

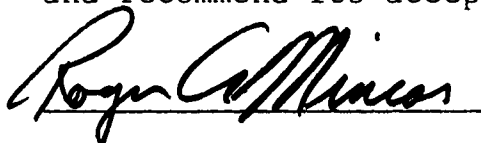
To the Graduate Council:

I am submitting herewith a thesis written by George C. Welborn, Jr., entitled "The Partitioning of Copper in the Activated Sludge Process as a Function of pH." I recommend that it be accepted in partial fulfillment of the requirements for the degree of Master of Science, with a major in Environmental Engineering.



Roy O. Ball, Major Professor

We have read this thesis
and recommend its acceptance:



Accepted for the Council:



Vice Chancellor
Graduate Studies and Research

Thesis
79
W434
cop. 2

THE PARTITIONING OF COPPER IN THE
ACTIVATED SLUDGE PROCESS AS A
FUNCTION OF pH

A Thesis
Presented for the
Master of Science
Degree
The University of Tennessee, Knoxville

George C. Welborn, Jr.

December 1979

1405188

ACKNOWLEDGEMENTS

I would like to thank Dr. Roy O. Ball and Dr. Roger A. Minear for their guidance and encouragement throughout this study and Dr. Richard L. Church for his advice and careful reading of this thesis. A note of thanks is due to many of my fellow students especially Raymond Rose for answering my constant questions, Dwayne Fry for his patience and endurance in assisting in many of the analytical procedures, and Mark Cantrell for his assistance in processing the data produced by this study.

A note of appreciation is due Mr. Bill Long and Mr. Jim Fry of the Civil Engineering staff for constantly repairing key equipment at a moment's notice. Without their assistance, I should still be attempting to finish this study. I would like to thank Diane Dombroski for her help in typing the early drafts of this thesis.

I should like to thank the University of Tennessee Civil Engineering Department and the Church and Dwight Company, Inc. for sponsoring this research under contract number R01-1331-19.

ABSTRACT

The uptake of metal onto the biomass in the activated sludge system has been reported to be a function of pH, metal concentration, organic ligand concentration and time of contact. All of the previous studies were carried out under batch conditions by adjusting a sample biomass to the desired pH and adding the copper. The test results indicated that as the pH increased more copper was adsorbed per unit of biomass.

In this study, complete mix continuous flow biological reactors, under steady state conditions, were used to observe the effect of pH on the partitioning of copper in the activated sludge system. The pH values studied, 6.3, 7.4, 8.4, and 8.9, were obtained using phosphate buffers. The copper concentrations were used 176 ppb and 17.5 ppb.

At the influent copper concentration of 176 ppb, the amount of copper adsorbed per unit of biomass increased from .33 $\mu\text{g Cu/mg MLVSS}$ at pH 6.4 to .42 $\mu\text{g Cu/mg MLVSS}$ at a pH of 8.9. At 17.5 ppb influent copper, the copper adsorbed increased from .08 $\mu\text{g Cu/mg MLVSS}$ at pH 6.4 to .11 $\mu\text{g Cu/mg MLVSS}$ at pH 8.9. The free ionic form remained less than 10^{-7} moles per liter at all conditions. The concentration of soluble copper did not vary appreciably as a function of pH but was controlled by the concentration of solids in the mixed liquor.

TABLE OF CONTENTS

SECTION	PAGE
I. INTRODUCTION	1
II. LITERATURE REVIEW	3
III. EXPERIMENTAL EQUIPMENT AND PROCEDURE	15
IV. EXPERIMENTAL RESULTS	26
V. CONCLUSIONS AND RECOMMENDATIONS	61
BIBLIOGRAPHY	63
APPENDIX	68
VITA	79

LIST OF TABLES

TABLE	PAGE
1. Experimental Conditions	17
2. Buffer Solutions (Mixed In Tap Water)	17
3. Sampling Procedure	21
4. Inorganic Carbon Concentration	27
5. Comparative Concentrations of Ca and Mg . . .	30
6. Summary of Kinetic Data	32
7. Kinetic Coefficients	37
8. Summary of Copper Data, Reactors 1, 3, 5, 9, and 10	40
9. Summary of Copper Data Reactors 2, 4, 6, 7, and 8	41
A-1. Reactor #1	69
A-2. Reactor #2	70
A-3. Reactor #3	71
A-4. Reactor #4	72
A-5. Reactor #5	73
A-6. Reactor #6	74
A-7. Reactor #7	75
A-8. Reactor #8	76
A-9. Reactor #9	77
A-10. Reactor #10	78

LIST OF FIGURES

FIGURE	PAGE
1. Typical Reactor and Sampling Points	16
2. Schematic of the Equipment Layout	18
3. Copper Analyses Procedure	24
4. Q Versus S Substrate Removal Kinetics	34
5. 1/Q Versus 1/S Substrate Removal Kinetics	35
6. S/Q Versus S Substrate Removal Kinetics	36
7. OBS. Yield Versus pH Substrate Removal Kinetics	38
8. Influent Copper Versus pH	42
9. Mixed Liquor Suspended Solids Vs. pH	43
10. Mixed Liquor Volatile Suspended Solids Versus pH	44
11. Soluble Copper Versus pH	46
12. Solid Complexed Copper Versus pH	48
13. Soluble/Influent Copper Versus pH	50
14. Soluble/Total Copper Versus pH	51
15. Soluble Copper/Soluble + Solid Complexed Copper Versus pH	52
16. Total Copper Versus pH	53
17. Solid Complexed Copper/MLSS Versus pH	54
18. Solid Complexed Copper/MLVSS Versus pH	55
19. Mixed Liquor Suspended Solids Versus Soluble Copper	57
20. Mixed Liquor Volatile Suspended Solids Versus Soluble Copper	58

I. INTRODUCTION

Past research on the partitioning of heavy metals in the activated sludge process indicates that the majority of the total metal present is complexed with the biomass and other organic matter present in the system. The amount of metal adsorbed is affected by the pH, the initial concentration of metal ions, the concentration of the organic solids and the contact time.

Cheng (11), using sludge acclimated to a dog food substrate under batch conditions, found that the amount of metal adsorbed per unit mass of organic material increased with increasing pH and with increasing initial metal concentration. This has prompted interest in the use of pH adjustment in altering the partitioning of metals in the activated sludge process by the addition of sodium biocarbonate.

The objective of this study was to monitor the changes in copper partitioning present in the continuous activated sludge process as a function of pH at constant alkalinity and determine if the results from batch tests also apply to steady state conditions. Ten complete mix continuous flow biological reactors were used to maintain different pH values using phosphate buffer systems.

The partitioning was observed by measuring the free copper, soluble copper, adsorbed copper, and total copper

fractions of the mixed liquor in the reactors at various pH values.

II. LITERATURE REVIEW

Why Copper Is of Concern

Unlike other environmental parameters such as biochemical oxygen demand (BOD) that are reduced through treatment, copper is a conservative pollutant that merely changes in chemical form or species. Standards have been established (Federal, state and local) for maximum concentrations of copper in drinking water, in wastewaters discharged into municipal sewers, and in treated effluents released into the environment. In addition, the Resource Conservation and Recovery Act (PL 94-580) may classify sewage sludges with excess metal content as hazardous wastes. Because of the increased cost of disposing sewage sludges classified as hazardous wastes, it would be informative to develop a predictive model in order to determine what form or species the copper will take during processing and how different parameters will affect the model.

Copper is ubiquitous in nature, but only through man's development has it become an environmental concern. Natural sources of copper include exposed rocks and soils in surface water, decomposition of vegetation and animal matter, and atmospheric fallout (3). During a recent survey of 1,557 surface water samples in the United States, copper analysis showed a mean value of 15 ppb with values ranging from 1 ppb to 280 ppb (1). Generally, a value of

greater than 50 ppb indicates that man's activities are affecting the stream and it is generally considered to be polluted (2). Man's activities are also reflected in the analysis of air particulates and rainfall. The copper measured in air particulates has been found to be on the order of .2 micrograms per cubic meter ($\mu\text{g}/\text{m}^3$) in one urban area, .1 $\mu\text{g}/\text{m}^3$ in rural areas, and .05 $\mu\text{g}/\text{m}^3$ in an oceanic environment (6,7). A similar increase was found in the copper concentration measured in rainwater which was found to vary from 3.1 ppb in rural areas to 21 ppb in urban areas (4,5).

Comparing the copper concentrations found in nature to the copper concentrations in municipal sewerage, again points out the affect that man's activities have on the amount of copper present. Extensive surveys indicate that the mean influent concentration of copper for municipal waste treatment plants is .10 mg/l with values ranging from less than .02 mg/l up to 3.36 mg/l (8). It has generally been accepted that industrial discharges are the major source of heavy metals in waterways and municipal wastewater. However, a recent study in New York City indicated that 47% of the total copper received at the treatment plants could be attributed to residential sources indicating that much of the copper in municipal wastewater may be inherent to our present water distribution system (40).

Not only does copper exist in increased amounts in water and air as a result of man's activities, but the soil

is also affected through using sewage sludge as a soil conditioner. This practice is a topic of growing concern due to findings that indicate in agricultural areas production might be inhibited by the copper added to the soil (10). Not only is the plant growth inhibited by the use of sewage sludge, but the form of copper present in the soil changes to more stable forms. Keeney and Walsh (9) reported that the extractable copper content of sludge-treated soil decreased with time, suggesting a change in copper to less soluble forms.

Biological Effects

Copper is one of several metals that are essential to microorganisms at low levels but inhibitory or toxic at higher levels. Different bacteria have different minimum levels of copper below which a deficiency of copper occurs preventing the attainment of maximum growth. An enzyme cofactor is just one of the necessary roles played by copper in cell metabolism. As an example, copper is an enzyme cofactor of cytochrome oxidase (19). Because copper is necessary to growth, many organisms are able to obtain the necessary amounts of copper even in very dilute solutions by concentrating the copper into their cells. Dugan and Pfister (17) reported that an activated sludge species (Zooglaea ramigera) is capable of concentrating copper from dilute solution to the extent that copper can represent 12.5 to 34% by weight of the organism. This

weight agreed with similar observations reported by Neufeld (18). In another example of the ability of organisms to concentrate copper, it was reported that Phytoplankton are able to achieve a concentration factor of 17,000 for copper (20). One site responsible for the majority of the copper accumulation in bacteria is the cell membrane. However, research has not been done to more definitively establish the exact mechanism of accumulation in the membrane.

In addition to the inhibitory effect of low levels, copper can also be inhibitory or toxic to organisms at high levels. Several pathways have been identified as possible reasons for the toxic influence of high concentration of copper on cell growth. The inhibitory effect of copper can result from denaturing of cellular constituents, the disruption of cell organization, or the inhibition of the formation of mercaptides in essential proteins (19).

Though some research has been conducted on the inhibitory mechanisms, most of the research has been concerned with the percentage removal of metal by the sludge and not the physichemical relationship between the biomass and the metal ions. Stones (21) studied the fate of metals during treatment by several biological processes. The results showed that activated sludge treatment removed around 80% of the copper initially present in the settled sewage. McDermott (22) reported that in the range of 10 to 25 mg/l of copper, 79% was removed through the secondary effluent with only 9-12% being accounted for by the primary.

Upon increasing the organic loading, 61% removal of copper was measured due to the increased sludge production at the higher organic loading. Stones (21) also worked with copper concentrations of less than 1 mg/l and found a removal of 79-85% by the activated sludge. Young (26) studied the form the copper took through the secondary stage of treatment. By filtering the effluent through a 45 μ filter, he determined that over 90% of the copper present was retained by the filter indicating that only 1% of the effluent copper was in the soluble form.

The toxic effects of copper on activated sludge systems has also been studied by authors incorporating various techniques. Sudo and Aiba (24) examined the 50% inhibition level of copper for three common activated sludge organisms and found that it was different for all three. Poon (25) and others studied similar effects in a comparison of the toxic levels for a mixed activated sludge culture with Geotrichum candidum.

Rudolfs (26) reported that at a copper concentration of 1 mg/l the aerobic metabolism of a mixed activated sludge culture was inhibited. It was reported by Barth (27) and others that a continuous copper concentration of 1 mg/l would affect the effluent quality from an activated sludge plant. Lamb and Tollefson (29) found a 90% decrease in substrate utilization by the activated sludge biomass as a result of 5 mg/l concentration of copper. They also showed that the toxicity decreased markedly with the increase of

suspended solids. An 80% decrease in substrate conversion at 210 mg/l total suspended solids was reduced to a 3% decrease by increasing the suspended solids to 4,000 ppm.

Biological Kinetics

General relationships for bacterial mediated processes have been developed by Lawrence and McCarty (15) using the mean cell residence time, θ_c , as the unifying parameter and developing relationships between θ_c , microbial growth and substrate utilization. There are two relations of interest in biological kinetic characterization. The first relates biological growth and endogenous metabolism to substrate removal rate.

$$\frac{dx}{dt} = Y \frac{df}{dt} - K_d X \quad \text{Equation 1.}$$

where

$\frac{dx}{dt}$ = net growth of microorganism, mg-VSS/l-day

Y = growth yield coefficient, mg of microorganism per mg of substrate utilized

$\frac{df}{dt}$ = rate of substrate utilization by microorganisms mg/mg-day

K_d = rate of endogenous respiration, mg/mg-day

X = concentration of microorganism, mg/l

Using VSS as the measure of microorganism concentration is the major shortcoming of Equation 1, because any influent solids or hydrated complexes in the mixed liquor can volatilize when measuring FSS, contributing to the VSS term and resulting in yield values that are higher than the true value. In spite of its

shortcomings, empirically developed Equation 1 has been shown to apply to aerobic and anaerobic systems as well as pure cultures (15).

The second basic equation relates the rate of substrate utilization both to the microbial mass in the reactor and to the substrate concentration surrounding the microorganisms.

$$\frac{df}{dt} = \frac{kSX}{K_s + S} \quad \text{Equation 2.}$$

where k = maximum rate of waste utilization per unit weight of microorganism per day

K_s = the waste concentration at which the rate of substrate utilization per unit weight of microorganism is one half the maximum rate, mg/l

S = concentration of utilized substrate, mg/l

At present, two operational and control parameters have found wide acceptance (15). The first is the substrate removal rate or food to microorganism ratio, q .

$$q = (\Delta f / \Delta t)_t / X_t \quad \text{Equation 3.}$$

where $(\Delta f / \Delta t)_t$ represents the mass of substrate utilized over a finite time period by a mass of microorganism, X_t .

By combining Equations 2 and 3 a useful relationship is obtained relating the food to microorganism ratio, q , to the substrate concentration (5).

$$q = \frac{kS}{K_s + S} \quad \text{Equation 4.}$$

The reciprocal expansion of Equation 4 enables the kinetic constants to be obtained by plotting the values of experimental data for $1/q$ vs. $1/s$.

$$1/q = (K_s/k) (1/s) + (1/k) \quad \text{Equation 5.}$$

The Y intercept of such a plot is equal to $1/k$ and the slope equivalent to K_s/k .

An alternative linear form of Equation 5 is obtained by multiplying through by S . By plotting (S/q) vs. S for experimental data the kinetic constants may be obtained by setting the Y intercept equal to $1/k$ and the slope equal to K_s/k .

The second control parameter, θ_c , has been termed the mean cell residence time or sludge age and is defined as

$$\theta_c = \frac{X_T}{(\Delta X/\Delta t)_T} \quad \text{Equation 6.}$$

where X_T = total active microbial mass in the system, mg
 $(\Delta X/\Delta t)_T$ = total quantity of active microbial mass removed daily including both those lost in the effluent and those wasted purposely.

By substituting Equations 1 and 3 into Equation 6, we obtain the familiar form of

$$\theta_c^{-1} = Y_q - K_d \quad \text{Equation 7.}$$

The constants of this relation can be obtained by plotting experimental data for substrate removal (q) vs. the inverse of the mean cell residence time. The Y axis intercept is

equal to K_d and the slope is equal to Y . With the kinetic coefficients (Y , K_d , K_s , k) determined from the plots of Equations 5 and 7, a rational design of the biological process for that waste can then be made.

Partitioning of Copper in Activated Sludge Systems

Various forms of copper have been studied by different authors in observing the partitioning of copper in the activated sludge process. The four most common forms measured are free copper, soluble copper, adsorbed copper and total copper. The free or ionic form of copper has two ionic states, the cuprous ion (Cu^+), an unstable ion, and the cupric ion (Cu^{+2}). The free form represents very little of the total copper due to the high affinity of copper to combine with organic and inorganic compounds present in the system, the organic complexes being more stable than the inorganic ones. Stiff (31, 32) found that the free copper represented only 3% of the total copper present in a copper-bicarbonate-carbonate system and less than 1% in various sewages, effluents, and water samples.

