

AN ABSTRACT OF THE THESIS OF

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Title: Factors Affecting the Adsorption of Heavy Metals by Activated
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Adsorptive removal of heavy metals by activated sludge bacteria is an important process for the control of heavy metals in wastewater treatment plant effluents and sludges. In this laboratory study, the objectives were to delineate the factors affecting metal uptake by activated sludge in order to provide the optimal metals partitioning for future use of sludge or effluent.

Activated sludge was cultured at solids retention times of 1, 5, and 15 days. Batch experiments of steady-state cultures were performed to determine the removal mechanism and to investigate the effects of θ_c , pH, and metal dosage upon the removal of cadmium, copper, and zinc by the biological solids.

The mechanism for metals removal was interpreted as adsorption. Constants for Langmuir (and Freundlich) adsorption equations and

metal-sludge "complexation" constants were determined, but the small number of data points makes this information tentative and subject to further study.

Experimental removal of copper by the activated sludge solids was maximized at pH 7 (85 percent removal). Zinc and cadmium removal were significantly more concentrated by the activated sludge bacteria at θ_c values less than 5 days than at higher θ_c values, while copper showed only minimal increase in the concentration factor with increasing θ_c values.

Hypothetical calculations indicated that $\text{NH}_3\text{-N}$ complexation of copper was significant at neutral pH values while soluble hydroxy-complexes were predominant at pH values of 8.5 and 10.

It was concluded that metals partitioning in activated sludge treatment facilities could be optimized by controlling the operational parameters θ_c and pH (and also by controlling NH_3 for copper at neutral pH values), and a methodology was presented for prediction of soluble metal and particulate metal speciation utilizing empirically determined stability constants for the metal-sludge "complexation". Correlation between calculated and empirical speciation of metals was highly dependent upon the solids concentration and the conditions of K'_s determinations.

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FACTORS AFFECTING THE ADSORPTION OF HEAVY METALS BY ACTIVATED SLUDGE BACTERIA

1. INTRODUCTION

Man has indiscriminately infected his biosphere with many toxic elements and compounds. Heavy metals are a few among the many recently recognized pollutants. The ability to remove and discard or reuse these elements is currently being studied throughout the nation in an attempt to alleviate the situation.

Biologically, heavy metals in trace amounts can be toxic to first level trophic organisms in two ways: acute toxicity, caused by short term lethal dosages, and chronic toxicity caused by long term non-lethal dosages. Chronic toxicity may cause non-lethal impairment or long term death. This latter form of toxicity is sometimes considered to be the more serious of the two.

Bioaccumulation of metals in the food chain is another threat to man. First level trophic organisms have been found capable of concentrating aqueous heavy metals by adsorption upon their surfaces. The organisms have the ability to concentrate these metals to a level many thousands of times greater than the maximum solubility of the metals in water. As these organisms are consumed, the metals bioconcentrate in various organs of the higher level organisms causing acute and chronic toxicity in them.

Extensive amounts of metals are lost in industrial wastewaters. Because of the ability of some organisms to concentrate heavy metals, they are the object of interest to many industries for possible recovery purposes. Activated sludge solids have been found to concentrate metals up to 10,000 times their weight in the aqueous phase (Neufeld and Hermann, 1975). Activated sludge is also the process most widely employed in the treatment of sewage.

Widespread use, prevalence in industrial and domestic wastewater, and the environmental effects of metals are reasons for studies of heavy metals removal by activated sludge bacteria. Cadmium, copper, and zinc, the metals selected for this study, occur naturally in the environment. Copper and zinc occur in about equal proportions and are required nutrients at ultra-trace concentrations. Cadmium is not nearly so abundant, but it is toxic at lower concentrations than copper or zinc, and it is also bioaccumulated which multiplies its threat to man.

There is then a need to delineate the factors affecting heavy metals uptake by activated sludge. In cases where the maximum effluent concentrations of metals would be exceeded, metals recovery or effluent reuse for irrigation or other applications are contemplated, it would be desirable to maximize metals removal. On the other hand, removal of metals by sludge solids needs to be minimized in cases where sludge disposal on land is contemplated and metals accumulation in soils and crops, or groundwater contamination are considered problems.

The following factors were studied to delineate metals removal by activated sludge bacteria:

1. The mechanism of removal for metals by activated sludge -- adsorption vs. metabolism,
2. The effects of pH upon metals concentration by the activated sludge bacteria,
3. The effects of the solids retention time (SRT) upon metals concentration by the activated sludge bacteria, and
4. Adsorption isotherms for metals removal by activated sludge bacteria.

2. BACKGROUND

2.1 Sources and Environmental Significance of Cadmium, Copper, and Zinc

2.1.1 Sources and Distribution of Metals in the Environment

Cadmium is a soft, bluish white metal in pure form. It is produced primarily from the zinc ore sphalerite (ZnS) (Hem, 1972; Weast, 1972). The most abundant mineral form of cadmium is greenockite, (CdS).

Some of the physical-chemical properties of cadmium are listed in Table 2.1. Cadmium is a very rare metal. Its abundance is about 1/1000th the abundance of zinc. Cadmium is not known to be essential to life processes and it is highly toxic to animals.

The distribution of cadmium in the environment depends upon the degree of pollution. Natural and polluted waters have similarly low levels of soluble cadmium (see Table 2.2). Sediment concentrations of cadmium vary widely and high concentrations of cadmium in the sediment are indicators of pollution (see Table 2.2). Natural soils have very low concentrations of cadmium 0.10 to 0.7 ppm (Hem, 1972) while some polluted sediments contain up to 220 ppm cadmium (Rohatgi, and Chen, 1976).

Electroplating comprises greater than 50 percent of the total industrial consumption of cadmium (Hem, 1972; Weast, 1972). However, the primary cause of environmental pollution by cadmium is the burning of

TABLE 2.1PHYSICAL/CHEMICAL PROPERTIES OF METALS (Weast, 1972)

<u>Metal</u>	<u>Atomic Number</u>	<u>Atomic Weight</u>	<u>Valence</u>	<u>Melting Point, °C</u>	<u>Boiling Point, °C</u>
cadmium	48	112.40	2	320.9	765
copper	29	63.546	1, 2	1083.4 ± 0.2	2567
zinc	30	65.37	2	419.58	907

TABLE 2.2

DISTRIBUTION OF METALS

Metal	Comments	Concentration	Ref.
cadmium	Distribution in soils	0.01 to 0.7 ppm avg. 0.06 (by weight)	Hem, 1972
	Distribution in fresh waters (polluted and non polluted)	0.001 mg/l (avg)	Hem, 1972
	Sediments of an industrially polluted river	128 ppm	Oliver, 1972
	Unpolluted sediments in an area surrounding a waste water plant outfall	2.6 ppm	Rohatgi and Chen, 1972
	Polluted sediments in an area surrounding a waste water plant outfall	220 ppm	Rohatgi and Chen, 1972
copper	Distribution in fresh water (polluted and non polluted)	0.07 to 0.12 ppm	Bender, '72
	Sediments of an industrially polluted river	236 ppm	Oliver, '72
	Unpolluted sediments in an area surrounding a waste water plant outfall	30 ppm	Rohatgi and Chen, 1972
	Polluted sediments in an area surrounding a waste water plant outfall	2580 ppm	Rohatgi and Chen, 1972
zinc	Distribution in soils	10 - 300 ppm avg. 50 (by wt.)	Hem, 1972
	Distributed in fresh waters (polluted and non polluted)	0.01 mg/l	Hem, 1972
	Storm water runoff	107 mg/l	Whipple, '77

Table 2.2 cont.

Metal	Comments	Concentration	Ref.
zinc	Unpolluted sediment in an area surrounding a waste water plant outfall	110 ppm	Rohatgi and Chen, 1972
	Polluted sediment in an area surrounding a waste water plant outfall	4220 ppm	Rohatgi and Chen, 1972

fossil fuels (Pitt, 1973). Because of their low boiling points (see Table 2.1), cadmium as well as zinc, enter the environment as metal fumes and become attached to particulates (Hem, 1972; Waganet, 1978; and Pitt, 1973). These metal particulates return to the earth and enter the aquatic system via stormwater transport. Surveys in urban areas have shown that about 50 percent of the cadmium entering receiving streams was from stormwater runoff (Davis and Jacknow, 1975, Pitt, 1973).

Copper has been used in industry for many centuries. It is an abundant reddish colored metal and is malleable, ductile, and a good conductor of heat and electricity. Copper occurs in many minerals: cuprite, malachite, azurite, chalcopyrite, and bornite (Weast, 1972).

Some of the physical-chemical properties of copper are listed in Table 2.1, copper is an essential nutrient, but becomes toxic for some species of fish at levels above 0.01 mg Cu/l in solution, though most fish can tolerate significantly higher concentrations of copper (Buhler, 1972).

Concentrations of soluble copper in polluted and non-polluted waters is low, occurring from 0.07 to 0.12 ppm (Bender, et al., 1972). Aquatic sediments adsorb and concentrate the copper from the waste streams and thus the low levels of soluble copper are maintained (see Table 2.2):

Large quantities of metals are lost in the waste streams of electro-

plating and brass/copper industries (see Table 3). Other sources of copper pollution are from mining wastes and aerosol contamination.

Zinc is similar to cadmium in many of its physical-chemical properties (see Table 2.1). It is a much more abundant metal and has been used for centuries in the formation of brass (Weast, 1972). Zinc is available from the ores sphalerite (sulfide), smithsonite (carbonate), calamine (silicate), and franklinite (zinc, manganese, and iron oxide). Zinc is a bluish white metal and is more brittle than cadmium but becomes malleable at temperatures of 100°C to 150°C. Zinc, like copper, is essential to the growth of organisms, but can be toxic at concentrations only slightly above the concentrations required for proper nutrition.

The low soluble zinc concentrations occurring in waters is consistent with the other metal concentrations. The metal is concentrated in the sediments near industrial and domestic waste water outfalls (see Table 2.2).

Industrial sources of zinc include electroplating, alloy processing, and galvanizing (Table 2.3). Zinc with a low boiling point, escapes high temperature processes as a metal fume, and attaches to particulate matter. In this form it is transported to streams via fallout and stormwater runoff (Whipple, 1977).

Industrial wastes and fossil fuel emissions are major sources of environmental metals contamination. Soluble metal concentrations in streams are generally low, but sediment concentrations may be greatly elevated in polluted waters.

TABLE 2.3

INDUSTRIAL SOURCES OF METALS POLLUTION

Metal	Industry	Conc. of metal and/or transport	Ref.
cadmium	electroplating	---	Hem, 1972
	silver solder	fumes	Hem, 1972
	zinc and copper refinement	fumes	Weast, 1972 & Hem, 1972
	laundries	0.002 to 0.134 mg/l	Klein, L.A., <u>et al</u> , 1974
	fabric dyeing	up to 25 mg/l	Rouse, 1976
	fossil fuel & other aerosol pollution	2 mg/kg, street soil	Pitt, 1973, & Biggs, 1973
copper	electroplating	4.5 mg/l	Nemerow, '72
	brass/copper in- dustries		Nemerow, '72
	bright dip	6.9 to 44.0 g/l	
	pickling so- lution	4.0 to 22.6 g/l	
	copper mining/ dust		Nemerow, '72
	Aerosol contami- nation, mining dust, fossil fuel, etc.	100 mg/kg, street soil	Pitt, 1973
zinc	galvanizing	---	Weast, 1972
	alloying	---	Weast, 1972
	electroplating	46.3 mg/l	Nemerow, '72
	metal plating	39-82 mg/l	Nemerow, '72
	rubber industries waste	---	Weast, 1972

Table 2.3 cont.

Metal	Industry	Conc. of metal and/or transport	Ref.
	brass/copper indus- try	---	Nemerow, '72
	bright dip	0.2 to 37.0 g/l	
	pickling solu- tion	4.3 to 41.4 g/l	
	motor industry	1.12 #/100 cars	Nemerow, '72

2.1.2 Toxicity

Wood (1974), in his investigation on the biological cycles for toxic elements, states that cadmium copper, and zinc come under the "very toxic and readily accessible" category of elements. Accessibility is the key word for increased hazards from metals.

Copper and zinc are known to be essential to life. Copper is essential in oxidative processes, enzymes, and hemocyanin. Zinc is required for the activity of many enzymes (Buhler, 1972). Thus for these metals, there is a minimal concentration required for growth beyond which the metals become inhibitory.

Cadmium, on the other hand, is not an essential element. Though cadmium forms coordination complexes with a variety of ligands in biological systems, no metabolic interconversions have ever been demonstrated for this element (Wood, 1974). Since there is no metabolic interaction, and thus no elimination of cadmium in waste products or formation of nontoxic compounds, cadmium will accumulate and become more concentrated as it ascends the food chain.

Heavy metals may exhibit the property of synergistic toxicity. If more than one metal is simultaneously ingested, even at individually non-toxic concentrations, the toxicity of the combined metals may be sharply increased (Cheremisnoff and Habib, 1972).

2.1.2.1 Industrial Sources of Toxicity

General pathways for cadmium exposure are ingestion via cadmium-

coated food containers, and fume inhalation. Acute toxicity from ingestion produces increased salivation, choking, persistent vomiting, abdominal pain, diarrhea, and tenesmus. Toxicity occurs at dosages of approximately 3 mg/day (Buhler, 1972; Fairhall, 1969). Symptoms usually begin within 15 to 30 minutes of exposure, but may take as long as three hours. Respiratory exposure to cadmium fumes can cause throat dryness, cough, headache, vomiting, and chest constriction (Buhler, 1972; Fairhall, 1969).

Chronic cadmium poisoning can occur from repeated respiratory or gastrointestinal uptake. Victims of chronic poisoning often exhibit a yellow ring on their teeth, and have kidney damage and proteinuria. Advanced cases have hypertension and testicular damage (Fairhall, 1969).

Zinc fumes can cause toxic reactions, but toxicity is dependent upon the type of zinc compound fumes inhaled. Zinc fumes inhalation most often results in symptoms similar to influenza, but more toxic fumes like zinc chloride burn the nasal passages as well (Fairhall, 1969). Ingestion and inhalation dosages for toxicity have not been determined.

Toxicity from copper is less common but does occur. Chronic copper toxicity exhibits itself in symptoms of dullness, depression, anorexia, and liver and kidney problems. Levels of copper for toxic reaction have not been established in man (Fairhall, 1969).

2.1.2.2 Toxicity in Aquatic Sediments

Toxicity of heavy metals to aquatic life forms is dependent upon

the form in which the metal exists (Andrew, 1977). Copper can be toxic as a free metal ion, or can reduce the toxicity of compounds such as cyanide through complexation (Stiff, 1971). Cadmium, at 0.003 ppm, is known to be toxic to some species of fish (Buhler, 1972), while Psuedomonas aeruginosa was resistant to as much as 400 ppm cadmium (Seyfrid, 1978). Zinc was found to be toxic to some fish at levels of 0.01 ppm (Buhler, 1972) but again tolerance levels may exceed this.

Copper toxicity to aquatic organisms has been studied by Singleton and Guthrie (1977) to establish patterns of toxicity. It was found that in both fresh water and brackish water, the addition of 2 ppm copper as copper sulfate caused an increase in the total colony forming units (TCFU) of the aquatic micro-organisms. However in contrast to this increased TCFU, the diversity of the originally existing colonies was significantly decreased in fresh water, with little or no decrease in the diversity of the brackish water organisms. It was concluded that the decreased diversity of the organisms would result in a significant decrease in the stability of the aquatic community.

2.1.2.3 Land Applications of Sewage Sludge Containing Metals

As the use of sludge for a soil conditioner or fertilizer became more widespread, the dangers of the heavy metals from this source became a major controversy. Bauer (1978) argues that cadmium uptake varies from plant to plant, even among species of corn. Leafy green vegetables and leaves of plants are known to accumulate cadmium and thus are to be

avoided as a crop in which sludge may be used as a fertilizer (Bauer, 1978; Page, 1978). Cattle foraging on sludge fertilized grass have been found to have a lower cadmium content in their tissues than found in control cattle (Bauer, 1978; Fitzgerald, 1978).

The ability of a plant to uptake metals from a metal compound depends on factors other than the solubility of the metal compound in water. Plant uptake of metals from soils depends on the portion of the metal that is "plant available" rather than the total metal content of the soils (Brown, 1975). The plant root causes chemical and enzyme reactions to alter the chemistry of the area surrounding the root (rhizosphere). The rhizosphere differs for each plant, and thus metal uptake will vary from plant to plant.

The guidelines for allowable metal loadings on soils vary widely. One guideline states an absolute value of 200 tons of sludge per year may be applied to soils without upset (Jorgenson, 1975). Garrigan's (1975) guidelines recommend that sludge applications be based upon the organic content of the soil, the soil type, the maximum permissible metal content of the sludge, and the cation exchange capacity of the soil (see Table 2.4).

2.1.2.4 Effects of Metals upon Sewage Treatment Processes

Industrial wastewaters, stormwater runoff, and domestic sewage all contain significant amounts of heavy metals. These waste streams

TABLE 2.4

PERMISSIBLE LEVELS OF METALS IN SLUDGES (from Garrigan, 1977)

<u>Metal</u>	<u>Max. Concentration ug metal/g sludge</u>	<u>Min. NH₄-N/element ratio</u>
cadmium	1% of zinc	500
copper	1000	10
zinc	2000	4

TABLE 2.5

TOXIC METAL LIMITS IN ANAEROBIC DIGESTORS

<u>Metal</u>	<u>Dosage for anaerobic toxicity mg/l</u>
cadmium	60 ^a -- 180 ^b
copper	170 ^b
zinc	103 ^b

^a(Mosey, 1971)^b(Mosey, et al, 1975)

are often combined in the municipal sewage treatment facilities, and can reduce the existing biological treatment efficiency.

The toxicity of heavy metals to a biological culture (such as activated sludge) is a function of the chemical form of the metals. The study by Barth et al. (1965) showed that primary treatment had two effects upon metals in activated sludge:

1. the total metal content of primary effluent is less than that of influent sewage because some metal is removed with the primary sludge, and
2. the chemical and physical characteristics of sewage alter the form of the dosed metal. For example, 90 percent of zinc added in a soluble form was converted to an insoluble form.

Several studies have considered heavy metals effects on secondary biological treatment processes. Cadmium and zinc soluble chloride compounds were observed to deflocculation of activated sludge at dosages of 20 and 50 ppm, respectively (Neufeld, 1976). Elemental copper at 1 ppm caused reduction of treatment efficiency by activated sludge. Activated sludge cultures which have been acclimated to copper-CN showed no reduction in treatment efficiency. Similarly, zinc-CN removal by activated sludge remained good after acclimation, but the bacteria would not acclimate to zinc-sulfate and treatment efficiency was reduced (Barth et al., 1965).

Anaerobic digestors are sensitive to metals. Metal dosages which cause acute toxicity of the biomass and levels of tolerance are listed in

Table 2.5. The toxicity can be reduced or eliminated if the metal is in a metabolically unavailable form.

