

(48) BIOTRANSFORMATIONS OF ARSENIC SPECIES IN ACTIVATED SLUDGE PROCESS

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In this study, the kinetics of arsenite [As(III)] oxidation and arsenate [As(V)] reduction by activated sludge was investigated by laboratory experiments. The initial reaction rate data were fitted to the Michaelis-Menten model. Then, in order to confirm the potential application of activated sludge as As bio-oxidant, As speciation analysis was carried out for wastewater samples collected from 11 stations in the Akiu-Onsen Wastewater Treatment Plant (AWTP), located in Sendai, Japan, which is operated under the oxidation ditch activated sludge process. As(III) and As(V) were the dissolved species found in the influent and were similarly distributed. In the oxidation ditch, As(III) was oxidised to As(V) after supply of oxygen by the aerator and As(V) was the main species in the final effluent (>98% of total species). In the return sludge pipe, As(V) was reduced to As(III). As was inefficiently removed by the process and the final effluent concentration was 260 µg/L. By jar-tests, it was confirmed that addition of 24 mg/L as ferric chloride to the AWTP effluent could remove more than 95% of the biologically oxidised As and decreased the residual total As concentration to less than 10 µg/L.

It was concluded that the activated sludge process is a reliable technology to biologically oxidise As(III) to As(V) prior to an additional treatment, such as coagulation process, to remove As from contaminated wastewaters.

Keywords: Arsenic biotransformation kinetics, activated sludge, arsenic removal, ferric chloride

1. INTRODUCTION

Arsenic (As) is a highly toxic element that exists in four oxidation states [-III (arsine), 0 (elemental), +III (trivalent), +V (pentavalent)] and occurs in inorganic or organic form. The form and oxidation state of As, on which its toxicity depends, are controlled mainly by the pH and redox conditions. The inorganic species, arsenite [As(III)] and arsenate [As(V)], are the predominant species found in most environments and are considered to be much more toxic than the organic ones¹⁾.

Natural processes and anthropogenic activities have contributed to the release of large amounts of As in the environment. In Japan, As has been ranked among the 12 major environmental pollutants²⁾ and the top 10 of the most released chemical substances³⁾. Its presence at a significant or trace concentration was reported in river⁴⁾, lake⁵⁾, groundwater⁶⁾, mine drainage⁷⁾, sediment⁸⁾ or hot

springs⁹⁾, as well as in seawater organisms^{10), 11)}. As is also found in many areas of the world and large-scale contamination continues affecting adversely the health of millions of people¹²⁾. Thus, the elimination of As from the contaminated sites, including water and wastewaters, has become a great challenge for scientists and engineers.

Coagulation process has been the most frequently used method to treat As-contaminated water and wastewaters in numerous pilot- and full-scale applications. It involves the addition of a metal coagulant, usually alum or ferric ions. This technology can reduce As concentrations to less than 50 µg/l and, in some cases, to below 10 µg/l¹³⁾.

Of different processes tested on laboratory scale to treat As-contaminated water or wastewaters, As(V) was generally removed more efficiently than As(III)¹⁴⁾⁻¹⁸⁾. Thus, when As(III) is present, it should be pre-oxidised to As(V). Processes applied to accomplish this step include ozonation, photo-

oxidation, or the addition of oxidising chemicals such as potassium permanganate, sodium hypochlorite, or hydrogen peroxide¹³. Actually, there is a growing interest in the application of biological oxidation processes as new alternatives to the use of chemical oxidants. Some microorganisms isolated from various complex biological systems, such as soils, sediments, sewage or marine and geothermal areas, have shown a high resistance to As and the ability to catalyse the oxidation of As(III) to As(V)¹⁹. Since the activated sludge is also a bacterial-rich environment, authors²⁰ have investigated its ability to control the transformation and the removal of As(III), As(V), monomethylarsonic acid [MMA(V)] and dimethylarsinic acid [DMA(V)] in batch experiments under aerobic and anoxic conditions. It was found that the overall arsenic removal was less than 25%. On the other hand, under aerobic condition, As(III) was oxidised to As(V), and MMA(V) was simultaneously methylated to DMA(V) and demethylated to As(III), which in turn was oxidised to As(V). Under anoxic condition, As(V) was reduced to As(III). All reactions occurred immediately after contact of the As species with the sludge and only with the presence of living microorganisms, showing that they were biologically mediated. It was then concluded that the activated sludge process could not be applied to remove As from wastewater by direct adsorption onto the sludge flocs but for the pre-oxidation of arsenic species to As(V).

