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ENVIRONMENTAL TRACE ANALYSIS OF ORGANICS IN WATER BY GLASS CAPILLARY COLUMN CHROMATOGRAPHY AND ANCILLARY TECHNIQUES

PRODUCTS OF OZONOLYSIS

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SUMMARY

A method employing a polymeric sorbent has been used for analysis of volatile organic components in water. Trace level organics are sparged from water with nitrogen gas and are concentrated on Tenax GC prior to analysis with either flame ionization gas chromatography or gas chromatography–mass spectrometry. Glass capillary columns were used to obtain maximum resolution of chromatographic peaks. Specifically, the method has been applied to a qualitative and quantitative study of the products which result from ozonization of secondary treated domestic wastewater. The principal volatile products of ozonolysis are *n*-hexanal, *n*-heptanal, *n*-octanal and *n*-nonanal. Representative samples contained 0.7 ppb** of *n*-heptanal and less than 1 ppb of any of the volatile compounds.

INTRODUCTION

The technique of concentrating trace level constituents in environmental samples with polymeric sorbents is well known. Previous applications of polymeric 2,6-diphenyl-*p*-phenylene oxide (Tenax-GC; Enka, Arnhem, The Netherlands) have included headspace analyses of biological fluids[1,2], direct concentration of trace constituents of air samples[3–7], analysis of tobacco smoke[8], headspace analyses of water[9,10], and direct sorption of organics in water samples[11].

When coupled with high-resolution gas chromatographic (GC) columns, the polymeric sorbents become an integral part of a sophisticated analysis system, which may also be coupled with a mass spectrometer and computer. The sorbent provides a means of concentrating the sample components and the use of glass capillary chromatography columns provides a means of separating the complex mixture of components routinely encountered in environmental sampling. A gas chromatograph–mass spectrometer–computer system provides both quantitative and qualitative data, as well as the capability to store and retrieve the large amounts of information generated

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** Throughout this article, the American billion (10^9) is meant.

in each experiment. In one phase of our environmental studies we are using this instrumentation to identify trace organics appearing in water and wastewater. In particular, the compounds formed when ozone is used instead of chlorine for disinfection of water are being studied.

Ozonolysis has been used in France and other European countries as a disinfectant for potable water[12]. This treatment has not been approved for similar use in the United States because drinking water standards in this country require a residual disinfection capability such as provided by chlorine treatment. Ozone, unlike chlorine, rapidly decomposes in the water distribution lines, and if there is later contamination a health hazard may result. For wastewater streams, however, the residual disinfectant criterion does not apply and thus ozonolysis may be a viable alternative treatment.

In light of recent reports of chlorinated hydrocarbons in potable water supplies[13,14], some of which may arise from chlorination of wastewater[15] entering rivers from which some cities take their drinking water, it is important to determine if other methods of treatment will limit or reduce the formation of such compounds. Some of these chlorinated compounds are known carcinogens and efforts are being made to reduce or eliminate them wherever possible. The Upper Thompson Sanitation District (UTSD) wastewater treatment plant at Estes Park, Colo., U.S.A., is a new facility in which ozone is being used as a chemical disinfectant of the biologically treated secondary effluent before discharge to the Big Thompson River. The determination of the chemical consequences of this kind of treatment is necessary in order to assess the impact of using ozone as a disinfectant, and to determine whether the products formed are less objectionable than those formed by chlorination[25].

EXPERIMENTAL

Sample collection and handling
       All glassware which was to be in contact with water samples was carefully washed with hot water containing Alconox detergent, rinsed three times with hot tap water, three times with deionized distilled water, once with acetone and then methanol, and finally three times with deionized distilled water. Glassware was then oven-dried at 110°. Samples were collected in 4-l amber glass bottles fitted with aluminum-lined caps. These bottles were rinsed with three aliquots of sample water, filled to overflowing and capped. Samples were stored in ice for transporting and maintained in the laboratory at 0–4° until analysis, usually within 1 or 2 days.

