

Removal of Bacteria from Water by Adhesion to Cross-Linked Poly(Vinylpyridinium Halide)

NARIYOSHI KAWABATA,^{1*} TAKAYA HAYASHI,¹ AND TSUGUO MATSUMOTO²

Laboratory of Environmental Chemistry, Department of Chemistry, Faculty of Polytechnic Science, Kyoto Institute of Technology, Matsugasaki, Sakyo-ku, Kyoto 606, Japan,¹ and Laboratory of Insect Pathology and Microbiology, Kyoto University of Textile Science, Matsugasaki, Sakyo-ku, Kyoto 606, Japan²

Received 6 December 1982/Accepted 29 April 1983

Cross-linked poly(vinylpyridinium halide) was found to have a novel and remarkable ability to remove bacteria from water. For example, when 10 g (wet weight) of cross-linked poly(*N*-benzyl-4-vinylpyridinium bromide) was contacted with 20 ml of suspensions of *Escherichia coli* (9.7×10^4 to 9.7×10^7 /ml), *Salmonella typhimurium* (8.0×10^6 to 1.1×10^7 /ml), *Streptococcus faecalis* (5.0×10^7 /ml), *Staphylococcus aureus* (8.1×10^7 /ml), and *Pseudomonas aeruginosa* (3.2×10^5 /ml) under stirring in sterilized physiological saline at 37°C, 99% of the viable cells of these bacteria were removed in 2 to 6 h. When suspensions of these bacteria (10^5 to 10^8 cells per ml) were passed through a column (20 mm by 100 cm) of cross-linked poly(*N*-benzyl-4-vinylpyridinium bromide) at 37°C with a flow rate of 0.8 to 1.4 bed volumes per h, 97 to 100% of the viable cells were eliminated from the suspensions during the treatment. Mechanistic studies demonstrated that cross-linked poly(vinylpyridinium halide) irreversibly captured these bacteria alive during the treatment. That is, total organic carbon was removed during the treatment, and the bacteria which adhered to the resin proliferated on the bacterial medium. The adhesion capacity was estimated to be 10^{10} cells per g (dry weight). Total organic carbon was also removed even when the bacteria were killed by heat treatment before the column studies.

Water disinfection processes include the removal and destruction of microorganisms by both physical and chemical means. The most popular process is the treatment with chlorine and other related chemicals, but the formation of trihalomethanes and other carcinogens is a serious defect in this procedure. During the course of study to develop an alternative method of disinfection with insoluble polymeric materials, cross-linked poly(vinylpyridinium halide) was found to have a novel and remarkable ability to remove bacteria from water. Mechanistic studies revealed that cross-linked poly(vinylpyridinium halide) irreversibly captured bacteria alive during the treatment.

Ion-exchange resins have been used to isolate microorganisms by direct treatment of cell suspensions with the resins (1, 2, 4, 9). In this case, the adsorption of microorganisms onto the resins was reversible, and microorganisms were isolated by desorption from the resins. On the other hand, cross-linked poly(vinylpyridinium halide) irreversibly captured bacteria. Insoluble synthetic materials of antimicrobial activity have been used as insoluble disinfectants (3, 5, 8). The polyiodide form of a strong base anion-exchange resin was extremely effective for this

purpose but exhibited very little tendency for adsorption of bacteria (8). However, cross-linked poly(vinylpyridinium halide) captured bacteria alive during the treatment. Thus, the methodology described in this paper appears to be a new concept of water treatment and of wide application in microbiology.

MATERIALS AND METHODS

Materials. Cross-linked poly(vinylpyridinium halide) was prepared by the reaction of the appropriate organic halide with vinylpyridine-divinylbenzene copolymer containing 72 mol% vinylpyridine by the procedure described previously (7). Cross-linked poly(vinylpyridinium halide) was disinfected with 70% alcohol followed by extensive washing with sterilized physiological saline to remove the alcohol from the resin.

Escherichia coli, *Salmonella typhimurium*, *Streptococcus faecalis*, *Staphylococcus aureus*, and *Pseudomonas aeruginosa* are used as important bacteria in public health. These bacterial species were incubated for 20 h at 37°C in nutrient broth. The bacterial cells were harvested by centrifugation and washed repeatedly with sterilized physiological saline.

