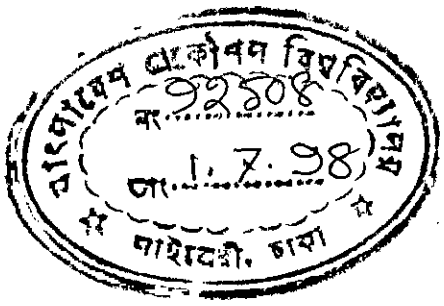
REMOVAL OF MICROORGANISM AND SUSPENDED MATERIAL BY COAGULATION AND FILTRATION



BY
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A thesis submitted to the Department of Civil Engineering of Bangladesh University of Engineering and Technology, Dhaka in partial fulfilment of the requirements for the degree

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I hereby certify that the research work reported in this thesis has been performed by me and this work has not been submitted elsewhere for any other purpose (except for publication).

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ABSTRACT

The quality of water is of vital concern for mankind since it is directly linked with human welfare. But majority of people are still not aware of it. People use surface water for various domestic purposes. Surface water is often physically and bacteriologically contaminated due to unhygienic practices. During floods the water quality deteriorates and treatment is required to make water potable.

The study was devoted to acquire a better understanding of the effectiveness of alum for removal of faecal coliform as well as kaolin particles. Different parameters affecting removal of faecal coliform and kaolin particles were investigated in this study such as alum dosage, pH, sedimentation time and mixing. Filtration with filter paper was performed after coagulation. Removal of faecal coliform was observed at different pH and sedimentation time.

Alum dosage was found effective for reduction of faecal coliform and kaolin particles. With increasing alum dosage faecal coliform and turbidity removal rate were found increasing. pH and sedimentation time also significantly affected the removal of faecal coliform and turbidity. Most of the experiments were performed at pH 4.5 and 6.0. It was observed that the removal efficiency at pH 6.0 was higher than that of pH 4.5.

At any alum dosage, removal of faecal coliform was found to increase with increasing sedimentation time. Moreover for the same removal of microorganism less sedimentation time was required for increased alum dosage. With increasing sedimentation time settlement rate was found to decrease. It was found that with increasing alum dosage potable water could be obtained within a short period of sedimentation.

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CHAPTER-1
INTRODUCTION

1.1 GENERAL

Water is essential to all life, both animal and plant. In order to survive, all animals and plants must have ample supply of water. It is difficult to imagine any clean and sanitary environment without water. Invariably, the progress of sanitation throughout the world has been closely associated with the availability of water. In regions where water is easily available, it is easier to achieve rapid and extensive improvement of public health.

Water spreads communicable diseases in essentially two ways. The first is the well known direct ingestion of the infectious agent when drinking contaminated water. The second is due to lack of sufficient water for personal hygiene purposes. Inadequate quantities of water for the maintenance of personal hygiene and environmental sanitation have been shown to be a major contributing factor in the spread of epidemic diseases. Approximately 80 percent of all sickness can be attributed to inadequate water supply or sanitation. For examples, diarrhoel diseases kill six million children in developing countries every year and affect upto 18 million people. People with water-borne diseases occupy half of all the hospital beds in the world. World Health Organization (WHO) estimates that about 25,000 people die per day from water-related diseases (Asian Environment, 1984).

Bangladesh the 144000 sq. km delta in the bay of Bengal, is inhabited by about 110 million people with low literacy rate and is among the lowest per capita income generating group in the world. Rivers, ponds, canals, ditches, etc. have been the sources of domestic water for ages. In comparison to existing tubewells, often these surface sources have been more convenient, easily accessible, socio-economically and culturally preferable and abundantly available to the majority of the population. But the practices of indiscriminate defecation and disposal of raw sewage into these surface water bodies have created an environment which is heavily polluted and is

of major concern because these water bodies have the potential to serve as reservoir or carrier of pathogens of water related diseases (Bilqis, et. al, 1988).

About 7.2 million people live in Dhaka City Corporation. To meet the demand of water for these people only about 189 deep tubewells and one small water works are now working whereas about 400 deep tubewells are required. Department of Environment has reported that 80% of diseases in Dhaka city are due to impure water. About 330-450 coliforms per 100 milliliter has been identified in water of residence and restaurant of Mugdapara. In the same areas about 10,000 coliform per 100 mililitre has been identified in water of community center (Bichitra, 1995).

Diarrhoel diseases, very common among children, are a leading child killer in developing countries. On a global basis, about 2.2 million children (under five) die from dehydration due to diarrhoel diseases each year (UNICEF, 1997) and need proper water treatment before use.

1.2 RATIONALE OF THE STUDY

Water is found in nature from two main sources : Ground water source and surface water source. Ground water has been extracted in our country by deep and shallow tubewells and also by ring wells. River, pond and ditches are the common sources of surface water in our country. River cover about eight percent of the country even in the dry season. During the monsoon, another 29 percent of the country is inundated, and most of the rest is water logged from frequent rain. Ponds and rivers are the primary water sources for bathing, washing, cleaning and cooking (MPO 1986).

Department of Environment (DOE) in 1984 reported that most of the main rivers, such as Buriganga, Shitalakhya, Balu, Jamuna, Padma, Halda and Surma which run through the crowded cities of the country have coliform counts above the international standard. According to the report all samples, mostly collected at

monthly intervals, showed coliform counts around the range of 2000 to 5000 colonies per 100 ml. Surface water sources receive highly polluted discharges from municipalities, industries and commercial centers located around the sources. The unhygienic sanitary practices of the people, agricultural drainage and annual floods aggravate the deterioration of the quality of surface water. Turbidity and suspended solids are found to increase in monsoon when the surface water receives additional organic and inorganic loads with the drainage of the surrounding plains (Ahmed, 1988).

During 15th to 19th May, 1995, Cyclone hit the coastal belt. After Cyclone, diarrhoea started in cyclone affected area. About 30 people lost their lives everyday. There was no potable water and condition was deteriorating tremendously (Bichitra, 1995). Flood is another natural calamity in Bangladesh. Every year large portion of this country is inundated by flood and it stays for 3-4 months. People, specially child, die due to diarrhoeal diseases which is attributed to contaminated water.

This study is intended to acquire a better understanding of the effectiveness of alum coagulation for removal of faecal coliform and suspended particles from water. Effectiveness of alum coagulation and filtration under a wide range of conditions were investigated in this study.

1.3 OBJECTIVES OF THE STUDY

Following are the specific objectives of the research :

1. To study the removal of faecal coliform by varying alum dosage and to obtain optimum coagulation condition.

2. To study the influence of various parameters such as pH, sedimentation time etc.on removal of faecal coliform and suspended particles.

3. To find out the relationship between the optimum alum dosage and residual turbidity and faecal coliform at a certain pH.
4. To study the removal of faecal coliform and kaolin particles by coagulation and filtration.

1.4 ORGANIZATION OF THE THESIS

The study is presented in five chapters, the first of which is introduction. Chapter 2 contains a brief and selective review of the relevant literature which provides mechanism of coagulation, chemistry of aqueous Al, factors influencing coagulation and indicator organism.

In chapter 3, methodologies adopted in the research work are described. Chapter 4 investigated the efffectiveness of coagulation for the treatment of highly turbid and bacterialogically polluted water. Filtration by filter paper is also included.

In chapter 5, attempts are made to bring the various strands of the study together to provide a deeper understanding of coagulation as conclusion and further studies are recommended.

CHAPTER -2
LITERATURE REVIEW

2.1 INTRODUCTION

Impurities in water normally are of two types, suspended and dissolved. The surface water are characterized by the suspended impurities whereas the ground water are generally free from the suspended matter but are likely to contain a large amount of dissolved impurities. Suspended solids in water may consist of inorganic or organic particles or of immiscible liquids. Inorganic solids such as clay, silt and other soil constituents are common in surface water. Organic material such as plant fibers and biological solids (algal cells, bacteria etc.) are also common constituents of surface waters. These materials are often natural contaminants resulting from the errosive action of water flowing over surfaces. The suspended matter often contains pathogenic or disease producing bacteria; as such surface waters are not considered to be safe for water supply without the necessary treatment. (Peavy et. al 1985).

Recently various methods have been adopted to make water potable and attractive to the consumers. In the case of surface waters, the treatment procedure is involve for removal of silt or turbidity, colour, taste, odour and bacteria. Moreover, the method of water treatment has to be selected on the basis of the character of the raw water to be treated. Plain Sedimentation, Sedimentation with Coagulation and Filtration are the common methods for surface water treatments in tropical country like Bangladesh (Masuduzzaman, 1991). In this chapter, mechanism of coagulation, aqueous Aluminium characteristics and factors influencing coagulation have been discussed.

2.2 COAGULATION

The term coagulation comes from the Latin coagulare, meaning to drive together. It is a chemical technique directed toward destabilization of colloidal particles. Colloids are very fine, lighter particles. The colloidal size range is generally regarded to extend from 1 manometer (10^{-9}m) to 1 micrometer (10^{-6}m). The removal of very fine, light, colloidal impurities from water is difficult to achieve in practice by the process of plain sedimentation. This can be greatly expedited by the addition to water of certain chemical compounds which were thoroughly mixed with woolly masses of flocculant precipitate enmeshing the suspended particles become heavier and finally settle out. This substances are

called coagulants and their process of reaction is coagulation. Rapid mixing is important at this stage to obtain uniform dispersion of the chemical and to increase the opportunity for particle to particle contact. The entire process occurs in a very short time probably less than a second and initially results in particles sub-microscopic in size.

The second stage of the formation of settleable particles from destabilized colloidal sized particles is termed flocculation. Thus in another words, we can say that coagulation is the name given to the actual combining action whereas flocculation is the term to indicate the process of building up of the larger and heavier particles to floc which will settle. In contrast to coagulation, where the primary force is electrostatic or inter-ionic, flocculation occurs by a chemical bridging or physical enmeshment mechanism. Flocculation is operationally obtained by gentle and prolonged mixing which converts the sub-microscopic coagulated particles into discrete, visible, suspended particles. At this stage, the particles are large enough to settle rapidly under the influence of gravity and may be removed from suspension by filtration (AWWA, 1971).

2.2.1 STABILITY FORCES

As a result of immense surface area, colloidal particles tend to rest in suspensnion, it will not settle down. Most colloidal particles in water and waste water are negatively charged. The principal mechanism controlling the stability of hydrobhobic (water hating) and hydrophilic (water loving) particles is electrostatic repulsion. In the case of hydrophobic surfaces, an excess of anions or cations may accumulate in the interface, producing an electrical potential that can repulse particulate of similar surface potential. For hydrophilic surfaces, typically electrical charges arise from dissociation of inorganic groups.

Again individual hydrophobic colloids have an electrical charge, a colloidal dispersion, like an ionic solution, does not have a net electrical charge. For electroneutrality to exist, the charge on the colloidal particle must be counterbalanced by ions of opposite charge contained in the dispersing phase. The ions involved in this electroneutrality are arranged in such a way as to constitute what is called the "Electric double layer".

To discuss the electrical double layer, it is convenient to start by considering the charge on the colloidal particle. The charge on the particle will be offset by ions of opposite charge which accumulate around the particle to form the outer

coating of the double layer. The high electrical potential on the particle surface will decrease to zero at some distance from the particle as a result of the accumulation of counter-ions.

A model proposed by Stern (1924) can be used to describe the distribution of the electrical potential in the vicinity of a colloidal particle. First, consider a charge as shown in the figure 2.1. The electrical potential created by this charge at the particle surface will attract counter-ions toward the particle. According to Stern, the distance of closest approach of these counter-ions to the particle will be limited by the size of the ions. He proposed that the centre of the closest counter-ions are separated from the surface charge by a layer of thickness Ω (a distance approximately equal to the hydrated radius of the ion) in which there is no charge. This layer is referred to as stern layer.

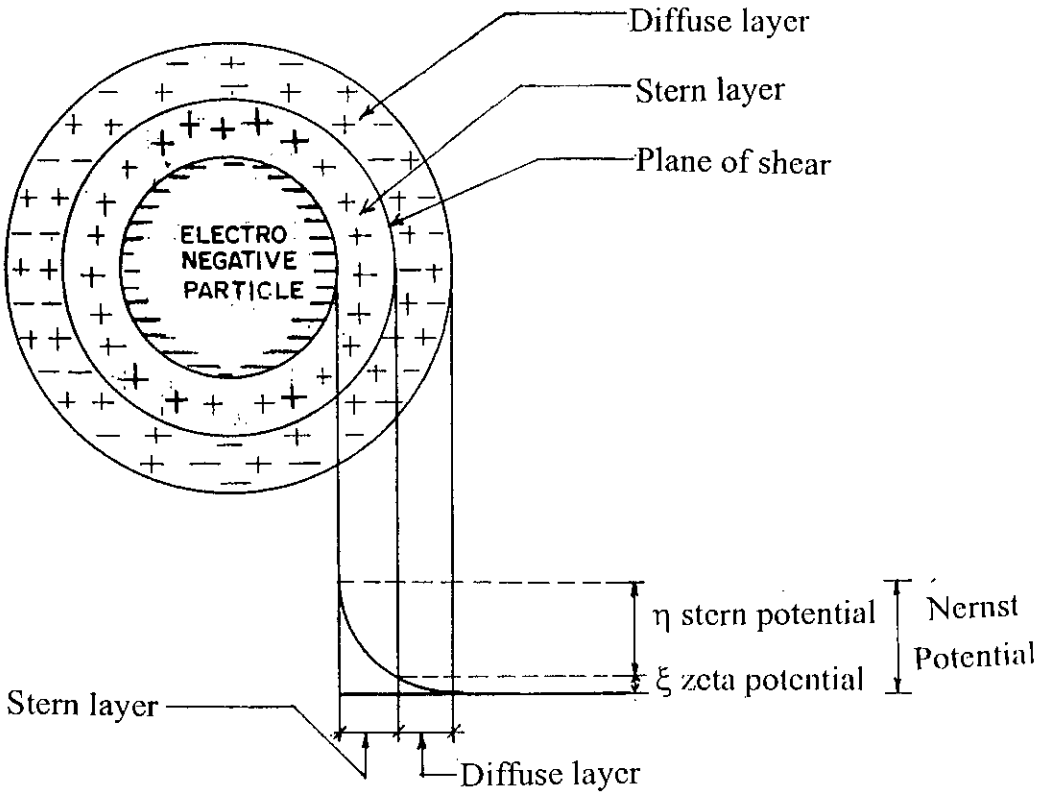


Figure 2.1: Guoy-Stern Colloidal Model (Source : Steel and McGhee,1979)

A consequence of the electrical double layer surrounding each particle in water is to create regions of electrical potential in a bulk solution that nominally has a zero potential. The electrostatic work required to transport a unit charge from this bulk solution through the phase boundaries of each layer surrounding a particle to any point measures the particle of that point. The potential increases sharply with increasing approach to the primary particle surface. The potential at the surface of the particle is called the total potential or Nernst potential, is represented by ψ. For certain surfaces of known composition, ψ can be calculated. However, its value is unobtainable for impure colloids of interest in water treatment.

The second potential of interest is called the Zeta Potential (ξ) and is located at the "Plane of Shear", that is at the boundary between the solvent adhering to the particle in its motion and that which can move with respect to it. This "plane of shear" essentially separates the water of hydration from free water.

The zeta potential is interesting and important because it is related to particle stability, and approximate values of zeta potentials can be calculated from easily measured electrophoretic mobilities. It should be pointed out that the zeta potential does not measure that total potential, but rather a portion of it.

The net effect of the existence of electrical double layers around particles is to inhibit the close approach of particles to each other and hence the double layers confer stability to the suspension. Both the thickness of the double layer and the surface charge density are sensitive to concentration and valence of the ions of the solution and hence stability of the suspension can be markedly affected by adding suitable ions to the solution.

The stability of colloidal material arises from the predominance of forces associated with the solid-liquid interface. Interfacial phenomenon have been mainly concerned with interactions originating from four different sources. These are :

i. Couloumbic force or electrostatics force - from the charge of the particle.

ii. Vander waals' force-attraction between particles irrespective of their charge and distance

iii. Chemical bonds

iv. Thermal random forces.