Soluble complexes are defined as those passing through a $.45 \mu$ filter. The majority of the soluble complexes in sewage treatment plant effluents consists of inorganic ions such as carbonates, chlorides, sulfates, nitrates, phosphates, sodium, potassium, calcium and magnesium. The organic compounds providing adsorption sites consist of proteins, carbohydrates, fats, oils,

surfactants, phenols and others (33). The soluble copper as a percent of total copper present in activated sludge systems has been found to vary from author to author. Stiff (32) reported that only 25% of the total copper was in the soluble form while Hass (13) reported values around 30%. Jenkins and Keight (34) found that the percent soluble described as the initial copper increased.

Several authors have reported that the majority of the complexed copper is adsorbed by the biomass. Barth, et al (24) observed this by studying the difference in the amount of copper received by a digester. More copper was contained in a combined sludge on a percent of solids basis, than by primary sludge alone. They pointed out that the greater amount of copper was due to the higher bacterial concentration of the combined sludge.

The amount of copper adsorbed by the biomass has been found to be affected by pH, copper concentration, and organic matter concentration. Cheng (11) reported that the uptake capacity increases with increasing pH, increasing metal ion concentration and increasing organic matter. This agreed with the work of Jenkins and Keight (34) who observed that the amount adsorbed increased with increasing initial concentration. Rudolfs and Zuber (35) also found the adsorption to be a function of the sludge concentration.

Chau (36) reported that the same complexing capacity of the biomass will be obtained under given conditions. He reported a complexing capacity of 1.8 to 2.5 microequivalents

of copper per liter. Kunkel and Manahan (37) reported that an activated sludge effluent contained a .9 mg/l complexing capacity.

Tenney and Stumm (38) postulated that the increase in metal uptake with increasing pH was due to negative charges on the bacterial surface. They considered the organisms to act as charged hydrophilic colloids. Because the net negative charge is a result of acid-base ionization of functional groups in the cell surface, the reaction between the bacteria and metal ions is a pH dependent stoichiometric relationship between the concentration of the biomass and the metal ions.

Though many authors have observed and reported the partitioning in the activated sludge process, few have attempted to derive a kinetic or equilibria model. Cheng (11) made use of stability constants obtained from batch tests for a rational basis for predicting levels of metal accumulation in sludge, and the effluent metal concentrations. Patterson, Shimada, and Haas (14) also made use of stability constants obtained from batch tests for developing a model predicting the dynamics of metal transport through a municipal treatment plant. Neufeld et al. (39) omitted the effect of pH when kinetic models and equilibrium relationships were developed for the accumulation of several heavy metals other than copper. None of these studies, however, observed or predicted the effect of pH on the partitioning of copper under steady state conditions.

Summary

The conclusions pertinent to this research that were obtained from the literature review are as follows:

1. The removal of copper in the activated sludge process is largely an adsorption phenomena. The majority of the copper present in the MLSS and the effluent SS is complexed with organic material.

2. Copper concentrations in excess of 1 mg/l inhibit the growth rate of bacteria in the activated sludge biomass. The extent to which the growth rate is inhibited increases as the concentration of organic solids is decreased.

3. The kinetic behavior of activated sludge processes may be described by two relations. By plotting experimental data, kinetic coefficients may be obtained and the variations in kinetic behavior under different environmental conditions predicted.

4. Using batch tests, more copper was found to be adsorbed per unit of biomass by increasing the contact time, the copper concentration in the system, pH of the system, or by decreasing the concentration of organic material present in the system.

5. No studies found in the literature attempted to show that the amount of copper adsorbed per unit mass of biomass increases as a function of pH under steady state conditions.

III. EXPERIMENTAL EQUIPMENT AND PROCEDURE

System Description

The activated sludge process was simulated with ten complete mix continuous flow biological reactors. Each reactor had a nine liter volume and was equipped with an adjustable internal clarifier. A schematic of the reactors used is shown in Figure 1.

The substrate for five of the reactors was spiked with Cu using a copper reference solution (Fisher Scientific Company, Cat. No. SO-C-194), consisting of copper oxide and dilute nitric acid. The substrate of the other five reactors was left at the natural copper concentration occurring in the feed solution. The experimental conditions of the reactors are listed in Table 1.

In order to maintain the pH at the desired levels without directly affecting the alkalinity, a phosphate buffer system was used. Equal volumes of substrate and buffer solution were fed to each reactor. The buffer solutions used to maintain each of the pH levels are listed in Table 2.

VOL = 9.0 LITERS

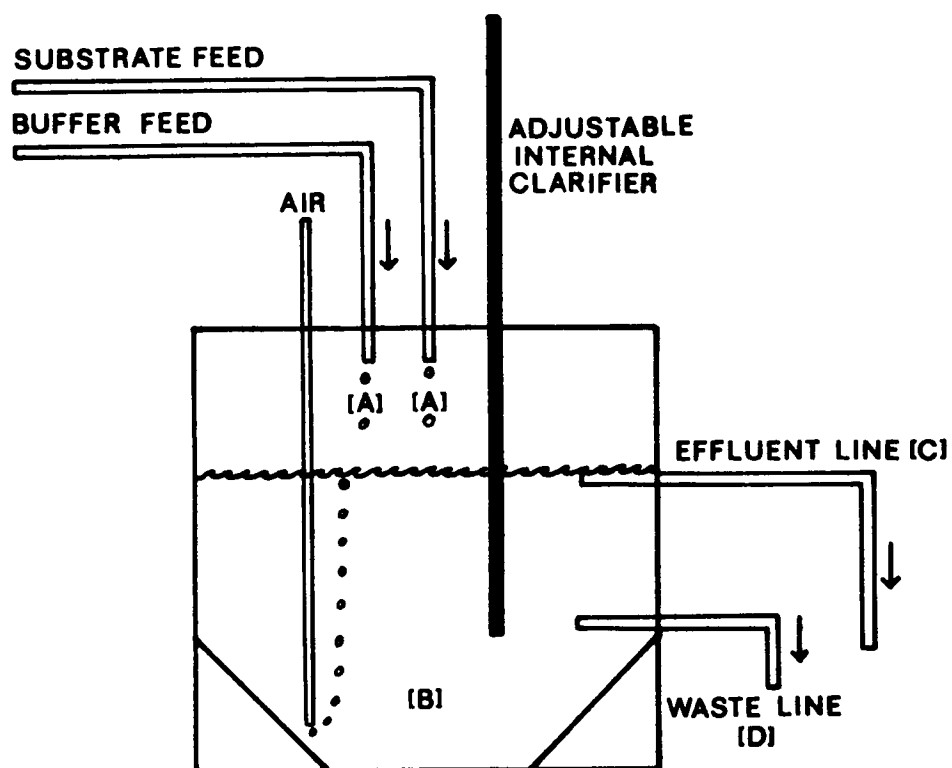


FIGURE 1. TYPICAL REACTOR AND SAMPLING POINTS

TABLE 1
EXPERIMENTAL CONDITIONS

Reactor Number	1	2	3	4	5	6	7	8	9	10
pH	9.0	9.0	8.3	8.3	6.4	6.4	7.3	7.3	7.3	7.3
Natural Cu	x		x		x				x	x
Cu spiked		x		x		x	x	x		

TABLE 2
BUFFER SOLUTIONS (MIXED IN TAP WATER)

pH	6.3		7.3		8.3		9.0	
Reactors	5,6		7,8,9,10		3,4		1,2	
Buffer solution	.01M H_2KPO_4		Tap H_2O		.015M K_3PO_4		.03M K_3PO_4	

As shown by the equipment layout schematic, Figure 2, each reactor had two influent lines. In order to simplify the pumping configuration, one pump was used to feed the reactors receiving added Cu and the other was used to feed the remaining reactors. Reactors 1, 3, 5, 9, and 10 were fed using a Cole-Parmer (Chicago, Illinois) 10 head pump with 7013 heads and reactors 2, 4, 6, 7, and 8 were fed by a Bulcher Dekastaltic Pump (Fort Lee, New Jersey).

The maximum feed rate from the Bulcher Dekastaltic Pump was found to be nominally 3.5 ml/min and the Cole-Parmer pump was adjusted to the same rate. The feed tubing was replaced every three days in order to prevent bacterial

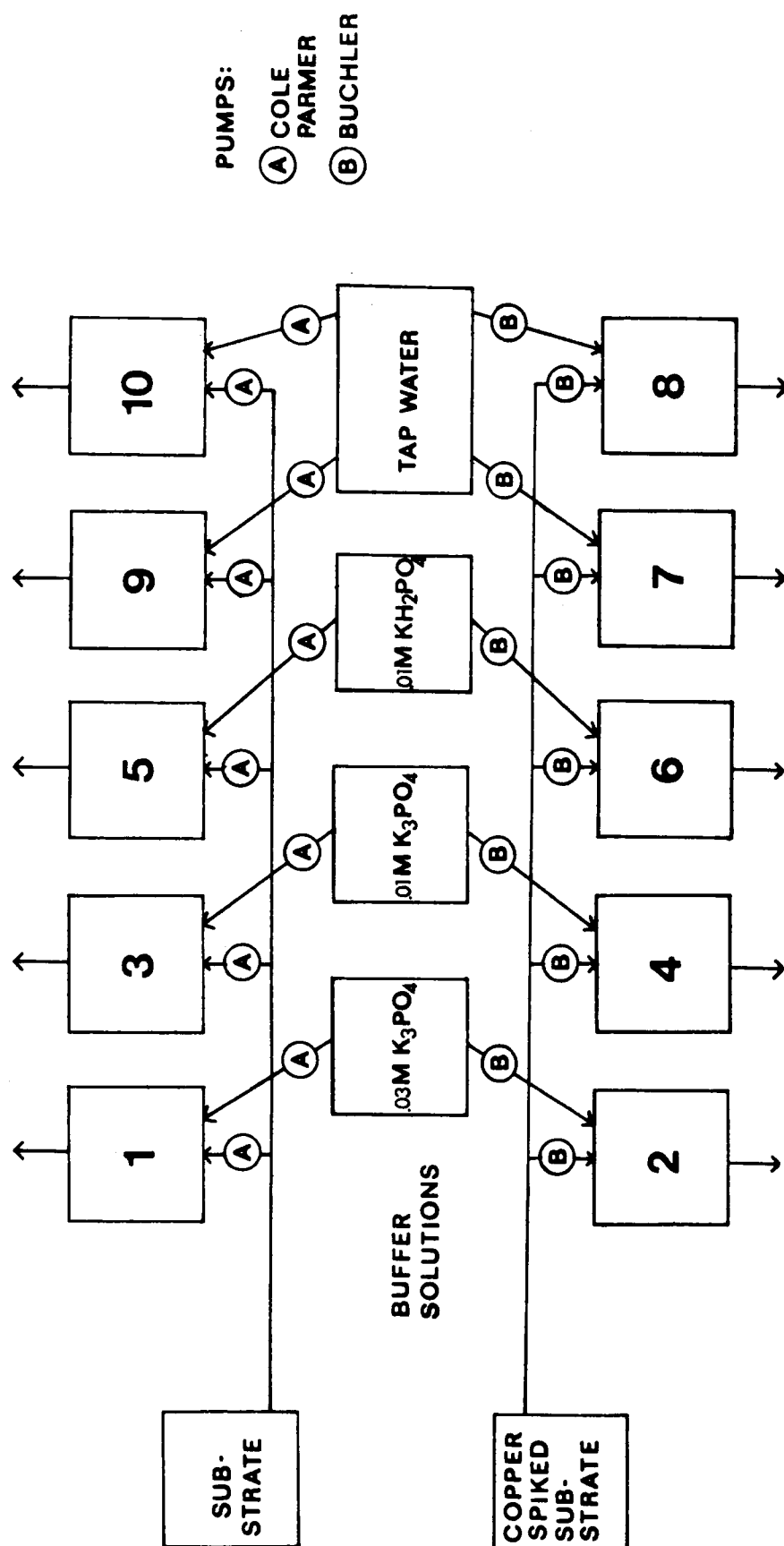


FIGURE 2. SCHEMATIC OF THE EQUIPMENT LAYOUT

growth inside the feed lines and to prevent the tubing from wearing out.

The substrate solutions were prepared and stored in a constant temperature room maintained at 2°C to prevent bacterial growth in the feed. One twelve ounce can of Vanilla Nutriment, a diet supplement drink produced by the Drackett Products Company (Cincinnati, Ohio), was diluted with tap water to a volume of fifteen gallons, resulting in a dilution of 1:160 in the feed barrels. This substrate concentration in conjunction with the equal volume of buffer solution resulted in a Nutriment substrate dilution factor of 1:320, having a nominal COD value of 400 mg/l. The results of the daily analytical data is presented in the Appendix. One of the substrate solutions was spiked with 20 ml of the 1000 ppm copper reference solution. This resulted in a feed concentration of copper of 176 ppb or .176 mg/l.

Reactor Maintenance and Sampling Procedure

The reactors were maintained at a sludge age of five days and at a hydraulic detention time of twenty hours. The air flow into the reactors was found to vary pH by the production of H_2CO_3 so the air flows were kept the same throughout the experiment. The sludge age was maintained by measuring the flow, MLSS, and effluent SS, calculating total mass per day and the total effluent mass, and wasting the volume of mixed liquor calculated from Equation 6 that

removes one fifth of the total solids from the system daily. The reactors were started with sludge from the secondary underflow of the Third Creek Wastewater Treatment Plant, Knoxville, Tennessee, and was acclimated to the nutriment substrate. The reactors were then run for more than three sludge ages to insure that steady state conditions had been reached before any copper analysis was attempted.

A list of the samples taken and a schedule of the analysis performed is given in Table 3. The sample points are shown on Figure 1.

Analytical Methods

The COD determinations were made immediately after sampling the combined influent streams. The soluble effluent COD was defined as the soluble metal passing through a Reeve Angel glass fiber filter (Whatman, Inc., Clifton, New Jersey, #934 AH) under a vacuum of 19 inches of mercury. The method used was the micro semi-automated analysis for COD as reported by Jirka and Carter (40) using the HACH Chemical Company COD Reactor and pre-mixed COD digestion reagent vials with teflon caps. The vials came pre-mixed and it was only necessary to add 2 ml of sample and .05 grams of mercuric sulfate catalyst to the vials. The vials were sealed and heated for two hours at 150°C and allowed to cool before being analyzed spectrophotometrically by a Bausch & Lomb Spec 20 Model 33-29-40 that was adapted

TABLE 3
SAMPLING PROCEDURE

Analysis Performed	Frequency			Sampling Point		
	Daily	MWF	Last 4 Days	A	B	C
COD		x	x	x	x	
pH	x		x		x	
Cu total			x	x	x	
Cu +2			x	x	x	
TSS	x		x		x	x
VSS	x		x		x	x
Flow	x		x			x
Temp.	x		x		x	
O ₂ uptake		x	x		x	
ISV		x	x		x	
Cu solids			x		x	
Cu soluble			x		x	

to accommodate the HACH COD vials. A standard curve was prepared using dilutions of a stock solution of potassium acid phthalate with a COD of 1,000 mg/l. The standards and the samples were analyzed by determining the increase in Cr (III) concentration at a wave length of 600 n. The maximum limit of detection for this method was determined to be 450 mg/l of COD. Because the influent COD samples had COD values of approximately 400 mg/l all the influent COD samples were diluted 1:2 using distilled water. To minimize the effects of refraction and reflection, the vials were cleaned and rotated to give the maximum transmittance reading. The 100% transmittance reading was defined as the value obtained from a CO₂ free distilled water blank. The 0% transmittance reading was set with the light path totally blocked. This method for COD determination was chosen over the standard wet chemical method due to the ease of preparation, acceptable precision, and reproducibility.

The total suspended solids analysis were defined as the solids not passing through the previously mentioned glass fiber filters under a vacuum of 19 inches mercury. The filters were tared and the nonfilterable solids heated to 105°C and 550°C for determination of total suspended solids and fixed suspended solids, respectively, in accordance with section 208 Standard Methods (30).

The total carbon analyses were performed on a Beckman 915 Total Carbon Analyzer using 20 microfilters (μ l) aliquots. The total carbon channel was standardized with potassium acid biphthalate and the inorganic channel standardized using a sodium bicarbonate solution. All samples were tested within an hour after they were taken.

Trace copper analysis was made by atomic adsorption spectroscopy using a Perkin-Elmer Model 403 (Norwalk, Connecticut). Most samples were analyzed using the graphite furnace atomization technique, however some samples did require the standard flame head.