2.2 Effects of SRT upon the Settling Characteristics of Sludge

The settling rates of activated sludge were studied to determine the effects of SRT (θ_c) and SVI (sludge volume index) upon the sludge settling characteristics (Bisogni and Lawrence, 1971). It was learned that oxygen utilization was higher for very low θ_c values, but remained constant at $20 \text{ mgO}_2 / \text{g solid/hr}$ for $\theta_c = 3$ to 12 days. The SVI was compared to the SRT and was found to be at a maximum value at $\theta_c = 3$ days. The SVI values were much lower at higher and lower θ_c values. Table 2.6 describes the sludge characteristics as a function of θ_c . The protozoan population in activated sludge is increased in older sludges (10 - 12 days) whereas in sludges of $\theta_c = 1$ to 3, the protozoans were virtually nonexistent.

2.3 Effects of SRT upon Soluble Organics

A study by Kim et al. (1976) considered the effect of activated sludge SRT upon the adsorption of soluble organics by carbon. High SRT sludges were found to produce effluents with lower amounts of nonadsorbable soluble organics than occurred in lower SRT systems. The effluent COD of the higher SRT sludges was also lowered. It was determined that SRT had an affect on the type of soluble organics present in the activated sludge system.

TABLE 2.6CHARACTERISTICS OF ACTIVATED SLUDGE SOLIDS AS A FUNCTION
OF θ_c^*

θ_c range (days)	Characteristics of the solids
0.25 to 2	Predominately dispersed growth
2 to 9	Well formed average size floc of low to medium density
9 to 12	Irrregular shaped floc particles of low density which looked as if they broke loose from larger floc particles (deflocculated) and pin floc

* (Bisogni and Lawrence, 1971)

These organics could affect metals removal by complexing the metals, thus decreasing the amount of metals adsorbed. This increases the apparent solubility of the metal.

2.4 Effects of Organics upon Metal Adsorption

The form and concentration of organic ligands are important because of their effects in changing the apparent solubility and chemical interactions of metals. Polygalacturonic acid, a natural polymer has been found to complex metals and increase their soluble concentrations by a factor of 10^2 (Jellinek and Sangal, 1972). Some organic ligands will complex trace metals in forms available to plants which increase growth, but other ligands will form more stable unavailable compounds which reduce growth (Bender et al. 1970).

Many metal-ligand complexes interfere with the adsorption of the metal. For example, the extent of adsorption of copper depends strongly upon the availability of organic ligands and surfaces. When complexing agents are absent, Cu^{2+} is substantially removed from solution by adsorption on hydrous metal oxides. Addition of complexing agents will result in partial or total release of adsorbed copper (Vuceta and Morgan, 1978). In an activated sludge laboratory study, the adsorption by a surface (activated sludge) proved to be more stable than chelation compounds (Cheng et al. 1975), thus little or no decrease in metal adsorption occurred.

Davis and Leckie (1978), reported three different effects of

complexing organic ligands on adsorption by iron oxides:

1. Complexing ligands that are not adsorbed decrease trace metal adsorption. The adsorption site on the surface compete with the soluble ligand for coordination of the metal ion. The magnitude of the effect depends on the relative intensities of the metal-ligand and metal-surface bonds and the concentration of the ligand.
2. When the major coordinating functional groups of an adsorbed complexing ligand are involved in the surface adsorption, then the ligand will decrease or have a minor effect upon metal adsorption. The magnitude of the effect on metal uptake again depends on the relative intensities of metal binding by the complexing ligand and the adsorbing surface.
3. When the adsorbed ligand has a strongly complexing functional group directed outward from the surface, trace metal-ligand complex is adsorbed or the ligand-oxide unit can adsorb metal. The functional groups may serve as additional adsorption sites or may stabilize adjacent sites. The adsorption site provided by an adsorbed complexing ligand with an outwardly directed functional group may have different energy of adsorption than an adjacent oxide adsorption site.

Many organics in solution can form chelates with metals. In some complexes, the solubility of the metal may be greatly increased. Other chelates may convert the metal to an insoluble form (as in humic

acid complexes) or detoxify a metal by forming a metal chelate in a biologically unavailable form (Manahan and Smith, 1973).

2.4.1 Organics in Secondary Effluents

Domestic sewage contains less than 0.1 percent organics by weight. These organics are mainly carbohydrates, amino acids, and fatty acid esters. Organics which are easily utilized by bacteria don't affect trace metals, but persistent organics can have very significant effects.

The organics in secondary effluents have been characterized by Manka et al. (1974) as shown in table 2.7.

TABLE 2.7

DISTRIBUTION AND CHARACTERIZATION OF ORGANICS
IN SECONDARY EFFLUENTS
(PERCENT)

	Pro- tein	Carbo- hy- drates	Anionic Deter- gents	Tanins & Lignins	Ether Ex- tract	Ful- vic Acid	Humic Acid	Hymotho- melanic Acid
Avg.	22.0	5.75	15.6	1.35	14.2	25.1	10.6	6.4
Range	18.9- 24.8	3.6- 8.0	11.2- 20.5	0.8- 2.4	10.3- 19.9	16.8- 29.6	3.6- 15.7	1.9- 10.3
Total % = 101								

Organics which combine to form complexes or chelates usually do so by an exchange of electrons. Examples of organics which are strong electron donors are shown in Table 2.8 (Faust and Hunter, 1971).

TABLE 2.8
 TYPES OF DONOR GROUPS

aliphatic amino	$R-NH_2, RR'NH, RR'R'N$
carboxylate	$-COO^-$
enolate	$\diagdown C=C-O^-$
alkoxide	$R-O^-$
phenoxide	$\diagup C=O^-$
mercaptide	$R-S^-$
phosphate	$-O-P(=O)(O^-)_2$
phosphonate	$-CH_2-P(=O)(O^-)_2$
aromatic amino	$-N=$

Weaker electron donors, such as carbonyl, ether, amide, thio-ether, and hydroxyalkyl do not form strong complexes on their own, but may be part of a ligand substitution with the electron donors listed in Table 2.8 to form very stable complexes and chelates.

In Table 2.7, note that over one-third of the organics are humic substances (humic and fulvic acids). Fulvic acid is a naturally occurring metal complexing agent that can complex divalent and trivalent metal ions. Fulvic acid can solubilize metals into a stable complex form from practically insoluble hydroxides and oxides (Faust and Hunter, 1971; Reuter and Perdue, 1977).

2.5 Bacterial Cell Wall as an Adsorption Surface for Heavy Metals

2.5.1 Cell Wall Characteristics

The main function of the cell wall is to provide rigidity and shape to the cell. However, the composition of the cell wall affects its interaction with the environment. The part of the cell wall which affects bioflocculation and metals adsorption is the slime layer or capsule (Forster, 1972; Steiner, 1975; Friedman et al. 1968). Many biopolymers including lipids, proteins, and nucleic acids have been reported to occur at activated bacterial sludge surfaces, the most common type being the polysaccharides (Steiner, 1975).

Polysaccharides can be composed of simple sugars such as glucose, galactose, and mannose, while other cell wall polysaccharides are composed of the corresponding amino sugars (Sistrom, 1969).

Gram negative bacteria (e.g. Pseudomonas spp., Flavobacter spp., and Achromobacter spp.) have been found commonly in activated sludge (Forster, and Lewin, 1972). Gram negative bacteria have a lower quantity of sugars in their exterior structures than gram positive bacteria, and may also be composed of up to 20 percent lipid (Steiner, 1975, Siström, 1969). Gram negative bacterial cell walls have a greater variety of the amino acids that occur in protein than gram positive bacteria.

2.5.2 Cell Wall Effects Upon Adsorption

The high molecular weight exocellular polymers of the biofloc in activated sludge which include polysaccharide, protein, RNA, and DNA provide many functional groups which may act as binding sites for heavy metals (Cheng et al. 1975). These polymers also enhance bioflocculation.

Soluble polymers have been found to play an important role in polyvalent metals removal. Metal-salt-polymer units attach to the neutral sugars and form a bridge, thereby removing metals and inducing flocculation (Steiner et al. 1975).

Polyvalent metal ions displace alkali metal ions and bound water in cell walls of certain Zoogloea species. This reduces the surface charge and aids flocculation (Dugan et al. 1970).

2.6 Previous Study of Heavy Metals Adsorption by Activated Sludge Bacteria

Neufeld and Hermann (1975) investigated the uptake of heavy

metals by activated sludge, and tried to establish the capability of metals acclimated activated sludge bacteria to remove very high concentrations of metals as treatment of industrial wastes. Sludge acclimated to a sterile synthetic soluble feed was slug dosed with metals concentrations from 30 to 1000 mg metal/l. The sludge was found to remove large quantities of metal (see Table 2.9). The study did not report whether or not mineral precipitation was a significant removal mechanism.

Lower dosages of metals and removal by activated sludge bacteria were investigated by Cheng et al. (1975). Batch reactors were employed for testing steady state activated sludge cultures. A summary of the initial kinetic tests is given in Table 2.10.

The results of the study showed that at lower metal concentrations, metal is removed by the biofloc through the formation of metal organic complexes, and that at higher metal concentrations, metal ion precipitation may cause additional removal.

Metal uptake capacity of the sludge was determined to be a function of pH. Uptake was found to increase as pH increased to a point where precipitation of metal hydroxides occurred.

The affinity of the biomass for metal ions was found to be stronger than that of competing organics in the supernatant. The preferred order of metal uptake by activated sludge was determined to be:

lead > copper > cadmium > nickel.

TABLE 2.9

METALS REMOVAL BY ACTIVATED SLUDGE
(Neufeld et al. 1975)

Metal	Dosage mg/l	Equilibrium Metals Conc. assoc. with solids, mg/l	Percent Removal
cadmium	30	24	80
	100	70	70
	300	234	78
	1000	350	35
zinc	30	18	60
	100	50	50
	300	165	55
	1000	90	9

TABLE 2.10

COPPER UPTAKE BY ACTIVATED SLUDGE
(Cheng et al. 1975)

Metal	pH	Initial Metal Concentration mg/l	Percentage uptake at	
			10 min.	120 min.
copper	5.8	2.1	82	85
		10.3	84	91
		25.2	81	90

3. THEORETICAL CONSIDERATIONS

3.1 θ_c and Related Models of Biological Growth

Lawrence and McCarty (1970) developed solids retention time (SRT) as the unifying parameter for the basis of biological treatment, design, and operation.

The basic equation for bacterial growth is

$$\mu = Y U - k_d \quad (3.1)$$

where $\mu = (dX/dt)/X$, the specific growth rate of bacteria,

$U = (dF/dt)/X$, the substrate consumption rate

Y = the growth yield coefficient, mass/mass,

k_d = the microorganism decay rate, time^{-1} ,

X = the microbial mass concentration, mass/volume,

(dX/dt) = the net growth rate of microorganisms per volume of reactor, mass/volume-time, and

(dF/dt) = the rate of microbial substrate utilization per unit volume.

Also

$$U = (dF/dt)/X = \frac{kS_1}{S_1 + K_s} \quad (3.2)$$

where k = the maximum rate of substrate utilization, mass substrate per mass solids-time

S_1 = the concentration of substrate surrounding the organisms, mass per volume, and

K_s = the half velocity coefficient, mass per volume.

θ_c , termed sludge age, is the mean solids retention time and is defined

as

$$\theta_c = X/(dX/dt) = \mu^{-1} \quad (3.3)$$

For a complete-mix reactor with cell recycle, equation 3.3 becomes

$$\theta_c = \frac{V X}{Q_w X_r + (Q - Q_w) X_e} \quad (3.4)$$

where Q_w = the flow rate of the fraction of the solids wasted, volume per time

X_r = the concentration of the solids wasted, mass per volume

X_e = the concentration of biological solids lost in the secondary clarifier effluent, mass per volume, and

Q = the forward flow rate, vol/time.

For a completely mixed-cell recycle reactor, a mass balance for the microbial mass in the reactor clarifier system yields

$$(dX/dt)(V) = Y (dF/dt) V - X_e Q - Q_w X_r - k_d X V \quad (3.5)$$

Substituting equation 3.2 and assuming steady state, this becomes

$$\frac{Y V X (k S_1)}{S_1 + K_s} = X_e Q + Q_w X_r + k_d X V \quad (3.6)$$

Further substitution by equation 3.4 and rearranging yields

$$S_1 = \frac{K_s (1 + k_d \theta_c)}{\theta_c (Yk - k_d) - 1} \quad (3.7)$$

S_1 , the concentration of the effluent substrate, is defined solely by θ_c (K_s , Y , k , k_d are fixed for a given microbial system).

The mixed liquor microbial mass concentration can be derived from a steady state mass balance given by equation 3.5 and evaluated for a time period of one θ_c :

$$Y(S_0 - S_1) Q \theta_c = k_d X V \theta_c + (X_e(Q - Q_w) + X_r Q_w) \theta_c \quad (3.8)$$

where S_0 = influent substrate concentration mass/vol.

Rearranging,

$$X = \frac{Y(S_0 - S_1)}{1 + k_d \theta_c} \left(\frac{\theta_c}{\theta} \right) \quad (3.9)$$

Equation 3.9 shows that the microbial population is dependent upon the influent substrate and θ_c . For a given waste strength, S_0 , X is solely a function of the operating θ_c value of the system.

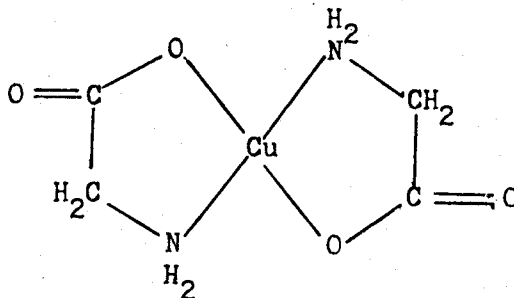
In summary, θ_c defines the waste strength of the effluent. Influent waste strength alters the microbial mass concentration, X . If the influent waste strength is constant, X will be defined by θ_c .

3.2 Chelation and Complexation

As defined by Stumm and Morgan (1970) combinations of cations with molecules or anions containing free pairs of electrons is called complex formation.

The formation can be electrostatic, covalent or a mixture of both. The metal cation is the central atom, around which the molecules or anions (called ligands) are arranged. When the ligand is composed of more than one atom, the atom responsible for the basic nature of the ligand is called the ligand atom. If there are more than one ligand atoms in a base, it can occupy more than one coordination position in the complex and is referred to as a multidentate complex former. Complexation with multi dentate ligands is called chelation, and the complexes are called chelates. An obvious feature of a chelate is the formation of a ring. For example, in the reaction between glycine and

$\text{Cu}\cdot\text{aq}^{2+}$ (Stumm and Morgan, 1970), a chelate with two rings is formed.



Glycine occupies two coordination positions, O⁻ and N⁻ are the donor atoms. If more than one metal atom (central atom) is coordinated, a polynuclear complex is formed.

When a metal combines to form a chelate compound, its stability is a function of the number of rings formed, i.e. the number of bonding sites. Stability is increased by a factor of 10^2 for each chelate ring formed (Faust and Hunter, 1971). If the metal is not attached to the coordination bonding site of the ligand, then the stability of the chelate is much less than if bonding occurred normally. This lessened stability may lead to the following (Faust and Hunter, 1971):

1. polynuclear complexes (bridges with more than one metal ion)
2. hydrolysis with the ligand being replaced by the hydroxyl ion
3. precipitation of the metal-hydroxide, which results in more stable complexes with the metal left in solution, and
4. different ligands substitute to form a mixed ligand complex.

3.3 Adsorption

Adsorption is the phenomenon of accumulation or concentration of a

substrate at a surface or an interface. The material being concentrated or adsorbed is the adsorbate, and the adsorbing phase is the adsorbent.

3.3.1 Isotherms

During adsorption, material from one phase is adsorbed by another. At equilibrium, q_e , the amount of solute adsorbed per unit of solid adsorbent, is a function of C , the concentration of solute at equilibrium. The relationship between q_e and C can be expressed by an adsorption equation (isotherm). The three most commonly used adsorption isotherms are the Brunauer, Emmett, Teller (BET) equation, the Langmuir equation, and the Freundlich equation (Weber, 1972).

3.3.1.1 The BET Adsorption Isotherm

The BET adsorption isotherm assumes multilayer adsorption and constant energy of adsorption. A typical BET isotherm is shown in Figure 3.1a. The equation for the BET adsorption isotherm is

$$q_e = \frac{B C Q^{\circ}}{(C_s = C) 1 + (B - 1)C/C_s} \quad (3.10)$$

where

Q° = the number of moles of solute adsorbed per unit weight of adsorbent in forming a complete monolayer,

B = the constant which is expressive of energy interaction with the surface, and

C_s = the saturation concentration of the adsorbate in solution

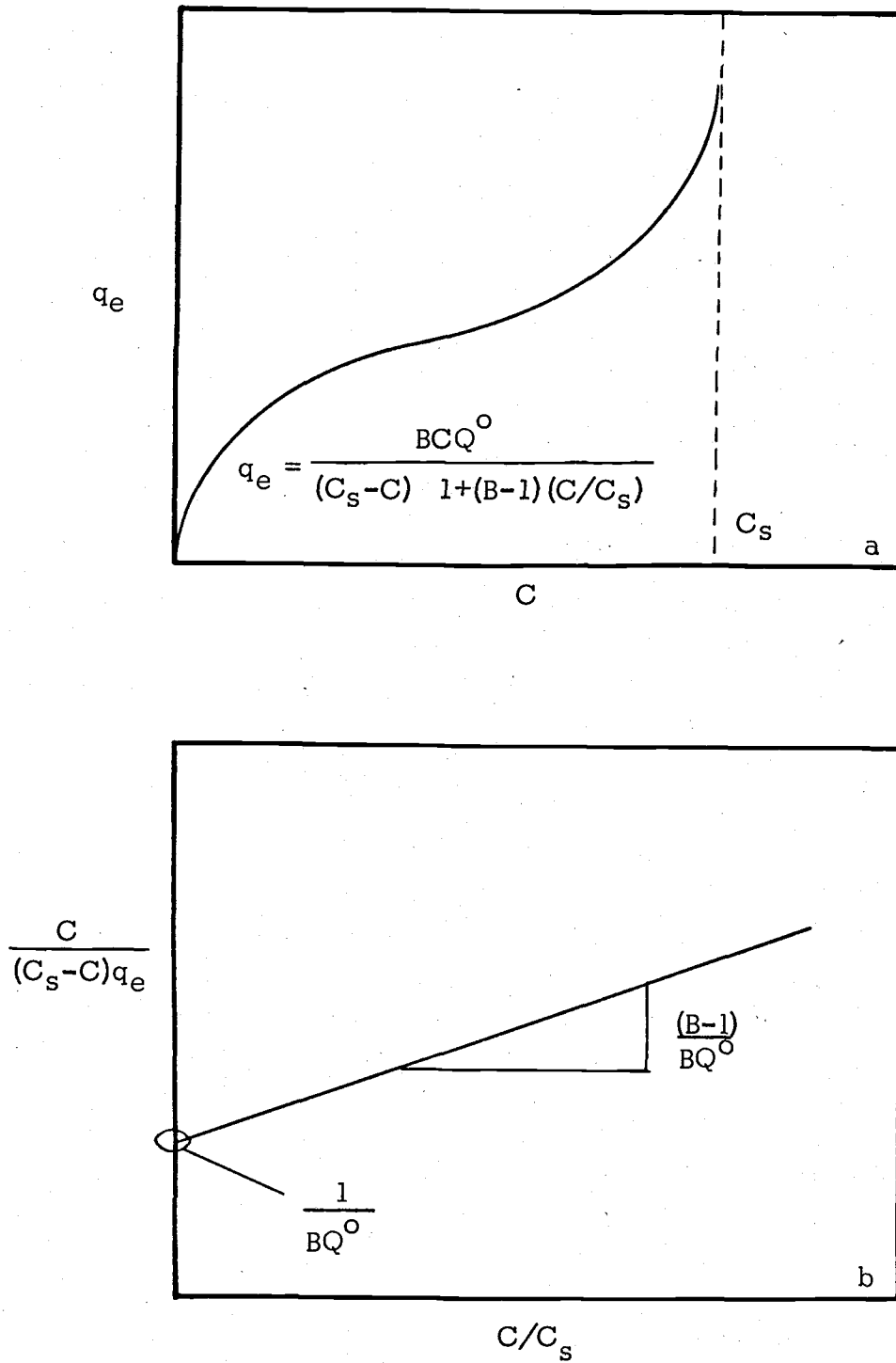
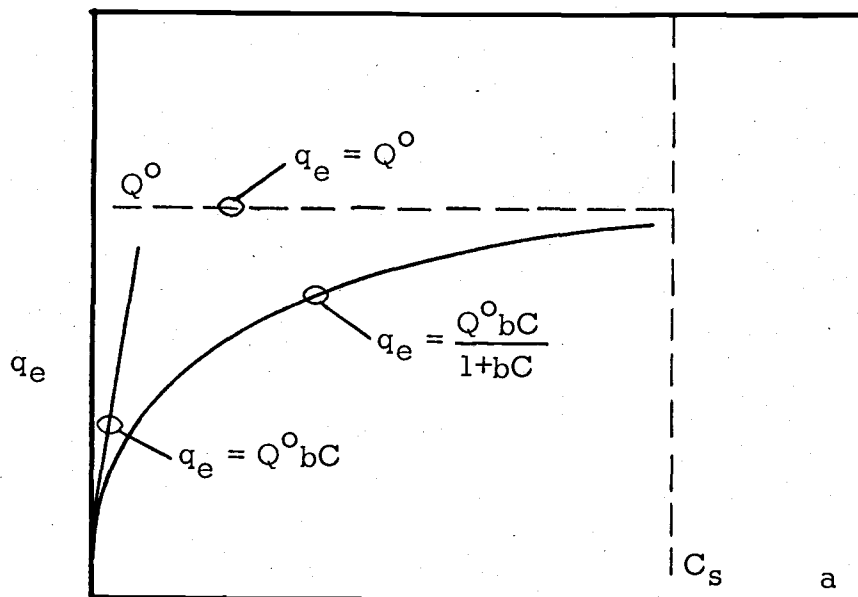


