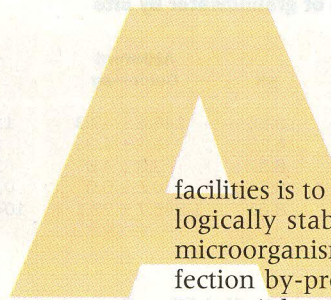




Biological stability of groundwater

A conventional treatment train with sand-anthracite columns provided better biological stability to finished water than a conventional train with GAC or membrane filtration.

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An important goal of water treatment facilities is to produce biologically stable water. Biologically stable water minimizes the regrowth of microorganisms and reduces the quantity of disinfection by-products in water distribution systems.¹ Bacterial regrowth is minimized because heterotrophic bacteria do not have a source of biodegradable carbon to oxidize. Disinfection by-products are reduced because there is a paucity of organic compounds to react with the disinfectant(s).^{2,3}

Water that is classified as biologically unstable promotes the growth of microorganisms, which in turn affects water quality and causes taste, odor, color, and turbidity problems.¹ Another adverse effect of unstable water is the growth of microorganisms and opportunistic pathogens that increase the heterotrophic plate counts (HPC) and potentially increase the coliform counts, causing noncompliance problems with

Conventional (e.g., coagulation, flocculation, and filtration) or membrane filtration treatment trains were used to remove organic compounds from groundwater. For the conventional train with sand-anthracite columns, the assimilable organic carbon (AOC) of the groundwater was reduced from $349 \pm 127 \mu\text{g/L C}$ to $54 \pm 51 \mu\text{g/L C}$. For the membrane filtration train, there was no statistical difference between the AOC of the raw water influent ($388 \pm 126 \mu\text{g C}$) and that of the membrane permeate ($334 \pm 156 \mu\text{g/L C}$), suggesting that this treatment produced biologically unstable water. Similar results were obtained using the heterotrophic growth response (HGR) method. Comparison of the biostability methods showed that HGR was positively correlated with AOC ($r = 0.52$; $P < 0.0001$; $n = 156$), indicating that AOC only partially explains the ability of heterotrophic bacteria to grow in water samples.

FIGURE 1 Schematic diagram of treatment trains and sampling sites

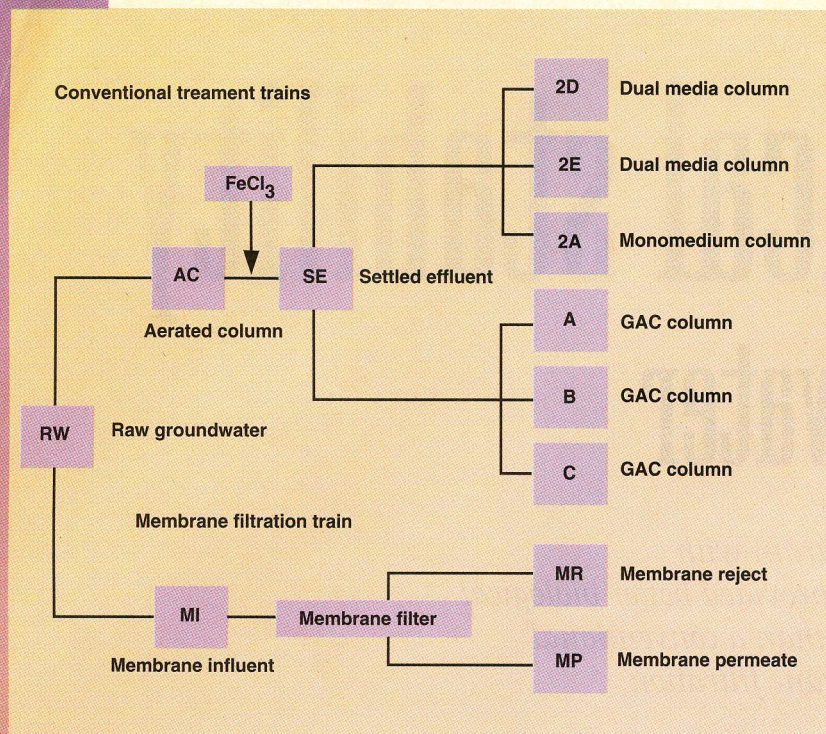


TABLE 1 Physicochemical properties of groundwater by site

Site	Turbidity ntu	pH	Apparent Color—cu	TOC mg/L
Raw groundwater	0.76 ± 0.08	8.85	146.8 ± 40.3	11.0 ± 0.1
Sand—anthracite columns	0.20 ± 0.06	6.16	6.2 ± 4.2	1.6 ± 0.3
GAC columns	0.20 ± 0.17	6.57	3.8 ± 4.6	1.3 ± 0.1
Membrane permeate	0.18 ± 0.05	8.77	0.0 ± 0.0	0.63 ± 0.1
Membrane reject effluent	3.97 ± 0.02	8.66	1,545.7 ± 22.1	104.2 ± 11.1