The Vander waals' forces of attraction, which opposes the repulsive forces, are largely due to universal attractive forces. The Vander waals' attractive energy of interaction, V_A, between two particles is inversely proportional to the second power of the distance separating the particles and thus decreases very rapidly with increasing intermolecular distance. The variation in particle attractive energy with particle separating distance is shown in figure 2.2

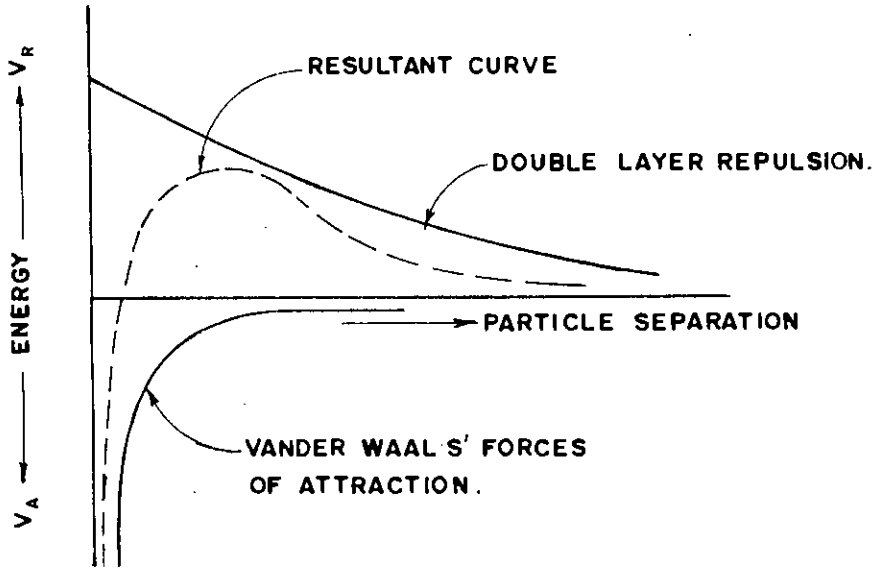


Figure 2.2: Schematic Diagram of Potential Curves (Source : Peavey et. al,1985)

The repulsion and attraction curves can be combined to form a curve representing the resultant energy of interaction. This curve indicates that repulsion forces predominate at certain distances of separation, but that if the particle can be brought to close enough together, the Vander waals' attractive forces will predominate and the particles will coalesce.

2.2.2 DESTABILIZATION OF COLLOIDS.

The destabilization of colloid can be accomplished by different mechanisms. Lamer (1964) has divided these into two categories.

a. Processes that effect a reduction of total potential energy of interaction between the electrical double layers of two similar particles.

b. Processes that aggregate colloidal particles into a three dimensional floc network by the formation of chemical bridges.

Lamer (1964) has designated the first as coagulation and the second as flocculation. The destabilization and aggregation of colloidal system have been explained by four models:

a. The double layer compression
b. Adsorption and charge neutralization
c. Adsorption and interparticle bridging
d. Enmeshment in a precipitate (sweep flocculation)

In practice, the colloidal destabilization is likely to be caused by more than one method. The mechanism of colloidal destabilization are briefly described in the following sections.

2.2.2.1 DOUBLE LAYER COMPRESSION

In this model the destabilization of colloids, only electrostatic interactions are considered significant. Ions of similar charge to the primary charge of the colloids are repelled and counter-ions are attracted. Destabilization by counter-ions is accomplished by compressing the diffuse layer surrounding the colloidal particles. The reduction of energy barrier between two particles enhance agglomeration. Ions acting only in this way are usually referred to as indifferent ions.

A mathematical model for the effect of indifferent electrolytes on the colloidal stability has been developed by Verwey and Over beek (1948). The main features of the model are as follows :

a) The concentration of coagulant necessary to destabilize a colloid is independent of the colloidal concentration and depends on the valence of the ions,

b) Charge reversal and restability of colloid cannot occur.

Gregory (1978) and Bratby (1980) have shown that high concentration of electrolyte in the solution results in high concentration of counter ions in the diffuse layer. This will cause a reduction in the thickness of the diffuse layer and consequently lowering the energy barrier which, effectively control the range over which electrical forces operate between particles.

2.2.2.2 ADSORPTION AND CHARGE NEUTRALIZATION

The charge on a colloid can be neutralized by the addition of molecules of opposite charge which have the ability to adsorb onto colloids. In the process of

destabilization of colloids, it is helpful to consider the energy involve in the electrostatic interaction between a colloid particle and a coagulant ion. Bonding of very short range type such as hydrogen bonding will occur. Also coordinating reactions and ion exchange reaction can take place and lead to ion adsorption and neutralization of the particle charge.

According to Stumm and O' Melia (1968), the destabilization of colloid by counter-ions brought about by the adsorption of counter ions of the surface of particles causing neutralization of charge. Amount of coagulant required to coagulate colloids is linearly proportional to the surface area of colloids and at excess coagulant dosage, restablization can occur, leading to charge reversal.

2.2.2.3 ADSORPTION AND INTERPARTICLE BRIDGING

Starch, cellulose, synthetic polymeric compounds are characterized by a large molecular size and most have multiple electrical charge along a molecular chain of carbon which can form bridges between particulate, thus destabilizing the suspension.

The chemical bridging theory proposes that a polymer molecule will become attached to a colloidal particle at one or more sites. Attachment may result from coulombic attraction if the polymer and particle are of opposite charge or from ion exchange, hydrogen bonding or Vander waals' forces if they are of similar charge. The tail of the adsorbed polymer will extend out into the bulk of the solution and can become attached to vacant sites on the surface of another particle to form a chemical bridge and the formation of a floc particles of good setting characterizes.

The early theory by Healy and Lamer (1962) indicated that the optimum dosage for maximum flocculating effect occurs when half of the available surface sites on the solid particles have been covered with polymer. For homogenous

suspensions there is a direct relationship between particle concentration and optimum polymer dosage. Polymer dosages that saturate the available surfaces of the dispersed phase produce restablization because no sites are available for the formation of polymer dosage.

2.2.2.4 ENMESHMENT IN PRECIPITATE

The use of a metallic salt as a coagulant in water treatment can lead to a precipitate of metal hydroxide. Colloidal particles may be enmeshed (sweep) in the precipitates. Edwards and Amirtharajah (1985) and Vik et. al, (1985) have developed a design and operational diagram for alum coagulation, where sweep and optimum sweep coagulation stages are distinctly marked.

Working with dispersed clay on water, Packham (1965) showed that the rate of precipitation of metal hydroxide increases with increasing concentration of the colloidal particles to be removed. This suggested an inverse relationship between the coagulant dosage and the concentration of colloids. Under such condition, the use of a coagulant aid can reduce the required dosage.

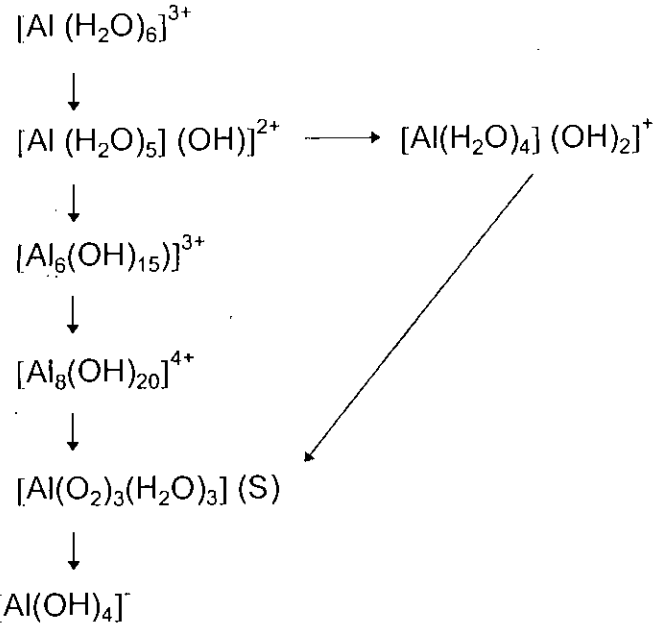
2.2.3 CHEMISTRY OF AQUEOUS Al

Aluminium salts are the most widely used coagulants in water treatment, the different aluminium species exhibits in water are directly associated with turbidity removal. Thus it is necessary to consider the aqueous chemistry of these coagulants.

There is little doubt that the aqueous chemistry of aluminium is complex and diverse because of numerous hydrolysis intermediates formed prior to precipitation of aluminium hydroxide, $Al(OH)_3$ (s). Matijevic et. al, (1964) provided a fairly clear description of the hydrolysis species of Al^{3+} and their

interactions with the colloids in the context of coagulation and restabilization. When aluminium salts are added to water, the metal ion Al^{3+} hydrates coordinating six water molecules and forming an aquometal ion, $Al(H_2O)_6^{3+}$. The aquometal ion can then react and form several hydrolysis species, some of the simplest being monomeric and dimeric hydroxocomplexes where coordination occurs with OH^- ligands that replace the six co-ordinated water molecules.

Stumm and Morgan (1962), Packham and Sheiham (1977) and others have given the stepwise conversion of the tripositive aluminium ion to the negative aluminium ion as pH increases are as follows :



Amirtharajah and Mills (1982), Vik,et. al.(1985) and others have reported a number of aluminum polymers that may possibly be involved in coagulation and flocculation process. These are :

$Al_{13}(OH)^{5+}_{34}$, $Al_7(OH)^{4+}_{17}$, $Al_8(OH)^{4+}_{20}$, $Al_6(OH)^{3+}_{15}$

$Al_2(OH)^{4+}_{2}$, $Al_2(OH)^{+}_{5}$, $Al_4(OH)^{4+}_{8}$, $Al_{13}(O)_4\,(OH)^{7+}_{24}$

$Al_3(OH)^{5+}_{4}$, $Al_{13}(OH)^{7+}_{32}$ and $Al_{14}(OH)^{8+}_{34}$, to name but few.

Dempsey et. al (1984) contend that the evidence for the existence of such polymers is myriad and indirect. However the degree of alkalinity in the solutions an important determinant in aluminium speciation. Other dominant factors include the concentration of reactants, contamination by other substances, pH, temperature and reaction time. Matijevic et. al (1964) have found that the hydroxo-metal complexes readily adsorb on surfaces and the charges that they carry may cause charge reversals of the surfaces that they adsorb on : second, the sequential hydrolysis reactions release H^+ ions, which lowers the pH of the solution in which they are formed ; and the concentration of the various hydrolysis species will be controlled by the final concentration of H^+ ions and is, by the pH. These findings are in keeping with the individual suggestions of O'Melia and Stumm (1967) and O'Melia (1972).

Hundt and O' Melia (1988) described the aqueous chemistry of Al using five monomers Al^{3+}, $Al(OH)^{2+}$, $Al(OH)_2^+$, $Al(OH)_3$ and $Al(OH)_4^-$; three polymers $Al_2(OH)_2^{4+}$, $Al_3(OH)_4^{5+}$ and $Al_{13}O_4\ (OH)_{24}^{7+}$ and a solid precipitate Al $(OH)_3$ (s). They also have discussed the influence of the chemical behavior of anions of Al in solution. The presence of anions with a strong affinity for Al, such as sulphate, greatly affects the Al species in solution. Sulphate, a tetrahedral polyvalent anion, tends to link OH-Al polymers together, but in a distorted arrangement. Therefore, most basic salts containing sulphate are necessary to produce a visible precipitate than when solutions containing chloride or nitrate salts are titrated. They also presumed a serving effect, that accelerates the formation of polymers and assists in the linking of planner complexes to form the solid lattice.

Sullivan and Singley (1968) have estimated the quantity of mononuclear species (monomers) at different pH, the estimates are given in figure 2.3. The dominant species upto pH 4.5 is Al^{3+}, from 4.5 to 8 it is Al $(OH)_3$ and above pH 8 it is $Al(OH)_4^-$. Hossain (1996) show that at pH 4.5 maximum +ve Al species form and at pH 6.0 most of the Al species are $Al(OH)_3$.

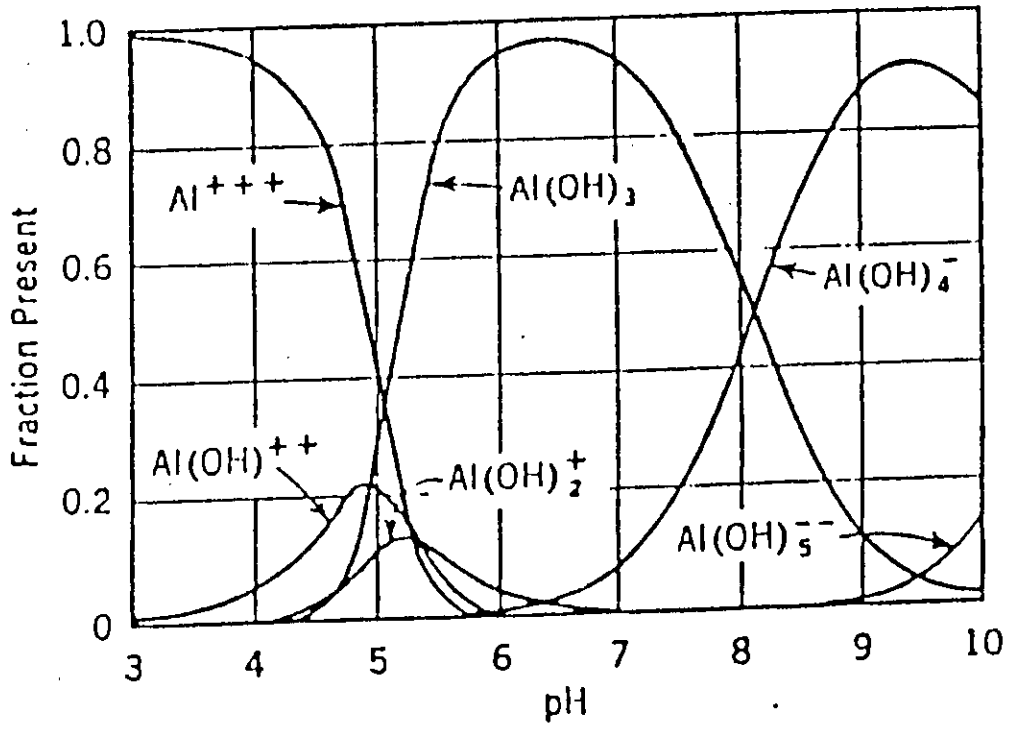


Figure 2.3 Species present as a function of pH for 1×10^{-4} M Aluminium perchlorate (Sullivan & Singley,1968).

Matsui and Tambo (1989) have estimated that the precipitated aluminium species has much larger size than 1μm and the volume of 1 mg of aluminium hydroxide has about 50 times greater volume than 1 mg of clay particles.

2.2.4 TYPES OF COAGULANTS USED

The Coagulants commonly used in water purification are alum or aluminium sulphate, copperas or iron sulphate and lime. Other less commonly used coagulants are sodium aluminate, ferric chloride, ferric sulphate, chlorinated copperas and activated silica etc.

Aluminium sulphate or alum is a grayish-white solid available in the form of lumps containing 17% aluminium sulphate. This is easily available and widely used in water works. Alum is most effective between pH ranges of 5.5 - 8.0. The alum produces good floc which removes suspended and colloidal impurities.

Copperas (ferrous sulphate) is a good coagulant, cheaper than alum and is good for waters whose pH is 8.5-11.0. Although less expensive than alum, these coagulants can cause colour problems if the precipitate is not removed completely.

Sodium aluminate is an alkaline compound and can be used in waters which are not alkaline. It reacts very rapidly and forms a good precipitate of aluminium hydroxide. It does not produce hardness in water and also removes colour. But it is costly and hence not widely used.

Ferric coagulants are good oxidizing agents. They remove hydrogen sulphate, taste and odorous from water. Chlorinated copperas is a mixture of ferric chloride and ferric sulphate obtained by chlorinating ferrous sulphate. It is a

good coagulant and is very effective even with less alkalinity of water. The floc produced is tough and easily settles down. This coagulant removes colour also.

2.2.5 FACTORS INFLUENCING COAGULATION

Optimum coagulation treatment of a raw water represents the attainment of a very complex equilibrium in which many variables are involved. Thus for a given water, there will be interrelated optima of conditions such as pH, turbidity, chemical composition of the water, type of coagulant temperature and mixing conditions.

2.2.5.1 EFFECT OF pH

pH is the most important variable that affects coagulation process in water treatment. Some investigators established that there is at least one pH range for any given water within which good coagulation -flocculation occurs in the shortest time with a given coagulant dosage. The extent of the pH range is affected by the type of coagulant used and by the chemical composition of the water as well as by the concentration of coagulant.

Aluminium sulphate, the most commonly used coagulant in water purification, is most effective between pH range of 5.5-8.0. The pH zone of coagulation by iron salts is generally broader. Interrelations between pH and required alum dosage to coagulate clay, residual aluminium, and turbidity of precipitated coagulant are shown in figure 2.4. Curve A shows that the minimum dosage to remove 50 mg/l clay turbidity occurs within a pH range of 6.8 to 7.8, residual aluminium after treatment with 200 mg/l alum, shown in curve B, is minimal at a pH of between 5.5 and 7.5; and flocculation of aluminium hydroxide in the absence of clay, shown in curve C, occurs most rapidly at a pH of about 7.2.

