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Water Research 39 (2005) 4899–4912

WATER RESEARCH

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Seasonal occurrence and degradation of 2-methylisoborneol in water supply reservoirs

Paul Westerhoff[a,*], M. Rodriguez-Hernandez[a], Larry Baker[b], Milton Sommerfeld[c]

[a]*Department of Civil and Environmental Engineering, Arizona State University, Box 5306, Tempe, AZ 85287-5306, USA*
[b]*Minnesota Water Resources Center, 173 McNeal Hall, 1985 Buford Avenue, St. Paul, MN 55108, USA*
[c]*Plant Biology, School of Life Sciences, Arizona State University, Box 1701, Tempe, AZ 85287-1701, USA*

Received 25 August 2004; received in revised form 28 June 2005; accepted 29 June 2005

Abstract

Methylisoborneol (MIB) and geosmin are cyanobacterial metabolites that occur at nanograms per liter levels in surface water supplies and are responsible for many taste and odor complaints about the aesthetics of drinking water. This study evaluated three water supply reservoirs with bottom-release (hypolimnion) outlet structures in Arizona. MIB concentrations were always higher than geosmin concentrations, but both followed similar seasonal trends. MIB concentrations increased from spring to late summer, and stratified vertically with depth in the water column; the highest concentrations were always in the upper 10 m of the water column. Thermal destratification in the autumn increased MIB concentrations released from the outlet of reservoirs and impacted downstream utilities for several months. By winter of each year MIB concentrations were non-detectable. Mass balance analyses on MIB indicated that in-reservoir reactions were more important in changing MIB concentrations than conservative hydraulic "flushing" of the reservoir. Maximum net loss rates for MIB in the field ($R_{F,max}$) were on the order of 0.23–1.7 ng/L-day, and biodegradation appeared more important than volatilization, photolysis or adsorption. Using lake water in laboratory experiments, bacterial biodegradation rates (R_L) ranged from 0.5–1 ng/L-day and were comparable to $R_{F,max}$ values. Based upon these rates, MIB concentrations in a reservoir would decrease by approximately 30 ng/L over a period of 1 month. This was the magnitude change in MIB concentrations commonly observed after autumn thermal destratification of the reservoirs.

Keywords: Cyanobacteria; Biodegradation; Reservoir; Taste and odor; MIB; Drinking water

1. Introduction

Many cyanobacteria (blue-green algae) produce intracellular and extracellular metabolites, such as biotoxins and/or taste and odor (T&O) compounds, impact water supplies (e.g., 2-methylisoborneol (MIB), trans-1,10-dimethyl-trans-9-decalol (geosmin)) (Carmichael, 1997; Gerber, 1979; Juttner, 1995; Kaas and Henriksen, 2000; Suffet et al., 1999). Planktonic and periphytic cyanobacteria, as well as some actinomycetes, produce MIB and geosmin in reservoirs, rivers, canals, and within water treatment plants (WTPs) (Gerber, 1965; Izaguirre and Taylor, 1995; Suffet et al., 1995). The

*Corresponding author. Fax: +1 602 965 0557.
E-mail addresses: p.westerhoff@asu.edu (P. Westerhoff), baker127@umn.edu (L. Baker), milton.sommerfeld@asu.edu (M. Sommerfeld).

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doi:10.1016/j.watres.2005.06.038

biological function of these algal metabolites is unknown, although they may be intermediates or by-products of pigment production (Bafford et al., 1993; Zimba et al., 1999). MIB and geosmin are of particular interest because they are unpalatable, imparting earthy/musty/moldy tastes and odors to drinking water. MIB and geosmin odors in drinking water are noticeable at concentrations of 2–10 ng/L (Wnorowski, 1992). A quarter to half of US all WTPs using surface water report problems with T&O compounds related to algae metabolites (Suffet et al., 1996). In a US nationwide survey of drinking water consumers, 52% of respondents expressed concern about "smell or taste" of water (WQA, 2001). In a similar survey of five southwestern cities, 60% of respondents were "somewhat concerned" or "very concerned" about the taste and smell of public water supplies (Baker and Wolf, 2003).

Treatment plants generally accomplish T&O compound removal by carbon adsorption or ozonation (Bruce et al., 2002; Lawton et al., 1998). These treatment processes greatly increase the costs of water treatment. Consequently, it is desirable to mitigate the occurrence of MIB and geosmin at their sources (Izaguirre and Taylor, 1995; Means and McGuire, 1986). MIB and geosmin often accumulate in surface water reservoirs, so knowledge regarding production and degradation of these compounds is valuable for developing management strategies to reduce their concentrations in upstream reservoirs before water arrives at treatment plants.

Some soil and aquatic bacteria are capable of biodegrading MIB and geosmin. The Cam Operon includes the primary genes responsible for biodegradation of these alcohols (Hoehn, 1965; Izaguirre and Taylor, 1998; Izaguirre et al., 1999; Oikawa et al., 1995; Trudgill, 1990). Although the kinetics of algal metabolite biodegradation in biological treatment systems have been evaluated and modeled (Nerenberg et al., 2000; Rittmann, 1995), very little is known about MIB or geosmin degradation in water supply reservoirs.

The goal of this paper is to better understand the significance of mechanisms responsible for affecting MIB and geosmin concentrations in water supply reservoirs. Seasonal T&O concentrations plus hydrological, chemical, and biological data were collected over a 3-year period for three water supply reservoirs near Phoenix, AZ. Mass balance analyses of T&O compounds in the reservoirs were used to calculate in situ field rates of net MIB production and loss. Laboratory experiments confirmed MIB and geosmin biodegradation using native lake organisms, and data were used to estimate biodegradation rates. The significance in changing MIB or geosmin concentrations within the reservoirs was addressed by comparing conservative mixing mechanisms (destratification, flushing water through the reservoir) and reaction mechanisms (production, biodegradation, volatilization, photolysis, sorption).

2. Methods and analyses

2.1. Site descriptions

The three reservoirs studied are major components of the water supply system for nearly 3 million inhabitants of the Phoenix, AZ, metropolitan area. The reservoirs are located in the semi-arid Sonoran desert within 50 km of the metropolitan area. The reservoirs range from oligotrophic to mesotrophic (Table 1). During thermal stratification the epilimnion is usually 5–15 m deep and accounts for 30–60% of the total reservoir volume. Important differences in reservoir operation may affect both the production and degradation of MIB and geosmin and the release of these compounds to downstream WTPs. The reservoirs supply water to approximately 15 WTPs, several major industries, and many agricultural users; water travels to these users via open, concrete-lined canals. A site location map for the reservoirs and other water supply infrastructure information is available elsewhere (Bruce et al., 2002).

Bartlett Lake is an on-stream reservoir on the Verde River located 3 km downstream of Horseshoe Lake. Horseshoe Lake was at less than 20% capacity during the study period. Most water enters the reservoir following snowmelt at higher elevations between February and May. Water is stored throughout the summer and released from October to April. Bartlett Lake as a single outlet near the bottom (hypolimnetic withdrawal) located at the downstream (i.e., outlet) end of the lake. Interactions between hydrology and dissolved organic carbon (DOC) in Bartlett Lake have been studied previously (Nguyen et al., 2002; Parks and Baker, 1997).

