There are two major differences in conditions between plant operation and laboratory experiments: (1) Whereas cyanobacteria are usually present along with the musty odor compounds in the raw water flowing into the plant, laboratory experiments are usually conducted with commercially available reagent chemicals of musty odor compounds in the absence of cyanobacteria. Because the cyanobacteria commonly incorporate musty odor compounds into their structures, under high pressure these intracellular compounds might be released from the cyanobacteria into the water during plant operation. It should be noted that the musty odor compounds has been already known to be released from the cyanobacteria by the addition of oxidative chlorine compounds such as hypochlorous acid (Ashitani et al. 1988, Ando et al. 1992), and accordingly the prechlorination process could cause the release of the odor compounds into the water. However, the difference in removal ratio of the odor compounds between real plants and laboratory experiments is observed even in the plants in which the prechlorination process is not employed. (2) Whereas musty odor compounds produced by cyanobacteria enter treatment plants, commercially available chemically synthesized musty odor compounds are used in most laboratory experiments. Thus, the adsorption properties of the compounds produced by cyanobacteria might be different from those of the chemically synthesized compounds.

Accordingly, our objectives were to investigate (1) the release of intracellular geosmin from cyanobacteria under high pressure in batch tests; (2) the release of intracellular geosmin from cyanobacteria in laboratory-scale MF operation; (3) the removal of both geosmin and cyanobacteria in pilot-scale experiments by using a hybrid system of PAC adsorption, coagulation, and ceramic MF; and (4) differences in adsorption of natural and chemically synthesized geosmin by PAC in batch tests.

Methods

Cyanobacteria used

Three types of geosmin-producing cyanobacterium were used. Two of the three cyanobacteria, *Anabaena planktonica* (NIES 817) and *Anabaena smithii* (NIES 824), were provided by the National Institute for Environmental Studies (NIES, Tsukuba, Japan). These cyanobacteria were cultivated in 5-L glass vessels in cefixime and tellurite (CT) medium (Watanabe & Ichimura 1977). The other cyanobacterium, which was identified as *Anabaena* sp., was isolated from Lake Sagami (Yokohama, Japan) and then cultivated in 2-L flasks of CT medium.

Batch pressurization tests

Anabaena smithii in its three different growth phases (logarithmic phase, stationary phase, and decline phase) was used for the batch pressurization studies. Culture medium containing the cyanobacterium was diluted with river water (Toyohira River, Sapporo, Japan; DOC 1.1 mg/L, OD260 0.027 cm^{-1}) to obtain an extracellular geosmin concentration of approximately 100 ng/L. The solution was pressurized at 400 kPa for 4 h in a batch cell by introducing compressed air to the cell. After pressurization, the solution was gradually depressurized to normal pressure by using a ball valve to prevent volatilization of geosmin by rapid depressurization. Before and after pressurization, the intracellular and extracellular geosmin were separately quantified as described below.

Laboratory-scale MF experiments

Cyanobacterium *A. smithii* in its stationary phase was used for the MF experiments. Figure 1 is a schematic of the MF experiment. Culture medium containing the cyanobacterium was spiked into water from the Toyohira River in a raw water tank at the proportion of 1:9 (v/v). The cyanobacterium-spiked river water was directly fed into a monolithic ceramic MF module (single tubular, nominal pore size 0.1 μm, effective filtration area 0.0008 m^2; NGK Insulators, Ltd., Nagoya, Japan) at a constant flow rate (125 $L/(m^2 \cdot h)$) by a peristaltic pump in dead-end mode. The MF experiments lasted for 4 h with no backwashing. Geosmin concentrations in the MF inflow and in the MF permeate were measured periodically.

Pilot-scale experiments in a hybrid system with adsorption, coagulation, and microfiltration

Anabaena sp. isolated from Lake Sagami was used in the pilot-scale experiments (Fig. 2). Culture medium containing the cyanobacterium was spiked into water from Lake Sagami in a raw water tank. The

cyanobacterium-spiked lake water was supplemented with powdered activated carbon (PAC) at a dose of 2 mg/L. Two types of PAC were used: one was commercially available PAC (abbreviated here as N-PAC, d_{50} 7.6 μm, Futamura Chemical Industries Co., Ltd., Gifu, Japan) as received, and the other PAC (abbreviated here as S-PAC, d_{50} 0.65 μm) was obtained by micro-grinding of the N-PAC. After 2 min of PAC contact time with the water in the tube, the water was supplemented with coagulant (polyaluminum chloride, PACl, 10% (w/w), Hieisyouten Co., Ltd., Nagoya, Japan) at a dose of 25 mg/L. After 2 min of PACl contact time, the water was fed into a monolithic ceramic MF module (multichannel tubular, nominal pore sizes 0.1 μm, effective filtration area 0.48 m^2; NGK Insulators, Ltd.) at a constant flow rate (125 L/(m^2·h)) in dead-end mode. The experiments lasted for 4 h with no backwashing. Geosmin concentrations in the MF inflow and in the MF permeate were measured periodically.

Batch adsorption tests

Geosmin produced by *A. planktonica* was used for the batch adsorption tests. Culture medium containing the cyanobacterium was filtered through a glass fiber filter with a 1-μm pore size (Toyo Roshi Kaisha, Ltd., Tokyo, Japan) to remove any cyanobacterium cells, and then the filtrate was stored as a stock solution of natural geosmin. After appropriate dilution of the stock solution with ultrapure water to obtain a geosmin concentration of approximately 100 ng/L, chemically synthesized d_3-geosmin was injected into the diluted solution at the same concentration as that of the natural geosmin. The solution was supplemented with sodium bicarbonate at 16.8 mg/L, and then the pH was adjusted to 7.0 by the addition of HCl. N-PAC was then added to the solution at a dose rate of 0.7 mg/L. Samples were periodically withdrawn from the solution at predetermined times, and geosmin and d_3-geosmin were quantified after the samples had been passed through a membrane with a 0.2-μm pore size (PTFE, Toyo Roshi Kaisha, Ltd.).