Figure 3.1, a. BET Adsorption Isotherm, q_e vs. C
 b. Linear Form of BET Adsorption Isotherm,
 $\frac{C}{(C_s - C)q_e}$ vs. C/C_s



C

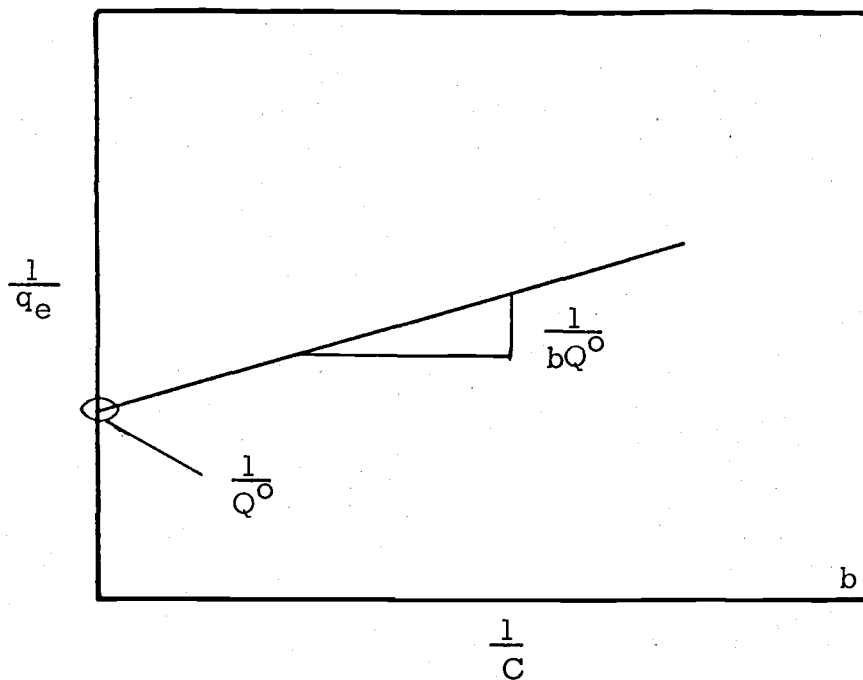


Figure 3.2, a. Langmuir Adsorption Isotherm, q_e vs. C

b. Linear Form of Langmuir Adsorption Isotherm,

$1/q_e$ vs. $1/C$

Equation 3.10, may be reduced to a linear form.

$$\frac{C}{(C_s - C)q_e} = \frac{1}{BQ^0} + \frac{B-1}{BQ^0} (C/C_s) \quad (3.11)$$

In Figure 3.1b, this straight line form is illustrated.

3.3.1.2 The Langmuir Isotherm

When the adsorption is primarily monolayer, the BET isotherm reduces to the Langmuir isotherm. This isotherm assumes monolayer adsorption and constant energy of adsorption. Equation 3.12 is the Langmuir adsorption relation.

$$q_e = \frac{Q^0 b C}{1 + bC} \quad (3.12)$$

where b is a constant related to the free energy of adsorption. Equation 3.12, may be linearized in the form:

$$\frac{1}{q} = \frac{1}{Q^0} + \frac{1}{bQ^0} * \left(\frac{1}{C}\right) \quad (3.13)$$

Equations 3.9 and 3.10 are illustrated in Figures 3.2a and 3.2b. The Langmuir isotherm can be further simplified. If the adsorbent is in excess (C is very small) then equation 3.12 becomes

$$q_e = Q^0 b C \quad (3.14)$$

and corresponds to the linear portion of the curve in Figure 3.2a. If the adsorbate is in excess, (C approaches C_s), then equation 3.12 becomes

$$q_e = Q^0 \quad (3.15)$$

which represents saturated monolayer adsorption.

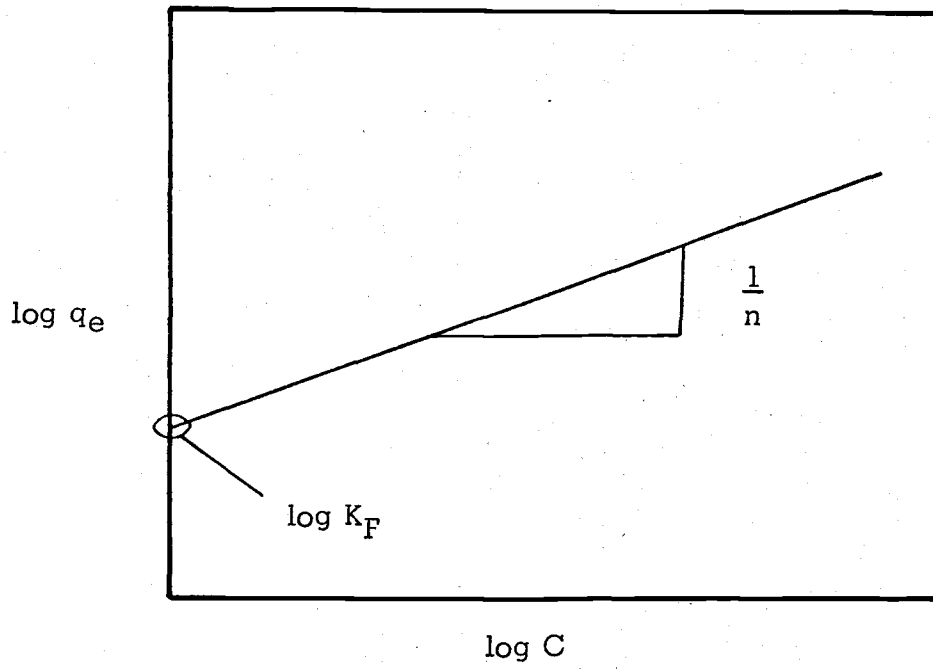


Figure 3.3, Linear Form of Freundlich Adsorption isotherm, $\log q_e$ vs. $\log C$

3.3.1.3 The Freundlich Isotherm

The Freundlich isotherm is a semi-empirical equation allowing for variations of surface adsorption energies. The Freundlich isotherm equation is

$$q_e = K_f C^{1/n} \quad (3.16)$$

where K_f and n are constants. The linear form of equation 3.16 is

$$\log q_e = \log K_f + \frac{1}{n} \log C \quad (3.17)$$

The straight line form is illustrated in Figure 3.3.

3.3.2 The Concentration Factor

Both the Langmuir and Freundlich isotherms can be further utilized in terms of a concentration factor, CF.

In the Langmuir isotherm, when the uptake of adsorbate by the adsorbent is linear (equation 3.14, Figure 3.2a) the slope of the line is a constant, Q^0/b . For this portion of the adsorption, the ratio of q_e to C must also be a constant. q_e/C represents the concentration of the adsorbate on the adsorbent per unit concentration of adsorbate in solution, and is dimensionally equivalent to the concentration factor, CF. CF is equal to a constant value only when the uptake of adsorbate is linear. As the concentration of the adsorbate in solution (C) increases, the uptake is no longer linear and CF decreases. At the point where $q_e = Q^0$, CF approaches zero. Therefore CF can be very misleading if not taken from the linear uptake phase of adsorption reactions.

The Freundlich isotherm can be manipulated similarly. If the adsorption is linear with respect to the adsorbate concentration in solution, then n approaches one and equation 3.16 reduces to

$$q_e = K_f C \quad (3.18)$$

CF would therefore be equivalent to K_f for linear adsorption.

3.3.3 Activated Sludge Metal Adsorption Model

Cheng et al. (1975) developed a model to relate ligand complexation and sludge adsorption of metals. The same equation may result from the following development. Metal in solution, M_s , is defined as the summation of the free metal ion, M , and the complexed metal in solution, ML_x . If M_s is thought of as the apparent equilibrium soluble concentration of the metal, then the equilibrium reaction for the metal-sludge adsorption may be expressed as



and the apparent stability constant of the metal-sludge complex is

$$K'_s = \frac{MS_y}{(M_s)(S)^y} \quad (3.20)$$

where

MS_y = the total metal bound to the sludge, moles metal

S = the concentration of MLSS, g/l,

y = the equivalent binding sites on the sludge per mole of metal, y = metal, mole/g, and

K'_s = the apparent stability constant of the reaction.

K'_s will change significantly if the character of complexing ligands in

the solution change, but for a given θ_c and a given influent substrate, K'_s should remain fairly constant.

Rearranging equation 3.20 gives

$$\log (MS_y/M_s) = \log K'_s + y \log S \quad (*) \quad (3.21)$$

which is linear for constant ligand concentration and pH. A log plot of MS_y/M_s versus S will yield the slope y and the apparent sludge complexation constant, K'_s , is the intercept.

(*) Please note that this equation utilizes S as the total MLSS, not the total minus MS_y .

4. EXPERIMENTAL PROCEDURES

4.1 Scope of Study

The experimental portion of this thesis was designed to accomplish the following specific objectives:

1. To establish the kinetics of adsorption
2. To establish the optimum adsorption pH
3. To determine whether or not adsorption is the primary removal mechanism
4. To determine if the metals adsorb competitively on activated sludge bacteria
5. To determine the effects of SRT upon metals adsorption

4.2 Experimental Apparatus

4.2.1 Complete-Mix Flow, Cell Recycle Reactor

The activated sludge cultures used in all experiments were developed in complete-mix, flow cell recycle reactors (Figure 4.1). This figure illustrates the aeration, mixing, internal settling, and influent and effluent aspects of the chemostat. Wasting was accomplished each day by removing the specified amount of solids from the culture to maintain the desired value of θ_c . Dissolved oxygen was maintained at approximately 2.0 mg/l.



Apparatus used for Complete-Mix, Continuous Flow Reactor with Cell Recycle.

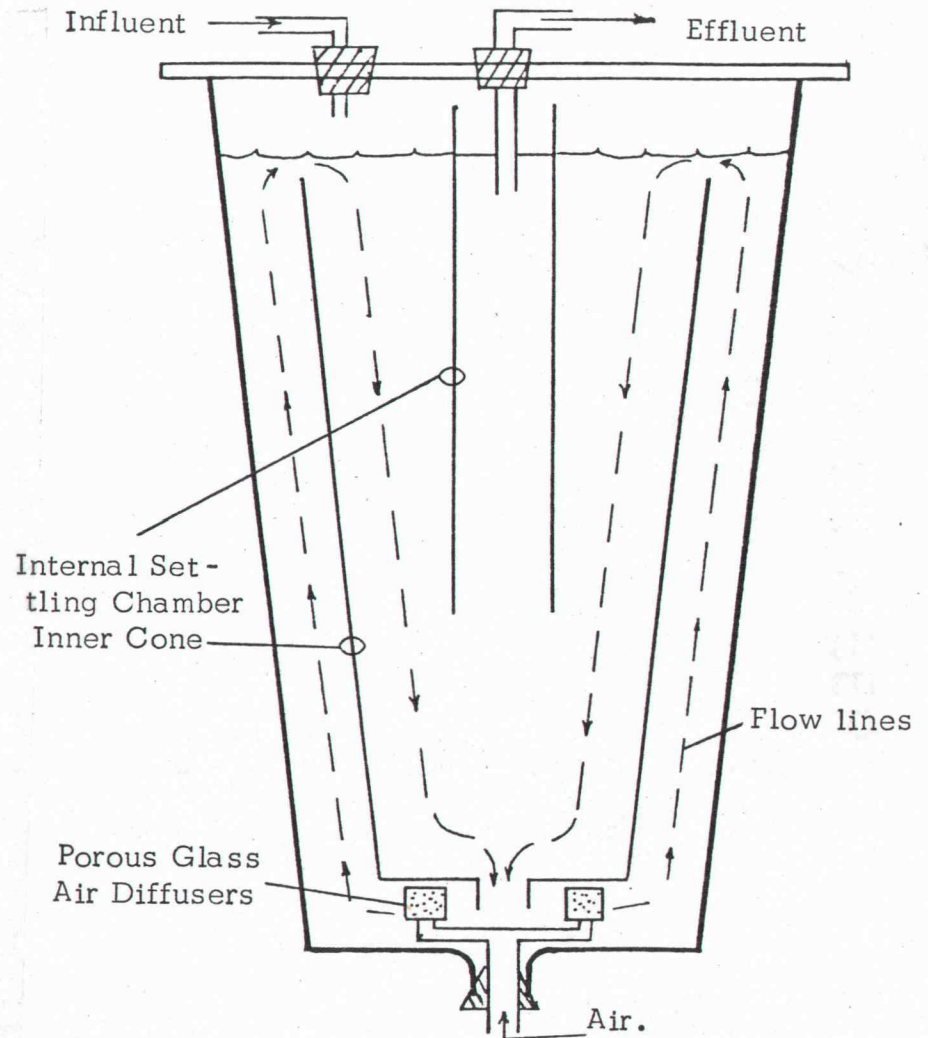


Figure 4.1, Schematic of Complete-Mix Continuous Flow Reactor with Cell Recycle

4.2.2 Complete-Mix Batch Reactors

Tests upon the steady-state cultures were performed using complete-mix batch reactors (Figure 4.2). Mixing was provided by filtered air delivered through a Pasteur capillary pipette. System pH was controlled by trial and error addition of NaOH or HNO₃ (0.1 N solutions).

4.3 Preparation of Cultures and Nutrient Feed

Laboratory reactors were seeded with activated sludge from the municipal sewage treatment plant, Albany, Oregon.

Nutrient feed was provided by using a combination of filtered primary effluent from the municipal wastewater treatment plant, Corvallis, Oregon and BBL Nutrient Broth stock solutions to achieve a nominal COD of 200 mg/l. The preparation procedure is as follows:

1. Primary effluent was collected, filtered through diatomaceous earth, and tested for the COD concentration.
2. Nutrient broth (BBL Nutrient Broth, 62.5 percent GelysateTM peptone, 37.5 percent Beef extractives) stock solution was prepared by dissolving approximately 20 g broth in glass-distilled water and autoclaving at 15 psi, 121°C for 15 minutes. This solution was tested for COD concentration, and stored at 4°C with no exposure to light.
3. The nutrient broth augmentation of the filtered primary effluent to reach 200 mg COD/l satisfied the following equation:

$$\frac{A * \text{vol}_A + B * \text{vol}_B}{\text{vol}_A + \text{vol}_B} = 200 \text{ mg COD/l} \quad (4.1)$$

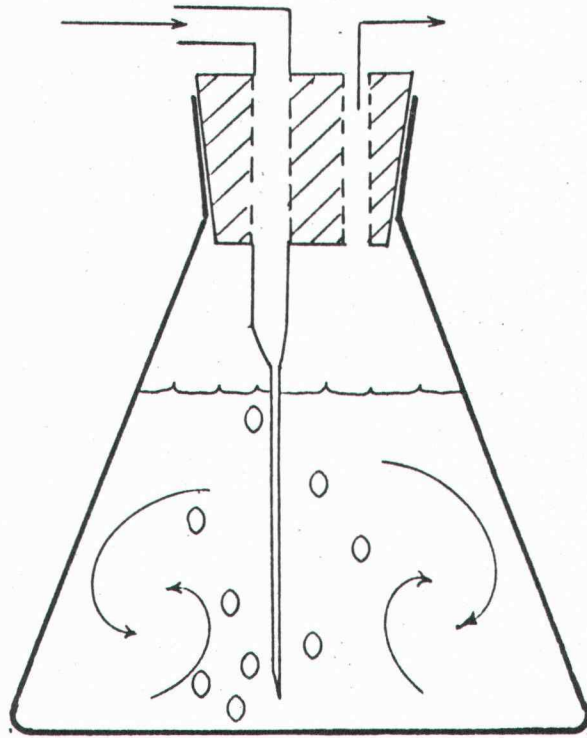


Figure 4.2

Schematic of Complete-Mix Batch Reactor

Batch Reactor Apparatus,
Laboratory Set-up

where A = the COD concentration in the nutrient broth solution,

vol_A = the volume of nutrient broth added to the primary effluent,

B = the COD concentration in the filtered primary effluent, and

vol_B = the volume of the filtered primary effluent.

The COD of the nutrient broth stock solution was high enough to render the volume of nutrient broth added negligible.

4. The final product was stored in a stainless steel tank at 4°C without exposure to light. Every two days, the tank was cleaned and the feed solution renewed.

5. The feed solution was buffered with 50 mg/l phosphate buffer to maintain an alkalinity of 50 to 100 mg/l as CaCO_3 and pH 6.8 to 7.2 in the reactors.

The feed was pumped into the reactors with peristaltic pumps at a rate adjusted to provide a hydraulic detention of 8 hours ($\theta_{\text{Hydraulic}} = 1/3$ day).