Saguaro Lake is the lowest of five reservoirs on the Salt River. Three of the upstream Salt River reservoirs have hydropower generation facilities, and pump-back piping from Saguaro to upstream reservoirs occurs during summer months to increase hydropower revenue: water released from upstream reservoirs during peak demand periods (daytime) is pumped back to the upstream reservoirs upstream during off-peak periods (nighttime). This operational mode results in a very short reservoir hydraulic residence time (HRT = outflow/volume = ~0.25 years). Saguaro Lake typically remains near capacity; very little (2–4 m) variation in surface elevation occurs throughout the year. Saguaro Lake also has one outlet near the bottom of the reservoir at the downstream end of the lake. Most releases occur in the summer ($>8.5\,m^3/s$) and cease around early October, with minimal flow ($\sim 0.1\,m^3/s$) between October and April.

Lake Pleasant is an off-stream water supply reservoir located on the Agua Fria river near the Central Arizona Project (CAP) canal in the Phoenix metropolitan area. Water pumped from Lake Havasu on the Colorado River travels to Lake Pleasant through an open, concrete-lined CAP canal approximately 200 km in

Table 1
Hydrologic and biological characteristics of three reservoirs at maximum pool elevations and hydrologic data during study period (1999–2002)

	Bartlett Lake	Saguaro Lake	Lake Pleasant
Surface area (km^2)	11.2	5.1	40.3
Volume (m^3)	2.2×10^8	0.85×10^8	10.5×10^8
Depth (m)	38	33	43
Surface elevation (m)	532	466	519
Hydrology during study			
Average annual inflow (m^3)	2.8×10^8	3.7×10^8	7.2×10^8
Average annual outflow (m^3)	2.6×10^8	3.6×10^8	7.0×10^8
Average HRT (years)	0.38	0.22	1.2
Average Secchi disk transparency (m)[a]	2.1	3.1	4.1
	[0.7–3.2]	[1.6–5.9]	[1.3–8.6]
Average chlorophyll-*a*	4.0	5.8	2.6
concentration (μg/L)[a]	[0.3–14]	[1–14]	[0.1–6]
Average total P (μg/L)[a]	19	19	8.5
	[2–93]	[3–85]	[0.6–40]
Average total N (mg/L)[a]	0.26	0.34	0.31
	[0.1–0.6]	[0.2–0.8]	[0.2–0.4]

[a]Range of observed values are included in brackets.

length. Lake Pleasant has two dual inflow/outflow release gates near the dam; they are located ~30 m apart within a single structure. Water pumped from the CAP canal fills Lake Pleasant from October to April of each year, and releases of Lake Pleasant water into the CAP canal occur between April and October. Downstream water demands are highest during the summer period. Natural drainage from the Agua Fria River supplies a small amount of additional inflow to Lake Pleasant. This was less than 5% of the total reservoir inflow during this study.

Daily inflow, outflow, and change in storage data were provided by Salt River Project (Bartlett Lake and Saguaro Lake) and CAP (Lake Pleasant). Storage volume versus depth relationships were used to compute reservoir volumes from daily elevation data.

2.2. Reservoir sampling and mass loading analysis

Monthly grab samples were collected at a depth of 1 m below the water surface from the reservoir inflows and outflows. Reservoir samples were collected using a Kemmerer sampler near the dam outlets, which correspond to the deepest part of the reservoir. Additional spatial samples were collected less frequently along transects from the dam to the inlet or in shallower coves. Discrete depth samples were collected at 5 m intervals. During stratification, discrete samples were combined (stainless steel container) into ''epilimnetic'' or ''hypolimnetic'' samples using thermal profiles as a guide. During non-stratified periods, discrete samples generally were collected throughout the water column. On several occasions, discrete samples were analyzed separately (not composited) to elucidate the fine detail of vertical profiles. Temperature, pH and dissolved oxygen (DO) profiles were measured in the field using a YSI Model 50B DO meter and YSI Model 60 pH meter (Yellow Springs, OH). Secchi disk (SD) (20 cm diameter) measurements were also taken during sampling events. Samples for MIB and geosmin analyses were stored in the dark at 4 °C in cleaned and ashed (550 °C for > 2 h) amber glass bottles until analysis. Samples collected for chlorophyll analysis were immediately filtered upon returning to the laboratory.

Based upon measured parameters, the field reaction rate for MIB for the entire reservoir (R_F) was calculated using the following equation:

$$\frac{dM}{dt} = Q_{in}C_{in} - Q_{out}C_{out} + \forall R_F, \quad (1)$$

where M is the mass of MIB (kg) in the reservoir (hypolimnion plus epilimnion); C_{in} and C_{out} are the MIB concentrations (kg/m^3) of water entering and leaving the reservoir, respectively; Q_{in} and Q_{out} are the flowrates (m^3/day); $\forall$ is the monthly reservoir volume (m^3); and t is time (days). A positive R_F value indicates net MIB production, and a negative value indicates net MIB loss.

2.3. Laboratory degradation experiments

Water samples from Saguaro Lake were collected at 5 m vertical depth increments, transported to the laboratory, and immediately filtered (ashed Whatman GF/F). Batch MIB and geosmin degradation experiments were conducted in 125 mL amber glass bottles incubated at 22 °C in the dark for up to 35 days. Control

samples were obtained by adding a biocide (100 mg/L sodium azide) to the samples; results were consistent with controls held in the dark at 4 °C. Geosmin (CAS#19700-21-1, Waco Ind., Japan) was added in select experiments. Additional experiments using a biological seed were conducted to increase bacteria and nutrient levels. The biological seed consisted of hypolimnetic water collected from Saguaro Lake enriched with 100 mg/L (as the compound) each of sodium nitrate, sodium phosphate, and sodium acetate. The seed solution was incubated in the dark for 7 days prior to the initiation of degradation experiments. The bacterial population was not enumerated, but the culture became milky-cloudy by the end of the incubation. One milliliter of biological seed was added to 125 mL of sample in select experiments. Samples remained aerobic throughout the experiments. Sample bottles were sacrificed over the following 10–38 days; at least 15% of the bottles were duplicated.

2.4. Analysis

MIB (CAS #2371-42-8) and geosmin (CAS #19700-21-1) standards were purchased from Waco Ind. (Japan). MIB and geosmin were analyzed with a headspace solid phase microextraction (SPME) fiber (Supelco #57348U). Non-filter water samples (25 mL) were placed into a 45 mL septum-capped vial containing 3.0 g of desiccated sodium chloride. An internal standard, 2-isopropyl-3-methoxypyrazine (IPMP), was added to the sample at a concentration of 10 ng/L. The SPME fiber was introduced into the headspace of the vial through the septum and maintained at 50 °C for 30 min with constant stirring of the vial using an automated SPME device (CombiPAL, CTC Analytics, Switzerland). Analysis of MIB and geosmin was performed on a Varian Star 3400 CX gas chromatograph and mass spectrometer. Compounds from the fiber were desorbed in the gas chromatograph and eluted from a column (MDN-5 capillary column; Supelco, Pennsylvania) into the mass spectrometer for selective ion storage (Lloyd and Grimm, 1999; Watson et al., 1999, 2000). The method detection limit (MDL) for MIB and geosmin was 0.5 ng/L.

Chlorophyll-*a* was measured (Standard Method 10200 H) on filtered (Whatman GF/C) water samples following extraction with 10 mL methanol, with spectrophotometric detection of chlorophyll-*a* and correction for phaeopigments and turbidity (APHA, 1995). Phytoplankton were identified using an Olympus BH2 microscope with the aid of a 1000 × oil immersion objective using wet mounts of living material and burn mounts of diatoms. Enumeration of taxa was performed by centrifuging 50 mL of water samples at ~3000 times-gravity for 10 min, and aspirating the supernatant to reduce volume to 10 mL. One milliliter homogeneous aliquots were placed in a 1 mL Sedqwich-Rafter counting cell. A minimum of 500 organisms or specimens in 40 fields were counted. Enumeration occurred under a total magnification of 200 × using a Zeiss inverted microscope and recorded as cells per mL of natural sample.