Measurement of intra- and extracellular geosmin

Each sample was divided and placed into two beakers: one for quantification of total geosmin, and the other for quantification of extracellular geosmin. For quantification of total geosmin, sodium hypochlorite was added to the sample solution at 20 mg/L to release the intracellular geosmin into the water by breaking down the cell walls of the cyanobacteria. After the mixture had been kept for 30 min at room temperature for reaction, an excess amount of sodium thiosulfate was added to the mixture to quench the unreacted sodium hypochlorite. Quantification of the geosmin in the mixture after the mixture had been passed by gravity through a glass fiber filter with a 1-μm pore size gave the total geosmin concentration (intracellular + extracellular geosmin). For quantification of extracellular geosmin, cyanobacterial cells were removed from the sample solution by passing the solution by gravity through a glass fiber filter with a 1-μm pore size. Quantification of the geosmin in the filtrate gave the extracellular geosmin concentration. Subtraction of the extracellular geosmin concentration from the total geosmin concentration gave the intracellular geosmin concentration.

Method of geosmin analysis

Geosmin was extracted from the sample solutions by the stir bar sorptive extraction (SBSE) method with a Twister stir bar (Gerstel GmbH, Mülheim, Germany), and then quantified by gas chromatography – mass spectrometry (GC–MS, 6890N gas chromatograph, 5973 mass spectrometry detector, Agilent Technologies, Palo Alto, CA, USA) equipped with a thermal desorption apparatus (TDSA, Gerstel). GC–MS was performed in selected ion monitoring (SIM) mode. d_3-Geosmin was used as an internal standard, except in the batch adsorption experiments on d_3-geosmin, in which 2-MIB was used as an internal standard. Detection of ion fragments of geosmin, d_3-geosmin, and 2-MIB occurred at m/z 112, 115, and 95, respectively.

Results and discussion

Batch pressurization tests

Figure 3 shows the changes in geosmin concentrations before and after pressurization at 400 kPa for 4 h. In the logarithmic and stationary phases, more geosmin was retained in the cells than was found outside the cells. This distribution tendency was similar to that in *Anabaena macrospora* (Negoro et al. 1988),

Fischerella muscicola (Wu & Jüttner 1988a), and *Oscillatoria tenuis* (Wu & Jüttner 1988b). In contrast, the intracellular geosmin concentration was much smaller than the extracellular geosmin in the decline phase: most of the geosmin existed outside the cell.

No changes were observed in the concentrations of either extra- or intracellular geosmin at any growth phase after pressurization of the cyanobacterium-containing solutions up to 400 kPa for 4 h, indicating that geosmin was not released from the cyanobacteria under high pressure at 400 kPa in static conditions.

Laboratory-scale MF experiments

Figure 4a shows the changes in extracellular geosmin concentration in the MF inflow and in the MF permeate. The extracellular geosmin concentration in the MF inflow (white circles) did not change substantially during the filtration. In the early stage of filtration the extracellular geosmin concentration in the MF permeate (gray circles) was almost the same as that in the MF inflow, meaning that the MF membrane alone could not remove the extracellular geosmin because its pores were much larger than the geosmin molecules. In contrast, approximately 800 ng/L of the intracellular geosmin in the MF inflow (data not shown) was completely removed by the membrane, because the cyanobacterium, which incorporated part of the geosmin in its structure, was so much larger than the membrane's pores that it was rejected by the membrane. Bottino et al. (2001) reported that a ceramic MF membrane with pores of 0.2 μm almost completely rejected eight types of algae.

After 3 h of filtration, the geosmin concentration in the MF permeate exceeded that in the MF inflow, and at the end of filtration it was still increasing. This result evidently indicated that the geosmin was released from cyanobacterial cells that had been accumulated on the membrane surface, possibly by the increased operation pressure; transmembrane pressure (TMP) gradually increased during the filtration and peaked at approximately 300 kPa (Figure 4b). When release of geosmin from the cyanobacteria was observed at 3-h filtration, the TMP was approximately 160 kPa. This value was smaller than the pressure (400 kPa) at which release of intracellular geosmin did not occur in the batch pressurization tests described above. One possible explanation for the discrepancy is as follows: the cyanobacterial cells were isotropically pressurized under stationary conditions in the batch pressurization tests. In contrast, each cell on the membrane was pressurized on one of its sides, but vented to the atmosphere on the other. This gradient in pressure might have forced the cells to compress and release the intracellular geosmin. Another explanation is that when the cyanobacterial cells were pressed against the disturbed, rough surface of the membrane on which foulant was accumulated, the particles of foulant trapped between the membrane and the cells might have acted as fulcrums in the water flow, exerting shear forces on the cells and thus breaking them open. No morphologic differences between the cyanobacteria before and after filtration were observed under an optical microscope; further study with an electron microscope (i.e. under higher magnification and resolution than with the optical one) is needed.

Regardless, under high pressure intracellular geosmin was not released in static conditions but was released in dynamic conditions. However, the precise relationship between TMP and the release of geosmin is not clear and further study is needed. Nonetheless, the difference in geosmin removal between actual plant operations and laboratory experiments with reagent geosmin is apparently due to the release of geosmin from cyanobacterial cells accumulated on the membrane.