Total organic carbon analysis
       An aliquot of each wastewater sample was placed in a clean vial and stored at 0–4° until analysis for "total organic carbon" using a Dohrmann Model 52D total organic carbon analyzer (Envirotech, Santa Clara, Calif., U.S.A.). This instrument measures the flame ionization response of organic material which is catalytically reduced to methane. Aliquots of water (1–30 μl) were analyzed in triplicate.

Headspace sampling
       Dynamic headspace concentration of volatile organic compounds was performed using a procedure and apparatus (Fig. 1) modified from that of Chesler *et al.*[9].

In this method volatile organic compounds are collected in a porous polymer bed after sparging the aqueous solution with a stream of nitrogen gas, previously purified by passing through a trap filled with Tenax-GC. Glass tubes containing Tenax-GC (60–80 mesh) were fabricated from standard 18 cm × 6.4 mm O.D. × 4 mm I.D. borosilicate glass stock. Each tube contained approximately 0.1 g of the polymeric sorbent, filling 7–8 cm of the tube.
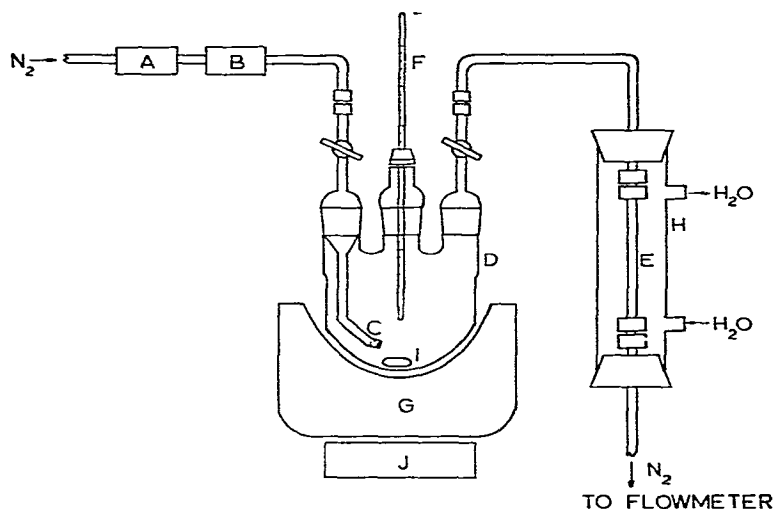

Fig. 1. Dynamic headspace sampling apparatus. Nitrogen gas was passed first through traps of molecular sieves (A) and Tenax-GC (B). The gas flow continued through a fritted glass tube (C) positioned at the bottom of the water sample in a 2-l round-bottom flask (D). Volatile components are sorbed by Tenax-GC in a glass tube (E) packed at both ends with glass wool. The sample was maintained at 70° (measured with a thermometer (F)) by a heating mantel (G). A cold water condenser (H) was used to cool the Tenax tube, maintaining an annular temperature of 10 ± 3°. Contents of the vessel were stirred with a glass-covered magnetic stirring bar (I) and electric motor with magnetic drive (J). Ground glass joints and standard Swagelok fittings with PTFE ferrules were used for all connections. Ground glass surfaces were not greased, but these joints were held tightly with springs.

The sparging flask containing 0.5–1.5 l of water was slowly heated to 70° during the first 0.5 h of gas flow (150 ml/min) and maintained at 70 ± 2° for 1.5 h. At that time the sorbent tube was disconnected from the vessel and reconnected directly to the nitrogen line for removal of residual water. The sorbent tube was maintained at 10 ± 3° during sample concentration and at room temperature during the water removal step. Drying required 1 h at a flow of 150 ml/min. The tubes were sealed with Swagelok caps and refrigerated at 0–4° until GC analyses were performed, usually within 1 or 2 days.

Preparation of open-tubular columns

Open-tubular GC columns were prepared from cleaned tubular borosilicate glass stock using a Hewlett-Packard Hupe-Busch 1045A capillary-drawing machine. Capillaries for preparation of support-coated open-tubular (SCOT) columns were drawn to 50 m with 0.5 mm I.D. and 0.8 mm O.D. The capillaries for preparation of wall-coated open-tubular (WCOT) columns were drawn to 60 m × 0.25 mm

I.D. × 0.35 mm O.D. Prior to column coating, interior surfaces were deactivated by three treatments with 1% benzyltriphenylphosphonium chloride in water. After removal of this solution by flushing with water, the column was heated at 250° while flowing nitrogen for 2 h.