Procedure. All procedures were carried out under aseptic conditions. Batch studies were performed with a glass tube (28 by 100 mm) connected with a stopper made of silicon rubber, a tube for sampling, and a

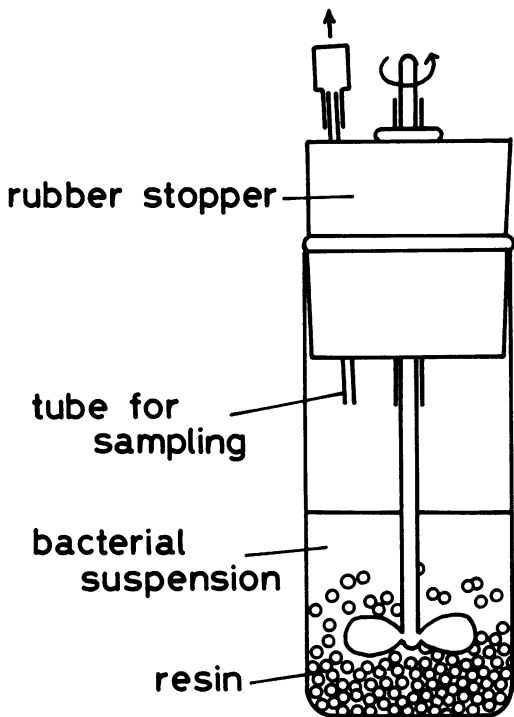


FIG. 1. Apparatus for batch studies of removal of bacteria from water by insoluble polymeric materials.

mechanical stirrer made by remodeling a hypodermic syringe (Fig. 1). In the glass tube was placed 20 ml of each suspension and 10 g (wet weight) of insoluble polymeric material, and the mixture was stirred at 120 rpm at 37°C. After a prescribed time, samples of the harvested cell suspensions were transferred to agar plates for the measurement of viable cell counts.

Column studies were carried out in two ways. A series of experiments were aimed at examining the change of viable cell counts during the contact with resins and were performed with a glass column (20 mm by 100 cm) at 37°C with a flow rate of 0.18 to 1.4 bed volumes per h (see Table 3). Another series of column studies was aimed at examining the change of the amount of total organic carbon (TOC) during the contact with resin and was performed with a glass column (10 mm by 30 cm) with a flow rate of 1.1 to 2.1 bed volumes per h (see Table 4 and Fig. 6 and 7). The relation of TOC to the viable cell counts for *E. coli* is shown in Fig. 2. The amounts of TOC of influent and effluent suspensions were determined by using the Sumitomo model GCT-12N TOC and total nitrogen apparatus.

RESULTS

Batch studies of removal of bacteria from water. Preliminary batch experiments were designed to establish the method to evaluate the effect of cross-linked poly(vinylpyridinium halide) for the removal of bacteria from water with cross-linked poly(*N*-benzyl-4-vinylpyridinium

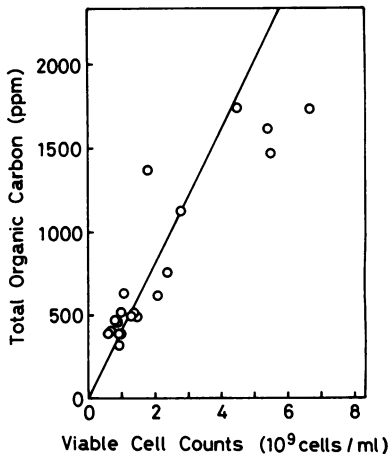


FIG. 2. Relation of TOC to viable cell counts of *E. coli* suspensions in sterilized physiological saline.

bromide) [BzVP(Br)] and *E. coli* (Fig. 3). Curve A of Fig. 3 shows that the viable cell counts obviously decreased in the presence of BzVP(Br). About 99% removal of *E. coli* was achieved in 2 h. On the contrary, curve B of Fig. 3 shows that substantial change in the viable cell counts was not observed even after 8 h in the absence of the resin.

Removal of *E. coli* by BzVP(Br) was carried out with a variety of initial viable cell counts (Fig. 4). Although the initial viable cell counts varied from 9.7×10^4 to 9.7×10^7 cells per ml, about 99% removal of viable *E. coli* was achieved in 2 h.