3. Results

3.1. Reservoir stratification and algae productivity

All three reservoirs were thermally stratified at depths of 5–15 m, typically from April to September or October. Surface temperatures ranged from 10 to 30 °C throughout the year. The maximum temperature difference between the epilimnion and hypolimnion ranged from 12 to 15 °C for Bartlett and Pleasant lakes to 7 °C or less for Saguaro Lake (Fig. 1A). Fig. 2 shows a representative vertical profile for several water quality parameters. The DO profile reveals stratification in Saguaro lake more clearly than temperature.

The shallowest SD transparencies were observed in November and December (0.6–1.5 m) (Fig. 1B). The deepest SD transparencies occurred between January and June. Lake Pleasant generally had higher transparencies than Bartlett or Saguaro lakes (Table 1, Fig. 1). Chlorophyll-*a* concentrations were typically less than 10 μg/L and varied seasonally. The hypolimnion (water from >15 m depth) of some reservoirs contained chlorophyll-*a*, indicating cell settling or migration from the epilimnion. Planktonic algae counts typically ranged from <20 to a maximum of 5000 cells/mL, but they were typically 200–500 cells/mL. Although total plankton counts were relatively constant across the seasons, the major algal groups did exhibit seasonal patterns. Diatoms were typically most abundant during the cooler months (October–March), whereas cyanophytes were typically most abundant from mid-summer to autumn (July–November). Cyanobacteria algae counts were always at <300 cells/mL (Fig. 1C). Of the cyanobacteria observed, the genera *Anabaena* spp., *Oscillatoria* spp., *Phormidium* spp. and *Pseudanabaena* spp. can produce MIB and *Anabaena* spp. and *Oscillatoria* spp. can produce geosmin. Some algae (*Oscillatoria* and *Cryptomonads*) are adapted to low light and may thrive below the epilimnion. Only a few algae blooms were observed during the 3 year study of the reservoirs, based upon visual observation (green-colored turbid water) and/or high cell counts (>1000 cells/mL). However, reservoir samples were collected only once or twice per month, and other blooms could have occurred between these sampling intervals.

3.2. Seasonal T&O occurrence

MIB and geosmin followed similar temporal and geographic distribution trends within the reservoirs.

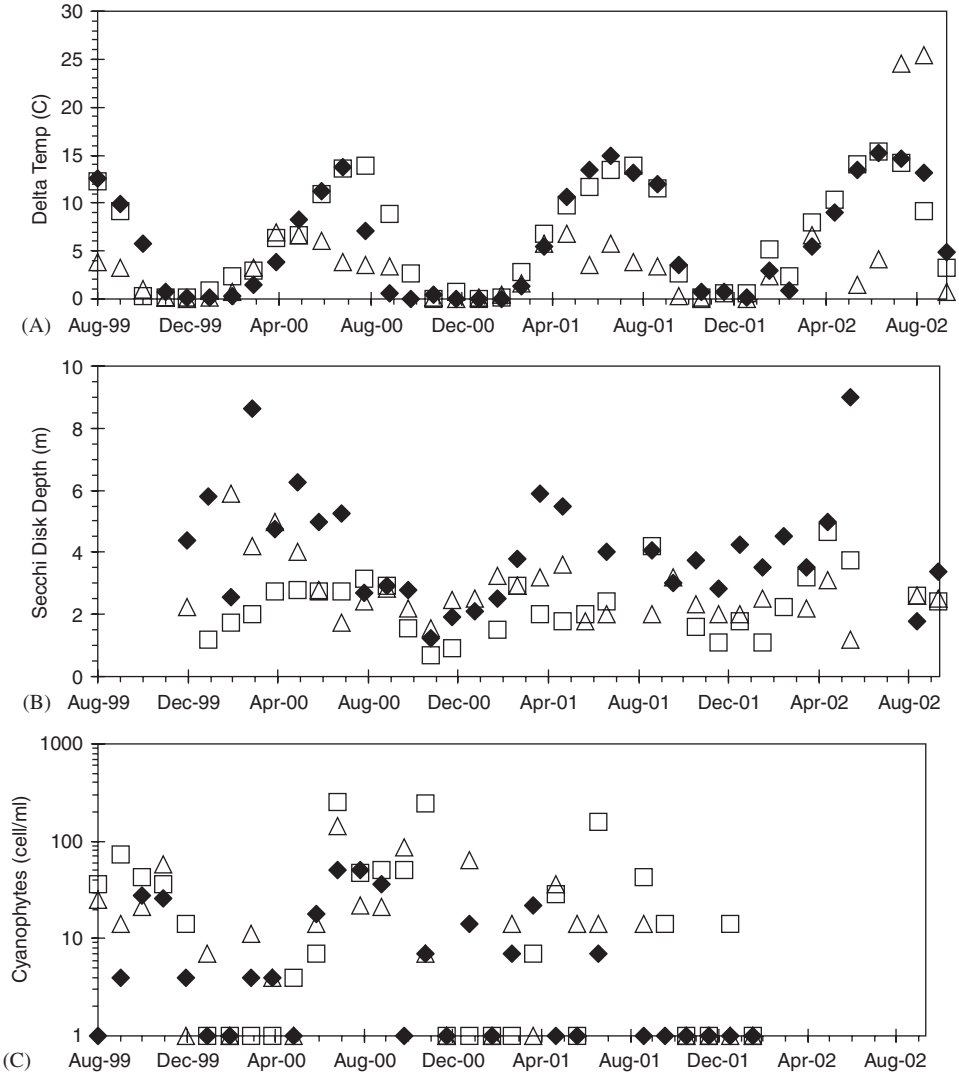

Fig. 1. Temporal patterns in hydrological and biological indicators for Bartlett Lake (□), Saguaro Lake (△), and Lake Pleasant (◆). Delta temperature represents difference between water temperature at depths of 5 and 30 m.

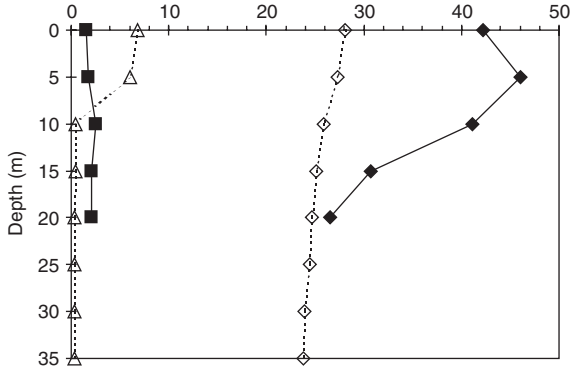

Fig. 2. Vertical profiles for MIB (◆, ng/L), geosmin (■, ng/L), dissolved oxygen (△, mg/L), and temperature (◇, °C) near the Saguaro Lake outlet (September 2000).