Pilot-scale experiments with a hybrid system of adsorption, coagulation, and microfiltration

The MF inflow contained approximately 40 ng/L of the intracellular geosmin as well as 55 ng/L of extracellular geosmin (data not shown). The extracellular geosmin concentration in the MF permeate was 20 ng/L at the beginning of filtration when the system was operated with N-PAC (figure 5), meaning that the geosmin was removed to a certain extent by N-PAC addition. However, the geosmin concentration in the MF permeate gradually increased with filtration time. The increase was probably due to the release of intracellular geosmin from cyanobacteria accumulated on the MF membrane, because coagulant dosing has been reported not to cause lysis of cyanobacterial cells and not to increase the geosmin concentration in the water (Velzeboer et al. 1995). The amount of N-PAC dosed to the system was insufficient for geosmin removal. In contrast, geosmin was completely removed from the water over the entire filtration period when the system was operated with S-PAC, even though the dose of S-PAC was exactly the same

as that of N-PAC. This is because the specific surface area and adsorption capacity of the S-PAC were much better than those of the N-PAC, thanks to the micro-grinding (Matsui 2004, 2005, 2006). The hybrid system using S-PAC adsorption, coagulation, and microfiltration could simultaneously and effectively remove both the cyanobacteria and the geosmin from the water.

Batch adsorption tests

Figure 6 shows changes in the concentrations of natural and synthesized d_3-geosmin. The concentrations of natural geosmin and d_3-geosmin decreased similarly with N-PAC contact time. No difference was observed between the remaining ratios of the two compounds, indicating that natural and synthesized geosmin behaved in the same manner with respect to PAC adsorption. Therefore, the difference observed in geosmin removal between actual plant operations and laboratory experiments does not result from a difference in adsorption characteristics between natural and chemically synthesized geosmin.

Conclusion

1. Intracellular geosmin was not released from cyanobacterial cells under the static conditions of the batch pressurization tests, but it was released under the dynamic conditions of the laboratory-scale MF experiments.
2. Extracellular geosmin was partly removed by the hybrid system with N-PAC dosing, but its removal rate decreased with filtration time, probably because of the release of intracellular geosmin. In contrast, the hybrid system with S-PAC dosing simultaneously and effectively removed both the cyanobacteria and the geosmin from the water.
3. No difference was observed in adsorption characteristics between natural and chemically synthesized geosmin.
4. The difference in geosmin removal between actual plant operations and laboratory experiments with reagent geosmin is probably caused by the release of geosmin from the cyanobacterial cells accumulated on the membrane under high pressure.

Acknowledgments

The authors thank Mr. M. Suzuki, Mr. Y. Nagai, Mr. F. Kanda, and Ms. N. Nigorikawa of the Waterworks Bureau, City of Yokohama, and Mr. Y. Kawase and Mr. S. Mima of NGK Insulators Ltd. for their assistance with the experiment. This study was part of collaborative research among Hokkaido University, the Waterworks Bureau of City of Yokohama and NGK Insulators, and was partly supported by research grants from the Ministry of Health, Labour and Welfare and the Ministry of Education, Culture, Sports, Science and Technology of the Government of Japan.

References

Ando, A., Miwa, M., Kajino, M. and Tatsumi, S. 1992 Removal of musty-odorous compounds in water and retained in algal cells through water purification processes, Water Science and Technology, **25** (2), 299–306.

Ashitani, K., Hishida, Y. & Fujiwara, K. 1988 Behavior of musty odorous compounds during the process of water treatment, Water Science and Technology, **20** (8-9), 261–267.

Bottino, A., Capannelli, C., Borghi, A. D., Colombino, M. & Conio, O. 2001 Water treatment for drinking purpose: ceramic microfiltration application, *Desalination*, **141**, 75–79.

Matsui Y., Murase R., Sanogawa T., Aoki N., Mima S., Inoue T. & Matsushita T. 2004 Micro-ground powdered activated carbon for effective removal of natural organic matter during water treatment. *Water Science and Technology: Water Supply*, **4** (4), 155–163.

Matsui Y., Murase R., Sanogawa T., Aoki N., Mima S., Inoue T. & Matsushita T. 2005 Rapid adsorption pretreatment with submicron powdered activated carbon particles before microfiltration. *Water Science and Technology*, **51** (6–7), 249–256.

Matsui Y., Sanogawa T., Aoki N., Mima S. & Matsushita T. 2006. Evaluating submicron-sized activated carbon adsorption for microfiltration pretreatment. *Water Science and Technology: Water Supply*, **6** (1), 149–155.

Negoro, T., Ando, M. & Ichikawa, N. 1988 Blue-green algae in Lake Biwa which produce earthy-musty odors, *Water Science and Technology*, **20** (8–9), 117–123.

Velzeboer, R., Drikas, M., Donati, C., Burch, M. & Steffensen, D. 1995 Release of geosmin by Anabaena circinalis following treatment with aluminum sulfate, *Water Science and Technology*, **31** (11), 187–194.

Watanabe, M. M. & Ichimura, T. 1977 Fresh- and salt-water forms of *Spirulina platensis* in axenic cultures, *Bulletin of Japanese Society of Phycology*, **25** (Suppl.), 371–377.

Wu, J. T. & Jüttner, F. (1988a) Effect of environmental factors on geosmin production by *Fischerella muscicola*, *Water Science and Technology*, **20** (8–9), 143–148.
Wu, J. T. & Jüttner, F. 1988b Differential partitioning of geosmin and 2-methylisoborneol between cellular constituents in *Oscillatoria tenuis*, *Archives of Microbiology*, **150**, 580–583.

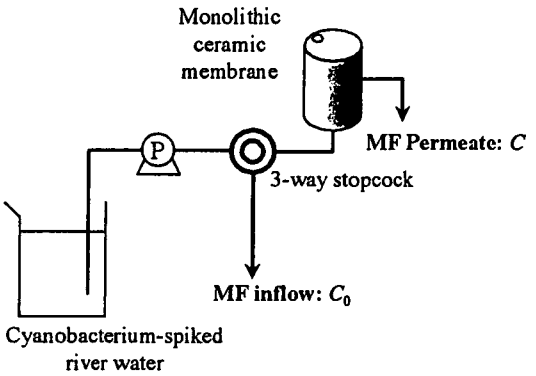

Figure 1 Schematic of the laboratory-scale MF experiment.

