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ENTERIC VIRUS REMOVAL FROM WATER BY COAL-BASED SORBENTS: DEVELOPMENT OF LOW-COST WATER FILTERS

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ABSTRACT

Using poliovirus type 1 (Sabin) and dechlorinated tap water, several coal-based sorbents were tested for their capacity to remove viruses from water. The sorbents included bituminous coal from Giridih, India, pretreated/impregnated with either alum, ferric hydroxide, lime or manganese dioxide. Filtrasorb-400, commercially available active carbon, was used as a reference. In batch tests, with input virus concentration of $2.34\text{-}2.83\times10^6$ PFU l^{-1} and sorbent concentration of 10 g l^{-1}, alum pretreated coal removed about 96% of the virus when pH of the water was between 6.3 and 8.9. Virus sorption was rapid and a plateau was reached in 30 min. Compared with the active carbon, alum pretreated coal exhibited greater sorption energy and about one log higher limiting poliovirus sorption capacity. Downflow column study indicated the potential of alum pretreated coal as a filter media for removing enteric viruses from water. A previous study showed this sorbent to be capable of removing enteric bacteria as well. Water filters prepared from such low-cost material may prove useful for domestic use in rural areas of India and other developing countries.

KEY WORDS

Drinking water; enteric viruses; coal-based sorbents; developing countries; low-cost water filters

INTRODUCTION

Presence of enteric viruses in drinking water constitutes an unacceptable health risk and hence has attracted serious attention of environmental health scientists and engineers. There is increasing evidence of enteric virus contamination of drinking water sources (Bitton and co-workers, 1985). Enteric viruses of human origin have, in fact, been isolated from potable waters of a number of communities, including certain urban centres in India (Rao and co-workers, 1981). It may be stated that the technology to produce virologically safe water is available; however, the present dilemma is that, primarily for financial reasons, such technology is not within the reach of the great majority of the world's population living in the rural areas of the developing countries (IAWPRC Study Group on Water Virology, 1983). The task that lies ahead is the development of low-cost reliable technology. Research on development and performance testing of low-cost indigenous materials that could be ultimately incorporated into domestic water filters to produce safe drinking water would be a logical and significant step forward in this direction.

Studies at the Indian Institute of Technology, Kanpur, indicated the potential of bituminous coal in removing model bacterial viruses from water (Oza and Chaudhuri, 1976, Sriramulu and Chaudhuri, 1976) and iron from ground water (George and Chaudhuri, 1977), hydrous iron/manganese-oxides impregnated bituminous coal in removing organics (humic acid) from water (Jayasimha, 1983) and raw as well as chemically pretreated/impregnated bituminous coal in removing heavy metals (cadmium and mercury) from water (Bhattacharya and Venkobachar, 1984; Karthikeyan and Chaudhuri, 1986; Kirankumar, 1984; Pandy and Chaudhuri, 1982). A recent study showed enhanced removal of Escherichia coli from water by chemically impregnated bituminous coal sorbents (Prasad, 1986).

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The results of a study to assess enteric virus (poliovirus) removal (sorption) potential of some coal-based sorbents (bituminous coal pretreated/impregnated with either alum, ferric hydroxide, lime or manganese dioxide), using a commercially available active carbon as a reference, are reported here.

