

VIRUSES IN WASTE, RENOVATED, AND OTHER WATERS

1972

LITERATURE ABSTRACTS

**U.S. ENVIRONMENTAL PROTECTION AGENCY
Office of Research and Monitoring
National Environmental Research Center
Cincinnati, Ohio**

VIRUSES IN WASTE, RENOVATED, AND OTHER WATERS

**Editor: Gerald Berg, Ph.D.
Editorial Assistant: F. Dianne White**

1972

**VIROLOGY SECTION
ADVANCED WASTE TREATMENT RESEARCH LABORATORY
OFFICE OF RESEARCH AND MONITORING
NATIONAL ENVIRONMENTAL RESEARCH CENTER
ENVIRONMENTAL PROTECTION AGENCY
CINCINNATI, OHIO**

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Ahmed, T. M. A. (1972). *Effect of Ultraviolet Irradiation on Chlorine Residuals in Polluted Waters.* **BIOCHEM J**, 128(1):39.

More than 50 samples of river water, polluted with sewage and industrial wastes containing high concentrations of suspended solids and dissolved impurities, were chlorinated and exposed to ultraviolet irradiation for six hours. About 60% of the chlorine dissipated in the first two hours. Less chlorine was lost at the greater depths. A reduction in pH level accompanied the chlorine loss.

Andriashvili, I. A., Tikhonenko, T. I., Gushchin, B. V. (1971). *Isolation, Concentration, Purification and Biological Properties of Enteric Phages.* **VOPR VIRUSOL**, 16(9-10):532-4. Russian.

Five phages of the F1 series were recovered from sewage on *Escherichia coli* K12F⁺. The phages were serologically unrelated to phages of the T and DD series. A method was developed for concentrating and purifying the phages.

Babov, D. M., Gubenko, L. T., Muromtseva, A. A., Yarotskaya, N. E. (1971). *Enterovirus Circulation in the Environment and Among the Population of Odessa.* **HYG & SANIT**, 36(3):388-91.

Summary appeared in 1971 edition of these abstracts. The paper was published originally in **GIG SANIT**, 36:54-7 (1971).

Bagdasaryan, G. A., Abieva, R. M. (1971). *Survival of Enteroviruses and Adenoviruses in Water.* **HYG & SANIT**, 36(3):333-8.

Summary appeared in 1971 edition of these abstracts. The paper was published originally in **GIG SANIT**, 36: 10-14 (1971).

Bell, J. A. (1972). *Viruses and Water Quality.* J AMER MED ASSOC, 219(12):1628.

The hazard presented by viruses in sewage and water, the effectiveness of treatment processes in removing them, and the development of detection methodology are discussed.

Bricout, F. (1972). *Viral Infections: Dissemination and Epidemiology.* NOUV PRESSE MED, 1: 1643-7. French.

The roles of water, food, and contact are discussed in a general review of viral epidemiology.

Calabro, J. F., Cosenza, B. J., Kolega, J. J. (1972). *Bacteriophages Recovered From Septage.* J WATER POLLUT CONTRL FED, 44(12):2355-8.

Two morphological phage forms were recovered from septage, one a short-tailed variety that infected *Citrobacter freundii*, *Escherichia coli*, and cell-wall mutants of *Salmonella typhimurium*, and the other a long-tailed variety specific for *Shigella flexneri*. Only rough host strains were susceptible.

Concentrations of linear alkyl sulfonate normally occurring in septage did not effect the phage noticeably.

Čerkinskij, S. N., Trahtman, N. (1972). *The Present Status of Research on the Disinfection of Drinking Water in the USSR.* BULL WHO, 46(2):277-83.

Recent research in the USSR aimed at evaluating methods for disinfecting drinking water and at determining the mechanisms of microbial inactivation are reviewed. The advantages and disadvantages of chlorine, ozone, and gamma irradiation are discussed, as are their effects on *Enterobacteriaceae* and on enteroviruses.

Cherkinsky, S. N., Lovtsevich, E. L., Ryabchenko, V. A. (1971). *The Sanitary Significance of Escherichia coli in Various Conditions of Water Decontamination in Respect to Enteroviruses.* HYG & SANIT, 36(3):329-33.

Summary appeared in 1971 edition of these abstracts. The paper was published originally in **GIG SANIT, 36: 7-10 (1971).**

Cliver, D. O., Herrmann, J. E. (1972). *Proteolytic and Microbial Inactivation of Enteroviruses.* WATER RES, 6(6):797-805.

Some enteroviruses are sensitive to proteolytic enzymes. Coxsackievirus A9 was inactivated by proteolytic bacteria, notably by *Pseudomonas aeruginosa*. This inactivation could be distinguished from adsorption or

aggregation of the virus particles. ^{14}C label from the viral coat protein, but not ^{32}P from the viral nucleic acid, was taken up by the bacterial cells.

Dardanoni, L. (1971). *Circulation of Wild Polioviruses*. ANN SCLAVO, 13(5-6):353-61. Italian.

Circulation of wild polioviruses in Italy was greatly reduced by the mass vaccination programs with attenuated poliovaccines which began in 1964.

Besides the dramatic reduction in reported cases of paralytic poliomyelitis, a great reduction in poliovirus recovery from man and sewage occurred.

DiGirolamo, R., Liston, J., Matches, J. (1972). *Effects of Irradiation on the Survival of Virus in West Coast Oysters*. APPL MICROBIOL, 24(6):1005-6.

Poliovirus 1 survived 400 krad of gamma radiation from a cobalt source in whole and shucked oysters. From 7 to 13% of the virus survived this dose.

Taste tests with shellfish that had undergone 400 krad of irradiation revealed that the oysters had undergone organoleptic changes that had rendered them unpalatable.

DiGirolamo, R., Wiczynski, L., Daley, M., Miranda, F. (1972). *Preliminary Observations on the Uptake of Poliovirus by West Coast Shore Crabs*. APPL MICROBIOL, 23(1):170-1.

West Coast shore crabs (*Pachygrapsus sp.* and *Hemigrapsus sp.*), when in seawater contaminated with poliovirus 1 or when allowed to feed on virus-contaminated mussels (*Mytilus californianus*), accumulated large amounts of the virus.

DiGirolamo, R., Wiczynski, L., Daley, M., Miranda, F., Viehweger, C. (1972). *Uptake of Bacteriophage and Their Subsequent Survival in Edible West Coast Crabs After Processing*. APPL MICROBIOL, 23(6):1073-6.

Edible West Coast crabs (*Cancer magister* and *Cancer antennarius*), when in seawater contaminated with coliphage T4, accumulated large amounts of the virus.

From 2.5 to 20% of coliphage T4 survived boiling for varying periods in contaminated crabs.

Heat penetration studies showed that, although internal temperature in the crabs was sufficient to inactivate the virus, the processing times normally used to cook crabs were not. These results suggest that processed crabs may serve as vectors for the dissemination of virus diseases if the crabs are harvested from a polluted area.

Dragan, L. P., Goregliad, N. I. (1972). *Distribution of Adenoviruses Among the Population of Kiev in 1967-70, Based on Studies of Sewage.* MIKROBIAL ZH, 34(1-2):49-50. Translation presently not available. Ukrainian.

Ellender, R. D., Morton, F., Whelan, J., Sweet, B. H. (1972). *Concentration of Virus from Water by Electro-Osmosis and Forced-Flow Electrophoresis. II. Improvement of Methodology and Application to Tap Water.* PREP BIOCHEM, 2(3): 215-28.