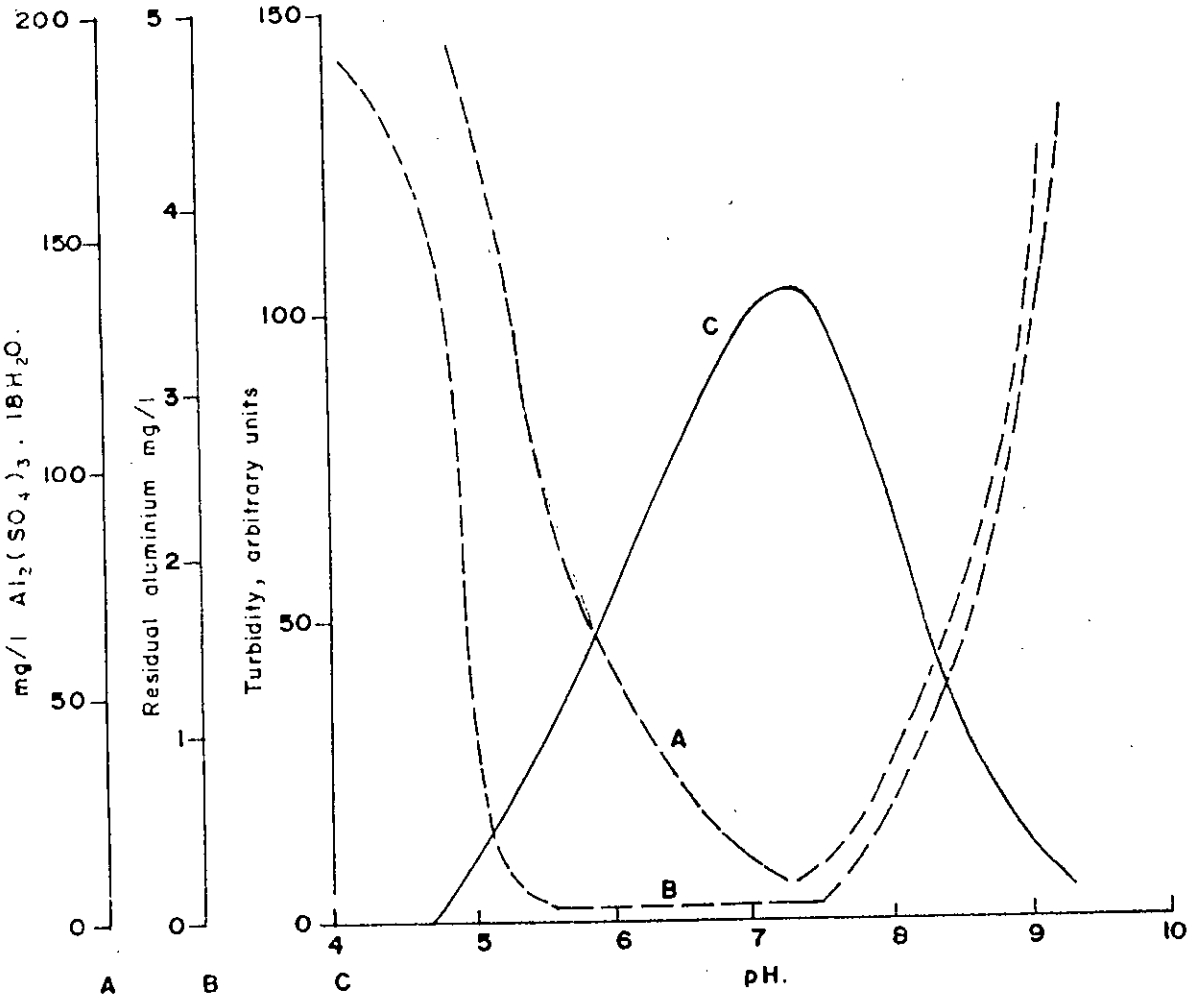


Figure 2.4 Comparison of the effect of pH on: Curve A, dosage of auminium sulphate required to halve turbidity of 50 mg/l clay suspension ; curve B, aluminium solubility as indicated by residual aluminium after coagulation with 200 mg/l $Al_2(SO_4)_3$. 18 H_20 ; curve C, flocculation of $Al(OH)_3$ as indicated by the turbidity 1 minute after mixing the reagents. [Source: Packham, (1963)]

Coagulation should, if possible, be carried out within the optimum pH zone for the particular water. For certain waters it may be necessary to adjust the pH with acid, lime, soda ash etc. to obtain the proper conditions. Addition of excess coagulant may in some plants be a more practical way to reduce pH to the favorable range then separate acid addition (AWWA,1990).

2.2.5.2 EFFECT OF SALTS

Natural waters are never pure water but are dilute solutions of inorganic salts of varying concentration and composition. The effects of these salts on the coagulation process have been extensively studied and have been shown to exert an influence, depending on the specific ion and its concentration. The principal effects of the presence of certain ions are to alter 1) the pH range of optimum coagulation (2) the time for flocculation (3) the optimum coagulant dosage (4) the residual coagulant in the effluent.

Extensive experimentation such as the foregoing has led to the following generalizations on the effect of ions on coagulation:

1. In general, coagulation with aluminium or iron salts is subject to greater interference from anions than from cations. Thus, ions such as sodium, calcium and magnesium have relatively little effect on coagulation.

2. Anions extend the optimum pH range for coagulation to the acid side to an extent dependent on their valency. Thus monovalent anions such as chloride and nitrate have relatively little effect, while sulphate and phosphate cause marked shifts in pH optima (AWWA, 1990).

2.2.5.3 EFFECT OF NATURE OF TURBIDITY

Turbidity in surface waters consists largely of clay and other mineral particles. The size range of most of these particles is about 0.2 to 5.0μ. Coagulation of such suspensions is accomplished relatively easily, one the proper pH has been determined. The following generalizations can be made.

1. A certain minimum of coagulant must be added for any clay turbidity concentration in order to provide an enmeshing mass of floc.

2. Some additional coagulant is generally required with increase in turbidity, but the dosage of coagulant will not increase linearly with increase in turbidity.

3. Paradoxically, with very high turbidities relatively smaller coagulant dosages are required because of the high collision probabilities, for the same reason the very low turbidities are frequently more difficult to coagulate.

4. The exchange capacity of the turbidity in water does not seem to be important although it has been suggested that increasing exchange capacity was the cause for increasing coagulant demand.

5. The organic matter frequently adsorbed on clay from natural stream waters does not increase coagulant demand.

6. A broad distribution of clay particle sizes is much easier to coagulate than a suspension containing a single or narrow range of particle sizes(AWWA, 1990).

2.2.5.4 EFFECT OF COAGULANT

One of the factors influencing coagulation is the coagulant. Alum is by far the most commonly used coagulant, iron salts can be used as well and in some instances have advantages over alum. A significant advantage of iron salts over aluminium is the broader pH range for good coagulation. Thus, in the treatment of soft coloured waters where colour removal is best obtained at acid pH's the use of iron salts is preferable. The greater insolubility of ferric hydroxide at alkaline pHs, in contrast to aluminium salts which form soluble aluminate ions at basic pH's, suggests that iron salts should be considered under these conditions. Sodium aluminate is, however, used for coagulation of softening precipitates at high pH (AWWA, 1990).

2.2.5.5 EFFECT OF TEMPERATURE

Difficulties in coagulation are encountered when water temperatures approach O°C. As temperature decreases, the viscosity of water increases and hence the rate of settling of floc is decreased. While decrease of temperature is known to decrease the rate of chemical reactions, this effect on coagulation is probably too small to be significant. It has been observed, however, that the optimum pH value is decreased by decreases in temperature and that this shift becomes more important with smaller dosages of coagulant.

The difficulties arising from cold temperatures can generally be overcome by (1) conducing the coagulation as near as possible to the pH optimum for that water at that temperature (2) increasing the coagulant dosage, which not only increases collision probabilities of particles but reduces that effect of optimum pH shift or (3) adding aids in the form of clays to increase floc density and coagulant aids such as activated silica and polyelectrolytes to increase floc strength and promote more rapid settling (AWWA, 1990).

2.2.5.6 EFFECT OF MIXING

Two stages of mixing or other means to create turbulence are generally used in a water plant. Rapid mixing to distribute the coagulant throughout the water being treated is frequently called "flash mixing". This rapid mixing is essential to uniformly disperse the coagulant and to promote collisions of coagulant particles with turbidity particles. Such high turbulence conditions should be maintained for 30 to 60 sec at the end of which the coagulant has hydrolyzed and has been adsorbed on the turbidity particles.

The second stage of floc growth or flocculation is accomplished by gentle stirring. Immediately following the addition of coagulant and during the flash mixing step, coagulation and precipitation produce finely divided suspensions. In a quiescent fluid, Brownian movement and the differences in settling velocities of various sized particles would result in ever-increasing size of floc. But floc growth by these means is exceedingly slow. Floc growth can, however, be hastened by stirring the water. This increases the number and opportunities for particle contacts by collision. The degree of agitation must be great enough to keep the floc particles suspended and in motion, but not so great that the floc is disintegrate by the shearing forces. This forces is called "flocculation". Detention times as little as 10 min but more frequently 30 to 60 min, are generally adequate to produce a floc that will settle in a reasonable time (AWWA, 1990).

2.2.6 JAR TEST TO DETERMINE OPTIMUM COAGULANT DOSAGE

This is a laboratory method to determine the optimum dosage of a particular coagulant which is required to be added to the raw water for coagulation and subsequent sedimentation in treatment plant.

The Jar test is performed using a series of glass containers that hold at least 1L and are of uniform size and shape. Six jars are used with a stirring device that simultaneously mixes the contents of each jar with a uniform power imput. Each of six jars is filled to the 1L mark with water whose turbidity, pH and alkalinity has been predetermined. One jar is used as a control, while the remaining five are dosed with different amounts of coagulants at different values until the minimum values of residual turbidity are obtained. After chemical addition, the water is mixed rapidly for about 1 minute to ensure complete dispersion of the chemicals, then mix slowly for 15 to 30 minute to aid in the formation of flocs. The water is next allowed to settle for approximately 30 minute so that floc may form, the smallest dosage of coagulant that produces good floc is taken as the optimum dosage for the particular waters (Aziz, 1975).

2.3 INDICATOR ORGANISM

Testing a water sample for pathogenic bacteria might at first glance be considered a feasible method for determining its bacteriological quantity. However on closer examination, this technique has a number of shortcomings that precludes its application. Pathogens are likely to gain entrance sporadically and they do not survive for very long period of time consequently they could be missed in a sample submitted to the laboratory. Although it is possible to detect the presence of various pathogens in water, the isolation and identification of many of these is often extremely complicated and seldom quantitative. Tests for specific pathogens are usually made only when there is a reason to suspect that those particular organisms are present at other times, the microbiological quality of water is checked using indicator organisms. An indicator organism is one whose presence presumes that contamination has occurred and suggests the nature and extent of the contaminants.

A number of microorganisms have been evaluated as indicators, including total coliforms, faecal coliforms, E. coli, faecal streptococci, pseudomonas

aerugionsa, enterococci and HPC. Yeast's have also recently been proposed as effective indicators. However total coliforms and the faecal coliforms have remain as the indicator of choice for decades, mainly because no other indicator has been proven to be more comprehensive than these two (Alam, 1996).

2.3.1 TOTAL COLIFORM

The term coliform organisms (total coliforms) refers to any rod-shaped, non spore forming, gram negative bacteria capable of growth in the presence of bile salts or other surface active agents with similar growth inhibiting properties, which are cytochrome-oxidase negative and able to ferment lactose at either 35 or 37°c with the production of acid, gas and aldehyde within 24-48 hours. Total coliform includes E. coli, Enterobacter, klebsiella and Citrobacter. These are present in the faeces of warm-blooded animals as well as soil and plant (WHO, 1984).

2.3.2 FAECAL COLIFORM

Faecal Coliform is a subgroup of total coliforms which ferment both lactose and other suitable substrates such as mannitol at 44.5 ± 0.2°c with the production of acid and gas. This more stringent conditions eliminate most of the non-faecal component while still permitting to faecal component to survive (WHO, 1984).

2.3.3 FAECAL STREPTOCOCCI

The other group of non-pathogenic organisms proposed as indicators of faecal contamination are the faecal streptococci. The varieties considered strictly as faecal in origin are : S. Faceless, S. Facecalis, Var. Liquefaciens, S. Facecalis Var. Zymogenes, S. Durans, S. Faecium, S. Bovis and S. Equinus. Large numbers of these non pathogens occur normally in faeces, their abundance

being on the some order of magnitude as that of coliforms. It has been thought that the faecal streptococcus group occurs only in the faeces of humans and other warm-blooded animals and therefore constitutes a more specific test for faecal contamination than the coliform group. However recent studies indicate that streptococci similar to the faecal streptococci may also be found on certain plants, plant products, and in waste from food processing plants. The strepto coccus group, although not replacing the coliform group as the standards is considered to be a confirmation that coliform organisms found in water samples are of faecal origin (Alam, 1996).

2.3.4 GUIDELINE VALUES FOR BACTERIOLOGICAL QUALITY

The guideline values for bacteriological quality established by WHO are given in table 2.1. These values are only a guide to those required to ensure bacteriological safe supplies of drinking-water whether piped, unpipped or bottled. Treated water entering the distribution system should be free form coliform organisms, however polluted the original raw water may have been. In practice, this means that it should not be possible to demonstrate the presence of any coliform organism in any sample of 100 ml.

The desirability of disinfecting all supplies of piped drinking-water before distribution should be considered . Supplies derived from protected sources which are distributed without disinfection should be similar in quality that of disinfected drinking water. No water entering a distribution system should be considered satisfactory if coliform organisms are detected in any sample of 100 ml. The presence of not more than 3 coliform organisms per 100 ml may be tolerated in occasional samples.

Bottled water must be at least as good in bacterial quality as unbottled potable water and thus contain no coliform organism. During an emergency it may be necessary either to modify the treatment of existing sources or to use

alternative sources of water temporarily. It may be necessary to increase disinfection at source or to rechlorinate during distribution. If the quality cannot be maintained. Consumers should be advised to boil the water during the emergency.

Recommended limits of total and faecal coliform for different types of water-use established by Millipore cooperation, U.S.A. are given in table 2.2.

Recently Department of Environment (DOE), Bangladesh has also set environmental quality standards. This standard has classified water into 7 categories according to use. The standards for total coliform and faecal coliform is shown in table 2.3.

Table 2.1 Guideline values for bacteriological quality

Organism	Unit	Guideline value	Remarks
A. Piped Water supplies			
A.1 Treated water entering the distribution system			
faecal coliforms	number/100 ml	0	turbidity<1 NTU; for disinfection
coliforms organisms	number/100 ml	0	with chlorine, pH preferably <8.0, free chlorine residual 0.2-0.5 mg/l following 30 minutes (minimum) contact
A.2 Untreated water entering the distribution system			
faecal coliforms	number/100 ml	0	
coliform organisms	number/100 ml	0	in 98% of samples examined throughout the year-in the case of large supplies when sufficient samples are examined
coliform organisms	number/100 ml	3	in an occasional sample, but not in consecutive samples
A. 3 Water in the distribution system			
faecal coliforms	number/100ml	0	

Organism	Unit	Guideline value	Remarks
coliform organisms	numbr/100 ml	0	in 95% of samples examined throughout the year - in the case of large supplies when sufficient samples are examined
coliform organisms	number/100 ml	3	in an occasional sample but not in consecutive samples
B. Unpiped water supplies			
faecal coliforms	number/100 ml	0	
coliform organisms	number/100 ml	10	should not occur repeatedly; if occurrence is frequent and if sanitary protection cannot be improved an alternative source must be found if possible
C. Bottled drinking - water			
faecal coliforms	number/100 ml	0	source should be free form
coliform organisms	number/100 ml	0	faecal contamination
D. Emergency water supplies			
faecal coliforms	number/100 ml	0	advise public to boil water in case
coliform organisms	number/100 ml	0	of failure to meet guideline values

Table 2.2 Recommended limits of total and faecal coliform

Type of Water	Total Coliform per 100 ml		Faecal Coliform per 100 ml	
	Desirable	Permissible	Desirable	Permissible
Potable and well water	0	1	0	0
Primary Contact water (Swimming)	<1000	<2400	<200	<1000
Secondary Contact Water (Boating, Fishing)	<5000	<1000	<1000	<5000
Treated Sewage Effluent	Coliform levels should not exceed those of water receiving the discharge			

Table 2.3 Bangladesh standard for microbial water quality (DOE, 1991)

Type of water use	Total coliform (#/100 ml)	Faecal coliform (#/100 ml)
Drinking	2	0
Recreational	200	NYS
Fishing	5000	NYS
Industrial	NYS	NYS
Irrigaion	1000	10
Livestock	100	NYS
Coastal	1000	NYS

The standard limits of microbial loading for effluents set by DOE is listed in Table 2.4

Table 2.4 Standard limits of total and faecal coliform in effluents

Type of effluents	Discharge into	Total coliform (#/100 ml)	Faecal Coliform (#/100 ml)
Sewage effluent		1000	NYS
Industrial effluent	Inland Surface Water	10000	100
	Public Sewer	10000	100
	Land	10000	100

2.4 GUIDELINE VALUES FOR TURBIDITY

High levels of tubidity can protect microorganisms from the effects of disinfecton and can stimulate the growth of bacteria. In all cases where water is disinfected, therefore, the turbidity must be low so that disinfection can be effective.