EXPERIMENTAL

Giridih bituminous coal (GBC), obtained through the National Environmental Engineering Research Institute, Nagpur, India, and Filtrasorb-400 (F-400) active carbon (Calgon Corp., Pittsburg, PA, USA), were crushed to a geometric mean size of 0.388 mm, washed several times in distilled water and dried overnight at 103°C before use in sorption experiments. The proximate and ultimate analyses of the coal were: moisture (at 60% R.H., 40°C) 4.6%, ash 18.8%, volatile 31.6%, fixed carbon 45.0%, and moisture 4.6%, ash 20.68%, carbon 61.2%, hydrogen 3.38%, sulphur 0.33%, nitrogen 1.31%, oxygen (by difference) 8.08%. For alum pretreatment, the method of Anderson and co-workers (1980) for alum loading of cation exchange resins was used. Crushed GBC (20 g) and 200 ml of 1 M alum ($Al_2(SO_4)_3.16\ H_2O$) solution at pH 3.2 were agitated in an end-over-end shaker @ 20 rpm for 7 d. For ferric hydroxide impregnation, the method of Anderson and co-workers (1982) for coating magnetite with ferric hydroxide gel was employed. Crushed GBC (20 g) was slurried in 100 ml distilled water to which 0.5 g of ferric chloride ($FeCl_3$) was added. Using 2 N sodium hydroxide, the pH of the slurry was then adjusted at a rate of 6 pH units h^{-1} to a final value of 11.5 and the final volume was made up to 200 ml. The slurry was allowed to settle for 15 min. Lime impregnation of GBC was carried out using a method reported by Cullen and Sivious (1982). Crushed GBC (20 g) in 100 ml lime slurry (100 g l^{-1}) was agitated in an end-over-end shaker @ 20 rpm for 2 h. For manganese dioxide (δMnO_2) impregnation, the method adopted by Kirankumar (1984) was used. Crushed GBC (24 g) was added to a solution of potassium permanganate (25.675 g in 325 ml distilled water) at 90°C in a water bath. The slurry was stirred for 10 min followed by slow addition of 300 ml of 2 M hydrochloric acid in 10 min with constant stirring. Stirring was continued for another 10 min followed by washing off excess precipitate with distilled water and 0.05 M perchloric acid till the supernatant became clear. The sorbents following pretreatment/impregnation were removed from solution, washed several times with distilled water, dried at 103°C and sieved to a geometric mean size of 0.388 mm.

Poliovirus type 1 (Sabin) pools were prepared as x10 concentrates from MA-104 cell cultures and were quantified by plaque assay in MA-104 cell cultures. Techniques for culture, maintenance and passage of MA-104 cells were described previously (Sattar and co-workers, 1984). For preparation of virus pools, cell monolayers in 490 cm^2 roller bottles were inoculated with poliovirus at a multiplicity of infection of one virus per 100 cells and maintained at 37°C in Eagle's minimal essential medium (MEM) with 1% fetal calf serum (FCS). Complete cytopathic degeneration of the monolayers occurred within 72 h. The cultures were frozen (-80°C) and thawed three times to aid in virus release from the infected cells. The virus suspension was clarified of large cellular debris by centrifugation (10 000 g for 15 min at 4°C), concentrated by ultracentrifugation (100 000 g for 120 min at 4°C) and suspended in sterile distilled-deionised water. Virus stocks were passed through 0.22µm cellulose acetate (Millipore) and 30 nm polycarbonate (Bio-Rad) membrane filters and stored at -80°C. Cell monolayers for plaque assay were prepared in 12-well plastic culture plates. Approximately $5x10^4$ viable cells suspended in 2 ml of growth medium (MEM with 5% FCS) were added to each well and the plates were incubated at 37°C in sealed plastic bags. Within 48 h the cell monolayers were ready to be used. The monolayers were washed with Earle's balanced salt solution (EBSS) and after virus inoculation (0.1 ml), the monolayers were maintained at 37°C under 0.6% agarose MEM overlay. The plaques were generally ready for counting after 48 h.

Experimental water was dechlorinated, filter sterilised Ottawa tap water. The relevant characteristics of the tap water were: turbidity 0.45 NTU, pH 8.6, total alkalinity 23.0 mg l^{-1} as $CaCO_3$, total hardness 54.8 mg l^{-1} as $CaCO_3$ (Ca 41.8; Mg 13.0), specific conductance 130.0 µsiemens cm^{-1}, and chloride 4.5 mg l^{-1} as Cl^-.

