Lawrence Berkeley National Laboratory
Recent Work

Title
OZONE OXIDATION OF ORGANIC SEQUESTERING AGENTS IN WATER PRIOR TO THE ANALYSIS OF TRACE METALS BY ANODIC STRIPPING VOLTAMMETRY (ASV)

Permalink
https://escholarship.org/uc/item/5x80m2bj

Author
Clem, Ray G.

Publication Date
1977

00 00 ..0 00 44 87 00 02 07 11 93

UC-4
UC-11

LBL-6315 Rev.
Preprint

Energy and Environment Division

Lawrence Berkeley Laboratory University of California/Berkeley
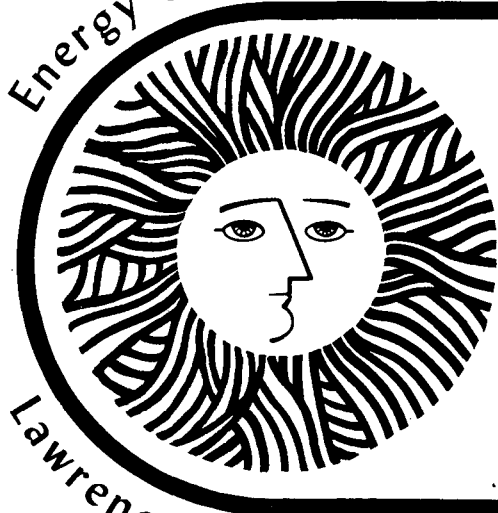
Ozone Oxidation Of Organic Sequestering Agents In Water Prior To The Analysis Of Trace Metals By Anodic Stripping Voltammetry (ASV)

Ray G. Clem and Alfred T. Hodgson

January 1977

LBL-6315 Rev.
c.)

DISCLAIMER

OZONE OXIDATION OF ORGANIC SEQUESTERING AGENTS IN WATER PRIOR
TO THE ANALYSIS OF TRACE METALS BY ANODIC STRIPPING VOLTAMMETRY (ASV)

Ray G. Clem* and Alfred T. Hodgson

Energy and Environment Division
Lawrence Berkeley Laboratory
University of California
Berkeley, California 94720

Brief

The temporal release of trace metals, Pb and Cd, from the sequestering

agents ETDA, APDC, humic acid, and tannic acid after treatment with

ozone is discussed. The new method is applied to bay water and sewage

effluent samples.

* Author to whom correspondence should be addressed.

OZONE OXIDATION OF ORGANIC SEQUESTERING AGENTS IN WATER PRIOR
TO THE ANALYSIS OF TRACE METALS BY ANODIC STRIPPING VOLTAMMETRY (ASV)*

Ray G. Clem and Alfred T. Hodgson

Energy and Environment Division
Lawrence Berkeley Laboratory
University of California
Berkeley, California 94720

ABSTRACT

Ozone is investigated as a general oxidant for trace organic
sequestering agents in water. The course of destruction of sequester-
ing agents is monitored by observing the liberation of free-ion Pb
and Cd at the part-per-billion level. The model compounds used for
developing the presented method are: EDTA, APDC, tannic, and humic
acids. The method is applied to the determination of Pb and Cd in
sewage effluent and in San Francisco Bay water. A method by which
humic acid having an initial ash content of >11% is reduced to <0.1%
is discussed.

—

—

-1-

Acidification to a pH between 1 and 2 is an essential first step in water sampling; it arrests biological growth; it effects the release of trace metals from naturally occurring, weakly acidic, organic sequestering agents such as humic and fulvic acids; and, it reduces metals loss through adsorption on the container wall. Unfortunately, in the presence of acid, the long-term stability of graphite electrodes used for anodic stripping voltammetry (ASV) is poor (1). Neutralization of the added acid prior to the analysis would, of course, result in the recomplexation of the released metals by the humic and fulvic acids. This intolerance of acid extends to other instrumental and chemical techniques as well. For the determination of trace metals with flameless atomic adsorption (AA), it is necessary to extract them if much salt is present or if the original metals concentrations are below the detection limits of the AA method. Neither of the extractants commonly used--dithizone or ammonium pyrrolidine dithiocarbamate (APDC)-- is effective in extracting all the metals of interest at pH 1 to 2. The recovery is generally the highest between pH 4 and 7. Similarly, the optimum pH range for chelating resins is on the interval between 5 and 9 (2). Chelating resins are intolerant of acid also. The lack of agreement among various techniques used in a recent inter-laboratory calibration study illustrates the great need for some means to deal with trace organics in water samples (3).

From the foregoing, what is clearly needed is a method for destroying naturally occurring sequestering agents rather than trying to accommodate them in the analytical procedure. Previous workers have recognized the problem and used such oxidizing agents as persulfate (4)

and UV photolyzed hydrogen peroxide (5). Both methods are efficacious, but both have drawbacks. Persulfate in acidic solutions in the presence of mercuric-ion rapidly destroys ASV electrodes (6); the last traces of it are difficult to remove; and, the reagent harbors trace metals. Similar problems exist for the peroxide method. These problems with solid and liquid oxidants have led us to investigate the use of the gaseous reagent, ozone.

In the heirarchy of oxidants in acid solutions, ozone stands third (E^O = 2,07V) behind only fluorine (E^O = 3.06V) and atomic oxygen (E^O = 2.42V). Despite this, it is seldom used in analytical procedures. As an oxidant for organic matter prior to trace metals analysis, ozone appears ideal. It reacts rapidly at room-temperature with organic matter at levels found in natural water samples. It inactivates sequestering agents over the pH range of 2 to 10; in this, it is unique among other reagents tried. The high purity oxygen gas used in its generation is inexpensive, readily available, and does not harbor trace metal impurities. The decomposition product of ozone in aqueous solutions is oxygen. For many applications, this gas is innoxious; however, if it is necessary to remove it, along with dissolved ozone, subsequent to electrochemical measurements, it can be displaced rapidly with Ar, N_2, or CO_2.

The requisite equipment for ozone production is inexpensive and simple to assemble. All that is needed is a high-voltage transformer and a corona discharge tube which can be fabricated easily from Pyrex tubing.

To illustrate the new method, we have selected ethylene diaminetetra-acetic acid (EDTA), APDC, tannic acid, and humic acid, all of which are widely recognized as powerful complexing agents for the metals, lead and cadmium, used in this study. The method is applied to bay water and sewage effluent samples.

Also, during the course of this work, we developed a method for reducing the ash content of technical-grade humic acid to < 0.1%. This procedure should be of general interest to anyone using instrumental techniques to develop methods for trace metals in water.

Experimental

The instrumentation used consisted of an MPI Electroanalyzer Model 1502B operated in the ASV Mode in conjunction with an ElectRoCell-ASV Model A-2000. The Lucite MPI ASV cell was modified to accept a quartz cell, fabricated in this laboratory's glass shop, having the same internal dimensions as the plastic one. The preparation of the electrode used was described previously (1). Data were recorded with a Mosely Model 7001A, X-Y recorder. Spectrometric scans were made with a Beckman Model DB recording, UV-Visible spectrophotometer. Fixed wavelength measurements were made with a Beckman Model DU. Both instruments were equipped with deuterium UV sources. A set of 4, matched, 1-cm quartz cells were used, and when needed, 0.9-cm quartz cell spacers were used to reduce the pathlength to 0.1-cm. A Beckman Model J-21B centrifuge was used in the purification of humic acid.

All water used was purified by first passing house-distilled water through a mixed-bed ion-exchange column before feeding it into a

double boiler quartz still. The quartz distilled water was collected and stored in a 10-1 linear polyethylene bottle equipped with a spigot of the same material. When first received over two years ago, this bottle was alternately and repeatedly leached with concentrated HCl and washed with quartz water to clean it and subsequently has never been used for anything other than storage of quartz distilled water. With the exception of humic acid and Eastman yellow label APDC, all chemicals employed were of reagent grade. The APDC was purified by dissolving it in 95% ethanol then precipitating it by adding a volume of diethyl ether equal to the volume of ethanol. The crystals were dried under vacuum.

Quartz-ware was cleaned by soaking it in concentrated HNO_3 at 55°C overnight when first received and by soaking for a minimum of 30 minutes when in routine service. The FEP-Teflon bottles used for sample collection and reagent storage were initially cleaned as recommended by Patterson (7) and as described above when placed in service. If the quartz-ware became contaminated with an oily film as described below, it was boiled in red-fuming HNO_3 then washed with water and soaked in concentrated nitric acid as described above.

Purification of Humic Acid

Humic Acid, technical grade, was purchased from Aldrich Chemical. This material proved to contain 11.6% ash. To separate the soluble humic acid from the insoluble matter (portions of plants, sand, dirt, etc.), place 50g of humic acid in a 1-1 beaker with 4g of NaOH in 800 ml of water, cover the beaker with Parafilm, then stir magnetically overnight. The next day, transfer the solution to

four 250 ml polyethylene bottles, then centrifuge at 7500 rpm for
55 minutes. The temperature of the sample is maintained at 20°C.
Decant the soluble humic acid into clean bottles and discard the
insoluble material. Add 20 ml of 12 $\underline{M}$ HCl to each of the bottles
to precipitate the humic acid. Centrifuge again as described above
to remove most of the sodium. Discard the decantate. Resolublize
the humic acid with 10 ml of concentrated ammonium hydroxide, stir,
then bring the volume up to 200 ml. Precipitate the humic acid again
with 20 ml of 12 $\underline{M}$ HCl. Centrifuge, resolubilize, precipitate, then
allow the humic acid to stand in the presence of the HCl overnight.
Centrifuge again and discard the decantate. Stir the humic acid with
200 ml of water. Centrifuge. Repeat this washing step, if necessary,
until the pH of the wash water is between 2 and 3. This "HCl-purified"
humic acid has an ash-content of 0.9%. The yield is ~80%.