A modified electroosmosis (EO) method was devised for concentrating viruses from distilled and tap waters. Selective membranes, which allow high rate water transport, demonstrated the importance of membrane technology to the EO procedure. The other modifications added were only secondary contributory factors.

With the technic as developed thus far, poliovirus 1, seeded into distilled or tap water at levels as low as 0.01 PFU/ml, was recovered. Reovirus, seeded at a level of 0.1 PFU/ml, was recovered. Physical concentration of diluent in all experiments averaged 40-fold. Problems were encountered when virus-containing tap water was concentrated, most likely, because of the high content of dissolved impurities which may increase viral inactivation or aggregation.

EO was more efficient than forced-flow electrophoresis as a dehydration procedure. However, the turbulent anode back-fractionation method did allow for an increase in the concentration of virus. Lower virus inputs may have accounted for the decrease in virus recovery compared with that in our previous studies where higher inputs were used.

Ellender, R. D., Sweet, B. H. (1972). *Newer Membrane Concentration Processes and Their Application to the Detection of Viral Pollution of Waters.* WATER RES, 6(6):741-6.

New classes of ultrafilter membranes (both cellulosic and synthetic polymeric varieties) for concentrating viruses from distilled water were tested in a model rapid flow, recirculating, thin channelized system under a positive pressure gradient (osmotic or compressed gas). In contrast to the adsorption-elution systems (membranes and polyelectrolytes), the method depends upon rapid transport of water and low molecular weight substances through the membrane and rejection of viruses and other high molecular weight materials. Because of flow rates, turbulence factors, and membrane properties, virus adsorption is minimized.

Polymeric membranes offer greater fluxes (dehydration rates) than cellulosic membranes. In tests with up to 20 liters of distilled water, greater than 50% recoveries of poliovirus 1 were obtained when virus inputs were 0.01 to 1 PFU/ml. Dehydration factors were 400X or greater. The reasons for virus loss are not understood, but may involve adsorption factors associated with the mechanical portions of the system.

England, B. (1972). *Concentration of Reovirus and Adenovirus from Sewage and Effluents by Protamine Sulfate (Salmine) Treatment*. APPL MICROBIOL, 24(3):510-12.

Protamine sulfate precipitated adenoviruses and reoviruses from sewage and effluents. Bovine albumin increased the efficiency of recovery of these viruses. The viruses were efficiently eluted from the precipitate with 1N NaOH. Eighty to 100% of exogenously added virus was recovered with this technic. Most enteroviruses were recovered inefficiently or not at all by this method.

The protamine precipitation method was at best as effective as the $\text{Al}(\text{OH})_3$ technic for recovering naturally occurring reoviruses from sewage and effluents, but superior to the insoluble polyelectrolyte (PE60) and CaHPO_4 adsorption methods.

Adenoviruses and reoviruses, frequently overgrown in plaque assays, were more readily recovered by tube assay methods.

Ergasheva, L. E., Artykov, M. S., Ilyinsky, I. I. (1972). *Sanitary-Virologic Features of Sewage in Certain Urban Sewerage Systems of Uzbek SSR*. GIG SANIT, 37(9):90-1. Translation presently not available. Russian.

Farley, C. A., Banfield, W. G., Kasnic, G., Jr., Foster, W. S. (1972). *Oyster Herpes-Type Virus*. SCIENCE, 178(11):759-60.

A herpes-type virus infection, the first to be found in an invertebrate animal, is reported in the oyster *Crassostrea virginica*. Intranuclear herpes-type viral inclusions were more prevalent in the oyster at elevated water temperatures of 28 to 30 C than at normal ambient temperatures of 18 to 20 C. The inclusions were associated with a lethal disease at the elevated temperatures.

Foliguet, J.-M., Doncoeur, F. (1972). *Inactivation of Enteroviruses and Salmonella in Fresh and Digested Wastewater Sludges by Pasteurization*. WATER RES, 6(11): 1399-1407.

In laboratory studies with seeded viruses and bacteria, thermal treatment of fresh and digested sewage sludges resulted in pasteurization. Reductions of more than 99.99% of poliovirus 1 and more than 99% of coxsackievirus B3 were obtained at 80 C in 10 minutes. The heat-up time was less than 10 minutes. Even greater reductions of *Salmonella paratyphi B* were demonstrated under these test conditions.

Francis, T. I., Wright, S. G., Onukogu, A. I., Okafor, E. E. (1972). *Clinical and Epidemiological Studies of an Epidemic of Jaundice in Aba and Onitsha, 1970*. W AFRICAN MED J, 21(4):43-6.

Eighty-four cases of jaundice occurred in two towns in the East Central State of Nigeria. Most of the cases were clinically compatible with infectious hepatitis, but a small group of patients had symptoms suggestive of mild yellow fever or other group B arbovirus infections, and a smaller group had hepatoma. These clinical findings were confirmed by serological data.

Epidemiological data suggested that the infectious hepatitis outbreak was waterborne.

Furuse, K., Watanabe, I. (1971). *Effects of Ultraviolet Light (UV) Irradiation on RNA Phage in H₂O and in D₂O*. VIROLOGY, 46(10):171-2.

The UV sensitivities of MS2 and Q β phages were slightly different in H₂O than in D₂O. There was little difference between the UV sensitivities of the whole phages and those of their RNA freed from coat proteins. Thus, the coat proteins apparently did not protect the nucleic acids of these phages against absorption of UV energy.

The inactivation of RNA phages by UV irradiation may be the result of pyrimidine hydration and pyrimidine dimerization or to changes in RNA configurations resulting from the secondary effects induced by hydration or dimerization.

Garibaldi, R. A., Murphy, G. D. III, Wood, B. T. (1972). *Infectious Hepatitis Outbreak Associated with Cafe Water*. HSMHA HEALTH REP, 87(2):164-71.

In May, June, and July 1970, 95 cases of infectious hepatitis occurred among residents of Polk County, Arkansas, and in surrounding counties in the State and in Oklahoma. Of the 95 patients, about 80% had patronized a cafe in Hatfield, Arkansas between mid-April and the end of July 1970. In a group of 470 area residents who had not been ill, only 19% had patronized the same cafe.

Epidemiologic and laboratory evidence incriminated cafe water as the most likely source of the epidemic. Of 78 hepatitis patients who had patronized the cafe, 97% had drunk water as compared with 55% of 53 non-ill patrons questioned. Although sanitary conditions inside the cafe appeared to be excellent, the tap water was contaminated with coliform bacteria. Fluorescein dye flushed down the toilet in the cafe appeared in the tap water 20 days later. Lateral seepage of effluent from a septic tank through underground shale fissures appeared to be the most likely route in contamination of the cafe water.

The cafe was closed voluntarily by the proprietress in June, and household contacts were treated with immune serum globulin. An immunization campaign for area residents was carried out in July to reduce the extent and severity of secondary spread.

Gentry, R. F., Braune, M. O. (1972). *Prevention of Virus Inactivation During Drinking Water Vaccination of Poultry*. **POULTRY SCI**, 51(4):1450-56.

Newcastle disease (ND) and infectious bronchitis (IB) drinking water type vaccines secured at the time of field application had sufficient viable virus to produce an immune response. IB vaccine with only 0.7 EID at the final drinking water concentration stimulated an immune response in 4-week old chickens.

IB virus titers were reduced in filtered tap water, both at room temperature and in an ice water bath. ND virus titers were also markedly reduced in filtered tap water at room temperature. Powdered skim milk protected against inactivation.

One mg/liter of chlorine or quaternary ammonium sanitizer inactivated ND, IB, and avian encephalomyelitis viruses. Skim milk neutralized the chlorine and the quaternary ammonium salt.

