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**FY2000  
RESEARCH SUMMARY  
OF  
WATER QUALITY CONTROL DEPARTMENT**

by

WASTEWATER SYSTEM DIVISION  
ADVANCED WASTEWATER TREATMENT DIVISION  
SLUDGE MANAGEMENT DIVISION  
WATER QUALITY DIVISION

National Institute for Land and Infrastructure Management,  
Ministry of Land, Infrastructure and Transport, Japan

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**Synopsis**

In this Research Summary, researches carried out by four Divisions of Water Quality Control Department in FY 2000 are briefly introduced.

Keywords : Wastewater System, Wastewater Treatment, Advanced Wastewater Treatment,  
Wastewater Sludge, Water Quality Control

## PREFACE

This Research Summary introduces briefly the researches carried out by the Water Quality Control Department of former Public Works Research Institute in Fiscal 2000. While the full content of each research is presented in Japanese in the Annual Reports of Water Quality Control Department, National Institute for Land and Infrastructure Management, Ministry of Land, Infrastructure and Transport (Technical Note of NILIM No.10), the abstract of each research is being published in both Japanese and English. Since we have several tens of foreign visitors to the Water Quality Control Department each year to exchange views on water quality control engineering each other, English version, though abstract, is considered useful for that.

Researches introduced in this Research Summary of FY 2000 were funded through Research & Study Expenses, Study Funds for National Pollutational Control Research Institutes, Environment Research Program Budget, Research Funds for Sewerage & Sewage Purification Programs, Research Funds for River Construction & Improvement Programs, Funds for Construction Technology Research and Development, Applications from Regional Development Bureaus, and Special Coordination Funds for Promoting Science and Technology. Although these funds have different purposes in each research from theory to practice, the results are obtained through intensive studies and are all useful to effectively promote water quality control. Hopefully, more intensive exchange of views in water quality control engineering is to be promoted through this Research Summary.



Eiichi Nakamura, Director

Water Quality Control Department

National Institute for Land and Infrastructure Management

Ministry of Land, Infrastructure and Transport

# CONTENTS

<b>Y.Inoue,Y.Suzuki,R.Uematsu,M.Suwa, R.Hiraide</b>	Sewerage system for tropical and subtropical countries	1
<b>Y.Inoue,R.Himono,T.Toyoda,M.Nose</b>	Rehabilitation of sanitary sewers	3
<b>Y.Inoue,M.Matsubara,T.Toyoda</b>	Utilization of urban stormwater in sewerage	5
<b>Y.Inoue,R.Uematsu,T.Yoshida, M.Morikane</b>	Benefit analyses of sewage works	7
<b>Y.Inoue,R.Himono,T.Toyoda,M.Nose</b>	Investigation of wastewater characteristics and deterioration of pipe system	9
<b>Y.Inoue,R.Himono,T.Toyoda,M.Nose</b>	Standard for discharge of domestic wastewater with high concentration	11
<b>Y.Inoue,Y.Suzuki,R.Uematsu, K.Takamura</b>	Optimum water cycle in urban areas	13
<b>Y.Suzuki,M.Ogoshi,H.Yamagata, S.Kim</b>	Biological decomposition of endocrine disrupting substances	15
<b>Y.Suzuki,T.Hatatsu,M.Nakamura</b>	Fate of pathogens in natural waters	17
<b>Y.Suzuki,T.Hatatsu</b>	Energy saving of nitrogen and phosphorus removal systems	19
<b>Y.Suzuki,M.Ogoshi,M.Suwa</b>	Risk management of pathogens in sewerage system	21
<b>Y.Suzuki,H.Yamagata,A.Kitanaka</b>	Removal of endocrine disruptors by wastewater treatment	23
<b>Y.Suzuki,M.Ogoshi,H.Yamagata</b>	Research on wastewater reuse system as an urban infrastructure	25
<b>H.Morita,S.Ochi,T.Kitamura</b>	Control of pathogenic microorganisms in wastewater system	27
<b>H.Morita,Y.Kawashima,Y.Ikeda</b>	Control of heavy metals in sludge treatment process	29
<b>H.Morita,Y.Kawashima</b>	Unit environmental load for LCA of sewerage system	31
<b>H.Morita,S.Ochi,T.Kitamura</b>	Identification of pathogenic microorganisms in wastewater system	33
<b>H.Morita,S.Ochi,M.Minamiyama</b>	Behavior of endocrine disruptors in sewage sludge	35
<b>M.Takahashi, Y.Suzuki,H.Morita,S.Ochi, R.Hiraide</b>	Reducing CH <sub>4</sub> and N <sub>2</sub> O gas emission from wastewater treatment facilities by improving applicability of core controlling technologies	37
<b>H.Morita,S.Ochi</b>	Systems for using the self-energy of sludge	39
<b>H.Morita,H.Saino</b>	Sewage sludge treatment with other sludge which differs from sewage sludge	41
<b>H.Morita,H.Tanaka, S.Ochi,M.Minamiyama,K.Komori, Y.Okayasu</b>	Development of technology for dioxin contaminated soil countermeasures	43
<b>H.Tanaka,K.Komori,M.Shirasaki, H.Ishii,K.Taketoshi</b>	Effects of sewerage in watershed	45
<b>H.Tanaka,A.Takahashi,Y.Yakou, M.Saito,T.Higashitani</b>	Evaluation methods of contamination discharged from urban wastewater affecting source water of drinking water supply	47
<b>H.Tanaka,K.Komori,M.Shirasaki, H.Ishii</b>	Research on the alteration of endocrine disruptors in water environment	49
<b>H.Tanaka,K.Komori,A.Takahashi, Y.Okayasu,Y.Yakou</b>	Novel technology for detection of micropollutants in wastewater	51
<b>H.Tanaka,K.Komori,Y.Okayasu, K.Taketoshi,Y.Yakou,T.Higashitani</b>	Technological development for the prevention of hazardous substances in sewerage	53
<b>H.Tanaka,K.Komori,A.Takahashi, Y.Yakou,M.Saito,T.Higashitani</b>	Evaluation of aquatic ecosystems affected by sewerage systems	55
<b>H.Tanaka,M.Shiraisaki,H.Ishii, K.Taketoshi</b>	Systematic way of surveying water quality of rivers	57

# SEWERAGE SYSTEM FOR TROPICAL AND SUBTROPICAL COUNTRIES

Y. Inoue, Y. Suzuki, R. Uematsu, M. Suwa and R. Hiraide

Wastewater System Division, Advanced Wastewater Treatment Division

Project period : 1996-2000

## OBJECTIVES

As a result of rapid economic growth, urbanization and shortage of sanitary facilities, the environmental deterioration became very serious in developing countries in tropical and subtropical regions. To restore this situation, the environmental sector in ODA is highly demanded by the developing countries. The aid on planing and construction of sewerage in the above countries, which is one of the most important works in environmental sector, has been expected more and more. The technology of sewerage originated from Japan is in high standard. However, it is not always appropriate to adopt it to developing countries, considering the respective natural condition, social / economic background and so on. Therefore this study aims to establish the most suitable sewerage system for developing countries in tropical and subtropical regions.

## RESULT

### SELECTION OF WASTEWATER TREATMENT PROCESSES

For developing countries in tropical and subtropical regions, in case of selection on treatment processes, given conditions such as the climate, effluent quality standard, affordable investment (initial and maintenance cost) and acquired land area are different from those for industrial countries. Therefore these developing countries have mainly applied stabilization pond, activated sludge, trickling filter, oxidation ditch and aerated lagoon processes. We analyzed these processes from the viewpoints of construction as well as maintenance / operation. Also we compiled design methods for stabilization pond and aerated lagoon processes, which are not common in Japan.

In stabilization pond process, anaerobic, facultative and maturation ponds are often used in series. We analyzed each characteristic and design method. Especially, there are many models for the design of facultative ponds. Some are complex and rely on data difficult to obtain. The most widely used models in practice are those of surface loading approach (including McGarry & Pescod equation and Mara equation) and Gloyna equation.

