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Critical Review of Virus Removal by Coagulation Processes and pH Modifications

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CRITICAL REVIEW OF VIRUS REMOVAL BY COAGULATION PROCESSES AND pH MODIFICATIONS

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FOREWORD

The Environmental Protection Agency was created because of increasing public and government concern about the dangers of pollution to the health and welfare of the American people. Noxious air, foul water, and spoiled land are tragic testimony to the deterioration of our natural environment. The complexity of that environment and the interplay between its components require a concentrated and integrated attack on the problem.

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This report is a state-of-the-art review of the literature concerned with the removal and inactivation of virus by chemical coagulation processes and pH modifications. Such information is necessary for a rational approach to the development and standardization of optimum treatment conditions for the removal and inactivation of viruses.

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ABSTRACT

Operation of advanced wastewater and water supply treatment plants to assure virological safety of the effluent relies on each unit process removing a finite number of viruses. These treatment plants frequently use chemical coagulation and precipitation at high pH with hydrated lime as part of the process. These treatment methods offer important opportunities for removal and inactivation of viruses from water and wastewater. This report is a literature review which examined the effectiveness of these processes in removing viruses.

Coagulation of water and wastewater for enteric virus removal should provide a removal of 90-99.999 percent of the influent viruses based on observations with polioviruses and Coxsackie A2. Either ferric or aluminum salts provide equal capability for virus removal when used in sufficient dosage. The required dosage is related to the conditions of the water and should be sufficient to provide a maximum removal of turbidity. The control of these processes for virus removal can be obtained by monitoring removal of turbidity. The pH of the water appears to influence virus removal and recommended pH values are 5-7 for virus coagulation with metallic coagulants where hydrated metal oxides are formed. The optimum pH where aluminum or ferric phosphates are the sole precipitate is between 5-6.5. Laboratory studies have shown a slight reduction in removal of viruses when organics are present as in treated effluents, but this is not supported by pilot plant data. Inorganics in the amounts in fresh waters do not appear to influence virus removal when floc formation is adequate.

Virus removal is unsatisfactory when polyelectrolytes are used as the primary coagulant in the usual concentration ranges of 1 to 2 mg/l. Virus removals are not increased when polyelectrolytes are used as coagulant aids if the coagulation process is otherwise acceptable. Where polyelectrolytes are used cationics would be preferred over the nonionics because of their greater density of positive charges. Anionic polyelectrolytes are not recommended since virus removal may be decreased in their presence.

Enteric viruses are inactivated at high pH and physically removed by absorption to precipitates formed under alkaline conditions in water. These viruses are, however, stable at pH values much lower than are obtained in either water and wastewater treatment. Virus inactivation in the absence of precipitates at high pH is affected by organics, inorganics, pH, contact time, temperature and type of virus. The exact effect of each parameter on virus inactivation cannot be determined from the present evidence, but the few viruses which have been studied indicate that an inactivation in excess of 90 percent should be obtained with a contact time of about 90 minutes at a pH of 11.5-12. Magnesium hydroxide and calcium hydroxylapatite have greater absorptive capacities for viruses than does calcium carbonate. Combined removal and inactivation of viruses in excess of 90-99 percent should be expected in the lime flocculation of a typical biologically

treated effluent with about 10 mg/l of phosphorus. Control of the process should be maintained by monitoring pH and turbidity with the objective of maximizing the turbidity removal.

Recommendations are included for application of these technologies to field situations.

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SECTION 1

INTRODUCTION

Operation of advanced wastewater and water supply treatment plants to assure virological safety of the effluent relies on each unit process removing a finite number of viruses. The sum of the removal in each of these barriers assures that safety of the finished water can be assumed. Principal processes in both advanced wastewater and drinking water treatment are coagulation, flocculation and sedimentation of the flocculent particles which are produced. It is known that viruses are removed in these processes to varying degrees. However, there has not been a systematic review of our present state of knowledge on the removal of viruses by these processes. This is particularly true in the removal of viruses by coagulation and flocculation under conditions where low or high pH conditions are obtained. Conditions of high pH are obtained during softening of water by precipitation and in waste water reuse systems for the precipitation of phosphorus, but the literature presents inconsistent data on virus removal by chemical coagulation, particularly with organic polymers. The apparent inactivation of viruses by pH is very high, however, a few investigators have presented results which show that the true inactivation may be significantly less than the apparent inactivation.

The principal objective of this project is to critically review the literature on removal or inactivation of viruses in water by chemical coagulation and by high and low pH.

SECTION 2

CONCLUSIONS

This literature review can be summarized and conclusions developed as shown below:

COAGULATION WITH METALLIC COAGULANTS

(1) Enteric virus removal from water and biologically treated effluents, based on observations with poliovirus and Coxsackie A2, will be from 90 to 99.999 percent.

(2) Other representative enteric viruses should be studied to confirm the coagulation removal prediction based on the poliovirus and Coxsackie A2 data.

(3) Either ferric or aluminum salts provide equal capability for virus removal when used in sufficient dosage. An optimum coagulant dose for virus removal cannot be determined. The dosage appears related to conditions of the water.

(4) Turbidity removal can be used as a process control for virus removal. Maximum virus removal occurs at or near the point of maximum turbidity removal. Minor reductions in virus removal may occur when coagulant dosage is adjusted for maximum turbidity removal rather than maximum virus removal.

(5) Water conditions will vary the amount of virus removal which may be expected. The few data available indicate that virus removal with metallic hydrated oxides is best at a pH of 5-7 and at a pH of 5-6.5 with aluminum or ferric phosphates. No conclusions can be drawn from the few data on virus removal variations caused by temperature variations, if in fact there is an effect. Laboratory studies have shown a slight reduction in removals when organics are present in moderate amounts (treated effluents) but this is not supported in pilot plant results. Inorganics, in amounts existing in fresh waters, do not influence virus removal when floc formation is adequate.

(6) The mechanism of virus removal appears to be one of a complex with the metallic cation at reactive virus sites, probably carboxyl groups among others, and incorporation into the floc.

(7) Laboratory and pilot plant data substantiate the reliability of this process for virus removal but confirming tests on a full plant have not been performed.

COAGULATION WITH POLYELECTROLYTES

(1) Virus removal is unsatisfactory when polyelectrolytes are used as primary coagulants in the usual concentration range of 1-2 mg/l.

(2) Virus removals are not increased when polyelectrolytes are used as coagulant aids if the coagulation process is otherwise acceptable.

INACTIVATION WITH pH EXTREMES WITHOUT PRECIPITATION

(1) Enteric viruses are inactivated at pH values of 10 to 12. The pH at which this occurs is evidently specific for each class of enteric viruses.

(2) Enteric viruses as a class are stable at pH values much lower than are obtained in water and wastewater treatment.

(3) The inactivation of viruses, particularly in the lower alkaline pH range where susceptibility has been observed, may be due to aggregation which causes a loss of infectivity, but is not true inactivation.

(4) Virus removal is affected by organics, inorganics, pH, contact time, temperature and type of virus. The magnitude of each parameter on the virus removal cannot be determined from the present evidence.

(5) The true inactivation of enteric viruses in this process, based on poliovirus 1 data, occurs by disruption of the protein capsid and loss of nucleic acid to the water. The nucleic acid may be partially degraded before its release to the water.

REMOVALS WITH PRECIPITATION AT HIGH pH

(1) Laboratory investigations have shown removals of poliovirus 1 up to 99.9 percent during water softening by precipitation and up to 99.1 percent during precipitation of calcium hydroxylapatite. Increased amounts of precipitates increased the virus removal.

(2) Pilot and full scale plant data show high removals of natural viruses during treatment of secondary effluents. Removals up to 99.9 percent or more have been obtained.

(3) The process appears reliable based upon the high removals obtained from several field and full scale studies.

(4) The questions posed above for removal of viruses by pH effects alone have not been answered in the high pH precipitation studies.

(5) Process control can be obtained by monitoring pH, turbidity, and suspended solids removal.

SECTION 3

CONVENTIONAL COAGULATION

BACKGROUND

A critical review of the literature on the removal of viruses by coagulation should answer a number of questions on the process. Among these questions are those shown below:

(1) Reliability of process. This concern can be examined by considering the following:

   (a) Plant scale data versus laboratory or pilot plant data. Similar results in full scale plant operation should be obtained under reasonably similar conditions in laboratory or pilot plants.

   (b) Reproducibility of data by different investigators. Results by various investigators removed from each other in time and place should be similar.

   (c) Delineation of factors affecting process. The coagulation process response to variables such as organic content, pH, temperature and salt content, among others, should be examined.

(2) Effectiveness of different coagulants. Differences between coagulants, if any should be determined. These differences would involve the following: coagulant dose, response to different viruses, effectiveness in waters with varying temperature, organic content, salt concentration, etc.

(3) Comparative removal of different viruses. Only a few of the more than 100 enteric viruses have been investigated to determine their response in the coagulation process. The ability of these tracer viruses to predict removals of all others should be assessed.

(4) Process control. Routine plant operation uses turbidity measurement as a control parameter. Virus removal may parallel turbidity removal and thus be monitored by measuring this parameter.

(5) Usefulness of coagulant aids. Polyelectrolytes have been used as coagulant aids for increased removal of turbidity and/or to obtain better settling of the floc formed in the process. They may also increase the effectiveness of virus removal.

(6) Determination of the mechanism of virus removal by coagulation. Knowledge of this mechanism would permit an extrapolation of virus removal from the few viruses which have been examined to the many which have not.

METALLIC COAGULANTS

This section of the review is restricted to the metallic coagulants, generally aluminum and iron salts, primarily used in "clean" waters representative of those entering a water treatment plant. Several cases of coagulation of an activated sludge effluent have been included because of their notable interest and careful control of the operation. The reported virus removals are those following the sedimentation phase.