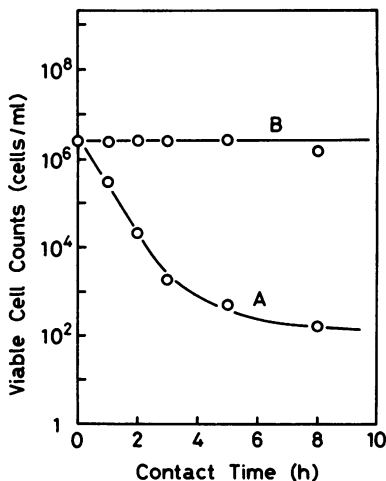


FIG. 3. Removal of viable *E. coli* from water by cross-linked poly(*N*-benzyl-4-vinylpyridinium bromide) in the presence (A) or absence (B) of resin.

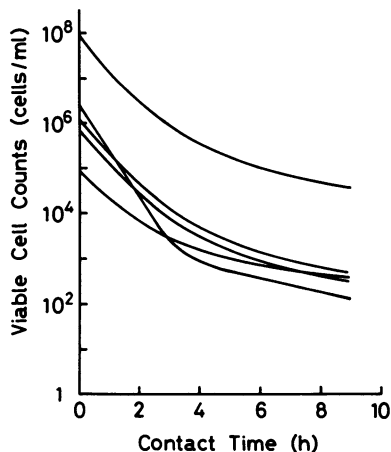


FIG. 4. Removal of viable *E. coli* from water by cross-linked poly(*N*-benzyl-4-vinylpyridinium bromide) carried out with a variety of initial viable cell counts.

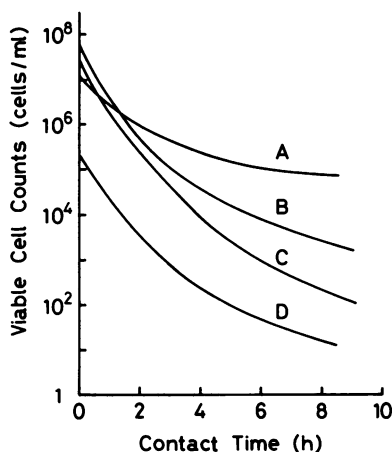


FIG. 5. Removal of viable cells of various bacteria from water by cross-linked poly(*N*-benzyl-4-vinylpyridinium bromide): *S. typhimurium* (A); *S. aureus* (B); *S. faecalis* (C); and *P. aeruginosa* (D).

The removal of other bacteria from water by BzVP(Br) was carried out in the same way (Fig. 5). About 99% removal of the viable cells of *S. faecalis*, *S. aureus* and *P. aeruginosa* was also achieved in 2 h. However, the removal of *S. typhimurium* was rather difficult, and 99% removal of the viable cells was achieved after 6 h.

Removal coefficient. To evaluate quantitatively the ability of insoluble polymeric materials for the removal of viable bacteria from water, we attempted to find a useful index of the ability. As can be seen in Fig. 3, 4, and 5, the relation of the logarithm of viable cell counts to contact time was linear in the early stage of contact. This result indicated that the removal process was a first-order rate process, similar to the case of quaternary ammonium triiodide ion-exchange sterilization process (6). Thus, we defined the removal coefficient based on the initial rate of the decrease of viable cell counts:

$$\text{Removal coefficient} = \frac{V}{Wt} \log [N(0)/N(t)]$$

Here, $N(0)$ is the initial viable cell count, $N(t)$ is the viable cell count at contact time t , V is the volume of viable cell suspension, W is the dry weight of insoluble polymeric materials, and t is the contact time. Table 1 shows the values of the removal coefficients obtained for the removal of various bacteria by BzVP(Br). Although the initial viable cell counts varied from 9.7×10^4 to 9.7×10^7 cells per ml, and the amount of resin was changed from 5 to 20 g (wet weight) (2.0 to 7.6 g (dry weight)), the coefficient for the removal of *E. coli* by BzVP(Br) ranged from 3.4 to 4.8 ml/g h. This result shows the utility of the removal coefficient.

Effect of the structure of cross-linked poly(vinylpyridinium halide). To examine the effect of the structure of cross-linked poly(vinylpyridinium halide) on the removal coefficient, various types of cross-linked poly(vinylpyridinium halide) were used to remove *E. coli* from water (Table 2).