All samples were centrifuged for separation of soluble and solid phases using an IEC International Centrifuged Model HT (International Equipment Company, Needham Heights, Mass.) operated at 9,000 rpm for twelve minutes. The centrate was filtered using .45 μ m membrane filters, Millipore type HA (Millipore Corp., Bedford, Mass.), acidified using ultra-pure nitric acid and stored for analysis at a later date. The solids were digested in the presence of ultra-pure HNO_3 and ultra-pure HCl on a hot plate in 125 ml flasks. The solids were then diluted to 40.0 ml and stored. The influent copper and total mixed liquor copper samples were acidified immediately after they were obtained and stored. All labware was cleaned with warm six normal HCl and rinsed thoroughly with deionized water. A schematic of the copper analysis procedure is shown in Figure 3.

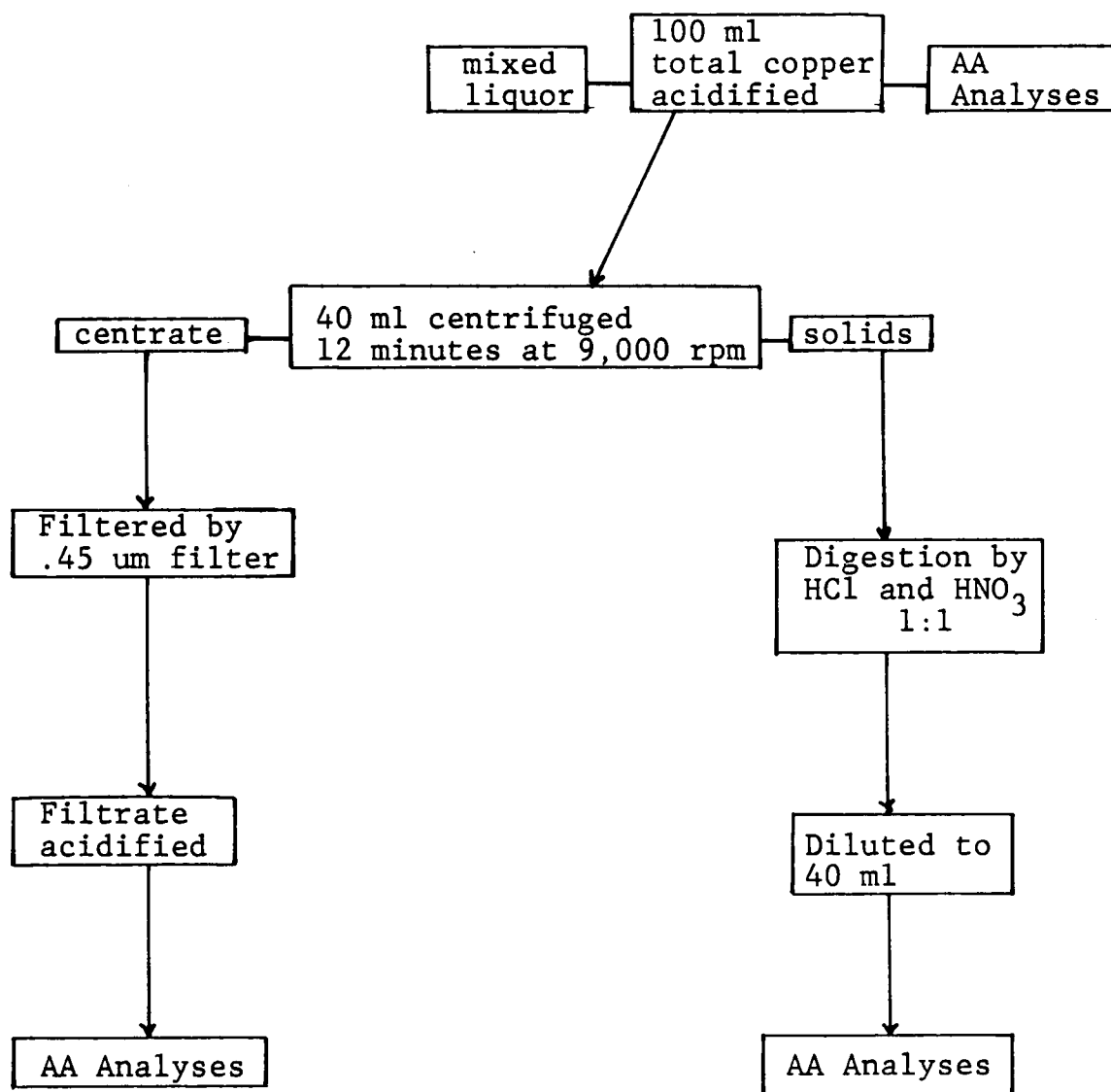


FIGURE 3
COPPER ANALYSES PROCEDURE

Copper samples analyzed using the graphite furnace technique used an ultraviolet wave length of 324.8 nm, a 10 microliter sample, drying at 150° for 34 seconds, charring at 550° for 22 seconds, and atomizing for 7 seconds at 2700°. A linear response was obtained from 10 to 200 ppb of copper. The flame analysis was performed using the optimum wave length of 324.8 nm and an air acetylene flame. The standards were prepared using plastic labware and mixed fresh as needed. All of the furnace sample cups used were checked to be free of copper contamination.

The activity of the activated sludge was monitored by determination of oxygen uptake. The mixed liquor was aerated to DO level of greater than 7 mg/l. A 300 ml sample of mixed liquor was removed from the reactor in a BOD Bottle. In order to determine the DO uptake rate, a YSI dissolved oxygen meter standardized against a sample of oxygen saturated distilled water was used to record several successive readings at 15-second intervals.

IV. EXPERIMENTAL RESULTS

System Performance

The reactors and internal clarifiers performed well in maintaining similar kinetic conditions in each of the ten reactors. Excessive losses of solids through the effluent were prevented and good solids settling in the secondary clarifier were obtained.

Some of the scatter in the substrate removal kinetics of the reactors was due to each reactor having two influent feed lines, one providing the copper and substrate with the other providing the buffer solution. If either one of the feed lines stopped or slowed, the influent characteristics changed substantially. The fluxuations could be minimized by each reactor being fed from one feed barrel and by one feed line.

It was not possible to use carbonate-bicarbonate buffer systems to control the pH because it was desired to maintain constant alkalinity. To hold the alkalinity variations of the influent feed to a minimum, the phosphate buffer systems in Table 2, p. 17, were used with acceptable results. The total inorganic carbon concentrations present in the influent feed and in the MLSS are given in Table 4.

As seen in Table 4, there is very little variation in the influent inorganic carbon concentrations with the exception of the concentration of reactors 5 and 6. The pH

TABLE 4
INORGANIC CARBON CONCENTRATION

Reactor	pH	Influent Inorganic Carbon (mg/l)	Mixed Liquor Inorganic Carbon (mg/l)
1	8.9	43.2	158.1
2	8.8	47.5	167.0
3	8.4	20.9	88.7
4	8.4	27.6	73.3
5	6.5	7.9	4.6
6	6.6	9.3	2.8
7	7.3	16.1	22.6
8	7.2	15.7	17.6
9	7.3	16.5	14.3
10	7.3	17.2	20.3

of the influent feeds to these two reactors was the lowest of all the feeds and the differences noted in the inorganic carbon concentration are a result of an increased transfer of CO_2 between the atmosphere and the buffer feed barrels due to the decreased pH. The changes in the influent inorganic carbon of the buffer barrels took place as equilibrium in the buffer solutions was approached over a period of time. The samples from the influent feeds were taken eight hours after the solutions were mixed.

The different values for the mixed liquor inorganic carbon shown in Table 4 are the result of carbon dioxide being introduced into the system from the atmosphere. Any increase in pH from the natural pH of reactors 7, 8, 9 and 10 will result in a shift in the form of the carbonate-bicarbonate system present in the system from CO_2 to

increased concentrations of $\text{CO}_3^{=}$ and OH^- . This shift results in a higher inorganic carbon mixed liquor concentration for the reactors maintained at the higher pH values.

The same relationship is found in reverse when the pH falls below the natural pH of reactors 7, 8, 9 and 10. As the pH is lowered, the concentrations of $\text{CO}_3^{=}$ and OH^- are lowered and the amount of CO_2 present increases, resulting in a decrease in the steady state inorganic carbon concentration.

Reactors 1 and 2 had noticeably higher MLSS and MLVSS values than the other reactors, resulting from a buildup of inorganic precipitates. In order to evaluate what the possible inorganic precipitates might be, a computer program developed at MIT was utilized. MINEQL, (the program name), was used for the calculation of the chemical equilibrium composition of aqueous systems. It did not take into account the effect of the organic material in the activated sludge systems. The concentrations of sodium, calcium, sulfate, iron, alkalinity, and magnesium for tap water were obtained from the yearly records maintained by the Knoxville Utilities Board. These values along with the concentrations of potassium, phosphate, copper and pH served as the input.

The output indicated that the possible precipitates were $(\text{Cu})_2\text{CO}_3(\text{OH})_2$, $\text{Ca}_5(\text{PO})_3\text{OH}$, $\text{Mg}_3(\text{PO}_4)_2$, $\text{Fe}(\text{OH})_3$, and

FePO_4 . The quantities of the iron precipitates were insignificant compared to the amounts of the other three precipitates and could not have been the cause of the elevated solids levels.

The program indicates that the amount of $\text{Ca}_5(\text{PO})_3\text{OH}$ that was precipitated was the same for all the phosphate buffered reactors. It also indicated that more $\text{Mg}_3(\text{PO}_4)_2$ was precipitated at pH 8.9 than at pH 8.4 and that none was precipitated at the lower pH values. More $(\text{Cu})_2\text{CO}_3(\text{OH})_2$ was precipitated in the reactors receiving substrate with added copper than the other reactors. At a pH of 8.9 and 8.4, twenty-four times as much copper is indicated to have precipitated than at pH 7.4, fifty times more copper is likely to have precipitated in reactors 7 and 8 than 9 and 10. At pH 6.4, no copper appeared to have been precipitated.

Because the program omitted the influence of the organic material, these values are only indications of the amounts that were precipitated. Though the precipitates would vary if the organic material had been included in the calculations, the amounts of precipitate in the reactors will still be relative to each other and the program therefore provides some indication as to the components of the solids.

To test the validity of the results of the computer program, the digested samples of adsorbed copper were analyzed for calcium and magnesium by atomic adsorption

spectrophotometry. The results are shown in Table 5 with 1.0 representing the relative concentration of magnesium and calcium at pH=8.9.

TABLE 5
COMPARATIVE CONCENTRATIONS OF Ca AND Mg

pH	8.9	8.3	7.4	6.4
Mg	1.0	.7	.3	.3
Ca	1.0	.63	.25	.25

The results indicate the buildup of solids is a result of the accumulation of $\text{Ca}_5(\text{PO}_4)_3\text{OH}$ and $(\text{Mg})_3(\text{PO}_4)_2$ that is more predominant at the higher pH values. The $(\text{Cu})_2\text{CO}_3(\text{OH})_2$ contribution to the total solids is considered significant in reactor 2 only based on marked increase in the adsorbed copper measurements for this reactor.

The increase in MLVSS for reactors 1 and 2 can also be explained by the presence of these inorganic precipitates. The hydrolyzed forms of $(\text{Cu})_2\text{CO}_3(\text{OH})_2$ and $(\text{Mg})_3(\text{PO}_4)_2$ disassociate at 200°C. When measuring the fixed solids, the temperature was increased from 105°C to 550°C so any of these forms present volatilized and their weight loss was counted as organic matter. Due to the apparently large amount of the inorganic precipitates present, significant error in the true MLVSS measurements for reactors 1 and 2 is very likely.

The problem with the inorganic precipitates can be greatly reduced through the use of distilled or deionized water. The difficulty encountered in measuring volatile solids can be reduced by measuring the solids change from a temperature above the disassociation temperature of the suspected precipitates to 550°C.

Biological Kinetics

In order to approach steady state conditions, the reactors were run for three sludge ages, fifteen days, before any analyses for copper partitioning were performed. The results of daily analyses performed for each reactor are given in the Appendix. In order to minimize the effects of sampling and analytical error, average values were calculated for each data point. Equation 8 was used to calculate the substrate removal rate for each reactor.

$$q = \frac{\bar{V}(\bar{S}_0 - \bar{S})}{X(9.0)} \quad \text{Equation 8.}$$

where $\bar{V}$ = average flow, l/day

X = MLVSS, mg/l

S_0 = influent COD, mg/l ($\bar{S}_0$ - average)

S = effluent COD, mg/l ($\bar{S}$ - average)

The average values for the kinetic data taken and for related calculated parameters are given in Table 6.

Because all the reactors were maintained at a five day sludge age, the range of sludge ages necessary to obtain the yield (Y) and endogenous decay coefficient (K_d)

TABLE 6

SUMMARY OF KINETIC DATA

Reactor	Flow	MLVSS	COD INF	COD EFF	1/COD EFF	Q	1/Q	S/Q	Yield	pH
1	12.0	1606.0	355.0	41.7	0.024	0.260	3.8	160.3	0.769	9.0
2	10.6	1456.0	394.0	35.0	0.029	0.290	3.4	120.5	0.689	9.0
3	11.5	1077.0	362.0	31.0	0.032	0.393	2.5	78.9	0.509	8.3
4	10.9	903.0	375.0	61.4	0.016	0.421	2.3	145.9	0.476	8.3
5	12.8	1283.0	362.5	43.3	0.023	0.354	2.8	122.3	0.565	6.3
6	10.2	1110.0	405.0	46.4	0.022	0.366	2.7	126.7	0.546	6.3
7	11.3	920.0	391.2	92.9	0.011	0.407	2.4	228.1	0.491	7.4
8	11.5	801.7	384.3	112.0	0.009	0.434	2.3	258.0	0.461	7.4
9	12.8	1001.7	375.0	81.7	0.012	0.416	2.4	196.1	0.480	7.4
10	11.0	1067.0	331.2	51.7	0.019	0.346	2.8	149.2	0.577	7.4

by plotting θ_c^{-1} vs. q was not available. By assuming that the rate of endogenous respiration was substantially less than the yield, Equation 7, upon rearranging, reduces to

$$Y_{\text{observed}} = \theta_c^{-1}/q \quad \text{Equation 9.}$$

Equation 9 was used to calculate the values for observed yield given in Table 6.

In order to verify that all ten reactors were operating at or near the same conditions, the values for S and q were plotted in Figures 4, 5, and 6. The plots in Figures 5 and 6 revealed a linear relationship that was determined using the least squares method for establishing linear regressions. The constants k and K_s obtained from these plots are given in Table 7.

The solid line in Figure 4 utilizes the kinetic coefficients obtained by plotting $1/q$ vs. $1/s$ in Figure 5 to show the relationship between the substrate concentration and the substrate removal rate. The agreement of the data points with this line indicates that similar conditions were existing in all ten reactors and that the changes in environmental conditions due to varying pH and copper concentrations did not effect the mixed culture adversely. The plot also indicates the dilute Nutriment was readily biodegradable as shown by the low K_s values listed in Table 7.