WHO recommended that the guideline value is 5 Nephelometric Turbidity Units (NTU) or 5 Jackson Turbidity Units (JTU) but preferably less than 1 NTU when

disinfection is practiced. Turbidity in excess of 5 NTU (5 JTU) may be noticeable and consequently, objectionable to consumers.

The environmental quality standards established by Department of Environment, Bangladesh has also classified turbidity according to 7 categories of use. The standard for turbidity is shown in table 2.5.

Table 2.5: Bangladesh standard for turbidity (DOE, 1991)

Type of water use	Turbidity (JTU)
Drinking	10
Recreational	10
Fishing	NYS
Industrial	50
Irrigation	NYS
Livestock	NYS
Coastal	75

NYS = Standard is not yet set.

CHAPTER-3
METHODOLOGY

3.1 INTRODUCTION

This chapter describes the methodology adopted in this research work. It describes preparation of water samples, preparation of stock cell suspension and the methodology used for determination of faecal caliform and turbidity. Of the two standard methods available for detection and enumeration of faecal caliform, Membrane Filter Method was used. To measure turbidity, Hach turbidimeter was used. Number of faecal coliform was expressed in number/100 ml and turbidity was expressed in nephelometic turbidity unit (NTU).

3.2 PREPARATION OF RAW WATER SAMPLE

Distilled water was used as the main source of water for the experiments. pH of Distilled water was 6.8 ± 0.2. Kaolin was used as suspended material for this study.

3.3 PREPARATION OF STOCK CELL SUSPENSION OF FAECAL COLIFORM

About 1L of sewage containing microorganism was brought from manhole. Small amount of potato chips were added to the suspension, oxygen was supplied continuously to the suspension for the growth of bacteria. After 24 hours 200 ml from the top of the suspension was drawn in air tight bottle and preserved in refrigerator to inhibit the growth of bacteria. This suspension was used within 4 days as described by Alam in 1996. Before starting the experiments initial concentration of microorganism was determined in each batch of experiments. After 4 days cell suspension was drained out and bottle was cleaned to prepare a new stock suspension with freshly collected sewage.

3.4 METHODS OF BACTERIOLOGICAL EXAMINATION OF WATER

Assessment of bacteriological water quality may be done either by testing for indicator organism or by detecting individual pathogen. However, due to ease of detection and enumeration, testing for indicator organism has now become the standard test for assessing bacteriological water quality.

There are two standard methods for detection and enumeration of indicator organism (APHA, 1985)

i Multiple Tube Method

ii. Membrane Filter Method

3.4.1 MULTIPLE TUBE METHOD

In the multiple-tube (MT) method, a series of tubes containing a suitable broth culture medium is inoculated with test portions of a water sample.

After a specified incubation time at a given temperature, each tube showing gas formation is regarded as "presumptive positive" since this indicates the possible presence of coliforms ; however, since gas may also be produced by other organisms, a subsequent confirmation test is advisable. The two tests are known respectively as the presumptive and confirmed tests.

For the confirmed test, a more selective culture medium is inoculated with material taken from the positive tubes. After an appropriate time interval, the tubes are examined for gas formation as before. The concentration of bacteria in the sample can then be estimated from the number of tubes inoculated, and the number of positive tubes obtained in the confirmed test. The most probable number (MPN) of bacteria present can be estimated using specially devised statistical tables. This technique is known as the MPN method.

3.4.1.1 FIELD EQUIPMENT FOR MPN.

i) Hot-air oven, ii) autoclave, iii) incubator, iv) water-bath, v) pH meter, vi) balance, vii) water distillation apparatus, viii) dilution bottles, ix) pipettes, x) media preparation equipment, xi) gas burner, xii) culture tubes containing inverted vials. xiii) test-tube racks, xiv) inoculation loop and holder and general laboratory equipment.

3.4.1.2 CULTURE MEDIA

Commercially available dehydrated media simplify the preparation of culture broth, and are therefore recommended for laboratory work. Various manufacturers produce these media as powders, which can then be easily weighed out, dissolved in distilled water, and dispensed into culture tubes prior to sterilisation.

Several different culture media are available for the presumptive test, for example :

- lauryl tryptose broth (LTB) ;
- MacConkey broth ;
- lactose broth

These three media are in common use in many countries. The selectivity of MacConkey broth and LTB depends respectively on the presence of bile salts and the surface-active agent, lauryl sulphate. lactose broth is a non-selective medium.

As a confirmatory medium for total coliforms brilliant green lactose bile broth (BGB) is most widely used.

To confirm the presence of faecal coliforms, either BGB broth or Escherichia coli (EC) broth is used.

Preparation of media : Media should be prepared in accordance with the manufacture's instructions. The general practice described below.

i. A given amount of dehydrated medium is dissolved in distilled water to obtain the double-strength or single-strength presumptive medium (for confirmatory analysis, only single strength medium is used).

ii. Requisite volume of medium is dispensed into culture tube containing an inverted Durham tube and the tube is then capped.

iii. Culture tubes are then sterilised in an autoclave or pressure cooker at 114^0C for 10 minutes (or in accordance with the manufacturer's specifications). It is particularly important that media containing disaccharide, e.g. lactose, are not autoclaved at higher temperatures.

iv. The sterilised medium should be stored at room temperature (approximately 25^0C), to maintain sterility. In addition, since several dyes are light-sensitive, the solution should be protected from exposure to light.

3.4.1.3 TEST PROCEDURE

Figure 3.1 shows the procedures involved in Multiple Tube test of water and waste water sample, together with the appropriate incubation times and temperatures. The procedure to be used for testing relatively unpolluted water, such as treated water from waterworks, is described below.

i. The paper wrapping is removed from the sample bottle.

ii. With the stopper in position, the bottle is shaken vigorously to achieve a homogeneous dispersion of bacteria. (If the bottle is completely full of water, about 20-30 ml of water is discarded from the bottle prior to shaking. This ensure thorough mixing).

iii. With a sterile 10 ml pipette, 10 ml of the sample is inoculated into each of five tubes containing 10 ml of presumptive broth (double strength). It is advisable to shake the tubes gently to distribute the sample uniformly throughout the medium.

iv. The tubes are then incubated at 35^0C or 37^0C for 24 hours.

v. At the end of the 24 hour incubation period, each tube examined for the presence of gas. Gas, if present, can be seen in the durham tube ; if none is visible, the tube gently shaken. If any effervescence (streams of tiny bubbles) is observed, the tube should be considered positive.

vi. The number of positive tubes after 24 hours is recorded in a table.

vii. Negative tubes are kept in incubator for a further 24 hour period. At the end of this period, the tubes are again examined for gas production as in above. Gas production at the end of either 24 or 48 hours incubation is presumed to be due to the presence of coliforms in the sample.

viii. The number of positive tubes after 48 hours is recorded in the table.

ix. The confirmed test should be carried out at the end of both the 24 hour and the 48 hour incubation. Using a loop, one or two drops from each presumptive positive tube is transferred to a corresponding sterile confirmative 10-ml tube containing, e.g. BGB broth. Before each transfer, the inoculation loop is sterilised by flaming and is then allowed to cool.

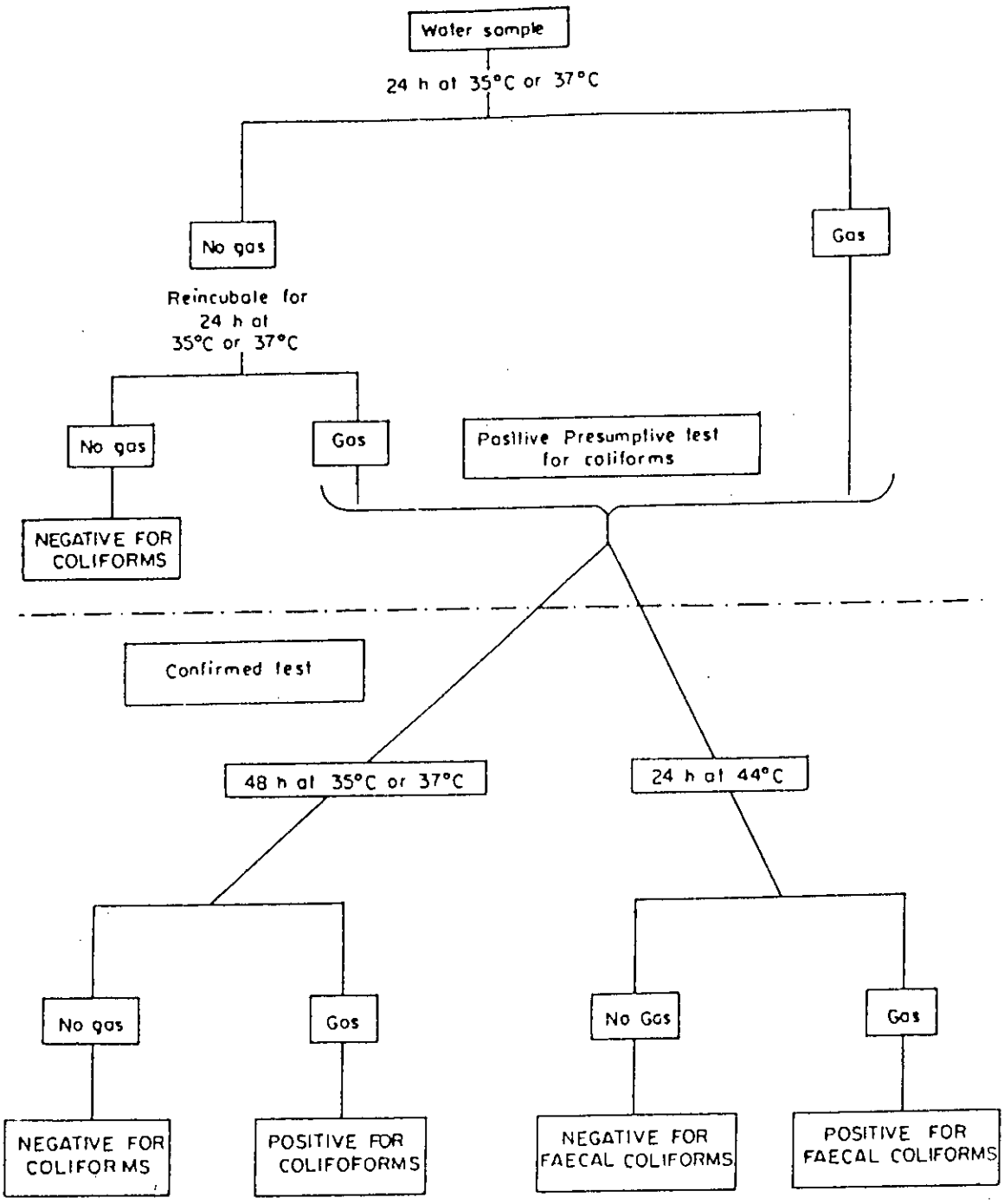


Figure 3.1 Procedures involved in presumptive and confirmed multiple tube tests.

x. If the presence of faecal coliforms is also to be investigated, subcultures in two tubes containing confirmative broth (e.g. BGB broth) should be prepared from each presumptive positive tube. EC medium is preferred in some areas for confirmation of faecal coliforms.

xi. To confirm the presence of coliforms, one subculture tube is incubated from each presumptive positive tube for 48 hours at 35^0C or 37^0C.

xii. The tubes are examined at the end of the 48 hour incubation period ; the presence of gas confirms that coliforms are present in the sample. The results recorded in the table.

xiii. To confirm the presence of faecal coliforms a second subculture tube is incubated form each presumptive positive tube for 24 hours at 44 ± 0.5^0C.

xiv. If at the end of 24 hours incubation, gas is found in the tubes, the presence of faecal coliforms is confirmed.

3.4.1.4 DETERMINATION OF MPN

The multiple-tube method allows one to estimate the density of presumed coliforms in the sample with an accuracy that increases, as do all statistical estimates, with the number of replicates examined. Results are expressed as MPN units per 100 ml, where MPN means the most probable number and reflects the statistical basis of the estimate. Only a series of replicants in which some tubes have positive and some tubes have negative results should be used to estimate numbers, since if all tubes are positive, one cannot know whether the initial number of cells placed in each tube was small or very large.

The statistical of the average number of coliforms per tube is based on the poission distribution.

$$P(X) = \frac{m^x e^{-m}}{X!}$$

Where P(X) is the probability of X, m is the mean, and X! is defined as 1 when X=0. In a dilution end- point assay, such as multiple-tube assay for coliforms, the average number of cells per tube can be calculated from the probability (frequency) of 0 cells per tube, i.e. from the frequency of negative tubes in a series of replicates giving both positive and negative results. In this case,

$$P(0) = e^{-m}$$

and m, the average number of coliforms per volume of sample used, is the natural logarithm of the fraction of negative tubes, P(0). Tables are available for the determination of MPN. For treated water, where five 10 ml portions are inoculated, the MPN can be found using Table 3.1.

Table 3.1 MPN of various combinations of positive and negative results when five 10-ml portions are used.

Number of tubes giving positive reaction out of 5 tubes of 10-ml each.	MPN
0	0
1	2.2
2	5.1
3	9.2
4	16.0
5	indeterminate

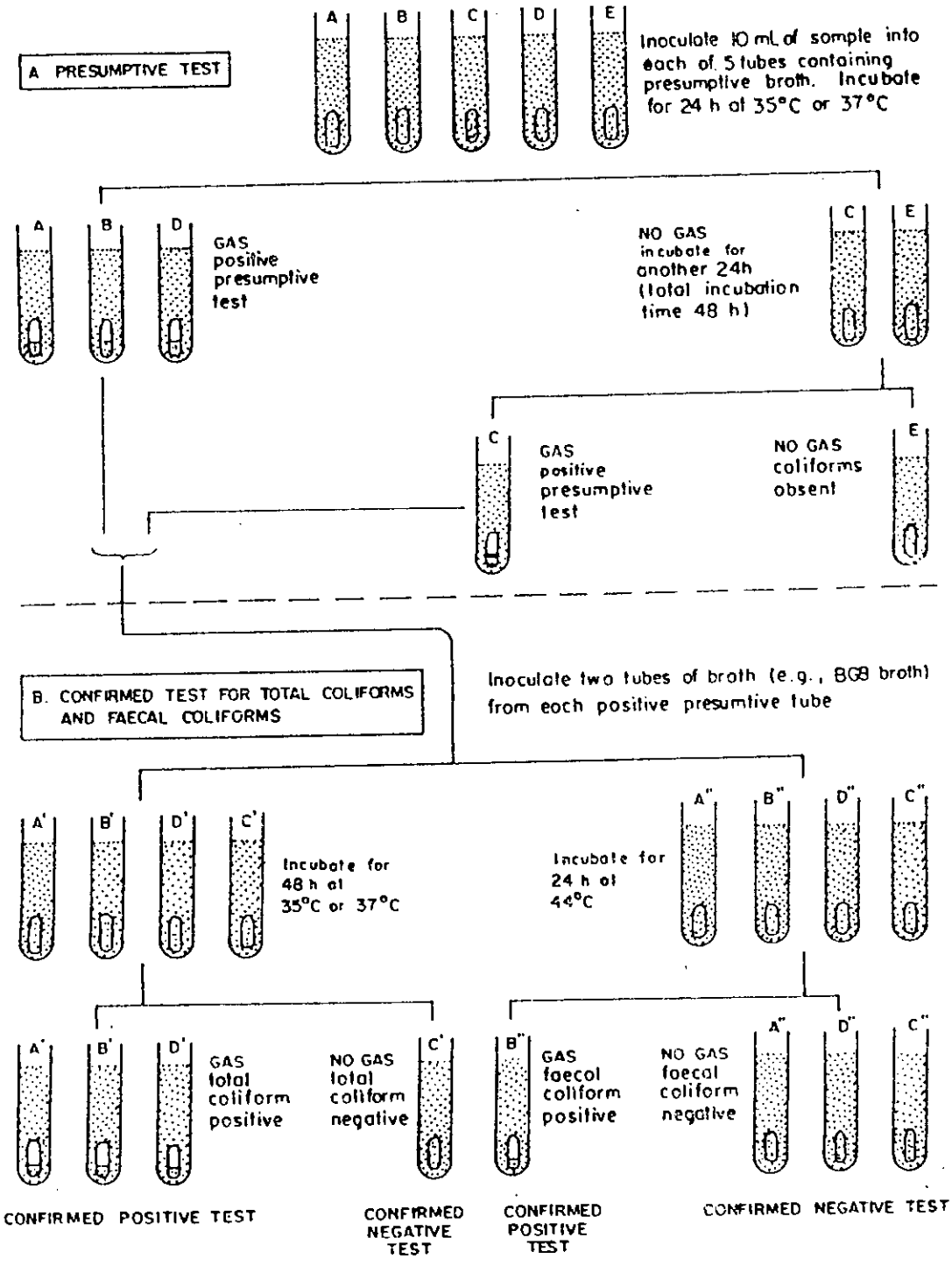


Figure. 3.2 Example of determination of total coliforms and faecal coliforms

An example is given in Figure 3.2 to show how the test results are interpreted. It will be seen that three confirmed positive tubes have been obtained for the total coliform group. From Table 3.1, the corresponding value of the MPN can be seen to be 9.2 total coliforms per 100 ml sample. As regards the test for faecal coliforms, there was only one confirmed positive tube. Consequently, the value of the MPN for this group is 2.2 per 100 ml.