Since MIB concentrations were higher than geosmin concentrations for >95% of the samples ($n = 132$) from the three reservoirs, this paper focuses on MIB. All three lakes exhibited trends between MIB concentration and water temperature in the epilimnion (Fig. 3). Saguaro and Bartlett Lakes exhibited exponential relationships, while Lake Pleasant had intermediate MIB levels at low temperatures and several instances of low MIB at higher temperatures. MIB concentrations >5 ng/L rarely occurred at water temperatures less than 20 °C. Thus cyanobacteria appear more likely, but not necessarily, to be found at warmer water temperatures so MIB is also more likely to be observed at warmer temperatures. However, if cyanobacteria occur (e.g., blooms) when it is colder then cooler water temperatures do not appear to inhibit MIB production. During the sampling event

illustrated in Fig. 2, MIB and geosmin samples were collected to only a depth of 20 m. The maximum concentrations of MIB occurred at a depth of 5 m. Other, deeper, vertical profiles indicated a gradual decline in MIB and geosmin concentrations to the reservoir bottom, and suggested higher net production of MIB in the epilimnion than hypolimnion. Gradual production of MIB occurs in the epilimnion over time. MIB was usually below detection limits near the bottom of the reservoir. Together these data support a conclusion that MIB was produced in the epilimnion, probably by cyanobacteria algae but possibly by actinomycetes as well.

Temporal trends of MIB concentrations in the lakes are shown in Figs. 4 and 5. MIB concentrations generally increased from April to September of each year, with higher concentrations in the epilimnion than in the hypolimnion. Stratification periods (temperature differential from top to bottom $>4\,^{\circ}C$) are indicated in Figs. 4 and 5. MIB concentrations near the surface declined around the time of thermal destratification due to mixing with low-MIB water from deeper in the reservoir, resulting in uniform MIB concentrations throughout the water column. By January of each year MIB and geosmin concentrations were below detection limits throughout the water column. Overall, temperature appears to be a critical factor in MIB and geosmin production and distribution throughout the water column for these three Arizona reservoirs, although it

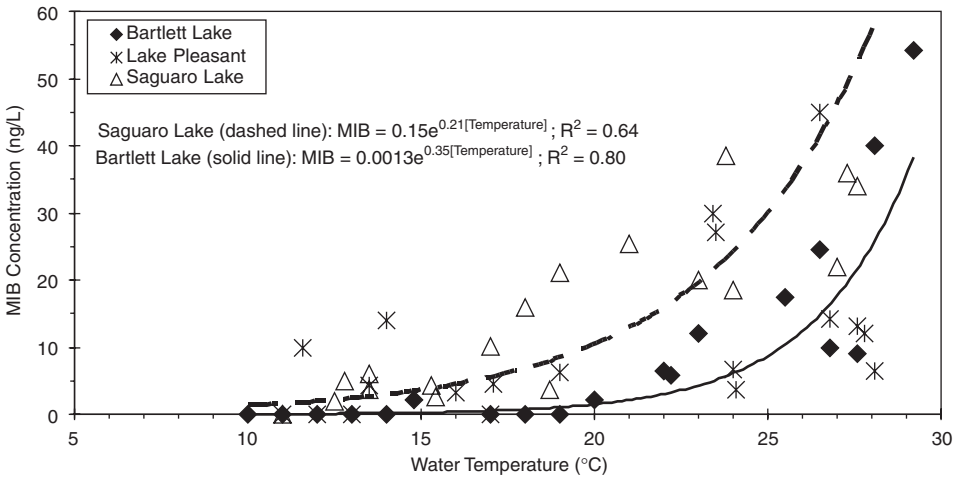

Fig. 3. Concentration of MIB in the upper 10 m of the water column in three lakes related to the water temperature at the same depth. Regression lines shown for Saguaro and Bartlett lakes.

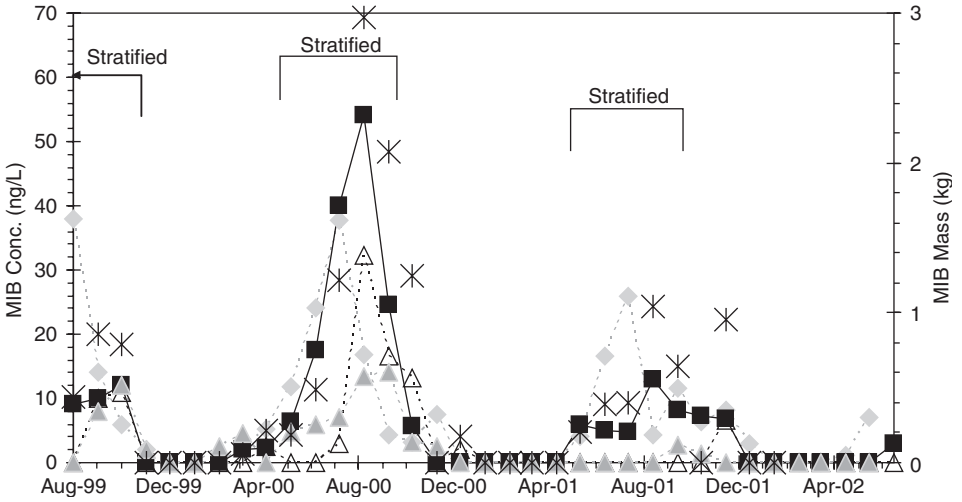

Fig. 4. Temporal trends in MIB concentrations at upstream (◆), downstream (▲), epilimnion (■), hypolimnion (△) sampling sites for Bartlett Lake. Total MIB mass in the reservoir (✳) is also indicated.

