Wat. Sci. Tech. Vol. 23, Kyoto, pp. 1125–1136, 1991.
Printed in Great Britain.

0273–1223/91 $0·00 + ·50

UTILIZATION OF MICROORGANISMS IMMOBILIZED WITH MAGNETIC PARTICLES FOR SEWAGE AND WASTEWATER TREATMENT

H. Ozaki*, Z. Liu** and Y. Terashima*

**Department of Environmental and Sanitary Engineering, Kyoto University, Kyoto 606, Japan*
***Henan Provincial Science Research Institute of Environmental Protection, Zhengzhou, China*

ABSTRACT

A novel method for sewage and wastewater treatment using microorganisms immobilized with magnetic particles was developed with a proposal of the treatment process. This process has the characteristic of having microorganisms in effluent recovered magnetically. Batch and continuous experiments for phenol removal were carried out using microorganisms immobilized with ferromagnetic particles after cultivation in the presence of phenol. In the continuous experiment, almost 100% of phenol in feed water was removed for at least 40 days, during which a developed apparatus, mainly consisting of a reactor and a separator with magnet, was operated without difficulty. In the mixed system of the immobilized microorganisms and activated sludge, phenol and easily biodegradable substance were removed simultaneously and efficiently. Thus the addition of the immobilized microorganisms to a reactor with activated sludge was considered to be able to endow the reactor system with further capability for treatment. Furthermore, it became apparent that a mixture of three kinds of microorganisms, immobilized with ferromagnetic particles and feeble magnetic particles respectively and without magnetic particles, could remove the objective substrate, and that the microorganisms immobilized with magnetic particles could be recovered using a magnet and a High Gradient Magnetic Filtration (HGMF) equipment.

These results show that a new process, in which the microorganisms in a reactor would become controllable and be maintained in high concentration, could be developed.

KEYWORDS

Wastewater treatment; immobilized microorganisms; polyaclylamide; recovery of microorgamisms; magnetic particles; magnetic separation; high gradient magnetic filtration; labeling technique

INTRODUCTION

Several types of sewage and wastewater treatment processes utilizing high concentrated microorganisms have been developed to remove pollutants more efficiently. The treatment process using immobilized microorganisms is one of the most interesting subjects in the field. The most common immobilization method for treatment use is to entrap microorganisms into a gel lattice prepared from one or two polymers, such as carrageenan, calcium alginate, polyacrylamide, polyvinyl alcohol and so on. Main advantages of the treatment, compared with conventional activated sludge process, are as follows;

1) high concentrated microorganisms are maintained in a reactor.
2) immobilized microorganisms can be protected from others and are not appreciably inhibited by toxic substances.
3) microorganisms which do not form flocks can work in a treatment process by use of immobilization method.

4) commonly used gel resulting from immobilization, more than 2 to 3mm in diameter, can be easily separated.

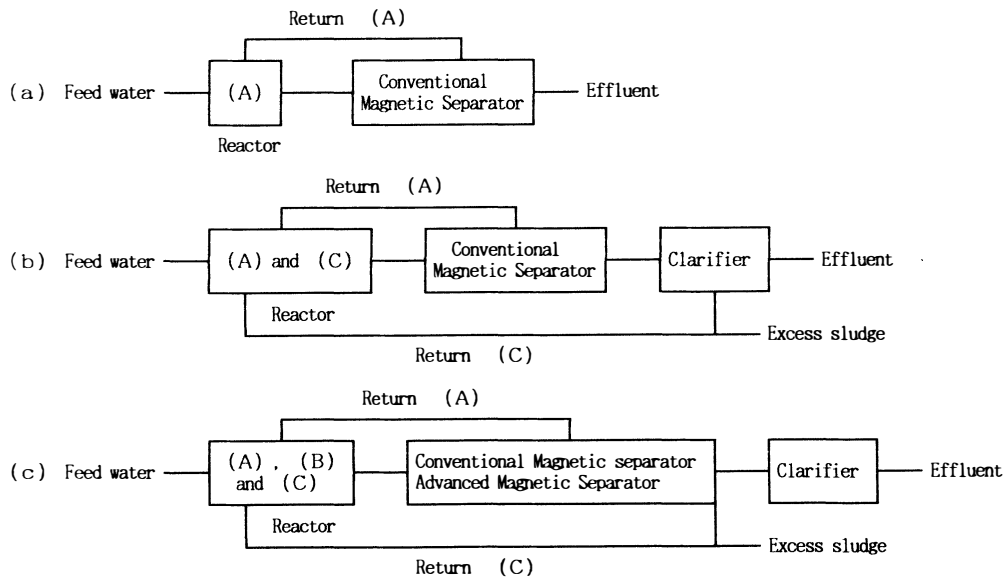
The process, however, has the following disadvantages;

1) the substrate removal efficiency is limited by the transport of reactants in a gel as support medium.
2) the gels of more than 2 to 3mm, mentioned in above item 4), cannot be suspended easily.
3) the activity of microorganisms may be become lower by immobilization.
4) particulate BOD cannot be removed.