The data from the literature have been divided to permit a general examination of coagulation conditions; enteric viruses versus bacterial viruses; and aluminum sulfate versus feric salts. In general the documented literature reports are those in which reasonable experimental control has been exercised. This has excluded several early reports such as those of Chang et al. (1), Carlson et al. (2) and Kempf et al. (3). These early reports among other things failed to note the importance of adding the viruses before the coagulation chemicals, failed to report data to evaluate the effectiveness of the coagulation process or failed to note that phosphate was a principal precipitate in studies where aluminum sulfate was used and not the hydrated aluminum oxide. Many other investigations have used materials such as aluminum oxides and phosphates and polyelectrolytes in solid forms as virus absorbers. Use of such materials in solid form does not duplicate the method of its use as a coagulant in water.

The data in Table 1 are for the coagulation of enteric viruses by metallic coagulants. Data for the removal of bacteriophages are presented in Table 2. Enteric virus data are available only for poliovirus 1, Coxsackie A2 and naturally occurring viruses (mainly Coxsackie B3 and B5). The data in Tables 1 and 2 are generally for the best removal results which were obtained by the various investigators. Factors which affect the reported removals will be presented in the following discussion.

Amount of Removal

The removals obtained for poliovirus 1 and Coxsackie A2 ranged from 48 to 99.999 percent. The 48 percent reported by Foliguet and Doncoeur (9) occurred when the clay concentration was low. These authors commented that better removals of viruses were obtained when they had an increased opportunity to absorb to clay particles. The virus removal then occurred with the concommittant removal of the clay. Lovtsevich et al. (18) in coagulating poliovirus 1 with $Al_2(SO_4)_3$ also noted better virus removal with higher initial turbidities. Other reports in Table 1 indicated higher orders of removal when the turbidity was at lower levels than that used by Foliguet and Doncoeur. The 70 percent removal of poliovirus 1 reported by Boardman and Sproul (6) was from a study where coagulation for virus removal was not optimized. Wolf et al. (4) reported 63 percent removal of poliovirus 1 and 46 percent for f2 phage in an activated sludge effluent when the aluminum

TABLE 1 COAGULATION OF ENTERIC VIRUSES WITH METALLIC COAGULANTS

Virus		Coagulant		Conditions of Coagulation					Removal				
Type	Initial Conc PFU/ml	Type	Dose mg/l or ppm	Type water	Turbidity	Temp °C	pH Start	pH End	Type Study	Virus %	Turbidity %	Ref.	Remarks
Polio 1(Vaccine)	596	$Al_2(SO_4)_3$	56	Act.sl.effl.	~10 FTU	29	7.1	6.8	P	63	~2.5 FTU	4	Al:P = .44 - 1
Polio 1(Vaccine)	113	"	NS	"	3.4 FTU	22	7.3	6.9	P	99.7+	1.5 FTU	4	Al:P = 7.1
Polio 1(Sabin)	$3\text{-}7x10^4$	$Al_2(SO_4)_3$	10	Spiked D.W.	50 mg/l clay	Room	NS	6.8	L	90	97	5	
Polio 1(Sabin)	$1\text{-}3x10^4$	$Al_2(SO_4)_3$	20	D.W.	None	22.5	7.0	7.0	L	70	-	6	Not opt. coag.
Polio 1(Chat)	130	$Al_2(SO_4)_3$ + Calgon WT 3000	76 + 0.2	Act.sl.effl.	4.6	24	7.3	6.9	P	95(avg)	10	7	Al:P = 1.7:1
Polio 1(Mahoney)	$10^{6.3}$ to $10^{8.37}$**	$FeCl_3$	66	Spiked demin.	45-178	13-21	7-7.7	6.6-6.9	P	99.7-99.999	0.6-3.8TU	8	
Polio 1(Mahoney)	$1.4x10^6$***	$FeCl_3$	60	Spiked demin.	25 mg/l clay 500mg/l clay	15-17	5-8	NS	P	48-99.7	NS	9	Poorer removal w/ low clay
Polio 1,2,3 (Glaxo oral vaccine)	$9.2x10^7$*	$Fe_2(SO_4)_3$	40	Polluted River	NS	NS	NS	NS	P	99.8	NS	10	
Coxsackie A2	$2.25x10^5$ +	$Al_2(SO_4)_3$	40	Spiked D.W.	0.4ml SiO_2	25	6.2	6.2	L	86	NS	11	
Coxsackie A2	"	"	60	"	"	"	"	"	"	96	"	"	
Coxsackie A2	"	"	80	"	"	"	"	"	"	97	"	"	
Coxsackie A2	"	"	100	"	"	"	"	"	"	99	"	"	
Coxsackie A2	$4.5x10^3$ +	$FeCl_3$	20	"	"	"	"	"	"	97	"	"	
Coxsackie A2	"	"	40	"	"	"	"	"	"	98	"	"	
Coxsackie A2	NS	$Al_2(SO_4)_3$	25	Ohio River	16-240	25	NS	6.7-7.3	L	99	1-5TU	12	
Coxsackie A2	NS	$Al_2(SO_4)_3$	25	Ohio River	140-255	15	NS	6.7-7.4	L	95	1-5TU	12	
Coxsackie A2	"	"	"	"	40-135	5	"	6.7-7.4	"	96	1-5TU	"	
Coxsackie A2	NS	$FeCl_3$	15	"	5-10	25	NS	8.1-8.4	L	95	0.1TU	12	
Naturally occurring (mainly Coxsackie B3 & B5)	.004 ++	$Fe_2(SO_4)_3$	40	Polluted River	NS	NS	NS	NS	P	No virus detected >88.3%	NS	10	

* Infectious particles (slug dose)

** ECP_{50}/ml

*** MPNCU/ml

\+ LD_{50}/ml

++ Infectious particles/ml

NS - Not stated

DW - Distilled Water

L - Laboratory

P - Pilot plant

TABLE 2 COAGULATION OF BACTERIOPHAGES WITH METALLIC COAGULANTS

Virus		Coagulant			Conditions of Coagulation		pH			Removal			
Type	Initial Conc PFU/ml	Type	Dose mg/l or ppm	Type Water	Turbidity	Temp °C	Start	End	Type Study	Virus %	Turbidity %	Ref.	Remarks
MS2	$2.5\text{-}5\times10^5$	$Al_2(SO_4)_3$	20-26	NS	120 mg/l clay	24-25	6.0	6.0	L	99.9	~97	13	
M. pyogenes phage	$5\times10^4\text{-}3\times10^6$	$Al_2(SO_4)_3$	80-100	Spiked D.W.	NS	25	6.2	6.2	L	99-99.8	NS	11	
M. pyogenes phage	NS	$Al_2(SO_4)_3$	25	Ohio River	16-240	25	NS	6.7-7.3	L	97	1-5 TU	12	
"	"	"	"	"	140-255	15	"	6.7-7.4	"	89	"	"	
"	"	"	"	"	40-135	5	"	6.7-7.4	"	84	"	"	
f2	$1\text{-}8\times10^5$	$Al_2(SO_4)_3$	25/21**	Natural Lake	2-4	23-25	8.1	7.3-7.4	L	99.9	96	14,15	
f2	NS	"	15/15**	Natural Surface	1.1-1.2	NS	NS	6.8	L	99.4	96	16	
f2	4.9×10^3	$Al_2(SO_4)_3$	53	Act.sl.effl.	~10	29	7.1	6.8	P	46	~2.5 FTU	4	Al:P = .44:1
f2	2.6×10^3	"	NS	Act.sl.effl.	3.4	22	7.3	6.9	"	99.8	~1.5 FTU	4	Al:P = 7:1
T4	$2.5\text{-}5\times10^5$	$Al_2(SO_4)_3$	20-26	NS	120 mg/l clay	24-25	5.2	5.2	L	98	~98	13	
MS2 phage	3.9×10^5	$FeCl_3$	50-60	NS	12.5 JTU	24-25	Optimum pH was 5.0		L	99.5	98	17	
M. pyogenes phage	$2\text{-}6.4\times10^5$	"	20-40	Spiked D.W.	NS	25	6.2	6.2	L	99.3-99.9	NS	11	
"	NS	$Al_2(SO_4)_3$	15	Ohio River	60-100	25	NS	7.1-7.4	L	86	5-10 TU	12	
f2	$0.7\text{-}1\times10^6$	$FeCl_3$	50/23**	Natural Lake	0.4-3	20-24	8.2	6.9*	L	99.4	92	14,15	
f2	$7\text{-}9\times10^5$	$Fe_2(SO_4)_3$	50/49**	"	2	22-24	8.3-8.4	7.5*	"	92	89	" "	
f2	1×10^6	$FeSO_4$	36	"	1-2	24-25	8.2	8.6*	L	94	Minor or increase	" "	
f2	NS	$FeCl_3$	40/34**	Natural Surface	1.1-1.2	NS	NS	6.8	L	99.1	95	16	
f2	NS	$Fe_2(SO_4)_3$	62/35**	"	1.1-1.2	NS	NS	7.2	L	99.91	90	16	
T4	$2.5\text{-}5\times10^5$	$Al_2(SO_4)_3$	20-26	NS	120 mg/l clay	24-25	5.2	5.2	L	98	~98	13	
Natural phage	54.2 (host-NS)	$Fe_2(SO_4)_3$	40	Polluted River	NS	NS	NS	NS	P	93	NS	10	
T4	$1.5\text{-}3\times10^{12}$ slugged into water	$Fe_2(SO_4)_3$	40	Polluted River	"	"	"	"	"	99.6	"	10	

* pH is opt. value for virus removal

** $\frac{\text{Opt. coag. dose for virus removal}}{\text{Opt. coag. dose for turb. removal}}$

NS - Not stated
DW - Distilled water
L - Laboratory
P - Pilot Plant

to phosphorus ratio was 0.44 to 1. Under such conditions no hydrated aluminum oxide would be expected to be formed with the precipitate consisting principally of aluminum phosphate (19). It may be inferred from Brunner and Sproul (20) that sharply reduced virus removals would be expected when the aluminum phosphate precipitation was done at Al:P mole ratios less than 1:1. When Wolf et al. (4) increased the Al:P ratio to 7:1 the poliovirus 1 removal increased to more than 99.7 percent and the f2 phage removal to 99.8 percent.