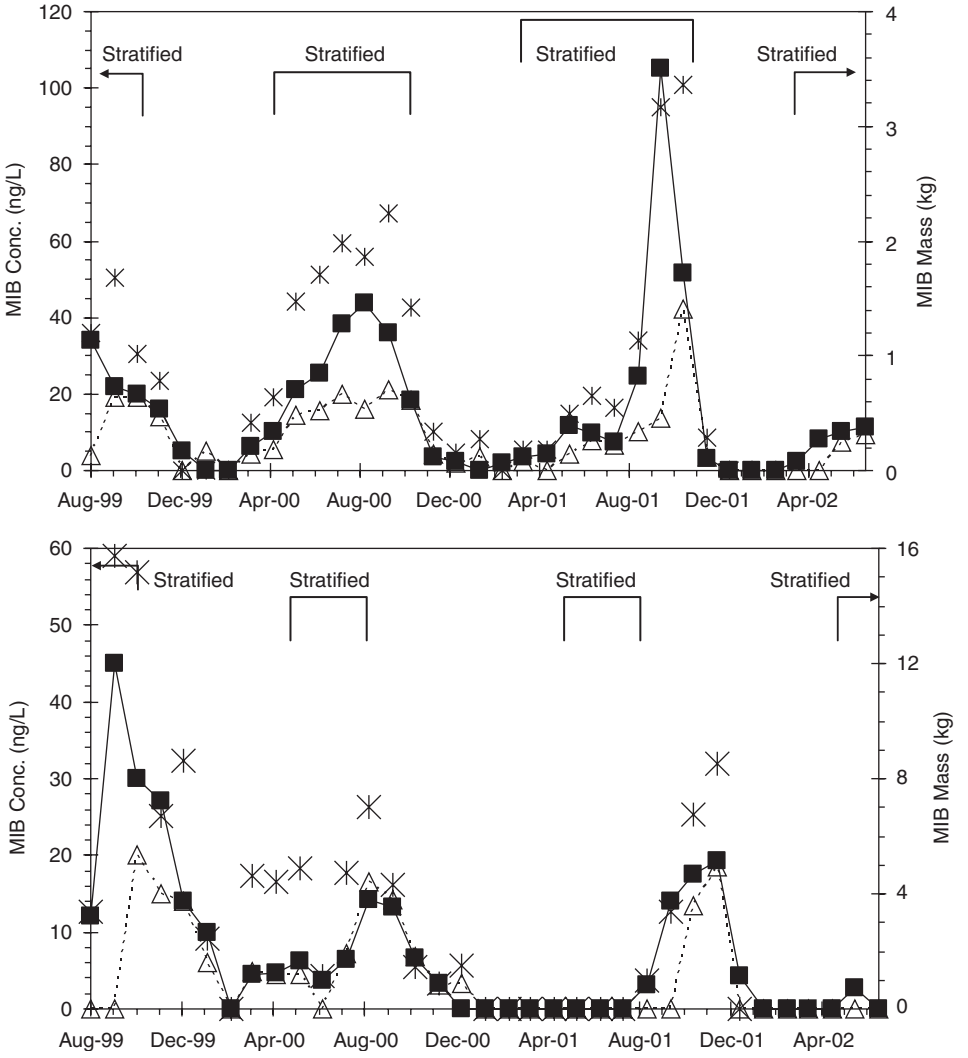

Fig. 5. Concentrations and masses of MIB in Saguaro Lake (upper) and Lake Pleasant (lower). Epilimnion (■) and hypolimnion (△) MIB concentrations are connected with lines, and the total MIB mass in the reservoir (✳) is also indicated.

is difficult to isolate just one mechanism (stratification, light availability, nutrient availability, etc.).

MIB concentrations in inflows and outflows for Saguaro Lake (Fig. 4) are typical of the other trends for the other two reservoirs. Inflow MIB concentrations frequently exceed epilimnion MIB concentrations within Saguaro Lake, and depending upon flowrates could represent a significant MIB source. Outflow from Saguaro Lake, via a bottom-release structure at the downstream end of the lake, exhibited low MIB concentrations throughout most of the year, except in the autumn following thermal destratification. This period of elevated MIB in the release constitutes the portion of the year when downstream utilities must remove MIB to improve the aesthetics of the drinking water.

3.3. Mass balance on T&O compounds in lakes

Mass balances for MIB were developed for each of the three reservoirs to estimate net MIB production or loss rates. Geosmin concentrations were not consistently high enough to conduct in-lake mass balances. The mass of MIB stored in the epilimnion and hypolimnion was computed for each month as the product of MIB concentration and associated volume. The mass of MIB in the lakes followed the same seasonal trend as MIB concentrations (Figs. 4 and 5) of increasing in the spring and rapidly decreasing in the autumn. Prior to autumn destratification the epilimnion contained most of the MIB mass.

Monthly MIB mass flow terms (into, out of, and reaction within the reservoir) are presented in Fig. 6 and

used in Eq. (1). The in-reservoir mass flow terms were calculated assuming MIB concentrations were geographically uniform throughout the two different "layers" of the reservoir that were sampled (0–10 m depth and 10 m to bottom). This assumption appears valid based upon relatively uniform MIB concentrations observed during intensive spatial sampling along transects and in coves. Inflow and outflow mass flow values deviate from "zero" in response to large inflows or reservoir releases (Fig. 6). Lake Pleasant has the largest volume and consequently high mass flow values. Positive values for mass flows indicate increasing MIB mass in reservoirs over a given time interval, whereas negative numbers represent decreasing MIB mass in the reservoirs. During many months the reaction term ($\forall R_F$) value is equal or greater than inflow or outflow mass flow rates which indicates significant in situ MIB production (positive value) or loss (negative value) depending upon the time of year. During some periods inflows account for equal or greater MIB mass increases than in situ net production of MIB.

The in situ reaction rate for MIB for the entire reservoir (R_F) was calculated based upon monthly MIB mass flowrates and monthly changes in reservoir volumes. R_F represents the difference between net MIB production and losses from biodegradation and other processes (discussed below). R_F values presented in Fig. 7 were typically positive during the spring and summer, reflecting MIB production by algae in the reservoirs. Negative R_F values indicate MIB losses. Net

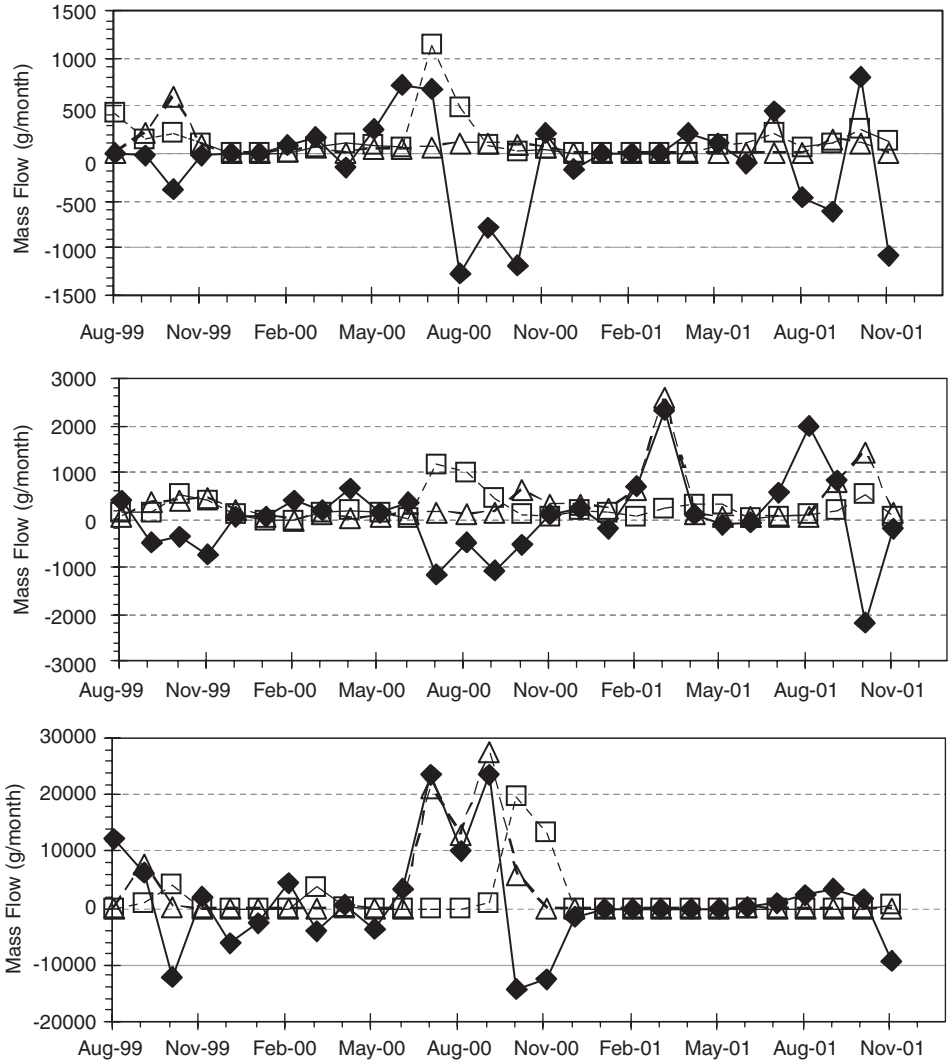

Fig. 6. Calculated MIB mass flow (g/month) into (□, $Q_{in}C_{in}$) and out of (△, $Q_{out}C_{out}$) of each reservoir, plus the mass flowrate reaction (♦, $\forall R_F$) for Bartlett Lake (top), Saguaro Lake (middle) and Lake Pleasant (bottom). All mass inflows and outflows are ⩾zero, while mass flowrates can be positive or negative and indicate either gain or loss of MIB, respectively.