state regulatory agencies.² Water with assimilable organic carbon (AOC) levels exceeding 50 µg/L C can be associated with increased incidence of coliform bacteria⁴ and therefore may pose a threat to public health. Bacterial growth that is fostered by the biological instability of water accelerates pipe corrosion,² leading to pipe deterioration and metal solubilization. Given the problems associated with biologically unstable water, treatment facilities require dependable methods to evaluate the quality of their finished water.

Several methods have been proposed to determine the biological stability of water.³ The major philosophical difference among these methods is the inoculum source. For example, whereas the AOC method is based on the growth of *Pseudomonas fluorescens* strain P17⁵ and *Spirillum* strain NOX,⁶ heterotrophic growth response (HGR)⁷ and biodegradable dissolved organic carbon (BDOC)⁸ methods are based on the growth of naturally seeded bacteria. The advantage of using specific bacterial strains to determine AOC is that these strains can be used as standards for all types of water samples. It is difficult to

compare measurements that depend on the growth of indigenous bacteria because the inoculum differs from one sample to the next.

Another important difference is that the AOC method is based on N_{max} of P17 and NOX in a water sample, whereas the HGR method is based on the difference between N_{max} and $N_{initial}$ of heterotrophic bacteria. The BDOC method provides information on the fraction of organic carbon that can be metabolized by bacteria within a period of a few days to a few months.⁸ The AOC method differs from the BDOC method in that easily available organic carbon (>0.1 mg/L) is assayed over a shorter time frame.

This study focused on examining the biological stability of groundwater processed by conventional and membrane filtration methods and investigating the relationship between AOC and HGR. The groundwater source was a deep aquifer (1,640–3,281 ft [500–1000 m]) located in Southern California. Water was pumped from the source to a pilot plant where it was treated by either conventional (aeration, coagulation, and

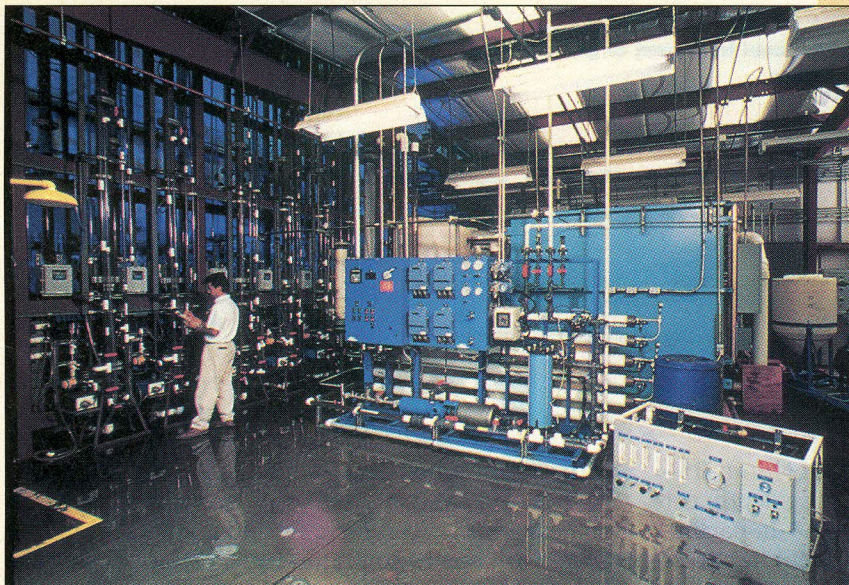
flocculation) or membrane filtration trains. High-molecular-weight humic compounds, which give the water a brown tinge, were removed by these treatments. Because different sites along the treatment trains varied in the amounts and kinds of biodegradable organic carbon as a function of the treatment process, the pilot plant provided a unique opportunity to monitor the biostability of groundwater.

Materials and methods

Bacterial strains and preparation of inocula.

Cultures of P17 and NOX* were streaked on R2A medium to ensure purity. A single colony of P17 and NOX was used to inoculate 100 mL of sodium acetate and sodium oxalate solutions, respectively. Inoculated solutions were incubated at 23°C until the organisms reached a stationary growth phase as determined by viable counts using the spread plate technique.⁹ The solutions were then kept in the dark at 4°C. Purity of the stock solutions was determined with every assay.