The ash content of the HCl-purified humic acid can be reduced
further by treatment with a mixture of HCl and HF. Redissolve the
above product with 10 ml of concentrated ammonium hydroxide, add some
water, 50 ml will do, precipitate the humic acid with a mixture of
20 ml of 12 $\underline{M}$-HCl and 10 ml of 42 $\underline{M}$-HF. Add 120 ml water. Let the
humic acid stand in contact with the HCl-HF acid for 2 days at room
temperature. Centrifuge for 10 min; decant the supernate; resolublize
with ammonia; precipitate the humic acid with 12 $\underline{H}$ HCl. Stir the
precipitated humic acid with 0.01 $\underline{M}$ HCl, centrifuge 10 minutes; repeat
this operation; then, finally wash it with water as described above.
The ash content of this "HF-purified" humic acid is 0.09%. The yield
is virtually 100%.

A portion of the product was dissolved by adding concentrated ammonium hydroxide drop-wise to the stirred solution to a pH of 4.7, then made to volume in a 100-ml flask. This was our stock solution. It contained 42.0 mg/ml as determined by weight from freeze-dried aliquots. Working solutions of this stock solution were made by dilution.

The humic acid stock and working solutions at pH 4.7 and at room temperature appear to be a good nutrient for microorganisms. To preserve these solutions, make them 0.1% v/v in chloroform. When not in use, store them in a refrigerator.

Ozone Generator and Ozonolysis Tube

The ozone generator or corona discharge tube was constructed completely of Pyrex glass tubing according to a basic published design (9). The inner tube was filled with mercury and contact to it was made with a length of stainless-steel wire. The outer tube was wrapped with 5 mil copper foil; the foil was secured to the tube with twisted copper wires, one of which served as a contact wire. Both the oxygen inlet and ozone outlet tubes were accessible at the top of the corona discharge tube. The discharge tube was immersed in a tube filled with ethylene glycol, and this tube, in turn, was immersed in a water-ice mixture contained in a large Dewar. The original method of cooling the discharge tube with tap water (9) was an obvious violation of present-day safety standards. The tube was powered with a 12 kV, 30 mA, 60 HZ transformer obtained from Jefferson Electric Co., Bellwood, Illinois. The transformer and assembled discharge tube were mounted in a ventilated Lucite plastic box. The box access

door was wired with a microswitch to turn the power to the transformer off if it were inadvertantly opened. Ozonolysis times of up to 1-hr could be preset with a CRA-LAB interval timer interposed between the line and the transformer.

In operation, oxygen at a flow rate of 3.7 l/min was introduced into the discharge tube through Tygon tubing. The ozone produced was lead into the gas dispersion tube with Teflon tubing. The 55 mm o.d. dispersion tube was 72 cm long and was constructed entirely of Supracil quartz. An 8-mm o.d. side-arm was used to lead the ozone gas into the bottom of the tube. The coarse quartz frit was fused to the dispersion tube and positioned 1 cm above the bottom. It was possible to treat 300 to 500 ml of sample in this tube.

All ozonolysis procedures must be conducted in a good fume hood since it, like other gaseous oxidents, fluorine, chlorine, and bromine, is highly irritating in the concentration generated. Furthermore, it must not be generated in the presence of gaseous reducing agents such as hydrogen, ammonia, or hydrocarbons.

Analytical Procedure

Place the assembled corona discharge tube into the Dewar. Pack crushed ice around the tube; add some water to improve the thermal contact. Let the tube stand for 30 minutes, then connect it to the ozonolysis tube and adjust the oxygen flow-rate to 3.7 l/min. Place 300 to 500 ml of sample acidified to pH 2 into the tube, then start the ozonolysis by setting the timer-relay. Stop the ozonolysis after 1 hour, but continue to pass oxygen through the sample for 5 minutes to sparge the solution of dissolved ozone. Stop the flow of oxygen,

then aliquot four 50-ml aliquots of sample into four clean polyethylene or FEP-Teflon containers. Add 300 μl of a mercuric nitrate solution containing 300 μg Hg^{2+}/ml to each container. Add increasingly larger spikes of Pb and Cd standards to the containers so that the Pb and Cd peaks are essentially double the initial peaks in the standard additions plot. Add 200 μl of saturated sodium acetate to each sample to bring the pH to 4.7. Mix each thoroughly. Buff the electrode before each analysis with metallographic Al_2O_3 (Linde Fine Abrasive Type A-5175) sprinkled over filter paper which has been attached to a microscope slide with rubber cement. Run each solution in the standard additions series. Plating times required will be typically 5 to 10 minutes. Make a plot of peak height vs the Pb and Cd added, then extrapolate the data to zero to quantify the Pb and Cd in the sample. Retain this curve as a calibration plot for other samples from the same source. Remake the calibration plot as often as is required to keep the results within the desired statistical limits.

Results and Discussion

Trace metals analysis of natural waters presents a challenge because the metals are distributed in many ways within a single sample: they are present in the free-ion state, are complexed by low molecular weight organic substances, are complexed at the surface functional groups of organic colloids such as humic acids, and even reside inside such colloids as will be shown below. All the foregoing substances are weakly acidic so their effects can be suppressed simply by acidifying the water sample, but this leads to problems with the stability of ASV electrodes (1).

Of the two previously used methods to destroy organic matter in water samples (4,5), persulfate oxidation and UV photolyzed hydrogen peroxide, only the latter seems to be a reasonable approach. Persulfate is difficult to purify of trace metals and, after oxidizing the sample with it, the last traces of oxidant are difficult to destroy. This reagent is most effective in acid solutions and, in the presence of mercuric ion, readily oxidizes graphite ASV electrodes (6). In contrast, UV photolysis of hydrogen peroxide with light from a high-pressure mercury lamp appears to be a workable approach (5) although it inherently has several disadvantages. The use of peroxide would present a source of contamination (our water samples were typically $\leq$1-ppb in level); sample evaporation from the heat produced by the UV lamp would be a problem; the 4 to 5 hour irradiation times required are rather lengthy; and, the presence of traces of peroxide could interfere in subsequent ASV determinations. However, the method can be used for neutral solutions.

A modification of the UV method--mercury sensitized photolysis using a low-pressure mercury vapor source (10)--was tried briefly but with limited success. It suffered from all the drawbacks described above for the high-pressure UV method. The low-pressure UV source did produce, however, a great deal of ozone. Because the lamp employed was new, the quartz envelope had not been damaged by short wavelength radiation. The use of ozone along was intriguing. Ozone has been used for destruction of organics in drinking water since the last century and, in recent years, for the treatment of sewage effuents (11), but very little use of it has been made for analytical purposes.

Judging from a summary of the field of organic/ozone chemistry (11), ozone appears to react with virtually all organic compounds. The functional groups capable of metal sequestering activity, and therefore of interest in the context of this report, are carboxyl, phenolic, sulfide, and amine groups.

Carboxyl groups seem to be an end product of the ozonolysis of many compounds and further attack on this group appears to proceed very slowly. Phenols are more reactive toward ozone than are most aromatics (12). Phenol, for example, is oxidized to CO_2, formic acid, glyoxal, and oxalic acid (13). Organosulfides are oxidized to sulfoxides and with further treatment to sulfones (14). Ozonolysis in neutral solutions of primary, secondary, and tertiary amines yields nitro- and nitroso-compounds in addition to various decomposition products (15-18). In acidic solutions, however, amines are unreactive toward ozone (19), resulting from the unavailability of the nitrogen electron pair.

In the initial experiments with ozone, the UV lamp was used as an ozone generator. The lamp output in the Schumann region diminished with time as a result of progressive short wavelength radiation damage to the quartz. For this reason and because there was no satisfactory way to dissipate heat from the lamp, the lamp was soon abandoned for a corona discharge tube which was capable of a higher and more constant output of ozone.

In a corona discharge tube, ozone is created by the reaction of atomic oxygen, produced in the high-voltage ac field applied across the discharge gap of the ozone generator, with neutral molecules of

diatomic oxygen. Since the ac field can destroy as well as produce

ozone, the yield is inherently low. This is fortunate in one respect;

ozone concentrations greater than 30% can detonate spontaneously (11).

Several factors combine to determine the yield, Y, of ozone per

unit of electrode surface according to the following relationships

developed for large industrial ozone generators (11);

$$Y \propto \frac{f\varepsilon V^2}{d} \tag{1}$$

where f is the frequency of the high voltage ac applied, ε is the

dielectric constant of the dielectric, V is the peak voltage applied

across the discharge gap, and d is the thickness of the dielectric.

The optimum applied voltage is proportional to the product of the

oxygen gas pressure and the width of the discharge gap. As production

of ozone is inversely related to the temperature in the region of

the discharge gap, efficient cooling of the discharge tube is essential.

The foregoing information indicates how a generator having a

higher ozone output than that employed here could be built, but the

consequences of a gas phase explosion or rupture of the dielectric

in an electric field of more than 10 kV is apt to be quite severe.

The ozone concentration at the output of our generator at a flow

rate of 3.7 l/min is 10 $\pm$ 1% as determined with the KI method (20).

This is significantly higher than that reported by previous workers (9)

and is likely the result of our use of an ice-bath as well as our

use of dry oxygen.

Water in the oxygen feed-gas is highly detrimental to the

production of ozone. In one experiment, oxygen saturated with water

vapor was fed into the discharge tube; the ozone content was reduced
to <3% and, upon switching back to dry oxygen, it did not increase
to the 10% level until after dry oxygen had passed through the tube
for more than a half hour.

The method used to mix the ozone with the sample is effective
but not as efficient as it could be. A coarse porosity frit was used.
If a medium or fine porosity frit material had been available and
used at the time the tube was fabricated, a much lower flow-rate of
oxygen for the same efficiency reported would have been possible.
Judging from the apparent increase in volume of the sample during
ozonolysis, at an oxygen flow-rate of 3.7 l/min, there is ~100 ml
of gas continually in contact with the sample solution. The amount
varies with the surface tension of the sample solution. The total
volume of the tube is 3 to 5 times greater than that of the sample
ozonized. This free volume is necessary since some samples, such
as humic acid and sewage effluents, have a great tendency to foam.
In fact, with those two samples, it was necessary to reduce the gas
flow rate in the initial stages of the ozonolysis and to repeatedly
wash down the walls of the tube with sample solution sucked up into
a pipet. In contrast, sea, bay, or drinking water samples presented
no problems.

