

Effect of Ozonation and Chlorination on the Mutagenic Potential of Drinking Water

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A wide variety of chemicals with potential mutagenic/carcinogenic actions have been identified in drinking waters (HUPER and PAYNE 1963; SIMMON et al. 1977). Whereas some are present as such in untreated water and are not eliminated during purification processes, some others, like the halomethanes, may occur as reaction products between naturally occurring hydrocarbons and chlorine applied to the water (STEVENS et al. 1976; ROOK 1974; BELLAR et al. 1974).

As an alternative to chlorine, ozone has been utilized for disinfection and color-removal of drinking water, particularly in Europe, but the effect of ozonation on the carcinogenic potential of various contaminants of drinking water is poorly defined. Some laboratory studies have determined the effect of ozone on pure chemicals in aqueous systems through the use of rapid in vitro microbiological bioassays. In one of these papers among 28 compounds that were analyzed only a few indicated some levels of mutagenic activity (COTRUVO et al. 1977). In two other studies mutagenic pesticides, aflatoxin B₁, some alkylating agents and aromatic amines were inactivated by ozone, whereas other chemicals like dimethylhydrazine were converted into stable mutagens (BURLESON et al. 1979; CANFIELD et al. 1979).

Although several authors have investigated the mutagenic activity of organic compounds present in drinking water with microbiological bioassays (LOPEZ et al. 1978; SCHWARTZ and SAXEMA 1979) no data were available about the mutagenic potential of water taken at various stages of a treatment plant which included both chlorination and ozonation.

We report in this paper some results that were obtained by testing by Salmonella/microsomes test non-volatile organic compounds of water of the treatment plant of Florence, Italy.

METHODS

The treatment plant, that supplies 90% of tap water of the town of Florence (500,000 pop.) uses water from the Arno River, outside and uphill from the town. Treatment capacity is $4 \text{ m}^3/\text{sec}$. The water is polluted mainly by domestic waste, industrial activities being rare in this uphill region. Although the River has irregular flow and therefore varying concentrations of pollutants, some average values of untreated water will be presented in what follows: turbidity 10-800 N.T.U.; pH 7.2-8.8; temperature $1-30^\circ \text{C}$.; anionic surfactants $0-0.5 \text{ g/m}^3$ L.A.S.; ammonia 0-2 mg/l; total coliform bacteria $10^3-10^6/100 \text{ ml}$.

The treatment scheme is the following: pre-chloration, addition of powdered activated carbon, coagulation and flocculation, decanting, rapid sand filtration, ozonation, and final chloration. Pre-chloration is made with 15% sodium hypochlorite, and the concentration of chlorine in the water varies from $2.5-7.5 \text{ g/m}^3$. Norit SA4 activated carbon is used at a final concentration of 15 g/m^3 . Flocculation-filtration is obtained with two different processes: in the first one (treatment A) decanting is obtained by means of a pulsating decanter in a mud bed, whereas in the second (treatment B) a Door static decanter is used. For flocculation aluminum polychloride is used up to a concentration of 30 g/m^3 . Sand filtration is carried out at a speed of $5 \text{ m}^3/\text{m}^2/\text{hr}$ with a sand bed 1 m thick. Ozonation is obtained with a plant of Dégremont Inc. in two steps, so that the final ozone concentration is of 0.4 mg/l, with a contact time of 8 min.

Water obtained at various stages of the treatment (untreated, after decanting with the pulsating decanter (treatment A) or with a Door static decanter (treatment B), after ozone) was passed through glass columns (1 x 50 cm) filled with 10 g (dry weight) of Amberlite XAD-2 resin. The resin, pre-washed with acetone/methanol/water, was kept in place by two glass

wool plugs. The upper plug was occasionally changed when the flow was reduced by partial clogging. One-hundred liters of water were passed through the columns at a flow rate of about 50 ml/min. The columns were then washed with 30 ml of bi-distilled water, dried with nitrogen, and eluted with 50 ml of methylene chloride and chloroform in sequence. After taking the solvents to dryness in a rotary evacuator, the non-volatile chemicals were re-suspended in DMSO. Variable amounts of DMSO, which will be referred to as water equivalent, were used for the determination of mutagenic activity.

Mutagenic activity was determined by means of the classic Salmonella/microsomes assay as described by Ames and co-workers (AMES et al. 1975), using Salmonella typhimurium strains TA1538 and TA100. Dose response curves, with and without metabolic activation, were obtained by plating different amounts of the DMSO concentrate. A least squares linear fit was calculated from dose response curves after detracting background mutation rate from each experimental point, and the theoretical mutagenic activity for 10 l of water equivalent was calculated. Each point was run in duplicate. The values of background mutation rate, expressed as means \pm S.D. were the following: 10.1 ± 6 and 21 ± 9 for strain TA1538 without and with S9 respectively; 80 ± 35 and 91 ± 46 for TA100 with and without S9 respectively. Blank values of mutagenic activity were obtained by passing distilled water through the columns and by applying the procedure just described. No mutagenic activity was measured after evaporation of the solvents.

