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Biological Treatment of Formaldehyde by Activated Sludge

(formaldehyde/activated sludge/formalin waste)

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The effect of formaldehyde (HCHO) on activated sludge was investigated. More than 3.3 mg/l of HCHO showed lag type toxicity for O_2 uptake of nonacclimated sludge. Acclimation studies for highly concentrated HCHO showed that activated sludge could remove 90% of 5,500 mg/l HCHO at 40 hours. Changes in pH, COD, and sludge weight during HCHO removal were investigated. In glucose-containing medium, sludge weight increased. Oxidation of glucose and increase of sludge weight occurred when HCHO was removed more than 80% and pH raised to 5.5. Optimal pH for HCHO removal by acclimated sludge was 6. Those results suggest that activated sludge oxidizes HCHO but are not utilized as a carbon source; addition of a carbon source such as glucose is necessary for treatment of highly concentrated HCHO.

Formalin is used as a disinfectant for laboratory tools and a preservative for pathologic specimens in hospitals and laboratories. The inflow of formalin waste often causes damage to biological waste water treatment plants. In Shimane Medical University, formalin waste of about 3% HCHO is recovered in large quantities. Recovered waste is treated by combustion, but the treatment is not economical. In recent years chemical treatments by slaked lime and hydrogen peroxide have been investigated as pretreatments before biological treatment for formalin waste

(1-3), but studies on biological treatment are few.

In this study, the effects of HCHO on activated sludge and the possibility of biological treatment for highly concentrated HCHO were investigated.

MATERIALS AND METHODS

Activated sludge

Activated sludge was collected from the aeration tank of the domestic waste water treatment plant of Shimane Medical University. The collected sludge was washed twice with medium prepared with the dilution medium of BOD (Biochemical Oxygen Demand) (4) shown in Table 1.

Table 1. Composition of Culture Media for O_2 uptake measurement

	(mg/l)
$MgSO_4 \cdot 7H_2O$	22.5
$CaCl_2 \cdot 2H_2O$	36.5
$FeCl_3 \cdot 6H_2O$	0.25
K_2HPO_4	21.8
KH_2PO_4	8.5
$Na_2HPO_4 \cdot 12H_2O$	44.6
NH_4Cl	1.7
Formaldehyde	0-500
Glucose	300
pH	7.2

Table 2. Composition of Culture Media for Acclimation Study

	Concentration (mg/l)							
Culture media	A	B	C	D	E	F	G	H
NH_4Cl	50	50	150	150	250	500	500	1500
$CaCl_2 \cdot 2H_2O$	7	7	21	21	35	70	70	210
$MgSO_4 \cdot 7H_2O$	5	5	15	15	25	50	50	150
KCl	7	7	21	21	35	70	70	210
KH_2PO_4	11	11	33	33	55	110	110	330
$Na_2HPO_4 \cdot 12H_2O$	29	29	87	87	145	290	290	870
$FeCl_3 \cdot 6H_2O$	1	1	3	3	5	10	10	30
Glucose	100	100	100	300	500	1000	1000	3000
Formaldehyde	100	100	300	500	1100	2500	3000	5500

pH was adjusted to 7.2

O_2 uptake curve

Various concentrations of HCHO containing the media shown in Table 1 were incubated by stirring at 20°C with 100 mg/l of activated sludge. O_2 uptake was measured and the curve recorded continuously by a coulometric BOD autoanalyzer (Yanagimoto Seisakusho Co., Japan).

Acclimation of activated sludge for HCHO

The culture media used for acclimation are shown in Table 2. These were prepared according to the method described for artificial waste by Sudo(5). Medium A was incubated by stirring at 20°C with 1,700 mg/l of activated sludge. When HCHO in medium A was almost completely removed, the components of B were added and incubated. In the same manner, the concentration of HCHO and other components was increased step by step to medium H.

Analysis

1) Formaldehyde: The concentration of HCHO was measured by the acetylacetone method (6).

2) COD (Chemical Oxygen Demand): Samples were filtrated through No.5C filter paper (Toyo Kagaku Sangyo Co., Japan) and measured according to the COD measurement method specified in the Japanese Industrial Standards (4).

3) Activated sludge weight: Samples were filtrated through GS-25 glassfiber filterpaper (Toyo Kagaku Sangyo Co., Japan), and the filterpaper was dried to measure the weight of the sludge.