SCOT columns were prepared[16] by mixing 0.5 g of OV-101 polydimethyl-siloxane (Applied Science Labs., State College, Pa., U.S.A.), and 2 g of Silanox 101 silica (Cabot, Boston, Mass., U.S.A.) in 100 ml of carbon tetrachloride and filling 25% of the column length with the suspension. The mixture was forced through the capillary and then out under nitrogen pressure and the residual solvent evaporated with nitrogen gas. The support was further coated by filling the column with 10% OV-101 in octane. The plug of the solution was forced out and the column conditioned by passing nitrogen at room temperature for 3 h, followed by temperature-programming the column to 200° at 1°/min, and maintaining the column at 200° for 12 h.

WCOT columns were prepared[17,18] by passing through the column a 10% solution of OV-101 in chloroform at a rate of 20 mm/sec. The plug of solution was forced out and nitrogen was passed through the column for 12 h at room temperature. Following this the temperature was programmed to 250° at 1°/min and held constant for 1 h.

Gas chromatography

A Varian Model 2400 gas chromatograph (Varian, Palo Alto, Calif., U.S.A.) equipped with dual flame ionization detectors (FIDs) and a Hewlett-Packard (Palo Alto, Calif., U.S.A.) Model 5982A gas chromatograph–mass spectrometer–computer system were modified with heated 10-port gas switching valves (Valco, Houston, Texas, U.S.A.). Additional modifications were made so that the FID instrument could accept a WCOT column and the gas chromatograph–mass spectrometer could be used with a SCOT column. Appropriate injection and detector port liners were inserted to reduce dead volume and a make-up gas tee union was installed for the FID. The gas chromatograph–mass spectrometer interface was modified so that the part of the transfer line of the chemical ionization inlet as far as the isolation valve was replaced with 0.3 mm I.D. glass-lined stainless-steel tubing (Scientific Glass Engineering, Houston, Texas, U.S.A.). A SCOT column was interfaced directly to the ion source without use of any carrier gas-separating device such as a membrane or jet separator.

Following sorption of volatile organic compounds on Tenax-GC the samples were thermally desorbed into a capillary column maintained at sub-ambient temperature. To accomplish this a Tenax tube was connected to the gas-switching valve and backflushed with helium while rapidly heating the tube to 250° with an aluminum block heater. The desorbed sample was condensed and thermally focused in the inlet end of the column, which was cooled by adding solid carbon dioxide to the GC oven to maintain it at −20°. After thermally desorbing 8–10 min at 2–4 ml/min carrier gas flow, the sample tube was switched out of the gas flow line, which effectively removes it from the analysis system. Rapid elution began as the temperature of the column oven was raised and separation of sample components was effected by temperature programs extending to 180°.

Mass spectral data were obtained and stored by the data system from m/e 35 to 400 every 2.2 sec during entire chromatographic experiments.

Quantitative studies and retention time characteristics were determined with

the FID instrument. Additional quantitative data were obtained with packed columns using both instruments.

Quantitative measurement procedures

Quantitative data for compounds of particular interest were obtained by sparging pure water to which known amounts of a given compound had been added. These samples were collected on Tenax GC and analyzed in capillary columns in the same manner as the other water samples.

The overall recovery efficiency of sparging, sorption and desorption was determined for the experimental conditions employed for representative compounds. Packed column chromatography was used for the efficiency of recovery measurements. An aliquot of a standard aqueous solution containing the same amount of the organic compound present in the entire sample taken for sparging was injected into the chromatograph and analyzed using the same temperature program. The peak from this chromatogram was taken as the norm against which recovery efficiencies were calculated.