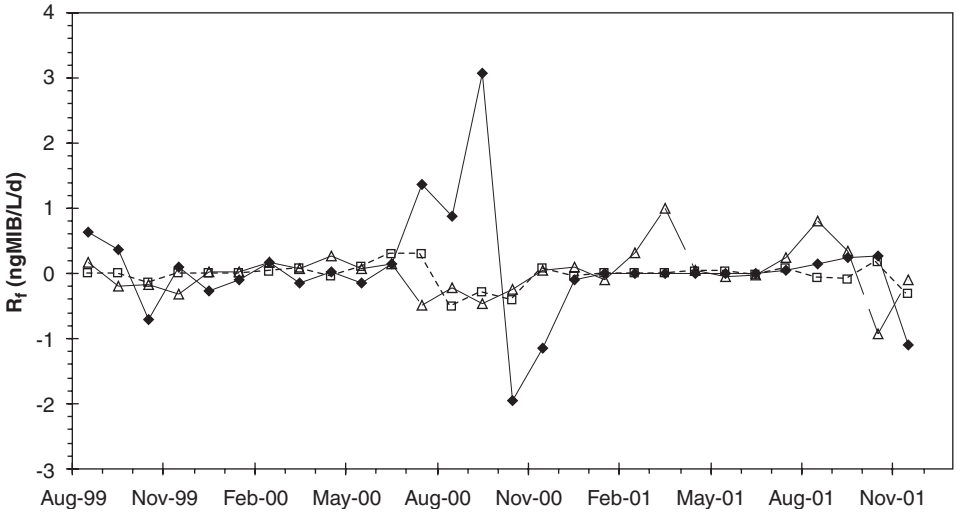

Fig. 7. Calculated R_F pseudo-zero-order rates (Eq. (1)) for MIB in Bartlett Lake (□), Saguaro Lake (△), and Lake Pleasant (♦). Positive (+) rates indicate production of MIB and negative rates (−) indicate a loss of MIB.

reaction rates (R_F values) ranged from –2 to 3 ng/L-day. R_F values were negative after thermal destratification; annual maximum loss rates ($R_{F,max}$) frequently occurred in October and November. Table 2 summarizes $R_{F,max}$ values.

3.4. Laboratory T&O compound degradation studies

MIB concentrations decreased during batch degradation studies (Fig. 8). Initial ambient MIB concentrations varied since samples were collected from different depths (corresponding to the vertical profile shown in Fig. 2). MIB concentrations remained nearly constant in the sodium-azide spiked controls or the 4 °C controls over the 36 days. MIB losses in the samples were attributed to biodegradation (see Section 4). A lag phase of about several days may have occurred for degradation in the epilimnetic samples (0 and 5 m), after which a nearly linear biodegradation rate was observed. Hypolimnetic samples with water from depths of 10, 15 and 20 m did not exhibit a lag phase and were characterized by the immediate onset of MIB biodegradation.

Addition of the biological seed to the 5 m epilimnetic sample shortened the lag phase for MIB degradation (Fig. 9A) and eliminated the lag phase for geosmin biodegradation (Fig. 9B). Pseudo-zero-order laboratory biodegradation rates (R_L, ng/L-day) were calculated based upon a linear regression of concentration versus time for each experiment, using only data recorded after the onset of observable degradation (post-lag phase for epilimnetic samples without the added bioseed). Linear regressions all had goodness of fit (R^2) values greater than 0.97. Representative linear fits of data are presented in Fig. 9. Rates for MIB biodegradation ranged from 0.8 to 1.2 ng/L-day with an average of 0.96 ± 0.15 ng/L-day ($n = 7$); these were statistically different ($\alpha < 0.05$) from the control experiments (Table 2). Geosmin

Table 2
Calculated pseudo-zero-order loss rates (ng/L-day) for MIB and geosmin from laboratory experiments (R_L), and annual maximum pseudo-zero-order loss rates (ng/L-day) of MIB from three reservoirs ($R_{F,max}$) for Autumn 1999 (†), Autumn 2000 (††), and Autumn 2001 (†††)

Source of data	MIB loss rate	Geosmin loss rate
Batch experiments (R_L *values*)		
Control ($MIB_0 = 46$ ng/L)	0.11	0.10
0 m ($MIB_0 = 42$ ng/L)	0.79	NA
5 m & duplicate ($MIB_0 = 46$ ng/L)	0.95 & 0.90	0.81
5 m + bioseed ($MIB_0 = 46$ ng/L)	1.1	0.90
10 m ($MIB_0 = 41$ ng/L)	0.80	NA
15 m ($MIB_0 = 31$ ng/L)	1.2	NA
20 m ($MIB_0 = 26$ ng/L)	1.0	NA
Field reservoirs ($R_{F,max}$ *values*)		
Bartlett Lake	$0.14^{\dagger}$, $0.51^{\dagger\dagger}$, $0.31^{\dagger\dagger\dagger}$	NA
Saguaro Lake	$0.31^{\dagger}$, $0.50^{\dagger\dagger}$, $0.90^{\dagger\dagger\dagger}$	NA
Lake Pleasant	$0.7^{\dagger}$, $2.0^{\dagger\dagger}$, $1.1^{\dagger\dagger\dagger}$	NA

NA = not available due to lack of data above geosmin detection limits. Initial MIB concentrations (MIB_0) for laboratory data are given in table; initial geosmin concentration was 22 ng/L.

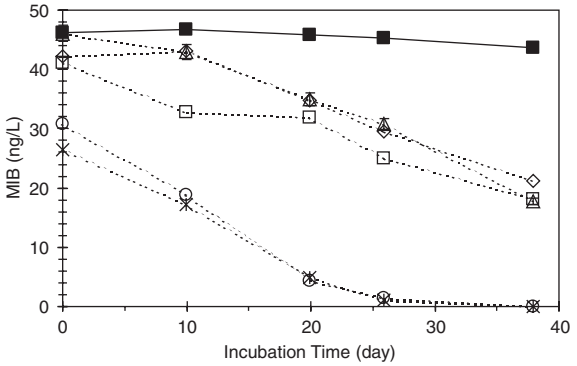

Fig. 8. Loss of MIB in batch experiments for samples collected from Saguaro Lake (September 2000; corresponds to samples presented in Fig. 2) from the surface (◇, 0 m) and at several depths (△, 5 m; □, 10 m; ○, 15 m; ✳, 20 m). Duplicate samples at 5 m (error bars show 1 std.dev.) and a sodium-azide spiked control sample (■) were conducted using water collected at a 5 m depth.

biodegradation rates were similar (Table 2). Over the naturally occurring concentration range considered, R_L values were statistically independent of initial MIB concentration ($\alpha = 0.05$). Initial sample collection depth and addition of a bioseed did not affect R_L values.

4. Discussion

4.1. Mechanisms for T&O compound loss in lakes

The loss of MIB mass between September and November may be due to several factors. The MIB mass balance analysis accounts for two important processes which would, therefore, not affect the MIB net reaction term (R_F): (1) thermal destratification of the reservoir with subsequent dilution of higher MIB concentrations in the upper parts of the reservoir by deeper water with lower MIB concentrations, and (2) flushing through of upstream water with lower MIB and geosmin and releases of reservoir water. R_F includes both MIB production and at least four loss mechanisms: (1) volatilization, (2) photolysis, (3) sorption, and/or (4) MIB biodegradation.