Earlier, Goldstone et al.²¹ have shown that the removal of As in a full-scale activated sludge process receiving wastewater containing less than 15 µg As/L as daily average concentration was only 34%. Furthermore, Watanabe et al.²² have investigated the behaviour of As in a biological wastewater treatment plant receiving As-contaminated wastewater. They have reported that the daily average of As concentration in the influent was 213 µg/L. Based on mass balance analysis, it was shown that only 14% of As contained in the influent was removed through the treatment processes.

The objective of this study was to evaluate the potential application of activated sludge as As bio-oxidant for the removal of As from wastewaters. First, an attempt was made to represent the kinetics of As(III) oxidation and As(V) reduction by activated sludge. Then, a full-scale As speciation analysis was carried out for wastewater samples collected from the Akiu-Onsen Wastewater Treatment Plant to validate the laboratory findings on As biotransformations by activated sludge. Because the behaviour and removal of total and dissolved As in the treatment plant have been already well covered in the work of Watanabe et al.²² as reported above, our research was focused principally

on the occurrence, the forms and the transformations of As species. Finally, jar-tests were conducted to obtain the optimum ferric chloride dosage to achieve residual As concentration meeting the Japan National Effluent Standard (100 µg/L) and the Japan Environmental Quality Standard (10 µg/L) for As.

2. METHODOLOGY

(1) Laboratory experiments: Kinetics of As(III) oxidation and As(V) reduction

Stock solutions of arsenite [NaAsO_2 , 1g As/L] and arsenate [$\text{Na}_2\text{HAsO}_4 \cdot 7\text{H}_2\text{O}$, 1g As/L], purchased from Kanto Chemicals (Tokyo, Japan), were prepared with deionised water and stored at 4°C before use.

Fresh activated sludge was collected from the aeration tank in the Tonan Wastewater Treatment Plant, located in Morioka, Japan, which is operated under the conventional activated sludge process. Activated sludges originating from the same treatment plant have been used during batch studies on As species behaviour²⁰. Upon arrival to the laboratory, a desired volume of sludge was washed three times with deionised water to remove soluble components. After settling, the liquid phase was removed while the solid phase was kept for the experiments.

Solutions containing approximately 50, 100, 250, 500 and 750 µg/L as As(III) or As(V) were prepared by diluting the stock solutions with deionised water. Solutions containing As(III) or As(V) were kept under aerobic or anoxic conditions, respectively. The pH was adjusted to 7 with 1M sodium hydroxide solution and 0.1M hydrochloric acid, and samples were collected to determine the initial As concentration in each solution. Then, solid phase of washed activated sludge with final concentration of about 1 g/L as suspended solids (SS) were suspended in the solutions containing As species in 2-L shaking flasks. The mixtures were agitated on a shaker table at 120 rpm for 30 minutes at 25°C. Samples were collected after 5, 10, 15 and 30 minutes from the start of the experiments, centrifuged for 10 min at 3500 rpm and filtrated with a 0.45 µm membrane filter (Millipore, Japan). Finally, the concentrations of As species in the filtrates were immediately measured to avoid changes in their forms in the samples.

(2) Field investigation: description of the Akiu-Onsen Wastewater Treatment Plant and sampling procedures

The Akiu-Onsen Wastewater Treatment Plant (AWTP) is located in Sendai, Japan. It receives about 2800 m³/day of municipal sewage and hot

spring wastewaters contaminated with As, which are treated via two parallel treatment systems, each consisting of two biological tanks operated under the oxidation ditch activated sludge process. The flow chart of one system in the AWTP is represented in Fig. 1. It consists of a grit chamber, two biological treatment tanks, a clarifier and a chlorination tank for disinfection. A part of the settled sludge is returned to the aeration tanks with the effluent from the grit chamber to maintain appropriate sludge concentration while the other part is treated via a sludge treatment system including thickening and chemical stabilisation. The treated wastewater is finally discharged to the Natori River.