Although small particles of gel less than 1mm in diameter are available for efficient treatment, relating to above item 1) of disadvantages, they can not be easily recovered.

The authors (1989) have developed a new treatment process using microorganisms immobilized with magnetic particles. In the process, the gels resulting from immobilization are made less than 0.25mm in diameter each, so that they can be suspended by aeration in a reactor. The microorganisms used, for example, autotrophs such as nitrifier or heterotrophs which are capable of decomposing recalcitrant substances. Those microorganisms immobilized with magnetic particles in the effluent from a reactor can be recovered magnetically, even if the gel particles including the microorganisms are small and they are mixed with activated sludge.

Figure 1 (a), (b) and (c) are examples of process flowsheet utilizing the immobilized microorganisms, under investigation by the authors. In process (a), the microorganisms immobilized with ferromagnetic particles (magnetite, Fe_3O_4) are suspended in a reactor, and those in effluent from the reactor are recovered by a conventional magnetic separator using permanent magnets, for returning to the reactor. In the mixture system of the immobilized microorganisms and activated sludge, in which recalcitrant substance and easily biodegradable substance as examples are removed simultaneously, a setting tank for activated sludge is installed in the next step of magnetic separator, as shown in Fig.1 (b). Figure 1 (c) is the process flowsheet in the case of adding another kind of microorganisms immobilized with feeble magnetic particles (hematite (α-Fe_2O_3) etc) into a reactor. The gels including feeble magnetic particles can be recovered by an advanced magnetic separator such as High Gradient Magnetic



(A) : Microorganisms immobilized with ferromagnetic particles
(B) : Microorganisms immobilized with feeble magnetic particles
(C) : Microorganisms immobilized without addition of magnetic particles or activated sludge

Fig. 1 Examples of process flowsheet utilizing microorganisms immobilized with magnetic particles

Filtration (HGMF), since they possess much lower magneticism than those including ferromagnetic particles (Owen et al., 1983). In the above process, at least three kinds of microorganisms can be separated and recovered individually after mixing, though the mixed microorganisms cannot be separated again in a conventional biological treatment process. From this meaning, it can be said that the method immobilizing microorganisms with magnetic particles is a kind of magnetical labeling technique of microorganisms, since they are distinguished from each other by magnetic characteristics.

The development of such an idea of water treatment will promote the establishment of the process in which the concentration of several kinds of microorganisms are controllable in a reactor. This paper presents the experiments and results on 1) treatablity of pollutants by using microorganisms immobilized with magnetic particles, 2) continuous treatment using the above process, and 3) recovery of the immobilized microorganisms.

MATERIALS AND METHODS

Preparation of Gel Including Microorganisms and Magnetic Particles

Acrylamide monomer was used as a main chemical for the immobilization, since polyaclylamide gel is more resistant to feeding by a pump or agitation by a stirrer than other kinds of gels. Polyaclylamide gel was mainly prepared in the manner reported by Chibata et al. (1974) and coauthors (Tosa et al., 1974), as shown in Fig.2, which is an example using five grams of wet biomass collected by a centrifuge. In the procedure ferromagnetic particles (magnetite: Fe_3O_3), paramagnetic particles (manganese pyrophosphate: $Mn_2P_2O_7$) or weak ferromagnetic particles (hematite: α-Fe_2O_3) were further added with chemicals of acrylamide monomer and so on. Suceptibilities of manganese pyrophosphate and hematite were 1.95×10^{-3} and 2.19×10^{-4} emu/cm^3/Oe, respectively.

The resulting gel was blended with a mixer on the market and sieved in the size of 74~250μm. The physical characteristics of the gel including magnetite (ferromagnetic particles) are listed in Table 1. The saturation magnetization of gel was estimated from that of magnetite

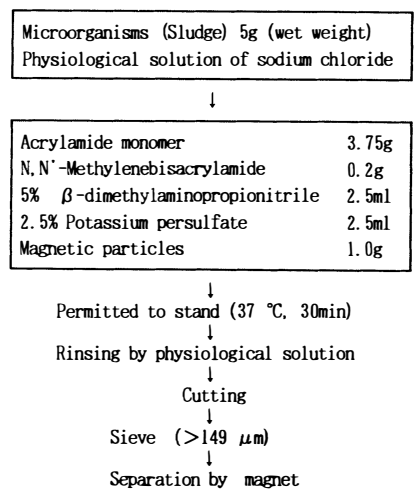


Fig. 2 Preparation method of gel including microorganisms and magnetic particles

TABLE 1 Physical characteristics of gel including microorganisms and magnetite

Apparent specific gravity	1.17 (g/cm^3)
Water content	83.8 (%)
Magnetite content	35.3 (mg/cm^3)
Estimated magnetization (at 1.0T)	15.9 (emu/dry-g)

(85.3emu/g) and the volume of gel. As mentioned in the section above, the gel including ferromagnetic particles can be easily separated by conventional magnetic separator. On the other hand, the gel including feeble magnetic particles can be separated only by advanced magnetic separator such as HGMF.