Virus neutralization tests on serums from 34 field flocks vaccinated against ND and IB without skim milk in the drinking water showed that 32% had not responded to the ND vaccine and 44% were still susceptible to IB. When skim milk had been added to the drinking water, all birds in 42 flocks had an immune response to the ND vaccine and only 2 (4.8%) failed to respond to IB vaccine.

The results indicated the value of adding skim milk to drinking water used for vaccinating chickens.

Globa, L. I., Lastovets, L. M., Rotmistrov, M. N., Golub, N. F., Radolits'ka, L. S. (1971). *Adsorbing Materials for Virus Removal from Water*. **DOPOV AKAD NAUK UKR SSR, Ser. B**, 33:1036-8. Ukrainian.

Polygoskite, bentonite, aglaporite, vermiculite, permutite, pyrophyllite, gypsum, and silica gel adsorb viruses from water, and may be used for that purpose prior to coagulation in water purification.

Globa, L. I., Lastovets, L. M., Rotmistrov, M. M. (1972). *Ability of Minerals to Adsorb Viruses from Water*. **MIKROBIOL ZH**, 34(1-2):64-5. Translation presently not available. Ukrainian.

Gloyna, E. F. *Disease Transmission Control as a Factor in Pond Design*. In "Waste Stabilization Ponds," Chapter 5, **WHO MONOGRAPH SER**, 60 (1971), 93-108.

Treatment facilities are not yet designed specifically for removing viruses and other pathogens, but more engineers and scientists are beginning to recognize that there is a problem.

The removal of BOD without regard to the destruction of disease-causing

agents is not enough. Pathogens must be destroyed in the wastewater treatment plant or diluted to a negligible risk level in the environment.

Grabow, W. O. K., Nupen, E. M. (1972). *The Load of Infectious Micro-Organisms in The Waste Water of Two South African Hospitals*. **WATER RES**, 6(12):1557-63.

Counts of microorganisms including pathogenic bacteria, viruses, and parasite ova in the sewage of two hospitals were compared with those in city sewage.

One of these hospitals is an isolation facility with stringent disinfection procedures. Counts of all the organisms tested, except *Pseudomonas aeruginosa*, were lower in the sewage of this hospital than in the sewage effluent of the town in which it is located. The other hospital is a general hospital with less stringent disinfection procedures. Counts of some organisms were slightly higher in the sewage of this hospital than in the sewage effluent of the city in which it is located.

Under normal conditions the wastewater of hospitals may need no treatment before disposal into city sewers. The discharge of primary hospital effluents into streams or their use for irrigation, however, should be considered with care.

Gromashevsky, L. V. (1972). *Evolution of Viruses and Viral Diseases*. **ZH MIKROBIOL EPIDEM IMMUN**, 12(12):48. Translation presently not available. Ukrainian.

Grushko, Y. M. (1971). *Non-Soviet Journals on Water Pollution*. **HYG & SANIT**, 36(2):251-3. (Same as *Problems of Sanitary Protection of Water Bodies in the Pages of Foreign Journals*, **GIG SANIT**, 36:73-5. Russian.)

This paper is a brief review of water pollution control literature, including some dealing with the viral problem, published in non-Russian journals.

Hall, M. W., Sproul, O. J. (1971). *Water Quality and Recreational Land Use*. **PUBLIC WORKS**, 102(3):52-6.

The increased demand for water-based recreation creates problems for water pollution control authorities. Degradation of surface waters by water-borne pollutants originating in shore-side camps and homes has received little attention. Disposal of treated wastewater which contains viruses and soluble plant nutrients that may be transported farther through the soil than organic materials and bacteria creates pollution problems for lakes and streams.

These problems are reviewed in some detail.

Hill, W. F., Jr., Akin, E. W., Benton, W. H., Metcalf, T. G. (1972). *Virus in Water. II. Evaluation of Membrane Cartridge Filters for Recovering Low Multiplicities of Poliovirus from Water.* APPL MICROBIOL, 23(5):880-8.

Poliovirus 1, added in large numbers to 100 gallons of tap or estuarine water containing 1,200 $\mu\text{g Mg}^{++}/\text{ml}$, were recovered completely in a Millitube MF cartridge membrane filter. Four elutions of 250 ml each with 5 X nutrient broth in 0.05 M carbonate-bicarbonate buffer at pH 9.0 recovered the virus from the cartridge. When 20 to 40 PFU of virus were added to 100 gallons of tap or estuarine waters, a two-step concentration procedure was used for virus recovery. Concentration first by the Millitube MF cartridge procedure followed by secondary concentration of the eluate by the two-phase polymer procedure resulted in virus recoveries of up to 67%.

Jordan, F. T. W., Massar, T. J. (1971). *The Influence of Copper on the Survival of Infectious Bronchitis Vaccine Virus in Water.* VET REC, 89(12):609-10.

Infectious bronchitis virus (chickens) survived longer in waters that did not have prolonged contact with copper pipes than in waters that did.

In laboratory experiments, the virus survived for more than six hours in water that contained less than 0.02 mg/liter of Cu^{++} (as CuSO_4), but only for two hours in similar water containing 0.2 mg/liter of Cu^{++} . The addition of iron filings (to displace the Cu^{++}) to the water, or the addition of powdered skim milk to a concentration of 0.1% increased virus survival time to at least six hours.

Kiseleva, L. F. (1971). *Survival of Polioviruses, Echoviruses, and Coxsackieviruses in Food Products.* VOPR PITAN, 30(11-12):58-61. Russian.

Enteroviruses survived for a long time in large numbers in tap water, milk, sour milk products, and bread. Hence, food products contaminated with enteroviruses may facilitate their spread among the population.

Konowalchuk, J., Speirs, J. I. (1972). *Enterovirus Recovery from Laboratory-Contaminated Samples of Shellfish.* CAN J MICROBIOL, 18(7):1023-9.

A sensitive reproducible method for recovering enteroviruses from the supernatant fluids of centrifuged homogenates of laboratory-contaminated shellfish meats is described. The supernatant fluids contained varying amounts of toxic materials that interfered with coxsackievirus B5 recovery in HEP-2 cells.

Most of the cytotoxicity was removed by precipitation with hydrochloric acid at pH 3.0 to 3.5. The acid-treated samples were diluted 1:4 in fetal bovine

serum and then mixed with cell suspensions to adsorb the viruses which were subsequently enumerated as plaques.

The method is applicable to oysters and especially to mussels and clams because of the high cytotoxicity of these shellfish. Other methods for concentrating sample fluids were also examined as a means for detecting enteroviruses in naturally contaminated shellfish.

Korsh, L. E., Bagdasaryan, G. A., Vlodavets, V. V. (1972). *Sanitary-Microbiological Studies in the Prevention of Bacterial and Viral Infection.* VESTN AKAD MED NAUK SSR, 27(4):71-8. Translation presently not available. Russian.

Kostenbader, K. D., Jr., Cliver, D. O. (1972). *Polyelectrolyte Flocculation as an Aid to Recovery of Enteroviruses from Oysters.* APPL MICROBIOL, 24(4):540-3.

A simple, rapid method for recovering enteroviruses from oysters is described. A polycation sewage flocculant promoted cohesion of oyster solids and thereby aided separation of these from the viruses. The suspension or extract obtained was inoculated directly into cell cultures or concentrated first for greater sensitivity.

Recovery of 80 to 100% of experimentally inoculated virus was achieved.