Samples were collected into plastic bottles pre-cleaned with HNO₃ and deionised water on December 12th in 2005 at 11 stations in the AWTP as indicated in Fig. 1, namely at the inlet of the treatment plant [st.1], at the inlet [st.2] and the outlet [st.8] of the oxidation ditch, in the oxidation ditch [st.3 to st.7], at the outlets of the clarifier and the chlorination tank [st.9 and st.10], and in the drawing pipe of the clarifier [st.11]. The samples from sts.1 to 10 were collected at near water surfaces in the AWTP (<30 cm in depth). The sample at st.11 was collected from a water valve of the return sludge drawing pipe close to the clarifier side. The second aerator (after st.5) was stopped in order to have a decreasing concentration of dissolved oxygen (DO) along the oxidation ditch. Typical properties of the samples collected at the different stations are reported in Table 1. The water temperatures, pH (pH meter F-12, Horiba, Japan), DO concentrations (DO

meter DOL-10, DKK, Japan) and ORP (pH/ORP meter HM-5S, TOA Electronics, Japan) were measured on-site during the samples collection. DOC concentrations were measured in our laboratory (TOC-500A, Shimadzu, Japan) using the on-site filtrated samples. The SS and COD_{Mn} values were obtained from the AWTP official data.

Samples were filtrated on-site with a 0.45 µm membrane filter.

Filtrates were preserved without acidification in cooler containers (4°C) until arrival to the laboratory, and then refrigerated before analysis conducted within 24 hours.

(3) Jar-tests

A ferric chloride stock solution (10 g FeCl₃/L) was prepared with FeCl₃ reagent (Kanto chemicals, Japan) and deionised water, and stored in an acid-washed dark glass container.

Samples were collected from the clarifier outlet [st.9] in the AWTP on November 14th in 2006 and preserved in cooler containers (4°C) until arrival to the laboratory to avoid the change in As species in the samples.

Upon arrival, the effluent from the clarifier was transferred into eleven 1-litre cylindrical glass beakers, and various volumes of the FeCl₃ stock solution were added to each beaker.

The concentrations of FeCl₃ added were 1, 2, 4, 5, 6, 8, 10, 25, 50, 75 and 100 mg/L. After mixing, the pH of each solution was adjusted to 7 with 1M sodium hydroxide solution. Finally, the beakers were placed on a jar-tester.

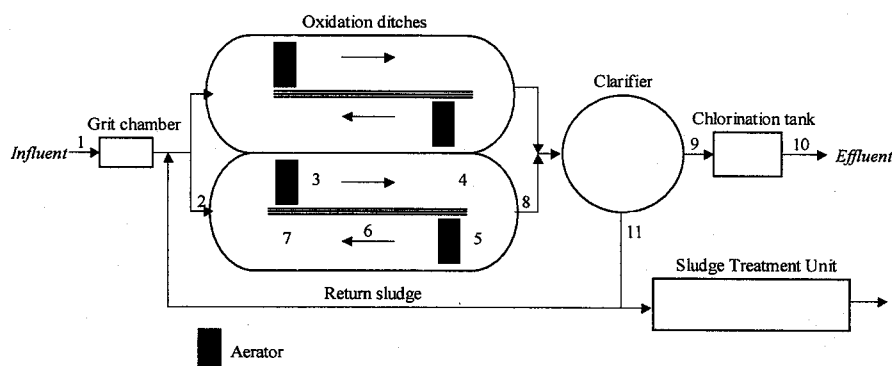


Fig. 1 Flow chart of the Akiu-Onsen Wastewater Treatment Plant (AWTP) and sampling locations.

Table 1 Sample characteristics.