Aerated lagoon process consists of complete mix aerated and partial mix aerated pond system. Solids in the complex mix aerated pond are kept suspended at all times. Oxygen requirements can be estimated using equations based on mass balances; however, the power input necessary to keep the solids suspended is much greater than that required to transfer adequate oxygen. On the other hand, in the partial mix aerated pond, no attempt is made to keep all solids suspended, therefore aeration serves only to provide oxygen for transfer. BOD removal and characteristics of both ponds are generally estimated using complete mix hydraulic model and first order reaction kinetics. The only difference is that reaction rate applicable to partial mix system is smaller than that to complete mix system.

## APPLICATION OF FACULTATIVE AND AERATED LAGOON PROCESSES

### 1) Facultative lagoon

The facultative lagoon was operated at the HRT of 30 days and BOD loading per unit area of 22.7 kg/(m<sup>2</sup> d). String type contact media was installed in the facultative lagoon during the latter half of experiment period. Average water quality of wastewater influent and removal efficiency are shown in Table 1. The removal efficiency of high water temperature time was better than that of the low time. The difference was 10% for BOD or COD and 20% for T-K-N or NH<sub>4</sub>-N, PO<sub>4</sub>-P. And the removal efficiency of the facultative lagoon with the contact media was improved.

Table 1 Influent quality and removal efficiency of facultative lagoon

	Influent (mg/L)	Removal efficiency of facultative lagoon (%)				
	Average concentration	All period	High water temperature	Low water temperature	With contact media	Without contact media
T-BOD	342.0	79.3	81.3	72.4	85.9	75.0
D-BOD	178.0	81.5	84.9	69.0	91.5	78.8
T-COD	461.0	61.2	66.7	49.1	65.3	54.6
D-COD	145.0	54.5	55.6	44.1	62.8	41.5
T-K-N	42.0	50.0	59.3	41.6	43.8	42.7
NH <sub>4</sub> -N	31.0	62.6	72.8	48.1	54.1	54.5
PO <sub>4</sub> -P	2.8	75.0	79.3	56.3	77.4	57.1

### 2) Aerated lagoon

The aerated lagoon was operated at the HRT of 5 to 15 days and the mixing power was from 30 to 60 Hz. The treated water quality was always under BOD 30 mg/L. High removal efficiency of T-COD and PO<sub>4</sub>-P was obtained during low mixing power time, because the suspended solids in reactor were settled. NH<sub>4</sub>-N concentration of treated water increased when the HRT was 5 days, because the HRT was not enough for the growth of nitrifiers (Table 2).

Table 2 Influent quality and removal efficiency of aerated lagoon

	Influent (mg/L)	Removal efficiency of aerated lagoon (%)				
	Average concentration	All period	HRT 15 days Mixer 60Hz	HRT 10 days Mixer 60Hz	HRT 10 days Mixer 30Hz	HRT 5 days Mixer 60Hz
T-BOD	344.0	82.7	91.7	91.7	91.7	—
D-BOD	180.0	86.6	99.1	97.9	95.0	—
T-COD	454.0	65.0	69.0	57.7	73.0	61.3
D-COD	150.0	67.5	77.8	76.1	71.2	63.4
T-K-N	40.0	57.6	79.4	74.7	68.4	46.4
NH <sub>4</sub> -N	31.0	70.6	98.4	98.8	84.9	54.9
PO <sub>4</sub> -P	2.8	46.4	42.9	46.7	73.3	37.0

# REHABILITATION OF SANITARY SEWERS

Y. Inoue, R. Himono, T. Toyoda, and M. Nose

Wastewater System Division

Project period: 1900 - 2000

## OBJECTIVES

Most cities have adopted separate sewer systems in recent years in Japan. By type of collection system, 2,113 cities had the separate sewer system and 24 cities had the combined sewer system at the end of 1998. At sewage facilities using the separate sewer system, various problems such as pipe corrosion by hydrogen sulfide, infiltration of storm water and underground water into pipes, and earth and sand accumulation in pipes occur as time elapses. Such problems are likely to increase as facilities become older. This study aims to clarify the causes of troubles that are not fully understood in sanitary sewers at present. The study also examines effective countermeasures for the principal troubles of sanitary sewers.

## RESULTS

### (1) Measurement of effect of countermeasures of infiltration of storm water

The effects of countermeasures for infiltration of storm water such as repair works were assessed by measuring the quantity of infiltration of storm water during rainfall (rate of infiltration) in the zone (99 houses, 2.5 ha) in a certain city.

Repairing house inlets and house connections was found to reduce the rate of infiltration by 73%, thus confirming the effect of such countermeasures.

### (2) Modeling of the infiltration of storm water

The rainfall-runoff simulation model (X P - SWMM) was applied to the reappearance of infiltration to the sewage pipe in the zone of the city.

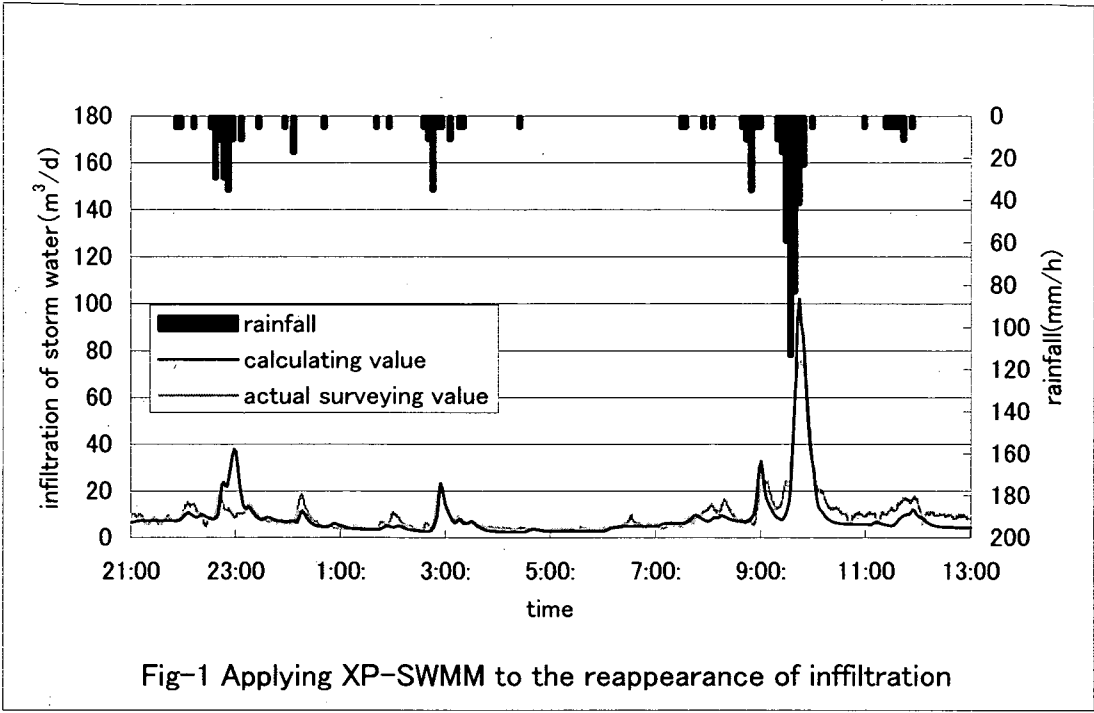
The height and time of the peak of infiltration could be reproduced faithfully by adjusting parameters such as impervious area rate and roughness coefficient as shown in Figure 1.

### (3) Effect on suppression of sulfide formation in the sewage pipe by nitrate injection

Nitrate injection is one method of suppressing sulfide formation in sewage pipes. We examined how to economically and effectively control the amount of nitrate injection by measuring the action of the sulfide and nitrate nitrite nitrogen (NOx-N) in a regional sewage pipe.