RESULTS

Fig.1 shows the O_2 uptake curve for activated sludge when various concentrations of HCHO were added to the glucose-containing medium. The O_2 uptake curves for 1.0 mg/l and 3.3 mg/l HCHO were similar to the control; however, the curves for 6.7 mg/l, 50 mg/l, 100 mg/l, and 500 mg/l HCHO showed 28, 30, 30, and 60-hour lags, respectively. After the lag, O_2 uptake occurred at a high rate due to the oxidation of glucose and HCHO. The results show that less than 3.3 mg/l HCHO has no effect on O_2 uptake, and more than 6.7 mg/l HCHO inhibits the normal function of activated sludge and causes an extension of lag time; but when the sludge was acclimated for each concentration of HCHO, it

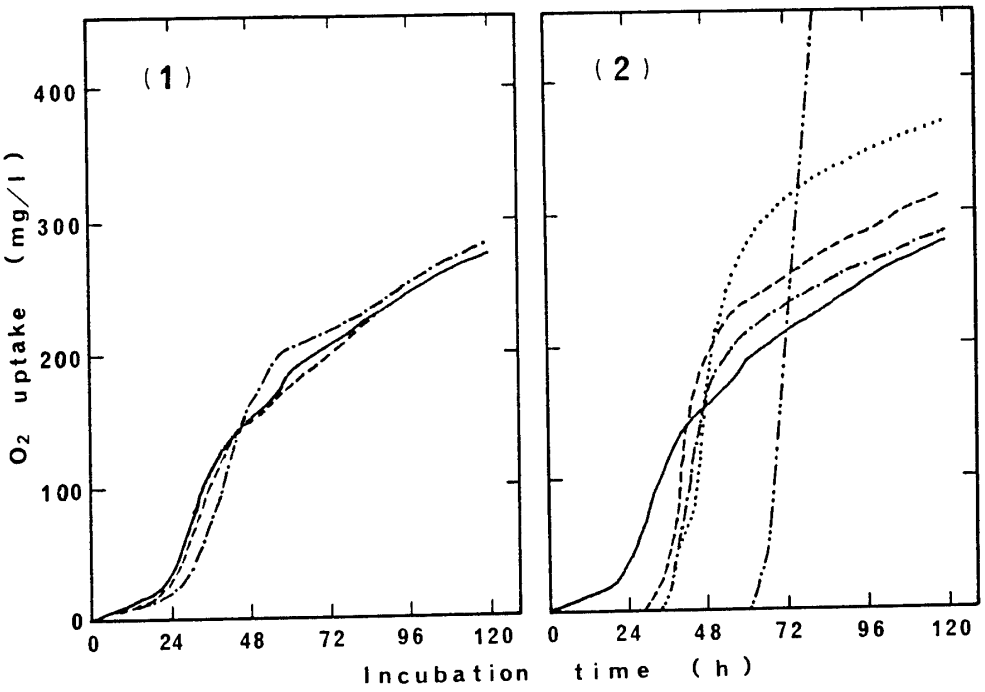

Fig.1. Effect of the concentration of HCHO on O_2 uptake by activated sludge.
HCHO concentration (mg/l)
(1) ——— :0.0, - - - - - :1.0, —·— :3.3
(2) ——— :0.0, - - - - - :6.7, —·— :50, ····· :100, —··— :500

showed strong activity.

Activated sludge collected from the aeration tank was incubated by stirring at 20°C for one to 24 hours with various concentrations of HCHO. After incubation, protozoa in the sludge were examined by microscope. *Vorticella convallaria*, *Epistylis* sp. and *Aspidisca lynceus* were predominantly observed. When the activated sludge was incubated for 24 hours with 2, 5, and 10 mg/l HCHO, the three species of protozoa behaved similarly to those in the control. The addition of 20 mg/l HCHO disturbed the ciliary movement of *Vorticella convallaria* and *Epistylis* sp. after one hour. The movement of *Aspidisca lynceus* was inhibited after one hour's exposure to 30 mg/l HCHO.