3.4.2 MEMBRANE FILTER METHOD

With the membrane filter method, the number of coliform organisms in water is determined by filtering a measured volume of the sample, or an appropriate dilution of it, through a membrane filter, usually made of cellulose esters. Bacteria in the sample are retained on or near the surface of the membrane, which is then incubated face upwards on a suitable selective medium containing lactose. All acid or aldehyde producing colonies that develop on the membrane are counted either as presumptive coliform organism or as faecal coliform organisms, depending on the temperature of incubation. Since it is not possible to detect gas production or membranes, it is assumed that all colonies that produce acid or aldehyde also produce gas. Colonies are counted and the result expressed in number of colony present in 100 ml of original sample. It is usual to incubate 2 membranes for each sample, one at 35-37°C and the other at 44-44.5°C (WHO, 1984)

3.4.2.1 FIELD EQUIPMENT FOR MF METHOD

Equipments used for MF method are:

i. Incubator, ii) rack for incubation of petridishes, iii) filter funnel, 100 ml capacity, iv) porous support for filter, v) filter support, vi) suction vessel, vii) syringe with two way valve for providing the vacuum needed for the filtration, viii) sample vessel, ix) forceps, x) bottle containing alcohol, xi) bottle containing sterile buffer, xii) plastic petridishes , xiii) pipettes.

3.4.2.2 CULTURE MEDIA

Various media can be used for the examination of coliform organisms by the membrane filtration method. Of these lactose tergital agar, lactose TTC, tergital agar and lauryl sulphate lactose broth may be used for coliform counts at 35°C or 37°c and MF-C broth at 44.5°C for faecal coliform counts. Although all these media rely on the fermentation of lactose for the detection of presumptive coliform organisms, the characteristic reaction varies with each medium. The characteristic metallic sheen of colonies on Endo-type media depends on the formation of aldehyde.

Preparing Media :

Sterile Phosphate Buffer water

i. 34.0 gm of potassium dihydrogen phosphate (KH_2PO_4) is dissolved in 500 ml of distilled water.

ii. It is then diluted to 1,000 ml with distilled water to produce 1 litre of stock buffer solution. Stock buffer is stored in refrigerator and is discarded if it becomes turbid.

iii. 1.25 ml of stock buffer solution and 5 ml magnesium sulphate is added to one litre of distilled water to make 1 litre of phosphate buffer water.

iv. the solution is sterilised by filtering through a Millipore GS (0.22 μm pore size) membrane filter, or by autoclaving in loosely capped container at 121^0C for 15 minutes at 15 psi.

v. the solution is stored in tightly capped containers on a cool, dark shelf or in a refrigerator.

Preparation of stock MF-C Medium

i. 100 ml of distilled water is added to 3.7 gm of dehydrated MF-C medium in a 250 ml screw cap Erlenmeyer flask.

ii. In a separate flask, 100 ml of 0.2N NaOH solution is added to 1 gm of bact-rosolic acid dehydrate to produce a 1% rosolic acid solution. For smaller quantities, 0.1 gm rosolic acid is added to 10 ml of 0.2N NaOH.

iii. 1 ml of 1% rosolic acid solution added into the rehydrated MF-C broth.

iv. The medium is heated to the boiling point in a loosely covered flask ; and then promptly cooled to below 45^0C

v. Final pH is adjusted around 7.4

vi. The media is dispensed at room temperature. The unused portion is stored at 2-10^0C and is discarded after two weeks.

3.4.2.3 **TEST PROCEDURE**

Different types of filtration and equipment exist, and the test procedure varies with the types of units. The general procedure is described below.

Determination of Faecal Coliform (FC)

i) The Erlenmeyer (side-arm) flask is connected to the vacuum source (turned off) and the porous support is placed in position. If an electric pump is used, it is better to put a second flask between the Erlenmeyer and the vacuum source, this second flask acts as a water trap and thus protects the electric pump.

ii) A petridish is opened and a pad is placed on it .

iii) With a sterile pipette 2 ml of selective broth medium (MF-C broth) is added to saturate the pad.

iv) The filtration unit is assembled by placing a sterile membrane filter on the porous support, using forceps sterilized by flaming.

v) The upper container is placed in position and is secured with the special clamps,

vi) Optimal volume of sample is poured into the upper container. If the test sample is less than 10 ml, at least 20 ml of sterile dilution water should be added to the top container before filtration applying the vacuum.

vii) After the sample has passed through the filter, the vacuum filter is disconnected and the container is rinsed with 20-30 ml of sterile dilution water. The rinsing operation is repeated after all the water from the first rinse has passed through the filter.

viii) The filtration unit is dissembled and using the forcep, the membrane filter is placed in the petridish on the pad with the grid side up. Care should be taken so that no air bubbles are trapped between the pad and the filter.

ix) The petridish is inverted for incubation.

x) The petridish is incubated at 44 ± 0.5°C for 18-24 hours with 100% humidity. Alternatively, tight-fitting or sealed petridish may be placed in water-proof plastic bags for incubation. The bags are then submerged in a water-bath maintained at 44 ± 0.5°C or 24 hours. The plastic bags must be kept below the surface of the water throughout the incubation period. They can be held down by means of a suitable weight, e.g. a metal rack.

3.4.2.4 COLONY COUNTING

Colonies of faecal coliform bacteria are of blue colour. This colour may cover the entire colony, or appear only in the centre of the colony. Colonies of other

colours should not be counted. The colonies can be counted with the aid of a lens, the number of faecal coliforms per 100 ml is then given by :

$$\text{Faecal coliform per 100 ml} = \frac{\text{No. of faecal coliform colonies counted} \times 100}{\text{ml of sample filtered}}$$

3.4.3 Reasons For Selecting Membrane Filter Method

Since its inception, membrane filter method has gained world wide acceptance because of its high degree of reproducibility, its ability of testing relatively larger volume of sample and for the savings in time to gain definite result.

The national training center of the U.S. Environmental Protection Agency (EPA) cited the following advantages.

- Results are obtained in approximately 24 hours, as compared with 48-96 hours for the standard fermentation tube method.

- Much larger, and hence more representative, samples of water can be sampled routinely with membrane filters.

- Numerical results from membrane filters have much greater precision (reproducibility) than is expected with the fermentation tube method.

- The equipment and supplies required are not bulky. A great many samples can be examined with minimum requirements for laboratory space, equipment and supplies.

- The MF technique costs about half as much as the MPN method per test. This means considerable saving to any laboratory routinely performing coliform analyses.

- It is the only approved coliform test that lends itself to field testing

- The alternative MPN method requires laboratory facilities for washing, autoclaving and incubation of many fermentation tubes, besides requiring many litres of culture media.

For the above mentioned advantages, the membrane filter method was selected for faecal coliform analysis.

3.5 Removal Of Suspended Materiel By Coagulation And Filtration

Kaolin solution of 400 mg/l, 1000 mg/l, 2000 mg/l, 3000 mg/l and 4000 mg/l were prepared. pH of the samples were maintained at 6.0. Alum of 5 mg/l, 10 mg/l, 15 mg/l, 20 mg/l, 25 mg/l, 50 mg/l, 125 mg/l and 250 mg/l were added in the samples containing kaolin solution of 400 mg/l and 1000 mg/l. 10 mg/l, 25 mg/l, 50 mg/l, 125 mg/l and 250 mg/l were added in remaining sample. Samples containing different alum dosages were stirred rapidly (80 rpm) for about 1 minute and then slowly for about 30 minutes and then allowed to settle for about 30 minutes. After 30 minutes sedimentation samples from the top were taken for turbidity determination. After coagulation and sedimentation samples containing kaolin concentration of 400 mg/l and 1000 mg/l were filtered by filter paper and residual turbidity after filtration were measured.

Samples of 400 mg/l and 1000 mg/l were taken in different beakers and pH were maintained at 4.5. Alum in 25 mg/l, 50 mg/l, 125 mg/l and 250 mg/l were added. After rapid and slow mixing samples were allowed to settle for about 30 minutes. After 30 minutes sedimentation residual turbidity were measured. Then filtration with filter paper were performed and then turbidity were measured by turbidimeter.

3.6 Removal Of Faecal Coliform at DIFFERENT pH And Sedimentation Time WITHOUT USING ALUM

500 ml of water sample of pH 6.8 were taken in six beakers. 1 ml of stock cell suspension of faecal coliform, described in section 3.3, were added in each beaker and stirred with stirrer. One beaker was used to determine the initial concentration of microorganism. Hydrochloric acid and sodium hydroxide were added to make sample of pH 3.6, 5.0, 5.6, 8.2 and 8.8. After stirring samples were allowed to settle for about 30 minutes. After sedimentation 10 ml sample from top were taken by pipette for microbial detection. Membrane filter technique was used for this purpose.

One beaker containing sample of pH 3.6 was observed over 24 hours. Initial concentration of faecal coliform was measured at 8.30 a.m. and faecal coliform population determination continued upto 9 p.m. and last reading was taken after 24 hours at 9 a.m. of the following day.

3.7 Removal Of Faecal Coliform By Coagulation And Filtration

500 ml water sample were taken in three beakers. 1 ml of stock cell suspension of faecal coliform were added in each beaker and stirred. Initial concentration of faecal coliform were counted. To determine the effect of sedimentation time on removal of faecal coliform, samples were maintained at pH 4.5 and 6.0. Different studies (mentioned in literature review) showed that at pH 4.5 maximum +ve Al species formed and at pH 6.0 maximum $Al(OH)_3$ formed. As volume of $Al(OH)_3$ is 50 times larger than clay particles. Therefore, rapid adsorption and sedimentation were resulted at pH 6.0 and charge neutralization were occured at pH 4.5. For this reason this study was emphasized on pH 4.5 and 6.0 only. 25 mg/l alum dosage were added in two beakers. Samples were stirred rapidly (80 rpm) for 1 minute and then slowly (20 rpm) for 30 minutes and allowed to settle for about 30 minutes. Both the samples were observed over 8 hours. First reading after sedimentation were taken at 10.00 am. and continued upto 6.0 p.m. Sample of pH 6.0 were

preserved upto 48 hours and faecal coliform were determined at 10:00 a.m. of following day and 10:00 a.m. of two days after sample preparation.

Moreover five beakers containing 500 ml distilled water were taken and stock cell suspension of 1 ml were added in each beaker. Initial concentration of faecal coliform were counted. Hydrochloric acid were added in four beakers to make samples of pH 6.0. Alum dosage in different concentration determined by jar test were added in different beakers. Samples were stirred rapidly (80 rpm) for 1 minute and then slowly (20 rpm) for 30 minutes and allowed to settle for 1 hour. After 1 hour sedimentation 10 ml from top of the sample were taken for faecal coliform detection. After coagulation samples were filtered by filter paper. 10 ml from each sample were drawn for microbial determination.

Again, five beakers were taken for preparation of sample with 500 ml distilled water and 1 ml stock cell suspension. Initial concentration of faecal coliform microorganism were determined. pH of the samples were maintained at 6.0. 25 mg/l, 50 mg/l 125 mg/l and 250 mg/l alum dosage were added in four beakers. Samples were stirred rapidly (80 rpm) for 1 minutes and slowly (20 rpm) for 30 minutes and sedimentation observed over 8 hours. After some interval bacterial concentration were counted. From this, coagulation condition were found at different alum dosage and different sedimentation time.

3.8 Removal Of Faecal Coliform And Suspended Material By Coagulation And Filtration At pH 6.0.

Kaolin solution of 1000 mg/l and 400 mg/l were prepared. About 500 ml distilled water were taken in one beaker and microbial concentration were counted. 500 ml of sample containing 1000 mg/l kaolin solution were taken in 5 beakers. 1 ml of sewage was added in each beaker and stirred. One beaker was used for determination of initial turbidity. Hydrochloric acid were added to make sample of pH 6.0. Then alum dosage of 25 mg/l, 50 mg/l, 125 mg/l and 250 mg/l were added in four samples. Samples stirred rapidly (80 rpm) for 1 minute and then

slowly (20 rpm) for 30 minutes and then allowed to settle for 30 minutes. After 30 minutes sedimentation 10 ml samples from each beaker were taken by pipette for faecal coliform detection and then turbidity were measured for each sample. Filtration was done by filter paper and turbidity were measured.

Same procedure was followed for samples containing kaolin concentration of 400 mg/l.

CHAPTER - 4
RESULTS AND DISCUSSIONS

4.1 INTRODUCTION

This study was intended to investigate the effectiveness of coagulation for the treatment of highly turbid and bacteriologically polluted water. Three types of sample were prepared for this purpose i) with kaolin ii) with microorganism iii) with both kaolin and microorganism. Coagulation depends on various factors such as pH, turbidity, chemical composition of water, type of coagulant, temperature and mixing conditions.

In this study an effort was made to determine the effect of various parameters, on removal of faecal coliform and suspended material. The parameters considered were Alum dosage, pH, Sedimentation time, Suspended Solids and Mixing time.

Filtration was a part of this study. Filtrations were performed after the coagulation and sedimetation with filter paper. Moreover, removal of faecal coliform was observed at different pH and sedimentation time without alum.

4.2 REMOVAL OF SUSPENDED MATERIALS BY COAGULATION AND FILTRATION

Two observations were done:

i. Effect of alum dosage on turbidity at pH 6.0
ii. Effect of alum dosage on turbidity at pH 4.5

4.2.1 EFFECT OF ALUM DOSAGE ON TURBIDITY AT PH 6.0

500 ml sample of kaolin concentration 400 mg/l, 1000 mg/l, 2000 mg/l, 3000 mg/l and 4000 mg/l were taken in different beakers. Hydrochloric acid were added in each beaker to make water sample at pH 6.0. Initial turbidity of all kaolin concentration were measured by turbidimeter. Initial turbidity of samples

containing kaolin concentration 400 mg/l, 1000 mg/l 2000 mg/l, 3000 mg/l and 4000 mg/l were 182, 445, 520, 650 and 814 NTU. After 30 minutes sedimentation turbidity of the samples were measured and these were 55, 112, 140, 175 and 210 NTU respectively.