4.4 Program of Experimentation

4.4.1 Batch Experiments

Batch experiments were run as follows:

1. Batch tests apparatus were set up as shown in Figure 3.2
2. The metal dosage solutions were prepared at high concentration to minimize the volumes added to the culture.
3. 200 ml samples of the steady-state activated sludge culture were

placed in each reactor, dosed with metal, and pH was adjusted as necessary.

4. A control test was performed for each set of experiments using the same culture with no metals added.

Samples for metal adsorption analysis were withdrawn as outlined in Table 4.1:

TABLE 4.1

SCHEDULE OF SAMPLING

20 hr. test:		4 hr. test:	
Sample #	Time elapsed, hrs.	Sample #	Time elapsed, hrs.
1	0	1	0
2	1/2	2	1
3	1	3	2
4	5	4	4
5	10		
6	20		

When it was found that the 20 hour test achieved the steady state adsorption within 60 minutes, and maintained it throughout the test period, the test schedule was altered to the 4 hr. tests.

The sample size for all the tests was 15 ml. Batch tests were run under the conditions listed in Tables 4.2 and 4.3.

4.5 Measurement of Zinc, Cadmium, Copper, and Iron

4.5.1 Sample Preparation

The 15 ml samples were withdrawn from the batch reactors by a

TABLE 4.2

SCHEDULE OF TESTS

Test No. *	θ_c	pH	Dosage
1	1	7	A
2	5	4	A
3	5	5	A
4	5	5.6	A
5	5	7.4	A
6	5	9	A
7	5	10	A
8	5	7	B
9	5	7	C
10	5	7	D
11	5	7	E
12	15	7	A
13	5	7	A**

*See appendix for raw data

**Autoclaved culture

TABLE 4.3

DOSAGES

Dosage label	metals dosage, mg/l		
	COPPER	ZINC	CADMIUM
A	1	1	0.5
B	10	10	5
C	100	100	50
D	0	100	0
E	0	0	50

volumetric pipette with an enlarged tip to insure against selective solids removal.

4.5.1.1 Total Metals

The following procedure was followed to prepare the total metals for atomic absorption spectroscope (AA) determination:

1. To a 15 ml sample, add 5 ml of concentrated distilled HNO_3 .
2. Evaporate to dryness in sand bed.
3. Repeat steps 1 and 2.
4. Cool to room temperature.
5. Add 3 drops concentrated HCl and enough 2% HNO_3 to bring the volume to 15 ml.
6. Wait for 30 minutes to resolubilize the metals.
7. Store the digested sample in nalgene bottles for AA determination.

4.5.1.2 Soluble Metals

1. Add 1/2 ml of distilled concentrated HNO_3 to an acid washed test tube.
2. Use a millipore filtration apparatus with a 0.45 μm filter that has been rinsed with 20 ml glass distilled water. Sample was filtered, and filtrate collection was provided by the test tube.
3. Filter the 15 ml sample until the membrane pad is nearly dry.
4. Remove the filtrate portion, and store for AA determination.

4.5.2 Preparation of Filter Blanks

Duplicate blanks for the filter membranes were run periodically to test for the metals content of the pads. A pad was prepared by filtering 20 ml of double distilled water through it, and then digesting it as in the total metals, section 4.5.1.1. The digested sample was redissolved to a volume of 15 mls and stored in nalgene bottles for AA determination.

4.5.3 Atomic Absorption Test Methods

All test methods used in this investigation are outlined in Standard Methods, part 300, section 301 (14th Edition, 1975).

4.6 Suspended Solids Measurement

Suspended solids were performed daily for SRT adjustments. They were needed to determine the amount of wasting required to achieve the given sludge age. The methodology for this test is as follows:

Apparatus: The apparatus used was a millipore filtration unit, glass.

Procedure:

1. Place a 0.45 μ filter pad into an aluminum weighing dish.
2. Dry at 103°C to 105°C for 4 hours.
3. Remove dish and pad and let cool in a desiccator.
4. Weigh immediately before use, and record the tare weight.

5. Filter sample volume, with rinsing of the funnel sides to get all solids onto the pad.
6. Remove filter pad with teflon tongs and replace in the same weighing dish.
7. Dry at 103-105°C for 4 hours.
8. Store in desiccator until cool.
9. Weigh pad and dish and record this value at tare + solids.

Calculation:

$$\text{mg/l suspended solids} = \frac{(B - A) * 1000}{\text{vol sample, ml}}$$

where A = tare weight, and

B = tare + solids.

4.7 Metals Determination Procedure

4.7.1 Instrument Description

Perkin Elmer Model 360 Atom Absorption Spectrophotometer with standard flame atomization was used for all metals determinations.

4.7.2 Operations and Control Settings

The operation of the unit was performed as described in the manufacturer's operating manual. The control settings used were as follows:

Metal	Wavelength, nm	Gas Mixture	Sensitivity check *
cadmium	228.8	air-acetylene	1.2
copper	324.7	"	4
zinc	213.9	"	0.8

*The sensitivity check is done by testing the concentration of the metal at this level. The instrument should read approximately 0.2 absorption units.

4.8 Reagents

4.8.1 Stock and Standard Solutions of Cadmium, Copper, Zinc, and Iron

All stock and standard solution of the metals were prepared as described in Standard Methods (Section 301.A 14th Edition, 1975). Stock solutions were prepared at 1000 mg/l. Standard solutions were prepared daily at the following concentrations: 0.1, 1, 2, 5, and 10 mg/l for instrument calibration.

4.8.2 Reagent Purification

All water used in cleaning and solution make-up water, was double distilled, with the second distillation through a glass still. All concentrated HNO_3 was distilled through a glass apparatus before use.

4.8.3 Glassware and Plastics Cleaning

Glassware and plastics were cleaned by soaking overnight in 6 N HNO_3 . It was then rinsed 6 times in double distilled water and allowed to dry.

4.9 COD Tests

The test used is described in Standard Methods, test 508, (14th Edition, 1975).

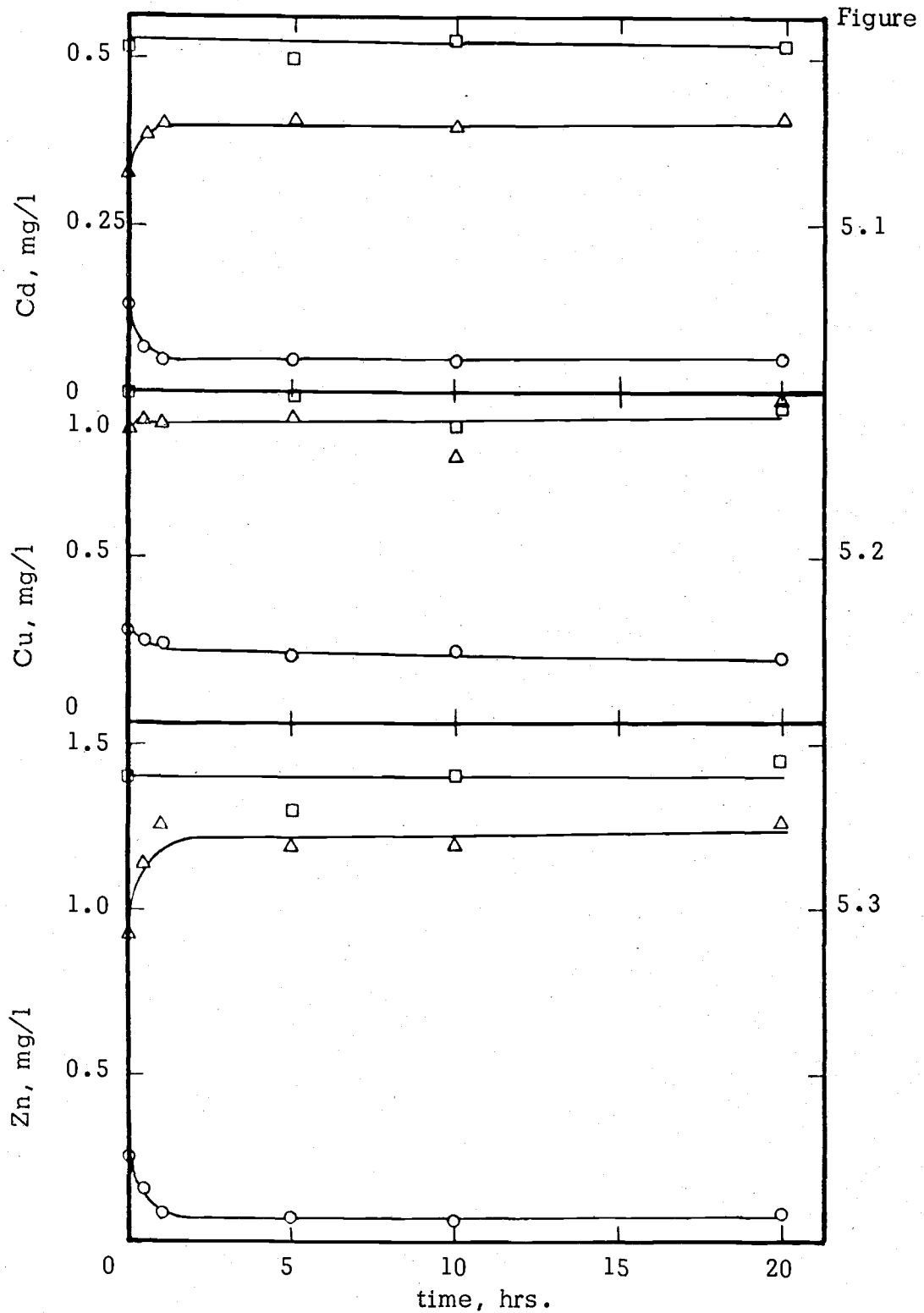
5. RESULTS

5.1 Adsorption Kinetics

In order to study the rate of uptake of metals by activated sludge bacteria, adsorption experiments over a 20 hour period were conducted (experiment 5). Samples withdrawn over a long time period showed that adsorption occurred swiftly and nearly to completion within the first 60 minutes of contact, and that subsequent uptake was minimal (Figures 5.1, 5.2, and 5.3).

Mass balances on total metals added (equal to soluble plus suspended metals) indicated unaccounted for losses of up to 15 percent for copper in the total metals determination. Cadmium and zinc mass balances indicate a gain of 7.5 and 6.8 percent, respectively, for total metals. Because of these differences, total metals in the kinetics study were calculated from the summation of soluble plus suspended metals. This "total metals" value remained constant throughout this experiment. Calculated in this way, copper removal was 80 percent. Cadmium removal was 88 percent and zinc removal was 95 percent (see Figures 5.1-5.3). Iron was totally associated with the solid phase of the culture in all experiments at a concentration of about 4.5 mg/l. Iron was not dosed in the experiments, and was monitored to determine the possible role of iron oxides adsorption uptake of other heavy metals by the solid phase.

Subsequent tests (except for high dosage experiments 8, 9, 10,



Figures 5.1, 5.2 and 5.3: Rate of Metal Uptake by Activated Sludge Solids

and 11), had mass balance correlations to within five percent and the totals were not adjusted.

5.2 pH

The adsorption of metals by activated sludge is highly affected by system pH. To investigate this dependence, pH was varied from about 4 to 10 in batch experiments on $\theta_c = 5$ day cultures dosed with 1 ppm zinc, 1 ppm copper, and 0.5 ppm cadmium (experiments 2, 3, 4, 5, 6, and 7). The effects of the pH variation on adsorption are shown in Figures 5.4 and 5.5. In order to normalize the varying solids concentrations for the pH adsorption experiments, the percent removal of metals was calculated as follows:

$$\text{percent removal} = \frac{Me_{SS}/MLSS}{Me_{TOT}/MLSS} \times 100 \quad (5.1)$$

where Me_{TOT} = the total amount of metal, suspended and soluble, mg/l, and Me_{SS} = the metal in the solid phase, mg/l.

The results indicate optimum adsorption of copper at approximately pH 7, and that the concentrations of cadmium and zinc attached to the solid phase increases with increasing pH. Figure 5.5 shows that zinc may begin to precipitate between pH 9 and 10, since the zinc CF increases steadily as pH increases. Cadmium, in contrast, shows a maximum CF at pH 9, and copper shows the same maximum adsorption pH as indicated in Figure 5.4 (pH 7).

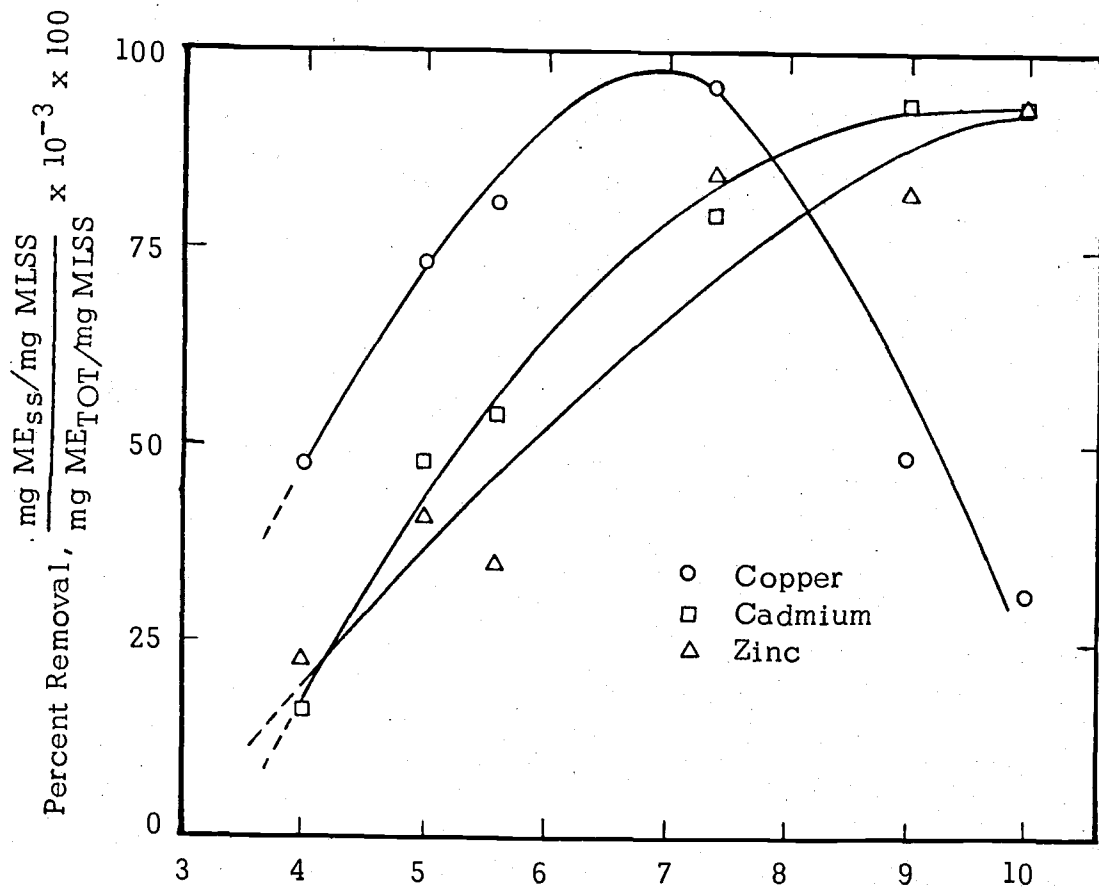


Figure 5.4 Percent Removal of Metals by Activated Sludge Solids vs. pH

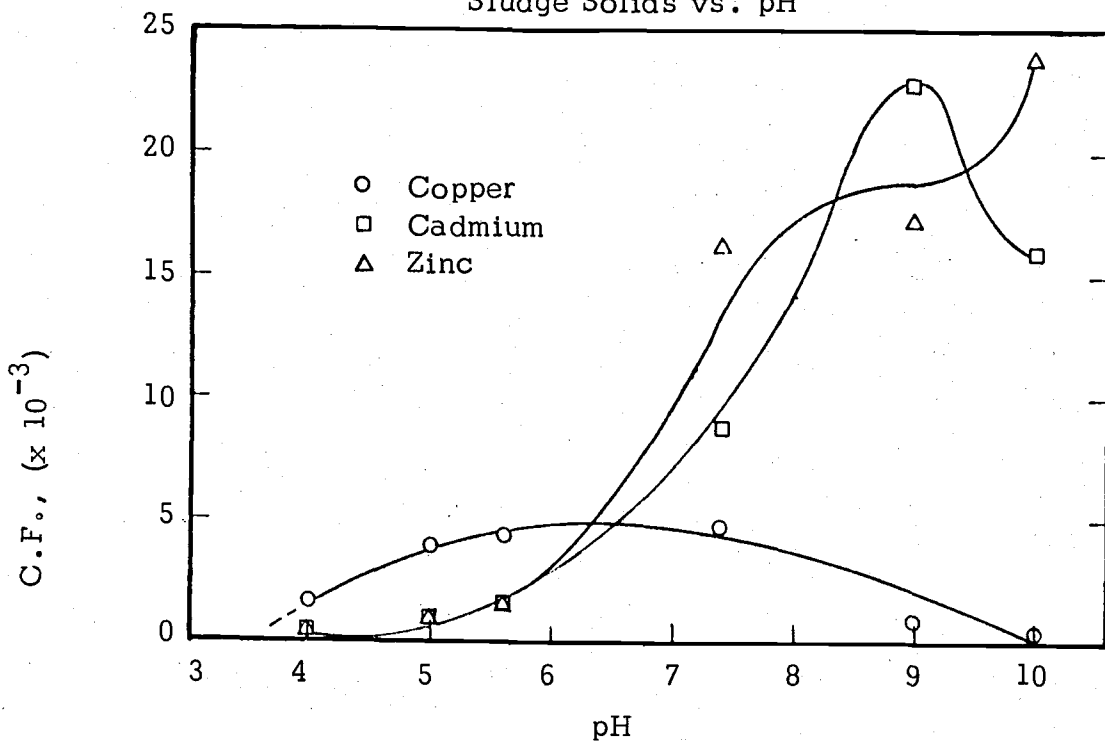


Figure 5.5 Concentration Factor, CF, vs. pH

5.3 Autoclaved Versus Live Cultures -- Adsorption Comparison

To determine if the primary mode of metals removal was adsorption, a metals uptake determination was conducted upon an autoclaved sample ($\theta_c = 5$ days, experiment 13). Because the suspended solids concentration for this experiment was 30 percent lower than that for the standard test (experiment 5), the data was normalized as previously described (section 5.2). Cadmium and zinc uptake by live cultures were essentially unchanged, 3 and 8 percent lower, respectively, than the corresponding uptake of the metals by the autoclaved cultures. Copper adsorption by live organisms was 22.8 percent greater than by autoclaved organisms (Figure 5.7).

5.4 Competitive Versus Noncompetitive Adsorption

Adsorption of cadmium and zinc as single solutes is illustrated in Figures 5.8 and 5.9. At a dosage of 50 ppm, cadmium removal by the activated sludge bacteria was 44 percent, while 57.7 percent of the zinc (dosed at 100 ppm) was adsorbed by the solids (Figure 5.6).

Copper adsorption experiments always contained zinc and cadmium in the ratio of Cu:Zn:Cd = 1.0:1.0:0.5, and single solute adsorption data for copper was not available.