Batch sorption tests were performed at room temperature (21°C) and the reaction mixture consisted of a total volume of 5 ml containing 0.05 g (10 g l^{-1}) of sorbent and an input poliovirus level of $2x10^6$ - $3x10^6$ plaque forming units (PFU) l^{-1}. This was placed in a 6 ml tube and agitated in an end-over-end shaker for a desired contact time. Thereafter, the tube was centrifuged (10 000 g for 5 min) and the supernatant sample (0.9 ml sample + 0.1 ml 10x EBSS) was titrated for unsorbed virus. pH of the experimental water was varied using sodium hydroxide or hydrochloric acid as required. For sorption equilibria studies, input virus level was varied in the range $4x10^6$ - $5x10^7$ PFU l^{-1}. For desorption tests to study the fate of the sorbed viruses, the sorbent (10 g l^{-1}) was loaded with virus using a procedure similar to the batch sorption tests. Following equilibrium virus sorption, the tube was centrifuged (10 000 g for 5 min) and 0.5 ml of the supernatant was withdrawn for estimation of virus loading. Thereafter, 0.5 ml of eluent (10% FCS or 10X tryptose phosphate broth at pH 9.5) was added to the tube, agitated for 30 min, centrifuged (10 000 g for 5 min) and the supernatant titrated for desorbed virus.

All experiments and virus titrations were performed in triplicate. Data are presented as arithmetic mean ± standard deviation (S.D.) in tables and arithmetic mean ± SEM in arithmetic plots.

Downflow column study was performed using dechlorinated tap water @ 0.09 $l\ h^{-1}$ in a 10 mm ID glass column with a sorbent bed depth of 8 cm and influent poliovirus level of 1.54×10^5 PFU l^{-1}. This provided a bed flowthrough time of 5.33 min. Following termination of the run, the bed was flushed with 15 ml of tryptose phosphate broth at pH 9.5 as eluent to assess the fate of the retained viruses.

RESULTS AND DISCUSSION

The results of a batch sorption screening test to assess poliovirus sorption potential of the coal-based sorbents (GBC, Alum-GBC, Ferric hydroxide-GBC, Lime-GBC and δMnO_2-GBC) and F-400 active carbon are shown in Table 1. Based on the results of bacterial virus sorption on bituminous coal (Oza and Chaudhuri, 1976), a 2 h contact time was employed in the screening test. Alum-GBC was the most promising sorbent in the pH range 6.3 - 8.9 and its poliovirus sorption potential was superior to that of F-400. Further sorption experiments were conducted using Alum-GBC and F-400.

It is difficult to explain the poliovirus sorption behaviour of the sorbents in terms of electrokinetic considerations. Isoelectric point (pI) of poliovirus lies in the range 6.6 - 8.2 (Gerba, 1984) whereas the pH of zero point charge (pH_{ZPC}) of the sorbents are: GBC 7.0 - 7.15, Alum-GBC 4.0, Ferric hydroxide-GBC 8.0 - 8.5, Lime-GBC 8.0, δMnO_2-GBC 4.5 and F-400 7.1. The overall charge of a surface will be positive below pI or pH_{ZPC} and negative above it; however, pockets of positive and negative charge will exist across the surface. Strong sorption of poliovirus onto Alum-GBC in the pH range 6.3 - 8.9 indicates probable existence of strong inherent attractive forces. As believed by Murray (1980), it is probably due to strong Lifshitz-van der Waals potentials which are larger than double-layer interactions at ionic strengths found in most natural waters.

Kinetics of sorption of poliovirus onto Alum-GBC and F-400 at pH 7.7 are shown in Fig. 1. For Alum-GBC, virus sorption was rapid and a plateau was reached in 30 min as compared to 90 min for F-400.