The removals reported in Tables 1 and 2 were from reports with higher influent virus concentrations than would be expected from a natural water source. A question of some significance is whether the percent removal is dependent upon the numbers of viruses in the influent stream. Guy et al. (10) reported removals in excess of 88 percent with naturally occurring enteric viruses in a pilot plant evaluation. Their study was done with only 0.004 infectious virus particles per ml in the influent water with a virus concentration technique detection limit of 0.00045 infectious particles per ml. Their study with 54.2 PFU/ml of naturally occurring phages provided removals of 93 percent. The Pomona virus study (7) also reported a poliovirus 1 removal of about 95 percent at a virus concentration of 1.3×10^2 PFU/ml in an activated sludge effluent using alum with a 1.7 to 1 Al:P ratio. Wolf et al. (4), on the other hand, obtained more than 99.7 percent removal of poliovirus 1 from an influent concentration of 113 PFU/ml. Their coagulation conditions were evidently better than that in the Pomona study since an Al:P ratio of 7:1 was used.

The laboratory and pilot plant work reporting on poliovirus 1, Coxsackie A2 and phage indicated that removals of these viruses at influent levels of 10^3 PFU/ml or more will be greater than 90 percent and may be as high as 99.999 percent. Sufficient studies have not been made with influent virus concentrations less than 100 PFU/ml to draw conclusions, but the available data indicate that the removals will be around 90 percent.

Type of Virus

Tables 1 and 2 present data on only two enteric viruses, poliovirus 1 and Coxsackie A2 plus, in one study, naturally occurring enteric viruses (principally Coxsackie B3 and B5) and four phages, MS2, f2, T4 and *Micrococcus pyogenes* phage plus the naturally phages in the Guy et al. (10) study. A comparison of the removal data between the two enteric viruses poliovirus 1 and Coxsackie A2 does not show marked differences between these viruses. The Coxsackie virus data for the spiked distilled water samples with low turbidity do not show the high removals obtained in certain of the polio studies. The aluminum sulfate coagulation of the Coxsackie virus with 40 to 100 mg/l gave removals from 86 to 99 percent, somewhat below the more than 99 percent removals with poliovirus obtained at lower coagulant doses. The Coxsackie data in natural river water with higher turbidities gave higher removals, 95 to 99 percent at lower coagulant doses. Data are available on too few enteric viruses to draw conclusions on whether there are real differences in their responses to coagulation.

Data in Table 2 show uniformly high removals by $Al_2(SO_4)_3$ coagulation for the f2, M. pyogenes phage and the MS2 phage. As discussed above, the 46 percent removal of the f2 observed by Wolf et al. (4) was caused by a known inadequate coagulation condition and is atypical. The T4 phage also showed a slightly lowered removal. The removal data for these phages, when coagulated using ferric salts, show few differences.

Too few enteric viruses have been examined to determine whether there are differences in their removability by the coagulation process. The bacteriophage data on four different phages do not show significant differences between themselves. Nor in fact, were there significant differences when their removals are compared with those of the enteric viruses in Table 1. The available data suggest that coagulation is relatively nonselective in virus removal.

Type of Coagulant and Dosage

The data in Tables 1 and 2 were obtained with $Al_2(SO_4)_3$, $FeCl_3$, $Fe_2(SO_4)_3$ and $FeSO_4$ and comparisons can be made of their effectiveness. These investigators were able to obtain enteric virus removals of 90 percent or more with each coagulant under the various conditions of their tests unless the coagulant dosage was known to be less than optimum. The phage data in Table 2 also show similar results except for the report on ferrous sulfate and the f2 phage. In discussing their ferrous sulfate work York and Drewry (15) and York (14) reported that virus removal with this coagulant was significant below 30 mg/l. It is also noted that minor turbidity removals occurred with this coagulant. These authors concluded that although an acceptable floc was formed and virus removal was in excess of 90 percent that since turbidity removal was poor the coagulant was not acceptable.

Coagulant dosages in practice are normally adjusted to minimize the turbidity leaving the sedimentation tank. It follows therefore that acceptable virus removals should desirably occur at these same concentrations. Data from Tables 1 and 2 have been extracted and presented in Table 3 to facilitate a comparison of the expected virus removal versus coagulant dosage. A comparison of these data do not show clearly a positive relationship between coagulant dosage and virus removal. It does appear, however, that in a given investigation increasing dosages to some optimum value increases the virus removal. This is presented rather clearly by the data from Chang et al. (11) when they increased removals of Coxsackie A2 from 86 to 99 percent with $Al_2(SO_4)_3$ dosages increasing from 40 to 100 mg/l. York (14) presented similar data but also showed that f2 phage removal decreased as each of his four coagulant dosages increased beyond the optimum value. Other investigators do not appear to have commented on this fact. York's observations are not unexpected if one considers that viruses are expected to behave, at least in part, as charged colloidal particles. Removals of such particles are often noted to decrease as the coagulant concentration increases and reversal of the colloidal charge occurs. He did not notice significant decreases in turbidity removals beyond the optimum concentration for its removal, however. The larger variation in coagulant dosages for the virus removals noted in Table 3 appears to be related to the

TABLE 3 VIRUS REMOVAL VERSUS METALLIC COAGULANT DOSAGE*

Coagulant Type	Dosage mg/l	Type Virus	Water System	Removal %	Reference
$Al_2(SO_4)_3$	10	Polio 1	Spiked DW	90	5
	20	"	DW	70	6
	40	"	Polluted River	99.8	10
	76	"	Act. sl. effl.	95	7
	40	Coxsackie A2	Spiked DW	86	11
	60	"	"	96	11
	80	"	"	97	11
	100	"	"	99	11
	25	"	Ohio River	95-99	12
	20-26	MS2	NS	99.9	13
	20-26	T4	NS	98	13
	15	f2	Natural surface	99.4	16
	NS	"	Act. sl. eff.	99.8	4
	25	"	Natural surface	99.9	14 & 15
	80-100	M. pyogenes phage	Spiked DW	99-99.8	11
	25	"	Ohio River	84-95	12
	15	"	Ohio River	86	12
$FeCl_3$	66	Polio 1	Spiked DW	99.7-99.999	8
	60	"	"	48-99.7	9
	20	Coxsackie A2	"	97	11
	40	"	"	98	11
	15	"	Ohio River	95	12
	50-60	MS2	NS	99.5	17
	50	f2	Natural lake	99.4	14 & 15
	40	"	Natural surface	99.1	16
	20-40	M. pyogenes phage	Spiked DW	99.3-99.9	11
$Fe_2(SO_4)_3$	40	Polio 1, 2, 3	Polluted river	99.8	10
	50	f2	Natural lake	92	14 & 15
	62	"	Natural surface	99.91	16
	40	T4	Polluted river	99.6	10

* Data extracted from Tables 1 & 2

DW - Distilled water
NS - Not stated

condition of the water. The data of Chang and co-workers (11) (12) show that dosages of only 25 and 15 mg/l for $Al_2(SO_4)_3$ and $FeCl_3$ respectively were required to obtain optimum removals of the Coxsackie A2. Dosages up to 100 mg/l for $Al_2(SO_4)_3$ and 40 mg/l for $FeCl_3$ were required for the spiked distilled water. Similar results can also be seen for $Al_2(SO_4)_3$ coagulation of the poliovirus in the various waters which were used.

These data support the conclusion that either aluminum sulfate or ferric salts will provide equal capability for virus removal. An "optimum" dosage for virus removal cannot be deduced from the data. The required dosage appears related to the characteristics of the suspending water.

Process control

In practice, dosages of coagulants are normally adjusted to minimize the turbidity leaving the sedimentation tank. Virus removals are normally thought to follow turbidity removal and process control for viruses is obtained by monitoring turbidity. Several investigators have presented evidence to show that increased turbidity removals are accompanied by increased virus removals (8) (13) (14) (15) (17). Chaudhuri and Engelbrecht (13) showed a marked parallelism between turbidity and T4 and MS2 phage removals using $Al_2(SO_4)_3$ as the coagulant. Manwaring et al. (17) observed similar results with coagulation of the MS2 phage with $FeCl_3$. Foliguet and Michelet (8) have also mentioned that high virus reductions were made in parallel with high turbidity reductions.

A close examination of certain of these data, however, shows that maximum turbidity removal may occur before the maximum virus removal is reached. Chaudhuri and Engelbrecht (13) showed that the maximum T4 bacteriophage removal lagged the maximum turbidity removal at pH 5.24 by about 5 mg/l of $Al_2(SO_4)_3$. This was not noted, however, at pH values of 6.17, 7.00 and 8.30. Shelton and Drewry (16) noted a decrease from 98 to 95 percent in the $FeCl_3$ dosage required for maximum virus removal compared to the dosage where the maximum turbidity removal was first reached.

York (14) and York and Drewry (15) have also shown that their virus reductions generally coincided with the turbidity reductions. York (14) did note, however, that the optimum coagulant dose for maximum turbidity removal was slightly lower than that reported for maximum virus removal. Portions of these data have been reproduced in Table 4. The $FeCl_3$ data showed a rather marked variation from the other coagulants with only 58 percent removal of turbidity when the maximum virus removal occurred. These data are confounded by the very low turbidity level of 0.38 TU in the raw water. Certain of his $FeCl_3$ coagulation data showed turbidity increases after coagulation and sedimentation. Difficulties in obtaining satisfactory settling of floc particles to yield reproducible data in these turbidity ranges are well established. This anomoly in his data may be safely discounted.

TABLE 4 DOSAGES FOR MAXIMUM REMOVALS OF f2 PHAGE AND TURBIDITY FROM NATURAL LAKE WATER*

Coagulant	Initial Turb.	Virus			Turbidity	
		Dosage	Max Virus Removal	Turb. Removal at indicated dosage	Dosage	Max Turb. Removal
	T.U.	mg/l	%	%	mg/l	%
$Al_2(SO_4)_3$	2.3 & 3.6	25	99.9	91-95	21	96
$FeCl_3$	0.38	50	99.4	58	23	92
$Fe_2(SO_4)_3$	2.3	50	92	92	49	89
$FeSO_4$	1.1 & 1.9	36	94	+	-	+

* From York (14)

+ Minor removals or increases of turbidity

The available data support the general conclusion that maximum virus removal will occur at or near the maximim turbidity removal. The evidence indicates that only minor reduction will occur if the coagulant dosage is adjusted to that for maximum turbidity removal.