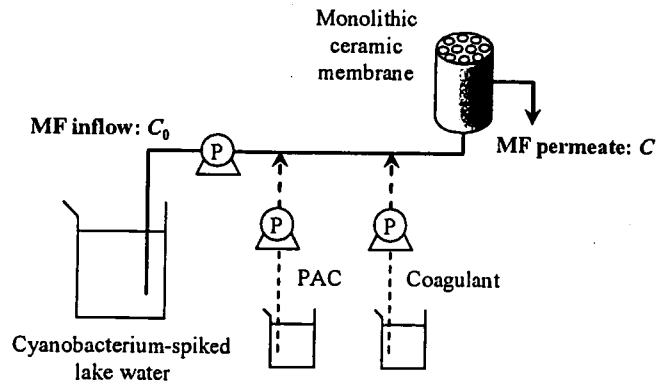

Figure 2 Schematic of the pilot-scale hybrid system with PAC adsorption, coagulation, and microfiltration.

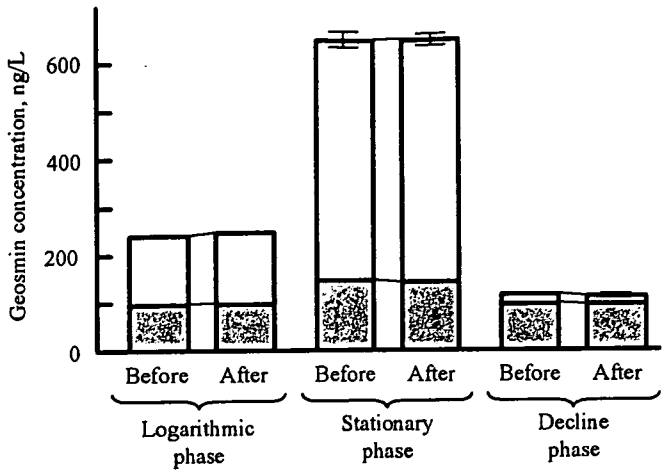

Figure 3 Changes in geosmin concentration before and after pressurization (400 kPa, 4 h). White and gray columns represent intra- and extracellular geosmin concentrations, respectively. Error bars represent standard deviation (n=3).

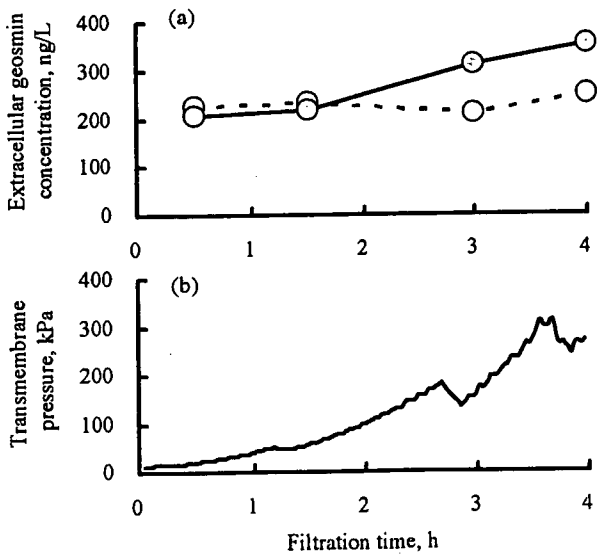

Figure 4 Changes in extracellular geosmin concentration in the MF inflow and in the MF permeate (a), and transmembrane pressure (b), in laboratory-scale MF experiments. White and gray circles represent the extracellular geosmin concentrations in the MF inflow and in the MF permeate, respectively.

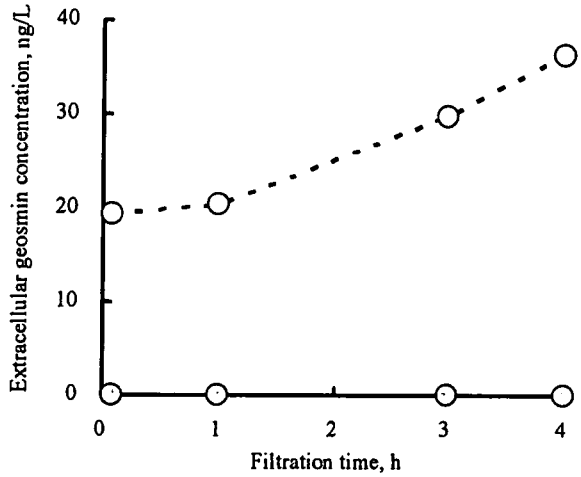

Figure 5 Changes in extracellular geosmin concentration in pilot plant experiments with a hybrid system that used PAC adsorption, coagulation, and microfiltration. White and gray circles represent N-PAC and S-PAC addition, respectively.

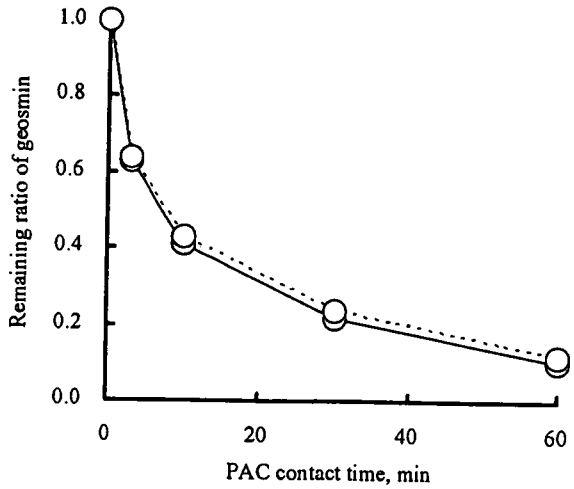

Figure 6 Changes in concentrations of natural and synthesized geosmin. Solid and open symbols represent natural and synthesized d3-geosmin, respectively.