The measured rate of evaporation of a 300 ml sample ozonized
at the recommended oxygen flow rate of 3.7 l/min is approximately
1%/hour. This rate is, of course, dependent upon the initial sample
volume and the average temperature of the sample during the ozonolysis
period. As suggested, if the discharge tube is chilled by immersion

in a quiescent ice-bath for 30 minutes before it is turned on, oxygen

flowing through it will have an exit temperature of about 17°C. This

temperature will not change after the oxidation is begun. Assuming

that under these conditions the vapor pressure of water in oxygen

is 14.5 mm and that the oxidizing gas becomes instantaneously saturated

with water vapor upon contracting the sample, then 3.4 g of water

per hour will be evaporated. This then sets a lower limit of about

300 ml on the sample size if it is not desirable to have a volume

loss of more than 1% during the ozonolysis. The temperature of the

solution at the end of a 1-hour ozonolysis time will be close to

that of the ozone. This, of course, depends upon its initial temperature.

The solubility of ozone in water is about ten times greater than oxygen

and is inversely related to the temperature. However, owing to the

small partial pressure of ozone in the oxygen stream, the ozone concentration

is usually on the order of only a few parts per million in the aqueous

phase. The temperature at which the sample solution is ozonized,

therefore, is a trade-off among increasing sample oxidation rates,

solution evaporation rates, and decreasing solubility and stability

of ozone in the sample at higher temperatures. To avoid these complications,

all the data presented were obtained at an initial ambient room-temperature

of ~20°C.

To demonstrate the efficiency of the method, we selected EDTA,

APDC, tannic and humic acids. All four compounds have the ability

to completely sequester lead and cadmium ions in neutral solutions.

Also, the latter two are of natural origin and thus are likely to

be found in natural waters. The first three compounds are relatively

simple, well characterized, small organic molecules which have all the
major functional groups reported to exist in the humic acid (21,22)
as discussed above. Humic acids, in contrast, range in molecular
weight from 16,000 to 300,000 (21). Ozonolysis of humic acid results
in a mixture of simple weak acids (23,24).

The EDTA was of special interest because, in unpublished studies,
one of us (RGC) found that it is extremely resistant to oxidation.
EDTA is very slowly and incompletely attacked upon treatment with ceric
perchlorate in 1 $\underline{M}$ perchloric acid at 80°C or with electrogenerated
Ag(II) in 1 to 5 $\underline{M}$ nitric acid at 0 to 5°C. The levels of EDTA, APDC,
humic and tannic acids ozonized were 3, 25, 25 and 100 ppm, respectively.
By either calculation or through amperometric titrations with lead it
was determined that these levels correspond to ligand concentrations of
10^{-5}, 1.5 x 10^{-4}, 10^{-6}, and 5 x 10^{-6} $\underline{M}$, respectively. These concentrations
approximated the organic content of bay or seawater. The measured
total dissolved organic carbon content in the San Francisco and connecting
bays was $\leq$9 ppm and that for open Pacific Ocean water was near 3 ppm.

All the simulated samples used were made 0.5 $\underline{M}$ in sodium or
potassium chloride to approximate seawater in chlorinity and some
were made 0.1 $\underline{M}$ in NaHCO$_3$ to approximate the pH of seawater.

Many different working solutions were made during the course
of this work. The lead and cadmium solution concentrations were held
at 20 $\pm$ 5 ppb each. These concentration levels were high enough that
the work did not have to be performed in clean-air enclosures thus
short plating periods could be employed. The FEP-Teflon containers
used have some exchange capacity (although very much lower than for

Pyrex), so the solutions were allowed to equilibrate with their containers

for several days before they were standardized and used. This loss

is much more of a problem for neutral to basic solutions than it is

for those stored at pH 2.

A pH of 4.7 (acetate buffer) was used for all results presented.

The reason for this is shown in Figure 1. At high pH values, lead

and cadmium peaks exhibit considerable overlap. This results from

complexations of the metals with hydroxide and carbonate ions which

cause the stripping peaks for both metals and especially lead to shift

to more negative potentials (25). At pH 4.7 the peaks are well separated

and the sensitivity is much improved.

It was found near the outset that working solutions stored at

neutral to basic pH values must be acidified to pH 2 then neutralized

to pH 4.7 with sodium acetate before being analyzed. Acidification

directly with acetic acid to pH 4.7 results in very poor reproducibility

and low values for lead and cadmium. Evidentially, the complexes

formed with carbonate and hydroxide are not completely eliminated

on mild acidification with acetic acid, and since the diffusion

coefficients for these complexes are considerably lower than for those

for the lead and cadmium solutions used for standard additions--usually

8 ppm in each metal and stored at pH 2 to 3--low values are obtained.

The variability of the results seems to depend upon whether the acetic

acid is added directly to the bulk solution or permitted to trickle

into the solution slowly down the side of the cell. Bicarbonate

solutions analyzed immediately after being prepared and acidified

to pH 4.7 with acetic acid do not exhibit this anomality. This appears

to be the first report of such behavior. Others should be aware of
this problem since it is possible to obtain ASV signals for Pb and
Cd from a neutral solution after ozonolysis.

After ozonolysis, the samples did not harbor residual oxidizing
agents. The ozone appeared to be completely swept out if oxygen was
continued to be passed through the solution for a few minutes after
the discharge tube was turned off. This was determined with moistened
starch-iodide test papers held in the exhaust gas from the ozonolysis
tube. Any organic per-acids or peroxides formed during ozonolysis
seemed to be completely eliminated upon acidification to pH 2 followed
with neutralization to pH 4.7 as suggested in the recommended procedure.
Samples treated in this way gave a negative test for oxidant with
a starch-iodide solution. The foregoing was true for all samples
tested below.

Given the oxidation potential of ozone (E^O = 2.07V - 0.059 pH),
it should oxidize chloride to chlorine (E^O = 1.4V) at pH 2, chloride
to hypochlorous acid at pH $\geq$3.3 but $\leq$7.3 (E^O = 1.5V - 0.059/2 pH),
and chloride to hyprochlorite ion at pH $\geq$7.3 (E^O = 1.7V - 0.059 pH).
We have been unable to detect any of the above predicted oxidants.
This is probably the result of unfavorable kinetics. It has been
reported that bromide is only partially oxidized to bromate in basic
solutions and iodide is oxidized to iodate, periodate, and some iodide
is lost from solutions as iodine pentoxide in carbonate solutions (9).
At pH 2, however, only elemental bromine and iodine should be liberated
and these would be swept from solution with the ozone. No problems
have been encountered from either of these two halides in bay or ocean

water samples because their natural abundances are quite low. The

bromine in seawater is 65 ppm: iodine is only 0.05 ppm.

   For three of the four model substances discussed below, data

are given for ozonolysis in both acidic and basic solutions. However,

in view of the greater time required to oxidize the sequestering

agents in basic solutions, adsorption effects noted for some of the

substances studied, and the necessity of acidifying the basic sample

to pH 2 before making the ASV scans at pH 4.7 as noted above, it is

recommended that all ozonolyses be conducted at pH 2. For all the

model compounds studied, the times given are for exhaustive ozonolysis.

   ETDA was the first chelating agent studied. Figure 2A shows

the results obtained upon ozonizing a solution having an initial

pH of 8.0 in bicarbonate. At the end of the experiment, the pH had

increased to 9.7 as a result of loss of CO_2. Acidification of the

sample remaining in the ozonolysis tube to pH 2 followed immediately

with neutralization to pH 4.7, did not result in complete recovery

for lead which would indicate that considerable adsorption of lead

on the walls of the quartz tube had occurred. Complete recovery was

not attained until the sample had sat at pH 2 overnight.

   Data taken in the early stages of the oxidation are meaningless.

The initial EDTA molar concentration is one to two orders of magnitude

greater than that of the metals. Even after the excess EDTA is destroyed

to the point that lead and cadmium peaks appear, the standard additions

plot is not linear. This perhaps is the result of the partially oxidized

EDTA having some residual complexing ability. Other problems arise

in the early stages of oxidation. Either an ozonide of EDTA is formed

which reacts with the electrode or an oxidation product is formed

which is strongly absorbed at the electrode surface. In either case,

the electrode sensitivity is greatly reduced in succeeding scans.

This appears to be a general problem encountered during method development

for EDTA, APDC, humic acid, and the sewage effluent samples discussed

below. No problems are encountered with tannic acid. If the ASV analysis

had been made only after the complete ozonolysis of the sequestering

agents, no loss of electrode sensitivity would be detected. This

may not be practical from a time standpoint, however.

The only effective way to restore the electrode sensitivity is

to buff it to expose new surface. Buffing with metallographic

Al_2O_3 as described above does not appreciably change the electrode

surface area, hence sensitivity, even after the electrode has been

polished dozens of times. This refurbishing procedure must be used

only for a hard-surface electrode such as the one employed here or

for glassy-carbon.

In comparing Figure 2A with Figure 2B, the most striking contrast

is the time required for the release of the metals. This of course

reflects the difference in the oxidation potential of ozone at pH 2

of 2.0V and at pH 9.7 of 1.5V. The ability of ozone to function at

high pH and to extract the hydrogen ions required for the oxidation

to proceed from a basic solution is quite remarkable.

Ultraviolet scans were made between 400 and 200 nm during the

course of ozonolysis of the neutral, 10^{-5} $\underline{M}$ ETDA solution in an effort

to detect nitrogen oxides, produced by oxidation of the amine

nitrogens (15), which would likely absorb at wavelengths longer than

that of EDTA (10). However, no evidence for this was found. EDTA
itself had a rather simple absorbance spectrum which was characteristic
of simple organic compounds. The first preceptible absorbance occurred
between 230 and 220 nm followed by a rapid exponential increase to a
value >2A at 220 nm. The shape of the curve remained unchanged during
the 4 hours oxidation time but the wavelength at which the absorbance
exceeded 1.0A gradually moved to shorter wavelengths during the oxidation.
This was probably the result of a slow loss of organic carbon (11).

APDC at pH 2 was the next complexing agent studied and was
selected to illustrate the action of ozone on the sulfide group.
Sulfide was rapidly and smoothly oxidized and complete recovery of
both lead and cadmium was attained within 45 minutes. See Figure 3.