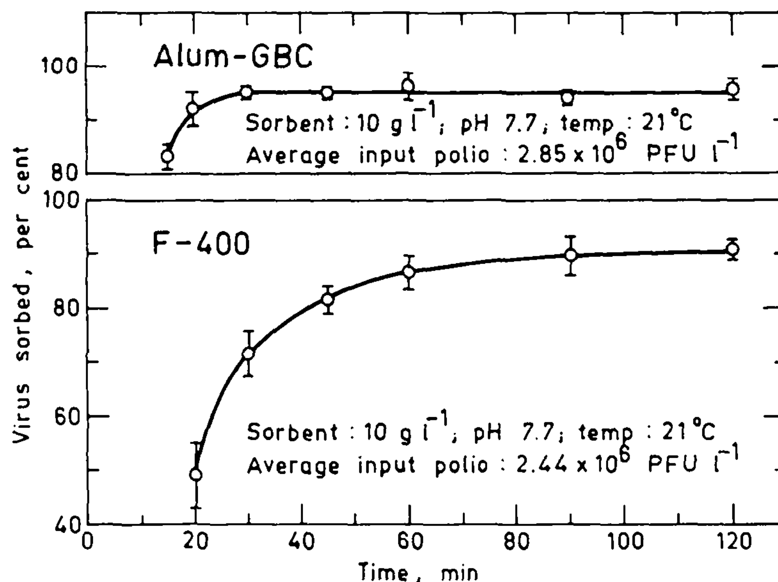

Fig. 1. Kinetics of sorption of poliovirus onto Alum-GBC and F-400

Table 1. Poliovirus sorption onto coal-based sorbents and active carbon

Sorbent	pH 6.3 Input: 2.37×10^{6} PFU l^{-1}		pH 7.7 Input: 2.34×10^{6} PFU l^{-1}		pH 8.9 Input: 2.83×10^{6} PFU l^{-1}	
	% Virus sorbed	Mean ±S.D.	% Virus sorbed	Mean ±S.D.	% Virus sorbed	Mean ±S.D.
GBC	82.33 79.37 75.30	79.00 ±3.53	78.33 72.09 75.90	75.44 ±3.15	No detectable sorption	
Alum-GBC	99.28 92.27 98.28	96.61 ±3.39	97.76 93.34 96.84	95.98 ±2.33	98.83 94.60 93.70	95.71 ±2.74
Ferric hydroxide-GBC	17.31 18.84 19.75	18.63 ±1.23	No detectable sorption		No detectable sorption	
Lime-GBC	No detectable sorption		No detectable sorption		No detectable sorption	
δMnO_2-GBC	63.38 69.57 69.13	67.36 ±3.45	35.35 29.34 27.83	30.84 ±3.98	70.00 66.86 63.33	66.73 ±3.32
F-400	97.18 93.56 96.57	95.77 ±1.94	93.26 93.17 96.10	94.18 ±1.67	92.74 91.63 95.18	93.18 ±1.82

Sorbent: 10 g l^{-1}; sorption time: 2 h; temp: 21°C

Figure 2 shows plots of poliovirus sorption (2 h) equilibrium data according to the linearised form of the Langmuir equation:

$$\frac{1}{q} = \frac{1}{Q^\circ} + \frac{1}{bQ^\circ} \cdot \frac{1}{C}$$

in which q = number of poliovirus sorbed per unit weight of sorbent at equilibrium (PFU g^{-1}), C = equilibrium concentration of poliovirus (PFU l^{-1}), Q° = number of poliovirus sorbed per unit weight in forming a monolayer or limiting sorptive capacity (PFU g^{-1}), and b = a constant related to energy of sorption. Alum-GBC exhibited a limiting sorptive capacity (4.7828×10^{7}PFU g^{-1}) about one log higher than that of F-400 (5.240×10^{6} PFU g^{-1}). Sorption energy for Alum-GBC was also greater than that of F-400 (b = 6.68×10^{-7} for Alum-GBC and 1.28×10^{-7} for F-400). Based on the data of sorption screening test as well as kinetic and equilibria studies, Alum-GBC may be regarded superior to F-400 active carbon as a sorbent for removing poliovirus from water.