When the three metals were dosed simultaneously, the results differed from those obtained for single solute adsorption. At a dosage of 100 ppm copper, 100 ppm zinc, and 50 ppm cadmium, only 1.5 percent

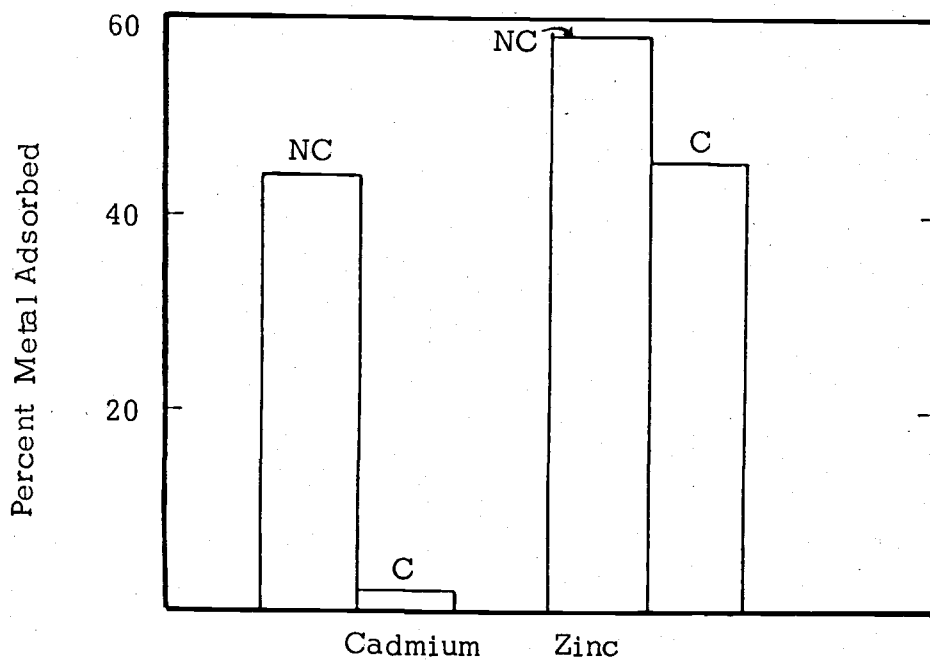


Figure 5.6 Competitive (C) vs. Noncompetitive (NC) Adsorption by Activated Sludge Solids

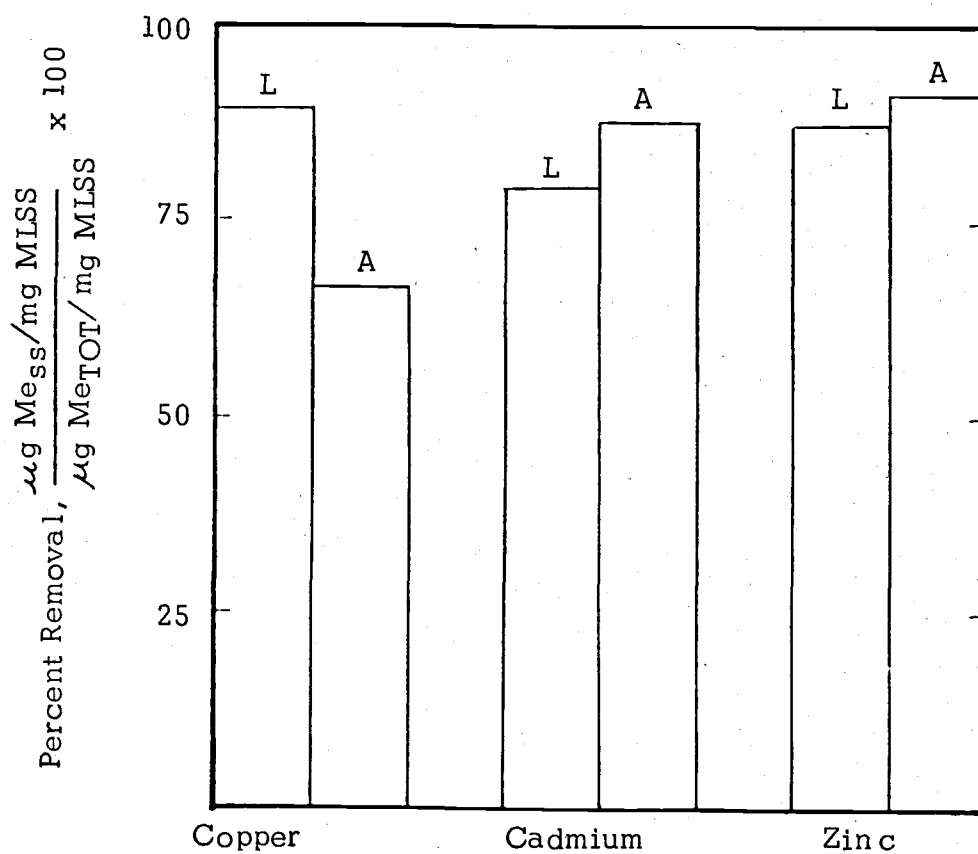


Figure 5.7 Comparison of Percent of Metals Removal by Live (L) and Autoclaved (A) Activated Sludge Solids

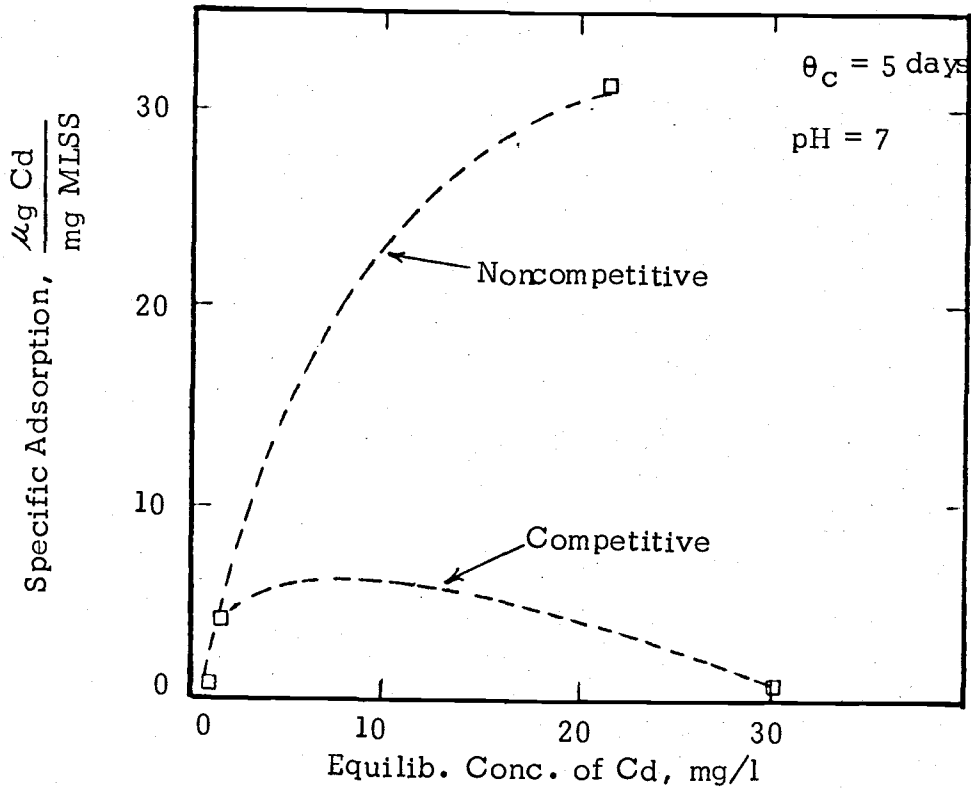


Figure 5.8 Cadmium Adsorption by Activated Sludge Bacteria

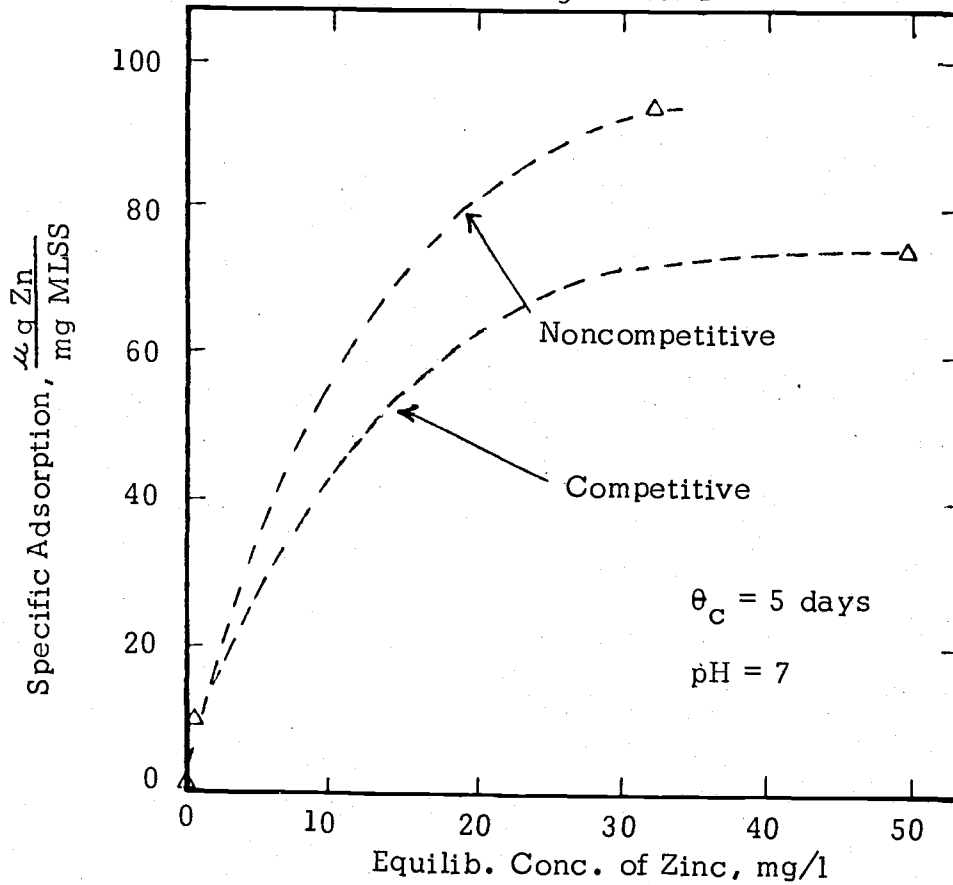


Figure 5.9 Zinc Adsorption by Activated Sludge Bacteria

of the cadmium and 44.5 percent of the zinc was adsorbed (see Figure 5.6). This constitutes 97 percent and 23 percent decrease in adsorption of cadmium and zinc, respectively. Because of limited data points, the observed adsorption phenomenon are only tentative, and are described in section 5.6.

The competitive adsorption experiments performed in this investigation were not typical in that all the metal concentrations in solution were increased by the same ratio rather than one metal concentration increasing while the others remained constant.

Figures 5.8 and 5.9 indicate that cadmium and zinc adsorption was decreased in competition with the other metals added. In these figures, the apparent Q^0 for the respective metals is lowered, though available data gives only tentative estimates.

5.5 Concentration Factor and θ_C

Concentration factor (CF) comparison was used only for data which occurred in the linear region of the adsorption isotherm (see Section 3.2). This limited the comparisons to tests of dosage "A" (1 ppm Cu, 1 ppm Zn, and 0.5 ppm Cd). Examination of Figure 5.11 shows the effects of varying θ_C upon the concentration factor. CF for zinc and cadmium decreases sharply with increasing θ_C (1 to 5 days). Subsequent change to $\theta_C = 15$ days causes only a minimal decrease in CF. The CF of copper adsorption however, showed a small but steady increase with increasing θ_C values.

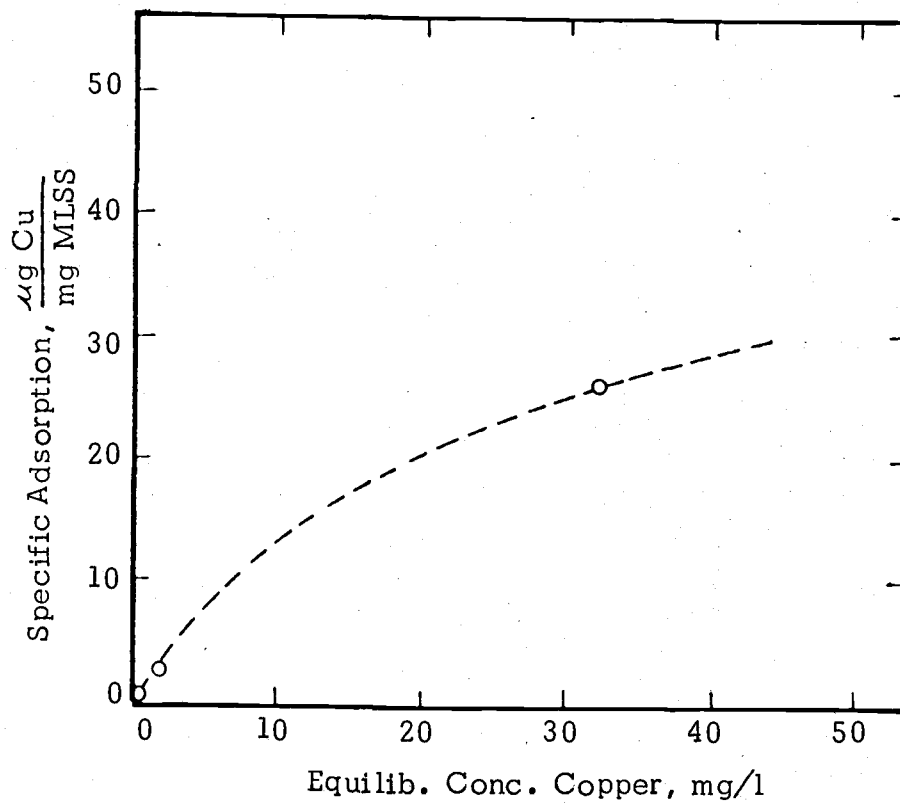


Figure 5.10. Copper Adsorption by Activated Sludge Solids

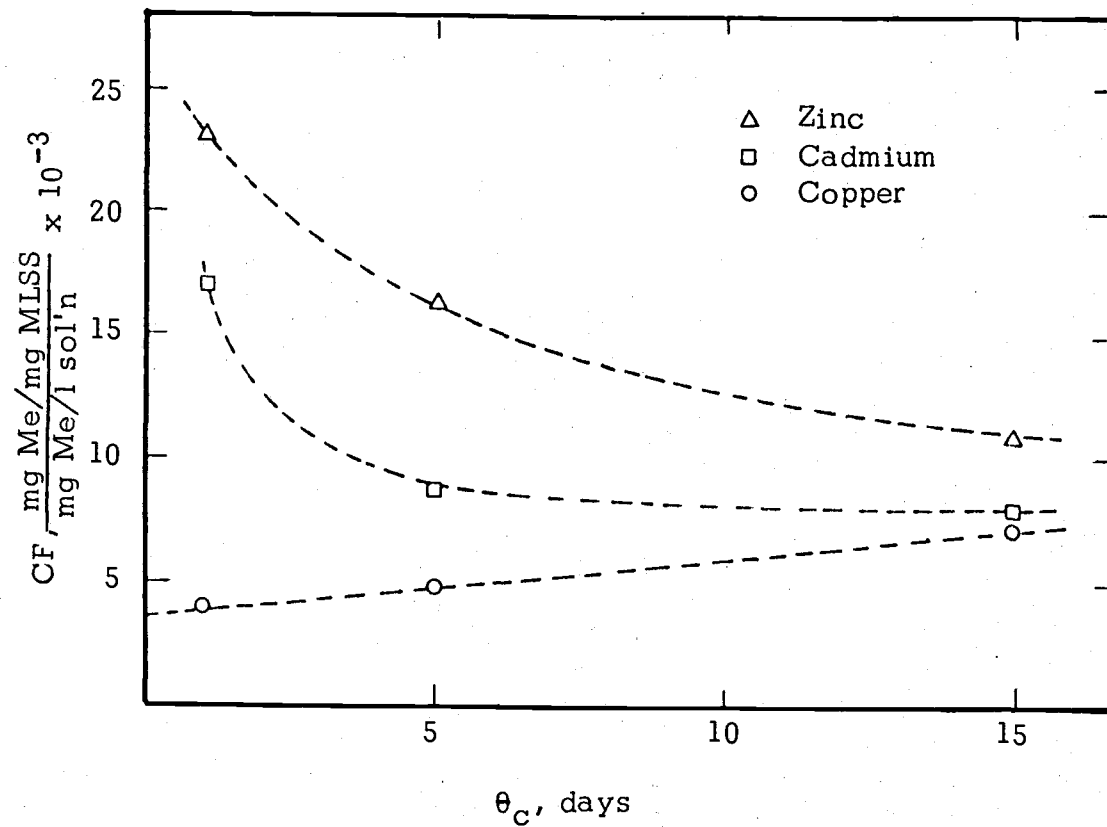


Figure 5.11 Concentration Factor, CF vs. θ_c

5.6 Adsorption Isotherms

Either Langmuir or Freundlich isotherms fit the data equally well. For interpretation purposes, the Langmuir equation was selected in this study. However, Freundlich isotherms and calculations of constants are included in Appendix II.

From the linearized Langmuir plot, Q^0 and b values were empirically determined and are listed in Table 5.1. Q^0 values were in the range of 50 to 100 ($\mu\text{g metal/mg solids}$) and b values ranged from a low of 0.05 for copper + 0.309 for zinc. These values are only tentative due to the low number of data points.