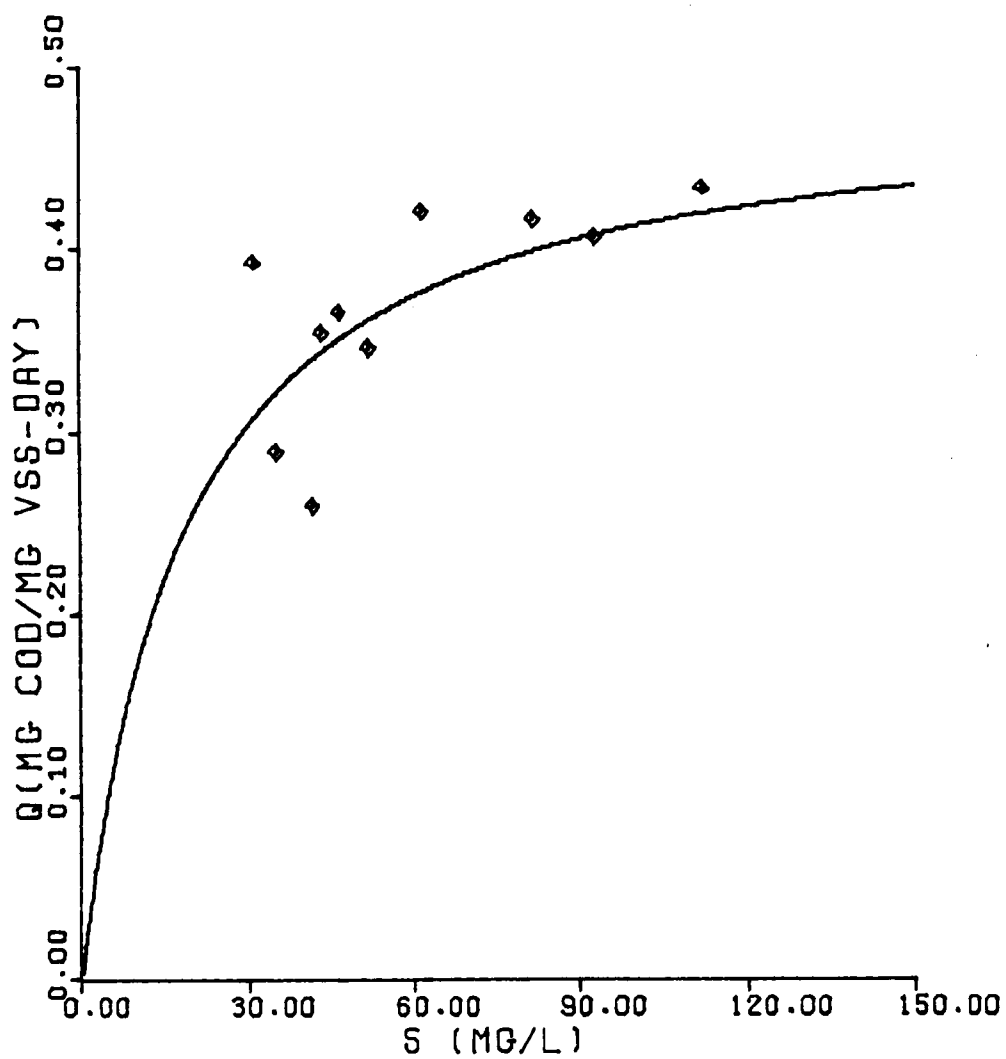


FIGURE 4

Q VERSUS S SUBSTRATE REMOVAL KINETICS

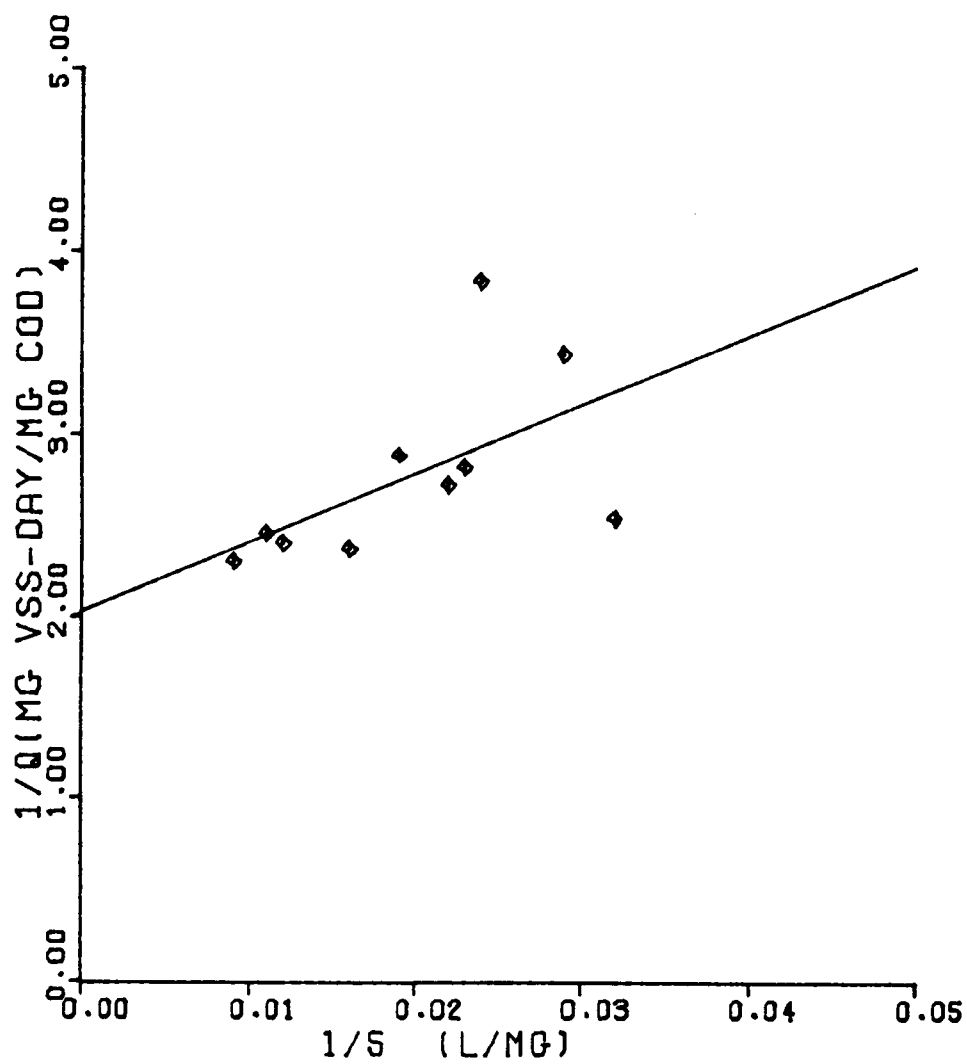


FIGURE 5

$1/Q$ VERSUS $1/S$ SUBSTRATE REMOVAL KINETICS

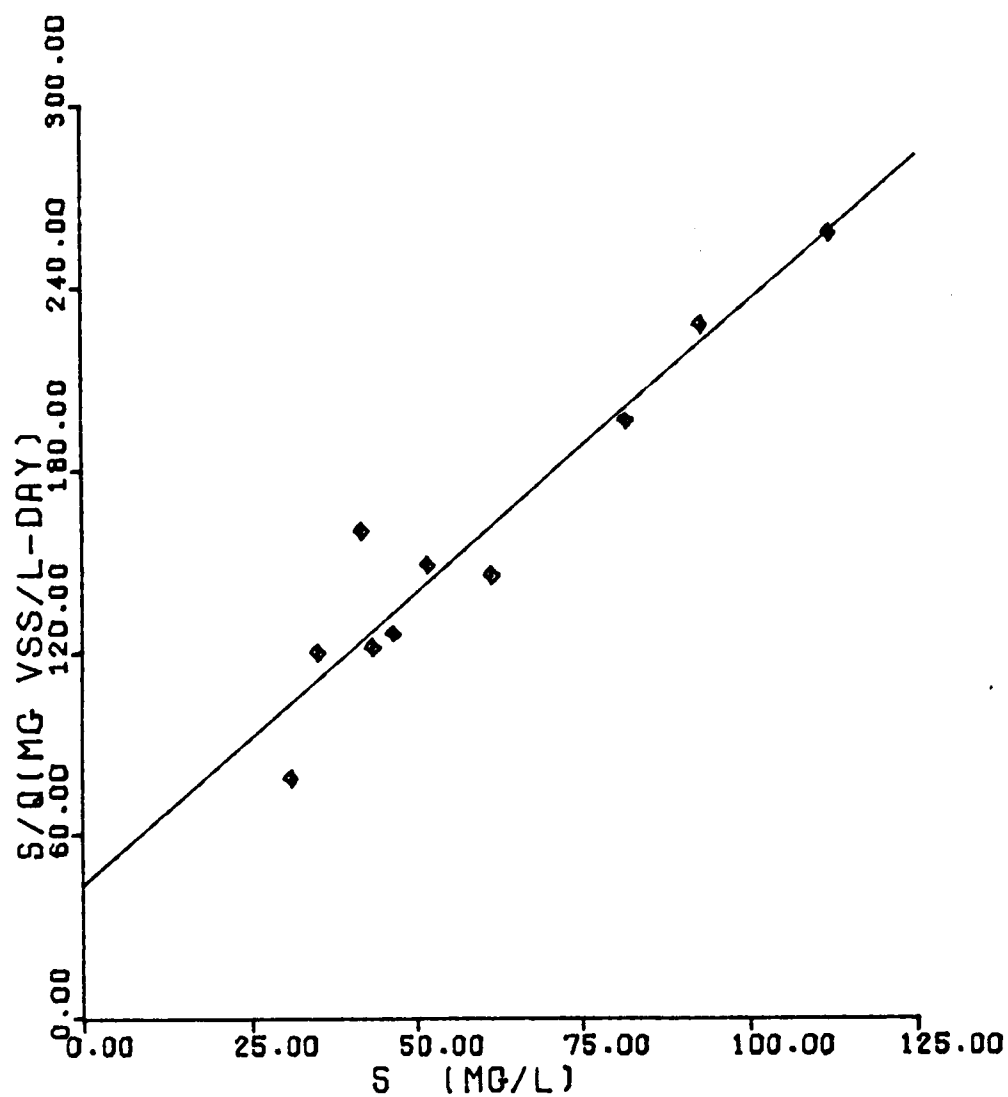


FIGURE 6

S/Q VERSUS S SUBSTRATE REMOVAL KINETICS

TABLE 7
KINETIC COEFFICIENTS

Method	k	K _s
1/q vs. 1/s	.487	18.1
S/q vs. S	.523	23.3

The observed yield values obtained using Equation 8 were plotted as a function of pH in Figure 7. The values were consistent with the exception of the higher yield values for reactors 1 and 2. The MLSS and MLVSS values are higher as a result of a buildup of hydrolyzed inorganic precipitates as discussed previously. The elevated yields are also a result of this. Substituting the average of the observed yield values from reactors 3 through 10 into Equation 8 and solving for MLVSS, gives calculated MLVSS values of 1225 mg/l and 1240 mg/l for reactors 1 and 2 respectively. These values more closely agree with the MLVSS values of the other eight.

Copper Partitioning

Because the results of the biological kinetics section indicate that all the reactors were maintained under similar substrate removal kinetics, the differences in the copper partitioning observed were assumed to be a result of the different pH values. A summary of all the data

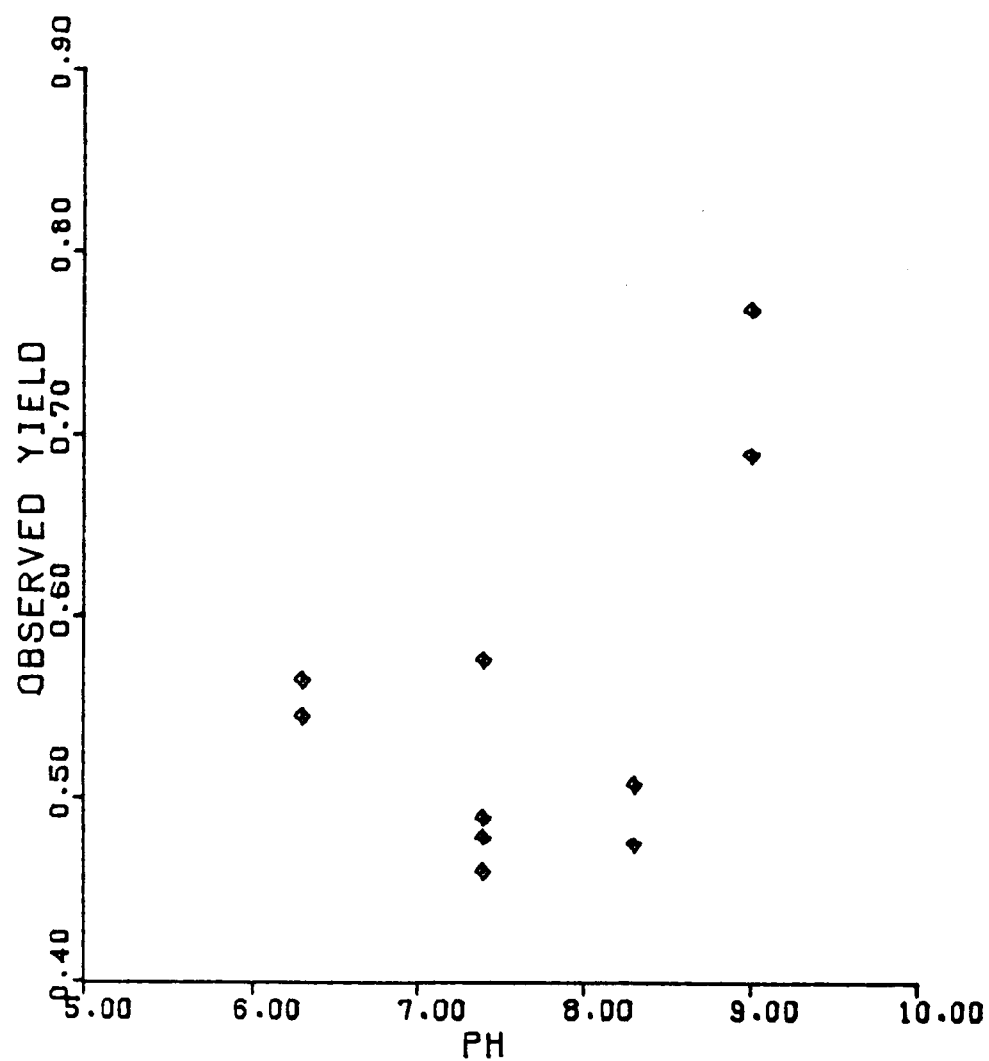


FIGURE 7

OBS. YIELD VERSUS pH SUBSTRATE REMOVAL KINETICS

pertaining to the analysis of copper partitioning is presented in Tables 8 and 9.

It was important that all five reactors in each group receive the same concentration of influent copper. To verify the amounts of copper in the influent feed, the data is plotted on Figure 8. The concentrations of copper to reactors 1, 3, 5, 9 and 10 clearly indicate that the same copper concentration was received by all the reactors in that group. The group of reactors with an elevated amount of copper shows that only a slight difference exists in the influent copper. The slight differences noticed are the result of pumping fluxuations and analytical procedure.

To compare the values of the MLSS and MLVSS, the data is plotted as a function of pH in Figures 9 and 10 respectively. In both figures, the reactors receiving the higher concentration of copper have lower solids values than the reactors at the same pH without the additional copper. The reason for the higher solids production in the reactors 1, 3, 5, 9 and 10 is that these reactors received more substrate per day on a mass basis as seen from Table 4 (page 27). The difference in the amount of substrate delivered resulted from the pump supplying these reactors operating at a slightly higher rate and the substrate these reactors received being slightly more concentrated. Substituting the larger mass of substrate into Equation 1 indicates that more bacteria is produced per day resulting in the higher concentrations of MLSS and MLVSS.

TABLE 8

SUMMARY OF COPPER DATA, REACTORS 1, 3, 5, 9, AND 10

	5	9	Reactor		1
			10	3	
pH	6.5	7.3	7.3	8.4	8.9
Influent Cu (PPB)	17.03	15.48	15.40	17.0	16.25
Soluble Cu (PPB)	14.8	12.6	13.2	13.0	13.4
Solid Complexed Cu (PPB)	112.6	68.8	53.0	122.1	189.9
Total Cu (PPB)	114.8	80.1	81.3	96.6	121.5
MLSS (mg/l)	1820.0	1412.5	677.5	1735.0	2797.5
MLVSS (mg/l)	1252.5	970.0	590.0	1175.0	1790.0
SUM Cu forms (PPB)	128.4	81.7	63.2	135.5	206.8
ΓMLSS μg Cu/mg MLSS	.0640	.0523	.0825	.0694	.0708
ΓMLVSS μg Cu/mg MLVSS	.0883	.0789	.0941	.1085	.1127
Predicted Solids Cu PPB	37.12	20.18	24.68	36.35	57.07
Cu Out (μg Cu)	439.9	361.7	518.8	452.6	604.9
Cu In (μg Cu)	217.9	198.0	452.6	195.5	195.0
Soluble/Influent	.82	.77	.86	.76	.81

TABLE 9

SUMMARY OF COPPER DATA REACTORS 2, 4, 6, 7, AND 8

	Reactor				
	6	7	8	4	2
pH	6.3	7.3	7.2	8.4	8.8
Influent Cu (PPB)	166.9	194.4	183.1	187.8	192.3
Soluble Cu (PPB)	85.6	118.9	115.0	102.4	94.4
Solid Complexed Cu (PPB)	348.4	322.4	255.1	219.6	623.2
Total Cu (PPB)	482.4	476.8	411.6	378.3	702.5
MLSS (mg/l)	1485.0	1130.0	980.0	1262.5	2342.5
MLVSS (mg/l)	1127.0	987.5	770.0	927.5	1535.0
Sum Cu Fractions (PPB)	440.8	438.6	871.4	312.1	726.9
MLSS (PPB Cu/mg)	.248	.300	.264	.169	.265
MLVSS (PPB Cui/mg)	.335	.390	.358	.229	.412
Predicted Solids (PPB)	461.0	473.2	434.9	517.1	576.0
Copper Out (μg)	1740.3	2165.8	2097.2	1380.1	2330.7
Copper In (μg)	1702.6	2196.2	2105.4	2046.5	2037.9
Soluble/Influent	.50	.60	.62	.54	.48