Lautier, F., Lavillaureix, J. (1972). *Recovery of Viruses From Polluted Waters, City Sewage, and from Urban and Rural Sewage Treatment Plants.* PATH MICROBIOL, 38(1):64-5. French.

Sixty-four samples of sewage, seven of river water (Ill River), and nine of treated effluents, all from the city of Strasbourg, were tested for viruses by the $Al(OH)_3$ adsorption procedure. Enteroviruses were recovered from eleven of the samples.

Lautier, F., Wilenski, A., Lavillaureix, J. (1971). *A Method for Detecting Viral Particles in Polluted Waters.* PATH MICROBIOL, 37:99-104. French.

Viruses were recovered from 100 ml volumes of sewage effluent by adsorption onto $Al(OH)_3$.

LeClerc, H. (1971). *Marine and Freshwater Pollution.* REV INTERN D'OCEANOGR MED, 24:155-70. French.

Pollution of marine and fresh water environments by microbes is discussed, with emphasis on *Vibrio*, *Clostridium*, *Salmonella*, *Mycobacteria* and viruses. The risk of infection to humans and methods for the prevention of microbial pollution are considered.

Legier, F. (1972). *Hygienic Problems in Swimming and Camping Areas*. OEFF GESUNDHEITSWES, 34(6):317-37. German.

Within a discussion of the sanitary requirements for swimming and camping facilities, the transmission of virus diseases, including the transmission of molluscum contagiosum and plantar warts, are noted.

Liebscher, S. (1970). *Enteroviruses in Swimming Pool Water*. Z GESAMTE HYG, 16(3):198-200. German.

Coxsackievirus B3 was recovered on HEP-2 cells from 20% of samples taken from an outdoor swimming pool. The virus caused meningitis and encephalitis in children. Bacteriological examinations of pool water disclosed high concentrations of coliforms.

Application of 0.3 to 0.5 mg chlorine/liter is not adequate for swimming pool disinfection.

Lindeman, S., Kott, Y. (1972). *The Effect of Chlorination on Enteroviruses in the Effluents of the Haifa Sewage Treatment Plant*. ISRAEL J MED SCI, 7(9):1111.

Two hundred and fifty ml of Haifa sewage effluent was passed through cellulosic membrane filters (0.45 μ m) and the filtrate, in turn, through an alginate ultrafilter (10 nm). The ultrafilter was dissolved in 3.8% sodium citrate, and viruses concentrated by this method were recovered by plaquing in primary *Cercopithecus aethiops* kidney cell cultures. About 35 to 45 PFU of viruses/100 ml of effluent was detected with this method as compared with only 3 to 5 PFU/100 ml with alum flocculation.

When 8 mg/liter chlorine were applied to effluents for one hour, no decrease occurred in virus numbers. However, after two hours of contact, virus numbers were decreased.

Long, W. N., Bell, F. A., Jr. (1972). *Health Factors and Reused Waters*. J AMER WATER WORKS ASSN, 64(4):220-5.

In a discussion of the possible health hazards associated with the reuse of renovated sewage, the virus problem is analyzed in some detail.

Lund, E. *Inactivation of Viruses*. In *Proceedings of the 6th International Conference on Water Pollution Research*, June 1972. Pergamon Press Ltd.

A number of chemical and physical treatments may inactivate viruses. Viruses may also become harmless spontaneously if kept outside proper cells for a sufficiently long period.

For most inactivation processes, we know little about the reactions taking place. The virus genome may not be able to take command in a cell. The virion may gain entrance into a cell, but fail to be stripped, or it may not get into a cell at all, or, the infectious nucleic acid may truly become inactivated.

Spontaneous inactivation of the small RNA viruses is a slow process in water at a temperature of 20 C or below. It is not an oxidation process. If it is the same process as thermal inactivation, it works faster on the proteins than on the nucleic acids. Heat-inactivated virus may contain infectious nucleic acid.

Viruses are inactivated faster at basic pH values. Alkaline inactivation may be the result of a process similar to thermal inactivation and thereby be one of protein denaturation, the result of splitting and uncoiling of proteins.

Spontaneous inactivation is accelerated in sea water. The virus inactivating capacity of sea water seems bound to sea water, and is not found in fresh water. Most workers point to one or more species of the marine microflora as the source of this inactivation. The nature of this process remains essentially obscure.

Viruses are sometimes more easily detected in primary sludge than in the water.

Chemical precipitation of sewage with calcium and aluminum salts may remove viruses from naturally contaminated waters, but the virus is essentially not inactivated. Contrary to what might be expected, virus may be demonstrable even when the precipitate has been obtained at pH 10.5 to 11.0. This must be considered in the disposal of sludge.

In what seems reasonable agreement both with the laws of physical chemistry and results empirically determined, the rate of inactivation of viruses by oxidizing disinfectants is dependent on oxidation potential.

Mack, W. N., Lu, Y. S., Coohon, D. B. (1972). *Isolation of Poliomyelitis Virus from a Contaminated Well*. HEALTH SERV REP, 87(3):271-4.

The water supply of a large restaurant was implicated as a source of infection in the illness of several patrons. A search for bacteria in the food and water source of the restaurant did not uncover the responsible agent.

However, poliovirus 2 was recovered from five gallons of the restaurant's well water concentrated by flocculation and ultracentrifugation.

Malherbe, H. (1971). *Viruses in Water*. S AFRICAN J SCI, 67(3)124-7.

The problem of viruses in sewage and water, particularly as it applies to the Union of South Africa, and the effectiveness of treatment technics are reviewed.

Markov, A. P. (1972). *Experience in the Sanitary-Virological Examination of Waste Waters.* **GIG SANIT, 37(2):94-5.** Translation presently not available. Russian.

Marks, H. C. *Residual Chlorine Analysis in Water and Waste Water.* In "Water and Water Pollution Handbook," Chapter 22, edited by L. Ciaccio. Marcel-Dekker, New York, New York (1972), 1213-47.

The measurement of chlorine and its compounds in water and sewage are reviewed in an extensive and detailed treatment of the subject.

Martin, A. E. (1972). *Medical Considerations in the Abstraction of Potable Waters from Polluted Sources.* **WATER TREAT & EXAM, 21(3):202-12.**

In a general review of the problem of pollution of water supplies, the need to protect against virus contamination of supplies, the means for doing so, and the standards designed for that purpose are discussed.

Morris, J. C. (1971). *Chlorination and Disinfection-State of the Art.* **J AMER WATER WORKS ASSN, 63(12):769-74.**

The state of the art of chlorination and disinfection with ozone, bromine, iodine, and other halogen species is assessed. The virucidal effectiveness of chlorine and some of its compounds is discussed. The relative value of ozone and the other halogens in different need situations is explored.

Morse, L. J., Bryan, J. A., Hurley, J. P., Murphy, J. F., O'Brien, T. F., Wacker, W. E. C. (1972). *The Holy Cross College Football Team Hepatitis Outbreak.* **J AMER MED ASSOC, 219(6):706-8.**

During a 15-day period in September and October 1969, an outbreak of infectious hepatitis affected the members of a college football team. Of 97 persons exposed, 90 were infected. Thirty-two experienced typical icteric disease, 22 were anicteric but symptomatic, and 36 asymptomatic players had significantly elevated serum glutamic pyruvic transaminase values (>100 units). Other athletes, using the same facilities but arriving six days after the established date of exposure, were unaffected.

The decision to obtain blood samples from the entire team as soon as the initial cases were recognized resulted in the demonstration of an unexpectedly high attack rate of 93%. Epidemiologic investigation revealed that an infected group of children in the neighborhood, an imperfect drinking water supply, a warm August day, a football team in training, and a local fire were links in the chain which resulted in this most unusual outbreak of infectious hepatitis.