TABLE 5.1

EMPIRICAL DETERMINATION OF LANGMUIR CONSTANTS Q^0 AND b
(Figures 5.12, 5.13, and 5.14)

Metal	Q^0 g metal/mg solids	b
Copper	50 to 100	0.05 to 0.1
Cadmium	50 to 100	0.093 to 0.186
Zinc	50 to 100	0.309 to 0.618

5.7 Metal-Sludge Stability Constants

Equation 3.20 was used as a basis for analyzing metal sludge "adsorption complexes." The linearized form, equation 3.21, was used to generate metal-sludge apparent adsorption constants as illustrated

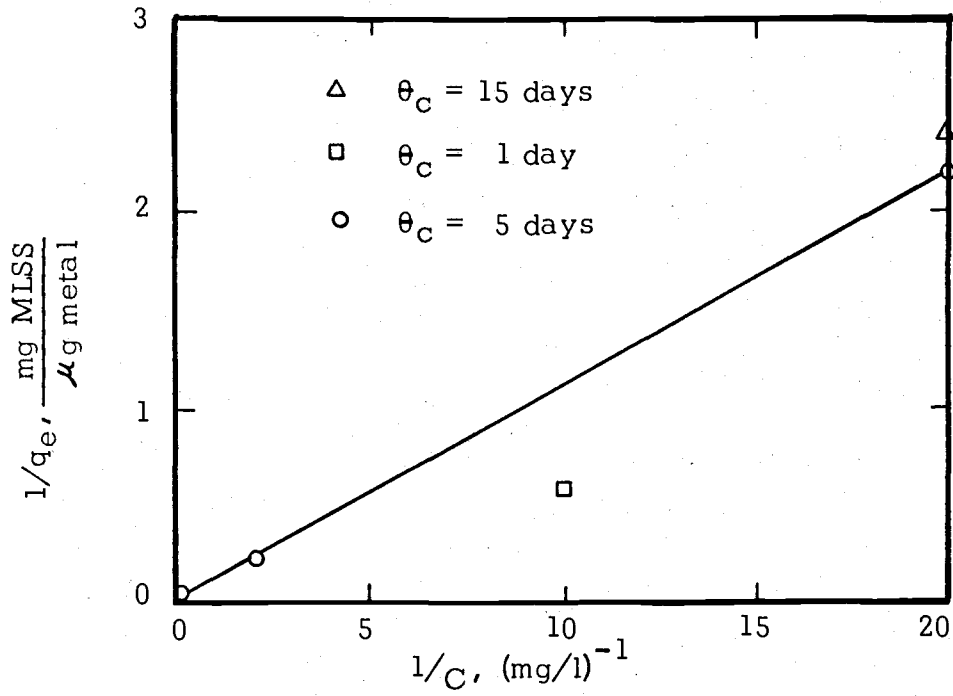


Figure 5.12 Linearized Langmuir Adsorption Isotherm for Cadmium

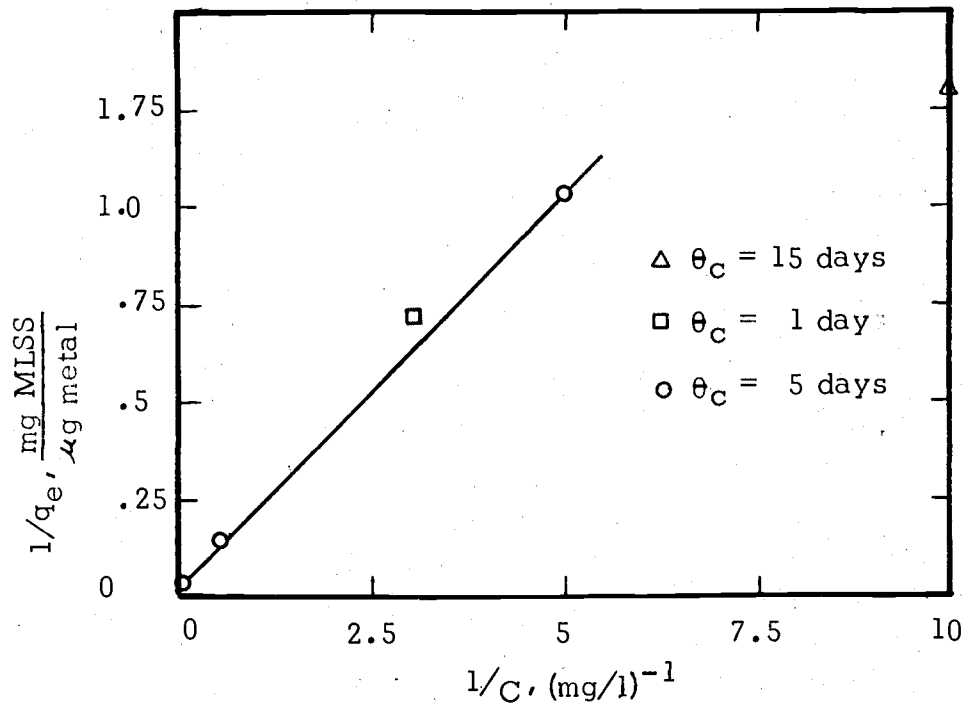


Figure 5.13 Linearized Langmuir Adsorption Isotherm for Copper

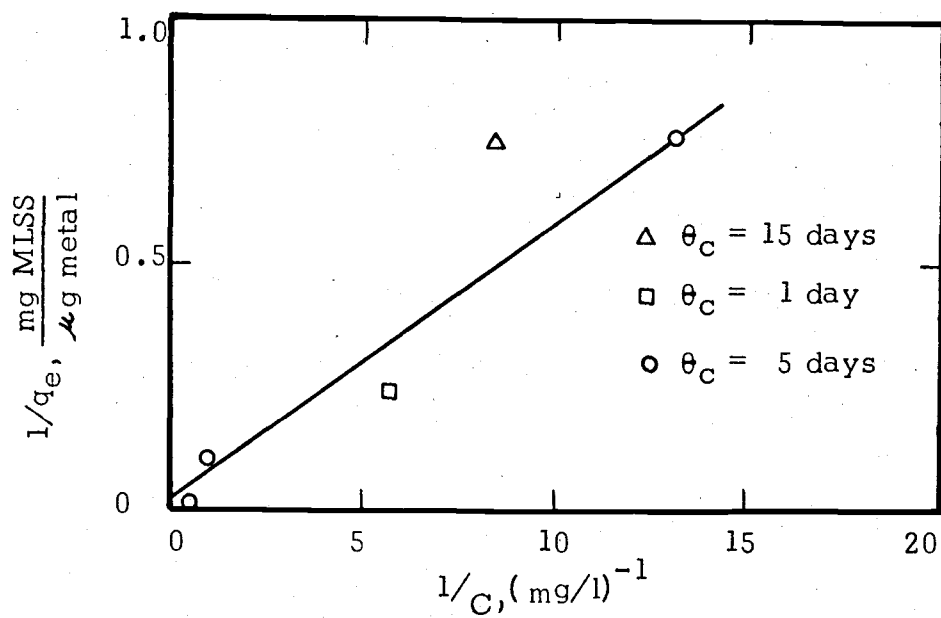


Figure 5.13a Linearized Langmuir Adsorption Isotherm for Zinc

in Figures 5.14, 5.15, and 5.16. Table 5.2 summarizes these constants. The difference between competitive and noncompetitive adsorption for cadmium is demonstrated by the lower stability constant (K'_s), for the competitive studies (Table 5.2). The difference between competitive and non-competitive adsorption of zinc is not so apparent. The equivalent binding sites on the sludge per mole of metal (y) are also listed in Table 5.2. Copper and zinc apparently have much smaller y values than does cadmium.

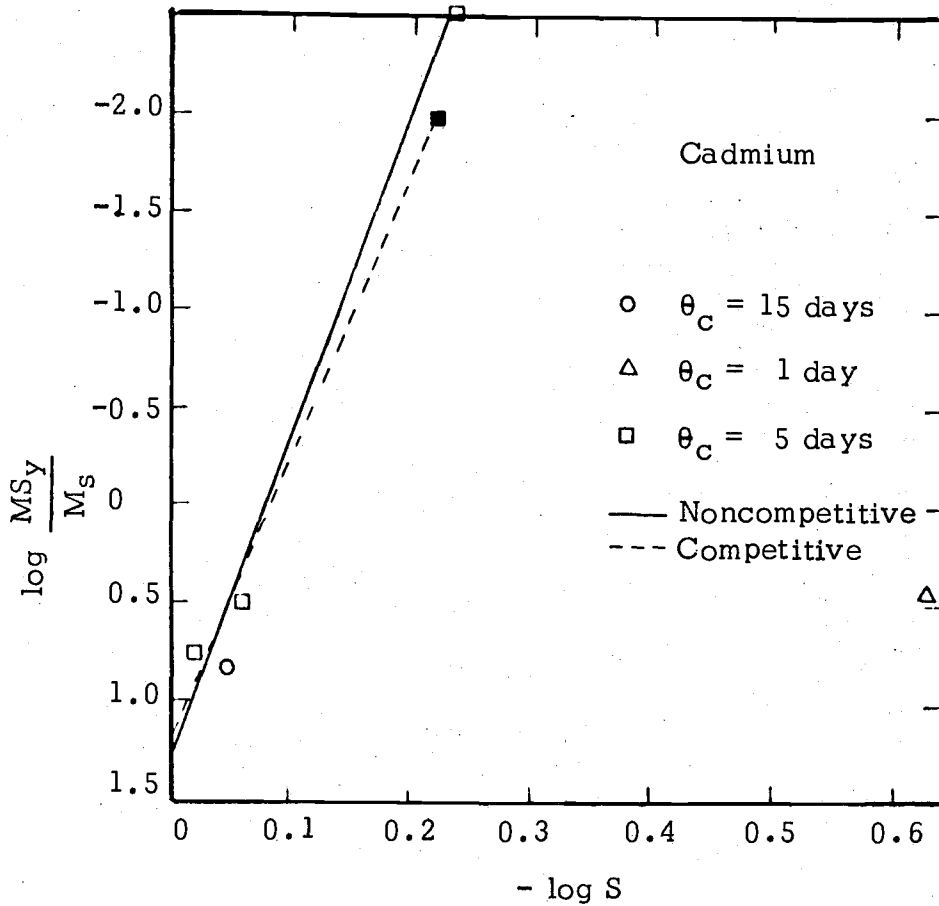


Figure 5.14 Linearization of Metal-Sludge Complexation for Cadmium

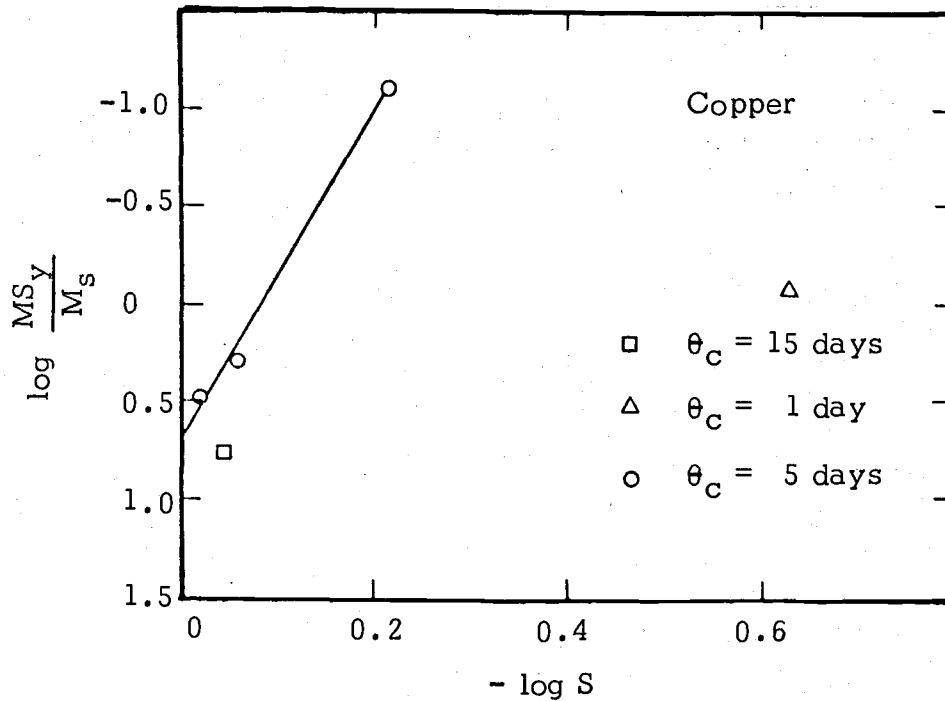


Figure 5.15 Linearization of Metal Sludge Complexation for Copper

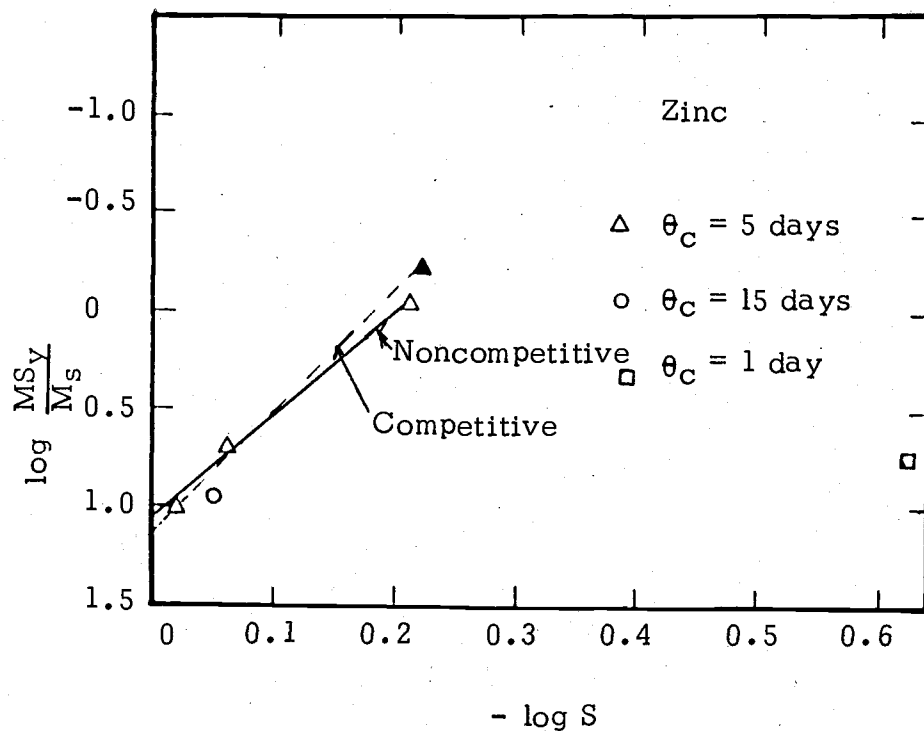


Figure 5.16 Linearization of Metal-Sludge Complexation for Zinc

TABLE 5.2

K_s VALUES DETERMINED EMPIRICALLY
(from Figures 5.14, 5.15, and 5.16)

Metal	$K_s, (1/g)^y$		$y, \frac{\text{equivalent sites/mole metal}}{\text{g solids}}$	
	noncompetitive	competitive	noncompetitive	competitive
Copper	---	4.4	---	8.3
Zinc	11.2	13.3	6.25	5.4
Cadmium	17.78	14.13	14.8	16.8

6. DISCUSSION OF RESULTS

6.1 Metals Partitioning by Control of pH and θ_c

6.1.1 Management of Metals: Partitioning Options

The two possibilities for management of metals in sewage are dependent upon the ultimate use of either the sludge or the effluent. In the case in which the sludge is to be used in land applications as a fertilizer or as a soil conditioner, metal concentrations of sludge must be held to a minimum. Conversely, reuse of sewage effluents for irrigation, or disposal into metal-sensitive waterways (e.g. sensitive due to trophic level organism toxicity or bioaccumulation, or eutrophication problems) must have minimum metals concentrations.

These partitioning options are mutually exclusive; it is not possible to have a minimum sludge-metal concentration and a minimum effluent metal concentration simultaneously. Minimum sludge-metal concentrations may be achieved only by sacrificing effluent quality and the converse is also true. Consequently, the effects of operational parameters (θ_c and pH) must be utilized in order to manage the metal distribution.

6.1.2 Management of Metals Partitioning Through Control of pH and θ_c

For the cases in which it is desirable to minimize the removal of heavy metals by activated sludge bacteria, the lowest possible

operating pH should be maintained for zinc and cadmium wastes and neutral pH values (6.8 to 7.2) should be avoided for wastes containing copper (see Figures 5.4 and 5.5, section 5.2).

Conversely, for maximum removal of zinc and cadmium metals by sludge bacteria, the pH should be maintained at maximum possible operating value, while copper removal is optimized at approximately pH 7 (Figures 5.4 and 5.5). Copper removal is also increased, somewhat, at high θ_c values, while zinc and cadmium removal is maximized at θ_c values less than 5 days (Figure 5.11).

6.2 Adsorption as the Primary Removal Mechanism

Adsorption is assumed to be the primary mechanism of metals removal by activated sludge bacteria due to the kinetics of uptake (Figures 5.1, 5.2, and 5.3) and because of the observed adsorption phenomenon for the autoclaved vs. live experiments (experiments 5 and 13, Figure 5.7). Kinetic studies showed rapid uptake characteristic of adsorption which is consistent with Cheng et al. (1975) and Parks, (1975). Zinc and cadmium removals by live cultures were very similar to, though slightly lower than, their respective removals by the autoclaved sludge, while copper removal by live organisms was 22.8 percent greater than by autoclaved organisms. This phenomenon could indicate the complexation of copper with the significant amounts of organics which are released by organisms when autoclaved. Copper is more strongly complexed by organic or cadmium or zinc, c, and thus is affected more.

6.3 Hypothetical Distribution of Metals as Determined from Empirical K'_s Values and Assumed Ligand Concentrations

In order to demonstrate the application of metal-sludge stability constants, a hypothetical speciation of metals in an activated sludge environment was calculated using empirically determined K'_s constants for metal-sludge "complexation" relationships.

The composition of the aqueous phase utilized for metals distribution calculations is given in Table 6.1 and is typical for weak wastewaters (Metcalf and Eddy, 1972). From this composition, the soluble metal complexes to be considered for speciation calculations were determined and are listed in Table 6.2 with the respective stability constants listed in Table 6.3. From this information it was possible to delineate the predominant species for a given metal under conditions for which the stability constant was determined (K'_s given for pH 7, $\theta_c = 5$ days, $Cu_{TOT} = 1$ ppm, $Zn_{TOT} = 1$ ppm, $Cd_{TOT} = 0.5$ ppm, and weak wastewater).

Tables 6.4 through 6.9 list the distribution fractions (percent) of predominant species for copper, cadmium and zinc. All species listed in Table 6.2 which were not listed in Tables 6.4 through 6.9 had negligible fractions.