The removal coefficient of BzVP(Br) proportionally depended on the content of the pyridinium group (Table 2). This result suggests that the ability of cross-linked poly(vinylpyridinium ha-

TABLE 1. Removal coefficient for the removal of various bacteria from water by cross-linked poly(*N*-benzyl-4-vinylpyridinium bromide)^a

Bacteria	$N(0)$ (cells per ml)	W (g dry wt)	Removal coefficient (ml/g h)
<i>E. coli</i>	9.7×10^4	3.8	4.4
	8.4×10^5	2.0 ^b	4.8
	8.4×10^5	4.0	4.2
	9.7×10^5	7.6 ^c	4.4
	1.7×10^6	3.8	3.4
	4.3×10^6	4.0	4.8
<i>S. typhimurium</i>	9.7×10^7	3.8	4.2
	8.0×10^6	3.7	2.1
<i>P. aeruginosa</i>	1.1×10^7	3.8	2.1
	3.2×10^5	3.3	4.2
<i>S. aureus</i>	8.1×10^7	3.9	4.4
<i>S. faecalis</i>	5.0×10^7	3.8	4.8

^a Carried out at 37°C. The content of the pyridinium group of the resin was 2.9 mmol/g (dry weight), and V , volume of viable cell suspension, was 20 ml; W , insoluble polymeric materials; $N(0)$, initial viable cell count. The amount of resin was 10 g (wet weight) unless otherwise stated.

^b The amount of resin was 5 g (wet weight).

^c The amount of resin was 20 g (wet weight).

TABLE 2. Removal coefficient for the removal of viable *E. coli* from water by various types of cross-linked poly(vinylpyridinium halide)^a

Cross-linked resin	Pyridinium group (mmol/g [dry wt])	N(0) (cells per ml)	W (g [dry wt])	Removal coefficient (ml/g h)
Poly(<i>N</i> -benzyl-4-vinylpyridinium bromide)	2.9			4.3 ^b
	1.5	3.7×10^7	3.3	2.0
	1.5	3.7×10^7	2.9	2.2
Poly(<i>N</i> -benzyl-4-vinylpyridinium chloride)	2.9	7.6×10^6	3.7	2.7
	2.9	3.7×10^7	3.6	4.2
	3.3	3.7×10^7	4.0	2.4
Poly(<i>N</i> -allyl-4-vinylpyridinium bromide)	3.3	3.7×10^7	4.0	2.4
Poly(<i>N</i> -propargyl-4-vinylpyridinium bromide)	2.6	3.7×10^7	5.6	2.0
	2.6	3.7×10^7	5.8	1.9
Poly(<i>N</i> -pentafluorophenylmethyl-4-vinylpyridinium bromide)	1.9	1.0×10^6	4.2	1.7
Poly(<i>N</i> -butyl-4-vinylpyridinium bromide)	2.8	2.9×10^6	3.6	2.2
Poly(<i>N</i> -octyl-4-vinylpyridinium bromide)	2.1	8.4×10^5	4.3	1.8
Poly(<i>N</i> -dodecyl-4-vinylpyridinium bromide)	1.4	1.0×10^6	4.3	1.3
Poly(<i>N</i> -hexadecyl-4-vinylpyridinium bromide)	0.9	6.5×10^7	5.8	1.3
Amberlite IRA-900 in the chloride form ^c	4.2 ^d	2.9×10^6	4.4	0.4
Amberlite XAD-4 ^e		4.7×10^6	5.2	0.3
Poly(4-vinylpyridine)		3.7×10^7	3.5	1.5

^a Carried out at 37°C; V, volume of viable cell suspension, was 20 ml; W, insoluble polymeric materials; N(0), initial viable cell count. The amount of resin was 10 g (wet weight).

^b The average value obtained from Table 1.

^c Commercial strong base anion-exchange resin.

^d The content of ammonium group.

^e Commercial macroporous resin with no ion-exchange functional group.

lide) to remove bacteria was derived from the presence of the pyridinium group.