5 mg/l, 10 mg/l, 15 mg/l, 20 mg/l, 25 mg/l, 50 mg/l, 125 mg/l and 250 mg/l alum dosage as determined by Jar test were added in different beakers containing kaolin concentration of 400 mg/l and 1000 mg/l. Each sample containing alum were stirred rapidly (80 rpm) for 1 minute and then slowly (20 rpm) for 30 minutes and then allowed to settle for 30 minutes. After 30 minutes sedimentation turbidity of each sample were measured. For each sample three enumerations were done and mean of these were taken. The results are shown in Figure 4.1. Moreover, 10 mg/l, 25 mg/l, 50 mg/l, 125 mg/l and 250 mg/l alum were added in the samples of kaolin concentration 2000 mg/l, 3000 mg/l and 4000 mg/l. After rapid mixing and slow mixing samples were allowed to settle for 30 minutes. After sedimentation turbidity were measured. The results are shown in Figure 4.2. A summary of results showing residual turbidity with varying alum dosage for different kaolin concentration are given in table 4.1

Table 4.1: Residual turbidity with variation of alum dosage at pH 6.0 after coagulation and sedimentation for samples at different kaolin concentration

Alum dosage (mg/l)	Residual turbidity (NTU)				
	Initial Kaolin concentration 400 mg/l	Initial Kaolin concentration 1000 mg/l	Initial Kaolin concentration 2000 mg/l	Initial Kaolin concentration 3000 mg/l	Initial Kaolin concentration 4000 mg/l
0	55	112	128	149	162
10	20	24	13.3	12.5	11
25	9.3	16.2	11	10.2	9.4
50	8.1	13.2	9.2	8.7	8.4
125	5.5	9.1	8.8	8.1	7.5
250	5.2	8.2	8.0	7.3	6.8

Fig-4.1 Effect of Alum Dosages on Turbidity after coagulation and sedimentation at pH 6.0

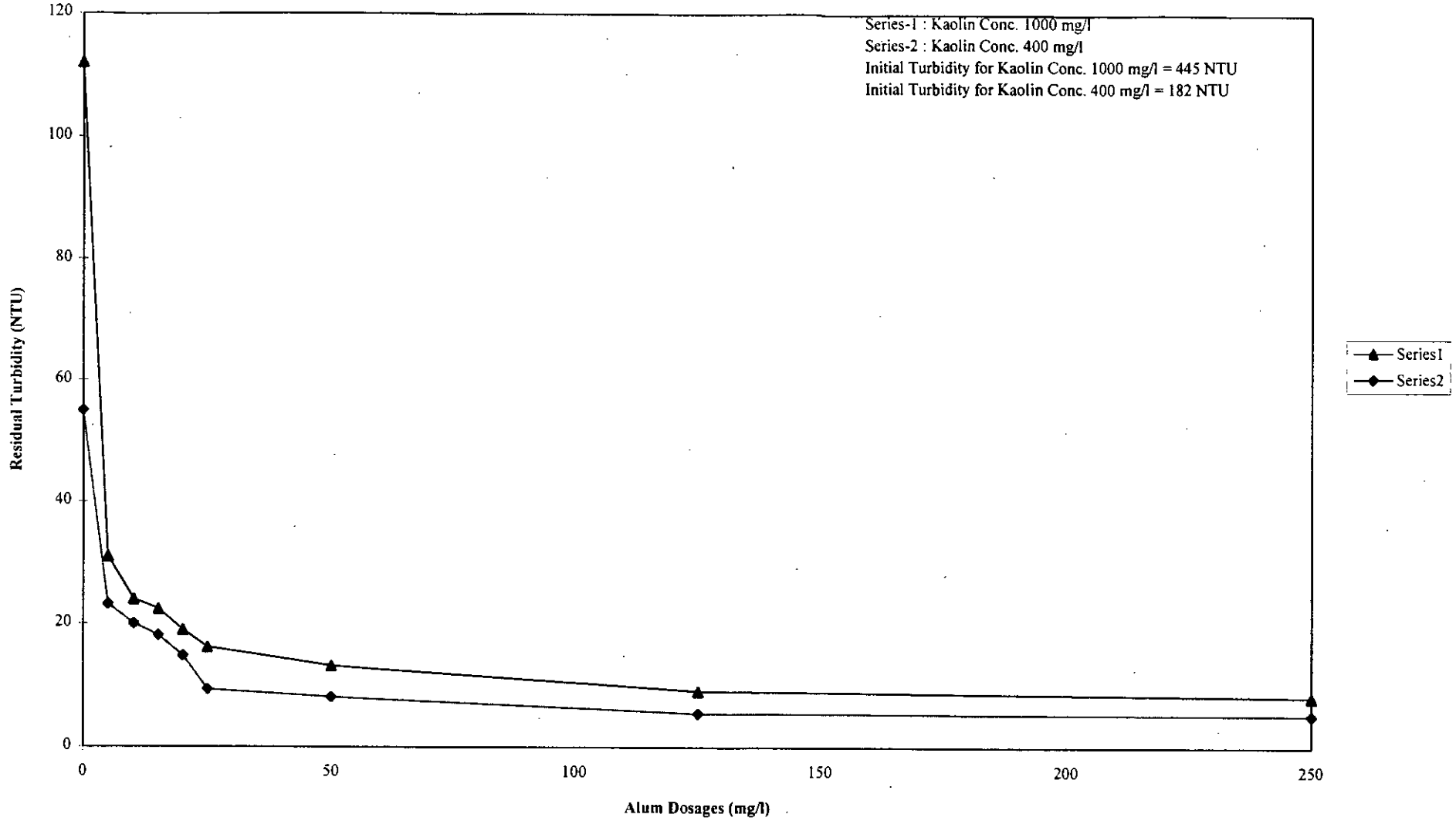
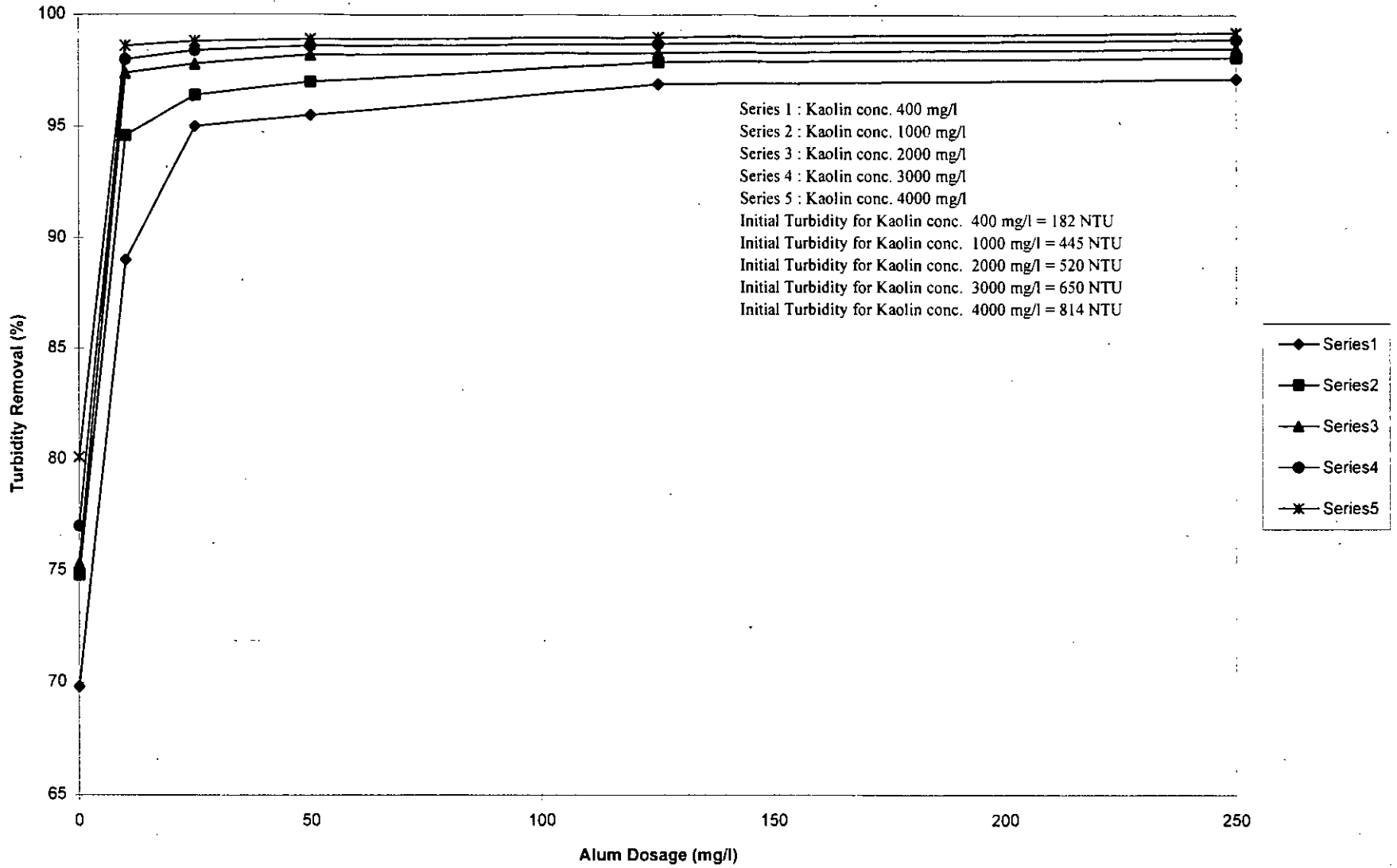


Fig.4.2 Effect of Alum Dosage on Removal of Turbidity after Coagulation and Sedimentation at pH 6.0



From Figure 4.1 it is observed that the residual turbidity decreases with increasing alum dosages. At low dosage the rate of removal of turbidity is more and rate of removal is decreasing with increasing dosage of alum. After 30 minutes sedimentation turbidity removal were 74.8% and 69.8% for kaolin concentration 1000 mg/l and 400 mg/l respectively. By addition of 5 mg/l alum turbidity removal were 93% and 87.2% for kaolin concentration 1000 mg/l and 400 mg/l respectively. Moreover, with addition of 250 mg/l alum, turbidity removal were 98.1% and 97.14% respectively. From table 4.1 it is found that at 400 mg/l kaolin concentration minimum residual turbidity obtained by addition of 250 mg/l alum dosages. But for higher kaolin concentration minimum residual turbidity cannot be obtained. Therefore it can be said that more alum dosages are required for higher kaolin concentration to attain minimum residual turbidity. From figure 4.2 it is found that optimum alum dosage for kaolin concentration 400 mg/l, 1000 mg/l, 2000 mg/l, 3000 mg/l and 4000 mg/l are 25 mg/l, 25mg/l, 10mg/l 10mg/l and 10 mg/l respectively.

Allowable limit for Bangladesh drinking water standard is 10 NTU. For initial kaolin concentration of 4000 mg/l, 3000 mg/l, 2000 mg/l, 1000 mg/l and 400 mg/l the alum dosages (without filtration) required to attain Bangladesh water standard limit are 18 mg/l, 26 mg/l, 50 mg/l, 105 mg/l and 24 mg/l respectively (not shown in the text).

Samples of kaolin concentration 400 mg/l and 1000 mg/l were filtered by filter paper after coagulation and sedmentation. The results are shown in Table A.2. From Table A.2 it is found that small amount of kaolin were retained in the sample. These results might vary if filter media could be used and need more studies in this area.

4.2.2 Effect Of Alum Dosage On Turbidity At pH 4.5

500 ml sample of kaolin concentration 1000 mg/l were taken in five beakers and 500 ml sample of kaolin concentration 400 mg/l were taken in another five

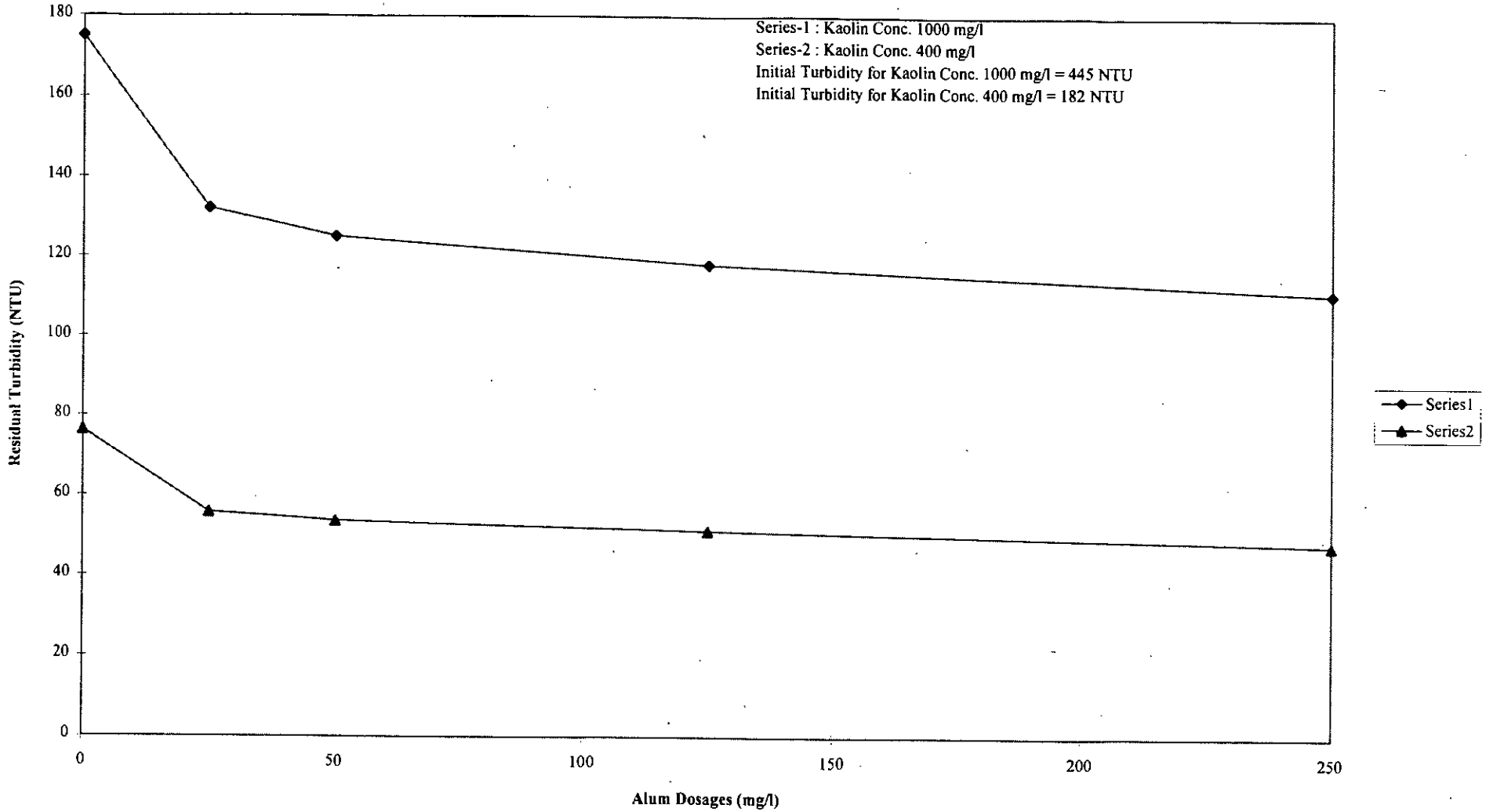
beakers. pH of the samples were maintained at 4.5. 25 mg/l, 50 mg/l, 125 mg/l and 250 mg/l alum dosage were added in four beakers containing kaolin concentration of 1000 mg/l. Samples were stirred rapidly (80 rpm) for 1 minute and then slowly (20 rpm) for 30 minutes and then allowed to settle for 30 minutes. Turbidity of the samples were measured. For kaolin concentration 400 mg/l similar procedure was followed. Initial turbidity of sample containing kaolin concentration of 1000 mg/l and 400 mg/l were 445 NTU and 182 NTU respectively. After 30 minute sedimentation, turbidity of both the samples were measured and these were 175 NTU and 76.4 NTU respectively. Alum in varying concentration were added in the samples. The results after coagulation and 30 minutes sedimentation are shown in Figure 4.3 and Table A.3.

From the Figure 4.3 it is found that by addition of 25 mg/l alum turbidity removal for kaolin concentration 1000 mg/l and 400 mg/l were 70.3% and 69.3% respectively. By addition of 250 mg/l alum turbidity removal for kaolin concentration 1000 mg/l and 400 mg/l were 75.0% and 73.6%.

Comparing two curves in figure 4.3 it can be said turbidity decreases with increase of alum dosage but removal rate of turbidity for kaolin concentration 1000 mg/l is higher than 400 mg/l. After coagulation and sedimentation samples were filtered by filter paper. The results are shown in Table A.4. From Table A.4 it is found that most of the kaolin particles are removed by filtration after coagulation.

From results of section 4.2.1 and 4.2.2 table 4.2 and 4.3 can be drawn. From table 4.2 and 4.3 it is seen that turbidity removal efficiency is higher in higher concentration of kaolin than lower concentration. After 30 minute sedimentation, 75.0% and 69.8% turbidity were removed for initial kaolin concentration 1000 mg/l and 400 mg/l at pH 6.0. The reason may be that in highly turbid water, collision probability increase therefore floc formation

Fig-4.3 Effect of Alum Dosage on Turbidity after coagulation and sedimentation at pH 4.5



increase. Another reason may be that some particles settle by sedimentation. Moreover, in the higher turbidity concentration the ratio of aluminium or aquo-aluminium complex which react directly with functional groups on the clay becomes slightly higher than the lower concentration suspension and the efficiency of coagulant dosage ration is improved. The same phenomenon had been observed by Tambo, (1990).