Station	1	2	3	4	5	6	7	8	9	10	11
Water temperature (°C)	20	17			17			17	17	17	17
pH	7.1	7.0			6.9			6.9	7.0	7.1	6.9
SS (mg/L)	164	-			2080 (MLSS)			-	-	1.6	3880
DO (mg/L)	2.1	0.2	1.4	0.3	0.1	0.0	0.1	0.2	2.7	5.6	3.8
ORP (mV)	260	150	320	350	340	340	310	350	400	650	330
COD _{Mn} (mg/L)	111	-	-	-	-	-	-	-	-	4.9	-
DOC (mg/L)	53	22	5.4	6.4	8.0	6.0	8.0	7.1	5.7	7.3	8.2

MLSS: Mixed Liquor Suspended Solids (-): data not available

The mixing conditions were: 1 min rapid mixing at 100 rpm, 30 min slow mixing at 45 rpm and 15 min quiescent settling¹⁴⁾. After settling, two samples were collected from the supernatant. One was not filtrated to analyse total As (T-As) concentration while the other was filtrated with a 0.45 µm membrane filter for dissolved As (D-As) concentration analysis.

(4) Analytical methods

Non-filtrated samples were analysed for T-As with an ICP-MS (HP4500, Yokogawa Analytical Systems, Japan) after digestion with HNO₃ and H₂SO₄^{23), 24)}, followed by filtration with 5B filter (Advantec, Japan) and dilution with deionised water.

D-As was directly determined with an ICP-MS and recovery tests for mixtures of standard and sludge filtrate can be found elsewhere²⁰⁾.

Analysis of As species was done by on-line coupling of HPLC (Shimadzu, Japan) with a Hamilton PRP-X100 anion exchange column (Hamilton, USA) and ICP-MS based on the method developed by Martinez-Bravo et al.²⁵⁾ for the simultaneous determination of Cr(VI), Se(IV), Se(VI), As(III), As(V), MMA(V) and DMA(V) in surface waters. This method was also shown to be suitable for the analysis of As species in the filtrate of activated sludge²⁰⁾. The detection limits were

0.214, 0.819, 0.668 and 0.183 µg/L for As(III), As(V), MMA(V) and DMA(V), respectively. These values were calculated by²⁶⁾:

$$\text{Detection limit} = \frac{3\sigma \times 10}{(S-B)} \quad (1)$$

where σ is the standard deviation in counts of a blank solution, S is the counts of a 10 µg/L standard solution and B is the counts of the blank background.

3. RESULTS AND DISCUSSION

(1) As(III) oxidation and As(V) reduction by activated sludge

We have already shown that no transformation of As species occurred within 12 hours in control solutions with no sludge and in solutions with autoclaved sludge²⁰⁾.

The time courses of the oxidation of As(III) to As(V) and the reduction of As(V) to As(III) at various initial concentrations of each species are shown in Figs. 2 and 3, respectively.

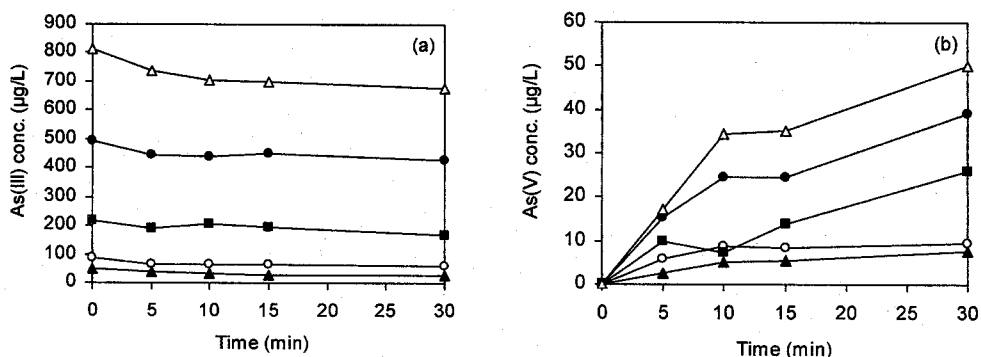


Fig. 2 Time course of the oxidation of (a) As(III) to (b) As(V) when As(III) was initially introduced.
[Initial As (µg/L); SS (g/L)] = ▲ [46; 0.87], ○ [89; 0.95], ■ [217; 1], ● [492; 0.84], △ [817; 0.88]