Water Conditions

The water conditions of general concern in virus coagulation appear to be those of pH, temperature and organic and inorganic chemical content.

pH--

Chang et al. (11) coagulated Coxsackie A2 in spiked distilled water with 80 ppm of $Al_2(SO_4)_3$ at 25°C and at final pHs of 5.5, 6.2 and 7.2. They obtained removals of 95, 97 and 99.0 percent respectively. These authors made a companion study with M. pyogenes phage at final pHs of 5.5, 6.2, 7.2 and 8.2. The respective removals were 85, 99.1, 97 and 80 percent. They concluded that the optimum pH range was 6.2 to 7.2.

Chaudhuri and Engelbrecht (13) coagulating MS2 and T4 phages with $Al_2(SO_4)_3$ at dosages from 20 to 100 mg/l at pHs of 5.24, 6.17, 7.00 and 8.30 found decreased removals as the pH was increased. They concluded that the optimum pH for coagulation of these viruses was 5.2. Using the MS2 phage with $FeCl_3$ at dosages from 20 to 100 mg/l with pHs of 5.0, 6.1, 7.0 and 8.0 Manwaring et al. (17) found very similar results to those of Chaudhuri and Engelbrecht. These investigators concluded that the optimum pH was 5.0 for coagulation of the MS2 with $FeCl_3$.

Chang et al. (11) have pointed out the importance of distinguishing the type of floc which is formed in the coagulation process in a virus study. They showed that the optimum pH for the removal of viruses in a system where aluminum phosphate is the precipitate is lower than where aluminum hydroxide is formed. They found that the optimum pH was 5.2 under conditions where $AlPO_4$ was formed when the pH varied over the range from 5.2 to 7.2. Their report is unclear on which virus was used in this study. On the other hand Brunner and Sproul (20) found an optimum pH of 6.4 in the coagulation of poliovirus 1 with $Al_2(SO_4)_3$ in a distilled water system when the Al:P mole ratio was 1:1. They investigated removals at pH values of 5.1, 6.4 and 7.3. Chang et al. (1) reported on the removal of M. pyogenes phage with 60 ppm of $Al_2(SO_4)_3$ in a phosphate buffered distilled water system. They reported that the removal decreased from 98 to 14 percent as the pH was increased from 5.2 to 8.2. The floc formation was also reported to decrease from fairly good to very poor over this pH range indicating that the precipitate formed was actually $AlPO_4$.

The available data are insufficient to draw conclusions on the optimum pH for enteric virus removal. It does appear, however, that the optimum pH for virus removal where aluminum phosphate is formed may be lower than where aluminum hydroxide is precipitated. The importance and usefulness of precise information on the optimum pH for virus coagulation is debateable for most situations. It would appear that in nearly every case coagulation will be optimized for removal of turbidity. Adjustment of pH to a value which might provide better virus removal will not receive high priority.

In cases where the raw water is known to have high virus concentrations control of pH to optimize their removal would be desirable and a laboratory investigation using the local water would be warranted.

Temperature --

Data on temperature effects on virus removal during coagulation are very few. The only data available appear to be those of Chang et al. (12) shown in Tables 1 and 2 for the coagulation of Coxsackie A2 and M. pyogenes phage with $Al_2(SO_4)_3$. These investigators noted a decrease in the removal of the Coxsackie A2 from 99 to 96 percent as the temperature decreased from 25° to 5°C. The phage removal decreased from 95 to 84 percent for the same temperature change. These authors did not consider the reduced removal of the Coxsackie virus to be significant. Reasons for the reduced removal of the bacterial virus were not given.

The importance of water temperature on the removal of viruses by coagulation cannot be determined with the available evidence.

Organic Content--

Chang et al. (12) reported that 20 ppm of gum arabic reduced the removal of Coxsackie A2 from 97 to 17 percent when 15 ppm of $Al_2(SO_4)_3$ was used. The removal of M. pyogenes phage was reduced from 92 to 0 percent. The gum seriously interfered with coagulation since no floc was produced. Manwaring et al. (17) reported a reduction in the removal of MS2 phage from 99 to 67 percent when 200 ml/l of wastewater effluent was added to a clean suspending medium when 60 mg/l of $FeCl_3$ was used. This was accompanied by a parallel decrease in the turbidity removal. No decrease was observed when up to 50 mg/l of bovine serum albumin was added. Chaudhuri and Engelbrecht (13) reported reduction from 99.8 to 94 percent in MS2 removal with 200 ml/L of wastewater effluent using 50 mg/l of $Al_2(SO_4)_3$. Parallel reductions in turbidity removal were also noted. Reductions of only 2 to 3 percent were noted in the coagulation of T4 with 50 mg/l of $Al_2(SO_4)_3$ in the presence of 50 mg/l of egg and bovine serum albumin, and 200 ml/l of wastewater effluent. Data in Table 1 from Wolf et al. (4) reported a 99.4 percent removal of poliovirus 1 in activated sludge effluent with a coagulant dosage to yield a 7:1 ratio of Al:P. The Pomona virus study (7) reported an average removal of 95 percent for poliovirus 1 when their dosage of $Al_2(SO_4)_3$ produced an Al:P ratio of 1.7:1. Foliguet et al. (21) also found that organic matter reduced poliovirus 1 removal during coagulation with $FeCl_3$.

The evidence supports the conclusion that virus removals may be reduced slightly from their highest values when organics are present, but coagulation is otherwise satisfactory. The data also show that chemical dosage modifications may be necessary in a plant to respond to raw water organic content changes.

Inorganic Material --

Chang et al. (12) speculated that the presence of calcium (24-54 mg/l) and magnesium (6-14 mg/l) may have slowed down the formation of the aluminum-M. pyogenes phage complex and thus reduced the removal of this virus over that obtained in a water with very low concentrations of these cations. These data are shown in Table 2 with the Ohio River water sample containing the higher concentrations of calcium and magnesium and the spiked distilled water with the low concentrations. A comparison of their data in Table 1 for the Coxsackie A2 virus for these waters does not show this effect. Manwaring et al. (17) found minor reduction in the removal of MS2 phage with $FeCl_3$ when calcium and magnesium present together were varied from 0-50 mg/l each. Chaudhuri and Engelbrecht (13) reported no reduction in the removal of T4 phage with $Al_2(SO_4)_3$ when the calcium and magnesium concentrations were changed in similar fashion.

Thorup et al. (5) obtained the data shown below in the coagulation of poliovirus 1 with 40 mg/l of clay using 10 mg/l of Al_2SO_4 or $Fe_2(SO_4)_3$ at pH 6.8 in distilled water with the indicated concentrations of $CaCl_2$:

Virus and Turbidity Removal

Ca(from $CaCl_2$)	Percent			
	$Al_2(SO_4)_3$		$Fe_2(SO_4)_3$	
mg/l	Virus	Turbidity	Virus	Turbidity
0	11	0	0	0
4	26	18	0	0
40	83	93	4	0
400	86	96	83	93
4000	79	94	85	93

Their data do not show significant variations in the removal of the poliovirus when acceptable turbidity removals were reached with calcium concentrations of 40, 400 and 4,000 mg/l.

The available data do not indicate a significant effect on the removals of viruses by coagulation when the inorganic concentration of the suspending water is varied.

Process Reliability

The reliability of the coagulation process using metallic salts cannot be judged by comparing removals obtained under laboratory and pilot scale conditions. Data from actual plant operations are required. However, there are no reports in the open literature on full scale conditions. Tables 1 and 2 cite six reports obtained from four different pilot plants (4) (7) (8) (9) (10). These plants were operated for removal of poliovirus 1 (except for Guy et al. [10] who used polio 1, 2 and 3) under a variety of conditions

using activated sludge effluent and spiked demineralized water. The removals obtained were consistently very high except for two reported instances. In these two cases Wolf et al. (4) obtained only 63 percent removal of poliovirus under known poor coagulation conditions. Foliguet and Doncoeur (9) attributed the 48 percent poliovirus removal in one of their runs to low levels of initial turbidity in the influent water to the pilot plant. With the exception of these two cases the removals in the four pilot plants ranged from 95 to 99.999 percent and are at least as good, and in most cases, better than the removals obtained from laboratory reports. It is also to be noted that these pilot plant reports were obtained by various investigators throughout the world. This gives increased evidence to the reliability of the process.

The coagulation process appears to be one which when satisfactorily operated will produce virus removals of up to 99.99 percent or more. However, this conclusion has not been confirmed by data from a full scale water treatment plant.

POLYELECTROLYTES

The reported data on the utilization of polyelectrolytes as primary coagulants have been summarized in Table 5. Thorup et al. (5) showed that the anionic and nonionic polyelectrolytes gave poor removals. They found no removals in distilled water but removals up to 40 percent with salt concentrations up to 126 mg/l of Na, K, Ca and Mg. None to poor or fair floc formation was observed. Chaudhuri and Engelbrecht (13) did not observe removal or inactivation of MS2 or T4 phages with anionics in concentrations of 1 and 1 to 5 mg/l respectively in deionized water.

Chaudhuri and Engelbrecht (13) observed a T4 and MS2 phage inactivation of about 80 percent with the cationics Primafloc C-7 and Catfloc in deionized water. They used the polyelectrolytes in concentrations of 1 mg/l except the Catfloc and T4 when 0.5 to 10 mg/l was used. They speculated that the "inactivation" may have been caused by a virus-host absorption interference such that one host bacterium cell was infected by more than one phage. Under such conditions the inactivation would not be real.

The data in Table 5 for cationic polyelectrolytes indicate limited removals of enteric viruses in concentrations of polyelectrolytes from 1 to 2 mg/l. Thorup et al. (5) and Cliver (22) reported minor removal of poliovirus 1 and Coxsackie B3 with Catfloc. Poor floc formation was noted under these conditions. High concentrations of cationics were reported to give removals of phages as high as 99.9 percent. Concentrations of the cationic materials to attain these removals for the MS2 and T4 were as high as 12.5 mg/l.