RESULTS AND DISCUSSION

Sampling and characterization of domestic wastewater

Typical headspace sample analyses of wastewater taken prior to and subsequent to chemical disinfection by ozone are presented in Fig. 2. The chromatograms A and B are presented as mass-resolved chromatograms according to the method of Biller and Biemann[19]. (The program was written by D. Pearce and provided by Hewlett-Packard.) This treatment of the data is useful to generate mass spectra free of peak overlap and extraneous peaks from column bleed. Although quantitative data are lost by such treatment, excellent spectra for qualitative identification and analysis are obtained. A typical total ion chromatogram, prior to data manipulation, is given as Fig. 2C. The very same chromatographic data treated by the Biller and Bieman method are shown in Fig. 2B.

The identification of each peak is based on comparison with spectra in the computerized library of spectra, followed by obtaining a spectrum from an authentic sample with our instrument. Identification of compounds for which quantitative results are presented have also been confirmed by retention time data.

The quantitative aspects of this study are especially interesting. The total organic loading of volatile and non-volatile compounds, as determined by total organic carbon analysis of the water discharged from the UTSD plant at the time of this study, was very low —in the range 5–15 ppm. By comparison, typical values of total organic carbon in the effluent of the Metro-Denver Sewage District No. 1 wastewater treatment plant range from 30 to 60 ppm. The amount of volatile material removed in the headspace procedure was such a small percentage of the total organic content that it apparently was not detectable by the total organic carbon method employed. Values of total organic carbon before and after headspace sampling were usually within the error limits established for each determination. Keith[20] has reported that all of the volatile compounds in heavily polluted industrial wastewater comprise less than 10% of the total organics present. If the same approximate percentage of organic compounds in treated domestic wastewaters is volatile, one might expect the total volatiles removed from these samples to be less than 1 ppm. Apparently a large
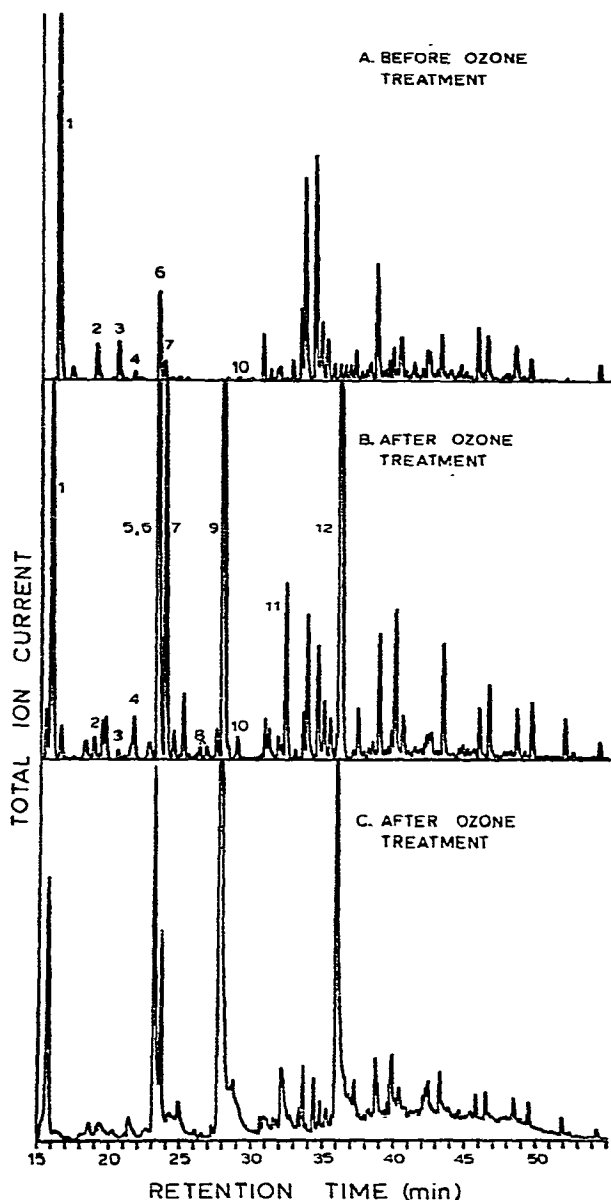
Fig. 2. Chromatograms of UTSD wastewater volatiles desorbed from Tenax-GC. The sample was desorbed onto the capillary column while the GC oven was maintained at −20° for 8 min. The oven was then temperature programmed to 170° at 4°/min. Helium carrier flow-rate was 4 ml/min. Mass spectra from m/e 35 to 400 were scanned and stored every 2.2 sec in the electron impact mode. (A) Mass-resolved chromatogram prior to ozonolysis. (B) Mass-resolved chromatogram subsequent to ozonolysis. (C) Same data as B prior to data reduction by Biller and Biemann technique. 1 = Chloroform; 2 = bromodichloromethane; 3 = dimethyl disulfide; 4 = toluene; 5 = *n*-hexanal; 6 = tetrachloroethylene; 7 = *n*-octane; 8 = xylene isomers; 9 = *n*-heptanal; 10 = dichlorobenzene isomer; 11 = *n*-octanal; 12 = *n*-nonanal.