RESULTS

Figure 1 shows mutagenic activity determinations obtained by testing water samples at different stages of the treatment plant. It is apparent that untreated water had a negligible mutagenic activity whereas significant, although low, mutagenic activity was induced by chlorination, both in treatment A and treatment B. It is interesting to note that the mutagens observed did not need metabolic activation to exert their genotoxic effect. The water after ozone treatment, on the contrary, showed some mutagenic activity

but considerably less than the chlorinated one.

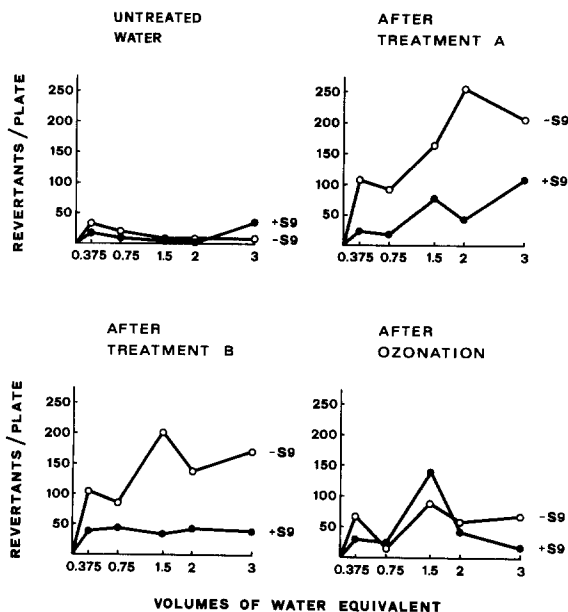


Figure 1. Mutagenic activity of various volumes of water obtained at different stages of the treatment plant. Tester strain: TA100.

Other results, obtained in different periods of the year, are shown in Fig.2, for strain TA1538 (top) and for strain TA100 (bottom). Untreated water did not show appreciable mutagenic activity, with the exception of experiment 1 in which some activity was observed with strain TA1538. In all other cases chlorination induced an increase of mutagenic activity, which was evident with both strains, with and without metabolic activation.

Although ozone did induce some increase of mutagenic activity over the levels observed with untreated water, when compared with the values of mutagenic activity observed after chlorination they were generally lower, with the exception of experiment 2 on strain TA100 with metabolic activation.

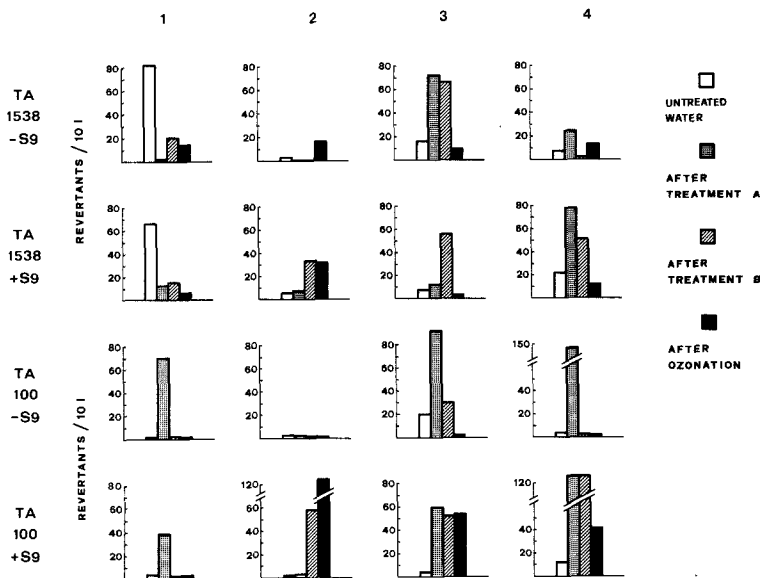


Figure 2. Mutagenic activity of 10 l of water at different stages of the treatment plant, determined with strain TA1538 and TA100, with and without metabolic activation. The water samples were obtained in the following months of 1980: 1=March, 2=June, 3=July, 4=October.

DISCUSSION

Ozone treatment may represent an alternative to chlorination for disinfection and odor removal in water purification processes. Chlorination is known in fact to cause the formation of carcinogenic halogenated hydrocarbons including chloroform and mutagenic chloramines (SCHICH and LEDERBERG 1976). Although the tests carried out so far by ozonating chemicals in controlled laboratory experiments do often show a decrease of the mutagenic activity, the reverse has been observed in some cases. The observations that we report indicate that in the treatment plant that was the object of the study, mutagenic chemicals were produced by chlorination and to a much lesser extent by ozone treatment. On the basis of this information, which is limited to a few preliminary measurements, ozonation seems a reasonable alternative to chlorination, with

the aim of minimizing the exposure to mutagenic chemicals in drinking water.

We have also to underline that the mutagenic potential of the untreated water of this plant was quite low, much lower than that reported by others in different locations in the U.S. (SCHWARTZ and SAXEMA 1979). The effect of ozonation on heavily contaminated waters remains to be determined.

We suggest the use of short-term mutagenesis procedures to assess the safety of alternative water treatment processes.

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