Polyelectrolytes have been used as coagulant aids with mixed success (5) (13) (14) (15) (21). Thorup et al. (5) found little improvement in T2 phage removal when 1 mg/l of an anionic and nonionic polyelectrolyte were used in a water which had been deliberately undercoagulated with 5 mg/l of $Al_2(SO_4)_3$. Under similar conditions they found that 1 mg/l of a cationic raised the T2 virus removal from 57 percent to 94 percent. They noticed little improvement

TABLE 5 VIRUS REMOVAL WITH POLYELECTROLYTES AS PRIMARY COAGULANT

Virus		Coagulant		Conditions of Coagulation					Removal				
Type	Initial Conc. PFU/ml	Type	Dose mg/l or ppm	Type Water	Turbidity	Temp °C	pH Start	pH End	Type Study	Virus	Turbidity	Ref.	Remarks
Polio 1(Sabin)	$3\text{-}7\times10^{4}$	Cationic Purifloc C32	1	Spiked D.W.	None	NS	6.8	6.8	L	0-36	-	5	
Polio 1(Chat)	NS	Cationic Catfloc	0.5-2.0	Ohio River	NS	NS	4.7	NS	L	0-40	NS	22	Poor to no floc
Coxsackie B3	NS	Cationic Catfloc	"	Ohio River	NS	NS	4.7	NS	L	0	NS	22	Poor to no floc
T2	$10^{6}\text{-}10^{7}$	Anionic Hercules CMC	1	Spiked DW	5 mg/l clay 5 mg/l infusorial earth	NS	7.0	7.0	L	0-41	NS	5	No or poor floc
T2	$10^{6}\text{-}10^{7}$	Nonionic Nacolyte 605	1	Spiked DW	"	NS	7.0	7.0	L	0-37	NS	5	No or poor floc
T2	$10^{6}\text{-}10^{7}$	Cationic Nacolyte 110	1	Spiked DW	"	NS	7.0	7.0	L	32-96	NS	5	Poor to good floc
f2	1×10^{6}	Cationic Drewfloc 21	1.5	Natural Lake	3.1	26	8.2	8.6	L	77	35	14,15	Fair Floc
f2	1×10^{6}	Cationic Catfloc	1-3.5	Natural Lake	5.3	19-24	8.2	8.2	L	99	40-60	14,15	Fair Floc
f2	NS	Cationic Catfloc	2.2	Natural Surface	1.1-1.2	NS	NS	7.5	L	92	Poor	16	Poor Floc
MS2	2.8×10^{5}	Cationic Primafloc C7	7.5	NS	12.5	24-25	5.9-6.0	5.9-6.0	L	99.1	98	13	
MS2	2.8×10^{5}	Cationic Catfloc	10	NS	12.5	24-25	5.9-6.0	5.9-6.0	L	99.6	97	13	
T4	5×10^{5}	Cationic Primafloc C7	7.5	NS	12.5	24-25	5.2-5.5	5.2-5.5	L	99.9	99	13	
T4	5×10^{5}	Cationic Catfloc	12.5	NS	12.5	24-25	5.2-5.5	5.2-5.5	L	99.5	99.1	13	

NS - Not stated

DW - Distilled water

L - Laboratory

in removal of T2 or poliovirus 1 with these materials when used in situations where the coagulation with $Al_2(SO_4)_3$ and $Fe_2(SO_4)_3$ was otherwise acceptable. Foliguet et al. (21) found that anionics and nonionics used as aids reduced the removal of poliovirus 1 coagulated with 60 mg/l of $FeCl_3$ from 99.7 percent to 92 percent with 0.25 mg/l of an anionic and to 89 and 95 percent with 0.25 and 1.0 mg/l of a nonionic. A cationic material did not change the removal obtained with $FeCl_3$ alone. Chaudhuri and Engelbrecht (13) did note an increase in T4 phage removal from 98 percent to 99.9 percent when about 1 mg/l of cationics were added. Little effect was noticed in a similar study with MS2 phage. York (14) and York and Drewry (15) observed little change in removal of the f2 phage when nonionics, anionics and cationics were used as coagulant aids. They did observe that utilization of a cationic might reduce the optimum dosage of $Al_2(SO_4)_3$ for virus removal. Others have made similar observations (16) (21). York and Drewry also noted that the effective coagulant dosage range for f2 removal was broadened with their anionic polyelectrolyte.

Utilization of polyelectrolytes in the usual dosage range of 1-2 mg/l does not appear to give significant virus removal when used as primary coagulants. Virus removals are not significantly increased when these materials are used in waters which are otherwise coagulated satisfactorily.

MECHANISM OF REMOVAL BY COAGULATION

Chang et al. (1) postulated that in the first stage of the flocculation process with alum the aluminum ions form an aluminum-virus precipitate. Under the "right" conditions the aluminum-virus precipitates are then incorporated or aggregated into the floc particles. They deduced the initial reaction from knowledge of aluminum-protein chemistry and surmized that the aluminum-virus complex was actually a salt of aluminum with a protein in the virus capsid. As mentioned above Chang et al. (12) found that gum arabic, a compound known to interfere with coagulation by coating the charged particle, interfered with coagulation and concluded that the formation of the virus-aluminum complex had been interrupted. The exact meaning of their data is in doubt because the gum arabic prevented the formation of any floc particles thereby preventing any possible incorporation into floc particles since they were not formed.

Chaudhuri and Engelbrecht (23) examined the initial aluminum-virus complex formation in the T4 bacteriophage. They determined the amount of aluminum which reacted with this virus at pH 5, 6 and 9 by exposing the virus to a known low concentration of aluminum, and measuring the residual aluminum after removal of the virus. They determined that 7,370, 6,200 and 6,300 atoms of aluminum were absorbed by the T4 phage at pH 5.0, 6.0 and 9.0 respectively. They stated without evidence that a satisfactory mass balance on the aluminum distribution was obtained. At pH 5.0 the smaller MS2 absorbed 4,600 atoms per particle. This initial binding reaction was found to occur in less than 30-40 seconds, the minimum resolution time in their system. They also estimated that the head of the T4 had 7,120 carboxyl groups, 1,980 hydroxyl groups and 6,930 ammonium and guanidinium groups. The match of the carboxyl groups with the number of absorbed aluminum atoms and the known involvement of these sites in binding aluminum

in casein and gelatin led them to conclude that this group was the most probable binding site on the virus.

The observation by Chang et al. (12) that gum arabic interfered with coagulation might be explained with the observations of Chaudhuri and Engelbrecht (23). Gum arabic has a high density of carboxyl groups which would act as competitive binding sites for the aluminum.

Cookson (24) has discussed the absorption of viruses to metal hydrolysis species. He pointed out that adsorption may occur through coordinated hydroxyl groups, covalent bonds between the trivalent atom and the virus, and from electrostatic forces. He points out that these mechanisms are dependent upon surface protein chemistry of the virus and the metal hydroxides. Aluminum or iron phosphate with many fewer hydroxyl groups on its surface would not be as active in virus removal as the metal hydroxide. Since different viruses generally have similar protein surface chemistry the conclusion that coagulation and removal of viruses is not specific for each virus is supported.

The mechanism by which polyelectrolytes coagulate and remove viruses appears less well understood. That the electrostatic forces are important is obvious from the very poor removals obtained from the anionic and nonionic polyelectrolytes (5) (13). These materials are negatively charged (anionic) or uncharged (nonionic) and offer little attraction for the virus which is negatively charged under most water pH conditions. Addition of cations in the suspending medium with these materials gave some removal. Whether this mechanism is one of the charge reduction permitting electrostatic attraction to occur or bridging of the cation which had been absorbed to either the virus or polyelectrolyte is unknown.

Adsorption of viruses to cationic polyelectrolytes occurs much more readily. Cookson, in discussing adsorption to the cationic polyelectrolyte PE 60, stated that adsorption to this material would occur at carboxyl or carboxylate salt groups through hydrogen bonding and by electrostatic attraction forces.

SECTION 4

VIRUS REMOVAL AND INACTIVATION BY pH AND LIME FLOCCULATION

BACKGROUND

Viruses are affected by changes in the pH of the suspending water and by any precipitates that may be formed during these pH changes. It is important to note that these effects may proceed simultaneously as the pH is changed. Viruses are inactivated or removed in some fashion as the pH is changed to that outside their stability range. This effect is time dependent. When precipitates are also formed the virus may absorb to the surfaces and be removed with the precipitate when it is removed from the water by sedimentation. These two effects will be considered separately in this review.

A critical review of the literature on removal and inactivation of viruses by pH and lime flocculation should address the following questions:

(1) Reliability of process. Reliability can be examined by considering the following:
   (a) Plant scale data versus laboratory or pilot plant data.
   (b) Reproducibility of data by different investigators.

(2) Delineation of factors affecting the process. It would appear that at least the following factors would be significant: (1) organic and inorganic content; (2) agglomeration of viruses as a loss of infectivity mechanism versus true inactivation; (3) pH level; (4) type of precipitate formed; (5) contact time; and (6) susceptibility of different viruses to the process.

(3) Process control. The utilization of pH, level of solids production and/or other mechanisms should be considered.

(4) Determination of the mechanism of virus removal.

pH EFFECTS

Low pH

Viruses tend to be more stable at pH values around neutrality. The enteric viruses which are of interest in water transmission also tend to be very acid stable. Salo and Cliver (25) showed that 1 log per week of Coxsackie A9 and poliovirus 1 was lost at pH 3.0 at 2°C. This inactivation increased to 1 log per 24 hours at pH 3.5 at 30°C. They also found that echovirus 7 underwent a 2 log loss in 48 hours over the pH range from 3 to 5 at 30°C. Robinson (26) found that several Coxsackie A and B strains survived for one day over the pH range from 2.3 to 9.4 at room temperature. It was reported by Ginsberg (27) that adenoviruses 1 and 2 lost 1 log of

titer in 30 minutes at 22-23°C at pH 3. He also found that adenovirus 3 lost between 2 and 3 logs under similar conditions. Using adenovirus 5 Fields and Metcalf (28) found a 30 percent loss of titer at pH 3.5 in 2 minutes. No further loss was noted in 180 minutes.