Although the difference in the content of the pyridinium group should be taken into consideration, BzVP(Br) and BzVP(Cl) were most effective among the resins examined. Cross-linked poly(*N*-alkyl-4-vinylpyridinium bromides) were less effective. The hydrophobic group did not strengthen the adsorptive behavior of the resin. The surface charge of the resin was expected to raise the adsorptive behavior. However, cross-linked poly(*N*-pentafluorophenylmethyl-4-vinylpyridinium bromide) was not very effective. The adsorptive behavior was not simply correlated with the ionic strength of the pyridinium group. The adsorptive behavior of the resin appeared to depend not only on the chemical structure of the functional group, but also on the hydrophilic nature, the degree of cross linking, and other physical properties of the resin matrix. Further investigation is required to clarify the mechanism of the sorption.

For comparative purposes, Amberlite IRA-900 in the chloride form provided by Rohm and Haas Co., Philadelphia, Pa., was used as a strong base anion-exchange resin. The resin was not very effective for the removal of *E. coli*. A reviewer advised us to use Ambergard resin, a product made by the same manufacturer and claimed to remove all kinds of microorganisms by adsorption without killing, instead of using

IRA-900. Unfortunately, however, the resin was not available in Kyoto. Ambergard resin might be more effective than IRA-900, although the chemical structure of the functional group would be similar.

Column studies of removal of bacteria from water. On the basis of the batch studies mentioned above, attempts were made to remove various bacteria by passage through a glass column packed with BzVP(Br) (Table 3). Except for the case with *S. typhimurium*, more than 99.999% of the viable bacteria were removed by passing through the column. The efficiency of removal decreased with the increase of flow rate, which may be due to the decrease of contact time.

Change of TOC amount during column studies. Attempts were made to elucidate whether cross-linked poly(vinylpyridinium halide) functioned as an insoluble polymeric disinfectant against bacteria but did not remove the dead cells or acted as a polymeric adsorbent or adhesive agent for the removal of bacteria. The change of TOC amount during the procedure was investigated. Although there are definitive ways of looking at the attachment of bacteria to surfaces, direct observation of the attachment to insoluble resins is difficult to investigate in contrast to the colonization of bacteria on the epithelium of organs with infections lesions. Column studies were carried out with suspensions of *E. coli* (10^8

TABLE 3. Removal of viable bacteria from water by a fixed-bed column packed with cross-linked poly(*N*-benzyl-4-vinylpyridinium bromide)^a

Bacteria	Flow rate (bed vol per h)	Total effluent (bed vol)	Viable cell counts (cells per ml)		Removal (%)
			Influent	Effluent ^b	
<i>E. coli</i>	0.8	4.8	1.0×10^7	0	100
	0.8	4.7	1.9×10^8	1.7×10^3	99.9991
	1.0	6.2	2.3×10^6	0	100
	1.0	5.8	2.5×10^6	0	100
	1.4	8.2	2.3×10^6	1.4×10^2	99.9994
<i>S. aureus</i>	0.8	4.6	1.0×10^7	0	100
	1.0	6.0	1.0×10^7	3.0×10^1	99.9997
	1.2	7.4	1.4×10^7	5.5×10^2	99.996
<i>S. typhimurium</i>	1.0	6.0	3.3×10^6	9.7×10^4	97
<i>P. aeruginosa</i>	1.0	6.3	6.3×10^5	0	100
<i>S. faecalis</i>	1.0	6.1	2.3×10^5	0	100

^a Carried out at 37°C for 5 to 6 h. The content of the pyridinium group of the resin was 2.9 mmol/g (dry weight).

^b The average value of the viable cell counts of effluent samples.

cells per ml) and a fixed bed containing BzVP(Br) with a flow rate of 1.1 to 1.2 bed volumes per h. The TOC amount of the influent suspensions was 40 to 50 ppm (40 to 50 µg/ml), but was quantitatively removed during the column studies. TOC was linearly correlated with cell counts (Fig. 2). The quantitative removal of TOC was thus concluded to be evidence of the quantitative removal of bacterial cells during treatment. Cross-linked poly(vinylpyridinium halide) was thus proved to have removed bacterial cells during treatment.