Tannic acid was the best behaved model compound studied. In
bicarbonate solution, lead was completely recovered and 88% of the
cadmium was found after 1 hour. After 2 hours the cadmium value was
98%. The tannic acid solution turned green upon being exposed to oxygen
but became completely decolorized in less than 15 minutes of ozonolysis.
However, some of the decomposition products are good complexing agents
as supported by the above data. Data taken for tannic acid at pH 2
are plotted in Figure 4A. Again, the rate of oxidation was much greater
under acidic conditions.

Figure 4B shows absorbance vs wavelength scans for tannic acid
in bicarbonate solution before and after ozonolysis. The peaks are
likely due to the benzene rings in the gallic acid portion of the
tannic acid structure. A scan of gallic acid at the 25 ppm level
is given for comparison to tannic acid before ozonolysis. The fact

that these peaks disappear after 15 minutes ozonolysis is probably the result of ring rupture on oxidation (12). Successive scans taken for tannic acid after the first 15 minutes ozonolysis are virtually superimposable.

As received, the humic acid was saturated with polyvalent elements and had virtually no exchange capacity for the metal-ions employed here, so, the first step was to devise a means for purification. Our initial efforts were guided by the work of Malcolm (26). Dissolution of the humic acid with sodium hydroxide followed by centrifugation effected a gross separation between the soluble humic acid and the insoluble mineral and vegetable materials. The soluble humic acid had an ash content of several percent and again had little exchange capacity for lead and cadmium. An emission spectrographic survey revealed the presence of Ca, Al, Mg, and Fe as major metallic constituents in the humic acid solution. Since humic acid is a cation-exchange material, all the above elements can be displaced with a strong acid. Hydrochloric acid was selected because it is non-oxidizing; it does not precipitate any of the above elements; and, it is removed readily from the humic acid on vacuum desiccation.

Data in Table 1 show the effect of repeatedly precipitating the humic acid with HCl, centrifugation, decanting the supernate, then resolublizing with ammonia water. The data indicate that most of the metals were released upon the first acidification. Subsequent treatments served to wash the humic acid. Allowing the humic acid to stand overnight in acid brought a second, but much smaller, increase in metals released; this is thought the result of elimination of metal

ions from the interior of the humic acid molecules. The ash content of the HCl-purified product is 0.9%.

An amperometric titration of the HCl purified humic acid with lead, using ASV for detection of the endpoint, indicated an exchange capacity of 1.89 μg Pb/1-mg of humic acid at a pH of 4.7 (acetate buffer). The uptake of lead was instantaneous in the 5 minutes time scale required to do the ASV run after each addition of lead, and the lead peak heights did not change over a half hour period. However, since we were also interested in determining the exchange capacity on allowing the lead and humic acid to equilibrate for a day or more, it seemed important to further reduce the ash content which, by spectrographic analysis, was found mainly to consist of Fe, Al, and Si.

Since these elements were not eliminated with the HCl treatment, it seemed likely that they were somehow entrapped within the humic acid molecules, perhaps in the form of clay particles, so that treatment with HF would be required to effect their dissolution, hence, their release. Such was the case. Contacting the HCl-purified acid with the 1.4 M HF, 1.2 M HCl mixture for 2 days at room temperature, resulted in a product containing <0.1% ash.

The foregoing procedures work quite well for the humic acid at our disposal; however, this is no guarantee that humic acids from other sources can be purified to such high levels. There are myriad numbers of acids which contain ash contents all the way up to 50% (26). Furthermore, the procedures we used are quite harsh and probably cause some hydrolysis of the starting material (21), but the purified material

served our purpose in illustrating the attack of ozone on a large
molecule with the subsequent release of trace metals.

Figure 5 shows the amperometric titration of the HCl and the
HF purified humic acids. The HCl purified acid was little degraded
by treatment with the HCl/HF mixture; the instantaneous exchange
capacity of the latter was 1.85 µg Pb/1-mg humic acid--essentially
the same as that above. All the long-term exchange capacity
experiments were made with the purified acid.

To quantify the long-term exchange capacity, a series of solutions
were made in which up to four times the amount of lead required to do
the instantaneous titrations was added. These solutions were allowed
to stand 24 hour, or more, in the dark at room temperature. The data
from the first series indicated that the complexing ability of the
humic acid <u>decreased</u> upon standing. Since humic acid is of natural
origin, it was thought that microbial degradation might be the cause of
the decrease. Chloroform has long been used to prevent the growth
of microbes in thiosulfate solutions with good success (27), so it
was tried with the result that the humic acid was stabilized. The
exchange capacity increased from the value above to 3.64 µg Pb/1-mg
humic acid to give an instantaneous to long-term exchange capacity
ratio of about 1:2. This ratio is similar to those we obtained with
ASV for the release of lead from sequestering agents in seawater upon
the intial acidification to pH 2 compared to lead values obtained
after the water sample had sat at pH 2 for 1 day or more. See
Figure 5. The increase in the exchange capacity with time is the
result of trace metals tunneling into the humic acid molecule.

The first attempt to ozonize humic acid at the 100 ppm level

in neutral solution was unsuccessful. Considering the small amount

of humic acid employed, the volume of tough, stable foam produced was

almost unbelieveable. Only after the humic concentration was decreased

to 25 ppm was it possible to proceed but then only at a reduced oxygen

flow-rate of <2 l/min. Also, it was necessary to repeatedly stop

the oxygen flow and to wash down the walls of the ozonolysis tube

with sample solution sucked up with a pipet. The most troublesome

stage was the first 30 minutes, after which it was necessary to wash the

walls down only every 10 minutes. The characteristic brown color of humic

acid was discharged in the first 30 to 45 minutes of ozonolysis.

Three hours of ozonolysis was required to attain recoveries of

96 and 99% for cadmium and lead, respectively. The course of oxidation

differed from that in acid solution discussed below because adsorption

of the oxidized products at the electrode did not appear to be a problem.

Predictably, foaming was of much less concern at pH 2 because

the humic acid was virtually all precipitated under these acidic

conditions. However, the foam that did appear carried the flocclant

precipitate to the surface of the solution and deposited it on the

walls of the ozonolysis tube. Frequent washing of the tube walls

with sample solution was required for about the first 30 minutes until

the precipitate was degraded sufficiently to be dissolved.

Adsorption of oxidation products at the electrode surface was

a severe problem. A standard additions plot taken after 1 hour in

which the electrode was not buffed between runs illustrates this.

See Figure 6. The loss of sensitivity occurred at a much greater

rate than for EDTA. Bowing of the curve was so severe that a linear extrapolation to zero was impossible. Only after the sample had been ozonized for 4 hours was a linear standard additions plot for lead obtained. Again see Fig. 6. Four hours is, of course, a long time to ozonize an actual sample, but we have reduced the time required through the use of calibration charts as described in the recommended procedure and illustrated below.

The adsorption phenomenon is a temporal problem. The longer the electrode was in contact with the sample solution, the greater the diminution of signal. This would indicate that if it were possible to use a single peak taken with a freshly polished electrode, rather than a series of peaks obtained in the usual standard additions plot, perhaps the problem could be circumvented by reducing the time the electrode is in the solution. Table 2 will illustrate this line of reasoning. The left hand portion of Table 2 shows the values of Pb and Cd obtained in a standard additions plot before humic acid was added. The peak heights of the Pb and Cd before any standard addition was made are also given. The agreement among the peak heights is quite good because the method used for polishing the electrode does not grossly affect the electrode surface area. Compare these peak heights with those obtained during the ozonolysis shown at the right of Table 2. Although 3 hours of ozonolysis were required to obtain a linear standard additions plot for cadmium and 4 hours for lead, the peak height of the first scan became constant after only 30 minutes for cadmium and after 60 minutes for lead.

In all the studies made above, the peak height of cadmium is little affected by the presence of the oxidized organic compound; the lead peak height, however, is always reduced some 10 to 20 percent in the presence of the products of ozonolysis. The foregoing suggests that if water is sampled from a single source, and if after being ozonized it is high in surface active agents, it would be advantageous to develop a master calibration chart from the ozonized sample rather than relying on the standard additions method to quantify the sample. This will result in significant time savings.

The ultraviolet absorbance spectrum of humic acid in neutral solution is similar in shape to that shown above for tannic acid after ozonolysis. In view of the larger diameter of the humic acid molecules, this kind of absorbance spectrum could be simply the result of increasing light scattering at increasingly shorter wavelengths. After ozonolysis, only simple weak acids remain (23,24). These have absorbance spectra similar to that of ozonized tannic acid.

The efficacy of the presented method was tested using water taken from the Berkeley Marina; sewage effluent taken from an 80 million gallon per day (MGD) primary treatment plant which serves both industrial and residential sources; and, sewage effluent from a 2 MGD secondary treatment plant which serves only residential sources. The major difference in the water sources is the dissolved organic content. As received, the samples were centrifuged at 7000 rpm for 55 minutes (the calculated particle size remaining in solution was $\leq 0.1\mu$ based upon a density of 2.5), acidified to pH 2 with HCl, then analyzed. The sewage samples behaved in a manner similar as humic acid.

Ozonolysis produced initially a great deal of foam which diminished as the oxidation progressed. Polishing the electrode after each ASV scan gave consistent results for the Pb and Cd peak heights after 1 and 2 hours of ozonolysis; for example, the lead peak for the 2 MGD sample was 26 and 28 for two scans taken after 1 hour and averaged 27 for the three scans taken after 2 hours. The results of these and other analyses are shown in Table 3. The standard deviations for the sewage samples are not as good as for those shown in Table 2. This is due to the fact that these samples contain components which are not completely characterized as is the case for the simulated samples. For example, the quartz cell invariably became coated with an oily film when running samples from the 80 MGD plant which necessitated boiling the cell in red fuming nitric acid. In addition, calcium phosphate or sulfate which might be stabilized by organics present in sewage effluents can precipitate after ozonolysis at pH 4.7. The sewage samples represent the worst possible case. Bay water taken from Berkeley Marina, in contrast, presented no particular problems, and the reproducibility obtained was better. The bay water was sufficiently devoid of organics that ASV signals could be obtained without ozonolysis, however, the reproducability of these results was not as good as those obtained after ozonolysis.