Fig.2 shows the acclimation of activated sludge for HCHO. The HCHO concentration was increased step by step from 100 mg/l to 5,500 mg/l. HCHO and other components are shown in Table 2. With the initial 100 mg/l HCHO, over 300 hours was required for complete removal (Fig.2-A), whereas only 8 hours was required when the same concentration was applied a second time (Fig.2-B). The HCHO removal rate was increased by acclimation. In the case

of a non-glucose medium, the removal rate was maximal at 2,500 mg/l (Fig.2-F), and the addition of 5,500 mg/l decreased the removal rate, which was about 50% at 300 hours (Fig.2-H). In the case of glucose-containing medium, the removal rate was maximal at 5,500 mg/l. This operation was carried out for about 40 days by using initially added activated sludge, which continued to remove HCHO throughout the operation.

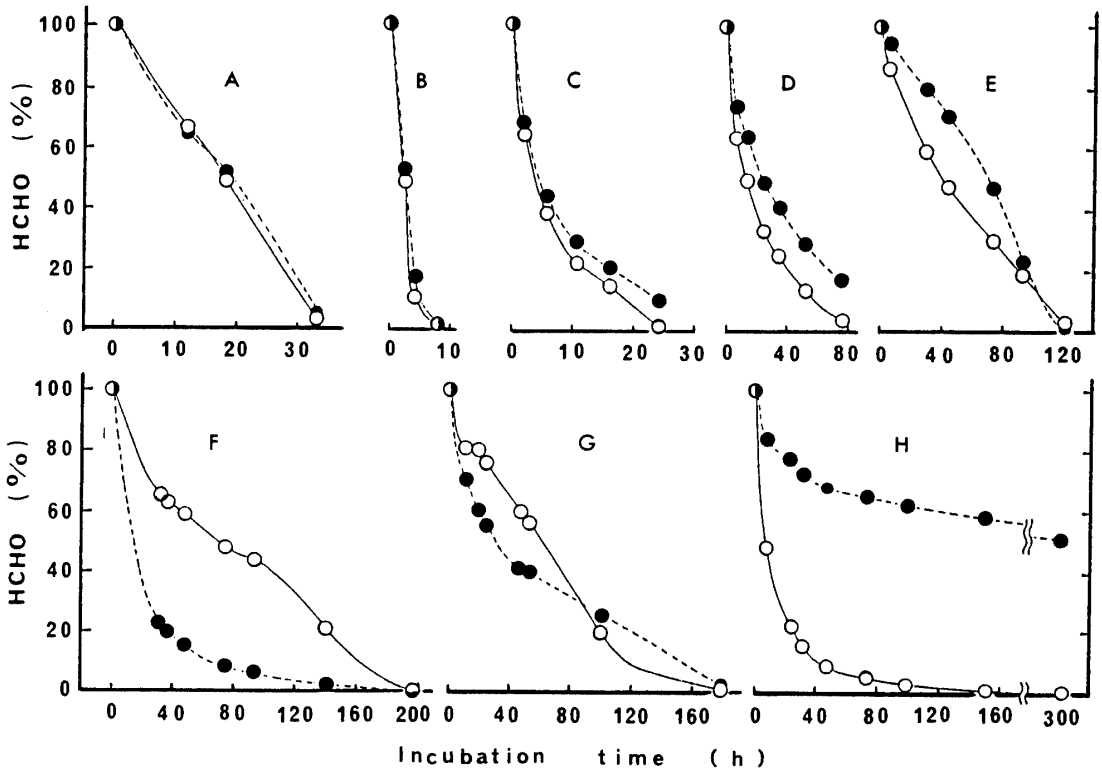

Fig.2. Acclimation of activated sludge for various concentrations of HCHO. The composition of culture media are shown on Table 2.
●---● :non-glucose, o——o :glucose-containing

Changes in pH, COD, and activated sludge weight during HCHO removal were investigated by using 300 mg/l HCHO-acclimated activated sludge. As shown in Fig.3, in non-glucose and glucose-containing media, HCHO removal and changes in pH were highly similar to each other. HCHO removal caused a decrease in pH. When about 50% of HCHO was removed, pH was minimal, and further removal caused an increase in pH. COD decreased with HCHO removal. Fig.4 shows changes in pH and sludge weight. In the case of non-glucose, no appreciable change in sludge weight was observed, but when the medium contained glucose, the weight increased about 200 mg/l when more than 80% HCHO was removed and pH was raised to about 5.5.