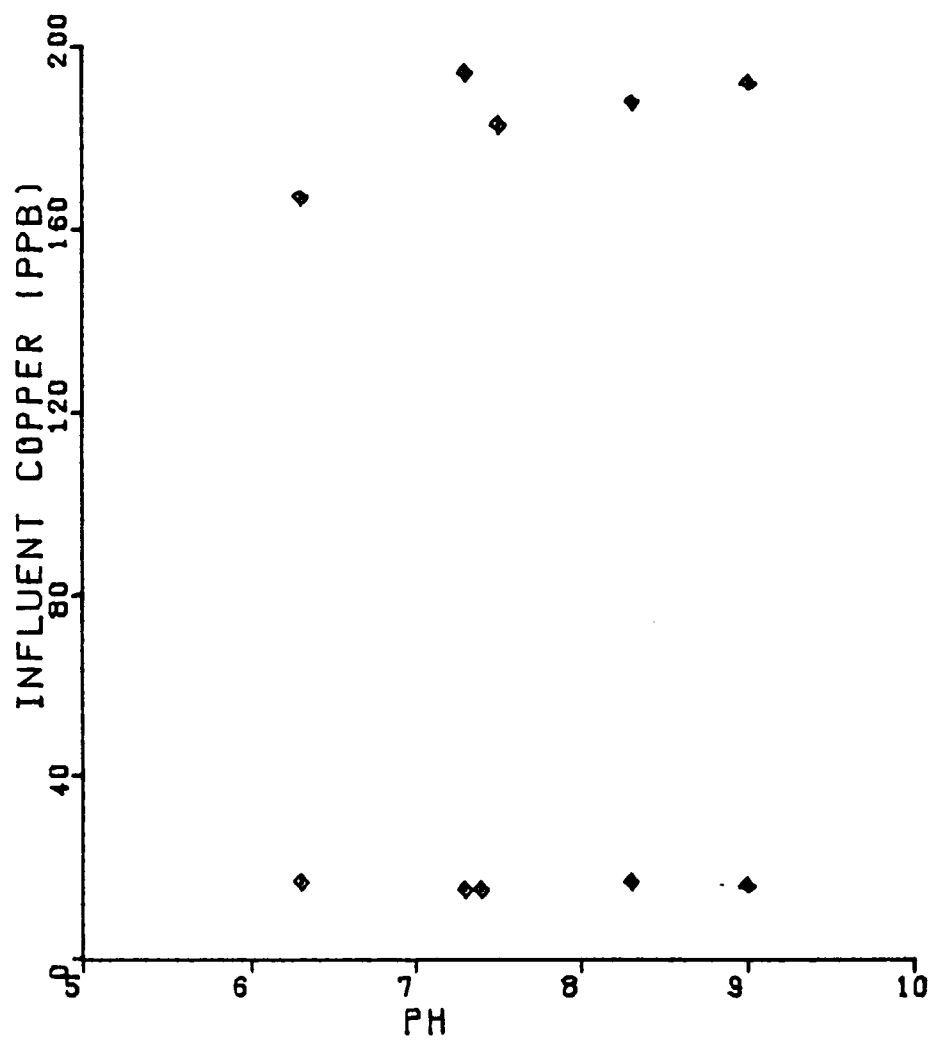


FIGURE 8
INFLUENT COPPER VERSUS pH

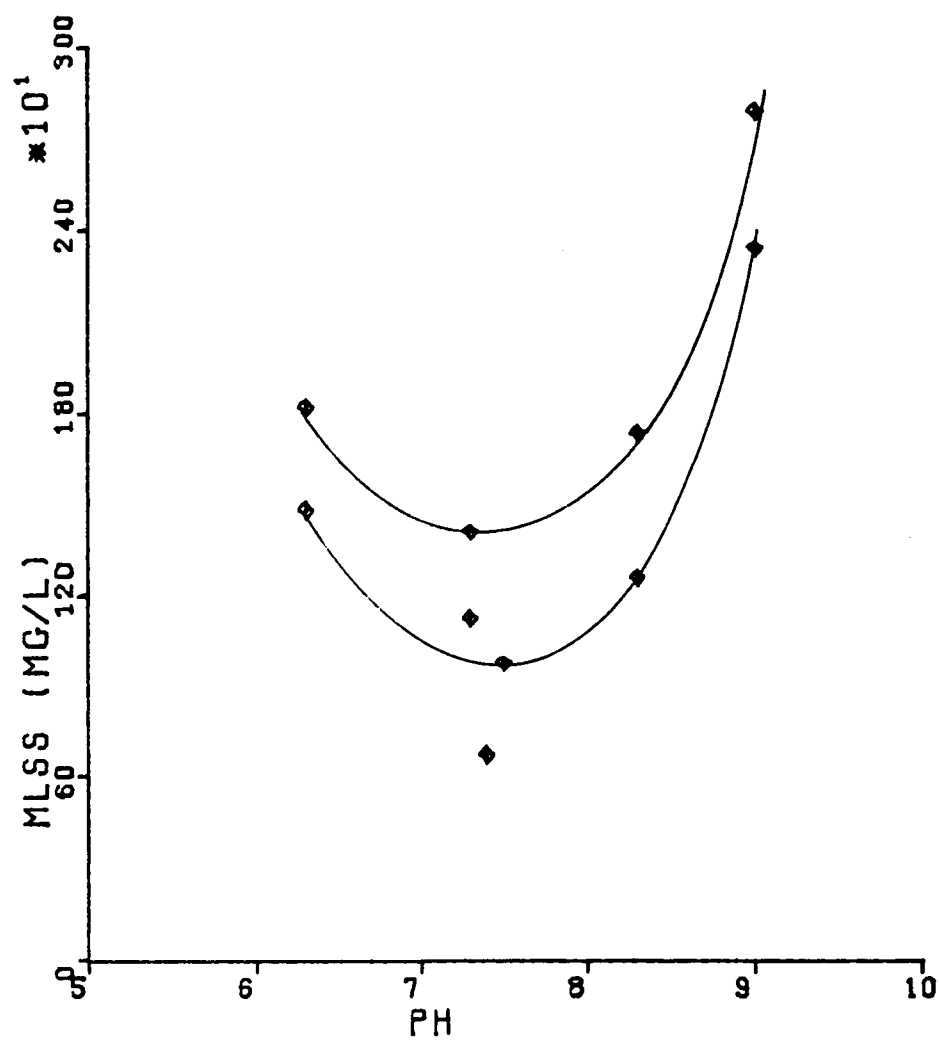


FIGURE 9
MIXED LIQUOR SUSPENDED SOLIDS VS. pH

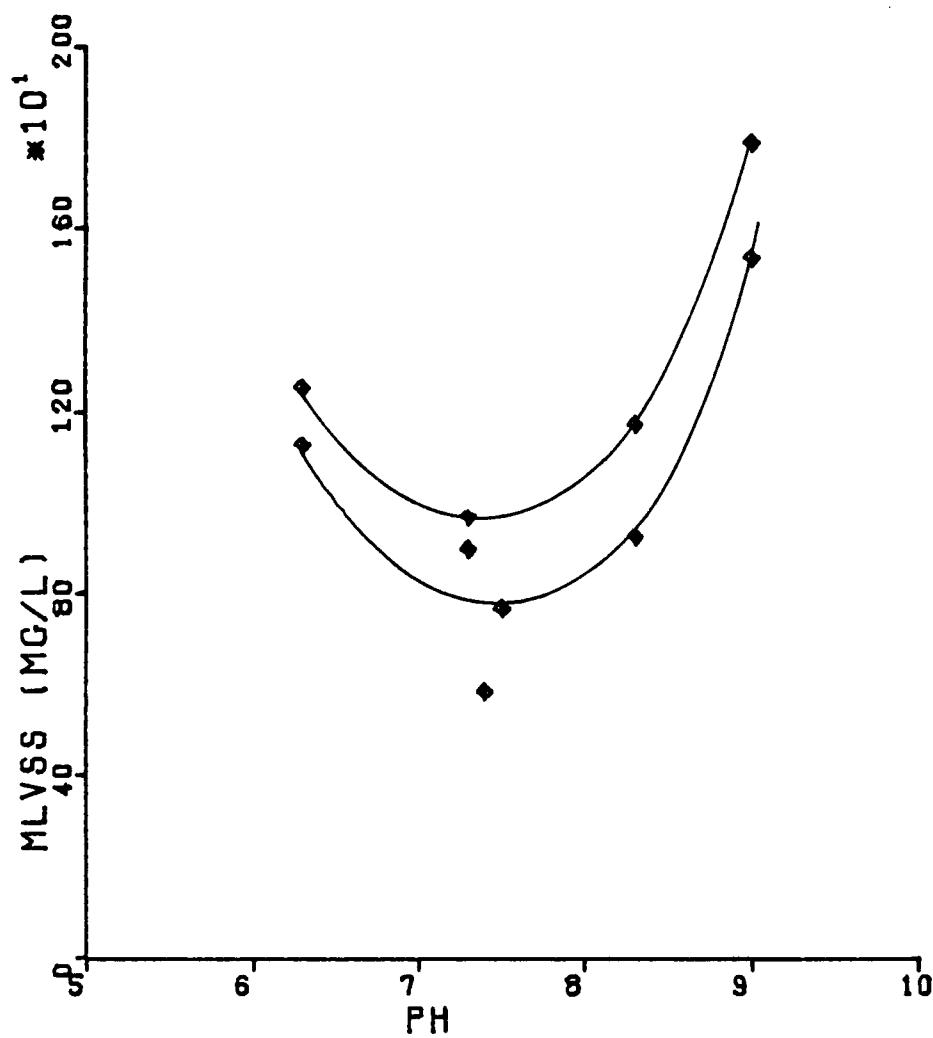


FIGURE 10

MIXED LIQUOR VOLATILE SUSPENDED SOLIDS VERSUS pH

The data plotted in Figures 9 and 10 also show that the production of solids varied as a function of pH with the reactors at pH 7.3 producing less solids and the reactors at pH 9.0 producing more solids. This is caused as discussed previously by the amounts of hydrolyzed precipitates which contribute to the MLSS and MLVSS values. The solids production appears to be very low for the reactors at pH 7.4 because no significant amounts of $\text{Ca}(\text{PO}_4)_3\text{OH}$ or $\text{Mg}_3(\text{PO}_4)_2$ were produced due to the absence of a phosphate buffer system at this pH.

The amount of copper present in the free ionic form was measured on several occasions through the use of a selective ion electrode. Analysis performed on the influent feed, the mixed liquor, and the effluent indicate that at all locations the amount of copper present in the free ionic form was less than 10^{-7} moles/liter. Measurements below this concentration were not possible because standards could not be prepared with less than this concentration of ionic copper. The high affinity of the free ionic form of copper to the organic material in the influent substrate and to the organic constituents in the mixed liquor resulted in the low measurement obtained.

An effort was made to monitor the change in the soluble copper present in the mixed liquor as a function of pH and influent copper concentration. A plot of the soluble copper as a function of PH is given on Figure 11.

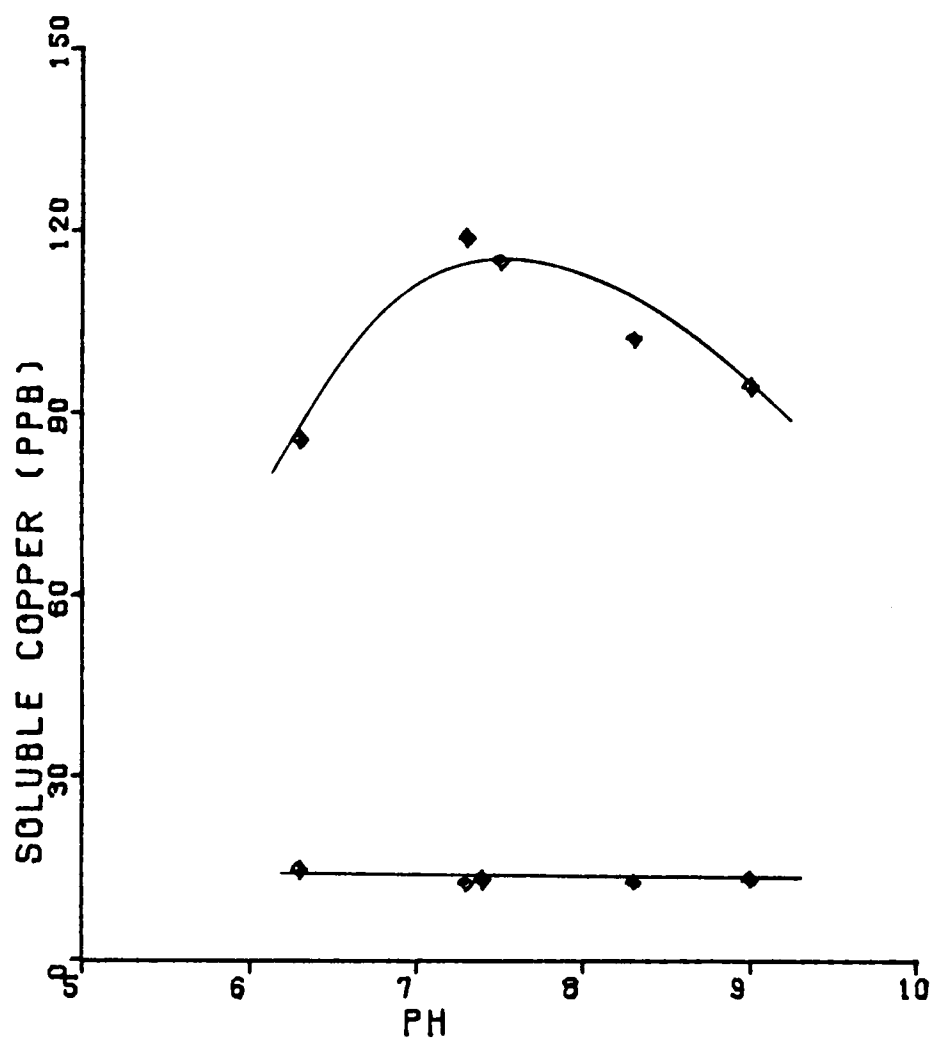


FIGURE 11
SOLUBLE COPPER VERSUS pH

The plot shows very little change in soluble copper as a function of pH for the reactors that received no additional copper in the substrate. In the reactors receiving the elevated copper concentrations a marked variation is apparent in the soluble copper present. Upon closer examination, the variation in soluble copper appears to be reflecting the variations in solids production as discussed earlier. The reactors with the lowest solids concentration had the highest soluble copper present and the reactors with the highest solids concentration had the least soluble copper. The reactors with the higher solids concentration have more organic material present and therefore more adsorption sites for which the copper present has a high affinity. The number of organic adsorption sites present and the high affinity of the copper molecules to these sites rather than the soluble forms explains the variations observed in Figure 11.

By plotting the concentrations of adsorbed copper in Figure 12, the same pattern was obtained as from the plots of the MLSS and the MLVSS. Because most of the copper present in all the systems was in the adsorbed form, it is to be expected that in the reactors with the higher solids concentration the most adsorbed copper would exist. To confirm that most of the copper present was in the adsorbed form and that it should closely parallel the solids present, the total copper concentrations were plotted as a function of pH on page 52. This figure follows closely the pattern observed in Figure 12, the adsorbed copper vs pH.

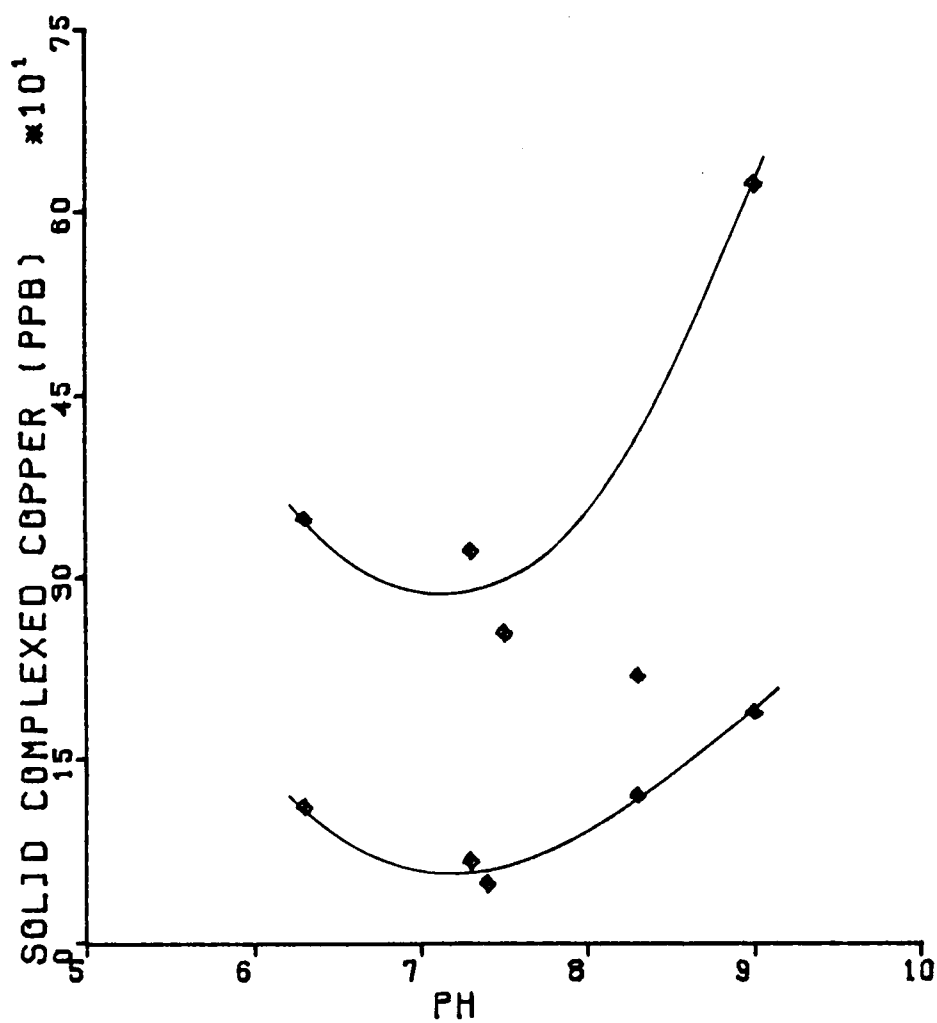


FIGURE 12

SOLID COMPLEXED COPPER VERSUS pH

In batch copper uptake tests performed by other authors, the amount of soluble copper varies as a function of pH due to the increased adsorption capability of the biomass. To determine if this was the case in a steady state system where the copper is continuously replaced, the ratio of influent copper was calculated and plotted on Figure 13. From this figure, no significant change in the soluble copper as a function of pH is indicated.