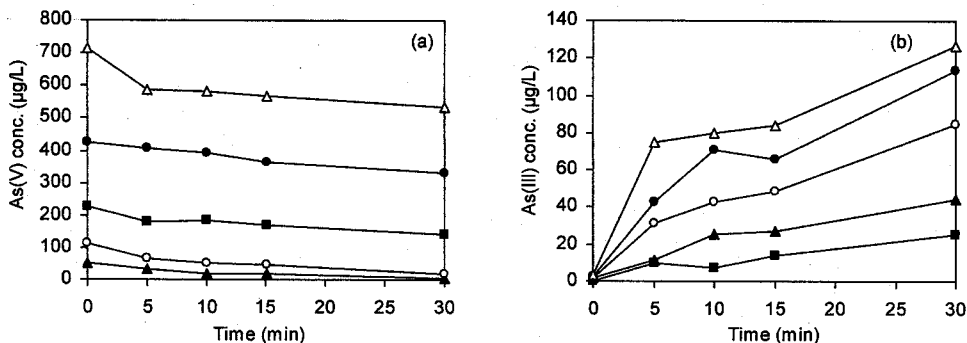


Fig. 3 Time course of the reduction of (a) As(V) to (b) As(III) when As(V) was initially introduced.
[Initial As (µg/L); SS (g/L)] = ▲ [54; 0.85], ○ [112; 0.93], ■ [228; 0.87], ● [428; 0.89], △ [715; 0.93]

Under aerobic condition, DO concentrations and the oxidation-reduction potential (ORP) at the start of the experiments and after 30 minutes varied from 6.5 to 2.5 mg/L and from 390 to 500 mV, respectively. Under anoxic condition, DO concentration was kept at less than 0.5 mg/L and ORP varied from 390 mV at the start of the experiment to 300 mV after 30 minutes.

The oxidation and reduction reactions began immediately after contact of the As species with the living microorganisms. Then, the amount of As(V) produced when As(III) was initially introduced and that of As(III) produced when As(V) was initially introduced were respectively increased with contact time and with the increase in the initial concentration of each species.

When the results obtained in As(III) oxidation experiments were compared with those in As(V) reduction ones, it was shown that under similar initial concentrations, the decrease in As(III) concentration was less than that in As(V) one [Figs. 2-(a) and 3-(a)] and that the amount of As(V) produced was less than that of As(III) produced [Figs. 2-(b) and 3-(b)]. In addition, the amount of product formed was less than that of reactant removed. The difference between the amounts of the reactant utilised and the product formed suggested that besides the oxidation-reduction reactions, slight adsorption of arsenic species have also occurred at the early stage of the experiments. However, it is difficult to determine if they occurred simultaneously or consecutively. The role of adsorption in the removal of As species by activated sludge is currently being investigated in our laboratory (manuscript in preparation).

(2) Kinetics of As(III) oxidation and As(V) reduction

It was reported that the genes encoding arsenite oxidase and arsenate reductase enzymes were found in a broad range of bacteria^{19), 27)-29)}. We have already

shown that As species transformations in the activated sludge process were biologically mediated²⁰⁾. Therefore, to determine the As reaction kinetics in the activated sludge, we tried to apply the Michaelis-Menten equation for enzyme kinetics. As(III) or As(V) was regarded as the substrate. The concentrations of the products [As(III) or As(V)] after 5 minutes were used to calculate the initial reaction rate of As(III) oxidation and As(V) reduction. The results were plotted against the initial substrate concentrations (Fig. 4). The relationships between the substrate concentration and the initial reaction rate were fitted to the Michaelis-Menten model³⁰⁾:

$$V = \frac{V_{\max} S}{K_m + S} \quad (2)$$

where V is the initial reaction rate (μg product formed/g SS/min), S is the initial substrate concentration ($\mu\text{g/L}$), V_{\max} is the maximum rate of the reaction ($\mu\text{g/g SS/min}$) and K_m is the Michaelis-Menten constant ($\mu\text{g/L}$) and is equal to the substrate concentration at which the reaction rate is one-half of the maximum one.

The Lineweaver-Burk plot (Fig. 5), a linear derivation of the Michaelis-Menten equation, was used to determine the constants V_{\max} and K_m . The kinetic parameters V_{\max} and K_m are 7 $\mu\text{g/g SS/min}$ and 500 $\mu\text{g/L}$ for As(III) oxidation and 30 $\mu\text{g/g SS/min}$ and 520 $\mu\text{g/L}$ for As(V) reduction.