Degradation of Phenol by Microorganisms Immobilized with Ferromagnetic Particles

Batch experiments. A series of batch experiments (Runs 1 to 5), shown in Fig.3, were conducted, to examine 1) the treatability of wastewater (phenol) only by microorganisms immobilized with ferromagnetic particles (Runs 1, 3 and 5), 2) the treatability in the mixture system of the immobilized microorganisms and activated sludge (Run 2), and 3) the recovery and reutilization of immobilized microorganisms from the mixture system (Runs 3 and 4).

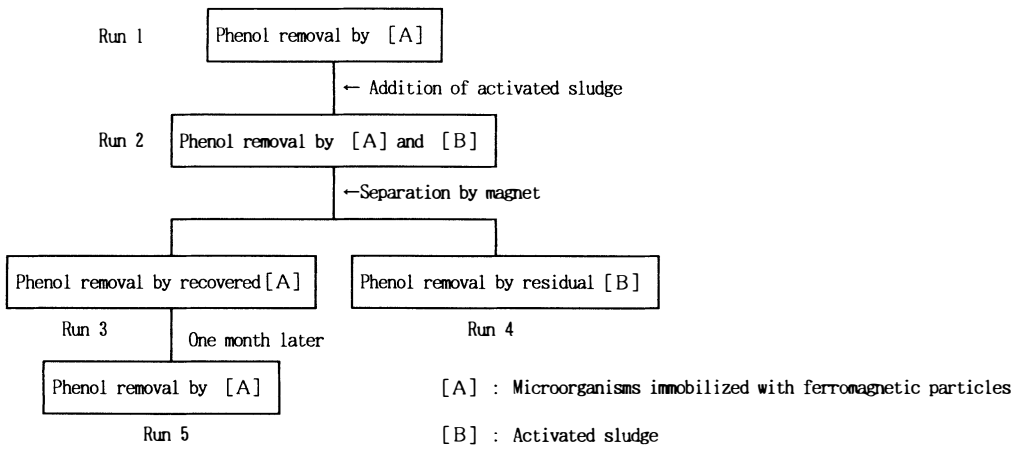


Fig. 3 Batch experiments (Runs 1 to 5)

After one month of Runs 1 to 4, Run 5 was carried out to examine the stabilization of immobilized microorganisms. The microorganisms used were cultivated in the presence of phenol for about one month before immobilization. On the other hand, the activated sludge of Run 2 was cultivated in the presence of only glucose. In each Run, simulated wastewater containing phenols and nutrients, which was selected as a representative wastewater, was in a moment added into a reactor (a 1 liter beaker) in which the immobilized microorganisms and/or activated sludge was suspended by aeration. The each initial concentration of phenol was about 80mg/ℓ. The gel particles including microorganisms and magnetite were recovered by using a horseshoe magnet which had a maximum magnet field of 1.5kG (Run 2). The degradation of phenol in the reactor with the elapse of time was monitored by a gas chromatograph equipped with flame ionization detector. In each experiment, the dissolved oxygen (DO) concentration was over 4.0mg/ℓ, pH was adjusted in the range of 7.0 to 7.4, and the concentration of microorganisms was estimated to be approximately 700mg/ℓ from the data of their phosphorous content.

Continuous experiments The continuous experiment of phenol treatment was carried out over a long term, using microorganisms immobilized with magnetite. The purpose of this experiment was to investigate the characteristics of substrate removal by the process described below, and recovery of immobilized microorganisms by a magnetic separator. Figure 4 shows the schematic diagram of the experimental set up, which consists of mainly a reactor (volume of 4 liter) and a magnetic separator (effective volume of 0.7ℓ). The separator had a disk of 200mm in diameter and 12mm in thickness, which was made of plastic and rotated at the speed of 2rpm. Eight bar magnets, each measuring 30×25×8mm, were sealed in the disk. A simulated wastewater containing phenols of about 60mg/ℓ and nutrients was fed under the condition of HRT of 6 hours. The mixed liquor including immobilized microorganisms was pumped up by a tube pump from the reactor to the magnetic separator. After the immobilized microorganisms which adhered on the magnets of a disk were scraped by a scraper on the wall of the disk, they were returned to the reactor by a wash out of influent fed on the scraper.