Mosley, J. W. (1972). *Viral Hepatitis: A Group of Epidemiologic Entities*. CAN MED ASSOC J, 106,(2)427-34.

This paper comprises a thorough sophisticated analysis and discussion of viral hepatitis including waterborne infectious hepatitis.

Nestor, I., Gostin, L. (1971). *Dissemination of Enteric Viruses in Urban Areas*. STUD CERCET INFRAMICROBIOL, 22(3):209-14. Rumanian.

Surveys carried out during the 1962-1969 period on the presence of enteroviruses in sewage, rivers, and in water supplies in towns and in other environments are summarized. Viruses were concentrated with ion exchange resins, and with the *Saccharomyces cerevisiae*, and aluminum hydroxide adsorption methods.

Nikolaevskaya, Z. S., Aizen, M. S. (1972). *Detection of Minimal Concentrations of Viruses in Large Volumes By Ultrafiltration Through Soluble Lanthanum-Aluminum-Alginate Ultrafilters*. VOP VIRUSOLOGII, 6:723. Russian.

Double filtration through bacterial asbestos filters treated with 0.05% aqueous sodium alginate followed by concentration by ultrafiltration through soluble La-Al-alginate ultrafilters facilitated recovery of small amounts of viruses from large volumes of water. The concentration factor was 400 to 500. There appeared to be no loss of viruses.

With this method, enteroviruses and other cytopathic agents were recovered from 37% of 1-liter effluent samples taken from a plant on the Moskva River, and from 37% of 3-liter river water samples taken at Kalinin on the Volga River

Okun, D. A. (1972). *Safe Drinking Water*. AM J PUBLIC HEALTH, 62(7):903-4.

Community drinking water supplies should be developed from the best sources available to minimize the risk of transmission of viral and other microbial diseases and to minimize the ingestion of toxic and other hazardous substances.

Olifson, L. E., Baltenko, E. N., Bukharin, O. V., Pozhar, V. N., Turovets, G. L. (1970). *On the Feasibility of Comprehensive Utilization of Industrial and Domestic and Fecal Effluents in a Mining Combine*. HYG & SANIT, 35(12):412-13. (Same as *Utilization of Industrial and Agricultural Wastewaters*, GIG SANIT, 35(12):92-4. Russian.)

Poliovirus 2, echovirus 7, and coxsackievirus B3 survived for three to 15

days in acid (pH 2.0) and alkaline pH (11.0-13.5) industrial wastewaters from mining and ore processing operations.

Palfi, A. B. (1971). *Virus Content of Sewage in Different Seasons in Hungary*. ACTA MICROBIOL ACAD SCI HUNG, 18(4):231-7.

Three hundred and seventeen viruses were recovered from 336 sewage samples collected in Hungary in 1969. Sixty-two were polioviruses of vaccine origin. Of the remaining 255 strains, 43% were reovirus 1, 21% were echovirus 7, and 18% were echovirus 11. These three virus types were prevalent throughout the year. The other strains recovered were identified as coxsackieviruses B1, B3, and B4, and echoviruses 1, 6, 12, 14, 19, and 20.

The greatest number of viruses were recovered in August. Fewer recoveries were made during October, November, and September. The number of recoveries was lowest in February and March, following compulsory vaccination programs.

In 1968, reovirus 1, coxsackieviruses B1 and B3, and echoviruses 6 and 7 predominated.

Pana, A. (1971). *Concentration of Enteroviruses from Water by Adsorption on Insoluble Polyelectrolytes*. NUOVI ANN IG MICROBIOL, 22(11-12):415-27. Italian.

The insoluble polyelectrolyte method yielded better virus recoveries from surface water than the $Al(OH)_3$ adsorption or alginate filter methods.

Pavoni, J. L., Tittlebaum, M. E., Spencer, H. T., Fleischman, M., Nebel, C., Gottschling, R. (1972). *Virus Removal from Wastewater Using Ozone*. WATER & SEWAGE WORKS, 119(12):59-67.

Seeded coliphage f2 was stable in secondary effluent for at least five hours, and was unaffected by flow or mixing in an ozone reactor.

The virus was inactivated completely in five minutes by a total ozone dosage of approximately 15 mg/liter which left a residual of 0.015 mg/liter. The rate of inactivation was greater for f2 phage than for bacteria. Carbonaceous material reacted with ozone more slowly than the phage, also. The mechanism of destruction of bacteria and viruses was probably oxidative. Ozone may be a general cytoplasmic oxidant which causes cell lysis and the release of soluble COD.

Standards for ozone use in wastewater are needed. Current disinfection standards are concerned only with chlorine. The contact times and dosage levels for chlorine are not applicable to ozone. *Escherichia coli*, presently the only indicator of disinfection efficiency, cannot be used always as a virus indicator.

Poduska, R. A., Hershey, D. (1972). *Model for Virus Inactivation by Chlorination*. J WATER POLLUT CONTR FED, 44(5):738-45.

A mathematical model to explain the process of virus inactivation by chlorination has been developed and is based on the principles of first-order, irreversible chemical kinetics for the reaction between chlorine and virus particles in solution. The viruses studied were poliovirus 1 and coliphages f2 and MS2.

Inactivation is dependent on the time of contact between viruses and chlorine, the concentration of chlorine, and the pH of the system. The process is complicated by the clumping of viruses which results in a heterogeneous inactivation system. Each of these factors was included in the model in order to quantify the kinetics of inactivation.

In the mathematical analysis, the virus clump sizes were numbered from 1 to N, beginning with clump size 1. The model consists of N independent exponential terms that describe the inactivation of each of the clump sizes in a system. An inactivation rate constant is associated with each term, and characterizes the rate of inactivation of each clump size.

Powers, E. L., Gampel-Jobbagy, Z. (1972). *Water-derived Radicals and Radiation Sensitivity of Bacteriophage T7*. INT J RADIAT BIOL, 21(4):353-9.

Ethanol scavenging of $\cdot\text{OH}$ and $\cdot\text{H}$ in buffered, X-irradiated suspensions of coliphage T7 saturated with either N_2 , N_2O , or O_2 resulted in partial sparing of the virus. In pure suspension, therefore, some of the inactivation of T7 phage appears to drive from $\cdot\text{OH}$ activity.

Radiation-induced sublethal damage to the phage may play a part in determining sensitivity.

Rao, V. C., Chandorkar, U., Rao, N. U., Kumaran, P., Lakhe, S. B. (1972). *A Simple Method for Concentrating and Detecting Viruses in Wastewater*. WATER RES, 6(12):1565-76.

A modified membrane filter method was developed for recovering viruses from sewage and sewage effluents. Small amounts of enteroviruses seeded into autoclaved sewage were adsorbed onto $0.45\ \mu\text{m}$, 47 mm diameter membrane filters at pH 3 and eluted at pH 8. Clarifying the sewage first by homogenization for four minutes in a Waring blender and centrifugation at 1,800 g and 9,230 g facilitated filtration with no loss of virus. Complete recovery of the seeded virus was achieved.

Enteroviruses seeded into fecal suspensions (BOD = 600 mg/liter) adjusted to pH 3 and containing 1,200 mg Mg^{++} (as MgCl_2)/liter were completely recovered.

During a one-year program of monitoring raw sewage from a middle

income community in Nagpur, a maximum of 3,150 PFU of viruses/liter were recovered during the monsoon season and 11,575 PFU/liter were recovered during the winter.