The plotting of the soluble copper as a fraction of the sum of the soluble copper and adsorbed copper indicates a dependency upon the amount of solids in the system. Figures 14 and 15 show the dependency of these parameters upon the variation of the solids values as discussed previously.

The amount of copper adsorbed per unit of biomass was found to increase with increasing pH under previous batch copper addition tests. To verify this in steady state conditions, isotherm data were calculated by dividing the concentration of copper present in the adsorbed form by the MLVSS. This ratio will be referred to as Γ_{MLSS} when the adsorbed copper concentration is divided by the MLSS and Γ_{MLVSS} when divided by the MLVSS. These parameters are plotted as a function of pH in Figures 17 and 18. The values for reactor 4 are not considered valid because of difficulties encountered in determining the adsorbed copper measurement. From Figure 18, it is seen that the amount of copper

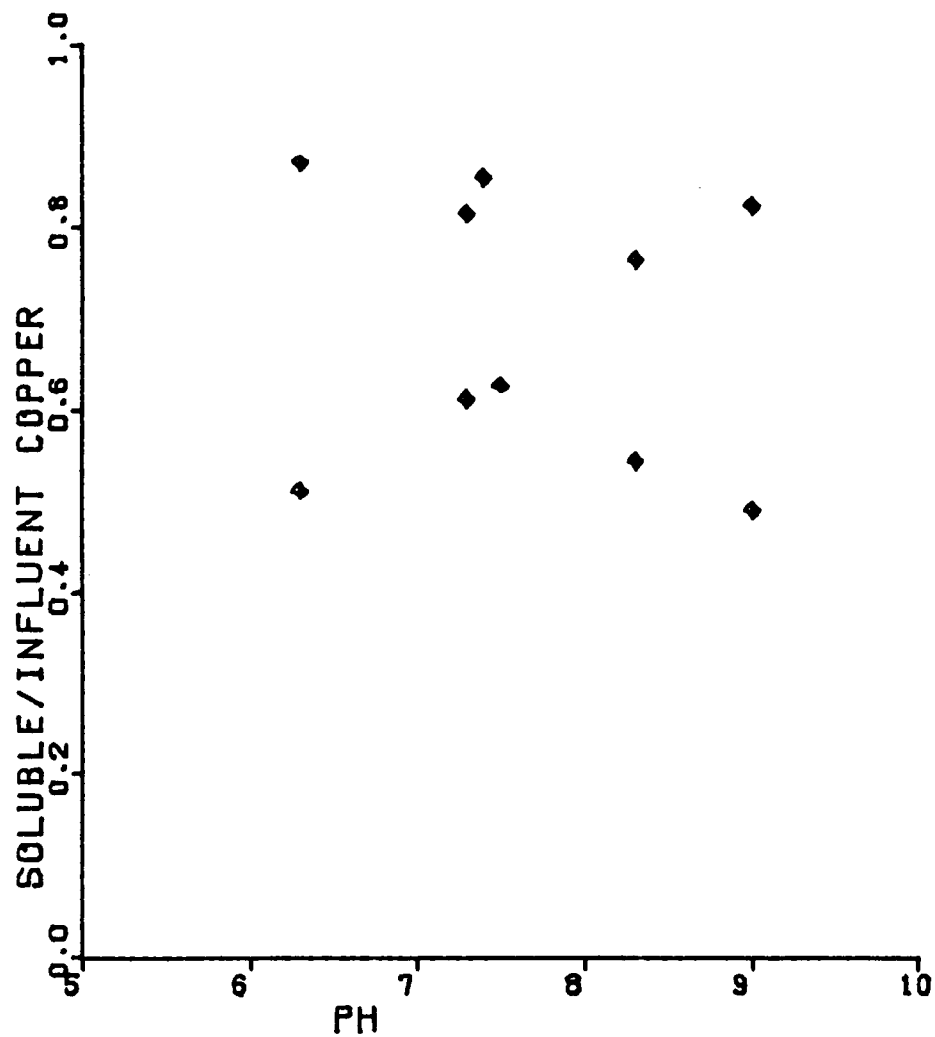


FIGURE 13
SOLUBLE/INFLUENT COPPER VERSUS pH

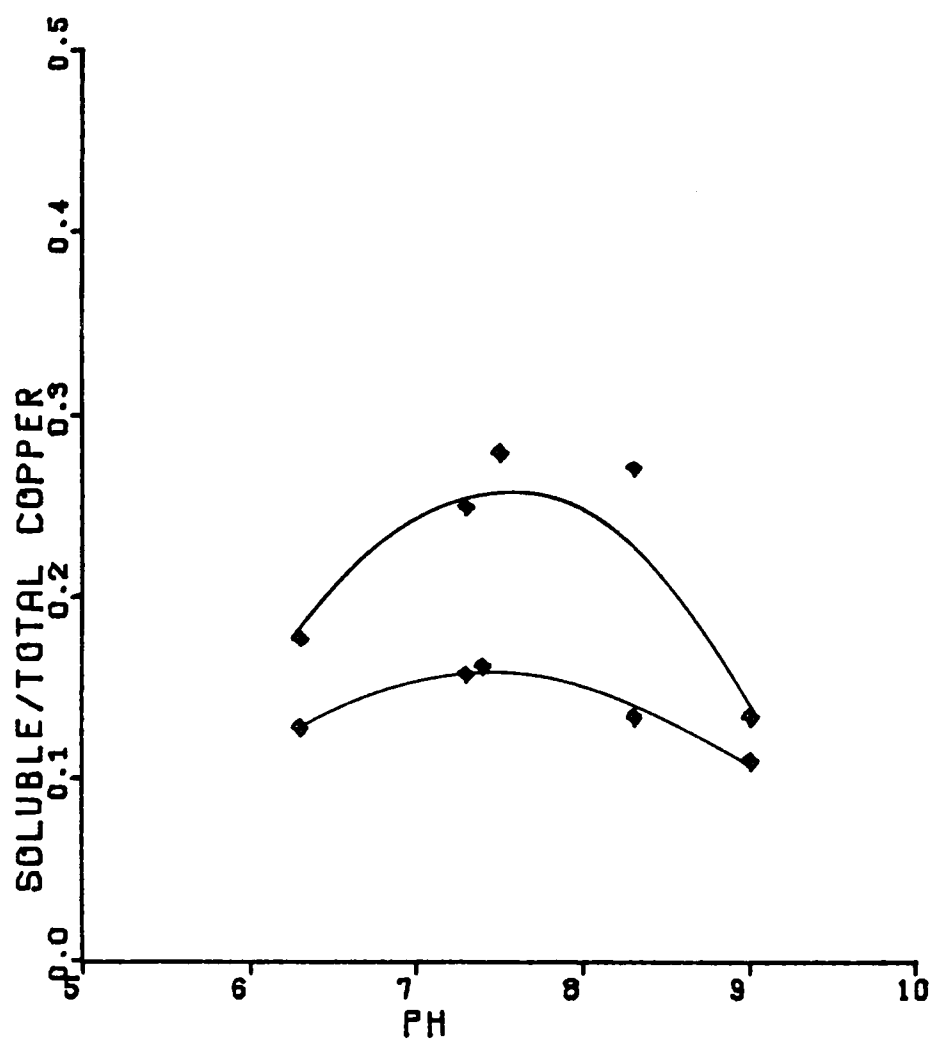


FIGURE 14

SOLUBLE/TOTAL COPPER VERSUS pH

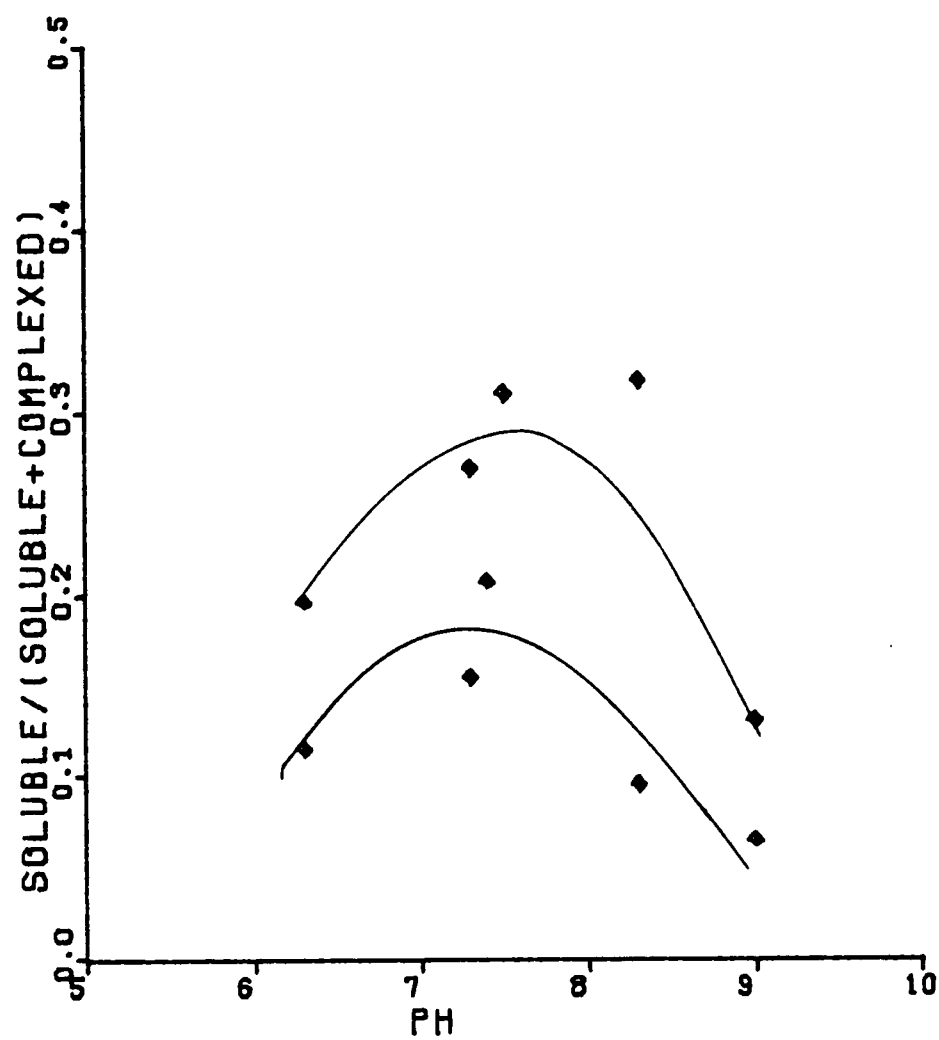


FIGURE 15

SOLUBLE COPPER/SOLUBLE + SOLID COMPLEXED COPPER VERSUS pH

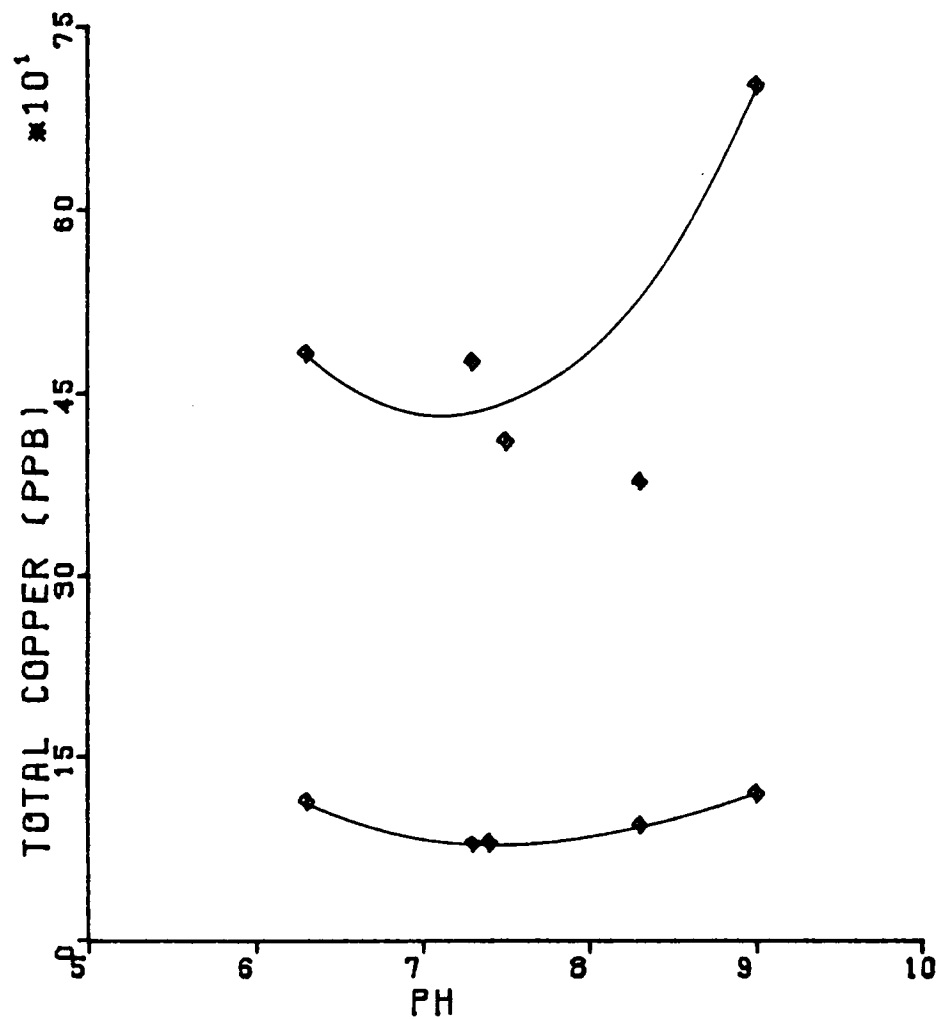


FIGURE 16
TOTAL COPPER VERSUS pH

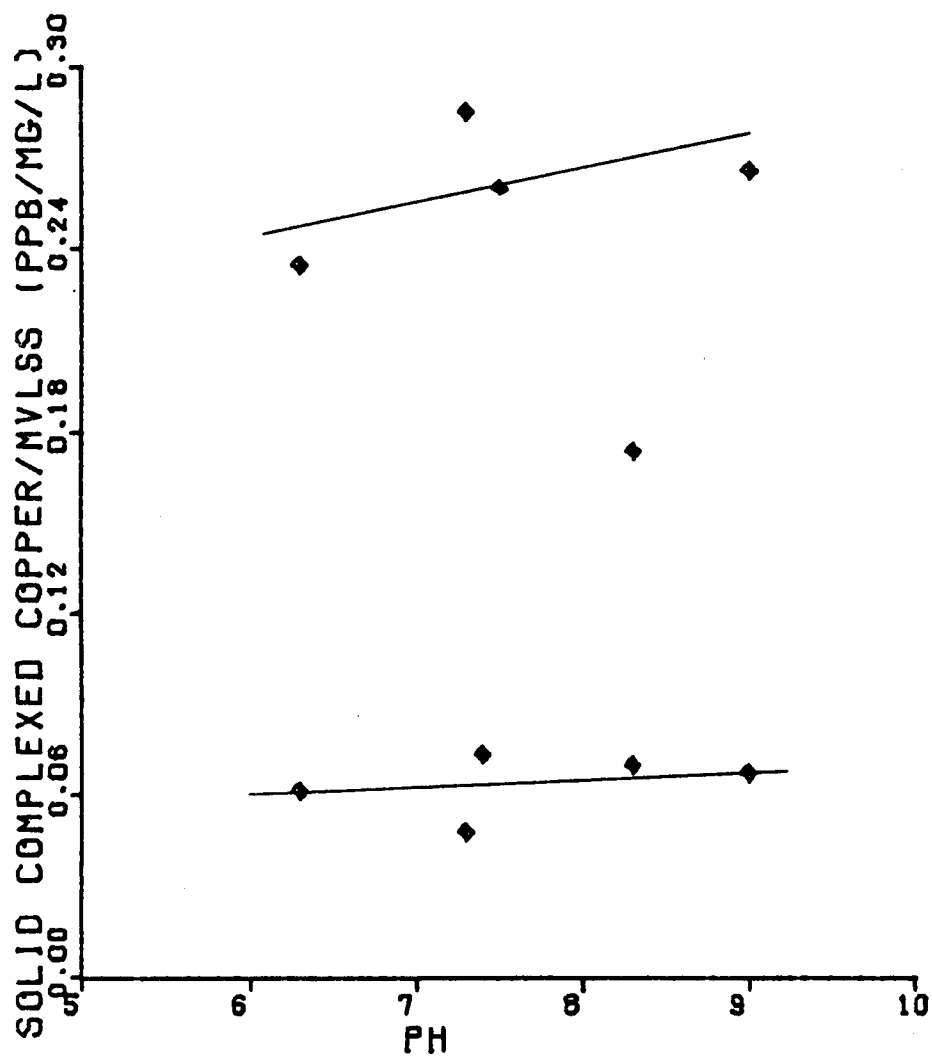


FIGURE 17

SOLID COMPLEXED COPPER/MLSS VERSUS pH

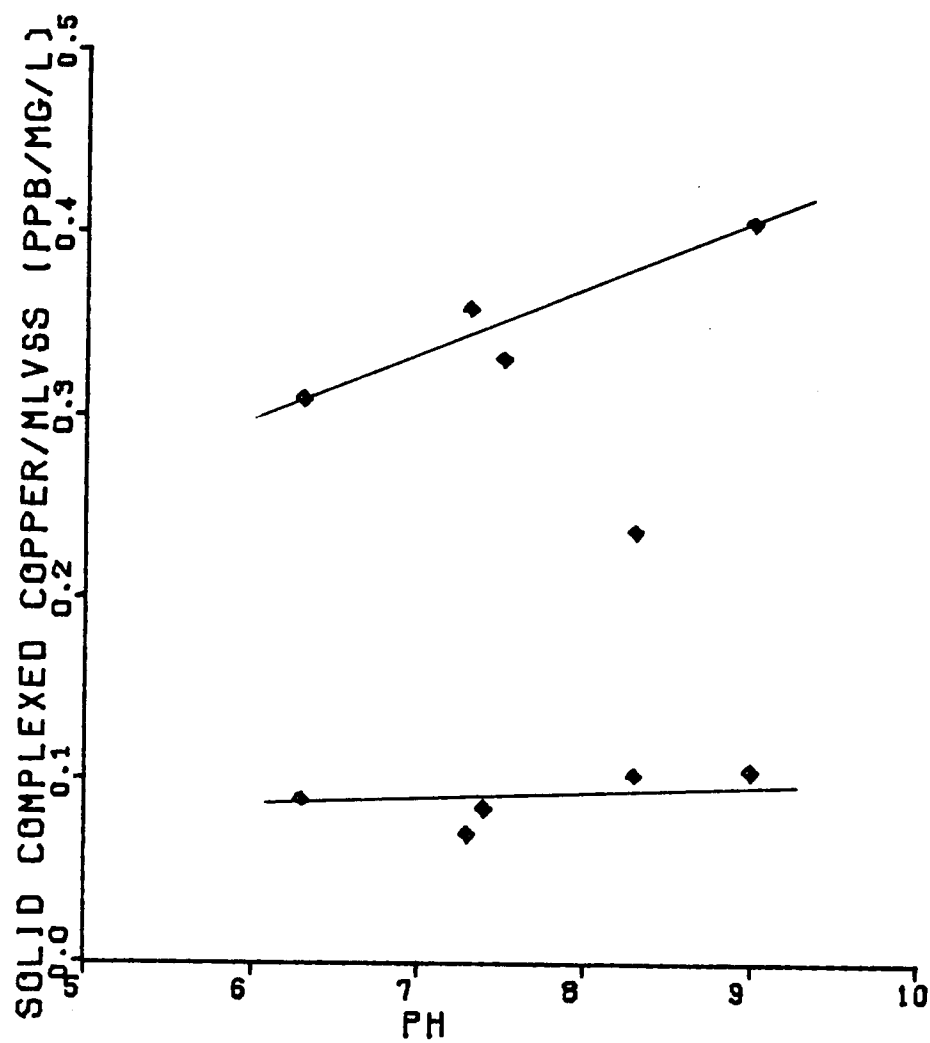


FIGURE 18

SOLID COMPLEXED COPPER/MLVSS VERSUS pH

complexed per unit of biomass increases with increasing pH for both concentrations. At higher levels of copper the amount adsorbed increased more than the lower concentration indicating that the extent that pH effects the adsorption capacity of organic material is governed by the copper concentration present.

To illustrate the effect that the concentration of copper present had on the adsorption capacity of the activated sludge biomass, r_{MLSS} and r_{MLVSS} were plotted in Figures 19 and 20 as a function of the concentration of soluble copper surrounding the biomass. The resulting isotherm indicates that the adsorption capacity is greatly affected by the extent to which copper is present in the system, which appears to depend on the soluble COD.

To check the accuracy of the analytical methods, a mass balance was performed upon each of the reactors. The copper data given in Tables 8 and 9, pages 40 and 41, were calculated using the concentration of copper in the influent and the volume of influent per day to obtain the mass of copper into the reactor. The copper out was calculated by summing the soluble and adsorbed copper removed from the reactors each day as follows:

$$Cu_{out} = Q[(r_{MLVSS})(Eff V_{ss}) + Cu_{sol}] + V[(r_{MLVSS})(MLVSS) + Cu_{sol}]$$

where Q = daily flow in liters

V = waste volume removed daily in liters

Cu_{sol} = soluble copper, ppb

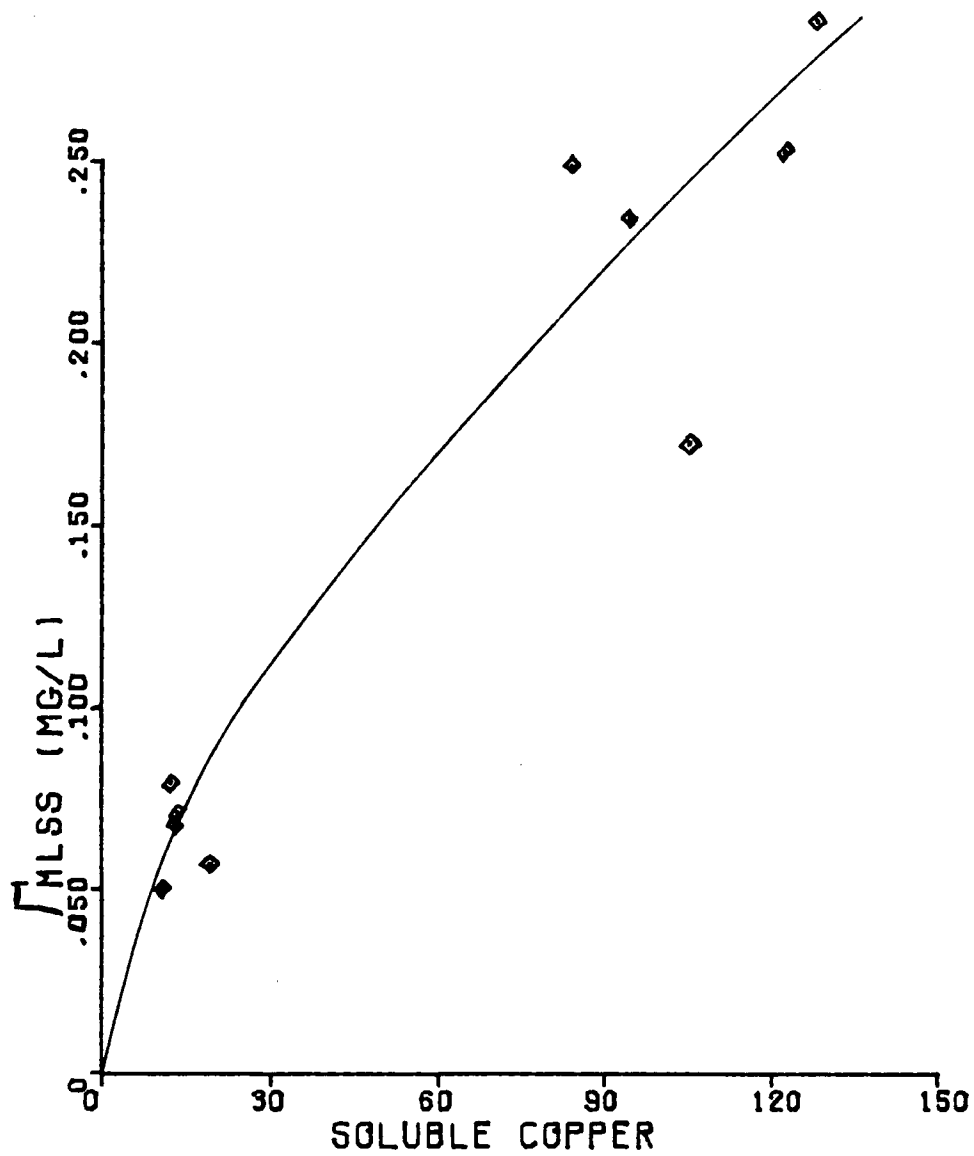


FIGURE 19

MIXED LIQUOR SUSPENDED SOLIDS VERSUS SOLUBLE COPPER

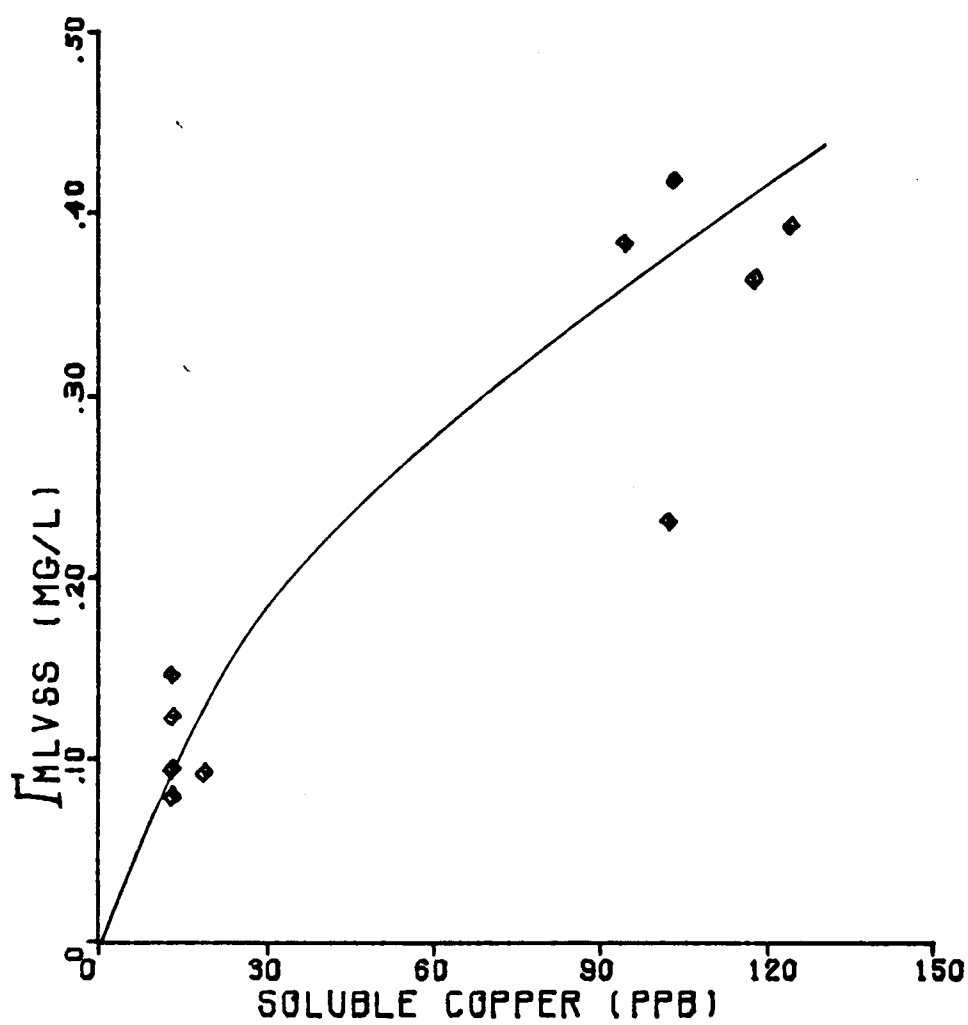


FIGURE 20
MIXED LIQUOR VOLATILE SUSPENDED SOLIDS
VERSUS SOLUBLE COPPER