Data on desorption of poliovirus from virus loaded Alum-GBC using fetal calf serum (FCS) and tryptose phosphate broth at pH 9.5 (TPB) as eluents are presented in Table 2. Murray and Laband (1979) also observed a decrease in infectivity following poliovirus sorption onto α-Al_2O_3 and radioactivity data indicated virus inactivation. However, elution of infectious viruses indicates the need for careful disposal of the sorbent following its use to remove viruses from water.

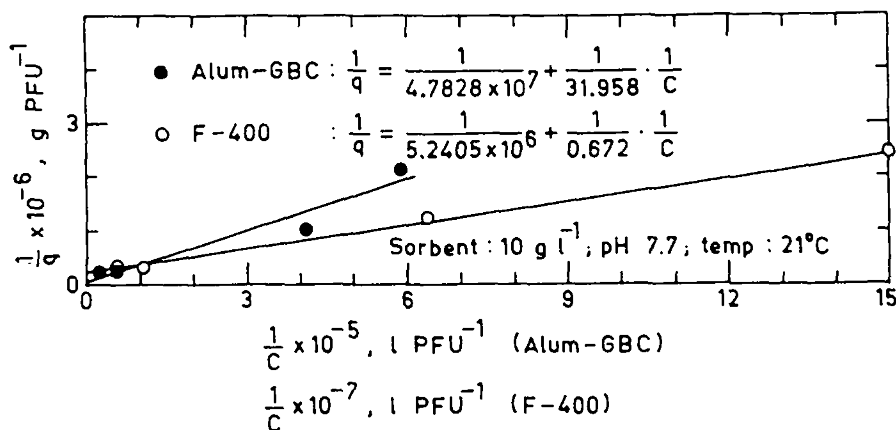

Fig. 2. Linearised plots of poliovirus sorption equilibria data according to Langmuir equation

Table 2. Desorption of poliovirus from Alum-GBC

Eluent	Input poliovirus	% Virus sorbed in 2 h	Mean ±S.D.	% Virus desorption	Mean ±S.D.
TPB	2.69×10^{6} PFU l^{-1}	99.48 99.57 99.28	99.44 ±0.15	48.40 48.50 58.13	51.68 ±5.63
FCS	3.32×10^{6} PFU l^{-1}	98.43 99.10 96.20	97.91 ±1.52	13.68 13.59 9.77	12.35 ±2.23

Sorbent: 10 g l^{-1}, pH 7.7; temp: 21°C; sorption time: 2h; desorption time: 30 min

Results of a downflow column study employing Alum-GBC as a filter bed are shown in Table 3. Consistently high removal of poliovirus further demonstrates the potential of Alum-GBC as a sorbent for removing viruses from water. Flushing of the column with tryptose phosphate broth at pH 9.5 eluted 12.68% of the retained viruses.

Table 3. Alum-GBC column study

Throughput volume, ml	% Virus sorbed
25	96.39
50	97.60
100	93.97
200	98.90
300	93.97
500	93.97

Influent virus: 1.54×10^{5} PFU l^{-1}; pH 8.6;
bed depth: 8 cm; flow rate: 0.09 l h^{-1}

A previous study by Prasad (1986) demonstrated the potential of Alum-GBC and several other low-cost sorbents for removing enteric bacteria from water. A 5 cm Alum-GBC bed with a flowthrough time of 5 min was able to remove nearly 100% *Escherichia coli* ($5x10^4$ CFU l^{-1}) from spiked dechlorinated tap water. It is likely that water filters prepared from low cost coal-based sorbents like Alum-GBC may prove useful for domestic use in rural areas of India and other developing countries. However, more studies are needed in this area, especially to test the ability of such sorbents in removing other significant enteric viruses such as hepatitis A and rotaviruses.

SUMMARY AND CONCLUSIONS

The present study showed the potential of a low-cost coal-based sorbent (alum pretreated bituminous coal) for removing poliovirus from water. Compared with active carbon, it exhibited faster virus sorption and higher limiting sorptive capacity. A previous study demonstrated the enteric bacteria removal potential of the sorbent. It is likely that water filters prepared from such low-cost sorbents may prove useful for domestic use in rural areas of India and other developing countries.