The lowest pH reached in water treatment is probably not lower than 4 to 5. The resistance of enteric viruses to inactivation at this pH range precludes consideration of low pH as a potential inactivation process.

High pH

Data on virus inactivation are presented in Table 6. Sproul et al. (29) Berg et al. (31), and Boardman (30) showed that little inactivation of poliovirus 1 was obtained at pH values up to about 10.5 at 20-25°C for exposure times up to 100 minutes. Sproul (29) and Berg et al. (31) found a poliovirus 1 loss of infectivity of 99 percent or more over the pH range from 11 to 12 with contact times of 90 to 100 minutes. Seemingly lower poliovirus 1 loss was noted by Sproul (32) using NaOH for pH adjustment in distilled water although he did show a 94 percent loss at pH 11.9. The pH at which significant loss of infectivity occurs for viruses other than poliovirus 1 cannot be readily determined. Ginsberg (27) found that adenovirus 1, 2, 3 and 4 were stable up to a pH of about 9. Adenovirus 5 was totally inactivated at a pH of 10.5 in 10 minutes. Robinson (26) found that several strains of Coxsackie A and B were stable for one day at pH 9.4.

The data in Table 6 for poliovirus 1 indicate that the loss of infectivity increases as the pH increases and as the contact time increases. The maximum inactivation observed was 99.8 percent in 5 minutes at 22-23°C. Echovirus 7 appears to behave similarly to poliovirus 1 although the data are very limited. The data on the adenovirus appear to indicate that these viruses may be more sensitive to alkaline pH values than either poliovirus 1 or echovirus 7.

Salo and Cliver (25) noted that the type of buffer system used and the salt content (in the molar concentration range) influenced the rate of inactivation by pH over the range from 3 to 9. Sproul et al. (29) noted that in their distilled water systems the cation associated with the hydroxide affected the loss of infectivity of the poliovirus 1. Loss of infectivity started at pH 10.5 when NaOH or $Ca(OH)_2$ was used but with KOH a pH of 11.5 was required. Berg and Berman (33) found that the time for 99 percent inactivation of echovirus 7 was increased from 1.3 minutes at pH 11.92 in BOD dilution water to 25.6 minutes at pH 11.87 in a 5 percent peptone solution. Inactivation comparisons at other pH values were also given.

Donovan (36) presented an interesting study on the loss of infectivity of f2 and T2 phages in settled activated sludge effluent under high pH conditions. His data shown in Table 7 indicate that the f2 virus was very sensitive to the pH of the system with virus reductions of 99.99 percent noted at a pH of 11.0 and above. These and other data which he reported tend to show that the virus reduction was temperature dependent when the pH was held constant since lower reductions were observed at 5°C than 22-27°C at pH 11.5.

TABLE 6 VIRUS REMOVAL AT HIGH pH WITHOUT PRECIPITATION

Virus Type	Initial Conc. PFU/ml	Temp °C	pH	Buffer or pH Source	Suspending Water	Contact Time Minutes	Type Study P,L,F	Removal or Inactivation %	Ref.
Polio 1 (Sabin)	$1\text{-}5x10^4$	21-22	11.9	$Ca(OH)_2$ or NaOH	DW	90	L	90	29
Polio 1 (Sabin)	$1\text{-}5x10^4$	21-22	12.2	KOH	DW	90	L	90	29
Polio 1 (Sabin)	NS	22.5	10.1	Na_2CO_3	DW	10	L	2	30
Polio 1 (LSc)	NS	25	10.1	$Ca(OH)_2$	Previously lime flocced sec. effl.	100	L	~0	31
Polio 1 (LSc)	NS	25	10.8	$Ca(OH)_2$	"	100	L	58	31
Polio 1 (LSc)	NS	25	11.1	$Ca(OH)_2$	"	100	L	99	31
Polio 1 (Mahoney)	$2.5x10^5$	22-23	11.5	NaOH	DW	30	L	7	32
Polio 1 (Mahoney)	$1.3x10^5$	22-23	11.9	NaOH	DW	30	L	59	32
Polio 1 (Mahoney)	$2.4x10^5$	22-23	12.1	NaOH	DW	20	L	94	32
Polio 1 (Mahoney)	$3.3x10^5$	22-23	12.5	NaOH	DW	5	L	99.8	32
Adeno 1	NS	22-23	11.2	NaOH	Organic medium	30	L	>99.9	27
Adeno 2	NS	22-23	11.1	NaOH	Organic medium	30	L	>99.9	27
Adeno 3	NS	22-23	10.2	NaOH	Organic medium	30	L	>99.9	27
Adeno 4	NS	22-23	11.0	NaOH	Organic medium	30	L	>99.9	27
Adeno 5	NS	NS	10.5	Glycine	NS	2	L	69	28
Adeno 5	NS	NS	10.5	Glycine	NS	5	L	88	28
Adeno 5	NS	NS	10.5	Glycine	NS	10	L	Total inact.	28
Echovirus 7	NS	25	11.23	NaOH	BOD dilution water	7	L	99.98	33
Echovirus 7	NS	25	11.49	"	"	4	L	99.99	33
Echovirus 7	NS	25	11.79	"	"	2	L	99.5	33
Echovirus 7	NS	25	11.92	"	"	1.5	L	99.9	33
T2	106	Room	10.8	$Ca(OH)_2$	DW	90	L	99.9^+	34
T7	$2\text{-}4x10^5$	22.5	10.1	Na_2CO_3	DW	10	L	42	35

NS - Not stated

DW - Distilled water

L - Laboratory

Table 7 LOSS OF INFECTIVITY OF f2 IN FLOC-FREE $Ca(OH)_2$ TREATED ACTIVATED SLUDGE EFFLUENT IN SIXTY MINUTES*

pH	Temperature °C Loss of Infectivity % 10°C	22°-27°C
7.0	48	58
8.0	30	56
9.0	61	78
10.0	56	98
11.0	99.99	~>99.9991
11.2	-	99.9999
12.0	>99.9999	>99.9999

*From Donovan (36)

When Donovan added EDTA to chelate the divalent cations in the pH adjusted effluent the f2 inactivation was four logs less than when it was not present. A similar test with $Ca(OH)_2$ in distilled water at pH 11.5 had less than one log of reduction in 5 minutes in the sample to which the EDTA had been added versus over 5 logs in less than 1 minute in the sample without EDTA. Studies in which NaOH, KOH, $Ba(OH)_2$ and Theorell-Stenhagen buffer were used to raise the pH to 11.5 showed that the valence of the cation, except for calcium, had no apparent effect on the inactivation. He concluded that the f2 virus clumped in the presence of the calcium and that a significant part of the "inactivation" was actually a loss of infectivity since a large portion of the viruses were no longer present as discrete particles. He also presented electron micrographs showing the aggregates of viruses in the calcium treated sample.

The f2 virus concentrations used by Donovan were evidently on the order of 10^6 to 10^{10} PFU/ml. The opportunity for collisions to permit aggregation of particles of similar diameters is proportional to the square of the number of particles. In Donovan's work the opportunity for collisions and possible aggregation of viruses was much larger than there would be in a field situation. In a biologically treated effluent or a contaminated surface water the opportunity for aggregation would be greatly reduced since virus concentrations in these cases would, at most, be on the order of 10^0 to 10^2 PFU/ml. The aggregation effect noted by Donovan is probably not important in the real world, or at least is of minor importance.

If the animal viruses are accurately modelled by the f2 bacteriophage, however, then a portion of the "inactivation" under high pH conditions may actually be only an aggregation of discrete particles. The actual inactivation or removal will be less than is otherwise thought. Additionally, the inactivation of viruses in a clumped state is more difficult than inactivation of discrete virus particles.

The few available data on virus inactivation by high pH are presently inadequate to determine the exact conditions under which this process will give high inactivations. It is readily obvious that viruses do undergo a loss of infectivity when a certain pH, which appears to be virus specific, is reached. The amount of loss to be attributed to aggregation, or true inactivation has not been determined. The salt content, particularly calcium, may be very significant. The influence of temperature, contact time, organic content, though significant, cannot be deduced from the data. The necessary pH for inactivation of a wide spectrum of viruses under any set of conditions is unknown.

LIME PRECIPITATION

The addition of $Ca(OH)_2$ to water will form various precipitates depending on the water characteristics. The precipitates of major importance are $CaCO_3$, $Mg(OH)_2$, and calcium hydroxylapatite, $Ca(OH)_2(PO_4)_6$, or some other precipitate of phosphate. The $CaCO_3$ and $Mg(OH)_2$ are formed in the softening of drinking water by the precipitation process. Calcium hydroxylapatite is formed during phosphate removal with $Ca(OH)_2$. $CaCO_3$ and

$Mg(OH)_2$ would also be formed in this latter reaction depending upon the concentrations of Ca and Mg.

Laboratory Evaluation

Virus removal and inactivation data under high pH conditions with the formation of a precipitate are presented in Table 8. In a laboratory water softening study using poliovirus 1 Wentworth et al. (37) found a maximum removal of 81 percent with 726 mg/l of $CaCO_3$, and 99.92 percent when 486 mg/l of $Mg(OH)_2$ as $CaCO_3$ were precipitated. Increased removals of the virus were noted as the pH increased although the virus removal with $CaCO_3$ was clearly more dependent upon the amount of precipitate formed. They also found that poliovirus removals were in excess of 99.9 percent at all tested levels when both $CaCO_3$ and $Mg(OH)_2$ were formed together. They attributed all of the virus removal to the incorporation of the virus into the precipitate and its removal by settling with the precipitate since their work was done at pH levels of 11.2 or below, a point where their poliovirus was not affected by pH alone. Their work with $CaCO_3$ was confirmed by that of Boardman (30). In a laboratory study Brunner and Sproul (20) were able to remove up to 99.1 percent of poliovirus 1 during the formation of calcium hydroxylapatite at a pH of 11.0 using a defined chemical medium. Using an activated sludge effluent they obtained a removal of 94 percent with a pH of 10.9. Berg et al. (31) using $Ca(OH)_2$ flocculation of a secondary effluent with a dosage of 200 to 500 mg/l of $Ca(OH)_2$ which gave pH values up to 11.0 obtained slightly higher poliovirus 1 removals, up to 99.9 percent. Removals of 92 percent were obtained at a pH of 9.3 using 200 mg/l of $Ca(OH)_2$.