Nitrate injection was done at the site of a pump, and the action of sulfide and NOx-N were measured at a manhole 7249 m downstream of the pump. Sulfide was detected when there was no NOx-N in the manhole, but sulfide was not formed when there was more than 0.5 mg/l NOx-N in the manhole.

Thus, when controlling sulfide formation by nitrate injection, the proper amount of injection can be maintained by monitoring NOx-N concentration in the location where hydrogen sulfide occurs.





# UTILIZATION OF URBAN STORMWATER IN SEWERAGE

Y. Inoue, M. Matsubara and T. Toyoda

Wastewater System Division

Project period: 1998 - 2000

## **OBJECTIVES**

In this study, we are focusing on stormwater utilization that has many effects. Considering stormwater as a resource of sewerage, we will solve technical problems for increasing the use of stormwater, especially from the viewpoint of sewage works. We will also quantitatively evaluate the effects of inundation control and CSO control by stormwater utilization, and will investigate the characteristics of stormwater runoff and pollutant load discharges in wet weather conditions to draw up suitable plans for inundation control and CSO control.

The main results obtained in FY 2000 are as follows.

## **RESULTS**

### **QUANTITATIVE EVALUATION OF STORMWATER UTILIZATION IN URBAN AREA**

We investigated the effects by conducting simulations for a typical city (land use, population density, etc.)

#### **(1) Effect of Inundation Control**

Stormwater utilization is not particularly effective, especially when the storage capacity is not sufficient. It seems to be effective for inundation control to decrease the storage level for utilization in the rainy season.

#### **(2) Effect of CSO Control**

Even if the storage capacity is not so large, stormwater utilization effectively decreases the number of overflows, especially in case of low rainfall events (less than 5 mm/hr). As a result, we can estimate that stormwater utilization has a greater effect on a pollutant load basis than a water volume basis for CSO control.

### **CHARACTERISTICS OF POLLUTANT LOAD DISCHARGE IN COMBINED SEWER SYSTEMS**

The Japanese government recognizes that CSO control is important to preserve the quality of receiving waters, and is discussing a new policy to implement CSO countermeasures more effectively. But many cities in Japan do not understand the characteristics of their sewer systems in wet weather conditions, and do not have sufficient data of sewer systems. The objective of this study is to identify the characteristics of pollutant load discharge and sewer deposit load through monitoring and modeling.

We conducted field surveys by setting three sites, two sites for flow monitoring and sewage sampling points (located at the end of a trunk sewer (171 ha) and at the middle of a trunk sewer (34 ha)), and the third one for rainfall monitoring. Analyzed parameters for sewage quality were BOD, COD<sub>Mn</sub>, SS, total nitrogen (TN), and total phosphorous (TP). Sewage was sampled during 5 rainfall events and 5 days of dry weather.

A comparison of wet weather loadings with dry weather loadings produced useful information.

- (a) In terms of pollutant load discharge from CSSs, surface (road, roof, etc.) residual loads are negligible in comparison with sewer deposit loads for BOD, TN and TP.
- (b) The ratio of sewer deposit loads of TN and TP to total daily loads in dry weather is smaller than that of BOD.
- (c) The ratio of sewer deposit loads of upstream areas to total daily loads in dry weather is larger than that of the whole study area for all parameters analyzed.

The PWRI (Public Works Research Institute) load model has been a principal model used to calculate pollutant load discharges from CSSs in Japan. By reflecting the result of (a) on the configuration of the model, the trends of TN and TP were well demonstrated. The model parameter Pp (sewer deposit load) that was calibrated by each rainfall event showed the same results as (b) and (c).

### HYDRAULIC CHARACTERISTICS OF PRESSURIZED STORM SEWERS

In order to prevent blowing-out of manhole covers and to eliminate accidents caused by entrapped air, the hydraulic characteristics of pressurized storm sewers must be investigated.

We constructed three models (1/1, 1/2, 1/4 scale) for hydraulic experiments to investigate the characteristics of water and airflow in various conditions. We can measure such items as water level (pressure), air pressure, flow rate of water, and flow rate of air through these models. Scale effects of air, design method for ventholes and design methods for pressurized storm sewers will be investigated hereafter.

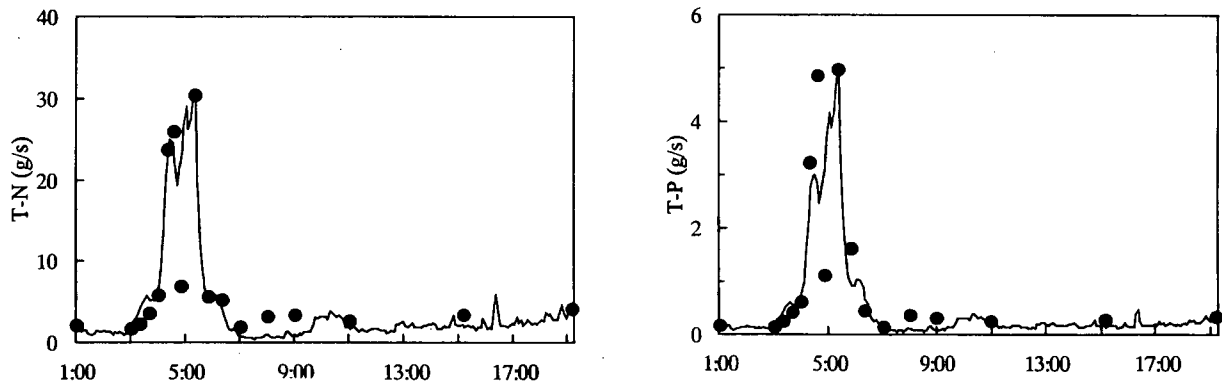


Fig. 1 Examples of the results of calculation by PWRI load model

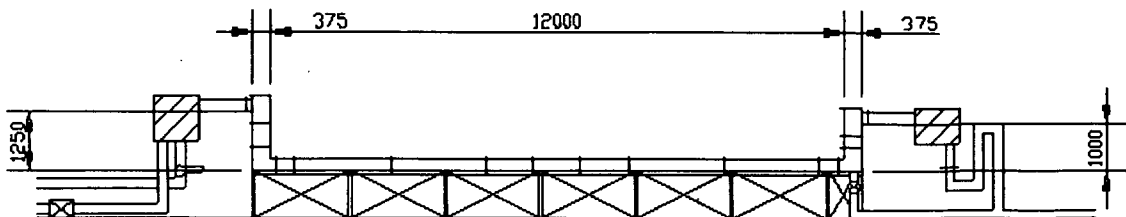


Fig. 2 Schema of experimental model (1/2 scale,  $\phi$  250 mm)