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REFERENCES

Anderson, N.J., R.J. Eldridge, L.O. Kolarik, E.A. Swinton, and D.E. Weiss (1980). Colour and turbidity removal with reusable magnetic particles. I. Use of magnetic cation exchange resins to introduce aluminium ions. Water Res., 14, 959-966.

Anderson, N.J., L.O. Kolarik, E.A. Swinton, and D.E. Weiss (1982). Colour and turbidity removal with reusable magnetic particles. III. Immobilised metal hydroxide gel. Water Res., 16, 1327-1334.

Bhattacharya, A.K., and C. Venkobachar (1984). Removal of cadmium (II) by low-cost adsorbents. J. Env. Eng. Div., Proc. Am. Soc. Civil Engrs., 110, 110-122.

Bitton, G., S.R. Farrah, C. Montague, M.W. Binford, P.R. Sheuerman, and A. Watson (1985). Survey of Virus Isolation Data from Environmental Samples. Department of Environmental Engineering Sciences, University of Florida, Gainesville, Florida, U.S.A.

Cullen, G.V., and N.G. Siviour (1982). Removing metals from waste solutions with low rank coals and related materials. Water Res., 16, 1357-1366.

George, A.D., and M. Chaudhuri (1977). Removal of iron from ground water by filtration through coal. J. Am. Water Wks. Assn., 69, 385-389.

Gerba, C.P. (1984). Applied and theoretical aspects of virus adsorption to surfaces. Adv. Appl. Microbiol., 30, 133-168.

IAWPRC Study Group on Water Virology (1983). The health significance of viruses in water. Water Res., 17, 121-132.

Jayasimha, B.N. (1983). Sorption of Humic Acid in Water on Hydrous Fe/Mn-oxides Impregnated Coal. M. Tech. dissertation, Indian Institute of Technology, Kanpur, India.

Karthikeyan, J., and M. Chaudhuri (1986). Enhancement of mercury (II) sorption from water by coal through chemical pretreatment. Water Res. (in press).

Kirankumar, T.V. (1984). Sorption of Mercury (II) on Coal Impregnated with Oxides of Manganese. M. Tech. Dissertation, Indian Institute of Technology, Kanpur, India.

Murray, J.P. (1980). Physical Chemistry of Virus Adsorption and Degradation on Inorganic Surfaces. U.S. Environmental Protection Agency, Cincinnati, Ohio, U.S.A.

Murray, J.P., and S.J. Laband (1979). Degradation of poliovirus by adsorption on inorganic surfaces. Appl. Env. Microbiol., 37, 480-486.

Oza, P.P., and M. Chaudhuri (1976). Virus-coal sorption interaction. J. Env. Eng. Div., Proc. Am. Soc. Civil Engrs., 102, 1255-1262.

Pandey, M.P., and M. Chaudhuri (1982). Removal of inorganic mercury from water by bituminous coal. Water Res., 16, 1113-1118.

Prasad, V.S. (1986). Development of Filtration/Adsorption Media for Removal of Bacteria and Turbidity from Water. Ph.D. dissertation, Indian Institute of Technology, Kanpur, India.

Rao, V.C., S.V. Waghmare, and S.B. Lakhe (1981). Detection of viruses in drinking water by concentration on magnetic iron oxide. Appl. Env. Microbiol., 42, 421-426.

Sattar, S.A., M.K. Ijaz, C.M. Johnson-Lussenburg, and V.S. Springthorpe (1984). Effect of relative humidity on the airborne survival of human rotavirus SA-11. Appl. Env. Microbiol., 47, 879-881.

Sriramulu, N., and M. Chaudhuri (1976). Dual-media coal-sand filter and its virus removal potential. Ind. J. Env. Health, 18, 77-86.