The foregoing work emphasizes the statement we made over 2 years ago that the adsorption of surface active compounds might well be the limiting factor in the on-line application of ASV to flowing systems (1). The recent work of Zirino further bears this out (28). It may be possible to incorporate ozonolysis in an on-line system

which would ozonize the water sample at the rate of 5 to 10 ml per minute provided a 10 to 15 minute delay between sampling and the output of data is acceptable.

Although the surface-active agents generated on less than exhaustive ozonolysis of samples high in organics forced us to modify the ASV standard additions method somewhat, it is likely that this method will be completely acceptable to those using solvent extraction or chelating resin preconcentration techniques prior to the analysis of metals by other instrumental methods.

As an outgrowth of the foregoing work, we are now building a 5 tube ozonizer which will permit a much higher sample through put and will be powered with a 60 kV transformer. The tubes will be wired in series so the voltage drop across each will be 12 kV.

The next logical extention of the method is to the digestion of solid or liquid biological samples. Nitric acid rarely has sufficient oxidative capacity to be used alone; the presence of H_2SO_4 and/or $HClO_4$ is required. However, calcium is precipitated in the presence of the former and potassium in the presence of the latter. It seems likely that if the sample were dissolved in warm concentrated nitric acid, then subjected to ozonolysis, complete digestion would occur. If the sample were not being prepared for analysis with ASV, then oxidation catalysts such as Ag or Hg could be included. The problems associated with the non-volatile acids could thus be circumvented.

References

1. R. G. Clem and A. F. Sciamanna, Anal. Chem. $\underline{47}$, 276 (1975).

2. J. P. Riley and D. Taylor, Anal. Chem. Acta. $\underline{40}$, 479 (1968).

3. W. G. Cox in "Proceedings of the Specialty Conference on Dredging and Its Environmental Effects," Am. Soc. of Civil Eng., publisher, New York, 1976, pp. 242-252.

4. W. R. Matson, Ph. D. Thesis, M. I. T. (1978).

5. F. A. J. Armstrong, P. M. Williams, and J. D. H. Strickland, Nature $\underline{211}$, 481 (1966).

6. R. G. Clem. Anal. Chem. $\underline{47}$, 1778 (1965).

7. C. C. Patterson and D. M. Settle, "7th Materials Research Symposium," NBS, Gaithersburg, MD, October 7-11, 1974.

8. R. G. Clem, G. Litton, and L. D. Ornelas, Anal. Chem. $\underline{45}$, 1306 (1973).

9. H. H. Willard and L. L. Merritt, Ind. Eng. Chem., Anal. Ed. $\underline{14}$, 486, 489 (1942).

10. J. G. Calvert and J. N. Pitts, "Photochemistry," Wiley, New York, 1967.

11. F. L. Evans, "Ozone in Water and Wastewater Treatment," Ann Arbor Science, Ann Arbor, MI, 1972.

12. H. R. Eisenhauer, J. Water Pollution Control Federation $\underline{40}$, 1887 (1968).

13. E. Bernatele and C. Frengen, Acta Chemica Scandinavica $\underline{15}$, 471 (1961).

14. P. S. Bailey, Chemical Reviews $\underline{58}$, 925 (1958).

15. P. S. Bailey, and J. E. Keller, J. Org. Chem. $\underline{33}$, 2680 (1968).

16. P. S. Bailey, J. E. Keller, D. A. Mitchard, and H. M. Wite, "Advances in Chemistry Series," No. 77, Am. Chem. Soc., Washington, D.C. (1968), p. 58.

17. P. S. Bailey, J. E. Keller, and T. P. Carter, J. Org. Chem. 35, 2777 (1970).

18. P. S. Bailey, D. A. Mitchard, and A. Y. Khashab, J. Org. Chem. 33, 2675 (1968).

19. L. Horner, H. Schaefer, and W. Ludwig, Chem. Berich. 91, 75 (1958).

20. F. J. Welcher, ed., "Standard Methods of Chemical Analysis," Vol. 2, part B, Van Nostrand, New York, 1962, p. 2461.

21. J. J. Alberts, J. E. Schindler, D. E. Nutter, and E. Davis, Geochim. et Cosmochim. Acta 40, 369 (1976).

22. A. Nissenbaum and D. J. Swaine, Ibid, p. 809.

23. M. D. Ahmed and C. R. Kinney, J. Am. Chem. Soc. 72, 559 (1950).

24. F. Dobinson and G. J. Lawson, Fuel 38, 79 (1959).

25. R. Ernst, H. E. Allen, and K. H. Mancy, Water Research 9, 969 (1975).

26. R. L .Malcolm, J. Res. U.S.G.S. 4, 37 (1976).

27. J. L. Kassner and E. E. Kassner, Ind. Eng. Chem. Anal. Ed. 12, 655 (1940).

28. A. Zirino and S. H. Lieberman, in "Analytical Methods in Oceanography," T. R. P., Gibb, ed., Advances in Chemistry Series 147, A. C. S., Washington, D.C., 1975, p. 82.

Table 1. Emission Spectrographic Analysis of the HCl Supernates[a]

Supernate No.[b]	Elements (µg/ml)										
	Ca	Al	Mg	Na	Fe	K	Ti	Mn	B	Sr	Si
1	500	200	500	500	50	<10	0.3	1.0	<1.0	0.5	<0.5
2	10	5	3	50	5	<10	0.1	<0.1	<1.0	≤0.1	<0.1
3	3	2	2	20	3	<10	<0.1	<0.1	<1.0	<0.1	<0.1
4[c]	3	5	2	<10	3	<10	<0.1	<0.1	<1.0	<0.1	<0.1
5	0.5	1	2	<10	1	<10	<0.1	<0.1	<1.0	<0.1	<0.1

[a]Total weight of humic acid is ~40 g.

[b]Total volume 800 ml each.

[c]Metals released after standing overnight.

Table 2.

Run	Baseline				Data for Humic Acid Ozonolysis				
	Cd ppb	pk.ht. mm	Pb ppb	pk.ht. mm	t (min)	Cd ppb	pk.ht. mm	Pb ppb	pk.ht. mm
1	14.7	64	18.9	56	30	–	64	–	36
2	15.8	68	19.2	58	45	–	67	–	41
3	15.5	67	19.7	59	60	–	67	–	41
4	16.0	69	19.5	57	90	–	67	–	45
					120	–	69	–	46
					180	16.2	69	–	46
					240	16.2	72	19.7	47
AV	15.5	67	19.3	58	AV	–	68	–	46
St'd Dev.	±0.57	±2	±0.35	±1.4	St'd Dev.	–	±2.4	–	±1
%St'd Error	±3.7	±3.0	±1.8	±2.4	%St'd Error	–	±3.7	–	±2.2
n = 4					n		6 from 45 through 240 min.		4 from 90 through 240 min.

Table 3. Analysis of Natural Samples

		Cd		Pb	
	Ozonolysis Time (hr)	ng/ml	pk.ht.(mm)	ng/ml	pk.ht.(mm)
Source: 80 MGD Treatment Plant					
Sample: 10/7 BOD: 250 ppm					
Date Run					
10-18	0	----	2	----	24
	0.75	non-linear plot	10	non-linear plot	24
10-21	1	2.3	10	10.6	36
	2	2.3	12	10.8	36
Source: 2 MGD Treatment Plant					
Sample: 3/18 BOD: 10 ppm					
Date Run					
3-21	0	----	2	----	double peak
	2	0.32	3	4.7	28
		0.29	3	4.0	27
		0.28	3	4.1	28
	Av.	0.30		4.3	
	St'd. Dev.	±0.02		±0.38	
Source: Berkeley Marina (San Francisco Bay Water)					
Sample: 11/15 BOD: not available					
Date Run					
11-19	0	----	8	0.83	30
11-20	0	0.11	15	0.62	37
		0.22	21	1.06	53
		0.19	11	0.93	52
	Av.	0.17		Av. 0.87	
	St'd. Dev.	±0.06		±0.23	
11-22	1	0.16	8	0.85	25
		0.16	7	0.78	26
		0.17	8	0.85	28
		0.12	10	0.70	33
	Av.	0.15		0.80	
	St'd. Dev.	±0.02		±0.07	

Figure Captions

Fig. 1.   Effect of pH on the resolution of Pb and Cd stripping peaks.

Upper curve: pH = 4.7 acetate buffer,

Lower curve: pH = 8.7 bicarbonate solution.

Fig. 2A.   Ozonolysis of EDTA in bicarbonate solution.

Solution: 0.1 $\underline{M}$ NaHCO$_3$, 0.5 $\underline{M}$ KCl.

Fig. 2B.   Ozonolysis of EDTA at pH 2.

Solution: 0.01 $\underline{M}$ HCl, 0.5 $\underline{M}$ KCl.

Fig. 3.   Ozonolysis of APDC at pH 2. Solution conditions same as

for 2B.

Fig. 4A.   Ozonolysis of tannic acid at pH 2. Solution condition same

as for Fig. 2B.

Fig. 4B.   Wavelength vs absorbance for tannic acid. Solution conditions

same as for Fig. 2A. Lower trace is for gallic acid, cell

pathlength = 0.1 cm.

Fig. 5.   Amperometric titration of humic acid with Pb.

Solution: 0.5 $\underline{M}$ KCl containing 378 µg humic acid per 15

ml and buffered to pH 4.7 with 0.03 $\underline{M}$ OAC$^-$/0.03 $\underline{M}$ NaOAc.

Top curve: HCl purified humic acid,

Middle curve: HF purified humic acid,

Bottom curve: HF purified humic acid that has been

equilibrated with added lead for more than 24 hours.

Fig. 6.   Standard additions plot of Pb and Cd in the presence of

humic acid. Upper curve: after 60 minutes ozonolysis.