The V_{\max} results show that the rate of As(V) reduction was faster than that of As(III) oxidation. During these experiments, the sludges used were originated from the same location and were prepared simultaneously by the same procedures. Therefore, the bacterial composition, concentration and activities in each mixture would be same at the start of the experiments. The difference in the reaction rate might then be due to the difference in the activities or concentrations of As-reducing and As-oxidising bacteria in the sludge.

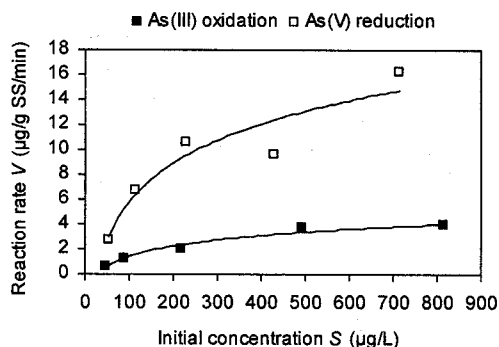


Fig. 4 Reaction rate of As(III) oxidation and As(V) reduction as a function of initial substrate concentration.

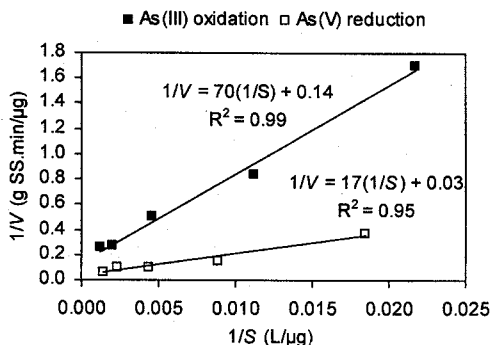


Fig. 5 Lineweaver-Burk plot for As(III) oxidation and As(V) reduction by activated sludge.

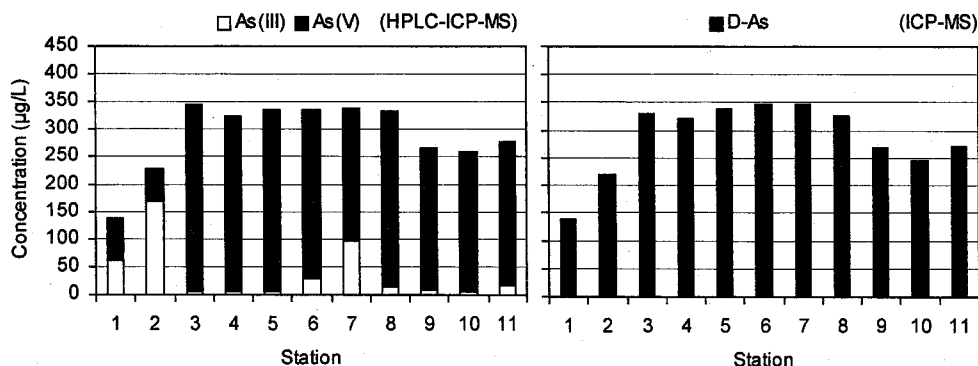


Fig. 6 Concentrations of As species measured by HPLC-ICP-MS and D-As measured by direct ICP-MS through the AWTP.

(3) As species occurrence and distribution through the AWTP

The concentrations of As(III), As(V) and D-As, in the AWTP are shown in Fig. 6. Watanabe et al.²²⁾ have reported that As concentrations in the influent fluctuated between 150 and 375 µg/L.

In the present study, the samples at the different stations were collected simultaneously. Therefore, the data are of use only for the analysis of the occurrence and distribution of As species through the treatment plant, and shall not be compared to evaluate the removal of As. No MMA(V) and DMA(V) were detected at any stations, and the sums of As species measured by the HPLC-ICP-MS system were comparable to the values of D-As obtained by direct measurement with ICP-MS for all samples.