# BENEFIT ANALYSIS OF SEWAGE WORKS

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Wastewater System Division

Project period: 1998 - 2001

## OBJECTIVES

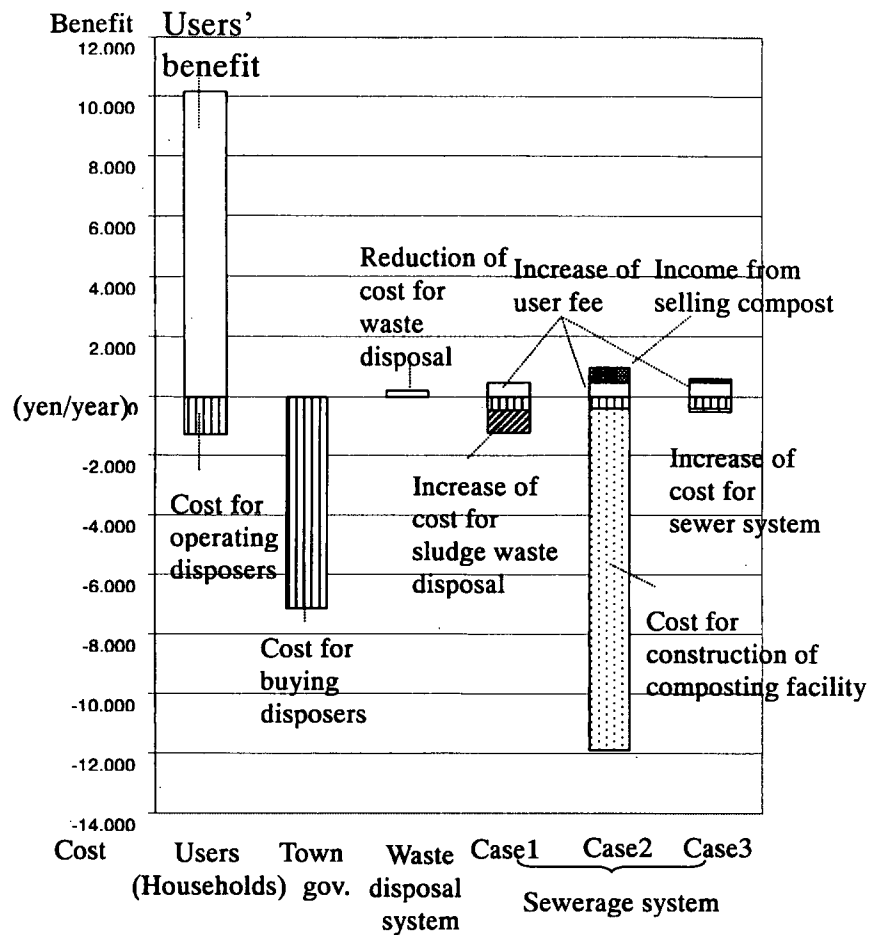
Food waste disposers, which are not allowed to be installed in areas served by sewage works in Japan, might adversely affect sewage systems. However, they could benefit users, reduce waste disposal costs, and have some environmental effects such as on the emission of CO<sub>2</sub> and recycling of waste. We carried out an economic evaluation of a test case of installing disposers in separate sewer system in Utanobori-cho, Hokkaido prefecture. Various effects of installing disposers were valued in monetary terms, and the benefit to users of disposers was valued using the contingent valuation method (CVM). Other effects on sewage and waste disposal systems were simulated based on reported data before starting to install disposers.

## RESULTS

The effects of installing disposers in Utanobori-cho were valued in monetary terms for each stakeholder, as shown in the figure. These were estimated assuming that disposers were installed in all houses in the area served by sewage works. The figure shows that the users' benefit would be larger than the total cost of sewage and waste disposal systems and buying disposers. The effect on the waste disposal system would be slightly positive, but the effect on sewage works systems would be negative because the increase in maintenance cost would be offset by the increase in sewer fees due to the larger volume of wastewater but the cost of disposal of sludge wastes would greatly increase without reusing them as a compost (case 1). If sludge wastes were reused as a compost, the cost of constructing the composting facility would be high (case 2), but the cost of disposing of sludge wastes could be reduced by reusing the sludge wastes (case 3).

The benefit of disposers to users was surveyed by CVM, and the willingness to pay (WTP) for using disposers for each household was estimated to be 950 - 1,260 yen on average per month.

These results were estimated based on the present situation of Utanobori-cho; the effect of disposers on sewage works and waste disposal systems remains unknown. These results are based on a case study and economic evaluation at present, and the effects of disposers should be monitored as their usage spreads.



**Figure** Cost-benefit analysis of installing disposers in Utanobori-cho  
(assuming 100% installation in sewerage system)

# INVESTIGATION OF WASTEWATER CHARACTERISTICS AND DETERIORATION OF PIPE SYSTEM

Y. Inoue, R. Himono, T. Toyoda, and M. Nose

Wastewater System Division

Project period: 2000 – 2002

## OBJECTIVES

Sewer systems serve 60% of the population, and emphasis on wastewater facilities is changing from construction to operation and maintenance. The length of sewer pipes has increased greatly in accordance with the increase in percentage of sewered population. Separate sewer systems were commonly used in sewerage facilities built in the mid 1960s to 1970s, which account for most of the pipe facilities now in existence. However, the characteristics of wastewater from the separate sewer system are different from those of the combined sewer system, and dilution of sanitary wastewater and flushing of the inside of pipes in rainy weather cannot be expected. Therefore, deterioration of the separate sewer system is likely to progress more easily in comparison with the combined sewer system. When a sewer system has deteriorated, renewal plans have been using knowledge gained in big cities. However, it is necessary to grasp the deterioration mechanism in order to draw up repair technology and renewal plans for separate sewer systems. Therefore, the deterioration of the sewer system must be investigated for acquiring knowledge about the influence of sanitary wastewater.

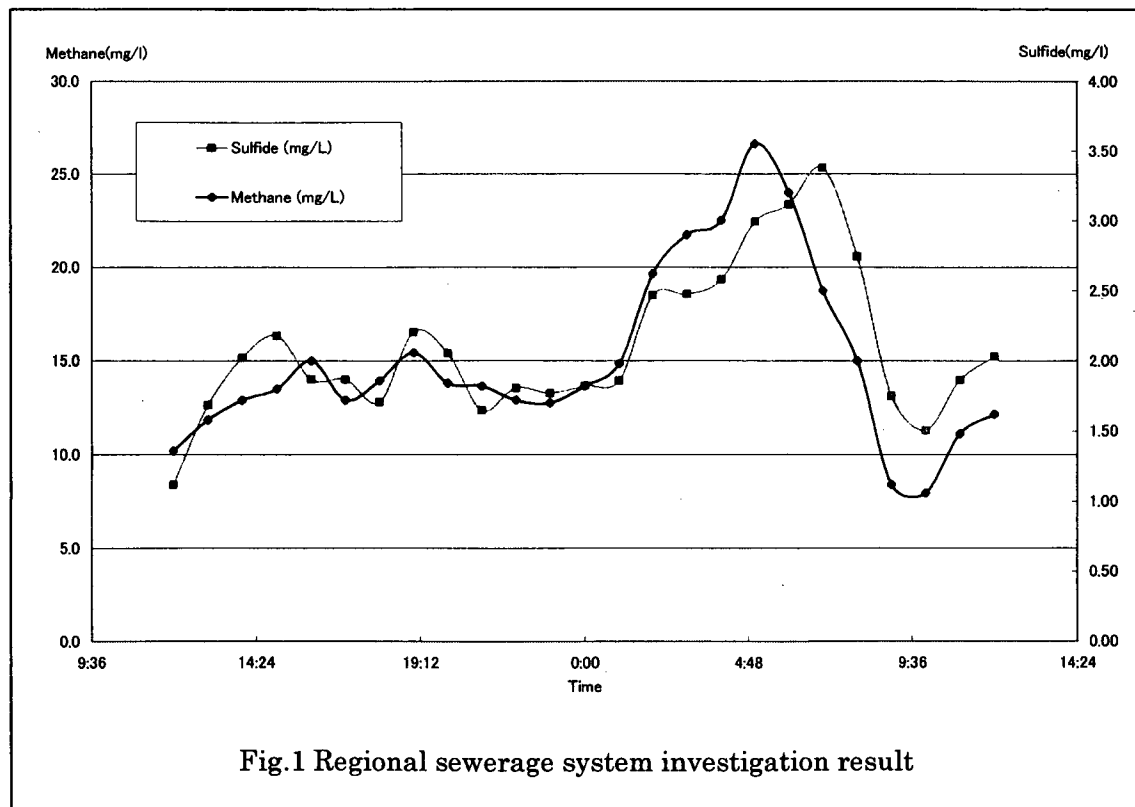
## RESULTS

### 1. Questionnaire survey on the sewer system of the regional sewerage system

A questionnaire survey on the sewer system in the regional sewerage system in the whole country was conducted. Answers were received from 149 treatment districts which had begun use by March, 1999. Treatment districts that reported deterioration of the sewer system accounted for 31% of the total. However, only 58% of treatment districts conduct corrosion investigations, and yet corrosion soon occurs in most treatment districts. There were many occurrences of corrosion in the downstream release point of the main sanitary wastewater. Sanitary wastewater generally becomes anaerobic and sulfide is likely to be formed. Furthermore, corrosion was discovered within 6 years in 40% of the treatment districts that carried out investigations, and rapid corrosion of the sewer system was reported to be likely.