Lower curve: after 240 minutes ozonolysis.
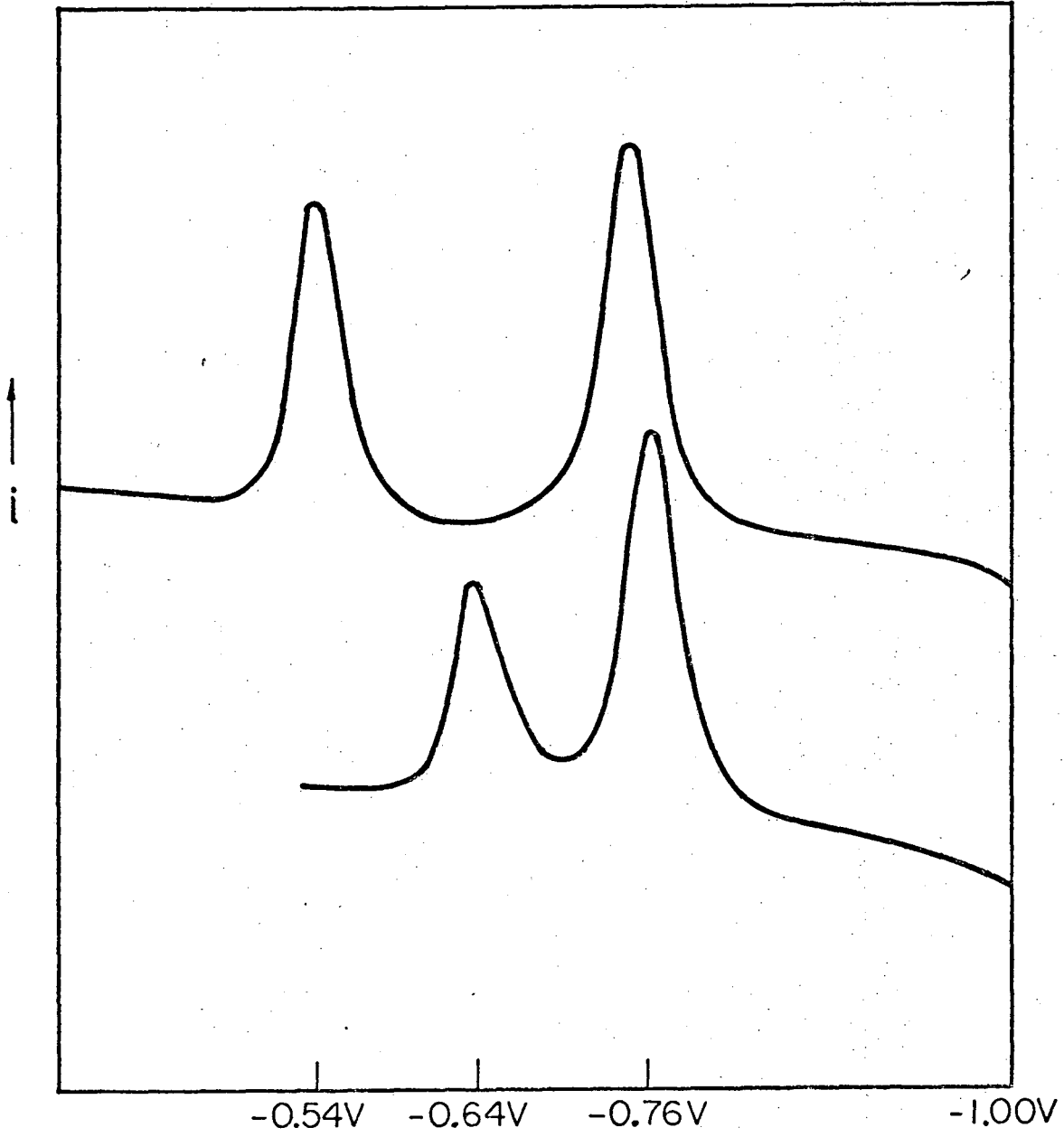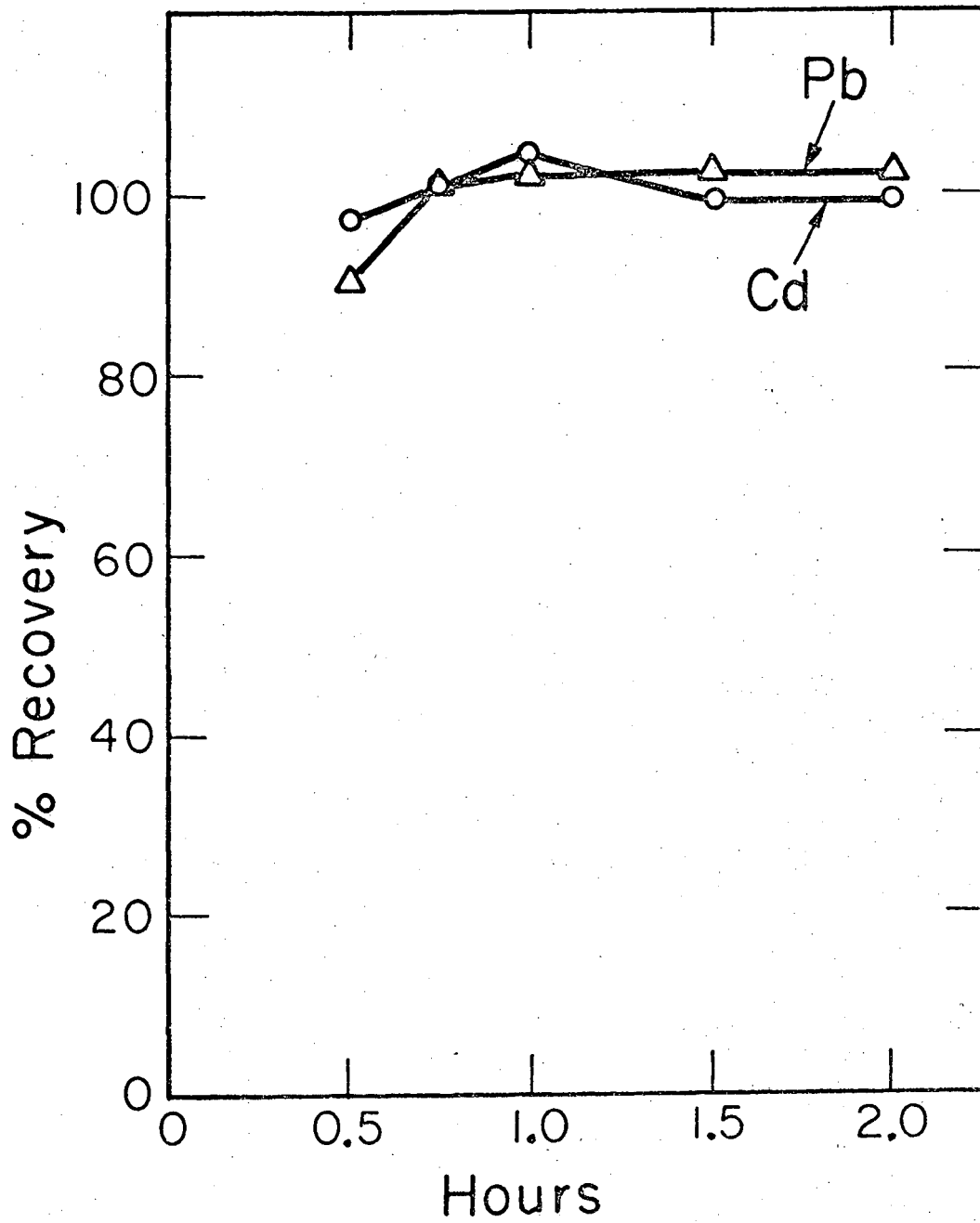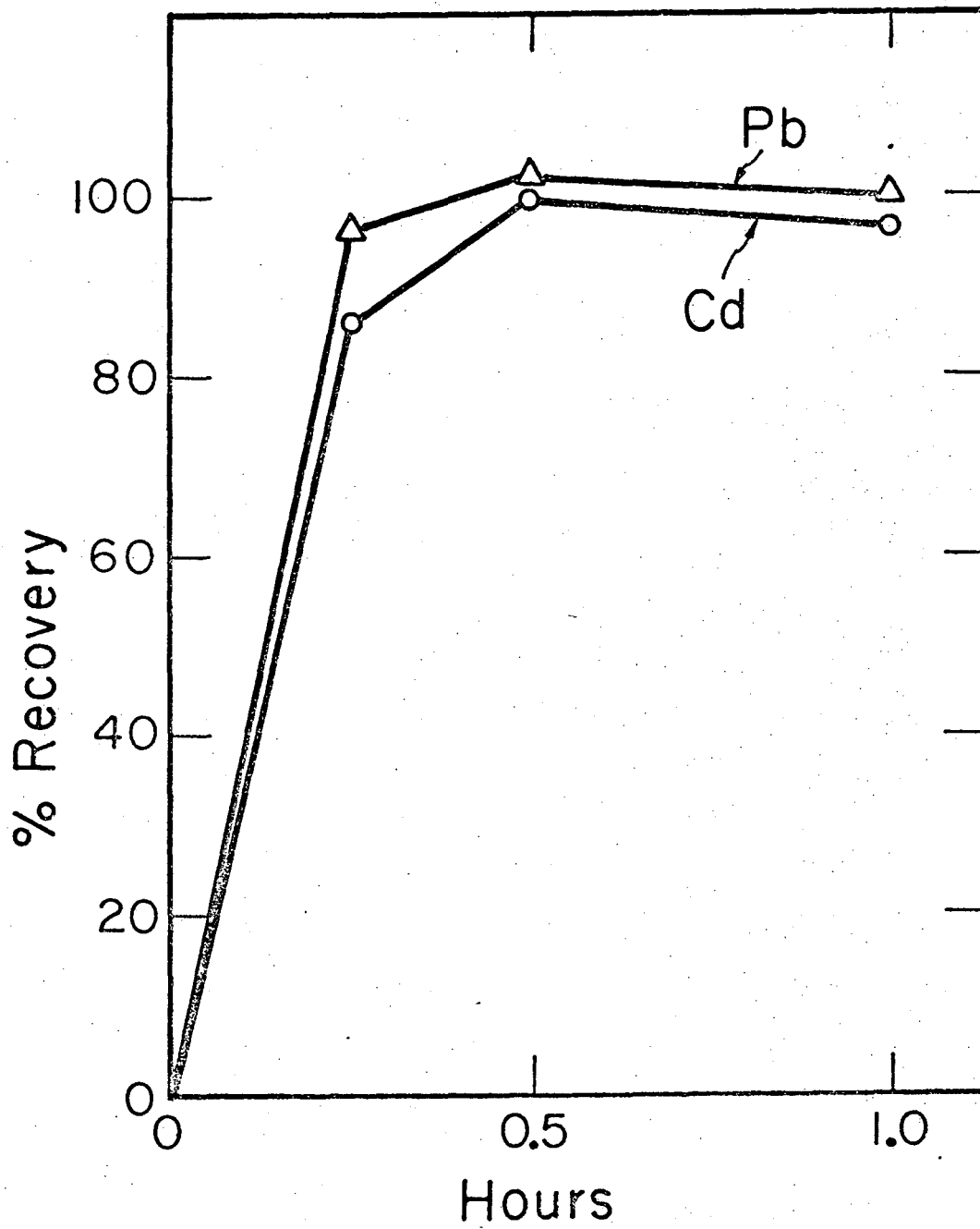
E vs SCE
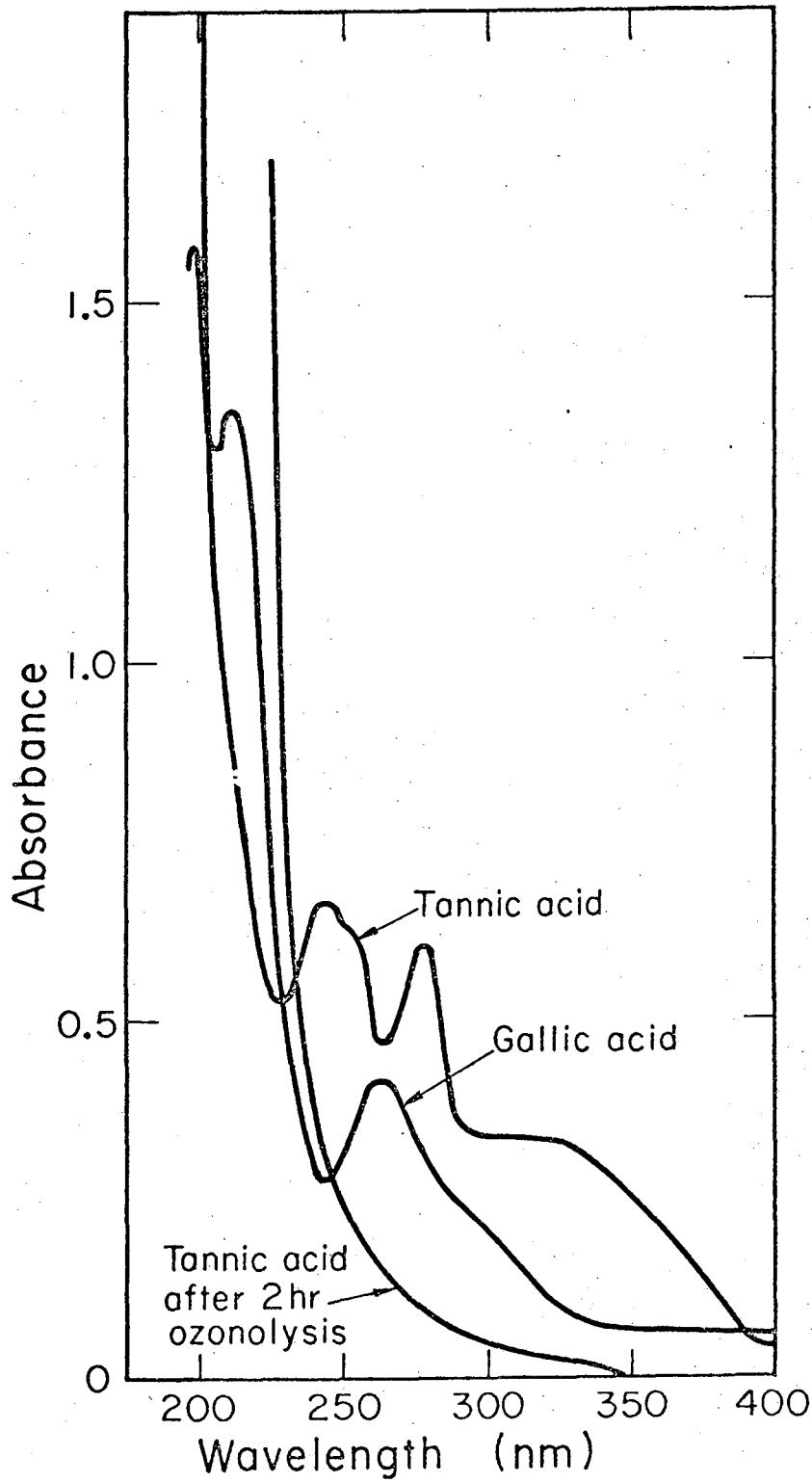
XBL774-3347