6.3.1 Hypothetical Copper Distribution

The hypothetical distribution of copper shown in Table 6.4 was comprised of 5 major species (greater than 10 percent of the total):

TABLE 6.1Assumed Values for Domestic Sewage Composition^a

$$\text{Cl}^- = 30 \text{ mg/l} = 8.5 \times 10^{-4} \text{ M}$$

$$\text{NH}_3 - \text{N} = 12 \text{ mg/l} = 8.57 \times 10^{-4} \text{ M}$$

$$\text{OrgN} = 8 \text{ mg/l} = 5.7 \times 10^{-4} \text{ M}^{\text{b}}$$

$$\text{Alk as CaCO}_3 = 50 \text{ mg/l} = 5 \times 10^{-4} \text{ M}$$

$$\text{Sulfate} = 10 \text{ mg/l}^{\text{c}} = 1.04 \times 10^{-4} \text{ M}$$

$$\text{Solids} = 0.945 \text{ g/l}$$

^a Metcalf and Eddy, 1972 unless otherwise listed

^b Assumed to be all fulvic acid

^c estimate

TABLE 6.2
List of Species Considered in Speciation
Calculation for Activated Sludge En-
vironment

Copper:	$\text{Cu}(\text{Cl})^+$, $\text{Cu}(\text{Cl})_2$, $\text{Cu}(\text{OH})^+$, $\text{Cu}(\text{OH})_2$, $\text{Cu}(\text{OH})_3^-$, $\text{Cu}(\text{OH})_1^-$, $\text{Cu}(\text{NH}_3)^{2+}$, $\text{Cu}(\text{NH}_3)_2^{2+}$ $\text{Cu}(\text{NH}_3)_3^{2+}$, $\text{Cu}(\text{NH}_3)_4^{2+}$, $\text{Cu}(\text{CO}_3)$, $\text{Cu}(\text{CO}_3)_2^-$, $\text{Cu}(\text{HCO}_3)^+$, $\text{Cu}(\text{Fulvic Acid})$, $\text{Cu}(\text{bacteria})$, Cu^{2+}
Cadmium:	$\text{Cd}(\text{Cl})^+$, $\text{Cd}(\text{Cl})_2$, $\text{Cd}(\text{Cl})_3^-$, $\text{Cd}(\text{Cl})_4^-$, $\text{Cd}(\text{OH})^+$, $\text{Cd}(\text{OH})_2$, $\text{Cd}(\text{OH})_3^-$, $\text{Cd}(\text{OH})_4^-$, $\text{Cd}(\text{NH}_3)^{2+}$, $\text{Cd}(\text{NH}_3)_2^{2+}$, $\text{Cd}(\text{NH}_3)_3^{2+}$, $\text{Cd}(\text{NH}_3)_4^{2+}$ $\text{Cd}(\text{CO}_3)$, $\text{Cd}(\text{bacteria})$, Cd^{2+} $\text{Cd}(\text{HCO}_3)^+$
Zinc:	$\text{Zn}(\text{Cl})^+$, $\text{Zn}(\text{Cl})_2$, $\text{Zn}(\text{OH})^+$, $\text{Zn}(\text{OH})_2$, $\text{Zn}(\text{OH})_3^-$, $\text{Zn}(\text{OH})_4^-$, $\text{Zn}(\text{NH}_3)^{2+}$, $\text{Zn}(\text{NH}_3)_2^{2+}$, $\text{Zn}(\text{NH}_3)_3^{2+}$, $\text{Zn}(\text{NH}_3)_4^{2+}$, $\text{Zn}(\text{CO}_3)$, $\text{Zn}(\text{HCO}_3)^+$, $\text{Zn}(\text{Fulvic Acid})$, $\text{Zn}(\text{bacteria})$, Zn^{2+}

TABLE 6.3

Stability Constants for Metal-Ligand
Complexes Used in Speciation Cal-
culations (25°C)

Metal	log β Me-Ligand Stability Constant ^(a)						
	Solids	OH ⁻	Cl ⁻	NH ₃	CO ₃ ⁼	HCO ₃ ⁻ ^(b)	Fulvic Acid ^(c)
Copper	0.643	6.0	0.6	5.8	6.1	2.7	-1.36
		17.1	0.4	10.7	9.1	-	
		14.0		14.7			
		16.5		17.6			
Cadmium	1.15	3.7	2.0	2.5	3.5	2.1	N/A ^(d)
		7.7	2.7	4.5			
		8.7	2.7	6.0			
		8.7	2.4	6.8			
				6.5			
			4.8				
Zinc	1.12	5.0	-0.5	2.2	3.9	2.1	-1.0
		11.1	-1.0	4.5			
		13.6		7.0			
		14.8		9.0			

(a) Benjamin, 1978, unless noted

(b) Long and Angino, 1977

(c) Singer, 1974

(d) N/A - not available

TABLE 6.4

Hypothetical Copper Speciation

pH 7 $\theta_c = 5$ days

SS = 0.945 g/l

Species	Fraction (percent)
Cu^{2+}	10.0
$\text{Cu}(\text{OH})^+$	1.0
$\text{Cu}(\text{OH})_2$	20.0
$\text{Cu}(\text{NH}_3)^{2+}$	27.1
$\text{Cu}(\text{NH}_3)_2^{2+}$	9.2
$\text{Cu}(\text{CO}_3)$	2.6
$\text{Cu}(\text{HCO}_3)^+$	2.5
Cu (bacteria)	27.6
	100.0

TABLE 6.5

Hypothetical Copper Speciation

pH = 7, $\theta_c = 5$ days, SS = 0.868 g/l

Species	Fraction (percent)
Cu^{2+}	11.6
$\text{Cu}(\text{OH})^+$	1.2
$\text{Cu}(\text{OH})_2$	23.2
$\text{Cu}(\text{NH}_3)^{2+}$	31.4
$\text{Cu}(\text{NH}_3)_2^{2+}$	10.6
CuCO_3	3.1
CuHCO_3^+	2.9
Cu (bacteria)	15.8
	99.8

TABLE 6.6

Hypothetical Cadmium Speciation

pH 7, $\theta_c = 5$ days, SS = 0.945 g/l

Species	Fraction (percent)
Cd^{2+}	15.1
CdCl^+	1.3
CdHCO_3^+	1.0
Cd (bacteria)	82.6
	<u>100.0</u>

TABLE 6.7

Hypothetical Cadmium Speciation

pH 7, $\theta_c = 5$ days SS = .868 g/l

Species	Fraction (percent)
Cd^{2+}	40.7
CdNH_3^{2+}	0.1
CdCl^+	3.5
CdHCO_3^+	2.6
Cd (bacteria)	53.3
	<u>100.2</u>

TABLE 6.8

Hypothetical Zinc Speciation
 pH 7, $\theta_c = 5$ days, SS = 0.945 g/l

Species	Fraction (percent)
Zn ²⁺	9.2
Zn(OH) ⁺	0.1
Zn(HCO ₃) ⁺	0.6
Zn (bacteria)	90.1
	<u>100.0</u>

TABLE 6.9

Hypothetical Zinc Speciation
 pH 7, $\theta_c = 5$ days, SS = 0.868 g/l

Species	Fraction (percent)
Zn ²⁺	13.7
ZnOH ⁺	0.1
ZnHCO ₃ ⁺	0.9
Zn (bacteria)	85.2
	<u>99.9</u>

Cu^{2+} , $\text{Cu}(\text{OH})_2$, CuNH_3^{2+} , $\text{Cu}(\text{NH}_3)_2^{2+}$, and $\text{Cu}(\text{bacteria})$. The speciation of copper is somewhat sensitive to the bacterial solids concentration; the $\text{Cu}(\text{bacteria})$ fraction for solids concentrations of 868 mg/l* is just half the $\text{Cu}(\text{bacteria})$ fraction for solids concentration of 945 mg/l*. The distribution for the other species was only minimally altered by solids variation.

Copper-ammonia species (CuNH_3^{2+} and $\text{Cu}(\text{NH}_3)_2^{2+}$) are the most predominant: 36.3 and 42.0 percent combined for solids of 945 mg/l and 868 mg/l, respectively. The fractions of $\text{Cu}(\text{bacteria})$ for the same conditions are 27.6 and 15.8 percent, respectively. These hypothetical fractions associated with the solid phase are much lower than experimentally observed (85 percent; Figure 5.2).

Because nitrification (the biological process of converting NH_4^+ to NO_3^- can occur at pH 7, 20°C , and $\theta_c = 5$ days; the laboratory conditions) and because the primary effluent was thought to be much weaker (60 percent weaker) than listed in Table 6.1, the hypothetical speciation of copper was recalculated with no ammonia present. The results showed $\text{Cu}(\text{bacteria})$ to be 43.2 percent and $\text{Cu}(\text{OH})_2$ to be 31.4 percent. This still does not account for the deviance between experimental and hypothetical removals by the solids and is offered only in partial explanation.

Expansion of speciation determinations showed that the predominant

*These concentrations (868 mg/l and 945 mg/l) correspond to two experimental solids concentrations (tests 5 and 8) involved in determining the K'_s value.

species was $\text{Cu}(\text{OH})_2$ (aq) (98 percent at pH 10) even when the calculations included some ammonia. The solubility product of $\text{Cu}(\text{OH})_2$ (S) was never exceeded (hypothetically) and this would account for the resolubilization of copper with increasing pH.

6.3.2 Hypothetical Cadmium Distribution

The hypothetical distribution of cadmium shown in Tables 6.6 and 6.7 (for solids of 945 mg/l and 868 mg/l, respectively) indicate that the major cadmium species are Cd (bacteria) and Cd^{2+} . For solids concentrations of 945 mg/l, the calculated Cd (bacteria) fraction (82.6 percent) corresponds well with experimentally determined solid-phase metal fraction (90 percent from Figure 5.1). However the sensitivity of this distribution model is obvious in the greatly lowered Cd (bacteria) fraction for solids of 868 mg/l. The difference between the solids concentrations is less than 10 percent, and is manifested in a 35 percent decrease in the Cd (bacteria) species for the lower solids concentration.

Expansion of the speciation calculations to include pH 8.5 and 10 show little change in the overall fractionation. The solubility product is never exceeded and therefore (hypothetical) precipitation does not occur.

The experimental removal of cadmium by solids is highest at pH 10 (Figure 5.4), whereas the hypothetical removal of metals by solids is lowest (70.2 percent) at pH 10. It may be that soluble cadmium

hydroxide species are adsorbed by the solids and thus removed, but the actual explanation is unknown.

6.3.3 Hypothetical Zinc Distribution

Tables 6.8 and 6.9 list the distribution of the calculated zinc species. Like cadmium, the major species are Zn^{2+} , and Zn (bacteria). Unlike cadmium, the effects of solids variation are less noticeable: only a 5 percent decrease in the Zn (bacteria) species for the lower solids concentrations.

Calculated fractions of Zn (bacteria) at pH 7 correspond very well with experimentally observed solids adsorption (85.2 to 90.1 percent calculated) compared to 95 percent observed (Figure 5.3)

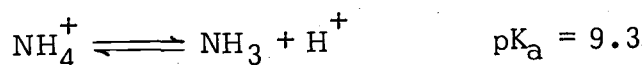
For increasing pH gross estimates indicate that zinc removal by bacteria is much less than experimentally observed removals: 90 to 95 percent experimentally compared to 70.0 and 0.5 percent hypothetical removal at pH 8.5 and pH 10, respectively.

Subsequent calculations showed that the hypothetical solubility products of $Zn(OH)_2(s)$ indicate that precipitation is occurring or is about to occur at pH 8.5 and 10. If precipitation does occur, then the fraction of solid phase metal would increase and hypothetical fractionation would be similar to that experimentally observed.

6.4 Effects of Ammonia and Amino Compounds upon Metal Adsorption

6.4.1 Effects of Ammonia

Ammonia is known to have high affinity for complexation with metals, especially copper. The concentration of ammonia-N in solution for weak domestic sewage is about 12 mg/l (8.57×10^{-4} M) (Metcalf and Eddy, 1972). Ammonia equilibrium in water is expressed by the following reaction:



Thus, NH_3 concentration is pH dependent. The concentrations of NH_3 with respect to pH at the given concentration are given in Table 6.10. Even though only 0.5 percent of the total ammonia-N was NH_3 , at pH 7, the Cu- NH_3 ligands comprised over 50 percent of the total speciation of copper hypothetically determined (Tables 6.4 and 6.5). As the NH_3 concentration increases with increasing pH, however no additional copper will be associated with it, in fact it decreases at pH 10 due to high $\text{Cu}(\text{OH})_2$ complexation.

Ammonia is thought to nitrify at laboratory conditions (pH 7, $\theta_c = 5$ days, and 20°C). This process converts NH_4^+ to NO_3^- and renders $\text{Cu}(\text{NH}_3)_x$ complexation negligible. Nitrification is facilitated at θ_c values greater than 5 days and could account for the increasing concentration factor of copper on bacteria with increasing θ_c observed in Figure 5.11.

6.4.2 Effects of Common Amino-Organic Compounds

Soluble organic nitrogen typically consists of amino acids, proteins, and nucleic acids. These compounds have amino ($-\text{NH}_2$) and carboxyl ($-\text{COOH}$) functional groups which differ in their acidity constant (pK) values. For example carboxyl and amino groups have pK values of approximately 2.0 and 9.0, respectively (Lehninger, 1970). The pH at which molecules containing amino, carboxyl, and other functional ionogenic groups have zero net charge is the isoelectric point. The isoelectric points of proteins, peptides, and amino acids are typically in the pH range of 4.5 to 6.5 (Lehninger, 1970), and above this pH value, the molecules have a negative net charge corresponding to protonation changes from $\text{R}-\text{NH}_3^+$ to $\text{R}-\text{NH}_2$ and $\text{R}-\text{COOH}$ to $\text{R}-\text{COO}^-$. Both of these transformations should increase the adsorption of metal ions.

In relation to the variable pH-adsorption phenomenon, organic nitrogen may affect the adsorption of metals depending upon the phase in which these organics exist. If there are larger quantities of functional groups upon the bacterial walls or on bacterially adsorbed organics than are in solution, then adsorption is likely to be enhanced with increasing pH. Since the polysaccharide structure of cell walls has largely carboxyl groups, the amino functional groups would effect the metals solubility according to the amino distribution in solid or soluble phase.

In addition to pH effects, the metal-organic stability constant for complexation with the functional groups must be favorable. Zinc, cadmium, and copper all have favorable complexation with the carboxyl groups, while copper has a higher affinity for amino groups than either cadmium or zinc and will be the metal most highly affected by them.

TABLE 6.10

Concentration of NH_3 For 8.57×10^{-4} M
Ammonia-Nitrogen
as a Function of pH

pH	NH_3 (M)	Percent of Total
7	4.273×10^{-6}	0.5
8.5	1.172×10^{-4}	13.7
10.0	7.144×10^{-4}	83.4

7. CONCLUSIONS

7.1 Management of Metals Partitioning in Sewage

The management of metals in domestic sewage may be enhanced by controlling pH and θ_C according to the following criteria:

1. To achieve minimum metal concentration in the effluent, metal concentrations must be maximized onto the solid phase:
 - a. Copper removal by bacterial solids is maximized at pH 7, while zinc and cadmium removal by bacterial solids is enhanced at the maximum possible operating pH.
 - b. Maximum zinc and cadmium concentrations on the solid phase occurs at θ_C values less than 5 days, while copper, only minimally affected by θ_C , increases in solids association slightly with increasing θ_C values.
2. For minimum metal concentration in the sludge, the metals are maximized in the soluble phase.
 - a. Maximum soluble copper concentrations occur at pH values greater and lower than pH 7. Soluble cadmium and zinc species are maximized by operating at the lowest possible pH values.
 - b. Cadmium and zinc metal-sludge association is minimized at high θ_C values (greater than 5 days) while copper-sludge association is the least at low θ_C values.

7.2 Prediction of Metal Speciation

The utilization of the experimentally determined K'_s values to predict the speciation of metals in sewage is highly dependent upon the following items:

1. Conditions for K'_s determinations (θ_c , pH, sewage composition and strength), and
2. solids concentration of the system.

Cadmium and copper speciation were highly affected by the solids variation. Further research to confirm the empirically determined K'_s values is needed.

7.3 Effects of Ammonia

Ammonia concentrations considered in the hypothetical speciation determinations showed high affinity for copper complexation at pH 7, but at increasing pH, the NH_3 complexation is negligible compared to the $\text{Cu}(\text{OH})_2$ aq. species which is thought to cause resolubilization of copper at high pH values.

High θ_c values were thought to achieve nitrification (NH_4^+ conversion to NO_3^-) and thus further reduce the effects of ammonia complexation.

Cadmium and zinc speciations were unaffected by ammonia complexation.

8. RECOMMENDATIONS FOR FURTHER STUDY

In order to utilize and expand the information obtained in this research, further study is required.

1. The values of Q^0 , b , and K'_s determined in this study are tentative being procured from a small number of data points. In order to provide useful values, confirmation of these constants is needed. In addition, K'_s should be investigated to determine if the stability constant can be applied more widely than it is now. (K'_s values vary for different pH values, θ_c values, solids concentration, waste composition and waste strength and may only be applied under these conditions.)
2. Detailed equilibrium calculations and experimental verification of metals distribution and speciation in the activated sludge system is needed in order to investigate the deviation of calculated from experimental metals distribution, and to further investigate speciation prediction capabilities.
3. Environmental management of metals could be furthered by extrapolation of the metal-bacterial solids adsorption phenomenon from this study into other environmental systems in which bacterial surfaces are important, e.g. organically polluted surface waters (sewage outfalls, etc.) and soil adsorption systems for wastewaters, etc.
4. Research to determine feasibility and methodology for metals recovery from sludge is needed to permit industrial reuse of metals and lessen their impact upon the environment if effective removal is possible.

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APPENDICES

APPENDIX I

List of Abbreviations and Symbols

- B constant expressive of energy interaction with the surface
- BET Brunauer, Emmett, Teller (adsorption isotherm)
- b constant, related to the free energy of adsorption
- C concentration of the solute
- CF concentration factor
- C_s saturation concentration of adsorbate in solution
- COD chemical oxygen demand
- Dosages
- A Cu = 1 ppm, Zn = 1 ppm, Cd = 0.5 ppm
- B Cu = 10 ppm, Zn = 10 ppm, Cd = 5 ppm
- C Cu = 100 ppm, Zn = 100 ppm, Cd = 50 ppm
- D Cu = 0 ppm, Zn = 100 ppm, Cd = 0 ppm
- E Cu = 0 ppm, Zn = 0 ppm, Cd = 50 ppm
- g gram
- k maximum rate of substrate utilization, mass substrate/
mass solids-time⁻¹
- k_d microorganism decay rate, time⁻¹
- K_f constant, (Freundlich equation 3.16)
- K_s half velocity coefficient, mass/vol
- K'_s apparent stability constant for metal-sludge complexation
- l liter
- M free metal ion

mg	milligram
MLSS	Mixed Liquor Suspended Solids
ML _x	complexed metal in solution
M _s	apparent equilibrium soluble concentration
MS _y	total metal bound to sludge, moles/l
n	constant (Freundlich equation 3.16)
Q	forward flow rate, vol/time
Q ^o	number of moles of solute adsorbed per unit weight of adsorbent in forming a complete monolayer
Q _w	Flow rate of the fraction of solids wasted vol/time
q _e	amount of solute adsorbed per unit of solid adsorbent
S	concentration of MLSS, g/l
S ₁	concentration of substrate surrounding the organisms = the effluent conc of complete-mix system, mass/vol
S _o	influent substrate concentration, mass/vol
SRT	solids retention time equivalent to SRT
SS	Suspended solids
t	time
TCFU	total colony forming units
u	substrate consumption rate, days ⁻¹
V	volume
X	microbial mass concentration, mass/vol
X _e	concentration of biological solids lost in the effluent, mass/vol
X	concentration of solids wasted, mass/vol
Y	growth yield coefficient, mass solids/mass substrate

y	equivalent binding sites on sludge per mole metal
θ_c	solids retention time
U	specific growth rate of bacteria
μg	microgram

APPENDIX II

TABLE 1Langmuir Straight Line
Calculations

Metal	θ_c	$\frac{1}{q_e}$ $\frac{\text{mg}}{\text{g}}$	$\frac{1}{C}$ $(\text{mg/l})^{-1}$
Copper	1	0.69	2.86
	5	1.03	5.0
	5	0.14	0.5
	5	0.02	0.03
	15	1.30	10
Zinc	1	0.59	10
	5	2.27	20
	5	0.22	1.15
	5	1.22	0.03
	5	0.03	0.05
	15	2.47	20
Zinc	1	0.24	5.5
	5	0.77	12.8
	5	0.11	0.91
	5	0.01	0.02
	5	0.01	0.03
	15	0.75	8.3

TABLE 2

Straight Line Freundlich Equation Data
(Equation 3.16)