## 2. Investigation of change in water quality of sanitary wastewater

The change in water quality of sanitary wastewater in conduits was investigated for the regional sewerage system. The longest conduit of the regional sewerage system is 56.9 km, and the average planned length is 20.6 km from the questionnaire survey. An example of the change in water quality inside the sewer system of the regional sewerage system is shown in Fig. 1. A change in time is shown about the sulfide and methane, which occurs when sanitary wastewater becomes anaerobic. It is put early in the morning from the midnight, and concentration rises with both materials as well. Because the sanitary wastewater flow rate is smallest in this time zone, sanitary wastewater is likely to become anaerobic upstream of the pumping place.



# STANDARD FOR DISCHARGE OF DOMESTIC WASTEWATER WITH HIGH CONCENTRATION

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Wastewater System Division

Project period: 2000 - 2004

## OBJECTIVES

The quality of wastewater discharging to the sewerage system is expected to change as living standards improve. In this research, we focus on wastewater from disposers installed in Utanobori, Hokkaido Prefecture, and examine their effects on sewers in terms of water quality and amount of sediments. Based on the results, a standard for the effluent from disposers to the sewerage system is proposed.

## RESULTS

The flow rate of sanitary wastewater and changes in its nature were researched in the pipe before and after installing a disposer, to grasp the load of the drainage from the disposer. Sedimentation inside the pipe is expected to increase as flow velocity reduces, so the conditions of accumulation were investigated by a television camera, the amount of accumulation was measured. Hydrogen sulfide is expected to form in the pipe due to the increased accumulation, so hydrogen sulfide was also measured.

### 1. Water measurements

Water was sampled for 24 hours in a zone where disposers had not been installed to investigate the influence on water quality. The load is calculated from flow rate and water measurements, as shown in Figure 1. There was no special difference though the load of the establishment zone, the non-establishment zone was compared. But when the load was calculated from the average flow rate for the year, the load of the zone where disposers were installed showed a high value of about 1.7 - 2.7 times that where disposers had not been installed.

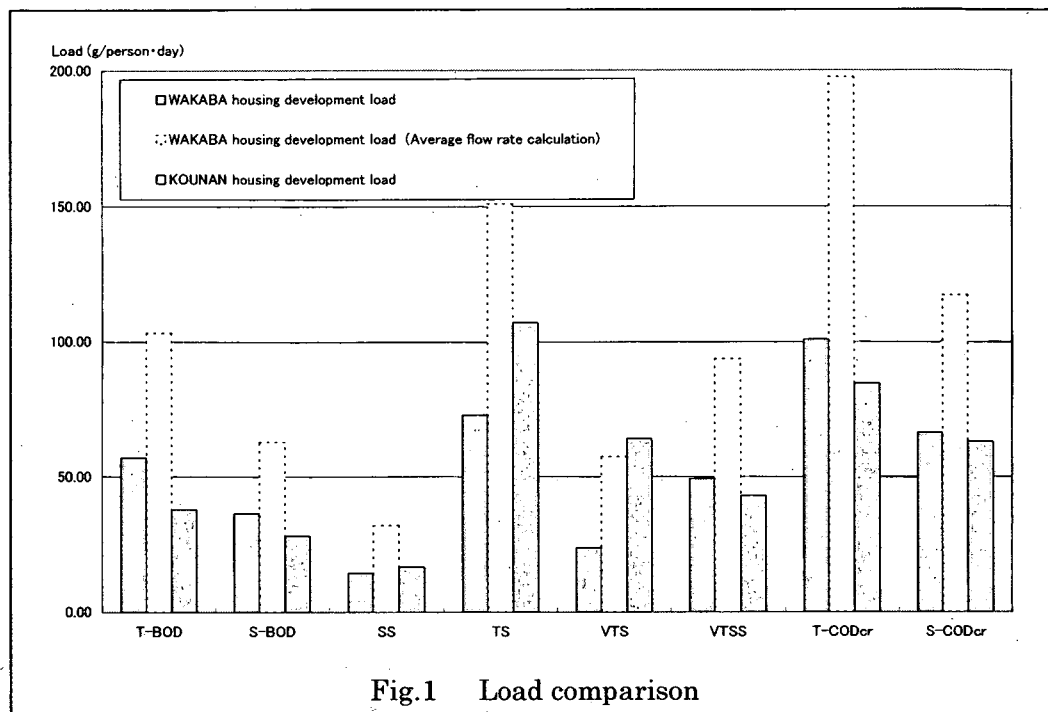
### 2. Hydrogen sulfide measurements

Hydrogen sulfide measurements showed that hydrogen sulfide did not form under usual flow conditions. However, when some pools occurred in the pipe, a small quantity of hydrogen sulfide formed in the disposer zone, so the installation of disposers is likely to lead to an increase in the formation of hydrogen sulfide.

### 3. Sediment measurements

A clear accumulation was confirmed by investigating the sediments by television camera in the disposer establishment zone. In the zone where disposers had not been installed, little accumulation was found. A simple comparison cannot be done based on factors such as the design of the pipe, but it was clear that more sediment accumulated in the zone where disposers had been installed. An

investigation of the sediment showed that there were more organic substances in the disposer establishment zone than in the zone where disposers had not been installed. Many egg shells were found in the sieve, which were likely to have come from the disposers.





# OPTIMUM WATER CYCLE SYSTEM IN URBAN AREAS

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Wastewater System Division, System Development Division

Project period: FY2000-2003

## OBJECTIVES

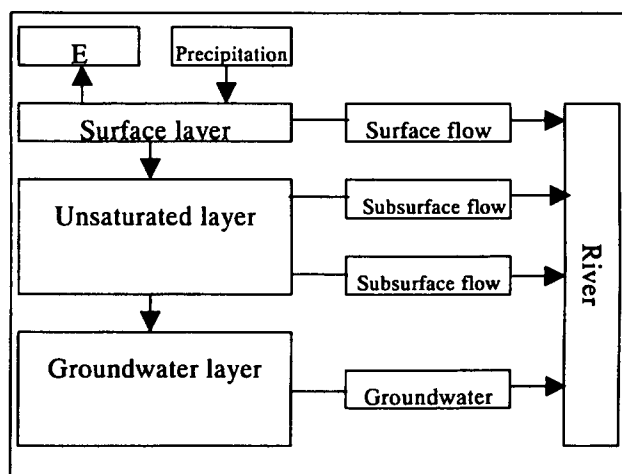
The sewerage system is one of the most important infrastructures in terms of improving living conditions, preventing inundation and conserving public water bodies. As the result of sewerage development, most water (wastewater or stormwater) in urban areas goes to public water bodies through the sewerage system. Recently people are showing more concern about environment (including water environment), which may offer higher quality of life. Therefore the sewerage system is required to play more active role in maintenance or restoration of a sound water cycle. Various efforts such as advanced treatment, CSOs control, utilization of treated wastewater, NPS control and infiltration / storage of stormwater are adopted. This study aims to establish the optimum water cycle system in urban areas through analyzing the most effective combination of the above measures in order to achieve desirable water quality/ quantity, considering cost performance

## RESULT

### INVESTIGATION ON HYDROLOGICAL MODELS

This fiscal year (2000) is the first one for the research period. Therefore we analyzed some hydrological models as methods for evaluating water cycle system quantitatively and selected one model to be adopted in a real watershed for the further research.

The run-off (water quantity) consists of surface runoff, fast subsurface runoff, slow subsurface runoff and groundwater runoff. Generally surface runoff / fast subsurface runoff are called as direct runoff, and slow subsurface runoff / groundwater runoff are called as base flow. The run-off models can be classified into lumped models or distributed models. The most common lumped one is so-called tank model. Distributed ones are generally modification of tank model (we call non-physically-based distributed model) or physically-based distributed models which solve



E: Evapotranspiration

Fig.1 Water circulation flow in river basin

equations by a numerical method. In most of run-off models of water pollutants into rivers, run-off pollutants accompanied by base flow are not taken into account, although not only point source but also non-point source

pollutions are considered. In this research, we examine water quality model including run-off pollutants by every flow in line with the above run-off (water quantity) model.

