using thin-layer chromatograph in hexane:diethyl ether mixture. (85:15,v/v) Quinone components were separated by reverse-phase HPLC(Shimadzu, LC-6A) and then identified and determined their concentration with some standard quinones.

## Results and Discussion

**Sludge characteristics of membrane separation bioreactor** Sludge taken from membrane separation bioreactor has light brown color with low settleability (SV30>80%). Observation of sludge through microscope showed that bulking of sludge in the system was caused by dispersed flocs under long sludge age operation. However, there was no existence of filamentous bacteria under this single completely-mixed tank operated under low F/M ratio.(Fig.2) Microbial observation also showed some protozoa which were generally found in high degree or complete oxidation process.

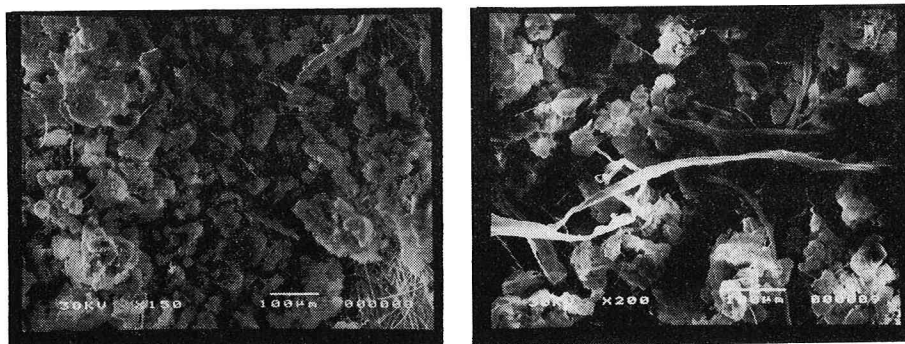


Fig.2. Microbial observation of sludge from membrane separation bioreactor through scanning electron microscope

**Bacterial flora in membrane separation bioreactor** Table 2. shows distribution of quinone composition together with their classification (after Hiraishi,1990). In quinone system of the sludge, Q-8 predominated ubiquinone system followed by Q-10 component. Other ubiquinone components presented in small percentage. MK-8( $H_4$ ) and MK-7 were the major menaquinone components in sludge. Interpretation of analysis showed that *Pseudomonas*, *Paracoccus* and *Coryneform* bacteria might be major groups of bacteria in the system. The results obtained showed some agreement with those profiles of batch anaerobic- aerobic system. (Hiraishi,1990) Ratio of menaquinone to ubiquinone varied in wide range of 0.5-1.5 depending on the system operation.

### Change of bacteria flora along process operation

Although analysis of sludge quinone system showed some fixed pattern in quinone composition, there was some fluctuation of quinone contents observed. Fig 3 showed some change in quinone system during deterioration of nitrogen removal caused by inhibition of nitrification from insufficient supply of oxygen. Decrease in ubiquinone content was observed in such period whereas menaquinone content remained almost constant. This might be due to sensitivity of ubiquinone contents of strict aerobes against oxygen concentration in the system.

Table 2. Distribution of quinone composition and identification of bacteria of sludge taken from membrane separation bioreactor

Quinone category and taxonomy	Percentage of distribution
Ubiquinone	100
Q-6	0.59-5.08
Q-7	1.29-3.15
Q-8	<i>Comamonas-Pseudomonas</i>
	<i>Alcaligenes</i>
Q-9	<i>Acinetobacter,Pseudomonas</i>
Q-10	<i>Paracoccus,Methylobacterium</i>
Menaquinone	100
MK-6	<i>Flavobacterium-Cytophaga</i>
MK-7	<i>Flavobacterium-Cytophaga</i>
	<i>Staphylococcus,Bacillus</i>
MK-8	<i>Aeromonas (with Q-8)</i>
MK-8(H <sub>2</sub> )	<i>Micrococcus,Coryneform</i>
MK-8(H <sub>4</sub> )	<i>Coryneform</i>
MK-9(H <sub>2</sub> )	
MK-9(H <sub>4</sub> )	

Note: Identification of bacteria was referred from Hiraishi (1990)

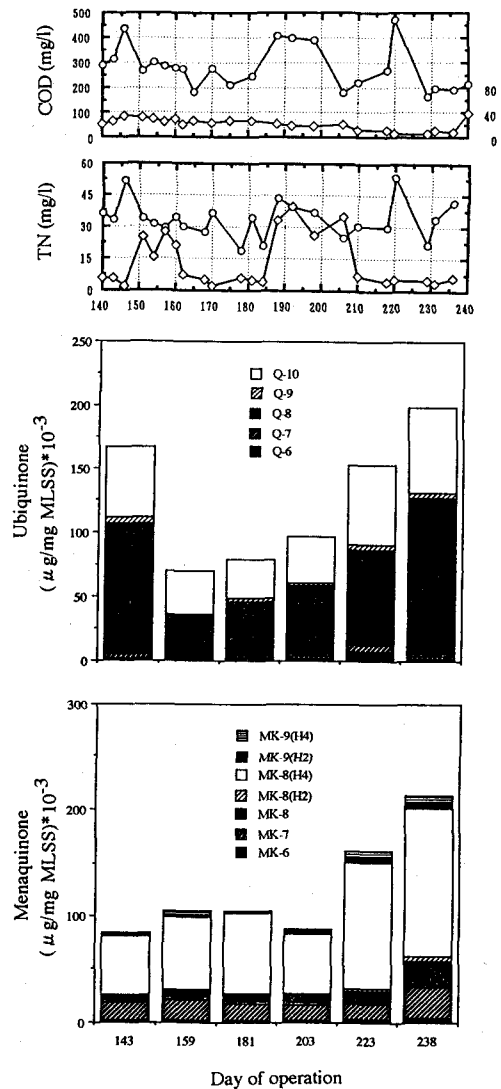


Fig 3. Change of bacterial flora along process operation

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