Fig 1
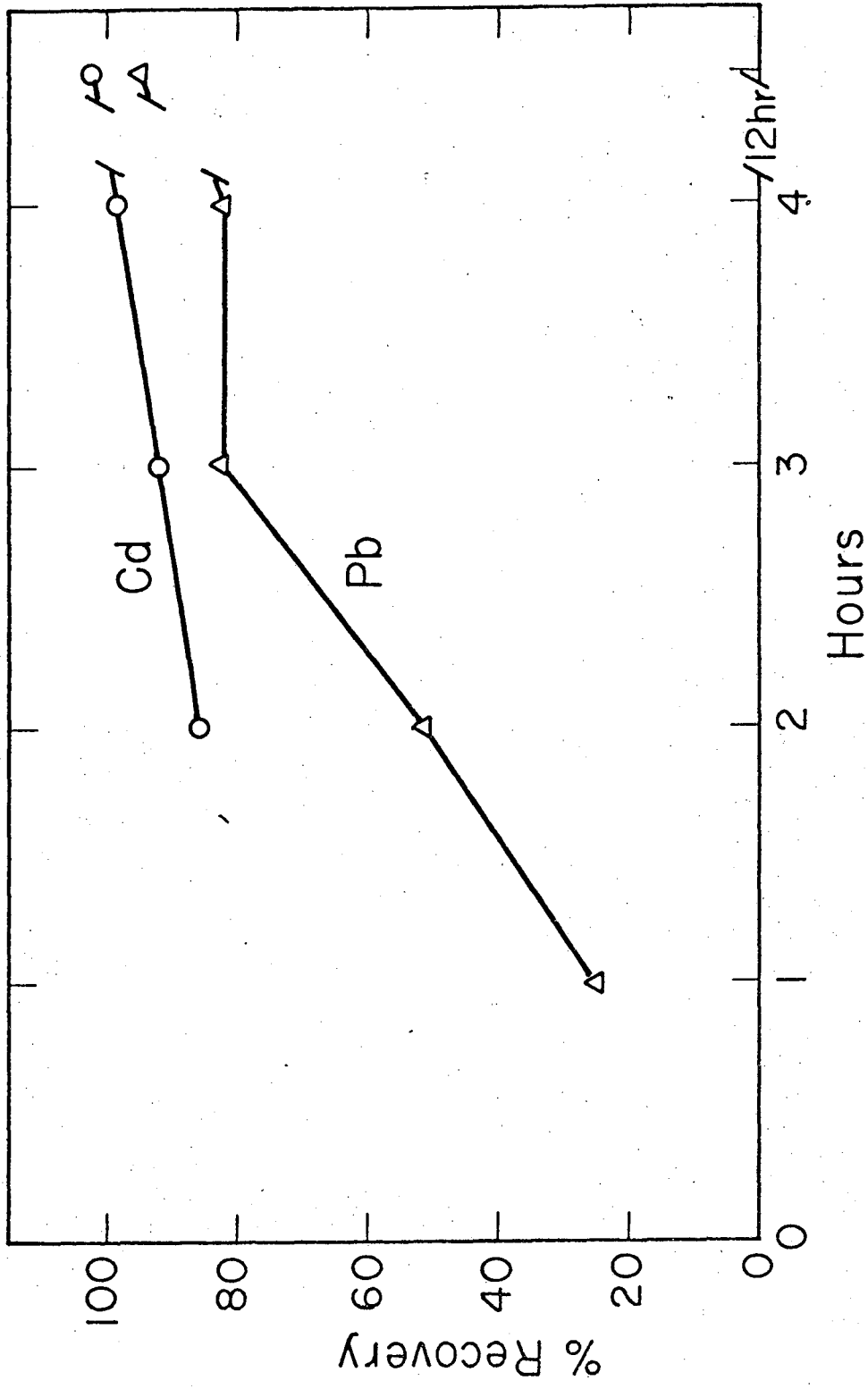
Fig 2a
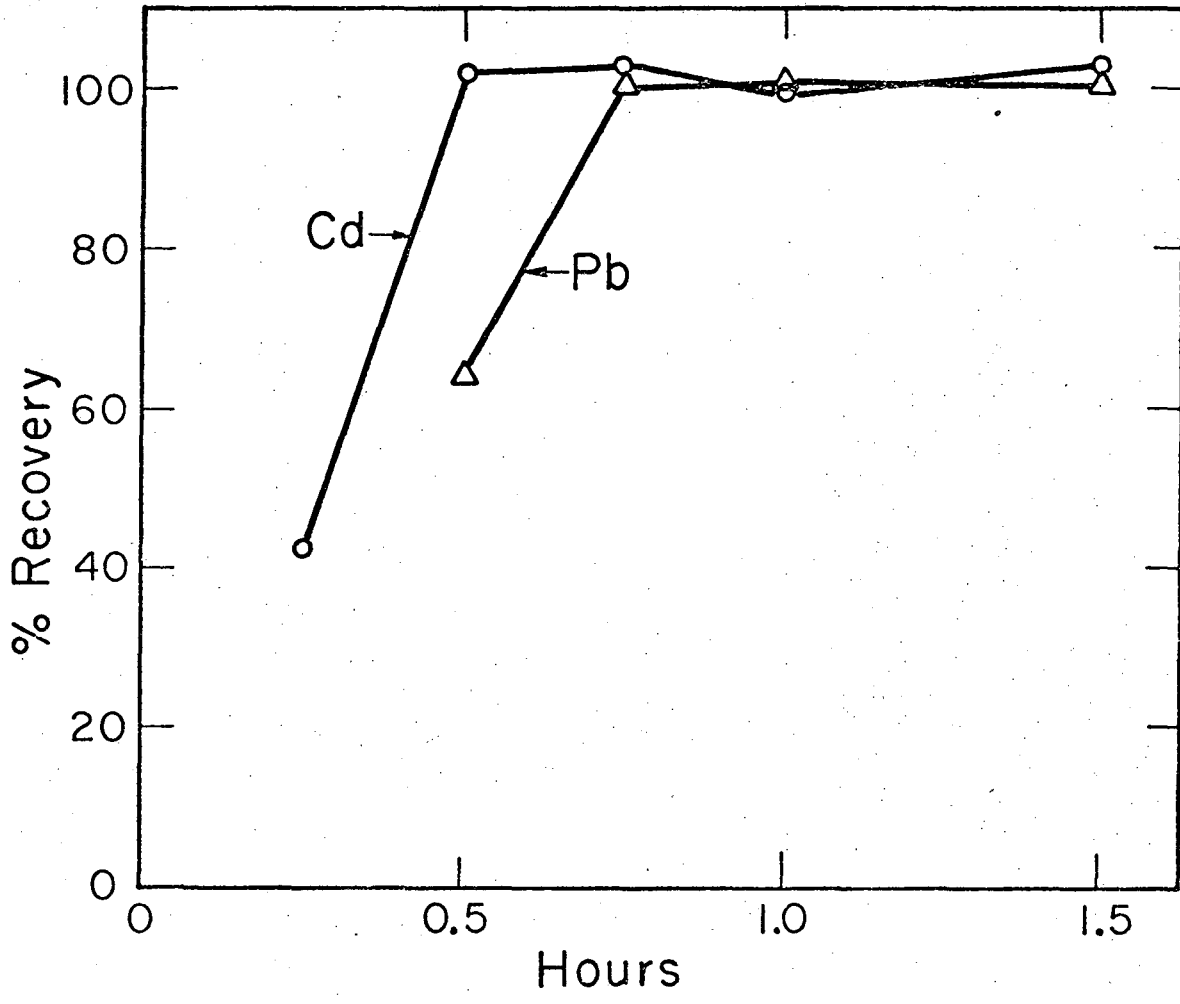
XBL774-3348