For the further research, non-physically-based distributed model will be adopted. Because it is impossible to evaluate change in water quantity and quality in watersheds resulted by proposed sewerage measures through using lumped models. In addition, it is complicated and difficult to analyze both of water quantity and quality by physically-based distributed model, considering necessity of establishing many parameters.

### ANALYSYS ON CONSTRUCTION COST OF PIPE SYSTEM

We estimated a construction cost of small sewer pipe every five years since 1985 based on the general design /working condition, considering the then estimate standard. Fig.2 (a) shows changes in the unit price of small pipe construction cost by open cut method. The cost increased from 1985 and reached about 1.5 times in 1995 and decreased by about 10% from 1995 to 2000. This change resembles a pattern of price movement in Japan.

In order to except influence of the price movement, we recalculated a construction cost of small sewer pipe using the price in 2000 (Fig.2(b)) . Earthwork cost increases by about 40 %, because excavation width increased due to revised estimate standard. Laying pipe work cost decreases by about 30%. Because a used pipe turned into a polyvinyl chloride pipe from concrete pipe, and a minimum diameter of sewer pipe turned into 200mm from 250mm.

On this calculation, the price movement influences a construction cost of small sewer pipe greatly. Besides the price movement, change of the estimate standard related to Earthwork affect the total cost much.

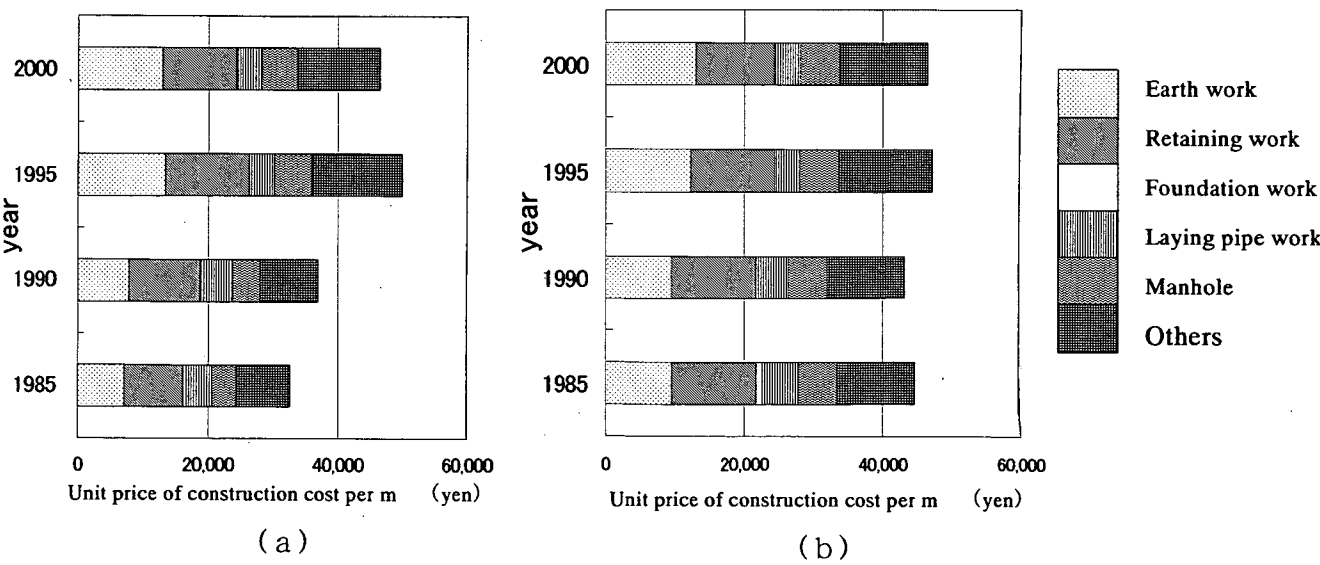


Fig.2 Cost of small pipe system by the open cut method

# BIOLOGICAL DECOMPOSITION OF ENDOCRINE DISRUPTING SUBSTANCES

Y. Suzuki, M. Ogoshi, H. Yamagata and S. Kim

Advanced Wastewater Treatment Division

Project Period: 1999 – 2000

## Objectives

Nonylphenol (NP) and 17  $\beta$ -estradiol (E2) were commonly detected in the final effluent of municipal wastewater treatment plant as the endocrine disrupting substances. So, these two substances are supposed to be degraded insufficiently by activated sludge. This study objects to find the fundamentals of removal and biological degradation of these two substances and also to find special microorganisms that can decompose these materials effectively. According to the result of the study in 1999, some microorganisms, which can decompose NP or E2, were separated from activated sludge. And most E2 degrading microorganisms have the ability to change E2 into estron (E1). The annual targets in the fiscal 2000 are as follows.

- 1) To find the special microorganisms which have the metabolizing ability of E1.
- 2) To identify the group of these microorganisms separated from activated sludge.
- 3) To find the accumulated part of these substances, if they are not metabolized in the activated sludge.
- 4) To examine the removal efficiency of these two substances by RO process.

This study project started in 1999, as a part of the comprehensive studies and developments on the water quality risk management in the aquatic environment.

## Methods and the results of search

The high E1 metabolizing microorganisms were searched from stored cultures, which were separated in the last year as E2 degrading microorganisms from the activated sludge in some MWTP. And we found some E1 degrading microorganisms.

According to the result of the morphological, kinematical and biochemical examinations, we identified the group of each two of the typical NP, E2, and E1 degrading microorganisms, as shown in Table 1.

About the accumulation part of NP, estrogen (E2 and E1) and related components in the activated sludge, we tried two kinds of step-by-step extraction methods, indicated in Fig.1. And the result of these examinations indicated the clear difference between NP and estrogen. That is to say, NP is accumulated in the activated sludge with fatty materials, but estrogen is seemed to be degraded and is not accumulated in the activated sludge like NP. And also there were differences among NP, NPEO and NPEC, about the accumulation part in the activated sludge, as shown in Fig.2.

Thinking from these experimental results, the removal of NP and relating substances are more important than the removal of estrogen. Because NP and relating substances are not well degraded by the activated sludge

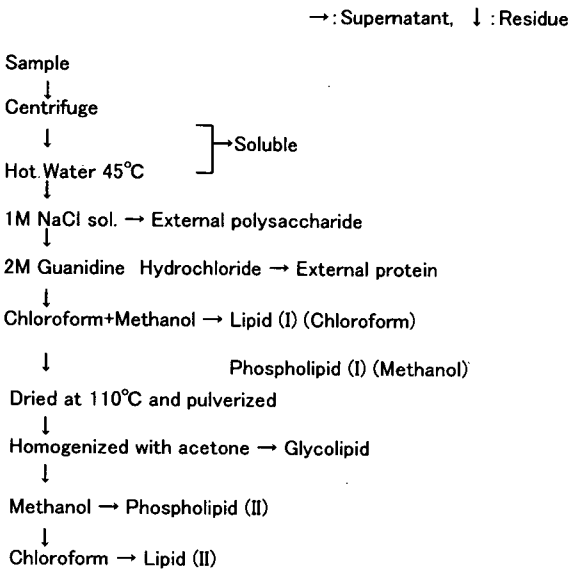
treatment in MWTP, and the sludge will become another pollution source of NP.

The basic removal efficiencies of NP and E2 by the RO treatment were examined using a loose RO membrane and two different NF membranes. The loose RO membrane indicated high removal efficiencies to both NP and E2 in the pH range from 5 to 9. But the removal rate of those two different NF membranes depended on pH, and their removal rates were relatively lower than that of loose RO membrane in the same pH range.