Release of a musty odor compound from cyanobacteria during ceramic microfiltration and its effective removal by a combination of activated carbon adsorption, coagulation, and ceramic microfiltration

T. Matsushita[1]*, Y. Matsui[1], D. Sawaoka[1] and K. Ohno[1]

[1] *Hokkaido University, Sapporo, Japan*

**Corresponding author, email taku-m@eng.hokudai.ac.jp*

ABSTRACT
The removal ratio of compounds causing musty odor in real plants is sometimes smaller than that achieved in laboratory experiments. We investigated whether this difference in removal ratios was due to the possible release of intracellular geosmin under high pressure from cyanobacteria coexisting on the filter membrane surface. We conducted batch pressurization tests with a cyanobacterium-containing solution, laboratory-scale MF experiments, and pilot-scale experiments designed to remove both the geosmin and cyanobacteria in a hybrid system that used powdered activated carbon adsorption, coagulation, and ceramic microfiltration. Release of intracellular geosmin from cyanobacteria accumulated on the membrane surface was observed in both the laboratory-scale MF experiments and the pilot-scale experiments, but not in the batch pressurization tests. Geosmin was still observed in the MF permeate when the hybrid system was operated with commercially available powdered activated carbon (PAC), and its concentration increased with filtration time owing to the continued release of geosmin. In contrast, operation of the hybrid system with micro-ground PAC completely removed the geosmin.

KEYWORDS
Cyanobacteria; geosmin; microfiltration; powdered activated carbon adsorption.

INTRODUCTION
In Japan, the number of people suffering from the adverse taste of drinking water peaked at more than 20 million in 1990. The number decreased with time thanks to the adoption of the advanced water treatment process such as an ozonation process followed by an activated carbon adsorption process. However, the number is still very large, approximately 4 million in 2006. Earthy and musty odor compounds are the predominant source of the adverse taste, accounting for 63% of all cases in 2006. Earthy-musty odor is due to the presence of geosmin or 2-methylisoborneol (2-MIB); these compounds are produced mainly by cyanobacteria in lakes or ponds in the upper parts of drinking water catchments and remain in the tap water. The remaining compounds are detected easily by the human's gustatory organ as bad taste even at very low levels (less than 10 ng/L). In this meaning, removing these compounds from the water is still of great interest in the field of drinking water treatment.

Cyanobacteria producing the earthy-musty odor compounds hold geosmin or 2-MIB in their cell structure. The earthy-musty odor compounds are known to be released from the cyanobacteria by the addition of oxidative chlorine compounds such as hypochlorous acid. Therefore, if the raw water containing the cyanobacteria which are rich in earthy-musty odor compounds enters into a drinking water treatment plant employing the conventional process with the pre-chlorination, the earthy-musty odor compounds are released from the cyanobacteria to the water. Because the earthy-musty odor compounds are not expected to be removed in the coagulation, sedimentation and sand filtration processes, the finished water is therefore contaminated by the earthy-musty odor compounds. To prevent the release of the earthy-musty compounds, it is recommended not to employ the pre-chlorination to the raw water containing cyanobacteria having such compounds.

Powder activated carbon (PAC) adsorption has been applied widely for the removal of compounds causing the earthy-musty odor for a long time. On the other hand, membrane technologies have also recently been developed, and are now being used in drinking water treatment plants. A technology that combines PAC adsorption with microfiltration (MF) is already being employed in working plants, with

excellent results. However, the engineers at these plants often raise the question of why the removal ratio of the earthy-musty odor compound in real plants is smaller than that achieved in laboratory experiments. Thus, although the operational parameters used in real plants (e.g., the PAC dose and PAC contact time) are determined from the results of laboratory experiments, the performance of the systems in these plants is sometimes below the expected value.

Adding commercially available regent grade chemical of the earthy-musty odor compound to water usually gives the raw water for the laboratory experiments: cyanobacterium is not contained in the raw water. In contrast, odor-producing cyanobacterium coexists with the odor compound in the raw water in the actual treatment plants. We assumed that the difference in removal ratio of the earthy-musty odor compound was due to the release of the compound from cyanobacterium. In particular, the release of earthy-musty odor compound under high operation pressure was focused.

Accordingly, our objectives were to investigate (1) the release of intracellular odor compound from cyanobacteria under high pressure in batch tests (static conditions); (2) the release of intracellular odor compound from cyanobacteria in laboratory-scale MF operation (dynamic conditions); and (3) the removal of both odor compound and cyanobacteria in pilot-scale experiments by using a hybrid system of PAC adsorption, coagulation, and ceramic MF.

MATERIALS AND METHODS

Regents

Geosmin and d_3-geosmin were purchased from Wako Pure Chemicals Industries, Ltd. (Osaka, Japan), and used without further purification.

Cyanobacteria used

Three types of geosmin-producing cyanobacterium were used. Two of the three cyanobacteria, *Anabaena planktonica* (NIES 817) and *Anabaena smithii* (NIES 824), were provided by the National Institute for Environmental Studies (NIES, Tsukuba, Japan). These cyanobacteria were cultivated in 5-L glass vessels in cefixime and tellurite (CT) medium (Watanabe and Ichimura, 1977). The other cyanobacterium, which was identified as *Anabaena* sp., was isolated from Lake Sagami (Yokohama, Japan) and then cultivated in 2-L flasks of CT medium.