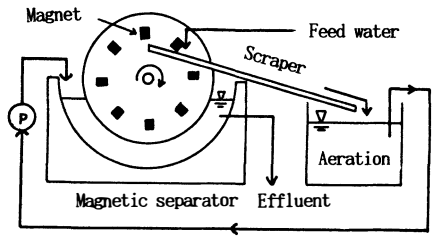


Fig. 4 Schematic diagram of set up for continuous treatment

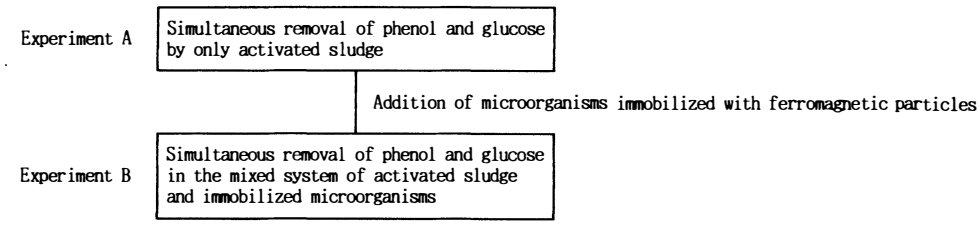


Fig. 5 Experimental procedure adding immobilized microorganisms

Furthermore, other continuous experiments (experiments A and B) were conducted using the immobilized microorganisms and activated sludge, in accordance with the procedure shown in Fig.5. A clarifier for activated sludge was set in the next step of magnetic separator illustrated in Fig.4. In experiment A, glucose and phenol were treated by only activated sludge which had been cultivated in the presence of glucose only. After 2 days, the microorganisms immobilized with ferromagnetic particles, which had been cultivated in the presence of phenol, were further added into the reactor (experiment B), under the same operating condition of experiment A. The concentrations of glucose and phenol in the feed water were about 100mg/ℓ and 300mg/ℓ, respectively. The concentration of glucose was measured by the anthrone method and that of CODcr was determined by an autoanalyzer (Technicon Autoanalyzer Ⅱ). The purpose of these experiments was to confirm the effectiveness of immobilized microorganisms' addition into a reactor with activate sludge. The recovery of gel particles including microorganisms and magnetite from a reactor, which had activated sludge and gel particles, was also examined.

Wastewater Treatment by Mixture of Immobilized Microorganisms

The experimental works were carried out on 1) the substrates removal by a mixture of microorganisms immobilized with ferromagnetic particles, microorganisms immobilized with feeble magnetic particles and microorganisms immobilized without magnetic particles, and 2) magnetic recovery of microorganisms immobilized with magnetic particles.

Substrate removal in batch experiment Table 2 indicates four kinds of gels resulting from immobilization, that is, gel including microorganisms and ferromagnetic particles (gels (3) and (6)), gel including microorganisms and weak ferromagnetic particles (gel (2)), gel including microorganisms and paramagnetic particles (gel (5)),and gel including activated sludge only (gel (1)). Here, gels (1) and (4) were the same, as well as gels (3) and (6). Those gels were classified in 2 groups; group A of gels (1), (2), and (3), and group B of gels (4), (5), and (6).

TABLE 2 A mixture of gels including microorganisms and magnetite

	Gel No	Immobilized Microorganism	Immobilized Magnetic Particle
Group A	(1)	Activated sludge	No addition
	(2)	Bacteria cultured in the presence of phenol	α-Fe_2O_3
	(3)	Bacteria cultured in the presence of triethylene glycol	Fe_3O_4
Group B	(4)	Activated sludge	No addition
	(5)	Bacteria cultured in the presence of phenol	$Mn_2P_2O_7$
	(6)	Bacteria cultured in the presence of triethylene glycol	Fe_3O_4

In the presence of mixed immobilized gels of each group, a batch experiment of each group was carried out in a reactor (1 liter beaker) under aerated condition, to examine the treatability of simulated wastewater including glucose, triethylene glycol and phenols. The concentration of microorganisms in each gel in a reactor was 100 to 200mg/ℓ.

Recovery of gels including microorganisms and feeble magnetic particles by HGMF The microorganisms immobilized with feeble magnetic particles can be separated only by an advanced magnetic separation technique, High Gradient Magnetic Filtration (HGMF). In HGMF, the strong magnetic force (Fm) expressed by eq.(1) acts on magnetic substances when they move through a magnetic field (H);

$$Fm=V \cdot M(H) \cdot dH/dx \qquad (1)$$

where V is the volume of the magnetic substance, and M(H) is the magnetization of the magnetic substance in a magnetic field (H). Thus magnetic substances are captured on a filter wire by the magnetic tractive force (Fm) that overcomes other competing forces of hydrodynamics and inertia etc (Oberteuffer, 1974, Terashima et al. 1986).

A schematic diagram of HGMF equipment, used in this study, is illustrated in Fig.6. The filter was 12.5mm in effective diameter, and was packed randomly with ferromagnetic stainless wire of 0.3mm in diameter. The suspension of the gel particles including microorganisms and hematite was passed, with up flow, through the filter placed in a high magnetic field (H) of 1.0T to investigate the effects of operating conditions on the recovery efficiency of gel particles by HGMF. The gel particles were 74 to 250μm in diameter. The flow rate, filter length and packing fraction of wire were varied in the range of 24.5 to 73.8m/h, 40 to 150mm and 2.6 to 4.8%, respectively. The recovery percents (R) of gel particles including Fe were evaluated by the following equation, in which the effects of mechanical trap of gel particles by filter were removed. The concentration of gel particles was estimated by measuring that of Fe included in gels after dissolving them.