Table 1 Identification Result of EDs Degrading Microorganisms

Degrading Component	E2		E1		NP	
Type No.	T45	T31	F5	T53	T3N2	FN1
Identified Genus	<i>Rhodococcus</i>	<i>Candida</i>	<i>Micrococcus</i>	<i>Rhodococcus</i>	<i>Micromonospora</i>	<i>Moraxella</i>

Extraction Method I



Extraction Method II

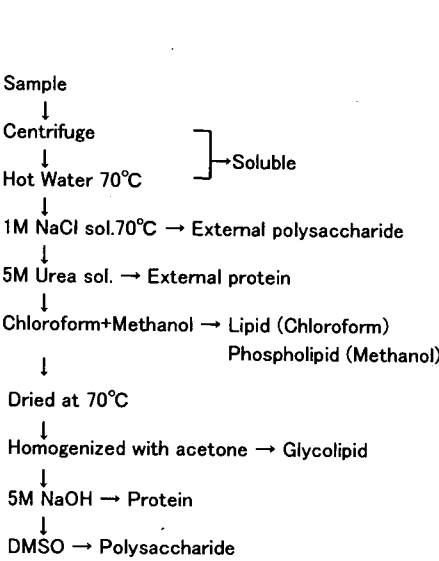


Fig.1. Extraction Method of NP and Estradiol in the Activated Sludge

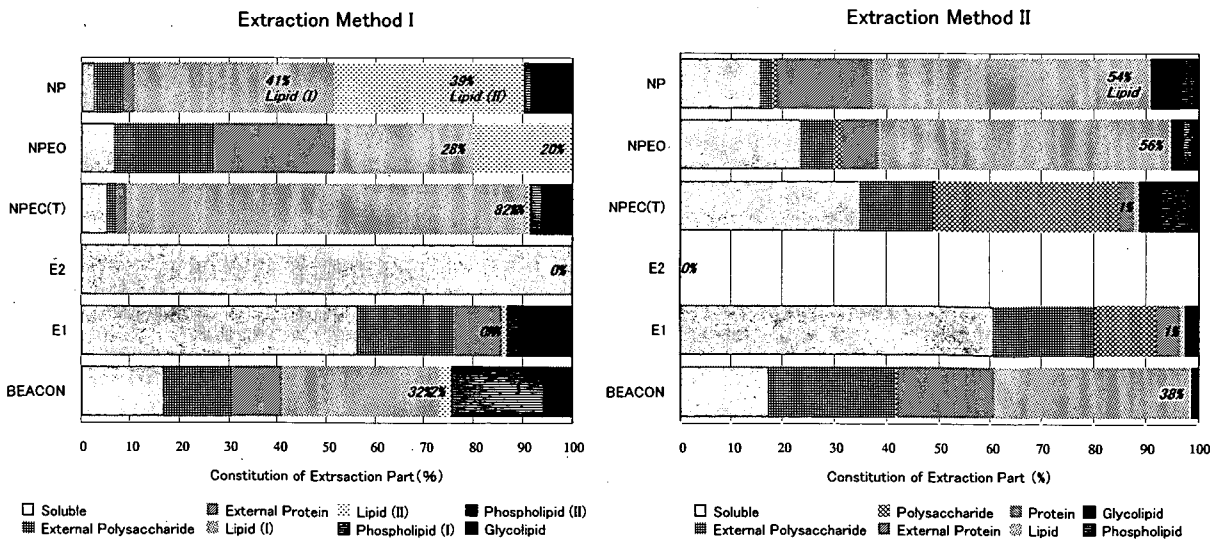


Fig.2. Result of EDs Extraction Test from the Activated Sludge

# FATE OF PATHOGENS IN NATURAL WATERS

Y. Suzuki, T. Hatatsu, and M. Nakamura  
Advanced Wastewater Treatment Division

Project period: 1998-2001

## OBJECTIVES

Construction of wastewater systems has improved public health, and decreased water-borne disease occurrence. Recently, people contact treated wastewater more frequently, because the number of wastewater treatment plants upstream of rivers is increasing and wastewater reuse is in progress. In addition, new water-borne diseases are coming out. Standards of treated wastewater quality have to be reviewed for safety. The objective of this study is clarifying the fate of pathogens in natural water. In this year, we evaluated the behavior of bacteria in the experimental channel, and applied RT-PCR (Reverse Transcription – Polymerase Chain Reaction) method to detect viruses in treated wastewater.

## RESULTS

### (1) The behavior of bacteria in the experimental channel

We experimented the fate of bacteria in the channel by measuring bacteria concentration along the flow. About 33% of *E.coli* that flowed in were removed in this process, and 99.9% of them were inactivated. (See Figure-1 )

Assuming the removal rate is proportional to the number of bacteria (Eq-1), the decrease velocity (k) was calculated at certain points of time.

$$\frac{dN}{dt} = -kN \quad \text{Eq-1}$$

N: number of bacteria

t: time

k: decrease velocity, time<sup>-1</sup>

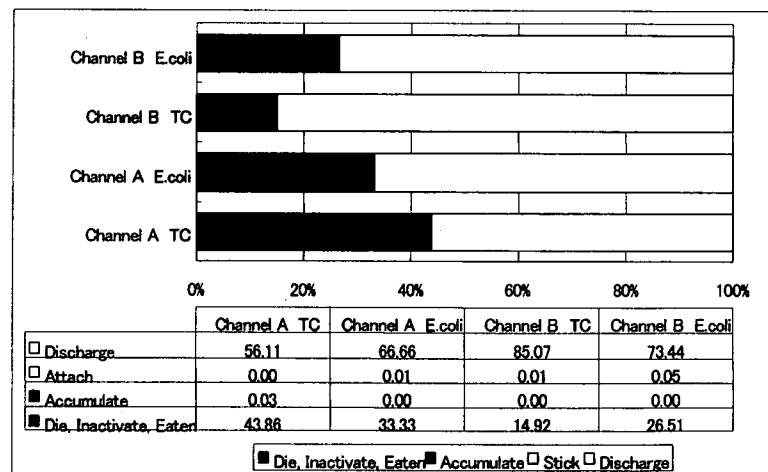


Figure-1 Mass balance of the *E.coli* in channel

The decrease velocity became smaller as time passed. In the case with gravel bed, the decrease velocity of *E.coli* was 0.08h<sup>-1</sup>, which was higher than the natural decrease velocity of 0.04h<sup>-1</sup>. Channel bed had influence on the decrease velocity.

### (2) Virus detection in treated wastewater by RT-PCR method

RNA coliphage Qβ was injected into treated wastewater, and detected by RT-PCR method. The results are as follows;

- The analytical detection limit of Q $\beta$  RNA by RT-PCR method was 0.5log higher in secondary or tertiary effluent compared with in Milli-Q water. This is because some kinds of substances in the treated wastewater disturb the enzymes for RT-PCR reaction.
- In treated wastewater and under high temperature, Q $\beta$  RNA concentration got lower significantly. It is suggested there are a lot of RNase (a kind of protein which decomposes RNA) in the secondary and tertiary effluent.
- To purification and extraction of Q $\beta$  RNA in treated wastewater, a commercial kit containing protein denaturant was applied. With the kit, RNA was extracted under ambient temperature, and most RNase was inactivated. The kit was effective to extract RNA with no decomposition.
- The applied commercial kit could not remove substances which disturb the RT-PCR enzymatic reaction.

# ENERGY SAVING OF NITROGEN AND PHOSPHORUS REMOVAL SYSTEMS

Y. Suzuki, T. Hatatsu

Advanced Wastewater Treatment Division

Project period: 1998-2001

## OBJECTIVE

Because nitrogen and phosphorus removal systems consume more energy than conventional activated sludge systems, the load to the global environment in the form of green house effect gas increases though the pollutant load to the local environment is reduced. For the purpose of reducing energy consumption of nitrogen and phosphorus removal systems, we investigated operation and design methods to improve energy efficiency of each process and the total system.