Rowland, A. J., Skone, J. F. (1972). *Epidemiology of Infectious Hepatitis*. BR MED BULL, 28(2):149-55.

The epidemiology, distribution, prophylaxis, and therapy of infectious hepatitis are discussed.

Ruschi, A. (1971). *Presence and Epidemiological Significance of Polioviruses in Sewage in Pisa (Italy)*. ANN SCLAVO, 13(5-6):370-6. Italian.

The frequency of poliovirus recovery from the sewage of Pisa in 1962 and 1963, prior to mass live-poliovirus vaccination programs, peaked during the summer months.

In 1965 and 1966, subsequent to the initiation of vaccination programs, poliovirus recovery from sewage peaked during the winter months.

Other enteroviruses were recovered with about equal frequency during both sampling periods, with peak recoveries occurring during the summer months.

Scarpino, P. V., Berg, G., Chang, S. L., Dahling, D., Lucas, M. (1972). *A Comparative Study of the Inactivation of Viruses in Water by Chlorine*. WATER RES, 6(8): 959-65.

The inactivation rates of poliovirus 1 and *Escherichia coli* by hypochlorous acid (HOC1) in phosphate buffer (pH 6), and by hypochlorite ion (OC1⁻) in borate buffer (pH 10) were determined at 5 C.

Hypochlorite ion inactivated poliovirus 1 about seven times more rapidly than HOC1, whereas HOC1 inactivated *E. coli* about 50 times more rapidly than OC1⁻. Thus, poliovirus 1 was inactivated more rapidly at pH levels where the free chlorine was in the form of OC1⁻ rather than in the form HOC1. Alkalinity at pH 10 did not inactivate poliovirus 1 during the test period.

A reassessment of the chemistry of HOC1 ionization may be in order to determine whether borate buffer alters the equilibrium of the reaction suppressing ionization, or whether it brings about the formation of virucidal forms heretofore undescribed.

Shah, P. C., McCamish, J. (1972). *Relative Chlorine Resistance of Poliovirus 1 and Coliphages f2 and T2 in Water*. APPL MICROBIOL, 24(4):658-9.

A mixture of ammonia chloramines inactivated poliovirus 1 and coliphage T2 more rapidly than they inactivated coliphage f2.

Shane, M. S., Cannon, R. E., DeMichele, E. (1972). *Pollution Effects on Phycovirus and Host Algae Ecology*. J WATER POLLUT CONTROL FED, 44(12):2294-2302.

Algal viruses of *Plectonema boryanum* were recovered from polluted stretches of the Christina River, but not from the unpolluted headwaters. Recovery of the viruses correlated well with chemical, physical, and biological pollution indicators.

Shelton, S. P., Drewry, W. A. (1971). *Virus Removal by Chemical Coagulation*. Water Resources Research Center No. 15, Department of Civil Engineering Research Series No. 12, University of Tennessee, 121 pages.

In waste and river waters, aluminum sulfate, ferric chloride, and ferric sulfate were effective coagulants, removing more than 99% of seeded coliphage f2 in at least one of the two waters. Cationic polyelectrolytes were unsatisfactory as primary coagulants because they did not form good flocs. Cationic, nonionic, and anionic polyelectrolytes improved coagulation with aluminum sulfate; however, the value of the polyelectrolytes is doubtful because of their cost. Sodium aluminate, in conjunction with aluminum sulfate, was effective for virus removal. Optimum dosages of the two for turbidity and COD reduction, however, did not correspond to optimum dosages for virus removal.

A simple mathematical relationship may exist between raw water COD and the optimum dosage of primary coagulants for virus and turbidity removal.

The colloidal titration technic and the Zeta-Meter methods for locating isoelectric points did not always produce the same results, and the optimum dosages for all parameters often did not correlate well with either method for isoelectric point location.

Shitskova, A. P. (1971). *Hygienic Aspects of Preventing Digestive Tract Diseases*. GIG SANIT, 36(12):71-7. Translation presently not available. Russian.

Shuval, H. I., Katzenelson, E. *The Detection of Enteric Viruses in the Water Environment*. In "Water Pollution Microbiology," Chapter 15, edited by R. Michael, Wiley-Interscience, New York, New York (1972), 347-61.

Enteric virus diseases, especially infectious hepatitis, may be waterborne. Outbreaks may be epidemic or sporadic. Methods for evaluating the virological safety of water supplies need to be developed.

Many enteroviruses survive longer than coliforms in rivers and in sea water. These viruses are also more difficult to remove or inactivate by sewage and water treatment methods than coliforms.

Thus, efficient methods for detecting viruses in water need to be developed. For this purpose, adsorption to particulates, adsorption onto membrane filters, and phase separation are the most promising methods at this time.

Singh, K. V., Bohl, E. H. (1972). *The Pattern of Enteroviral Infections in a Herd of Swine. CAN J COMP MED, 36(7):243-8.*

Viruses were recovered in porcine kidney cell cultures from the feces of 26 pigs, 34 to 64 days old, in a healthy herd of swine. The pigs within each of four litters infected began shedding viruses in their feces at about the same time. The type of virus initially recovered was usually the same.

Subsequently, waves of infection with different enteroviruses occurred during the following six months. At least six antigenically different viruses were recovered from the herd over a 26-month period. Most, and perhaps all, of these viruses apparently belonged to the enterovirus group. No disease was associated with the infections.

The colostrum and milk of sows contained significant amounts of enteroviral antibodies. Prior to nursing, the serums of new-born pigs contained no enteroviral antibodies, but shortly after nursing high titers of such antibodies developed. Antibodies were detected also in the feces of suckling pigs.

Slack, J. G. (1972). *Water Reclamation from Sewage Effluent: Experimental Studies in Essex. WATER TREAT & EXAM, 21(3):239-58.*

Full-scale plant tests in a standard inverted pyramidal upflow tank, at a particle-settling velocity of 0.5 mm/sec, showed that about 99 to 99.9% of the viruses in sewage and about 95 to 99% of the *Escherichia coli* and other coliforms present were removed by treatment with 150 mg/liter of aluminum sulfate and 250 mg/liter of powdered carbon.

Sorber, C. A., Malina, J. F., Jr., Sagik, B. P. (1972). *Quantitative Procedure for Evaluating the Performance of Water and Waste Water Treatment Processes at Naturally Occurring Virus Levels. ENVIRON SCI & TECHN, 6(5):438-41.*

A modified, standardized insoluble polyelectrolyte technic was used to evaluate the removal of coliphage T2 from water and wastewater at levels down to 1×10^{-4} PFU/ml. Extrapolation of the results below this level is possible if sufficient quantities of water are available.

The efficiency of the method for concentrating the virus is pH-dependent. At the optimal pH, a constant virus recovery of 25% was achieved at phage concentrations of 10^3 to 10^{-4} PFU/ml.

Sorber, C. A., Malina, J. F., Jr., Sagik, B. P. (1972). *Virus Rejection by the Reverse Osmosis-Ultrafiltration Processes*. WATER RES, 6(11):1377-88.

Asymmetrical cellulose acetate membranes of the type used in ultrafiltration and reverse osmosis rejected from 99.2% to over 99.999% coliphage T2 and a poliovirus present in feed solutions of PBS or lake water.

A relatively constant 0 to 7.6 PFU/liter penetrated the membranes used at average feed water virus concentrations of 9.6×10^1 to 6.8×10^8 PFU/liter.

Penetration of the membranes by viruses appeared to be a random phenomenon.

Sproul, O. J. (1972). *Virus Inactivation by Water Treatment*. J AMER WATER WORKS ASSN, 64(1):31-5.