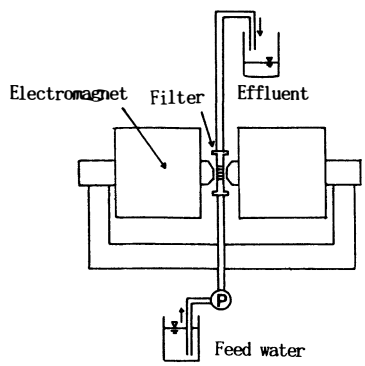
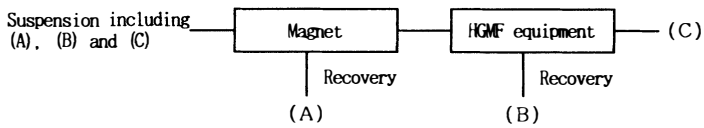


Fig. 6 Schematic diagram of HGMF equipment

$$R(\%)=\frac{\text{concentration of gel particles (Fe) in effluent under the magnetic field of 1.0T}}{\text{concentration of gel particles (Fe) in effluent under no magnetic field}}\times 100 \quad (2)$$

Recovery of Specified Kinds of Immobilized Microorganisms Separation and recovery of specified kinds of gels, that is, specified kinds of microorganisms, from the mixed gels of each group shown in Table 2, were examined in the procedure of Fig. 7. By this method of recovery, the gel (3) is expected to be recovered at first by a magnet, and secondly the gel (2) or (4) by HGMF equipment described above. The gels without the addition of magnetic particles cannot be separated magnetically. The recovery of gel particles including magnetite was conducted by using a horse shoe magnet in a 1 liter beaker. The HGMF equipment was operated under the following condition: induced magnetic field of 1.0T, flow rate of 24.5m/hr, filter length of 150mm and wire packing fraction of 2.6%. The recovery efficiency of gel particles including magnetite, and hematite or manganese pyrophosphate was obtained in the same manner as was described above by measuring the concentration of Fe or Mn included in the gels after dissolving them.



(A) : Gels including ferromagnetic particles and microorganisms
(B) : Gels including feeble magnetic particles and microorganisms
(C) : Gels without addition of magnetic particles

Fig. 7 Experimental procedure for recovery of gels resulting from immobilization

RESULTS AND DISCUSSION

Degradation of Phenol by Microorganisms Immobilized with Ferromagnetic Particles

Figure 8 shows the batch treatment results of Runs 1 to 4 indicated in Fig. 3. The immobilized gels of Runs 1 to 3 include ferromagnetic particles. The batch experiment of Run 2 was conducted in the mixture system of the immobilized gel of Run 1 and activated sludge which had been cultivated in the presence of glucose. The gel particles of Run 3 were recovered from the mixture system of Run 2, at the efficiency of more than 95.0% by a magnet. The phenol in each reactor of Runs 1 to 3 decreased obeying zero-order kinetics, as can be often seen in the

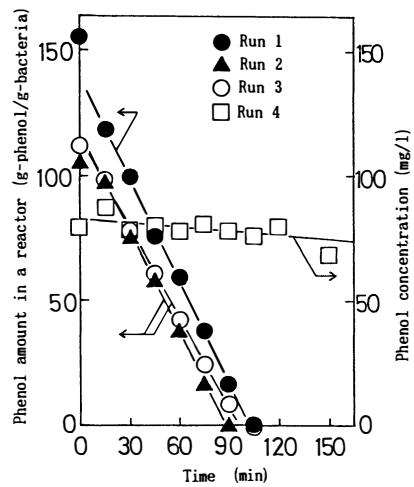


Fig. 8 Batch treatment results of phenol (Runs 1 to 4)

TABLE 3 Removal rate constant of phenol

Run 1	1.36×10^{-3}	(min^{-1})
Run 2	1.33×10^{-3}	(min^{-1})
Run 3	1.20×10^{-3}	(min^{-1})

treatment using activated sludge cultivated sufficiently in the presence of phenol. The results of Run 5, which was conducted after two months of Run 3, was similar to those of Runs 1 to 3. On the other hand, the activated sludge could remove little phenol in Run 4. Table 3 lists the removal rate constants of Runs 1, 2, 3, and 5. Those of Runs 1, 2, and 3 were almost the same. The larger value of Run 5 was probably caused by the progress of microorganisms cultivation. From the above results the following were found: 1) Phenol could be removed by the immobilized microorganisms and similarly by the mixture of those and activated sludge (Run 1 and 2), 2) The gel including ferromagnetic particles could be easily recovered by a magnet (Run 2 and 3), 3) The recovered microorganisms could be reutilized (Run 3), 4) The immobilized microorganisms leaked little from the gel to a reactor (Run 4), and 5) the immobilized microorganisms could maintain the activity removing phenol for at least 2 months (Run 5). Thus, it has become apparent that the microorganisms immobilized with ferromagnetic particles could be used in an activated sludge system, without undergoing an inhibition by coexistence with activated sludge.