portion of the organics in wastewater consists of humic acids and other non-volatile compounds.

Qualitatively, the volatile component elution profiles are remarkably similar for samples taken before and after ozone treatment, with the exception that prominent aldehyde peaks are found in samples taken after ozonization. The presence of aldehydes in the ozonized wastewater is an important, though not unexpected finding. The appearance of these compounds apparently constitutes the only significant change in the makeup of the volatile organic compounds in the treated water. Laboratory studies[21] of the ozonization of oleic acid give some indication of the types of compounds which may give such aldehydes upon ozonolysis. GC–MS analysis following ozonolysis of oleic acid in water shows that n-nonanal is a principal product. Molecules containing unsaturated hydrocarbon chains may be oxidatively cleaved by ozone to form an aldehyde with an appropriate number of carbon atoms. Substances such as oleic acid, unsaturated fats, olefins, and other unsaturated compounds are known to be present in wastewater, so any of these could be the aldehyde precursors. Organic chemists have for many years used ozonolysis of unsaturated compounds in organic solvents to locate the position of double bonds by identifying the products formed upon cleavage. The present work indicates that the same reactions occur in water through which ozone is bubbled. In addition, n-octane appears to be formed during ozonization of the wastewater (peak 7 in Fig. 2B). We have identified n-octane as a minor product of the ozonization of oleic acid, and nonanoic acid and nonanedioic acid are formed as well.

The chlorinated hydrocarbons found in these samples are of particular interest. One objective of the ozonolysis project at the UTSD plant is to determine whether ozonolysis is a viable alternative to chlorination for disinfection in wastewater treatment. Chlorine treatment is known to be a source of chlorinated hydrocarbons in the water disinfection process[15]. Although the origin of water sampled in this study is believed to be one of the cleanest sources possible (this first-use water arises primarily from the high-elevation watershed of the Rocky Mountains), a significant portion of the volatile organic content of the water consists of chlorinated hydrocarbons. The ozonolysis process, in which one could conceive of some chloride being oxidized to Cl_2, does not appear to be the source of these compounds. There is no chance of appreciable industrial contamination of the water supplies in this relatively pristine mountainous region. Preliminary studies of some of the domestic potable water influent for the plant suggest that at least some of the halocarbon loading is caused by chlorination of the drinking water supply. It is also important to note that the ozonolysis process does not change the concentration of the two major chlorinated constituents of these samples. Integration of appropriate single ion chromatograms shows that the amounts of chloroform and tetrachloroethylene are identical in samples taken before and after ozonolysis.

The presence in water of carcinogenic chlorinated compounds, even at very low concentrations, is a matter of growing concern. Some of the compounds are known to be carcinogenic in animals and give positive Ames tests[22] (indicative of mutagenicity in Salmonella bacteria), but the extent of the threat posed to humans is more difficult to assess. Very recently an epidemiological study[23] supported by the Environmental Protection Agency has revealed that there is a 44% higher incidence of gastrointestinal and urinary tract cancer among people in seven counties of New

York state who drink chlorinated drinking water than in a matched group in the same counties drinking unchlorinated water. While this does not prove, of course, a cause and effect relationship, it is clear that chlorination of water should come under increasing scrutiny and other alternatives for disinfection should be sought. The concentration levels of representative halogenated hydrocarbons in the final effluent discharged to the Big Thompson River are much lower (by a factor of 5 to 10) than in the effluent to the Platte River from the Denver plant in which chlorine is used for disinfection rather than ozone. It should be mentioned that chlorine treatment of water also causes the formation of aromatic hydrocarbons. After bubbling chlorine gas through wastewater from Denver, we have found that toluene, ethylbenzene, and *o*-, *m*-, and *p*-xylene are present in the volatile fraction removed by sparging. These compounds are present in much greater concentrations than seen in blanks or in aliquots of the same samples prior to treatment with chlorine. We are presently trying to identify possible precursors of these aromatic hydrocarbons. Drinking water supplies should be examined for the presence of toluene, benzene, and other aromatic hydrocarbons.