Batch pressurization tests

Anabaena smithii in its three different growth phases (logarithmic phase, stationary phase, and decline phase) was used for the batch pressurization studies. Culture medium containing the cyanobacterium was diluted with river water (Toyohira River, Sapporo, Japan; DOC 1.1 mg/L, OD260 0.027 cm^{-1}) to obtain an extracellular geosmin concentration of approximately 100 ng/L. The solution was pressurized at 400 kPa for 4 h in a batch cell by introducing compressed air to the cell. After pressurization, the solution was gradually depressurized to normal pressure by using a ball valve to prevent volatilization of geosmin by rapid depressurization. Before and after pressurization, the intracellular and extracellular geosmin were separately quantified as described below.

Laboratory-scale MF experiments

Cyanobacterium *A. smithii* in its stationary phase was used for the MF experiments. Fig. 1 shows a schematic of the MF experiment. Culture medium containing the cyanobacterium was spiked into water from the Toyohira River in a raw water tank at the proportion of 1:9 (v/v). The cyanobacterium-spiked river water was directly fed into a monolithic ceramic MF module (single tubular, nominal pore size 0.1 μm, effective filtration area 0.0008 m^2; NGK Insulators, Ltd., Nagoya, Japan) at a constant flow rate (125 $L/(m^2 \cdot h)$) by a peristaltic pump in dead-end mode. The MF experiments lasted for 4 h with no backwashing. Geosmin concentrations in the MF inflow and in the MF permeate

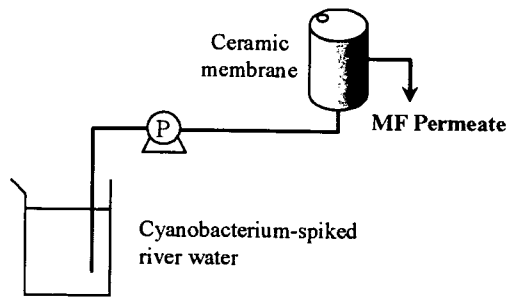

Fig. 1 Schematic of the laboratory-scale MF experiment.

were measured periodically.

Pilot-scale experiments in a hybrid system with adsorption, coagulation, and microfiltration

Anabaena sp. isolated from Lake Sagami was used in the pilot-scale experiments (Fig. 2). Culture medium containing the cyanobacterium was spiked into water from Lake Sagami in a raw water tank. The cyanobacterium-spiked lake water was supplemented with powdered activated carbon (PAC) at a dose of 2 mg/L. Two types of PAC were used: one was commercially available PAC (abbreviated here as N-PAC, d_{50} 7.6 μm, Futamura Chemical Industries Co., Ltd., Gifu, Japan) as received, and the other PAC (abbreviated here as S-PAC, d_{50} 0.65 μm) was obtained by micro-grinding of the N-PAC. After 2 min of PAC contact time with the water in the tube, the water was supplemented with coagulant (polyaluminum chloride, PACl, 10% (w/w), Hieisyouten Co., Ltd., Nagoya, Japan) at a dose of 25 mg/L. After 2 min of PACl contact time, the water was fed into a monolithic ceramic MF module (multichannel tubular, nominal pore sizes 0.1 μm, effective filtration area 0.48 m^2; NGK Insulators, Ltd.) at a constant flow rate (125 L/(m^2·h)) in dead-end mode. The experiments lasted for 4 h with no backwashing. Geosmin concentrations in the MF inflow and in the MF permeate were measured periodically.

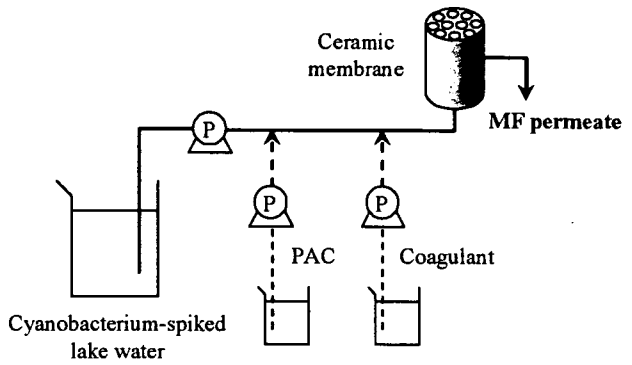

Fig. 2 Schematic of the pilot-scale hybrid system with PAC adsorption, coagulation, and microfiltration.

Measurement of intra- and extracellular geosmin

Each sample was divided and placed into two beakers: one for quantification of total geosmin, and the other for quantification of extracellular geosmin. For quantification of total geosmin, sodium hypochlorite was added to the sample solution at 20 mg/L to release the intracellular geosmin into the water by breaking down the cell walls of the cyanobacteria. After the mixture had been kept for 30 min at room temperature for reaction, an excess amount of sodium thiosulfate was added to the mixture to quench the unreacted sodium hypochlorite. Quantification of the geosmin in the mixture after the mixture had been passed by gravity through a glass fiber filter with a 1-μm pore size gave the total geosmin concentration (intracellular + extracellular geosmin). For quantification of extracellular geosmin, cyanobacterial cells were removed from the sample solution by passing the solution by gravity through a glass fiber filter with a 1-μm pore size. Quantification of the geosmin in the filtrate gave the extracellular geosmin concentration. Subtraction of the extracellular geosmin concentration from the total geosmin concentration gave the intracellular geosmin concentration.

Method of geosmin analysis

Geosmin was extracted from the sample solutions by the stir bar sorptive extraction (SBSE) method with a Twister stir bar (Gerstel GmbH, Mülheim, Germany), and then quantified by gas chromatography – mass spectrometry (GC–MS, 6890N gas chromatograph, 5973 mass spectrometry detector, Agilent Technologies, Palo Alto, CA, USA) equipped with a thermal desorption apparatus (TDSA, Gerstel). GC–MS was performed in selected ion monitoring (SIM) mode. d3-Geosmin was used as an internal standard. Detection of ion fragments of geosmin and d_3-geosmin occurred at m/z 112 and 115, respectively.