XBL774-3346

Fig 2b

XBL774-3345

Fig 3

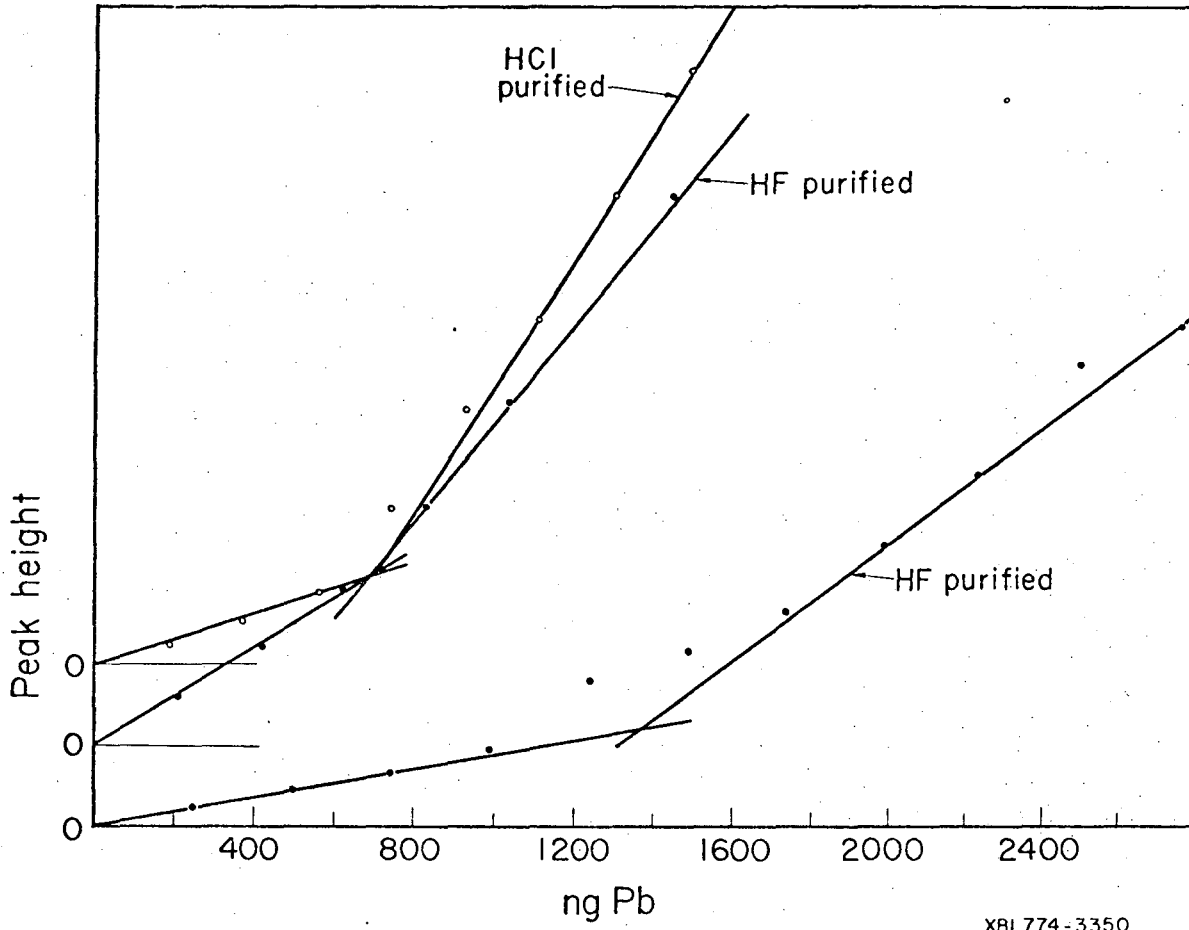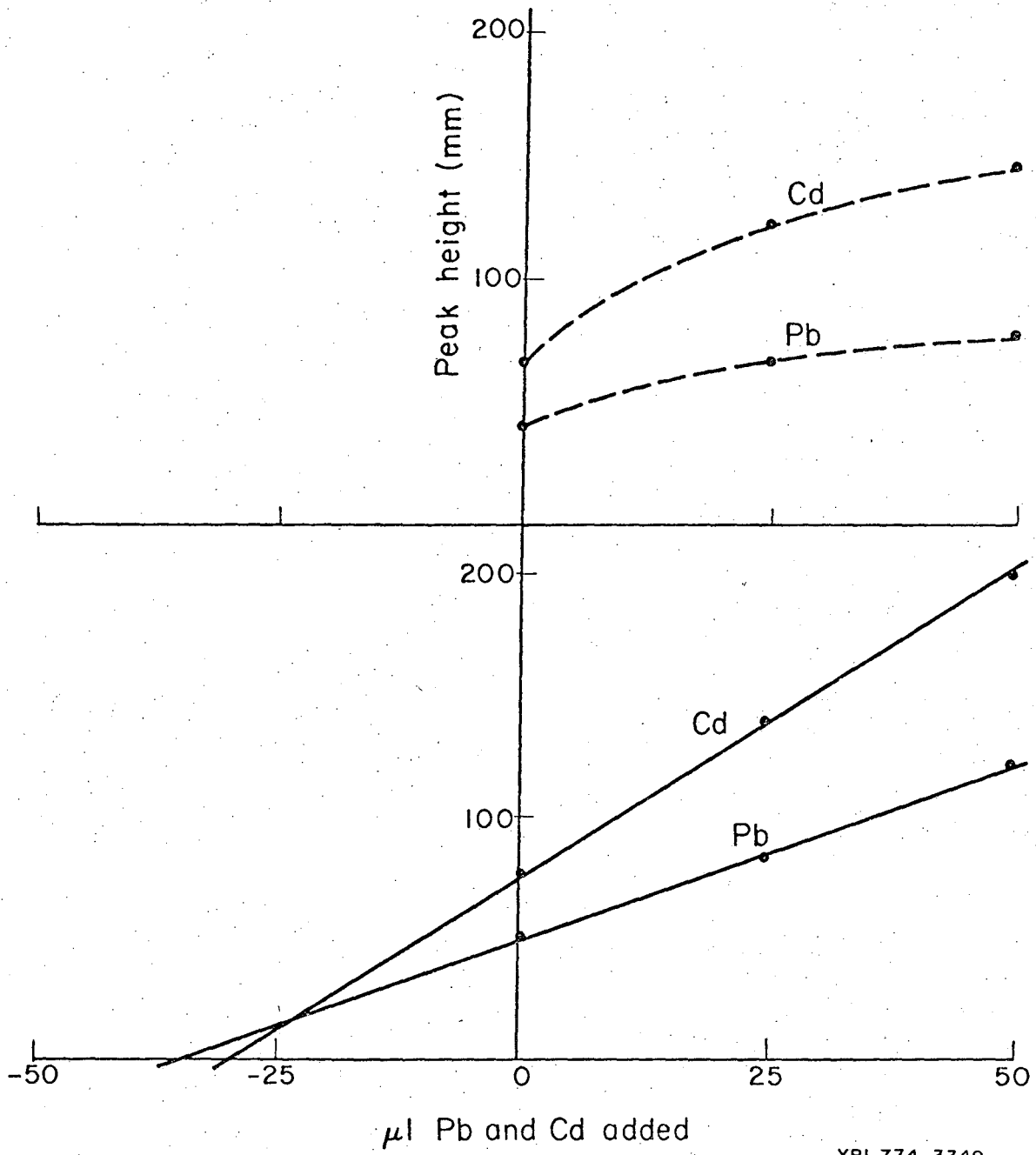XBL774-3343

Fig 4a

Fig 4b

XBL774-3344

Fig 5

μl Pb and Cd added

XBL 774-3349

Fig 6