Fig. 7 shows the distribution between As(III) and As(V) through the AWTP. Both As(III) and As(V) were present in the influent (st.1) and accounted each for about 50% of the sum of As(III) and As(V) ([As(III)+As(V)]). Then, the distribution between As(III) and As(V) varied greatly depending on the station where the sample was collected. As(III) accounted for more than 70% of [As(III)+As(V)] at st.2 whereas As(V) accounted for almost 100% of [As(III)+As(V)] at st.3. In the oxidation ditch, it is

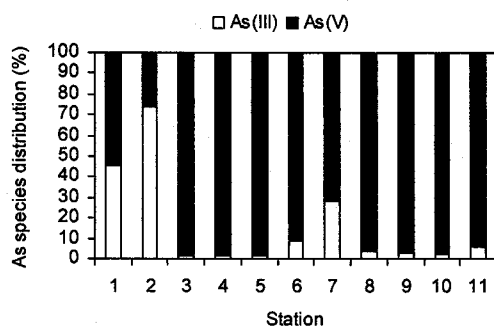


Fig. 7 Distribution between As(III) and As(V) through the AWTP.

clearly shown that the proportion of As(III) was increased as the sampling location was distant from the aerator (st.6 and st.7). Finally, As(V) accounted for more than 95% of [As(III)+As(V)] at st.8 and st.9, for 93% at st.11 and for almost 100% at the final outlet of the treatment plant (st.10), with concentration equal to 260 µg/L (Fig. 6).

D-As measured in the influent and the effluent were in agreement with the range of As concentrations in the AWTP reported by Watanabe et al.²²⁾. Under the pH and redox conditions at the different stations (Table 1), As(III) would be present as uncharged H_3AsO_3 , and As(V) as anionic $H_2AsO_4^-$ or $HAAsO_4^{2-}$ ³¹⁾. It is accepted that As(V) is stable at high ORP values and is found in well oxygenated environments, whilst As(III) is the predominant form in reducing environments³²⁾. Thus, despite of the redox condition at st.1 (Table 1), the significant proportion of As(III) suggests that As was released from the contamination source essentially as As(III), and that it was not completely oxidised to As(V) probably because of a more reducing condition in the sewerage system.

The variations in the distribution of As species at the different stations show that oxidation of As(III) or reduction of As(V) easily occurred in the AWTP. These variations could be closely related to the variations in the DO concentrations and ORP values shown in Table 1. After supply of oxygen by the aerator (st.3), As(III) present at st.2 and st.7 was almost completely oxidised to As(V), and the depletion of DO along the oxidation ditch has led to the partial reduction of As(V) to As(III). On the other hand, the sample at st.2 contained both wastewaters from the influent and from the return sludge pipe. In the latter, a reducing condition was created, as shown by the difference in the DO concentrations and ORP values between st.11 and st.2 (Table 1). Therefore, the high proportion of As(III) would be due to the reduction of As(V) present at st.11 to As(III) in the return sludge pipe. Unfortunately, it was impossible to collect samples

in the return sludge pipe to check the reduction process of As(V) through it.

It has been reported that the oxidation process of As(III) to As(V) via $O_{2(g)}$, and the abiotic reduction of As(V) via chemical processes [usually dissolved sulfide (H_2S , HS^-) as electron donor] in surface waters, at near neutral pH values, are generally slow^{31), 32)}. In the AWTP, although a high DO concentration was found at st.1 compared to st.3, complete oxidation of As(III) did not occur. At this station, the concentration of suspended solids, thus the microorganisms present, was relatively low when compared with the solid concentrations in the oxidation ditch and the return sludge pipe (Table 1). It appears from these observations that the distributions of As species in the treatment plant were not only dependent on the variations in DO concentrations but also significantly affected by the activated sludge. We have shown that the transformations of As species in activated sludge occurred only in the presence of living microorganisms and were accelerated by increasing the sludge concentrations from 100 to 700 mg/L as SS²⁰⁾. Therefore, it is also suggested that As(III) oxidation and As(V) reduction in the AWTP were microbially catalysed and were accelerated by higher concentration of sludge in the oxidation ditch (Table 1) compared with the batch experiments.

The transformations of As species were very fast and reversible, and no metabolite, such as the methylated forms of As, was produced at any stations. It is likely that the oxidation of As(III) and reduction of As(V) in the activated sludge could be a mean of detoxification processes¹⁹⁾. However, several isolated bacteria have been shown to gain energy for growth from the oxidation of As(III)^{28), 29), 33), 34)} or from the reduction of As(V)^{27), 35)}. Therefore, further investigation will be required to identify the As-oxidising/reducing bacteria in the activated sludge and to determine if energy is gained from the reactions.