RESULTS AND DISCUSSION

Batch pressurization tests

Fig. 3 shows the changes in geosmin concentrations before and after pressurization at 400 kPa for 4 h. No changes were observed in the concentrations of either extra- or intracellular geosmin at any growth phase after pressurization of the cyanobacterium-containing solutions up to 400 kPa for 4 h, indicating that geosmin was not released from the cyanobacteria under high pressure at 400 kPa in static conditions.

Laboratory-scale MF experiments

Fig. 4c shows the changes in extracellular geosmin concentration in the MF inflow and in the MF permeate. The extracellular geosmin concentration in the MF inflow (white circles) did not change substantially during the filtration. In the early stage of filtration the extracellular geosmin concentration in the MF permeate (gray circles) was almost the same as that in the MF inflow, meaning that the MF membrane alone could not remove the extracellular geosmin because its pores were much larger than the geosmin molecules. In contrast, the intracellular geosmin, which is indicated as the white column in Figure 4a, was completely removed by the membrane, because the cyanobacterium, which incorporated part of the geosmin in its structure, was so much larger than the membrane's pores that it was rejected by the membrane. Bottino et al. (2001) reported that a ceramic MF membrane with pores of 0.2 μm almost completely rejected eight types of algae.

After 3 h of filtration, the geosmin concentration in the MF permeate exceeded that in the MF inflow, and at the end of filtration it was still increasing. This result indicated that the geosmin was released from cyanobacterial cells that had been accumulated on the membrane surface, possibly by the increased operation pressure; transmembrane pressure (TMP) gradually increased during the filtration and peaked at approximately 300 kPa (Fig. 4b). When release of geosmin from the cyanobacteria was observed at 3-h filtration, the TMP was approximately 160 kPa. This value was smaller than the pressure (400 kPa) at which release of intracellular geosmin did not occur in the batch pressurization tests described above. One possible explanation for the discrepancy is as follows: the cyanobacterial cells were isotropically pressurized under stationary conditions in the batch pressurization tests. In contrast, each cell on the membrane was pressurized on one of its sides, but vented to the atmosphere on the other. This gradient in pressure might have forced the cells to compress and release the intracellular geosmin. Another explanation is that when the cyanobacterial cells were pressed against the disturbed, rough surface of the membrane on which foulant was accumulated, the particles of foulant trapped between the membrane and the cells might have acted as fulcrums in the water flow, exerting shear forces on the cells and thus

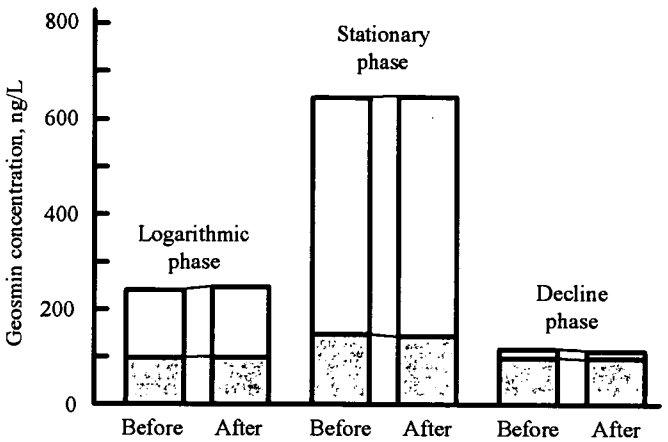

Fig. 3 Changes in geosmin concentration before and after pressurization. White and gray columns represent intra- and extracellular geosmin concentrations, respectively.

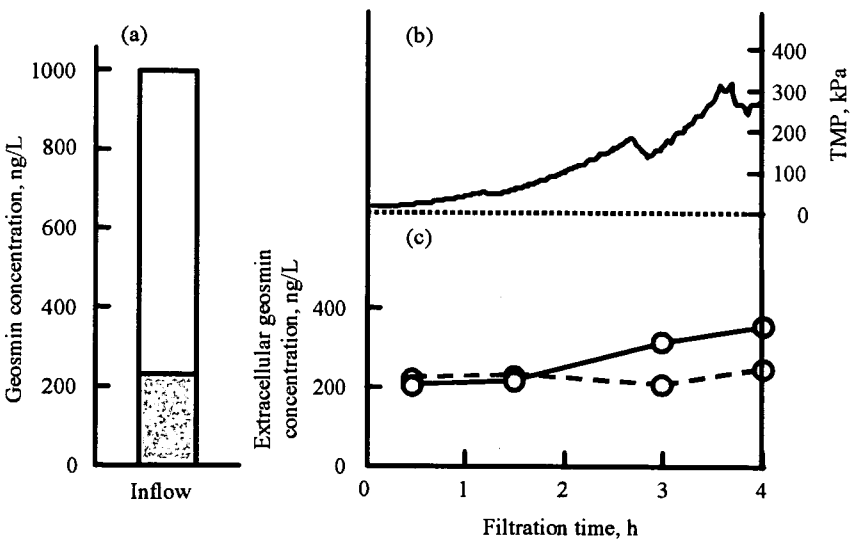

Fig. 4 Changes in geosmin concentration in the MF inflow and in the MF permeate, and transmembrane pressure (TMP), in laboratory-scale MF experiments. (a) Intra- and extracellular distribution of geosmin in the MF inflow. White and gray columns represent intra- and extracellular geosmin concentrations, respectively. (b) Changes in transmembrane pressure (TMP) with filtration time. (c) Changes in extracellular geosmin concentration in the MF inflow and in the MF permeate. White and gray circles represent the extracellular geosmin concentrations in the MF inflow and in the MF permeate, respectively.

breaking them open.