Viability of bacteria after contact with cross-linked poly(vinylpyridinium halide). Another series of experiments was carried out to elucidate whether cross-linked poly(vinylpyridinium halide) maintained bacteria alive or killed them on removal. Suspensions of viable *E. coli* were passed through a glass column containing BzVP(Br), and the resin bed was extensively washed with sterilized physiological saline. The resin bed was then taken out from the column and inoculated into nutrient broth. The washing solution was also cultured, but multiplication of *E. coli* was not observed in the solution. However, adsorbed or adhered *E. coli* on BzVP(Br) proliferate on bacterial media. Thus, we concluded that cross-linked poly(vinylpyridinium halide) captured these bacteria alive during the treatment, although the proportion of live bacteria was obscure.

Irreversible nature of removal of bacteria from water by cross-linked poly(vinylpyridinium halide). After suspensions of *E. coli* were passed through a glass column containing BzVP(Br), the resin bed was washed with sterilized physiological saline. This washing solution was cultured, but multiplication of *E. coli* was not observed.

We made many efforts to regenerate exhaust-

ed BzVP(Br) for the removal of *E. coli*: (i) extensive washing with absolute ethanol followed by extensive washing with sterilized physiological saline; and (ii) washing with aqueous hydrochloric acid (pH 3) followed by extensive washing with sterilized physiological saline. Despite these efforts, exhausted BzVP(Br) did not remove *E. coli* again. Attempts to regenerate BzVP(Br) thus ended in failure.

These observations strongly suggested that the removal of bacteria from water by cross-linked poly(vinylpyridinium halide) was irreversible, and made us consider that the resin acted as a polymeric adhesive agent rather than as a polymeric adsorbent for the bacterial cells.

Adhesion capacity. To evaluate quantitatively the ability of cross-linked poly(vinylpyridinium halide) to remove bacteria, column studies were also performed with bacterial suspensions of much higher concentrations. Based on the linear relationship shown in Fig. 2, the quantity of bacteria was evaluated by the quantity of TOC. Figure 6 shows the plot of TOC of the effluent suspension toward the effluent volume in the case of the removal of *E. coli* from water by BzVP(Br). In this case, the percentage of the removal of TOC was not very high, and significant leakage was observed during the treatment. This incomplete removal of TOC was mainly due to the too high concentration of bacterial cells in the influent suspension. When bacterial suspension of lower concentrations were used as the influent suspensions, the percentage of the removal was much improved. Under such conditions, however, quantitative evaluation of the removal ability required much larger amounts of the influent suspensions and much longer experimental time, and it would be very difficult to keep viable cells alive throughout the column studies.

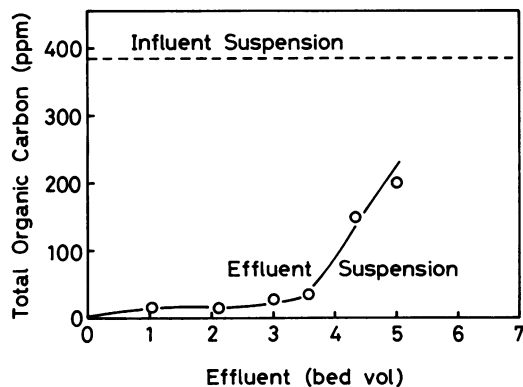


FIG. 6. Removal of *E. coli* from water by a fixed bed of cross-linked poly(*N*-benzyl-4-vinylpyridinium bromide). The flow rate was 1.1 bed volumes per h, and the viable cell counts of the influent suspension were 1.2×10^9 cells per ml.

The adhesion capacity was then calculated by an approximation, by using the following equation:

$$\text{Adhesion capacity} = C(0) \times [V(H) - V(0)]/W$$

where $C(0)$ is the influent concentration of bacteria, $V(0)$ is the void column volume, $V(H)$ is the effluent volume when the effluent concentration reached half of the influent concentration, and W is the dry weight of the resin (Table 4). Here, the adhesion capacity was determined based on the amount of adhered TOC. In the case of *E. coli*, however, the amount of adhered cells was estimated based on the relation of TOC to viable cell counts (Fig. 2 and Table 4). The adhesion capacity of BzVP(Br) for these bacteria appeared to be 10^{10} cells per g (dry weight).

Removal of killed bacteria from water by cross-linked poly(vinylpyridinium halide). Attempts were also made to clarify whether the adhesion to cross-linked poly(vinylpyridinium halide) was limited to viable bacterial cells. The column studies were performed with *E. coli* suspensions which were heated at 70°C for 50 min before the studies. The breakthrough curve is illustrated in Fig. 7, and the adhesion capacity is given in Table 4. This experimental observation clearly indicated that the adhesion to cross-linked poly(vinylpyridinium halide) is not limited to viable bacterial cells.