Figure 9 shows the results of continuous experiments on phenol treatment using the microorganisms immobilized with ferromagnetic particles. The phenol was not detectable in the

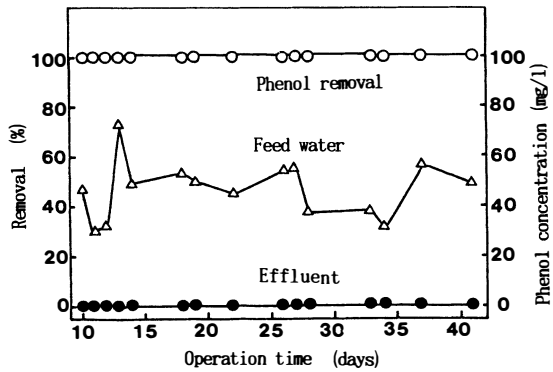


Fig. 9 Phenol removal in continuous experiment

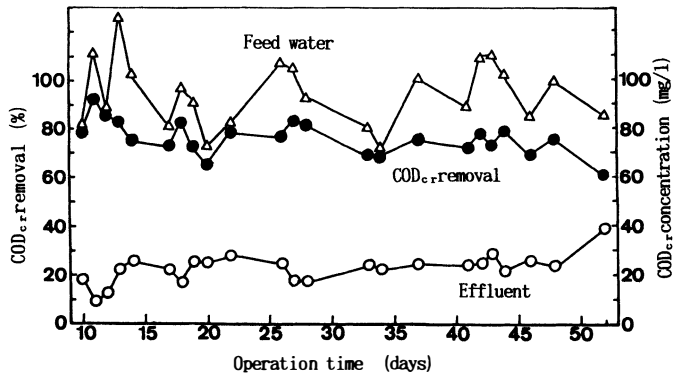


Fig. 10 CODcr removal in continuous experiment

effluent for about 40 days, and the experimental set up, illustrated in Fig. 4, worked without difficulty. However, the removal percents of CODcr were 70 to 85, as shown in Fig. 10. The CODcr in the effluent was considered to have resulted from the metabolism of microorganisms and hydrolysis of gel. Thus, a future subject would be a development of preparation method for gel which is resistant to hydrolysis and wear caused by agitation and pumping.

The result of another continuous treatment, which was carried out in the procedure of Fig. 5, are shown in Fig. 11. The immobilized microorganisms were added at the time indicated by the dotted line in Fig. 11 (Experiment B). Almost all the glucose in feed water could be removed during the experimental period, as a matter of course, since activated sludge was in the reactor in both experiments A and B. As can be seen from the result of experiment A, the activated sludge uncultivated in the presence of phenol can normally, to some extent, remove phenol from the initial stage. After adding the immobilized microorganisms, the removal efficiency of phenol increased more than in experiment A by about 40%. This result suggests that the addition of immobilized microorganisms is able to improve a treatment capability of an activated sludge system. Furthermore, it should be noted that the immobilized microorganisms can be recovered magnetically from the effluent since they possess magnetization.

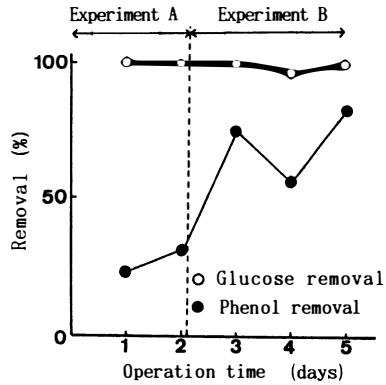


Fig. 11 Effect of immobilized microorganisms addition to activated sludge reactor on phenol removal

Wastewater Treatment by Mixture of Immobilized Microorganisms

Substrate removal in batch experiment The batch treatment results of group A in Table 2 are shown in Fig. 12. Group A consisted of three kinds of gels including microorganisms cultivated in the presence of glucose, tryethylen glycol and phenol, respectively. The concentrations of three substrates decreased with an elapse of time, although the substrate removal rates were different from each other because of the difference of microorganisms concentration included in each gel. The results of group B were similar to those of group A. Those results show that immobilized microorganisms in each gel worked sufficiently for the removal of objective substrate in such a mixture system of immobilized gels. The treatment method proposed in this study, which aims at the recovery of specified kinds of microorganisms from a mixed microorganisms system after substrate removal, is expected to stimulute the development of a new treatment process in which the concentrations of several kinds of microorganisms are controllable. For realizing the above process, it is necessary to establish a technique separating immobilized microorganisms individually which possess different magnetization.