Metal	θ_c	$\log q_e$ $q_e \left(\frac{\text{mM}}{\text{mgMLSS}} \right)$	$\log C$ mM
Copper	1	-4.64	-2.27
	5	-4.8	-2.51
	5	-3.96	-1.51
	5	-3.09	-0.304
	15	-4.92	-2.82
Cadmium	1	-4.82	-3.05
	5	-5.41	-3.36
	5	-4.39	-2.11
	5	-5.14	-0.573
	5	-3.56	-0.718
	15	-5.44	-3.36
Zinc	1	-4.20	-2.55
	5	-4.70	-2.9
	5	-3.85	-1.77
	5	-2.95	-0.118
	5	-2.84	-0.297
	15	-4.69	-2.74

TABLE 3

Empirical Determination of K_f and n , from Figures
1, 2, and 3 (Appendix)

Metal	K_f	n
Copper	1.66×10^{-3}	1.225
Cadmium	1.20×10^{-3}	1.25
Zinc	2.34×10^{-3}	1.25

TABLE 4

Metal-Sludge: Calculated Values

Metal	θ_c	MS_y mM/l	M_s mM	$\log \frac{MS_y}{M_s}$	S g/l	log S
Copper	1	0.357	0.535	-0.176	0.236	0.63
	5	.9677	0.306	0.500	0.945	-0.02
	5	6.25	3.06	0.310	0.868	-0.06
	5	3.31	49.7	-1.18	0.608	-0.22
	15	0.90	0.153	0.770	1.112	-0.046
Cadmium	1	0.236	0.089	0.42	0.236	-0.63
	5	0.246	0.044	10.75	0.945	-0.02
	5	2.37	0.774	0.49	0.868	-0.06
	5	0.296	26.7	-1.95	0.608	-0.22
	15	0.267	0.044	0.78	1.112	-0.046
Non-comp.	5	10.53	19.3	-.26	0.579	-0.237
Zinc	1	0.993	0.2	0.70	0.236	-0.63
	5	1.25	0.12	1.02	0.945	-0.02
	5	8.13	1.68	0.68	0.868	-0.06
	5	45.79	76.18	-0.22	0.608	-0.22
	15	1.46	0.18	0.91	1.112	-0.046
Non-comp.	5	58.0	50.48	0.06	0.608	-0.216

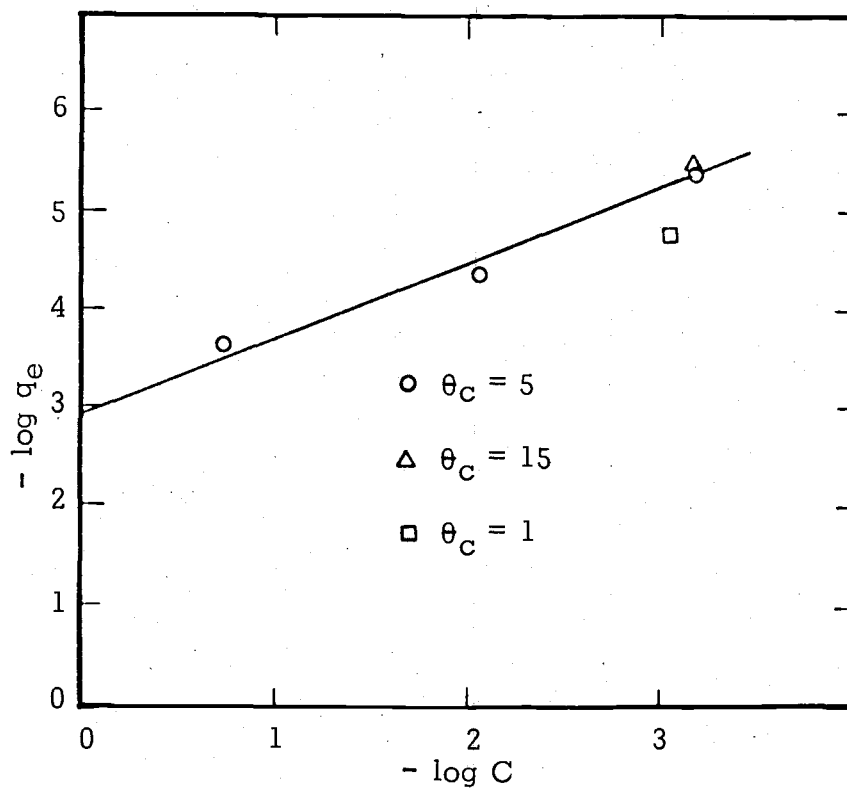


Figure 1 Linearization of Freundlich Adsorption For Cadmium

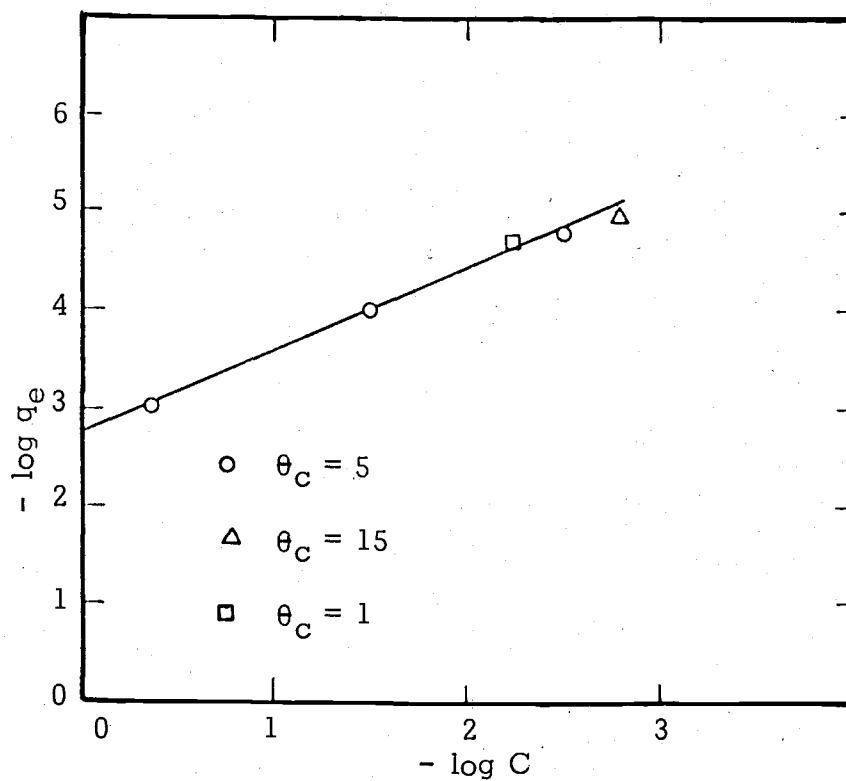


Figure 2 Linearization of Freundlich Adsorption for Copper

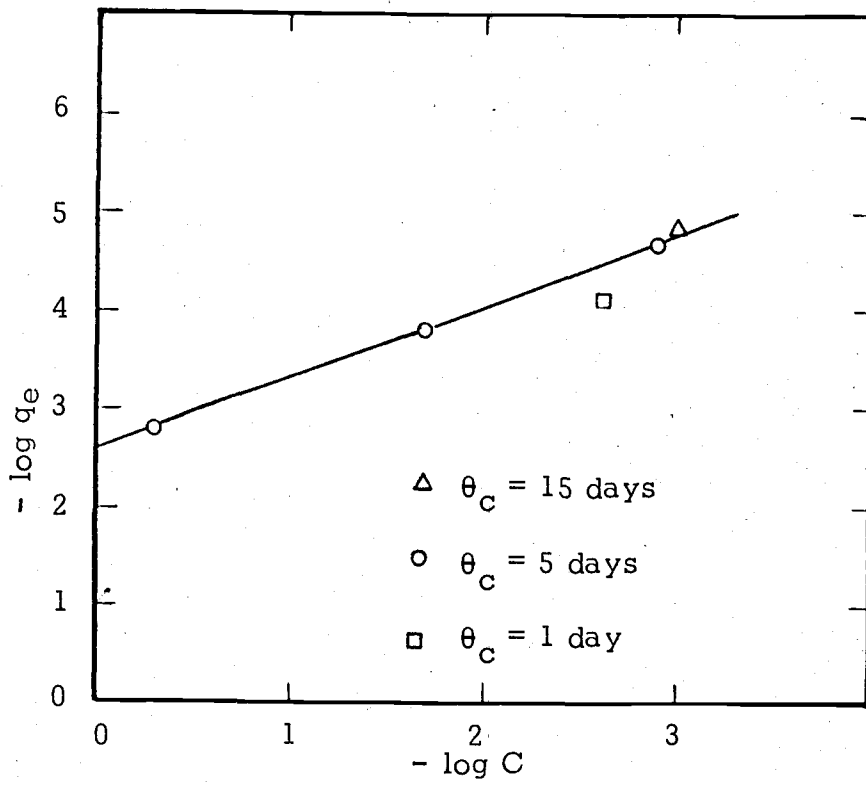


Figure 3 Linearization of Freundlich Adsorption for Zinc

TEST NO. 1 DATE May 13, 1979 DOSAGE A pH 7 SOLIDS 236 θ_c 1

SAMPLE DESCRIPTION	CADMIUM mg/l			COPPER mg/l			ZINC mg/l			IRON mg/l		
	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT
Background	0	0	0	0	0	0	0.2	0.15	0.26	0	1.5	1.5
T = 0 hrs.	0.05	0.42	0.47	0.3	0.3	0.675	0.2	1.0	1.1	0	1.5	1.5
T = 1 hr.	0.14	0.36	0.495	0.35	0.21	0.73	0.2	0.9	1.07	0	1.5	1.5
T = 2 hrs.	0.1	0.32	0.475	0.37	0.43	0.635	0.2	1.0	1.25	0	1.5	1.5
T = 4 hrs.	0.05	0.45	0.47	0.32	0.32	0.65	0.12	1.0	1.07	0	1.5	1.5

APPENDIX III

TEST NO. 2 DATE 4/29/78 DOSAGE A pH 4 SOLIDS 608 θ_c 5

SAMPLE DESCRIPTION	CADMIUM mg/l			COPPER mg/l			ZINC mg/l			IRON mg/l		
	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT
Background	0	0	0	0.05	0.07	0.035	0	0.3	0.35	0.1	2.35	2.5
T = 0 hrs.	0.31	0.15	0.455	0.35	0.42	0.95	0.69	0.27	1.03	0	2.55	2.6
T = 1 hr.	0.32	0.11	0.4	0.37	0.41	0.95	0.8	0.22	1.12	0	2.4	2.4
T = 2 hrs.	0.35	0.07	0.43	0.38	0.41	0.92	0.79	0.3	1.07	0	2.5	2.6
T = 4 hrs.	0.32	0.05	0.43	0.45	0.4	0.85	0.82	0.2	1.07	0	2.4	2.6

TEST NO. 3DATE 4/12/79DOSAGE ApH 5SOLIDS 778 θ_c 5

SAMPLE DESCRIPTION	CADMIUM mg/l			COPPER mg/l			ZINC mg/l			IRON mg/l		
	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT
Background	0.015	0.005	0.01	0.01	0.09	0.02	0.05	0.17	0.22	0.1	2.3	2.2
T = 0 hrs.	0.225	0.225	0.51	0.25	0.82	0.93	0.67	0.4	1.07			
T = 1 hr.	0.24	0.25	0.5	0.22	0.8	1.09	0.38	0.6	1.11			
T = 2 hrs.	0.25	0.23	0.5	0.30	0.79	1.13	0.47	0.4	1.19			
T = 4 hrs.	0.35	0.138	0.5	0.5	0.55	1.01	0.8	0.25	1.10			

TEST NO. 4 DATE 3-4-78 DOSAGE A pH 5.6 SOLIDS 934 θ_c 5

SAMPLE DESCRIPTION	CADMIUM mg/l			COPPER mg/l			ZINC mg/l			IRON mg/l		
	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT
Background	0.018	0.01	0	0.035	0.12	0.05	0.31	0.47	0.85	-	-	-
T = 1 hr.	0.08	0.225	-	0.16	0.67	-	0.8	0.77	-	-	-	-
T = 2 hrs.	0.075	0.268	-	0.12	1.16	-	1.16	0.74	-	-	-	-
T = 5 hrs.	0.207	0.19	-	0.26	0.75	-	1.24	1.02	-	-	-	-
T = 10 hrs.	0.14	0.24	-	0.16	0.84	-	0.72	0.80	-	-	-	-
T = 24 hrs.	0.13	0.25	0.43	0.20	0.85	0.4	0.75	0.75	2.375	-	-	-

TEST NO. 5 DATE 3-17-78 DOSAGE A pH 7.4 SOLIDS 945 θ_c 5

SAMPLE DESCRIPTION	CADMIUM mg/l			COPPER mg/l			ZINC mg/l			IRON mg/l		
	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT
Background	0.012	0.012	0.025	0.06	0.02	0.15	0.4	0.26	0.37	0.2	2.0	3.5
T = 0	0.14	0.375	0.51	0.28	0.89	1.0	0.24	0.93	1.41	0.26	5.2	4.5
T = 30 min.	0.07	0.40		0.25	0.92		0.165	1.16		0.19		
T = 1 hr.	0.052	0.41		0.24	0.915		0.092	1.28		0.17	4.95	
T = 5 hrs.	0.052	0.425	0.50	0.206	0.93	0.97	0.08	1.2	1.29	0.155	4.7	4.48
T = 10 hrs.	0.05	0.42	0.52	0.22	0.78	0.90	0.06	1.22	1.4	0.155	4.51	4.46
T = 20 hrs.	0.052	0.44	0.53	0.201	0.98	0.95	0.08	1.29	1.48	0.1	4.6	4.52

TEST NO. 6DATE 4-12-79DOSAGE ApH 9SOLIDS 778 θ_c 5

SAMPLE DESCRIPTION	CADMIUM mg/l			COPPER mg/l			ZINC mg/l			IRON mg/l		
	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT
Background	0.015		0.01	0.01	0.9	0.92	0.05	0.17	0.22	0.1	2.3	2.2
T = 0 hrs.	0.02	0.47	0.51	0.47	0.7	1.18	0.07	0.92	1.09	0.1	2.5	2.58
T = 1 hr.	0.025	0.5	0.515	0.54	0.63	1.01	0.07	0.98	1.08	0.05	2.35	2.6
T = 2 hrs.	0.03	0.43	0.5	0.65	0.42	0.97	0.08	0.90	1.08	0.1	2.2	2.55
T = 4 hrs.	0.02	0.49	0.52	0.67	0.3	0.92	0.05	0.98	1.08	0.05	2.3	2.68

TEST NO. 7DATE 4-15-78DOSAGE ApH 10SOLIDS 890 θ_c 5

SAMPLE DESCRIPTION	CADMIUM mg/l			COPPER mg/l			ZINC mg/l			IRON mg/l		
	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT
Background	0	0	0	0.07	0.07	0.08	0.08	0.13	0.22	0.15	1.5	2.5
T = 0 hrs.	0.01	0.48	0.49	0.62	0.4	1.06	0.02	0.97	1.03	0.35	2.35	2.7
T = 1 hr.	0.05	0.42	0.46	0.6	0.2	0.96	0.05	0.6	1.05	0.15	1.89	2.6
T = 2 hrs.	0.04	0.44	0.47	0.63	0.4	1.05	0.03	0.97	1.05	0.1	2.46	2.7
T = 4 hrs.	0.03	0.27	0.4	0.75	0.23	0.94	0.15	0.65	0.98	0.1	1.8	2.7

TEST NO. 8 DATE 4-18-79 DOSAGE B pH 7 SOLIDS 868 θ_c 5

SAMPLE DESCRIPTION	CADMIUM mg/l			COPPER mg/l			ZINC mg/l			IRON mg/l		
	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT
Background	0	0	0	0.01	0.07	0.07	0	2.15	2.25	0.15	2.5	2.65
T = 0 hrs.	0.65	3.2	5	1.1	5.95	9.1	1.12	8	9.75	0.2	2.75	2.8
T = 1 hr.	1.0	3.05	5	1.75	5.15	7.7	1.4	7.4	10	0.15	2.5	2.6
T = 2 hrs.	0.75	0.95	5	1.8	1.2	7.1	0.8	2.0	10	0.1	0.8	2.6
T = 4 hrs.	0.85	1.8	5	2.25	2.65	8.9	1.1	4.3	10	0.15	1.35	2.87

TEST NO. 9DATE 4-29-78DOSAGE CpH 7SOLIDS 608 θ_c 5

SAMPLE DESCRIPTION	CADMIUM mg/l			COPPER mg/l			ZINC mg/l			IRON mg/l		
	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT
Background	0	0	0	0.05	0.07	0.035	0	0.3	0.35	0.1	2.35	2.5
T = 0 hrs.	32	0.6	40	42.5	41	97.5	48	48	97	0	3.55	3.55
T = 1 hr.	32	0.55	33.5	32.5	38.5	85	48	48	100	0	3.5	3.5
T = 2 hrs.	29.5	0.5	30.5	34.5	26	102	52	44	100	0	3.45	3.5
T = 4 hrs.	29	0.5	32.5	30.5	32	92.5	51	41	100	0	3.4	3.5

TEST NO. 10 DATE 4-29-78 DOSAGE D pH 7 SOLIDS 608 θ_c 5

SAMPLE DESCRIPTION	CADMIUM mg/l			COPPER mg/l			ZINC mg/l			IRON mg/l		
	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT
Background	(See Test 9)											
T = 0	0.4	0.17	0.72	0.15	0.2	0.7	31.5	55	95	0	2.5	2.5
T = 1 hr.	0.35	0.17	0.5	0.15	0.2	0.7	32	60	100	0	2.5	2.5
T = 2 hrs.	0.45	0.21	0.65	0.15	0.2	0.7	35	57	98	0	2.5	2.4

TEST NO. 11DATE 5-2-78DOSAGE EpH 7SOLIDS 579 θ_c 5

SAMPLE DESCRIPTION	CADMIUM mg/l			COPPER mg/l			ZINC mg/l			IRON mg/l		
	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT
Background	0	0.02	0.02	0	0	0	0	0.5	0.5	0	1.0	1.0
T = 0 hrs.	26.5	16.2	40									
T = 2 hrs.	21.5	14.5	36.5									
T = 4 hrs.	21.5	21	41									

TEST NO. 12DATE 5-2-79DOSAGE ApH 7SOLIDS 1112 θ_c 15

SAMPLE DESCRIPTION	CADMIUM mg/l			COPPER mg/l			ZINC mg/l			IRON mg/l		
	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT
Background	0.04	0.05	0.05	0	0.09	0.08	0.15	0.47	0.47	0.1	1.3	1.5
T = 0 hrs.	0.25	0.3	0.5	0.25	0.77	1.04	0.62	0.87	1.47	0	1.35	1.4
T = 2 hrs.	0.05	0.45	0.5	0.1	0.92	1.0	0.15	1.47	1.6	0	1.35	1.47
T = 4 hrs.	0.05	0.45	0.5	0.1	0.8	1.0	0.1	1.48	1.57	0	1.6	1.45

TEST NO. 13

DATE 4-29-78

DOSAGE A

pH 7

SOLIDS 608 θ_c 5

SAMPLE DESCRIPTION	CADMIUM mg/l			COPPER mg/l			ZINC mg/l			IRON mg/l		
	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT	SOL	S.S	TOT
Autoclaved	0.06	0.4	0.5	0.32	0.55	0.96	0.05	1.2	1.32	0.05	2.5	2.45