(4) As removal from biologically treated wastewater by coagulation with ferric chloride

Watanabe et al.²⁵⁾ have reported that As-removal capacity of the AWTP is low (14% removal). On the other hand, it was shown in the previous paragraph that As concentration in the final effluent of the AWTP exceeded the Japan National Effluent Standard ($<100 \mu\text{g/L}$) and the Japan Environmental Quality Standard ($<10 \mu\text{g/L}$) for As. However, when the oxidation ditch is kept sufficiently aerated, dissolved As species present in the influent could be almost completely oxidised to As(V), and the kinetics of As(III) oxidation followed the Michaelis-Menten model. Therefore, the use of FeCl_3 as coagulant was tested in order to remove the As biologically oxidised by the activated sludge.

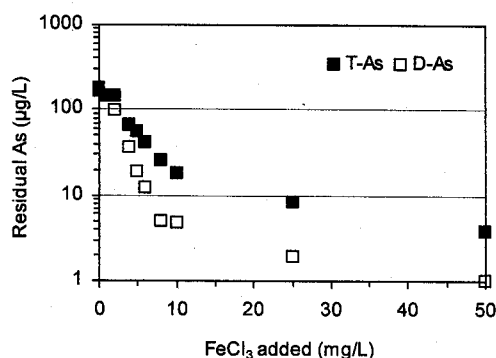


Fig. 8 Concentrations of residual As in the supernatant after settling as a function of FeCl_3 concentrations.

Under all experimental conditions, good separation of liquid and solid phase was observed after the settling time of jar-tests.

Fig. 8 represents the residual T-As and D-As in the supernatant after settling as a function of coagulant dosage. The initial T-As before jar-tests was $168 \mu\text{g/L}$. When no FeCl_3 was added, T-As in the supernatant was $163 \mu\text{g/L}$. As FeCl_3 concentration was increased, a decrease in residual T-As was observed. The required coagulant dosages to meet the effluent standard and environmental standard for As were 3 mg/L and 24 mg/L, respectively. 50 mg FeCl_3/L was sufficient to remove $159 \mu\text{g T-As/L}$, corresponding to 97.5% removal.

When residual D-As, rather than T-As, is considered, a lower dosage of FeCl_3 is enough to meet the Environmental Quality Standard. This shows that the amount of FeCl_3 used would be saved if the supernatant is filtrated before its release into the receiving water to remove the remaining P-As.

Hering et al.¹⁴⁾ have investigated the removal of As from freshwater systems by ferric chloride in bench-scale experiments. They have found that at a neutral pH, the addition of 4.9 mg FeCl_3/L could achieve nearly 100% removal at various initial As(V) concentrations up to $100 \mu\text{g/L}$, which is comparable to our results showing that a removal of $100 \mu\text{g T-As/L}$ could be achieved by the addition of 4.2 mg FeCl_3/L to the effluent containing various organic and inorganic matters.

4. CONCLUSIONS

In this study, it was shown that the activated sludge process could be effectively used to bio-oxidise As(III) to As(V) as a pre-treatment of As-contaminated wastewater. The main points drawn from this investigation are:

- (1) The kinetic data of As(III) oxidation and As(V) reduction could be fitted to the Michaelis-Menten model.
- (2) As(III) and As(V) were found to be similarly distributed as the main dissolved As species at the inlet of the Akiu-Onsen Wastewater Treatment Plant whereas As(V) was the dominant species at the final outlet (>98% of total species).
- (3) The activated sludge could not remove As from the wastewater efficiently. However, it can control the biotransformation of As species well. When the aeration tank was kept sufficiently aerated, the activated sludge process could be used as a reliable technology to rapidly bio-oxidise As(III) to As(V), reducing the toxicity of As in the effluent released to the receiving water body.
- (4) Additional treatment of the wastewater containing bio-oxidised As using coagulation process with FeCl₃ achieved high As removal efficiency (>95%). A residual As concentration less than 10 µg/L can be obtained by addition of 24 mg FeCl₃/L to the clarifier effluent (initial concentration: 163 µg/L).

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