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Water samples were collected from different sites along the conventional and membrane treatment trains at the Irvine Ranch Water District pilot plant.

Media and solutions. The sodium acetate solution consisted of 0.341 g/L anhydrous sodium acetate (100 mg/L acetate-C). The sodium oxalate solution consisted of 0.526 g/L sodium oxalate (100 mg/L oxalate-C). Both solutions were autoclaved at 115°C for 15 min and stored in 1-L Wheaton bottles at 4°C.

Mineral salts stock solution consisted of 0.171 g K_2HPO_4 , 0.767 g NH_4Cl , and 1.444 g KNO_3 in 100 mL of AOC-free water. The stock solution was diluted 1:10 and pasteurized at 67°C for 1 h. One mL of the diluent was added to 500 mL of sample.

R2A* plates were prepared following the manufacturer's specifications. Phosphate-buffered water used for diluting P17, NOX, and heterotrophic bacteria was prepared following *Standard Methods*.⁹

All distilled water was deionized and double-distilled using a 6-L automatic distiller.† The distilled water stored in a 20-L glass container was referred to as AOC-free. All stock solutions and dilution buffers were prepared using this water.

Preparation of bioassay vessels and other glassware. Glassware was cleaned using the acid-wash method of van der Kooji et al.⁵ Glassware cleaned by this method was referred to as AOC-free glassware. Plastic caps from the collection bottles were washed with laboratory detergent,‡ soaked in distilled water for 1 h, rinsed six times with distilled water, and then dry-autoclaved (121°C for 30 min). The AOC assay was conducted using 1-L Erlenmeyer flasks. The HGR assay was conducted using 2-L sample bottles.

To ensure that the AOC-free glassware was free of organic carbon, the authors evaluated the washing procedures. Six acid-washed flasks were filled with 500 mL of 10 g/L yeast extract and incubated for 12 h at 23°C. The flasks were then rinsed three times with sterile AOC-free water. Two flasks were cleaned by acid-washing followed by baking at 300°C for 8 h, two flasks were just baked for 8 h, and the remaining two flasks served as controls. The different clean-

ing procedures were evaluated by determining the N_{max} of P17 for each set of flasks. The experiment was replicated three times.

Enumeration of the bioassay organisms. Viable plate counts were determined by serially diluting the sample (if required) with sterile phosphate buffer.⁹ One hundred μ L of each diluent was spread on an R2A plate using a sterile glass rod. Each series of dilutions was plated in triplicate. The spread

plates were inverted and incubated at 23°C for two days for P17, five days for NOX, and seven days for HPC. The dilution containing between 30 and 300 colonies was selected for enumeration.

Operating conditions and water sample collection sites. A schematic diagram of the treatment trains is shown in Figure 1. The coagulant dosage for the conventional treatment train was 120 mg/L $FeCl_3$. The hydraulic loading rate on the filters was 8 gpm/sq ft (5.4 mm/s). A flow feed of 40 gpm (2.5 L/s) was employed. For the membrane treatment train, the recovery rate was set at 90 percent. A flux of 20 gpd/sq ft (0.009 mm/s) and a feed pressure of 90 psi (621 kPa) were used. Approximately 1.5 mg/L of antiscalant foulant was used.

Water samples from different sites along the conventional and membrane treatment trains were collected between the months of July 1993 and July 1994. Collection bottles consisted of 2-L heat-resistant glass AOC-free bottles. Water samples were kept in ice until they were processed (usually within 6 h) in the laboratory.

The following sites were sampled along the conventional treatment train with sand-anthracite columns: raw groundwater (RW); aerated water column (AC); settled effluent (SE); dual-media column 2D (0.8-mm-diameter sand, 26.4-cm packing depth and 1.6-mm diameter anthracite, 2.4-m packing depth), column 2E (0.6-mm-diameter sand, 26.4-cm packing depth and 1.2-mm-diameter anthracite, 1.3-m packing depth), and monomedium column 2A (1.2-mm-diameter anthracite, 2.4-m packing depth) (Figure 1). The conventional treatment train with granular activated carbon (GAC) columns had the following sample sites: RW, AC, SE, and three various-sized GAC columns (column A§—0.9-mm

*Difco Laboratories, Detroit, Mich.

†Megapure distilling apparatus, Corning Inc., New York, N.Y.

‡KWIP, Santa Monica, Calif.

§Filtratorb 300, Calgon Corp., Pittsburgh, Pa.