Regardless, under high pressure intracellular geosmin was not released in static conditions but was released in dynamic conditions. However, the precise relationship between TMP and the release of geosmin is not clear and further study is needed. Nonetheless, the difference in geosmin removal between actual plant operations and laboratory experiments with reagent geosmin is apparently due to the release of geosmin from cyanobacterial cells accumulated on the membrane.

Pilot-scale experiments with a hybrid system of adsorption, coagulation, and microfiltration

Whereas the extracellular geosmin concentration in the MF inflow (gray column in Figure 5a) was 55 ng/L, that in the MF permeate (white circles in Fig. 5b) was 20 ng/L at the beginning of filtration when the system was operated with N-PAC, meaning that the geosmin was removed to a certain extent by N-PAC addition. However, the geosmin concentration in the MF permeate gradually increased with filtration time. The increase was probably due to the release of intracellular geosmin from cyanobacteria accumulated on the MF membrane, because coagulant dosing has been reported not to cause lysis of cyanobacterial cells and not to increase the geosmin concentration in the water (Velzeboer et al., 1995). The amount of N-PAC dosed to the system was insufficient for geosmin removal. In contrast, geosmin was completely removed from the water over the entire filtration period when the system was operated with S-PAC, even though the dose of S-PAC was exactly the same as that of N-PAC. This is because the specific surface area and adsorption capacity of the S-PAC were much better than those of the N-PAC, thanks to the micro-grinding (Matsui et al., 2004, 2005, 2006). The hybrid system using S-PAC adsorption, coagulation, and microfiltration could simultaneously and effectively remove both the cyanobacteria and the geosmin from the water.

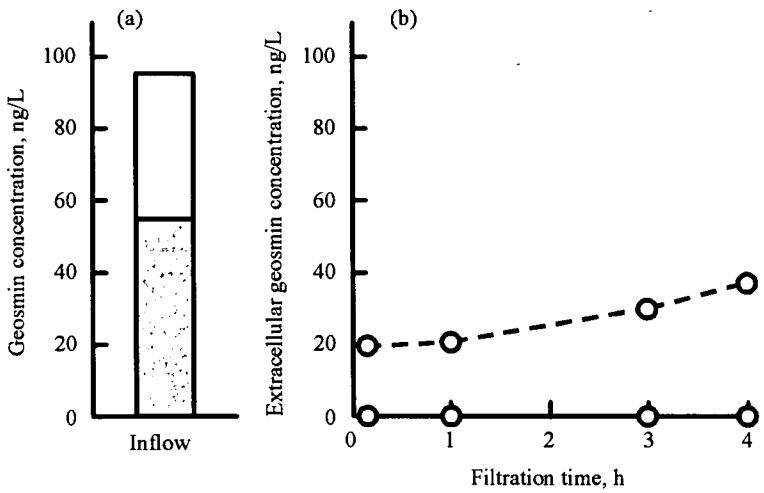

Fig. 5 Changes in geosmin concentration in pilot plant experiments with a hybrid system that used PAC adsorption, coagulation, and microfiltration. (a) Intra- and extracellular distribution of geosmin in the MF inflow. White and gray columns represent intra- and extracellular geosmin concentrations, respectively. (b) Changes in extracellular geosmin concentration in the MF permeate. White and gray circles represent N-PAC and S-PAC addition, respectively.

CONCLUSION

1. Intracellular geosmin was not released from cyanobacterial cells in static conditions, but released in dynamic conditions.
2. The difference in geosmin removal between actual plant operations and laboratory experiments with reagent geosmin is probably caused by the release of geosmin form the cyanobacterial cells accumulated of the membrane under high pressure.
3. Extracellular geosmin was partly removed by the hybrid system with N-PAC dosing, but its removal rate decreased with filtration time, probably because of the release of intracellular geosmin.
4 In contrast, the hybrid system with S-PAC dosing effectively removed the geosmin from the water.

ACKNOWLEDGEMENTS

The authors thank Mr. M. Suzuki, Mr. Y. Nagai, Mr. F. Kanda, and Ms. N. Nigorikawa of the Waterworks Bureau, City of Yokohama, and Mr. Y. Kawase and Mr. S. Mima of NGK Insulators Ltd. for their assistance with the experiment. This study was part of collaborative research among Hokkaido University, the Waterworks Bureau of City of Yokohama and NGK Insulators, and was partly supported by research grants from the Ministry of Health, Labour and Welfare and the Ministry of Education,

Culture, Sports, Science and Technology of the Government of Japan.

REFERENCES

Bottino, A., Capannelli, C., Borghi, A. D., Colombino, M. and Conio, O. (2001) Water treatment for drinking purpose: ceramic microfiltration application, Desalination, 141, 75–79.

Matsui Y., Murase R., Sanogawa T., Aoki N., Mima S., Inoue T. and Matsushita T. (2004). Micro-ground powdered activated carbon for effective removal of natural organic matter during water treatment. Water Science and Technology: Water Supply, 4 (4), 155–163.

Matsui Y., Murase R., Sanogawa T., Aoki N., Mima S., Inoue T. and Matsushita T. (2005) Rapid adsorption pretreatment with submicron powdered activated carbon particles before microfiltration. Water Science and Technology, 51 (6–7), 249–256.

Matsui Y., Sanogawa T., Aoki N., Mima S. and Matsushita T. (2006). Evaluating submicron-sized activated carbon adsorption for microfiltration pretreatment. Water Science and Technology: Water Supply, 6 (1), 149–155.

Velzeboer, R., Drikas, M., Donati, C., Burch, M. and Steffensen, D. (1995) Release of geosmin by Anabaena circinalis following treatment with aluminum sulfate, Water Science and Technology, 31 (11), 187–194.

Watanabe, M. M. and Ichimura, T. (1977) Fresh- and salt-water forms of Spirulina platensis in axenic cultures, Bulletin of Japanese Society of Phycology, 25 (Suppl.), 371–377.