The copper partitioning analyses were not performed on the effluent so for the calculations it was assumed that r_{MLVSS} is constant for both the MLVSS and the effluent VSS. The data given in Tables 8 and 9, pages 40 and 41, show that the calculated effluent copper closely agrees with the influent copper for reactors 2, 6, 7 and 8. This indicates that these reactors were definitely at steady state when the tests were made. The influent copper and effluent copper values do not agree as well for reactors 1, 3, 5, 9 and 10, the reactors that received no additional copper. The difference in these parameters does not indicate that reactors 1, 3, 5, 9 and 10 were not at steady state. It does indicate that the procedures used to measure the copper fractions were much more susceptible to contamination at the lower concentration of copper than at the higher concentration. Contamination resulting in an increase of only 4 ppb in the effluent copper concentration would cause a substantial error in the mass of copper out. Assuming that using r_{MLVSS} in calculating the effluent adsorbed copper was too high, the effluent copper value for the reactors with no added copper would be lowered substantially more than the effluent copper value for the other values.

Also given in Tables 8 and 9 are values for the predicted values for the solid complexed copper obtained from:

$$\text{Predicted copper} = (\text{influent copper} - \text{effluent soluble copper})_{\theta} - \theta_c \text{ where } \theta \text{ is the hydraulic detention time.}$$

The values indicate poor agreement with the actual values of adsorbed copper and the predicted values. The values for the reactors receiving no additional copper indicate that more copper was accumulated by the bacteria than should have if the copper acted as a particulate substance. This indicates that at low concentrations, the copper is not a result of only particulate buildup, but also bacterial accumulation.

V. CONCLUSIONS AND RECOMMENDATIONS

Conclusions

1. The amount of copper present in the free ionic form was less than 10^{-7} moles/liter. This is due to the high affinity of the copper for the organic material in the activated sludge process.

2. The amount of soluble copper present does not vary appreciably with pH but is greatly affected by the concentration of solids in the mixed liquor. Treatment plants with effluent copper concentrations in excess of imposed standards could lower the effluent concentration by increasing the suspended solids removal. This would lower the copper concentration in the effluent because the majority of the copper present is adsorbed to the organic material present.

3. The adsorption capacity of the organic material in the activated sludge process increased with increasing pH when measured as micrograms of copper per milligram of MLVSS, the same trend shown in previous batch copper uptake studies. Treatment plants with excessive copper concentrations in their waste sludge could lower the concentration of copper by lowering the pH of the mixed liquor.

4. The amount of copper adsorbed under steady state conditions is controlled to a greater extent by the influent copper than the pH. This indicates it would be better to control the copper at its source rather than to try to change

the form the copper takes in the mixed liquor through pH adjustment.

Recommendations

1. The research performed in this paper should be repeated using de-ionized water which will greatly reduce the amount of inorganic precipitates present and allow for a quantitative relationship to be determined between the copper and the MLVSS.

2. Carry the work of Neufeld (18) and others a step further by developing a kinetic and equilibria model for copper that accounts for the change in adsorption capacity due to pH change.