Volatilization of MIB ($H_c = 5.76 \times 10^{-5}$ atm m^3/mole) and geosmin ($H_c = 6.66 \times 10^{-5}$ atm m^3/mole) from reservoirs can be related to the compounds' Henry's constants (H_c), depth of water in the reservoir, and average wind speed. Using a 10 m depth and an average wind speed of 0.5 m/s, the half-life of MIB or geosmin due to volatilization would be approximately 1 year, too long to be important in the context of the observed changes in total (or upper 10 m) MIB mass in the reservoirs.

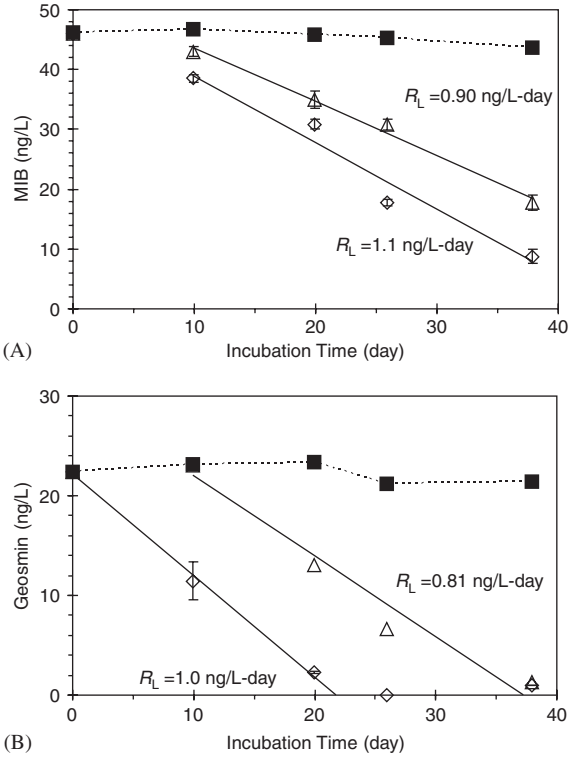

Fig. 9. Loss of MIB (A) and geosmin (B) experiments with water collected at a depth of 5 m from Saguaro Lake (September 2000; corresponds to samples presented in Fig. 2). Control sample (■) has negligible degradation compared to the 5 m sample without the added bioseed (△) or with the added bioseed (◇). Geosmin samples were spiked to 23 ng/L. Error bars show 1 std.dev. Lines represent fitted laboratory pseudo zero-order rates (R_L).

Photolysis of MIB and geosmin is possible, but no direct studies have quantified this sunlight-mediated photolysis. However, direct photolysis of MIB and geosmin was studied using medium-pressure mercury lamps and very high-energy dosages (10,000 mJ/cm^2) were required to remove 90% of the initial concentrations (Mofidi et al., 2000). In these study lakes it is unlikely that UV light will penetrate deep enough into the water column (Wetzel, 1983), although much of the MIB production could occur near the surface and therefore MIB would be exposed to some light. However, photolytic loss of MIB and geosmin is probably negligible.

Sorption or partitioning of MIB and geosmin onto particulate matter could occur. In fact WTPs add powder-activated carbon (PAC) to adsorb and remove MIB and geosmin (Bruce et al., 2002). PAC adsorbs MIB and geosmin through hydrophobic partitioning. Octanol-water partition coefficients (K_{OW}) for MIB (Log $K_{OW} = 3.1$) and geosmin (Log $K_{OW} = 3.7$) indicate moderate levels of hydrophobicity and sorption to

suspended material could occur. During this study Arizona was in a prolonged drought and turbidity associated with runoff into the reservoirs probably did not significantly change SD depths. Thermal destratification could suspend some inorganic material or detritus. Although suspended sediment analysis was not specifically conducted as part of the analytical procedures, the reservoirs did not appear to turn "dirt-colored" during thermal destratification. Although further research would be required to quantify sorption onto inorganic suspended material and organic detritus in the lakes may be required, this mechanism does not appear significant for our study. However, algae and bacteria cellular matter produced throughout the year, and especially following thermal destratification (as indicated decreased SD depths), could sorb MIB and geosmin. Because it is difficult to separate sorption from strict biological metabolism, these two processes will be considered jointly as biodegradation (discussed below).

Biodegradation of MIB and geosmin appears the most likely loss mechanism for several reasons. First, during laboratory experiments biodegradation was the dominant mechanism since other potential mechanisms were minimized. Algal growth and concurrent MIB and geosmin production were minimized by dark incubation; volatilization losses were minimized by use of closed vessels. Biosorption of MIB or geosmin onto bacterial biomass was likely unimportant, as negligible MIB or geosmin removal occurred in samples held at 4 °C or treated with sodium azide (control samples). Thus the laboratory experiments demonstrate the potential for native bacteria to degrade MIB and geosmin. Second, annual maximum negative R_F values ($R_{F,max}$) coincided with thermal destratification and usually followed periods of high MIB concentrations. In the lakes, indications of higher biological activity, including decreasing SD transparencies, increasing algae counts and increasing TN concentrations, accompanied autumn thermal destratification. During the period of annual maximum MIB loss ($R_{F,max}$), the primary substrate for heterotrophic bacteria may be organic matter released during algae growth or lyses (Plummer and Edzwald, 1998; Nguyen et al., 2002). For example, roughly 40–50% of the DOC obtained from laboratory cultures of green algae is biodegradable in bioacclimated sand reactors (Sommerfeld et al., 2001). Conditions in the lakes proceeding thermal destratification are conducive for bacterial growth and biodegradation of MIB and geosmin.

The increased biomass associated with organic cellular metabolites present after thermal destratification probably serve as the primary carbon source for bacterial growth. MIB is probably degraded in the reservoirs as a secondary substrate, similar to how MIB biodegrades in biofilm reactors (Rittmann et al., 1995). A secondary substrate may yield no energy to the cell (i.e., co-metabolism) or negligible energy due to its low concentration. Higher bacterial activity would occur with elevated primary substrate and nutrient levels, increasing the rate of MIB utilization. If MIB were degraded as a secondary substrate, R_L in laboratory experiments would increase with addition of primary substrate. This did occur in our experiments (e.g., bioseeded experiments), although we investigated only one level of nutrient addition. Others have observed a lag period prior to degradation of MIB by natural lake bacteria and postulated that it represented the time for growth of MIB degraders (Izaguirre et al., 1988a, b). With only limited bacterial counting data, higher bacterial counts were observed in that work when MIB was used as a sole carbon substrate (initial MIB = 23 mg/L) compared to cultures without MIB. The authors concluded that other unidentified organic or inorganic nutrients were required for MIB degradation at environmentally relevant concentrations.