Fig.5 shows the effect of pH on HCHO removal by 1,000 mg/l HCHO acclimated sludge. The optimal pH for HCHO removal was about

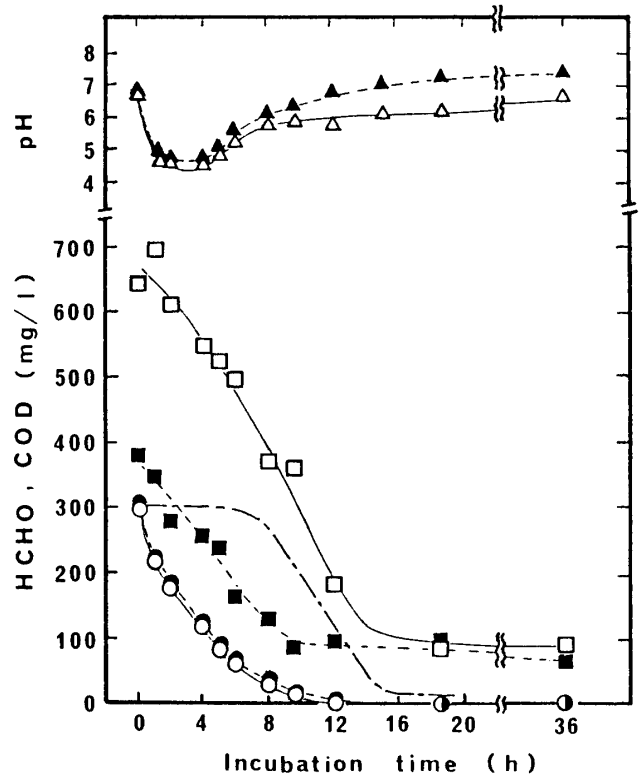

Fig.3. Time course of HCHO(●,○) and COD(■,□) removal by activated sludge and changes in pH(▲,△).300 mg/l of HCHO(●,■,▲) or 300 mg/l of HCHO and 300 mg/l of glucose(○,□,△) containing media were incubated by stirring at 20°C with activated sludge acclimated for 300 mg/l of HCHO. Curve(—·—) shows COD difference between glucose-containing(□) and non-glucose(■).

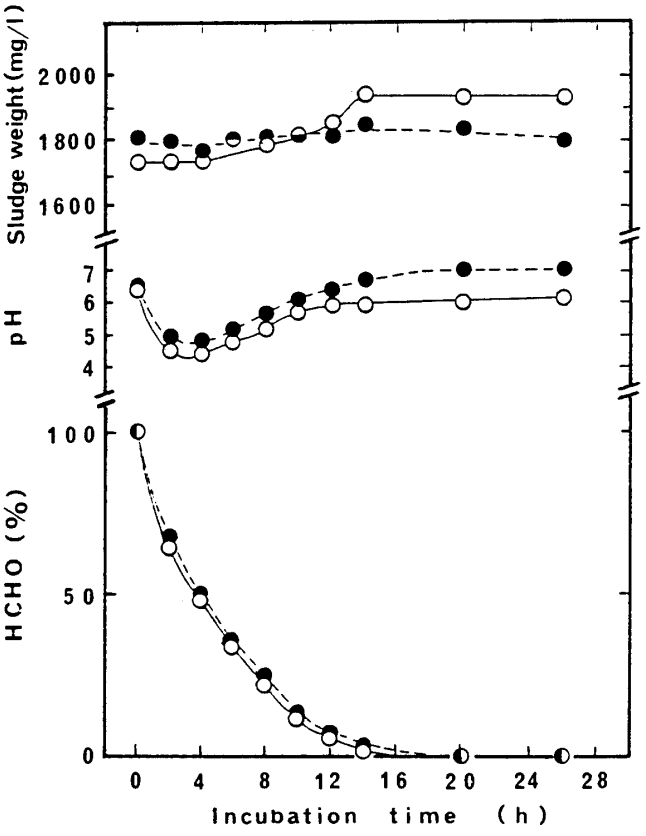

Fig.4. Time course of HCHO removal by activated sludge and changes in pH and sludge weight. 300 mg/l of HCHO(●) or 300 mg/l of HCHO and 300 mg/l of glucose(○) containing media were incubated by stirring at 20°C with activated sludge acclimated for 300 mg/l of HCHO.