Recovery of gels including microorganisms and feeble magnetic particles by HGMF In general, recovery efficiencies of magnetic substances by HGMF are affected mainly by operating conditions such as an induced magnetic field, a flow rate, a packing fraction, and a filter length. All the recovery experiments in this study were conducted under the maximum magnetic field (1.0T) which could be induced by the HGMF equipment used, since the magnetic tractive force acting on feeble magnetic substances is directly proportional to the magnetic field on the basis of the equation (1). An example of the recovery of gel particles by HGMF is shown in Fig. 13, with experimental conditions. The gel particles included hematite as a feeble magnetic substance. The ordinate of the figure indicates the recovery of Fe included in gel, that is, that of gel including hematite. When breakthrough of filter is reached, the gel captured on wires is removed by backwashing of filter under no magnetic field.

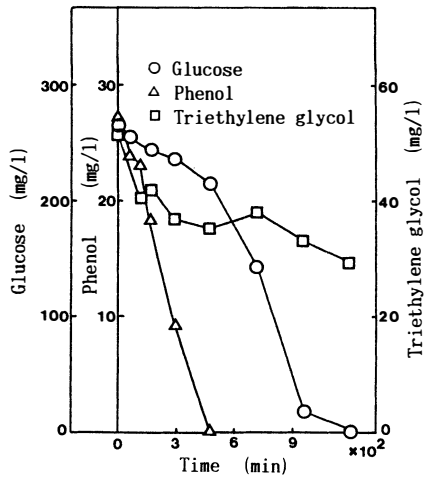


Fig. 12 Substrate removal in batch experiment

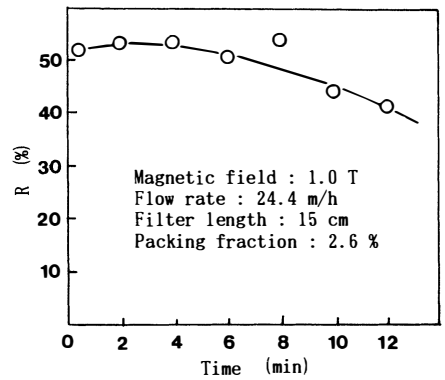


Fig. 13 An example of recovery of gel particles including microorganisms and hematite by HGMF

Figure 14 shows the relationship between the recovery efficiency and flow rates. As had been expected, the removal efficiency decreased with an increase of flow rate. A too low flow rate might cause more mechanical and gravitational capture of magnetic gels on a filter, although a comparatively high recovery efficiency was obtained under low flow rate. Further, a high flow rate would disturb the attachment of magnetic gels on a filter, as can be seen in Figure 14. It is easy to obtain better removal efficiency in the range of flow rate, as described below. The packing fraction little affected the recovery efficiency of magnetic gels in the examined range of 2.6 to 4.8%. The low packing fraction is, however, considered to be desirable for preventing a mechanical capture of magnetic gels. As shown in Fig. 15, the recovery efficiency increased with an increase of filter length. To obtain better removal efficiency, repeated passages of magnetic gels to a filter are considered to be effective from the result.

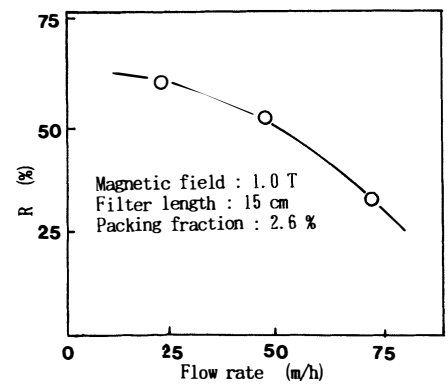


Fig. 14 Effects of flow rate on recovery efficiency of gel particles

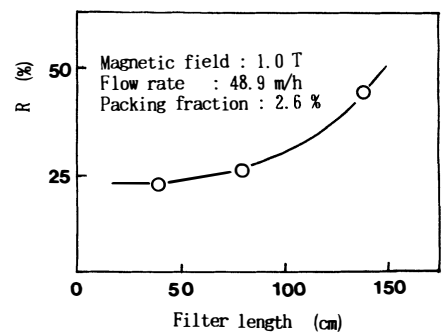


Fig. 15 Effects of filter length on recovery efficiency of gel particles

Recovery of specified kinds of immobilized microorganisms Figures 16 (a) and (b) show the results regarding magnetic separation of gels of groups A and B, listed in Table 2. Over 95.0% of gel particles including magnetite (gel (6)) was recovered by a magnet (Fig.16 (b)). The recovery efficiency of gel (3) was also applied to that of gel (6), from the fact that the value of gel(3) could not be obtained directly since gels (2) and (3) in group A included hematite and magnetite of ferrous compound, respectively. The recovery efficiencies of gel particles including feeble magnetic particles were 64.6% (gel (2), Fig. 16 (a)) and 33.7% (gel(5), Fig. 16 (b)). These values, though not so high, are expected to be enhanced under the