Further evidence for MIB and geosmin biodegradation is available from the literature. *Bacillus cereus* has been suggested as the key bacillus species responsible for degradation of earthy-musty odors in lakes and sand filters (Danglot et al., 1983; Hoehn, 1965; Ishida and Miyaji, 1992; Macdonald et al., 1987; Oikawa et al., 1995; Saadoun and El-Migdadi, 1998). Hoehn (1965) took this in situ degradation concept a step further by validating the feasibility of bioseeding a reservoir (10 km^2) with mass cultures (9 m^3 lots) of *B. cereus* applied to the lake's surface and shoreline (Silvey, 1964). The bioseeding, or bioaugmentation, successfully decreased the threshold number for earthy-musty odors, but a "decaying vegetation" odor was detected. Therefore, bacterial degradation could be responsible for MIB degradation in the lakes. This is supported by observations of population cycles of gram positive heterotrophic bacilli that appeared to increase when concentrations of earthy-musty odors were highest (Hoehn, 1965). The disappearance of the earthy-musty odors was followed by decreases in bacillus populations. Natural bacterial populations in lake water were hypothesized to be responsible for MIB and geosmin degradation after observing rapid (3–5 weeks) declines in high geosmin (>150 ng/L) and MIB (inoculated at 1000 ng/L) concentrations (Means and McGuire, 1986). Loss rates in these systems were estimated to be between 5 and 50 ng/L-day for both MIB and geosmin. The 5 ng/L-day loss rate occurred in a lake and may have been due to a number of factors combined: dilution after reservoir destratification, hydraulic flushing with imported trans-basin water, and/or bacteria biodegradation. Separately, a loss rate of nearly 50 ng/L-day removal was estimated from MIB inoculated lake water and may have been impacted by a primary carbon source (i.e., methanol) present in the MIB solvent.

Results from the current study may only be applicable to similar lakes in semi-arid regions or oligotrophic reservoirs with comparable algae biomass densities. Higher biomass densities could stimulate growth of natural lake bacteria and potentially increase the rate of MIB biodegradation as a secondary substrate, or increase the partitioning of MIB and geosmin onto cellular material. Likewise, sorption may be important in lakes with high suspended particulate concentrations.

4.2. Comparison between field and laboratory MIB degradation rates

$R_{F,max}$ values during the autumn period were approximately half the value of lab-based R_L values (Table 2). This result could be due to a number of factors, including: (1) differences in bacterial growth conditions between the lab and field (nutrient levels, temperature, type of organism), (2) minor contributions from other MIB loss mechanisms in the reservoir (photolysis, volatilization, sorption to suspended sediment), (3) simultaneous production and degradation of MIB and geosmin in the reservoir, (4) assumptions regarding spatial heterogeneity of MIB and geosmin throughout the reservoir, and/or (5) calculation of the field rates due to use of approximately monthly intervals between sampling. Laboratory rates may therefore represent an upper limit on the loss of MIB due primarily to biodegradation, under the conditions evaluated for these waters. MIB loss rates on the order of 0.5–1 ng/L-day appear common in the lakes.

4.3. Implications for lake management

Understanding the hydrologic and biogeochemical processes that lead to changes in MIB and geosmin concentrations in water supplies is important for improving the aesthetic qualities of finished drinking water, because it can be an economic burden to WTPs to remove these T&O compounds. This paper explored three important concepts. First, thermal destratification of reservoirs can increase MIB and geosmin concentrations in the hypolimnion due to mixing. While bottom-release reservoirs may have low MIB and geosmin concentrations throughout the summer, the compounds produced in the upper 5–15 m of the water column can contribute to rather rapid MIB and geosmin increases throughout the deeper waters of the reservoir, and in the outflow of the reservoirs, during and after autumn thermal destratification. In our study, the weakly stratified reservoirs (Saguaro Lake) experienced a more gradual increase of MIB and geosmin in water released from outlet at the bottom of the reservoir than did the more strongly stratified Bartlett Lake and Lake Pleasant, which had increases of 20–30 ng/L over the course of 15–30 days. In order to implement cost effective in-plant treatment (i.e., PAC application) it is important to predict the timing of when rapid changes in MIB and geosmin concentrations are likely to occur in water released from upstream reservoirs. Monitoring thermal stratification changes and MIB or geosmin depth profiles are important and can be used to estimate increases in MIB or geosmin after thermal destratification.

Second, the concept of mass flow analysis allows delineation between sources and losses of MIB or geosmin in water supplies. For Saguaro Lake, it was shown that MIB inflow from upstream reservoirs could be at least as important as MIB production within the reservoir itself. Thus, it may be fruitful to control algae growth in upstream reservoirs as well as in a terminal storage reservoir. Mass flow analyses also suggest that "flushing" of the reservoirs was not a major mechanism for MIB loss, and that in situ losses were more important. Different outcomes are possible for reservoirs with high water throughput (i.e., flushing) or continuously flowing rivers that serve as water supplies. However, MIB mass flow analysis in these other settings could aid in identifying reaches with the greatest net MIB production. The authors recently implemented this approach for water flowing in concrete-lined canals, where periphyton cause increases in MIB and geosmin along the length of the canal. There are numerous inflows and outflows in the canals. While MIB concentrations increased along the length of the canal, mass flow analyses facilitated computation of aerial MIB production (ng/L-day m^2). Canal reaches with high MIB aerial production were then successfully treated (copper and/or mechanical brushing) to prevent MIB production (Hu et al., 2003).

Third, MIB and geosmin reaction rates could be used with mass balances to predict the duration of T&O episodes and allow for improved management of water supplies. For the reservoirs in this study, thermal destratification was a major cause for increased MIB concentrations in water released from the bottom of the reservoirs to water utilities. Local utilities wanted to know how long the MIB episodes would last, and based upon a MIB loss rate of 0.5–1 ng/L-day and the MIB concentration after conservative mixing of the reservoirs during autumn destratification a duration of 30–40 days was estimated. This estimate allowed water utilities to continue ordering and applying PAC. Understanding the onset and duration of MIB in the reservoirs has recently resulted in operational changes in Lake Pleasant to avoid high MIB concentrations in water delivered to downstream water utilities. Upon onset of thermal destratification, and increased MIB concentrations in the water released from the bottom of the reservoir, the operator ceases release of water from the reservoir and uses water from an alternative source (pumped water from the Colorado River). Similar strategies could be used to manage other multiple-source water supply systems.

5. Conclusions

This study examined seasonal patterns of MIB production and degradation in three reservoir systems. For reservoirs in series, MIB mass contributions from upstream can be important. Within reservoirs MIB concentrations accumulate in the epilimnion during the warm summer months (August and September). Following autumn thermal destratification, MIB concentrations decline for 2–3 months to near-zero levels. In laboratory tests, R_L values ranged from 0.3 to 1.0 ng/L-day (average 0.4 ng/L-day). Observed $R_{F,max}$ values in the reservoirs were of the same order of magnitude as laboratory degradation rates (R_L) using reservoir water. Biodegradation, presumably heterotrophic aerobes, appeared to be more important mechanism for MIB and geosmin loss compared against volatilization, photolysis, or sorption. Thermal destratification and maximal chlorophyll-*a* concentrations provide bacteria with inorganic nutrients and primary carbon substrates (biodegradable cellular material) conducive to increased microbial growth and subsequent co-metabolism of ng/L concentrations of MIB and geosmin. Developing a better understanding of the factors controlling bacterial degradation of MIB may lead to management practices. Future work should focus on monitoring both algae and bacteria populations in conjunction with T&O compounds and other algal metabolites (i.e., cyanotoxins).

Acknowledgments

This research was supported by the City of Phoenix (Arizona) in cooperation with the Salt River Project and Central Arizona Water Control District. Post-doctoral researchers (Darlene Bruce, Qiang Hu, Mario Esparza-Soto), graduate students (Tom Dempster, My-Linh Nguyen, Kirsten Hintze, Michelle Cummings, Samanth Dawson), and technicians (David Lowry, Marisa Masles) assisted in sample collection and analysis. Helpful comments from an anonymous reviewer are greatly appreciated.

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