The unidentified peaks in the chromatograms show mass spectra generally characteristic of hydrocarbons with carbon numbers from approximately 9 to 11. Only relatively small decreases are observed in the amounts of these after ozonization. It is possible that the apparent decrease in concentration of these components is caused in part by physical rather than chemical processes. Ozone in air is bubbled through the wastewater and may drive off some of the volatile components in a manner similar to that by which samples are spårged for analysis.

Samples taken at other times than those represented by Fig. 2 have been found to contain several other components at low levels. These include trichloroethylene, *n*-heptane, carbon tetrachloride, and one of the dichloroethane isomers.

In experiments designed to identify some of the less volatile components of UTSD wastewater samples, an interesting constituent was found. Caffeine was identified as one of the predominant components of an ether extract of a wastewater sample that had undergone secondary treatment but had not been ozonized. The data in Fig. 3 summarize the results of experiments in which caffeine in aqueous solution was ozonized in the laboratory rather than in the plant. Details of this work will be reported elsewhere[21].

In addition to the compounds shown in Fig. 3, at least six others, as yet unidentified, minor products have been found. It is not expected that these compounds are sufficiently volatile to be removed from solution by the headspace method. These laboratory experiments and those concerning ozonolysis of oleic acid, however, have shown the complexity of mixtures which may result from using a strong oxidizing agent like ozone as a chemical disinfectant in water treatment.

Quantitative measurements

The headspace sampling method used for these analyses has proven to be a highly reproducible means of collecting trace level volatile components from water. Concentration and analysis of head space samples from three aliquots of the same sample has shown that chromatographic peak heights for seven compounds measured are reproducible within ±8.2, 1.0, 9.7, 5.3, 0.6, 2.0 and 6.3 % (relative standard deviations).

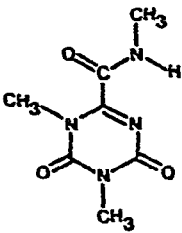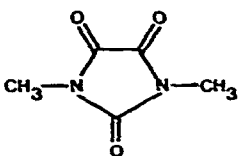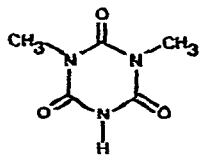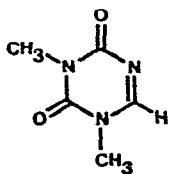Others[10,11,24] have reported the efficiency with which some organic compounds
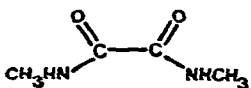
Compound	M. Wt.	Percent
1	198	33%
2	142	31%
3	157	9%
4	141	8%
5	116	1.5%

Fig. 3. Major products from the ozonolysis of caffeine in water.

are concentrated and recovered in systems similar to the one used in this work. Chloroform recoveries in the system used here are 40%, while those of *m*-xylene are 75%, toluene are 70%, *p*-dichlorobenzene are 60%, and *n*-heptanal are 30%. The concentrations of these compounds in representative UTSD wastewater samples after ozonization are 0.2 ppb of chloroform and 0.7 ppb of *n*-heptanal. In these samples the concentration of each of the volatile organic compounds was less than 1 ppb.

An examination also has been made of the reproducibility of retention time

data for several peaks in the chromatograms. It is important to establish the reliability of the method for routine analyses in which a sophisticated system such as GC–MS with its identifying capability might not be available. The least reproducible retention times occur early in the chromatograms at program temperatures near or below ambient temperature. In these cases, retention times are reproducible to within *ca.* ±5 % (relative standard deviation). At higher temperature the retention times are reproducible within ±1 % or less

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