Nearly all water treatment processes in present use are under investigation to define better their virus-removal capacity. Recent efforts have defined some basic mechanisms of virus removal.

If the virus problem in water supplies is as large as the recent scientific and popular literature indicates, then research needs and research efforts stand in the same relationship as a mountain to a molehill. No research effort at all in the past 10 years has been given to virus removal by filtration. In some plants, over the same period, this process has become the only treatment unit, performing the roles of flocculation chamber, sedimentation tank, and filter.

Stille, W., Kunkel, B., Nerger, K. (1972). *Shellfish-Transmitted Hepatitis*. DTSCH MED WOCHENSCHR, 97(2):145-7. German.

Within a period of three years (May 1968 to October 1971) 425 patients with viral hepatitis were studied at the University Clinic in Frankfurt. In 34, the disease apparently followed consumption of oysters or other shellfish during travel outside of Germany.

Sweet, B. H., Ellender, R. D. (1972). *Electro-Osmosis: A New Technique for Concentrating Viruses From Water*. WATER RES, 6(7):775-9.

Concentration of viruses by electroosmosis in a Canalco CF-3 unit was possible only in small scale operation (5-liter quantities). The technic was useful with poliovirus 1 inputs of 0.01 PFU/ml and reovirus inputs of 0.1 PFU/ml or greater only.

In distilled water, under optimal conditions with cellulose acetate membranes and flux rates of 3 ml/cm²/hour, physical concentrations of approximately 40-fold were achieved in 2.5 to 3 hours with up to 80% virus recovery. Results with tap water were not as good.

Because of the mechanical complexity of the system—its power requirements, need for osmotic driving solutions and pumping and refrigeration

systems—the method is not economical for large scale use. The system may serve best as a second step procedure following initial concentration of large volumes of water by either ultrafiltration or other processes. Because it desalts and dehydrates, the method may also be useful for concentrating viruses from estuarine waters.

Sweet, B. H., McHale, J. S., Hardy, K. J., Klein, E. (1971). *Concentration of Virus from Water by Electro-Osmosis and Forced-Flow Electrophoresis*. PREP BIOCHEM, 1(1):77-89.

Poliovirus 1 was concentrated from water in Cananco Model CF-3 Electrophoretic Filter/Concentrator (modified after Bier) by electroosmosis and forced-flow electrophoresis.

Water was removed at a rate up to 0.8 ml/hr/cm² of membrane area by electroosmosis. With 12-14 V/cm (5-6 amps) and adjusted pumping rates, 20-fold concentration was achieved without virus loss.

During forced-flow electrophoresis, the virus, negatively charged in an alkaline buffer, moves toward the positive pole. At 20 V/cm and with adjusted pumping rates, the best concentration achieved was 3-fold with 10-fold dehydration. Virus spill-over at the cathode and virus adsorption at the anode were responsible for the poor results, but this may be overcome by adjustment of the voltage coupled with adjustment of the pumping rates.

Voltage (30 V/cm) and current (6 amps) have no detrimental effects on viral stability. These techniques appear to be more rapid and gentle than other methods for concentration of virus and may be scaled up for practical use.

Tarabčák, M., Kratochvíl, I., Milošovicová, A. (1971). *Effect of Vaccination with Live Poliovirus Vaccine on the Circulation of Enteroviruses in the Population*. J HYG EPIDEMIOL MICROBIOL IMMUNOL, 15(7):258-66.

Stools from randomly selected healthy children in East Slovakia were tested quarterly in 1959 (before mass vaccination) and from 1960 to 1966 (after mass vaccination) to determine alterations in the circulation of polioviruses and other enteroviruses in the population. More than 7,300 stools were tested.

Since 1963, monthly tests of sewage from nine localities in Kosice and in the Saca housing estate have been done. Almost 400 sewage samples were tested.

In 1959 (before mass vaccination), poliovirus detection in the stools of healthy children correspond to the mean monthly incidence of poliomyelitis from 1951 to 1959.

Since the initial mass vaccinations, no cases of poliomyelitis have been reported in East Slovakia. Vaccination of the young is carried on routinely. Circulation of wild polioviruses was suppressed and vaccine strains disappeared from the population within three to four months after vaccination.

Mass vaccination with live poliovaccine reduced the circulation of non-polioviruses at the time of vaccination and for two to three months thereafter as compared with 1959.

The same types of viruses recovered from stools were recovered from sewage.

The circulation of enteroviruses in a population can be determined by detection of these viruses in sewage.

Taylor, F. B. (1972). *The Holy Cross Episode*. J AMER WATER WORKS ASSN, 64(4):230-2.

The series of events is described that led to an outbreak of infectious hepatitis in the Holy Cross football team after they consumed water from a contaminated line.

Venosa, A. D., *Ozone as a Water and Wastewater Disinfectant: A Literature Review*. In "Ozone in Water and Wastewater Treatment", Chapter 5, edited by F. L. Evans III. Ann Arbor Science Publishers, Inc., Ann Arbor, Michigan (1972), 83-100.

In a general review of water and wastewater disinfection by ozone, the inactivation of viruses by this oxidant is discussed.

Victorin, K., Hellström, K.-G., Rylander, R. (1972). *Redox Potential Measurements for Determining the Disinfecting Power of Chlorinated Water*. J HYG, 70(6):313-23.

In chlorine demand-free water, inactivation of *Escherichia coli* by sodium hypochlorite, monochloramine, dichloramine, halazone, chloramine T, cyanuric acid+sodium hypochlorite and cyanuric acid+monochloramine correlated better with redox potential than with the amount of available chlorine. For individual pure chlorine compounds, available chlorine generally correlated better than the redox potential with bacterial inactivation.

Wachs, B. *Hygienic Standards For Sewage Effluents*. In "Fundamental Measurements and Effluent Conditions of Sewerages in Canal Stations, Purification Plants, and Receiving Waters," edited by H. Liebman. Verlag R. Oldenbourg, Munich, Germany, (1970), 50-78.

Diseases caused by viruses and other infectious agents are discussed, along with water standards for drinking and swimming.

Wacker, W. E. C., Riordan, J. F., Snodgrass, P. J., Chang, L. W., Morse, L. J., O'Brien, T. F., Reddy, W. J. (1972). *The Holy Cross Hepatitis Outbreak. Clinical and Chemical Abnormalities.* ARCH INTERN MED, 130(9):357-60.

Of 97 men exposed at the same time to infectious hepatitis virus in drinking water, one-third developed icteric disease, 60% developed anicteric disease, and 7% showed no evidence of disease.

An elevated serum glutamic pyruvic transaminase (SGPT) level was the most frequent chemical abnormality in the anicteric group. Serial measurements of the levels of SGPT, serum glutamic oxaloacetic transaminase (SGOT), ornithine carbamyl transferase (OCT), serum alkaline phosphatase, bilirubin, and cholesterol in the icteric group demonstrated the time course and extent of these abnormalities in this homogeneous population. The levels of serum bilirubin, alkaline phosphatase, and OCT decreased in parallel and were normal by 70 days after exposure. The SGPT and SGOT levels remained elevated longer.

By 145 days after exposure, some patients still had elevated SGPT and SGOT levels. Transient hypercholesterolemia occurred in a significant number of patients reaching a maximum 55 days after exposure. The mean value was normal 25 days later.

Wallis, C., Henderson, M., Melnick, J. L. (1972). *Enterovirus Concentration on Cellulose Membranes.* APPL MICROBIOL, 23(3):476-80.