BIBLIOGRAPHY

BIBLIOGRAPHY

- (1) Water Quality Criteria 1972. 1972. National Academy of Sciences, National Academy of Engineering, Washington, D.C.
- (2) McKee, J. E. and Wolf, H. W. 1971. Water Quality Criteria. State Water Resources Control Board, California.
- (3) Williams, S. H., Aulenbach, B. and Clesceri, N. C. 1974. Sources and Distribution of Trace Metals in Aquatic Environments. Aqueous-Environmental Chemistry of Metals. Ann Arbor Pub. Inc., Ann Arbor, Michigan.
- (4) Rancitelli, L. A., Perkins, R. W., Tanner, T. M. and Thomas, C. W. 1970. Precipitation Scavenging. Atomic Energy Commission, Symposium Series 22. Washington, D.C.
- (5) Williams, S. L., Culon, E. M., Kahberger, R. and Clesceri, N. L. 1973. Bioassay Techniques and Environmental Chemistry. Ann Arbor Sci. Pub., Ann Arbor, Michigan.
- (6) National Air Pollution Control Administration. 1966. Air Quality Data from the National Air Sampling Networks and Contribution State and Local Networks, 1964-1965. Cincinnati, Ohio.
- (7) U.S. Public Health Service. 1962. Air Pollution Measurements of the National Air Sampling Network--Analysis of Suspended Particles, 1957-61, Washington, D.C.
- (8) Cotten, J. M. 1977. Trace Metal Removal by Wastewater Treatment. Technology Transfer. National Air Pollution Control Administration, Cincinnati, Ohio.
- (9) Keeney, D. R. and Walsh, I. M. 1975. Heavy Metal Availability in Sewage Sludge Amended Soils. Jn. Proc. Int. Conf., Heavy Metals in the Environment, Toronto, Canada.
- (10) Cunningham, J. D., Keeney, D. R., and Ryan, J. A. 1975. Yield and Metal Composition of Corn and Rye Grown on Sewage Sludge Amended Soil. Journal of Environmental Quality, 4:448-454.

- (11) Cheng, M. H. 1973. Interactions of Heavy Metals in the Activated Sludge Process. Ph.D. Thesis, Illinois Institute of Technology.
- (12) Mikolaitis, J. C. 1977. Complexation and Removal of Copper in a Secondary Effluent. M. S. Thesis, Illinois Institute of Technology.
- (13) Haas, C. N. 1974. Soluble Phase Chemistry of Trace Metal Transport In Secondary Wastewater Treatment Systems. M.S. Thesis, Illinois Institute of Technology.
- (14) Patterson, J. W., Shimada, P. and Haas, C. N. 1975. Heavy Metals Transport Through Municipal Sewage Treatment Plants. Paper presented at the Second National Conference on Complete Water Reuse.
- (15) Lawrence, A. S. and McCarty, P. L. 1970. Unified Basis for Biological Treatment Design and Operation. Journal American Society Civil Engineers, 96, SAE, 754.
- (16) Sawyer, C. N. and McCarty, P. L. 1967. Chemistry for Sanitary Engineers. McGraw-Hill, New York.
- (17) Dugan, P. and Fister, P. Implications of Microbial Polymer Synthesis in Waste Treatment and Lake Eutrophication, Proc. 5th Int. Water Poll, Res. Conf. (July 1970).
- (18) Neufeld, R. D. and Hermann, E. R. Heavy Metal Removal by Acclimated Activated Sludge, Journal Water Poll. Control Fed., 47:310 (1975).
- (19) Lehninger, A. L. 1970. Biochemistry. Worth, New York.
- (20) Bowen, H. J. M. 1966. Trace Elements in Biochemistry. Academic Press, New York.
- (21) Stones, T. 1958. The Fate of Copper During the Treatment of Sewage. Institute of Sewage Purification Journal and Proceedings, part 1:82-3.
- (22) McDermott, G. N., Moore, W. A., Post, M. A. and Ettinger, M. B. 1963. Effects of Copper on Aerobic Biological Sewage Treatment. Journal of the Water Pollution Control Federation, 35:227-41.
- (23) Jirka, A. M. and Carter, M. J. 1975. Micro Semi-Automated Analysis of Surface and Wastewaters for Chemical Oxygen Demand, Analytical Chemistry, 47:1397.

- (24) Sudo, R. and Aiba, S. 1973. Effect of Copper and Hexavalent Chromium on the Specific Growth Rate of Ciliata Isolated from Activated Sludge, Water Research, 7:1301.
- (25) Poon, C. P. C. and Bhayani, K. H. 1971. Metal Toxicity to Sewage Organisms, Journal of the Sanitary Engineering Division of the ASCE, 97:161.
- (26) Rudolfs, W. 1950. Review of the Literature on Toxic Materials Affecting Sewage Treatment Processes, Streams and BOD Determinations, Sewage and Industrial Wastes, 22(9):1157-1191.
- (27) Barth, E. B., Ettinger, M. B., Salotto, B. V. and McDermott, G. N. 1965. Summary Report on the Effects of Heavy Metals on the Biological Treatment Processes, Journal of the Water Pollution Control Federation, 37(1):86-96.
- (28) Patterson, J. W. 1971. Modes of Toxicity Analysis in Activated Sludge. Presented before the Division of Water, Air and Waste Chemistry of the American Chemical Society.
- (29) Lamb, A. and Tollefson, E. L. 1973. Toxic Effects of Cupric, Chromate and Chromic Ions on Biological Oxidation, Water Research, 5:585-99.
- (30) American Public Health Association. 1975. Standard Methods for the Examination of Water and Wastewater. 14th edition, Washington, D.C.
- (31) Stiff, M. J. 1971a. Copper/Bicarbonate Equilibria in Solutions of Bicarbonate Ion at Concentrations Similar to Those Found in Natural Water, Water Research, 5:171-6.
- (33) Metcalf and Eddy, Inc. 1972. Wastewater Engineering. McGraw-Hill Book Co., New York.
- (34) Jenkins, S. H., Keight, D. G. and Ewins, A. The Solubility of Heavy Metal Hydroxides in Water, Sewage and Sewage Sludge--II: The Precipitation of Metals by Sewage, Journal Air and Water Poll., 8:537 (1964).
- (32) Stiff, M. J. 1971b. The Chemical States of Copper in Polluted Fresh Water and a Scheme of Analysis to Differentiate Them, Water Research, 5:585-99.

- (35) Rudolfs, W. and Zuber, A. L. Removal of Toxic Materials by Sewage Sludge, Sewage and Industrial Wastes, 25(2):142 (1953).
- (36) Chau, Y. K. 1973. Complexing Capacity of Natural Water--Its Significance and Measurement, Journal of Chromatographic Science, 11:579.
- (37) Kunkel, R. and Manahan, S. E. 1973. atomic Absorption Analysis of Strong Heavy Metal Chelating Agents in Water and Wastewater, Analytical Chemistry, 45: 1465-8.
- (38) Tenney, M. W. and Stumm, W. Chemical Flocculation of Microorganisms in Biological Waste Treatment, Journal Water Poll. Control Fed, 37(10):1370 (1965).
- (39) Neufeld, R. D., Gutierrez, J. and Novak, R. A. 1977. A Kinetic Model and Equilibria Relationship for Heavy Metal Accumulation, Journal of Water Pollution Control Federation, 49(3):489.
- (40) Klein, L. A., Lang, M., Nash, N. and Kirschner, S. L. 1974. Sources of Metals in New York City Wastewater, Metal Finishing, July, 34.

APPENDIX

TABLE A-1

REACTOR #1

Date	1/18	1/20	1/23	1/25	1/27	1/28	1/29	1/30	1/31
Influent									
COD (mg/l)	---	400	460	330	330	400	260	340	320
Cu (ppb)	---	---	---	---	19.9	13.9	19.4	11.8	0
Effluent									
V (l)	10	13	10	13	12	11	14	12	13
TSS (mg/l)	240	150	90	170	0	150	90	170	110
VSS (mg/l)	---	---	80	100	-	50	60	160	60
FSS (mg/l)	---	---	10	70	0	100	30	10	50
Wasted Vol (l)	.83	1.2	1.5	1.15	1.8	1.0	1.45	1.2	---
Mixed Liquor									
COD (mg/l)	45	50	45	25	30	30	50	60	40
MLSS (mg/l)	1980	2510	2340	2730	3020	2760	2840	2570	2560
MLVSS (mg/l)	---	---	990	1540	2000	1690	1940	1530	1550
MLFSS (mg/l)	---	---	1350	1190	1020	1070	900	1040	1010
pH	8.8	8.85	8.5	9.4	9.0	8.9	9.019	8.860	8.9
Temp °C	19	19	19	18	18	18	18	16	18
Cu total (ppb)	---	---	---	---	54.4	56.2	90.4	45.2	---
Cu sol (ppb)	---	---	---	---	11.82	7.93	7.77	26.1	---
Cu solids (ppb)	---	---	---	---	---	---	165.8	2.4	---
O ₂ uptake	---	---	---	---	---	---	---	---	---

TABLE A-2
REACTOR #2

Date	1/18	1/20	1/23	1/25	1/27	1/28	1/29	1/30	1/31
Influent									
COD (mg/l)	365	400	---	330	490	530	200	430	350
Cu (ppb)	---	---	---	---	210	175	190	194	---
Effluent									
V (l)	12	10	*	10	11	10	10	9	11
TSS (mg/l)	120	140	*	400	220	210	170	310	110
VSS (mg/l)	---	---	*	280	220	100	140	270	60
FSS (mg/l)	---	---	*	120	0	110	30	40	50
Wasted Vol (l)	1.3	1.3	1.8	.25	1.0	1.1	1.3	.74	---
Mixed Liquor									
COD (mg/l)	25	45	*	30	30	20	25	65	40
MLSS (mg/l)	2720	2370	2800	2050	2310	2450	2500	2110	1990
MLVSS (mg/l)	---	---	1510	1290	1580	1600	1660	1300	1250
MLFSS (mg/l)	---	---	1290	760	730	850	840	810	740
pH	8.8	8.7	8.3	8.7	8.8	8.8	8.8	8.8	8.8
Temp °C	19	19	19	18	18	18	18	16	18
Cu total (ppb)	---	---	---	---	751.1	740.3	740.2	579.4	---
Cu sol (ppb)	---	---	---	---	66.5	83.8	96.1	131.4	---
Cu solids (ppb)	---	---	---	---	---	629.9	671.2	571.4	---
O ₂ uptake	---	---	---	---	---	---	---	---	---

*Batch fed due to pump failure.

TABLE A-4

REACTOR #4

Date	1/18	1/20	1/23	1/25	1/27	1/28	1/29	1/30	1/31
Influent									
COD (mg/l)	365	400	---	340	390	360	140	340	430
Cu (ppb)	---	---	---	---	270.1	135.3	56.4	157.8	---
Effluent									
V (l)	11.5	11	*	11	12	10	10.5	10	13.5
TSS (mg/l)	90	140	*	290	30	150	200	160	150
VSS (mg/l)	---	---	*	280	30	70	170	160	130
FSS (mg/l)	---	---	*	10	0	80	30	0	20
Wasted Vol (l)	1.3	1.1	1.8	.2	1.6	.9	.4	.5	---
Mixed Liquor									
COD (mg/l)	50	30	*	20	85	85	85	75	120
MLSS (mg/l)	1640	1720	1960	1580	1420	1370	1230	1030	1390
MLVSS (mg/l)	---	---	1200	920	1060	970	940	740	790
MLFSS (mg/l)	---	---	760	640	360	400	290	290	600
pH	8.1	8.3	7.9	8.1	8.3	8.4	8.5	8.5	8.4
Temp °C	19	19	19	18	18	18	18	16	18
Cu total (ppb)	---	---	---	---	394	382	363	374	---
Cu sol (ppb)	---	---	---	---	118.6	79.0	106.0	105.8	---
Cu solids (ppb)	---	---	---	---	---	225.2	214	---	---
O ₂ uptake	---	---	---	---	---	---	---	---	---

*Batch fed due to pump failure.

TABLE A-5
REACTOR #5

[illegible]

TABLE A-6

REACTOR #6

Date	1/18	1/20	1/23	1/25	1/27	1/28	1/29	1/30	1/31
Influent									
COD (mg/l)	365	400	*	320	780	410	480	450	410
Cu (ppb)	---	---	---	---	158.2	211.3	135.0	163.2	---
Effluent									
V (l)	9	9	*	12	12	9	9.5	11	13
TSS (mg/l)	350	610	*	590	0	200	270	120	160
VSS (mg/l)	---	---	*	490	0	140	260	120	160
FSS (mg/l)	---	---	*	100	0	60	10	0	0
Wasted Vol (l)	.1	0	1.8	0	1.8	.9	.4	.9	---
Mixed Liquor									
COD (mg/l)	35	30	*	35	45	40	45	95	120
MLSS (mg/l)	1490	1150	1980	1360	1710	1590	1430	1210	930
MLVSS (mg/l)	---	---	1360	1070	1390	1110	1030	980	830
MLFSS (mg/l)	---	---	620	290	320	480	400	230	100
pH	6.45	6.7	6.6	6.6	6.5	6.5	6.6	6.6	6.6
Temp °C	19	19	19	18	18	18	18	16	18
Cu total (ppb)	---	---	---	---	448.9	493.5	504.7	157.8	---
Cu sol (ppb)	---	---	---	---	65.2	79.9	84.8	112.4	---
Cu solids (ppb)	---	---	---	---	---	382	348	315	---
O ₂ uptake	---	---	---	---	---	---	---	---	---

*Batch fed due to pump failure.

TABLE A-7

REACTOR #7

Date	1/18	1/20	1/23	1/25	1/27	1/28	1/29	1/30	1/31
Influent									
COD (mg/l)	360	400	*	380	350	380	480	450	330
Cu (ppb)	---	---	---	---	191.5	202.8	202.8	180.3	---
Effluent									
V (l)	14.0	8.0	*	11.5	10.0	11.0	12.0	10.5	13.0
TSS (mg/l)	120	170	*	150	-	190	240	150	220
VSS (mg/l)	---	---	*	150	0	130	240	150	200
FSS (mg/l)	---	---	*	0	0	60	0	0	20
Wasted Vol (l)	1.1	1.2	1.8	.9	1.8	.4	0	.5	---
Mixed Liquor									
COD (mg/l)	65	160	*	50	85	125	115	120	90
MLSS (mg/l)	1940	1690	2240	1510	1300	1150	1060	1010	920
MLVSS (mg/l)	---	---	1350	1180	1110	820	820	840	750
MLFSS (mg/l)	---	---	890	300	190	330	240	170	170
pH	7.2	7.2	7.1	7.1	7.3	7.1	7.3	7.4	---
Temp °C	19	19	18	18	18	18	18	16	18
Cu total (ppb)	---	---	---	---	526.9	504.7	426.6	448.9	---
Cu sol (ppb)	---	---	---	---	127.2	133.1	111.4	104.2	---
Cu solids (ppb)	---	---	---	---	---	337.3	337.3	292.5	---
O ₂ uptake	---	---	---	---	---	---	---	---	---

*Batch fed due to pump failure.

TABLE A-8
REACTOR #8

Date	1/18	1/20	1/23	1/25	1/27	1/28	1/29	1/30	1/31
Influent									
COD (mg/l)	400	400	*	380	360	380	550	360	410
Cu (ppb)	---	---	---	---	191.5	157.8	225.2	157.8	---
Effluent									
V (l)	12	10	*	12	12	12	13	11	10
TSS (mg/l)	90	60	*	130	130	170	200	120	190
VSS (mg/l)	---	---	*	130	40	130	200	120	190
FSS (mg/l)	---	---	*	-	90	40	0	0	0
Wasted Vol (l)	1.3	1.5	1.8	.8	.8	0	0	.6	---
Mixed Liquor									
COD (mg/l)	55	45	*	85	120	150	125	120	100
MLSS (mg/l)	1670	1720	2110	1280	1020	980	1010	9120	940
MLVSS (mg/l)	---	---	1260	1070	920	650	730	780	660
MLFSS (mg/l)	---	---	850	210	100	330	280	130	280
pH	7.1	7.13	7.2	7.0	7.2	7.2	7.2	7.2	7.1
Temp °C	19	19	19	18	18	18	18	16	18
Cu total (ppb)	---	---	---	---	471.2	421.1	382.0	372.0	---
Cu sol (ppb)	---	---	---	---	111.2	129.0	106.8	113.0	---
Cu solids (ppb)	---	---	---	---	---	270.1	258.4	236.4	---
O ₂ uptake									

*Batch fed due to pump failure.

VITA

George Calvin Welborn, Jr. was born in Chattanooga, Tennessee in January 3, 1952. After graduating from the McCallie School in Chattanooga in May 1970, he attended Vanderbilt University from September 1970 to May 1974, studying Electrical Engineering. After receiving a Bachelor of Engineering degree from Vanderbilt, he was employed by Hensley-Schmidt, Inc., a consulting engineering firm, in Chattanooga, Tennessee. In January 1977, he was transferred to the Knoxville office of Hensley-Schmidt in order to work part time while participating in the Environmental Engineering graduate program at The University of Tennessee, Knoxville, receiving a Master of Science degree in December 1979.