These laboratory data indicate that poliovirus 1 removals in excess of 90 to 99 percent are possible with increased removals associated with increased amounts of precipitate. It would appear from the pH removal data without precipitate formation that a portion of the removals obtained in these studies at pH values of 11 or more was due to the pH effect and not by incorporation into the precipitate. Larger removals were associated with the precipitation of $Mg(OH)_2$ and calcium hydroxylapatite rather than the $CaCO_3$. Thayer and Sproul (34) felt that the negative charge of the $CaCO_3$ precluded high removals with this material. The presence of the organics in the treated effluents used in several studies did not appear to interfere with the poliovirus removal when compared to that obtained in a clean water system. Sattar et al. (39) using $Ca(OH)_2$ at pH 11.5 obtained high poliovirus 1 removals in the presence of the organics in raw sewage. In the Berg and Berman (33) study with echovirus 7, however, the very high concentration of organics (5 percent peptone) did interfere with the virus removal when no precipitates were formed. These data would indicate that organics may interfere in this process when present in sufficient concentration.

Insufficient data have been reported to determine whether inorganic concentrations affected the virus removal. Additionally data are not available from these studies to judge whether a portion of the observed removals can be ascribed to aggregation associated with calcium (or other ions) as discussed by Donovan (36).

TABLE 8 VIRUS REMOVAL AT HIGH pH WITH PRECIPITATION

Virus Type	Initial Conc. PFU/ml	Temp °C	pH Start	pH End	pH Source	Suspending Water	Contact Time Minutes	Type Study L,P,F	Removal or Inactivation %	Ref.	Remarks
Polio 1(Sabin)	$2.5\text{-}7.5\times10^4$	Room	NS	8.0-9.0	$Ca(OH)_2$	DW w/$Ca(HCO_3)_2$	91	L	9 → 81	37	181 → 725 mg/l $CaCO_3$ ppt only
Polio 1(Sabin)	"	"	"	10.3-10.6	NaOH	DW w/$MgCl_2$	91	L	99.3 → 99.92	37	56 → 486 mg/l $Mg(OH)_2$ as $CaCO_3$
Polio 1(Sabin)	"	"	"	10.8-11.2	$Ca(OH)_2$ + Na_2CO_3	DW w/$CaCl_2$ + $MgCl_2$	91	L	99.9 → 99.993	37	Excess lime soda ash softening w/initial hardness 200 → 500 mg/l as $CaCO_3$
Polio 1(Sabin)	NS	22.5	10.1	10.1	Na_2CO_3	DW w/$CaCl_2$	10	L	84	30	ppt of 300 mg/l $CaCO_3$
Polio 1(Vaccine)	8×10^4	19	NS	11.7-11.9	$Ca(OH)_2$ + $FeCl_3$ (54 mg/l)	Act.sl.effl.	~120	P	Total inact. (none detected)	4	
Polio 1(Vaccine)	4×10^4	18	NS	11.0-11.3	$Ca(OH)_2$ + $FeCl_3$ (27 mg/l)	Act.sl.effl.	~120	P	Total inact. (none detected)	4	
Polio 1(LSc)	$3.3\text{-}5.5\times10^4$	NS	NS	9.3 → 11.0	$Ca(OH)_2$	Sec.effl.	Between 75-90	L	92.3 → 99.9	31	Lime flocculation w/200-500 mg/l $Ca(OH)_2$
Naturally occurring enteric	0.027	NS	7.6	11.4	$Ca(OH)_2$ +~0.1 mg/l Dow A23	Trick. filter effl.	NS	F	97.7	38	
"	0.0045	"	"	"	"	"	"	"	99.88	38	
Polio 1(Sabin)	$3\text{-}5\times10^4$	18-25	NS	11.0	NaOH	DW w/$CaCl_2$ + $HPO_4^{=}$	91	L	97	20	26.5 mg/l $PO_4^{=}$ ppt
Polio 1(Sabin)	"	"	"	11.0	"	"	"	"	99.1	"	56.7 mg/l $PO_4^{=}$ ppt
Polio 1(Sabin)	6.2×10^4	18-25	NS	10.9	$Ca(OH)_2$	Act.sl.effl.	91	L	94	20	44 mg/l $PO_4^{=}$ ppt
Polio 1(Sabin)	$1.4\text{-}8.2\times10^4$	28	NS	11.5	$Ca(OH)_2$	Raw sewage	75	L	Total inact. (none detected)	39	8 trials
Polio 1(Sabin)	$2.0\text{-}5.0\times10^4$	4	NS	11.5	$Ca(OH)_2$	Raw sewage	75	L	Total inact. (none detected)	39	4 trials
Naturally occurring enteric + polio 1(Sabin) in some runs (filtrable)	0.21	~20	NS	9.7	$Ca(OH)_2$	Screened raw	144 to 174	P	92	40	
"	0.16	~25	NS	10.5	$Ca(OH)_2$	"	"	P	97	40	
"	0.25	~24	NS	11.5	$Ca(OH)_2$	"	"	P	99.6	40	

TABLE 8 (continued)

Virus Type	Initial Conc. PFU/ml	Temp °C	pH Start	pH End	pH Source	Suspending Water	Contact Time Minutes	Type Study L,P,F	Removal or Inactivation %	Ref.	Remarks
Naturally occurring enteric	0.70*	NS	NS	11.1±.2	$Ca(OH)_2$	Act.sl.effl.	50	F	Total inact. (none detected)	41	Coliphage infl. 43 PFU/ml Removal 99.95%
"	0.65*	NS	NS	10.5±.4	"	"	50	F	>99.98	41	Coliphage infl. 40 PFU/ml Removal 96%
"	0.60*	NS	NS	10.2±.1	"	"	50	F	Total inact. (none detected)	41	Coliphage infl. 31 PFU/ml Removal 78%
"	0.58*	NS	NS	9.6±.1	"	"	50	F	>99.98	41	Coliphage infl. 35 PFU/ml Removal 57%
f2	7.2×10^5	19	NS	11.7-11.9	$Ca(OH)_2$ + $FeCl_3$	Act.sl.effl.	~120	P	Total inact. (none detected)	4	54 mg/l $FeCl_3$ used
f2	2×10^4	18	NS	11.0-11.3	$Ca(OH)_2$ + $FeCl_3$	Act.sl.effl.	~120	P	Total inact. (none detected)	4	27 mg/l $FeCl_3$ used
T2	1×10^6	23-33	NS	8.1-9.2	$Ca(OH)_2$	DW + $Ca(HCO_3)_2$	91	L	25 → 57	34	Ppt of water w/ 100, 200 & 300 mg/l initial Ca as $CaCO_3$
T2	1×10^6	23-33	NS	10.5-10.6	NaOH	DW + $MgCl_2$	91	L	99.998-99.99992	34	Ppt of water w/ 100 → 500 mg/l initial Mg as $CaCO_3$
T2	1×10^6	23-33	NS	10.7-10.9	$Ca(OH)_2$ + Na_2CO_3	DW + $MgCl_2$, $CaCl_2$, $NaHCO_3$	91	L	99.95-99.993	34	Excess lime soda ash softening w/ initial hardness 300 → 500 mg/l as $CaCO_3$

* $TCID_{50}$/ml

NS - Not stated

DW - Distilled water

L - Laboratory

P - Pilot plant

F - Full scale

In general, contact times were not varied in these studies so no conclusions can be made on this parameter. It would be expected, however, that increased contact time at higher pH values would increase the observed inactivation from the pH effect based on Berg and Berman's data.

Pilot Plant and Full Scale Evaluation

Data from several pilot plant and full scale evaluations of virus removal under high pH precipitation conditions have been presented in Table 8. Wolf et al. (4), using $Ca(OH)_2$ flocculation of an activated sludge effluent, were not able to detect any of the 4-8 x 10^4 PFU/ml of influent poliovirus 1 in their settled effluent. It should be noted that $FeCl_3$ was used in their two tests. They also were not able to detect any of the 0.2 to 7.2 x 10^5 PFU/ml of f2 in the effluent under the same test conditions. Their pH values of 11.0 - 11.3 and 11.7 - 11.9 were well within the range when removals from the pH effect alone would be expected. Hiser (40), flocculating raw screened sewage with $Ca(OH)_2$, found increasing removals of filtrable viruses with increasing pH, reaching 99.6 percent at pH 11.5. His study showed that naturally occurring enteric veruses underwent the same percentage removal as did a spiked poliovirus 1. In this investigation the suspended solids were removed from the influent and effluent samples before concentration of the viruses for plaquing. Since the virus removal was a logarithmic function of the pH Hiser felt that the pH was the primary factor in the removal of the filtrable virus fraction.

Full scale test data from the Water Factory 21 investigations showed that 98 percent of the naturally occurring enteric viruses were removed based on the Buffalo Green Monkey (BGM) cell line and 99.9 percent on the Primary African Green Monkey cell line (38). These data were based on the influent and effluent virus titers occurring 50 percent of the time. The titer occurring 50 percent of the time was obtained from a probability distribution plot of the number of viruses in each of their samples taken over the entire sampling period. This report is of particular interest since it is from a 15 mgd plant over a 17 month sampling period. One hundred influent samples (some evidently were parallel samples) and 32 effluent samples were examined for virus content. The authors pointed out that only 5 percent of the 0.027 PFU/ml isolated in the influent on the BGM cell line could be confirmed upon passage in this cell line. In their study effluent samples were analyzed for viruses by two independent laboratories. One laboratory found only 3 positive samples while the other found 28 positives. Unfortunately none of these positives were confirmed. All of their percentage removal data are based on unconfirmed plaques, i.e. plaques which were not regrown in the same or another cell line to assure that the plaques had been caused by viruses and not by some extraneous effect. Their data also show that removals based on the virus titers equalling or exceeding that occurring 90 percent of the time were 99.3 and 96 percent based on the PAG and BGM cell lines respectively. The confirmed plaquing study done on the influent indicated 4 A and 5 B strains of Coxsackie, 10 strains of Echo, 3 strains of polio, 3 strains of Reo and 5 unknown plaques in 45 positive samples. Only 3 positive effluent samples were confirmed indicating 2 echo strains and polio 1. Unfortunately, while a rather broad spectrum of viruses were present in the influent insufficient effluent data were reported to draw

conclusions on whether all of the strains were equally susceptible to removal by this process.