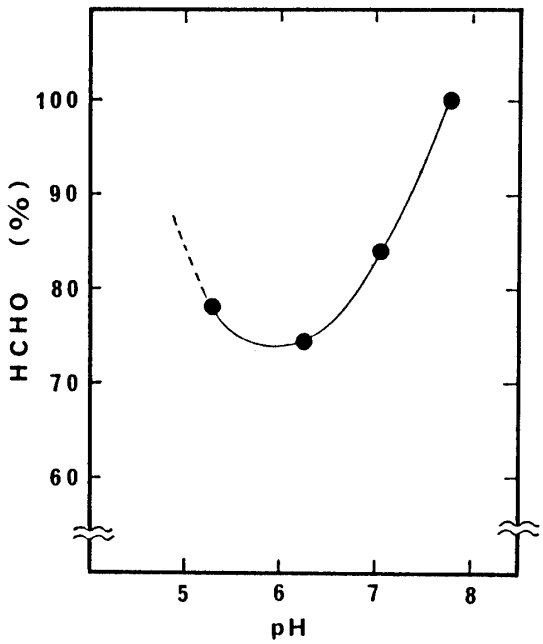

Fig.5. Effect of pH on HCHO removal by activated sludge. 400 mg/l of HCHO containing media in 0.1M phosphate buffer adjusted to various pH values were incubated by stirring at 20°C for 24 hours with 350 mg/l of activated sludge acclimated for 1,000 mg/l of HCHO.

6. No change in the pH of the medium was observed during the operation.

DISCUSSION

Various disinfectants often cause damage to biological waste water treatment plant when they flow into it. However, microorganisms in the plant can achieve decomposition and assimilation ability for certain concentrations of disinfectants by acclimation. Utilizing this property, many studies on the biological treatment of these wastes have been carried out (7-9). The present study concentrated on the biological treatment of HCHO by activated sludge.

Inoue (10) classified various chemical substances into three groups of high, middle, and low rate degradation by calculation of the ratio BOD / TOD (Theoretical Oxygen Demand), and reported that HCHO belonged to the middle group at both low and high concentration, and toxicity concentration for acetic acid degrading bacteria was 2 mg/l.

The O_2 uptake measurement is generally used for acute

inhibition tests of a toxic substance (11). More than 6.7 mg/l HCHO showed lag type toxicity for O_2 uptake of unacclimated activated sludge, but less than 3.3 mg/l HCHO did not have any effect on it. Ciliated protozoa, good indicators for the maintenance of activated sludge (12,13), were not affected by incubation with less than 10 mg/l HCHO for 24 hours. These results suggest that the inflow of less than 3 mg/l HCHO does not cause damage to treatment plant.

Acclimation studies for highly concentrated HCHO showed that where glucose is present, activated sludge could remove 90% of 5,500 mg/l HCHO at 40 hours.

As shown in Fig.3, 50% HCHO removal caused a decrease in pH, and further removal caused an increase in pH. It is suggested that the decrease in pH was caused by oxidation of HCHO to formic acid, while the subsequent increase was caused by oxidation of the formic acid. Oxidation of glucose in glucose-containing medium was presumed by calculation of COD difference between glucose-containing and non-glucose media, as shown in Fig.3. It started when HCHO was almost removed and pH was raised to 5.5. In glucose-containing medium, sludge weight increased with glucose oxidation, but in non-glucose medium, no increase was observed. Those results suggest that HCHO was oxidized but not utilized as a carbon source by the activated sludge.

The optimal pH of 1,000 mg/l HCHO acclimated sludge was about 6. However, during removal of 5,500 mg/l HCHO, the pH in the medium was lower than 4 and the minimum pH was 2.7, but HCHO was removed at a considerably high rate. Therefore, it is presumed that the optimal pH of highly concentrated HCHO-acclimated sludge is lower than 6.

Some methylotrophs, which are defined as microorganisms utilizing reduced C_1-compounds as the sole source of carbon and energy, have been reported to assimilate HCHO (14,15). On the other hand, HCHO-oxidizing microorganisms which are not able to utilize HCHO as a carbon source have been isolated from soil and sewage mud (16,17). The present acclimated activated sludge had similar properties to the latter.

Sayama (8) reported that although unacclimated activated sludge could not decompose 1,000 mg/l HCHO, activated sludge acclimated by increasing HCHO step by step could decompose it within 24 hours. In the present study, acclimated sludge could decompose 5,500 mg/l HCHO. Therefore, acclimation by increasing

HCHO step by step and adding glucose as a carbon source are important for the treatment of highly concentrated HCHO.

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