Cellulose nitrate membranes adsorb viruses from water in the presence of salts. Trivalent salts were more effective than divalent salts. Thus, 0.5 mM $AlCl_3$ was as effective as 50 mM $MgCl_2$.

For testing 500 gallons of water, only 0.24 kg of $AlCl_3$ was required in contrast to 20 kg of $MgCl_2$. Virus was eluted from membranes that had an area of 486 cm² with 250 ml of pH 11.5 buffer. Lowering the pH of the eluate and adding $AlCl_3$ permitted the virus to be reabsorbed on a 4 cm² membrane from which it could be eluted in 1 ml of the high pH buffer.

This procedure provided the basis for concentrating small amounts of viruses from large volumes of water.

Wallis, C., Homma, A., Melnick, J. L. (1972). *Apparatus for Concentrating Viruses from Large Volumes.* J AMER WATER WORKS ASSN, 64(3):189-96.

An apparatus for recovering viruses from large volumes of water has been devised which consists of a series of textile filters capable of removing debris and certain soluble materials that interfere with virus recovery, cellulose membranes or an insoluble polyelectrolyte to which viruses adsorb and from which they can be eluted, pumps, and other ancillary equipment. The apparatus is capable of filtering 300 gallons of water/hour.

More than 60% of 100 PFU of poliovirus 1 seeded into 100 gallons of tap water were recovered by this method.

Wallis, C., Homma, A., Melnick, J. L. (1972). *A Portable Virus Concentrator for Testing Water in the Field*. WATER RES, 6(10):1249-56.

A system is described for concentrating viruses from large volumes of water. The system consists of a water pump, an electric generator, a series of clarifiers, a virus adsorbent, a virus reconcentrator, a 5- and 1-gallon pressure vessel with a small tank of nitrogen as a source of positive pressure, and ancillary equipment, all mounted on 2-wheel carts for easy portability.

The system was standardized with small amounts of poliovirus. The virus was added to dechlorinated city tap water in an amount so small it could not be detected unless the virus was first concentrated.

In the system, raw tap water containing virus was serially passed through clarifying filters with porosities of 5 to 1 μm to remove particulate matter, and then through a 1- μm cotton textile filter to electrostatically remove submicron ferric and other heavy metal complexes. These filters did not detectably remove viruses. Salts were then added to the running tap water to enhance adsorption of the viruses to a fiberglass or cellulose acetate filter. Raw water was processed at the rate of 33 gph, with complete removal of the virus from the water. Eighty percent of the virus was eluted from the adsorbent.

Wallis, C., Homma, A., Melnick, J. L. (1972) *Concentration and Purification of Influenza Virus on Insoluble Polyelectrolytes*. APPL MICROBIOL, 23(4):740-4.

Influenza virus was concentrated and purified rapidly by adsorbing it onto and eluting it from an insoluble polyelectrolyte. The influenza virus was first stabilized at pH 4 to 5, since viruses adsorb to the polyelectrolyte more efficiently in this pH range. A precipitate which forms in influenza harvests under acid conditions in the cold was removed by ammonium sulfate at a concentration which trapped the precipitate but not the virus. Ammonium sulfate-treated influenza virus in allantoic fluid was readily concentrated on the polyelectrolyte. Elution yielded a virus concentrate essentially free of nonviral proteins.

Walton, G., Becker, R. J., Champlin, R. L., Faust, S. D., McCabe, L. J., Pearson, H. E., Pogge, F. W., Weiser, P. W., Wolf, H. W. (1972). *Community Water Pollution R&D Needs*. J AMER WATER WORKS ASSN, 64(4):211-15.

In a general report on research and development needs, the American Water Works Association Committee on Pollution Parameters notes the current state of knowledge on viruses in water, and recommends expanded studies on the survival and persistence of viruses in that environment.

Watson, J. T., Drewry, W. A. (1971). *Adsorption of f2 Bacteriophage by Activated Carbon and Ion Exchange*. Water Resources Research Center Report No. 14, Department of Civil Engineering Research Series No. 11, University of Tennessee, 238 pages.

In batch tests and in column studies at variable flow rates, activated carbon (Pittsburgh and West Virginia Nuchar C-190) and an anion resin (Amberlite IRA-402) adsorbed seeded coliphage f2 from a synthetic river water and from trickling filter effluent. A cation resin (Amberlite IR-122) was not an effective virus adsorbent. Virus removal could be described by a first-order equation.

On a weight basis, West Virginia Nuchar C-190 was the most effective adsorbent in both the batch tests and in the column experiments. However, in the columns, Amberlite IRA-402 achieved the best virus removals in both test waters when the same depth of adsorbents was used. This removal exceeded 99.9% in both test waters when the flow rate was 0.5 gpm/sq ft. The removals were considerably greater in the synthetic river water than in the wastewater effluent.

Competition for adsorption sites by other organic matter was a major factor in the wastewater effluent experiments. Diffusion seemed to play an important role in the virus adsorption process in the synthetic river water column studies.

Westman, W. E. (1972). *Some Basic Issues in Water Pollution Control Legislation*. AM SCIENTIST, 60(11-12):767-73.

The need for more intensive attention to viruses is considered in a discussion of water pollution control legislation.

Zarma, M. (1972). *Fecal Pollution of Sea Water*. MICROBIOL PARAZITOL EPIDEMIOL, 17(5-6):203-29. Rumanian.

The survival of viruses in sea water is briefly discussed in a review dealing primarily with other sea pollution problems.

Zikmund, V., Čech, M., Ěemin, K., Perman, J. (1972). *Epidemic of Infectious Hepatitis in a School Caused by a Waterborne Agent*. CS EPIDEMIOL, 21(7):197-202. Czechoslovakian.

Early in 1970, an epidemic of infectious hepatitis occurred in a Basic Nine-Year School in Liberec. The infection appeared to have been transmitted by water from a well supplying the school kitchen. About one-third of those exposed became ill.

The water may have been contaminated by a cesspool from a small house in which a case of infectious hepatitis had occurred.

At least 23 cases of hepatitis were documented, and there may have been as many as 44. Tests for Australia antigen on acute and convalescent serums were negative.

Zillich, J. A. (1972). *Toxicity of Combined Chlorine Residuals to Freshwater Fish*. J WATER POLLUT CONTRL FED, 44(2):212-20.

Laboratory studies showed that quantities of chlorine below measureable levels added to nontoxic effluents containing thiocyanate produced a toxicant. The toxicant was probably cyanogen chloride. The threshold concentration of this compound for rainbow trout appeared to be 0.08 mg/liter.

Chloramine concentrations of a few tenths mg/liter were lethal to warmwater fish such as sunfish, bullheads, and minnows.

Chloramine concentrations of 0.06 to 0.08 mg/liter were lethal to trout.

Chloramine concentrations of 0.085 mg/liter nearly eliminated the spawning of the fathead minnow, and concentrations of 0.043 mg/liter significantly reduced their reproduction.

Chlorinated effluents were toxic after dilution to 2 to 4%. Average concentrations of 0.16 to 0.21 mg/liter residual chlorine caused complete kills of fathead minnows. As little as 0.07 mg/liter caused a partial kill of the test fish, and 0.04 to 0.05 mg/liter constituted the threshold concentration in these wastes.

An extensive field survey of fish populations in four Maryland streams showed that chlorinated effluents significantly reduced the species diversity and total number of fish below the outfalls. After the toxic effects had been eliminated and the organic matter decomposed, the subsequent deoxygenation of the receiving streams caused species shifts and many of the more sensitive fish disappeared.

Thiosulfate rendered toxic chlorinated compounds nontoxic. This has been proven in laboratory and field situations.