DISCUSSION

Experimental results presented in this article demonstrated that cross-linked poly(vinylpyridinium halide) has a novel and remarkable ability to remove bacteria from water. This methodology appears to be a new concept of water purification.

TABLE 4. Capacity of cross-linked poly(*N*-benzyl-4-vinylpyridinium bromide) for the adhesion of bacteria in water^a

Bacteria	Flow rate (bed vol per h)	Influent TOC (ppm)	Adhesion capacity	
			mg TOC per g (dry wt)	10^{10} cells per g (dry wt) ^b
<i>E. coli</i>	1.1	370	6.0	1.5
	1.5	410	5.6	1.4
	2.1	300	5.1	1.3
	1.7	230	4.7	1.2
	1.6	230 ^c	4.7	1.2
<i>S. typhimurium</i>	1.5	790	1.6	
	1.2	450	1.3	
<i>S. faecalis</i>	1.5	290	4.0	

^a Determined by the continuous-flow column method. The content of the pyridinium group of the resin was 2.9 mmol/g (dry weight).

^b Amount estimated based on the relation of TOC to viable cell counts which is shown in Fig. 2.

^c In this case, the suspension of *E. coli* was heated at 70°C for 50 min immediately before being passed through the column, and the absence of viable cells was ascertained under these conditions.

Mechanistic studies revealed that the resin acted as a polymeric adhesive agent for bacteria. The quantitative removal of TOC was observed during contact of the bacterial suspension with resin. Several experimental observations strongly suggested the irreversible nature of the adhesion.

Mechanistic studies also revealed that the resin captured bacteria alive during the treatment. The bacteria which adhered to the resin proliferated on the bacterial medium. Bacteria

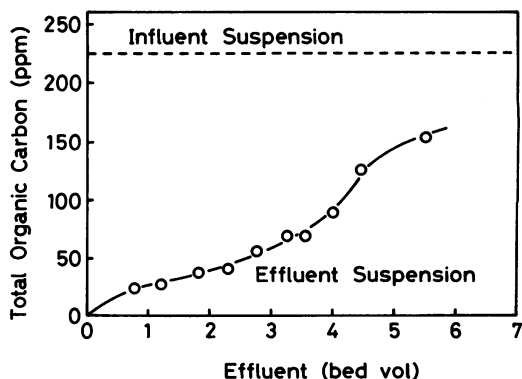


FIG. 7. Removal of heat-killed *E. coli* from water by a fixed bed of cross-linked poly(*N*-benzyl-4-vinylpyridinium bromide). The flow rate was 1.6 bed volumes per h, and the viable cell count of the influent suspension before the heat treatment at 70°C for 50 min was 5.8×10^8 cells per ml.

killed by heat treatment were also removed by the resin in a manner similar to the case of viable cells.

The ability of resin to remove bacteria from water was evaluated in two ways. The rate of bacterial removal was evaluated by the removal coefficient defined in this article. The coefficient was demonstrated to be a useful index of the removal ability. The amount of bacterial removal was evaluated by the adhesion capacity. The capacity was approximately evaluated based on the amount of removed TOC during a continuous flow column study. The adhesion capacity was estimated to be 10^{10} cells per g (dry weight) based on the relation of TOC to the viable cell counts.

The difference in the adhesion capacities for viable and killed bacteria was insignificant, and physicochemical interaction appeared to be much more important than physiological action in this adhesion.

The ability of resin, evaluated by the removal coefficient, proportionally depended on the content of the pyridinium group. The ability of resin to remove bacteria from water was thus concluded to derive from the presence of the pyridinium group. The removal coefficient of the resin depended on the chemical structure of the pyridinium group, and BzVP(Br) was most effective among the resins examined. The adhesion on the resin also depended on the nature of the bacteria. Experimental results indicated that both the rate and capacity of the bacteria removal were low for *S. typhimurium* when compared with those for the other bacteria used in this work. The mechanism of adhesion requires further investigation.