Table 4.2: Removal of turbidity at pH 4.5 and 6.0 at different alum dosage for initial kaolin concentration 1000 mg/l

Alum dosage (mg/l)	% removal of turbidity	
	At pH 6.0	At pH 4.5
0	75	60.0
25	96.4	70.3
50	97.0	71.9
125	97.9	73.5
250	98.1	75.0

Table 4.3: Removal of turbidity at pH 4.5 and 6.0 at different alum dosage for initial kaolin concentration 400 mg/l

Alum dosage (mg/l)	% removal of turbidity	
	At pH 6.0	At pH 4.5
0	69.8	58.0
25	95.0	69.3
50	95.5	70.5
125	96.9	71.8
250	97.14	73.6

From table 4.2 and 4.3 it is found that turbidity removal at pH 6.0 is higher than pH 4.5. From the study of Sullivan and Singley, (1968), it is found that at pH 6.0, 95% of Al species is $Al(OH)_3$ whether at pH 4.5 $Al(OH)_3$ is only 3%. From the study of Hossain, (1996), it is found that maximum +ve Al species form at pH 4.5. Therefore, it can be said from this study that the removal of kaolin at pH 6.0 may be due to adsorption of kaolin on $Al(OH)_3$. At pH 4.5 the removal may be mostly for charge netralization and slightly for adsorption.

4.3 Removal Of Faecal Coliform at different pH And Sedimentation Time without using alum.

Removal of faecal coliform were observed in two conditions.

i) Variation of pH

ii) Variation of sedimentation time

4.3.1 Removal Of Faecal Coliform With Variation Of pH

The samples were prepared by mixing sewage of 1 ml from stock cell suspension with 500 ml of distilled water in six beakers. Initial concentration of faecal coliform were calculated from one beaker. To determine the effect of pH on removal of faecal coliform the removal were observed at pH level 3.6, 5.0, 5.6, 8.2 and 8.8.

Hydrochloric acid and sodium hydroxide were added in the sample to maintain required pH. After stirring samples were allowed to settle for 30 minutes. After 30 min sedimentation 10 ml sample were taken from each beaker for faecal coliform determination. The initial concentration of faecal coliform was 39600/100ml.

Table 4.4 : Removal of faecal coliform at different pH without coagulation

pH	Faecal coliform 100 ml	Removal of faecal coliform (%)
3.6	22470	43
5.0	38660	2.3
5.6	39000	1.5
8.2	39240	0.9
8.8	38880	1.8

From table 4.4 it is observed that the removal of faecal coliform at pH 3.6 was 43%. Removal of faecal coliform in other pH, were not significant. At pH 5.0 removal of faecal coliform was 2.3% whereas in strong alkaline environment (at

pH 8.8) the removal was 1.8%. Similar observations were performed by Alam, (1996). It may be said that faecal coliform decay rate in acidic environment is higher than alkaline environment.

4.3.2. Removal Of Faecal Coliform With Variation Of sedimentation Time At pH 3.6

To determine the effect of sedimentation time on removal of faecal coliform, the removal for sample of pH 3.6 (as stated in section 4.3.1) were observed for 24 hours. The initial concentration of faecal coliform was 39600/100 ml. After 24 hours the concentration reduced to 4/100 ml. The removal of faecal coliform was 99.9%.

Table 4.5: Removal of faecal coliform with sedimentation time at pH 3.6 without coagulation

Time (hr)	No. of Faecal coliform/100 ml	% Removal of Faecal coliform
1/2	22470	43
1	17120	57
3/2	14980	62
2	11770	70
4	4500	88
6	1500	96
8	1300	97
12	300	99.2
24	4	99.9

The removal rate of faecal coliform showed two distinct phase; the initial phase when the faecal coliform population reduced rapidly upto 2 hours then reduced slowly. In initial phase the faecal coliform removal was 70.0% and remaining 29.9% faecal coliform was removed in subsequent 22 hours. Most of the organism die within short period in acidic environment at pH 3.6.

4.4 Removal Of Faecal Coliform By Coagulation And Filtration

Three observations were performed to determine the removal of faecal coliform by sedimentation and coagulation.

i. Effect of sedimentation time on removal of faecal coliform at particular alum dosage at pH 4.5 and 6.0.
ii. Effect of alum dosage on removal of faecal coliform by sedimentation and filtration at pH 6.0.
iii. Effect of sedimentation time on removal of faecal coliform at different alum dosage at pH 6.0.

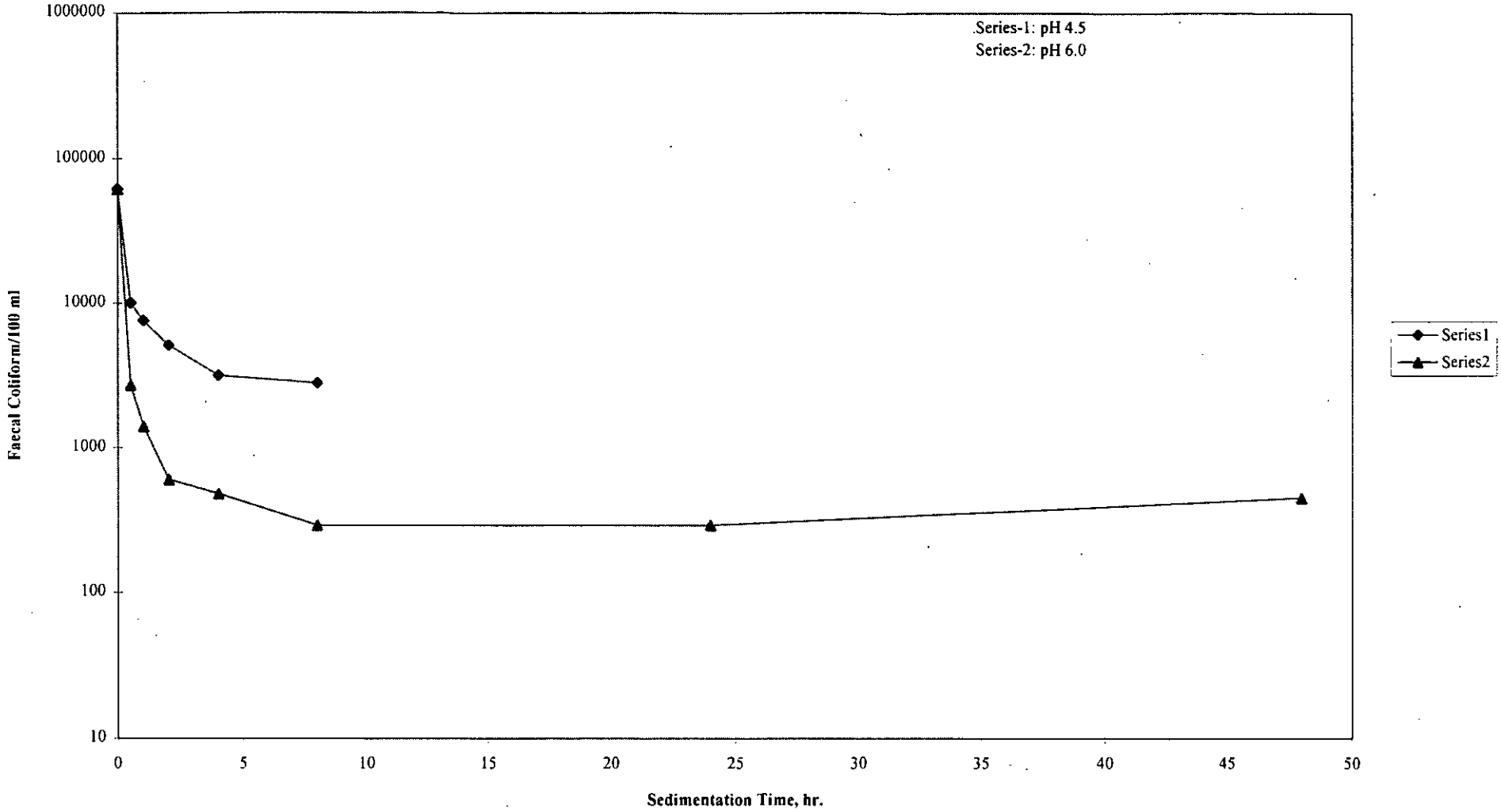
4.4.1 Effect Of Sedimentation Time On Removal Of Faecal Coliform At Particular Alum Dosage At pH 4.5 And 6.0.

500 ml water sample were taken in three beakers. 1 ml of stock cell suspension was added in each beaker and stirred. Initial concentration of microorganism were calculated. Hydrochloric acid were added to make sample at pH 4.5 and 6.0. 25 mg/l alum dosage were added in two sample. Samples were stirred rapidly (80 rpm) for 1 minute and then slowly (20 rpm) for 30 minutes and then allowed to settle for 30 minutes. After 30 minute sedimentation 10 ml from both the samples were taken for faecal coliform determination. Faecal coliform concentration of the sample at pH 4.5 were counted upto 8 hours thereafter samples were drained off. But sample at pH 6.0 were maintained for 2 days.

Initial concentration of faecal coliform for both the samples were 61700/100ml. Results at different sedimentation time are shown in Figure 4.4 and Table A.5.

From Figure 4.4 it is found that at pH 6.0 number of faecal coliform after 30 minute sedimentation was 2700/100 ml and number of faecal coliform after 8 hours sedimentatin was 290/100 ml. Therefore, 95.6% faecal coliform was

Fig.4.4 Effect of sedimentation time on removal of faecal coliform at 25 mg/l alum dosage at pH 4.5 and 6.0



removed by coagulation and 30 minutes sedimentation and 99.5% faecal coliform was removed by coagulation and 8 hours sedimentation. At pH 4.5, number of faecal coliform after 30 min sedimentation was 10000/100 ml and after 8 hours sedimentation number of faecal coliform was 2800/100 ml. 83.8% faecal coliform was removed by coagulation and 30 minutes sedimentation and 95.5% faecal coliform was removed by coagulation and 8 hours sedimentation.

From Figure 4.4 it is also found that at pH 4.5 number of faecal coliform decreases with increase of sedimentation time but upto 4 hours sedimentation substantial amount of faecal coliform were removed, then rate of removal decreases with increase of sedimentation time. At pH 6.0 number of faecal coliform decreases upto 8 hours sedimentation then number of faecal coliform increases.

It is apparent from two curves in Figure 4.4 that faecal coliform removal rate at pH 6.0 is higher than pH 4.5. At pH 6.0 the Al species are mostly $Al(OH)_3$, (Sullivan and singley, 1968 and Hossain,1996). The volume of $Al(OH)_3$ is 50 times larger than the same quantity of clay particles (Matsui and Tambo, 1989) as mentioned earlier. Large number of microorganisms were adsorbed on $Al(OH)_3$ mesh and settled down. But at pH 4.5 maximum +ve Al formed and small amount of $Al(OH)_3$ formed as mentioned earlier in the work of Sullivan and Singley (1968). Small amount of organism may adsorb in the $Al(OH)_3$ mesh and charge neutralization may takes place at pH 4.5. More studies are needed on the behavior of charge of organism.

Moreover, at pH 6.0 curve initially dropped abruptly, then slowly moved downward and then again moved upward. Settlement of $Al(OH)_3$ is faster in initial stage and then become slower. The reason for upward movement of the curve is that microorganism may regrowth or microorganism comes out from sediment of the sample with sedimentation time.

4.4.2 Effect Of Alum Dosage On Removal Of Faecal Coliform By Sedimentation And Filtration at pH 6.0

500 ml of water sample were taken in five beakers and stock cell suspension of 1 ml were added in each beaker. Initial concentration of microorganism were determined. Hydrochloric acid were added to make pH of 6.0. 25 mg/l, 50 mg/l, 125 mg/l and 250 mg/l alum dosage as selected by jar test were added in four beakers. Samples were stirred rapidly (80 rpm) for 1 minute and then slowly (20 rpm) for 30 minutes and then allowed to settle for 1 hour. After 1 hr. sedimentation 10 ml sample from top of each beaker were taken for faecal coliform determination. Initial concentration of faecal coliform was 57600/100 ml. The results obtained from coagualtion and sedimentation are shown in Figure 4.5 and Table A.6

From figure 4.5 it is found that by addition of 25 mg/l alum dosage faecal coliform reduced to 900/100 ml i.e. 98.4% faecal coliform was removed. By addition of 50 mg/l alum dosage faecal coliform population reduced to 500/100 ml which means that 99% faecal coliform was removed. By addition of 125 mg/l alum dosage 99.8% faecal coliform was removed. One faecal coliform colony was found in the filter paper where samples were treated by 250 mg/l alum dosage. Therefore number of faecal coliform remained in the sample was 10/100 ml.

From figure 4.5 it can also be seen that alum dosage increases faecal coliform removal rate increases. Samples were maintained at pH 6.0. At pH 6.0 maximum $Al(OH)_3$ formed which gave large volume of mesh where microorganisms were adsorbed in large quantity and settled down.

Samples after Coogulation and 1 hour sedimentation were filtered by filter paper. From figure 4.5 it is found that number of faecal coliform remained in the sample were 11, 10 and 2 per 100 ml where coagulation were done previously

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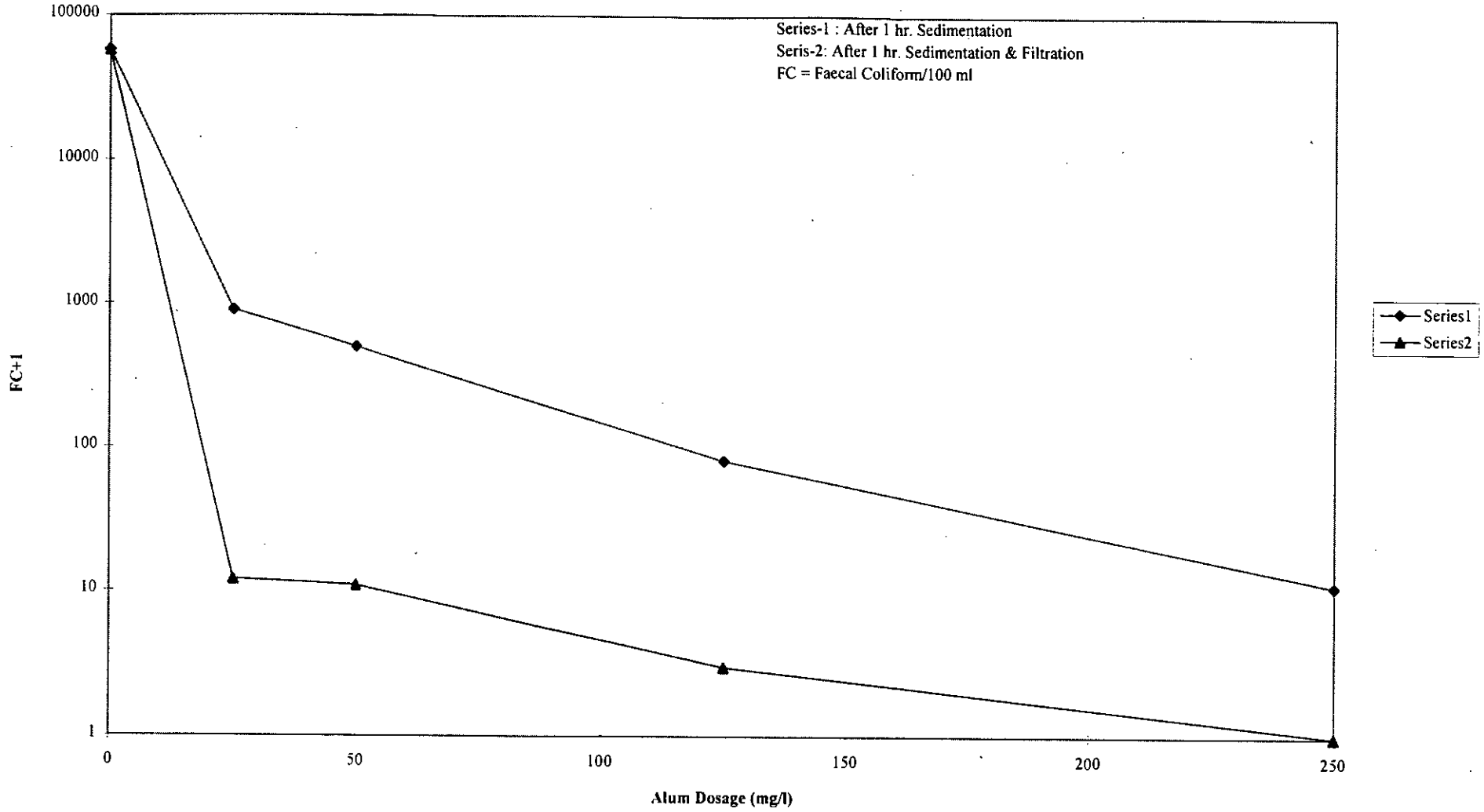
Fig. 4.5 Effect of Alum Dosage on removal of Faecal Coliform by Sedimentation & Filtration at pH 6.0



by addition of 25 mg/l, 50 mg/l and 125 mg/l alum dosage respectively. But no faecal coliform was found in the filter paper for sample treated with 250 mg/l alum dosage.