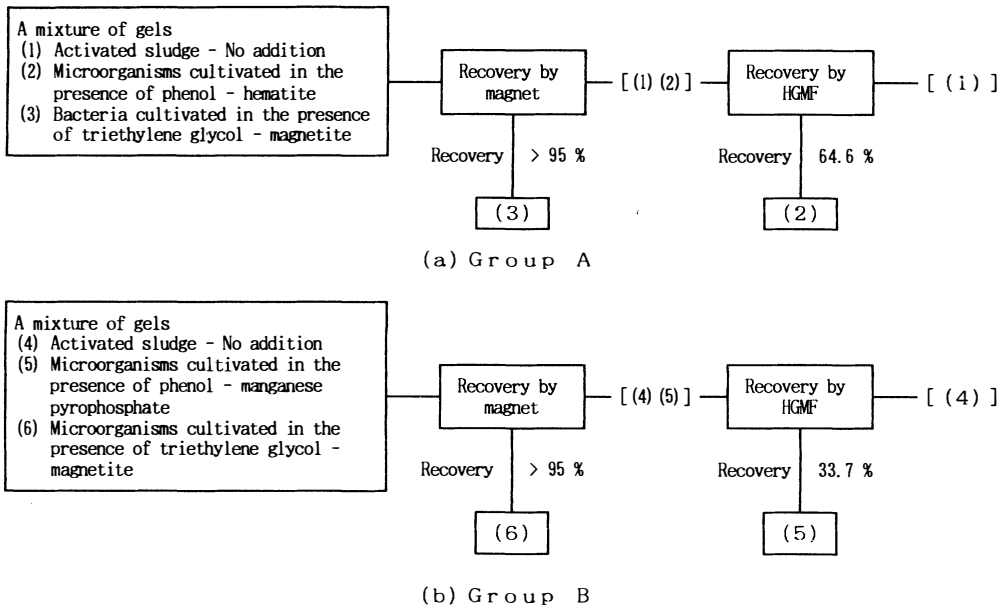


Fig. 16 Recovery of gels resulting from immobilization

better operating conditions such as higher magnetic field and repeated passage of magnetic gels to a filter. The microorganisms recovered in the procedure of Fig. 16 would be returned to a reactor directly or after storing.

CONCLUSION

(1) In batch experiment using the microorganisms immobilized with ferromagnetic particles after cultivation in the presence of phenol, phenol was removed in accordance with zero order reaction in the system of the immobilized microorganisms alone and of a mixture of those and activated sludge. The immobilized microorganisms, which maintained the activity removing phenol for at least 2 months, could be recovered easily from the reactor with activated sludge, by using a magnet at the efficiency of nearly 100%.

(2) In continuous experiment using above immobilized microorganisms, almost 100% of phenol in feed water was constantly removed for 40 days, during which a developed apparatus, mainly consisting of a reactor and a magnetic separator, was operated without difficulty. Over 95% of the immobilized microorganisms was recovered from effluent by the separator.

(3) The addition of above immobilized microorganisms into a reactor with activated sludge improved the treatability of phenol. Such a usage of immobilized microorganisms was considered to be able to endow the activated sludge system with further capability for treatment.

(4) A mixture of three kinds of immobilized microorganisms, which consists of microorganisms immobilized with ferromagnetic particles and feeble magnetic particles respectively, and microorganisms immobilized without an addition of magnetic particles, could remove the objective substrate, without inhibiting each other.

(5) From the mixture described above, over 95% of gels including ferromagnetic particles was recovered by a magnet. The gels including feeble magnetic particles were recovered using HGMF equipment, at the recovery efficiency of 33.7 or 61.6%. These values, though not so high, are expected to be enhanced under better operating conditions.

(6) The results described above show that a new process, in which the microorganisms in a reactor would become controllable and be maintained in high concentration, could be developed.

ACKNOWLEDGEMENT

We would like to thank Mr. S.Imabayashi and Mr. H.Koshikawa for their help with the experiments.

REFERENCES

Chibata, I., Tosa, T. and Sato, T. (1974). Immobilized aspartase-containing microbial cells: Preparation and enzymatic properties. Applied Microbiology, 27, 878-885.

Owen C.S. and Lindsay J.G. (1983). Ferritin as a label for high-gradient magnetic separation. Biophysical Society, 42, 145-150.

Oberteuffer, J.A. (1974). Magnetic Separation: A view of principles, devices, and application. IEEE Transactions on Magnetics, MAG-10, 223-238.

Ozaki, H., Liu, I. and Terashima, Y. (1989). Wastewater treatment using bacteria immobilized with magnetic particles. Proc. of the 10th Symposium of AESER, 369-374.

Terashima, Y., Ozaki, H. and Sekine, M. (1986). Removal of dissolved heavy metals by chemical coagulation, magnetic seeding and high gradient magnetic filtration. Water Reseach, 357-545

Tosa, T., Sato, T., Mori, T. and Chibata, I. (1974). Basic studies for continuous production of L-aspartic acid by immobilized Escherichia coli cells. Applied Microbiology, 27,