Although chlorine is a strong disinfectant against microorganisms, it reacts with a variety of organic impurities in water and converts them into trihalomethanes and other carcinogens; effective nonchlorine disinfectants have received increasing attention. From this point of view, the methodology described in this article provided a possibility for new alternative concepts of water purification. Use of the resin has some advantages. The resin usually does not react with organic impurities in water, and it does not add any chemical into water. The resin removed not only viable cells but also killed cells of bacteria from water, and the quality of the treated water was better than those of usual disinfection. The removal of whole cells was more suitable than usual disinfection in cases in which bacterial pathogens resist disinfectants, and the formation of trihalomethane and other carcinogens is not desirable. However, the new methodology has some weak points. The removal was much too slow, as can be seen in Fig. 3, 4, and 5. Therefore, in the column studies, a slow

rate of flow was necessary for complete removal of bacteria from water. It was extremely difficult to regenerate the exhausted resin, and we had to give up repeated use of the resin.

Experimental results presented in this article also demonstrated that cross-linked poly(vinylpyridinium halide) captured bacteria alive during the treatment. The adhesion appears to be irreversible. This nature of the resin appears to be of wide application in microbiology and biotechnology. We are now attempting to immobilize bacteria and activated sludge by this resin to apply the new methodology to the production of useful materials and wastewater treatment. Although various methods are known for the immobilization of enzymes and bacterial cells, continuous efforts are being made to pursue new materials and techniques for the immobilization (10). Immobilization to a solid matrix by chemical reaction is apt to result in damage to the enzyme activity. Entrapment in the gel of polymeric substances is apt to result in a decrease of the contact of substrates with enzymes and bacterial cells. Adsorption onto ion-exchange resin is reversible. However, immobilization on BzVP(Br) can be realized without chemical reaction and entrapment, and the adhesion is irreversible. Surface-immobilized, live bacteria entities on this resin are expected to show useful properties. We are also expecting the resin to be powerful material for the search of various microorganisms present in the natural world. It is noteworthy that BzVP(Br) has an excellent ability for removing sodium alkylbenzenesulfonates from aqueous solutions (7). The adsorption capacity reached 900 mg/g (dry weight), and was not affected by the presence of inorganic salts, alkali, and acid.

LITERATURE CITED

1. Daniels, S. L. 1980. Mechanisms involved in sorption of microorganisms to solid surfaces, p. 7-58. In G. Britton and K. C. Marshall (ed.), Adsorption of microorganisms to surfaces. John Wiley & Sons, Inc., New York.
2. Daniels, S. L., and L. L. Kempe. 1966. The separation of bacteria by adsorption onto ion exchange resins. Chem. Eng. Prog. Symp. Ser. 62(69):124-148.
3. Folsome, C. E., S. A. Leslie, and R. K. Thoms. 1955. Control of chronic lower genital tract infections. The use of anion exchange resin. Obstet. Gynecol. 6:531-537.
4. Freeman, R. R. 1964. Separation of cells from fluids. Biotechnol. Bioeng. 6:87-125.
5. Isquith, A. J., E. A. Abbott, and P. A. Walters. 1972. Surface-bonded antimicrobial activity of an organosilicon quaternary ammonium chloride. Appl. Microbiol. 24:859-863.
6. Kao, I. C., D. E. Robker, L. T. Fan, A. Snyder, and L. E. Erickson. 1972. Analysis and properties of a quaternary ammonium triiodide ion exchange sterilization process. J. Ferment. Technol. 50:438-445.
7. Kawabata, N., and T. Morigaki. 1980. Removal and recovery of dodecylbenzenesulfonate from aqueous solution by crosslinked poly(N-benzyl-4-vinylpyridinium halide). Environ. Sci. Technol. 14:1089-1093.
8. Lambert, J. L., G. T. Fina, and L. R. Fina. 1980. Prepara-

- tion and properties of triiodide-, pentaiodide-, and hepta-iodide-quarternary ammonium strong base anion-exchange resin disinfectants. *Ind. Eng. Chem. Prod. Res. Dev.* **19**:256-258.
9. **Rotman, B.** 1960. Uses of ion exchange resins in microbiology. *Bacteriol. Rev.* **24**:251-260.
10. **Venkatasubramanian, K., and W. R. Vieth.** 1979. Immobilized microbial cells. *Prog. Ind. Microbiol.* **15**:61-86.