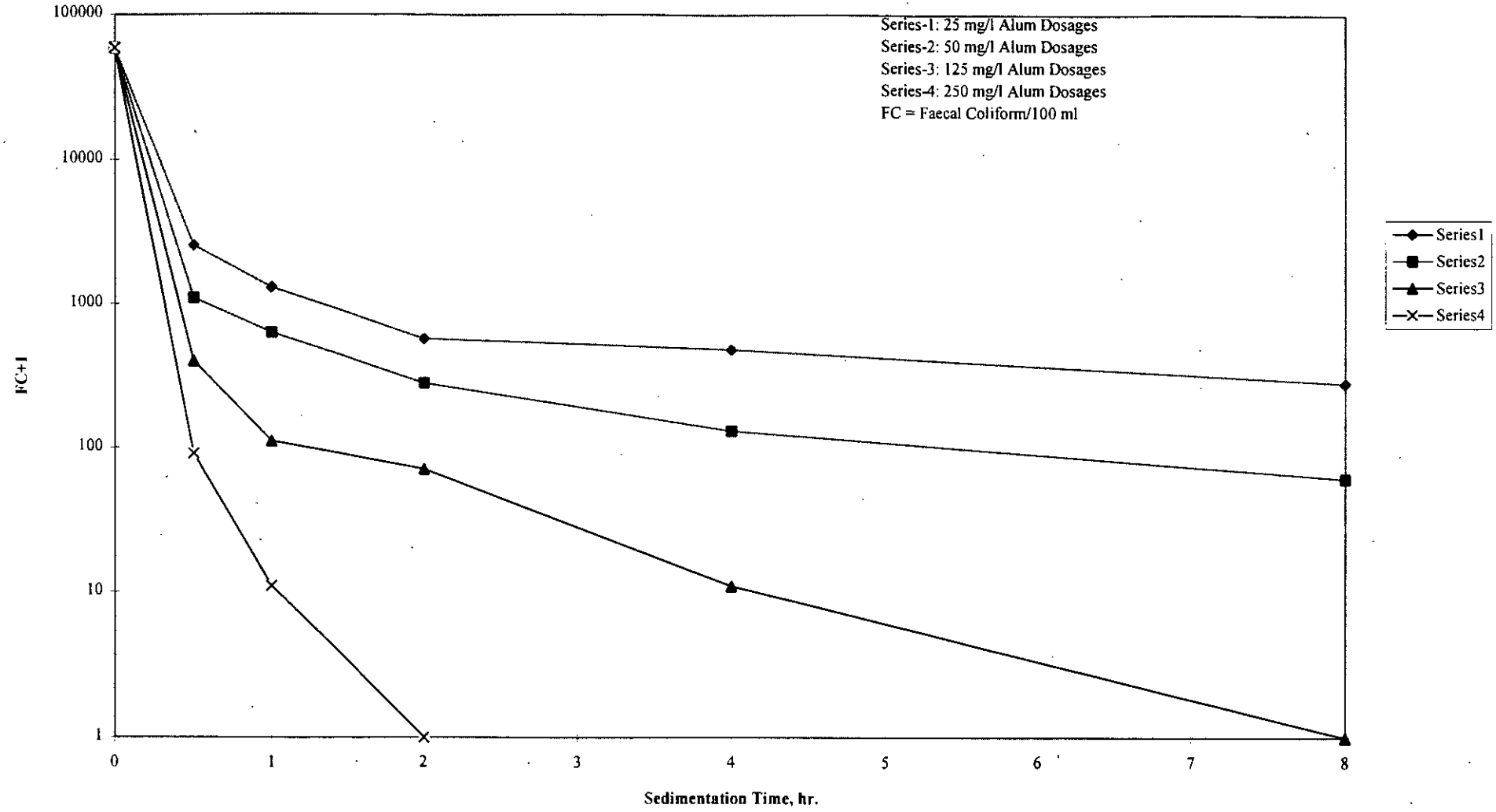
4.4.3 Effect Of Sedimentation Time On Removal Of Faecal Coliform At Different Alum Dosage At pH 6.0.

The samples were prepared by mixing sewage (1ml) from stock cell suspension with 500 ml of distilled water in five beakers. Initial concentration of faecal coliform were determined from one beaker. pH of the samples were maintained at 6.0. Jar test were performed with different alum dosage with rapid mix at 80 rpm for 1 minute and slow mix at 20 rpm for 30 minutes. Residual faecal coliform determination were performed over 8 hours sedimentation period. Initial concentration of faecal coliform was 59000/100 ml. The results after coagulation and sedimentation are shown in Figure 4.6 and Table A.7.

From figure 4.6 it is found that all the curves drop suddenly by addition of alum dosage and then become flatter. From curve 1 it is observed that by addition of 25 mg/l alum dosage faecal coliform removal rate upto 2 hours is significant and then become mostly constant. By addition of 25 mg/l alum dosage and 8 hours sedimentation number of faecal coliform reduced to 280/100 ml i.e. percentage removal of faecal coliform was 99.5.

From curve 2 it is found that upto 4 hours sedimentation period faecal coliform removal rate is higher and then become slower. 99.8% faecal coliform was removed by addition of 50 mg/l alum and 8 hours sedimentation. From curve 3 in Figure 4.6 it is found that by addition of 125 mg/l alum, faecal coliform retained after 4 hours sedimentation was 10/100 ml but no faecal coliform was found after 8 hours sedimentation. From curve 4 it is seen that within 2 hours all the faecal coliform was removed by addition of 250 mg/l alum dosage.

Fig. 4.6 Effect of Sedimentation Time on removal of Faecal Coliform at Different Alum Dosage at pH 6.0



From figure 4.6 it is observed that as alum dosage increases removal rate of microorganism increases and at any alum dosage removal of faecal coliform increases with increasing sedimentation time. Moreover, for the same removal of microorganism less sedimentation time required for increased alum dosage. Hossain, (1996) and Sullivan and Singley, (1968) found that maximum Al species form at pH 6.0 is $Al(OH)_3$. And Matsui and Tambo, (1989) observed that the size of precipitated aluminium species is larger than 1μm and the volume of 1 mg of aluminium hydroxide is about 50 times greater volume than 1 mg of clay particles. As alum dosage increases the amount of $Al(OH)_3$ i.e. adsorption of microorganism increases. Moreover with increasing sedimentation time settlement rate is decreasing. Therefore with increasing alum dosage potable water can be obtained within snort period of sedimentation.

4.5 Removal Of Faecal Coliform and Suspended Material By Coagulation And Filtration at pH 6.0

Kaolin solution of 1000 mg/l and 400 mg/l were prepared. Water sample of 500 ml were taken in one beaker. Sewage (1 ml) from stock cell suspension were added and then faecal coliform concentration were counted and found 47600/100 ml. 500 ml of sample containing 1000 mg/l and 400 mg/l kaolin solution were taken in ten beakers and 1 ml of sewage were added in each beaker. After stirring two beakers of ditterent kaolin solution were used to determine initial turbidity. pH of all the samples were maintained at 6.0. Alum dosage of 25 mg/l, 50 mg/l, 125 mg/l and 250 mg/l were added in four beakers of 1000 mg/l kaolin solution and another four beakers of 400 mg/l kaolin solution. Samples stirred rapidly (80 rpm) for 1 minute and then slowly (20 rpm) for 30 minutes and then allowed for sedimentation for 30 minutes. Initial turbidity of the sample containing kaolin concentration of 1000mg/l and 400 mg/l were 447 NTU and 184 NTU respectively. Residual turbidity after coagualtion and 30 minutes sedimentation are shown in Table A8. These results are almost same of section 4.2.1.

To count faecal coliform colony 10 ml samples from each beaker were taken after coagulation and sedimentation. After 24 hours of incubation no faecal coliform colony was found in any sample. This may be due to very high concentration of kaolin particles. In higher turbid water microorganisms come in contact with kaolin and form floc rapidly. Faecal coliform were adsorbed and entraped in flocs and at low dosage of alum (25 mg/l), all microorganism were removed.

After coagulation and 30 miniutes sedimentation, the samples were filtered by filter paper. The residual turbidity results are tabulated in Table 4.6. From Table 4.6 it is found that most of kaolin particles are removed by coagulation and filtration.

Table 4.6 : Residual turbidity with variation of alum dosage at pH 6.0 after coagulation and filtration

Alum dosage (mg/l)	Residual turbidity (NTU)	
	Kaolin concentration 1000 mg/l	Kaolin concentration 400 mg/l
25	1.8	0.69
50	1.25	0.60
125	0.73	0.37
250	0.62	0.35

CHAPTER-5
CONCLUSIONS AND RECOMMENDATIONS

5.1 CONCLUSIONS

The objective of this research work was to obtain optimum coagulation condition for removal of microorganism and suspended material. Faecal coliform group of bacteria was used as an indicator of microbial water quality. In this study the removal of faecal coliform and kaolin particles were observed at varying alum dosage, pH and sedimentation time. Effectiveness of filtration was investigated in this research but due to time constraint only filter paper was used for this purpose.

The following conclusions can be drawn from this research work :

1. Increase in alum dosage enhances removal rate of faecal coliform and at any alum dosage removal of faecal coliform increases with increasing sedimentation time. Moreover, for the same removal of faecal coliform less sedimentation time is required for increased alum dosage. As alum dosage increases, precipitation of $Al(OH)_3$ increase and adsorption of microorganism on $Al(OH)_3$ increases. Moreover, with increasing sedimentation time settlement rate decreases. Therefore, with increasing alum dosage potable water can be obtained within short period of sedimentation.

2. Samples containing faecal coliform only, large amount of alum dosages are required for removal of all faecal coliform whereas samples containing both kaolin particles and faecal coliform small amount of alum dosages are required for removal of all faecal coliform. For initial faecal coliform concentration of 59000/100 ml all the faecal coliform are removed by addition of 250 mg/l alum dosage and 2 hours sedimentation at pH 6.0. For samples containing faecal coliform concentration of 47600/100 ml and kaolin particles of 400 mg/l, all the

faecal coliform are removed by addition of 25 mg/l alum dosage and 30 minute sedimentation only.

3 In highly turbid water, some suspended particles settle by natural sedimentation. From this study it is found that after 30 minutes turbidity removal by natural sedimentation for initial kaolin concentration of 1000 mg/l and 400 mg/l were 74.83% and 69.8%, respectively. Moreover almost all of the kaolin particles are removed by coagulation, sedimentation and filtration.

4. Removal of turbidity increases with increasing alum dosage. By addition of 5 mg/l alum dosage and 30 minutes sedimentation turbidity removal was 93% and by addition of 250 mg/l alum dosage and 30 minute sedimentation turbidity removal was 98.1% for sample containing 1000 mg/ kaolin concentration and maintained at pH 6.0. Removal of turbidity increases with increasing kaolin concentration. By addition of 250 mg/l alum and 30 minutes sedmentation turbidity removal were 98.1% and 97.14% for kaolin concentration 1000 mg/l and 400 mg/l respectively.

5. For sample containing higher kaolin concentration most of kaolin particles were removed by sedimentation and small amount of alum dosage were required to obtain optimum condition. Optimum alum dosage for kaolin concerntratin 400 mg/l, 1000 mg/l, 3000 mg/l and 4000 mg/l were 25,25,10, 10 and 10 mg/l.

6. In samples containing microorganism removal of faecal coliform in acidic environment is higher than in alkaline environment without any treatment.

7. In sample containing microorganism the removal rate of faecal coliform at pH 3.6 showed two distinct phase: in the initial phase faecal coliform population reduced rapidly upto 2 hours, then reduced slowly in the second phase. In initial phase the faecal coliform removal was 70.0% and remaining 29.9% faecal coliform was removed in subsequent 22 hours.

5.2 RECOMMENDATIONS

1. In this study, removal of faecal coliform were observed at pH 6.0 and pH 4.5. At pH 6.0, the Al species are mostly Al($OH)_3$ (Sullivan and Singley, 1968 and Hossain, 1996). As the volume of $Al(OH)_3$ is 50 times larger than the same quantity of clay particles (Matsui and Tambo, 1989), large number of microorganisms were adsorbed on $Al(OH)_3$ mesh and settled down. But at pH 4.5, positively charged Al^{3+} is the principal species and small amount of Al $(OH)_3$ is formed. Small amount of organism may adsorb in the $Al(OH)_3$ mesh and charge neutralization may take place. More studies are needed to observe the behavior of charge of organism.

2. Two types of kaolin concentration (400 mg/l and 1000 mg/l) were used in this study. As highly turbid samples were used, all the faecal coliform was removed by any alum dosage. This experiment can be repeated using varying concentration of kaolin particles and coagulation condition to study the effect of turbidity.

3. Filter paper was used for filtration of suspended material and microorganism. Filtration may be done using different filter media.

4. Throughout the experiment, the samples after addition of alum dosage were stirred rapidly (80 rpm) for one minute and then slowly (20 rpm) for 30 minute. This condition can be varied to study its effect on removal.

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APPENDIX A

Data of Different Experiments

Table A.1 : Residual turbidity with variation of alum dosage at pH 6.0 after coagulation and sedimentation for samples containing kaolin particles

Alum dosage (mg/l)	Residual turbidity (NTU)	
	Initial Kaolin concentration 1000 mg/l	Initial Kaolin concentration 400 mg/l
0	112	55
5	31	23.2
10	24	20
15	22.4	18.1
20	19	14.8
25	16.2	9.3
50	13.2	8.1
125	9.1	5.5
250	8.2	5.2

Table A.2 : Residual turbidity with variation of alum dosage at pH 6.0 after coagulation and filtration for samples containing kaolin particles

Alum dosage (mg/l)	Residual turbidity (NTU)	
	Initial Kaolin concentration 1000 mg/l	Initial Kaolin concentration 400 mg/l
25	1.9	0.74
50	1.3	0.68
125	0.78	0.41
250	0.69	0.39

Table A.3 : Residual turbidity with variation of alum dosage at pH 4.5 after coagulation and sedimentation for samples containing kaolin particles

Alum dosage (mg/l)	Residual turbidity (NTU)	
	Kaolin concentration 1000 mg/l	Kaolin concentration 400 mg/l
0	175	76.4
25	132	55.8
50	125	53.7
125	118	51.3
250	111	48.0

Table A.4 : Residual turbidity with variation of alum dosage at pH 4.5 after coagulation and filtration for samples containing kaolin particles

Alum dosage (mg/l)	Residual turbidity (NTU)	
	Initial Kaolin concentration 1000 mg/l	Initial Kaolin concentration 400 mg/l
25	2.4	2.1
50	1.8	1.3
125	1.1	0.82
250	0.73	0.72

Table A.5: Number faecal coliform at different sedimentation time at 25 mg/l Alum dosage at pH 4.5 and 6.0.

Sedimentation time (hr)	No. of faecal coliform/100 ml	
	pH 4.5	pH 6.0
1/2	10000	2700
1	7550	1400
2	5100	600
4	3150	480
8	2800	290
24	-	290
48	-	450

Table A.6: Number of faecal coliform at different alum dosage at pH 6.0

Alum dosage (mg/l)	No. of faecal coliform after 1 hr. sedimentation per/100ml	No. of faecal coliform after 1 hr sedimentation and filtration per/100 ml
25	900	11
50	500	10
125	80	2
250	10	0

Table A.7: Number of faecal coliform with variation of alum dosage and sedimentation time at PH 6.0.

Alum dosage (mg/l)	No. of faecal coliform/100 ml				
	t=30 min.	60 min.	120 min.	240 min.	480 min.
25	2550	1300	570	480	280
50	1100	630	280	130	60
125	400	110	70	10	0
250	90	10	0	0	0

Table A.8 : Residual turbidity with variation of alum dosage at pH 6.0 after coagulation and sedimentation for samples containing both microorganism and kaolin particles

Alum dosage (mg/l)	Residual turbidity (NTU)	
	Initial Kaolin concentration 1000 mg/l	Initial Kaolin concentration 400 mg/l
0	113	56
25	15.7	8.5
50	12.5	7.2
125	9.6	4.7
250	7.5	4.48