Grabow et al. (41) in a full scale test (1.2 mgd) using an activated sludge effluent did not find any enteric viruses in the effluent when the pH was 11.1 ± 0.2. Removals in excess of 99.98 percent were obtained when the pH was as low as 9.6 ± 0.1. Parallel coliphage removal was 99.95 percent at pH 11.2 ± 0.2 decreasing to 57 percent at pH 9.6 ± 0.1. The 99.98 percent removal of enteric viruses at pH 9.6 ± 0.1 is surprisingly high and unexpected based upon the laboratory data. No information was presented on the amount of precipitate formed during the treatment.

The removals obtained in these reports indicate good reliability of the process under field operating conditions.

PROCESS CONTROL

The control of this process to maximize virus removal most clearly relates to the pH. The evidence shows that increasing pH causes increased inactivation. The point has been made above, however, that the pH necessary for inactivation of a wide spectrum of viruses under any set of conditions is unknown. Development of this information would permit pH to be used as a control parameter.

High removal of the suspended solids resulting from the precipitates which form in this process is also required. This phase of the process can be adequately monitored by suspended solids and turbidity measurements.

MECHANISM OF REMOVAL BY HIGH pH AND PRECIPITATES

Removal of viruses by high pH conditions is a two fold process in which irreversible inactivation occurs due to the pH effect and a physical removal by incorporation into the precipitates. It is well to point out at this point that Donovan (36) has shown that part of the so called inactivation of high concentrations of the f2 bacteriophage which may be caused by pH alone is really only a loss of infectivity caused by aggregation in the presence of calcium. Each aggregate then represents only one infectious unit rather than the number of infectious units that each virus in a deaggregated state would have. When these aggregates are subsequently exposed to a lower pH they may deaggregate and manifest themselves singly.

Poliovirus has been shown to be inactivated under alkaline conditions by a disruption of the capsid and a loss of the RNA to the water (42) (43) (44). Boeye and Van Elsen (42) suggested that the RNA when released from the capsid was either already degraded or that it was quickly degraded upon its release. Their work was done at pH 10, and at 30°C and higher temperatures. Information does not appear to be available to determine exactly how the proteins in the capsid are attacked to bring about this disruption. Extrapolation of this mechanism to enteric viruses other than polio should be done with care. Anderson and Stephens (45) have shown that T6 bacteriophage underwent a general loss of structural integrity following exposure to pH 9.6. Protein sheaths covering the tail structures were "triggered"

in some cases and had become disintegrated from the base to the head. Separated heads, tails and tail base plates were observed in electron micrographs.

Satisfactory explanations for the mechanisms of virus removal by calcium carbonate and $Mg(OH)_2$ precipitates have not been developed. Ca and Mg will react with carboxylate ions of proteins which would provide a virus cation complex which could then be incorporated into the precipitate (34). The negative charge of the calcium carbonate would minimize the electrostatic attractive forces between it and the virus particle.

Calcium hydroxylapatite probably removes viruses by some of the same mechanisms as aluminum hydroxide (24). The hydroxylapatite is hydrolyzed but possesses a negative charge. The importance of the reactions at the hydrolysis groups is probably pointed up by the lack of such sites on calcium carbonate which removes viruses to a lower degree.

SECTION 5

TECHNOLOGY APPLICATION FOR VIRUS REMOVAL

BACKGROUND

Virus removal or inactivation is complicated by several virus phenomena: their small size - as low as 15 μm; the many different types which may be present - over 100 enteric viruses are known; the state in which they exist - as discrete particles or embedded within or attached to solid particles; and the difficulty of detecting them in very small concentrations. When viruses are embedded within certain solids, such as their host cells, they are extremely resistant to inactivation by chemical disinfectants (46) (47). In this case one must depend primarily upon the physical removal of the cell by coagulation and sedimentation and/or filtration since the required dosage for inactivation by ozone and chlorine are well beyond that normally used.

This section will discuss the utilization and application of coagulation and high pH for virus removal and inactivation under field operating conditions. In cases where our present state of knowledge is incomplete best estimates will be applied to the situation.

COAGULATION

The coagulation process is relatively non specific in terms of its ability to remove viruses when the principal precipitate formed is hydrated metallic oxide. This is the usual material formed in waters low in phosphate. In wastewater effluent phosphate is present in larger amounts and aluminum and ferric iron will react preferentially with the phosphate before hydrated oxides are formed. Viruses will absorb to aluminum or ferric phosphates and are subsequently removed with the phosphate although they are less well absorbed on phosphate than on hydrated oxides. Molar ratios of aluminum or iron to phosphate in excess of 1:1 are desirable to provide excess metal for the formation of the hydrated oxide. Enteric virus removal from 90 to 99.999 percent should be obtained in a water which has been well coagulated, flocculated and settled.

The coagulants of choice are either alum, ferric chloride or ferric sulfate. Insufficient experience has been gained with ferrous sulfate to recommend its use at this time. Coagulant dosages which will give the minimum turbidity level after treatment are recommended. Polyelectrolytes are not generally useful in increasing the virus removal which would otherwise be obtained when a water is properly coagulated and flocculated with metallic coagulants. In cases where the coagulation with the metallic coagulant is inadequate due to insufficient dosage or poorly settling floc a polyelectrolyte which will cause the floc to settle should give satisfactory virus removal. Cationic polyelectrolytes are preferred over the nonionic form because of their greater density of positive charges. Anionic polyelectrolytes are not recommended since a report has shown that virus removal is

decreased in their presence. Polyelectrolyte addition may decrease the coagulant dosage needed for virus removal. It would appear desirable to verify this conclusion in each specific application, however.

Better and easier removals of viruses generally occur when turbidity is present. Turbidity in excess of 10 to 15 turbidity units would appear desirable to obtain this effect. This effect appears related to two factors: (1) the coagulation process and subsequent settling of the floc is facilitated when turbidity is present, and (2) the turbidity presents an increased surface area for adsorption of the virus which would not otherwise be present. Viruses are known to absorb readily to turbidity surfaces in water and are thereby removed when the turbidity is removed. Maximum removal of turbidity is also desirable in order to best prepare the water for subsequent disinfection. As mentioned above, viruses embedded within body cells, fecal material, etc., are difficult to inactivate with chemical disinfectants and a water with very low residual turbidity facilitates the disinfection process.

Virus removal by coagulation is slightly affected by water conditions. The temperature and inorganic content probably affect virus removal in a minor way provided that the coagulation and floc formation are otherwise acceptable. The affect of pH has not been well established where metallic hydroxides are formed but a pH on the acid side, between 5-7, appears desirable. Where aluminum phosphate is formed the optimum pH is lower, between 5 to 6.5. Organic content, as in a treated secondary effluent, may reduce slightly the virus removal otherwise expected. In such cases there does not appear to be an acceptable remedy except to optimize the coagulant dosage to assure the lowest residual turbidity possible.

REMOVAL AND INACTIVATION WITH HIGH pH

Viruses are inactivated at high pH and are physically removed by adsorption to precipitates formed at alkaline pHs in water. Most enteric viruses, however, are stable under acid conditions at pH values below those of any interest in water or wastewater treatment. The inactivation obtained at high pH is a function of both the pH and the contact time so the higher the pH within the lethal range and the longer the contact time the greater the expected inactivation. Removal by adsorption to precipitates is a function of type of precipitate and the amount available.

The pH for virus inactivation varies with the type of virus. The few which have been studied indicate that this may range from pH 10 to 12 for an inactivation in excess of 90 percent in 60 to 100 minutes of contact time. A pH of 11.5 to 12 with contact times of about 90 minutes should produce inactivations in excess of 90 percent for most viruses. It is to be emphasized that it is necessary for the virus itself to be exposed to this pH condition and that for viruses embedded with solid particles this probably will not occur. Such particles and their viruses would require removal by sedimentation. The exact effect of increasing contact time has not been determined but longer times will increase inactivation. In the absence of other information a 90-minute contact time appears acceptable.

The principal precipitates formed in water and biologically treated wastewater when lime is added to elevate the pH are calcium carbonate, magnesium hydroxide and calcium hydroxylapatite. Magnesium hydroxide and calcium hydroxylapatite have greater adsorptive capabilities for viruses than calcium carbonate. Adsorption of viruses to these precipitates with subsequent removal will occur well below the lethal pH inactivation range. Increased volumes of precipitates increases the virus removal.

Combined removals and inactivation of viruses in excess of 90 to 99 percent should be expected in the lime flocculation of a typical biologically treated effluent with about 10 mg/l of P. Removals of about 90 percent or more of the viruses should be expected from a typical drinking water excess lime softening plant with a raw water which contains about 25 mg/l or more of magnesium as Mg. Control of the process can be obtained by monitoring pH and turbidity. Turbidity removal should be maximized. Organics in the concentrations expected in a biological effluent should not interfere with the process.

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16. ABSTRACT

Operation of advanced wastewater and water supply treatment plants to assure virological safety of the effluent relies on each unit process removing a finite number of viruses. These treatment plants frequently use chemical coagulation and precipitation at high pH with hydrated lime as part of the process. These treatment methods offer important opportunities for removal and inactivation of viruses from water and wastewater. This report is a literature review which examined the effectiveness of these processes in removing viruses.

17. KEY WORDS AND DOCUMENT ANALYSIS

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