## RESULTS

(1) In the operating conditions, measurement of  $K_La$  was performed by three kinds of steady state methods.  $K_La$  values calculated from mass balance and off gas measurement methods were almost the same. In the case of respiration rate measurement method, the  $K_La$  value the same as other two values in the lower reaches (13,16 cell), but it differed greatly in the upper stream (Figure 1). The method of calculation from respiration rate needs to keep the DO during batch test as it is in the operating condition. But, in the case of  $DO \approx 0\text{mg/L}$  in the aeration tank, it is difficult to reflect the actual condition. When actual DO is low, respiration rate of activated sludge differs a lot. Therefore, it is not appropriate to use the methods of respiration rate.

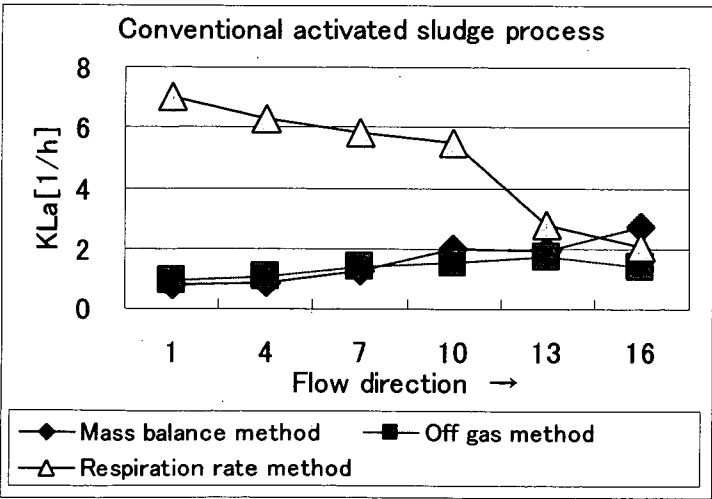


Figure 1  $K_La$  values along flow direction with three measurement methods

(2) We investigated the change of  $K_{La}$  in the aeration tank. Aeration intensity is basically the same through the aeration tank except for the anaerobic cells.  $K_{La}$  increased along flow direction, and the values were high in the cells with nitrification (Figure 2). Therefore, it was suggested that biological reaction had affected  $K_{La}$ .

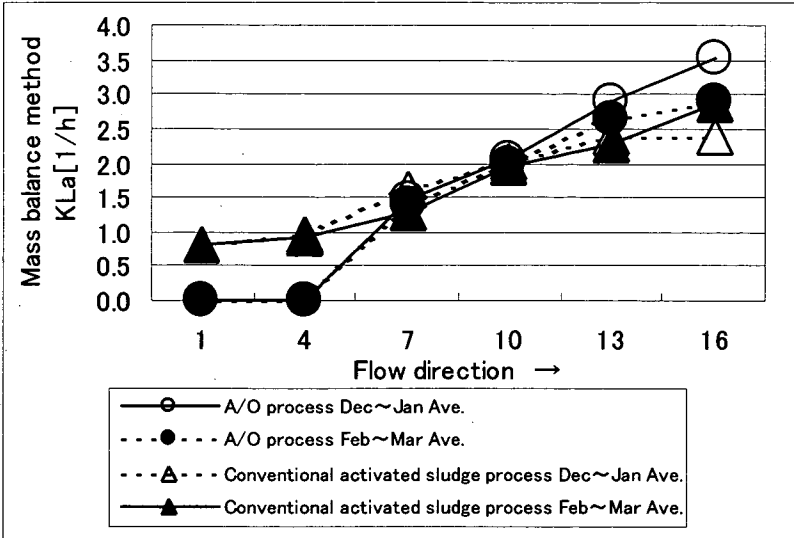


Figure 2 Change of  $K_{La}$  in the aeration process

(3) In the nitrification experiment using immobilization media, the nitrification rates measured with batch method and steady state method were compared. Both methods showed an equivalent nitrification rate and the difference between the measuring methods was not observed. However, for the measurement with batch method, it was required to make substrate and DO equivalent to real situation.

(4)  $K_{La}$  was measured in the nitrification experiment using immobilization media. In the batch method,  $K_{La}$  did not reflect the respiration rate of nitrification. Therefore, as compared with respiration rate calculated from mass balance, the respiration rate of the batch test became low.

(5)  $K_{La}$  values obtained from the mass balance of the immobilization experiment showed that oxygen transfer efficiency was higher with nitrification.



# RISK MANAGEMENT OF PATHOGENS IN SEWERAGE SYSTEM

Y. Suzuki, M. Ogoshi and M. Suwa  
Advanced Wastewater Treatment Division

Project period : 1998-2001

## OBJECTIVES

Generally, it is said that the disinfection tolerance of pathogens such as viruses and protozoan is stronger than that of the coliform assumed to be a present disinfection indicator. Most of pathogens might be removed by the activated sludge processing when they flow in the sewerage treatment plants. But there is a possibility that the infection risk at the downstream region increases by the existence of some pathogens which cannot be inactivated by usual disinfection processes.

In this research, the investigation of the source of pathogens in the watershed, and the improvement of the removal ability of pathogens for the case of an outbreak of pathogens were carried out for the reduction of pathogens in treated wastewater.

## RESULTS

This year examined the disinfection tolerance of the pathogens, the effect of sand filtration to remove them by using the pilot plant test, and the effect of inactivating pathogens in disinfection processes such as chlorination, ultraviolet rays and ozonation.

The examination result is shown as follows.

(1) To inactivate 90, 99, and 99.9%(1,2,3log) of polio virus in tertiary treated wastewater by chlorination, about 2.5, 7.9, and 13.2mg/l of chlorine were needed in 15 minutes of contact time. To inactivate 90, 99% of polio virus in tertiary treated wastewater with NH<sub>4</sub>-N added, 7.0, 16.9mg/l of chlorine were needed. To inactivate 90% of polio virus in secondary treated wastewater, 14.5mg/l of chlorine were needed.

(2) To inactivate 90, 99, and 99.9%(1,2,3log) of polio virus in tertiary treated wastewater by ozonation, about 7.6, 12.7, and 17.8mg/l of ozone were needed in 15 minutes of contact time. To inactivate 90% of polio virus in secondary treated wastewater, 15.7mg/l of ozone were needed.

(3) To inactivate 90, 99, and 99.9%(1,2,3log) of polio virus in tertiary treated wastewater by ultraviolet rays, about 570, 1170, and 1780mWs/cm<sup>2</sup> of ultraviolet rays were needed. To inactivate 90 and 99% polio virus in secondary treated wastewater, 970 and 1960mWs/cm<sup>2</sup> of ultraviolet rays were needed.

(4) Total coliform, fecal coliform and *Streptococcus faecalis* were less tolerate to be inactivated by chlorine,

ozone and ultraviolet rays than polio virus.

(5)It was thought that there was no synergy effect of inactivating virus by the combination of disinfection. However, combination of disinfection could be effective for reduction of disinfection byproducts, because less density of chemicals is needed to disinfect by combination of disinfection processes than by a single disinfection process at the same ratio.

(6)It was thought that the decrease rate of the virus did not rise greatly even if the contact time with the residual chlorine was long in consideration of the water supply process to the recycling facilities.

(7)The decrease ratio of virus through sand filtration was 28-56%. When the filtration speed was twice larger (100-200m/d), the decrease ratio was a half.

# REMOVAL OF ENDOCRINE DISRUPTERS BY WASTEWATER TREATMENT

Y. Suzuki, H. Yamagata and A. Kitanaka  
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Project period: FY1999-2001

## OBJECTIVES

Recently, concern has grown about the effects of endocrine disrupters (EDs) on human health and ecosystems. In sewage treatment plants, it is necessary to reduce effectively the EDs in the influent. For this purpose, we carried out an experimental study on: (1) the effective removal of EDs by controlling solid retention time (SRT) and hydraulic retention time (HRT) in standard activated sludge process pilot plants, and (2) the high-rate removal of EDs by tertiary treatment such as ozonization, coagulation-sedimentation, activated carbon treatment and chlorination.

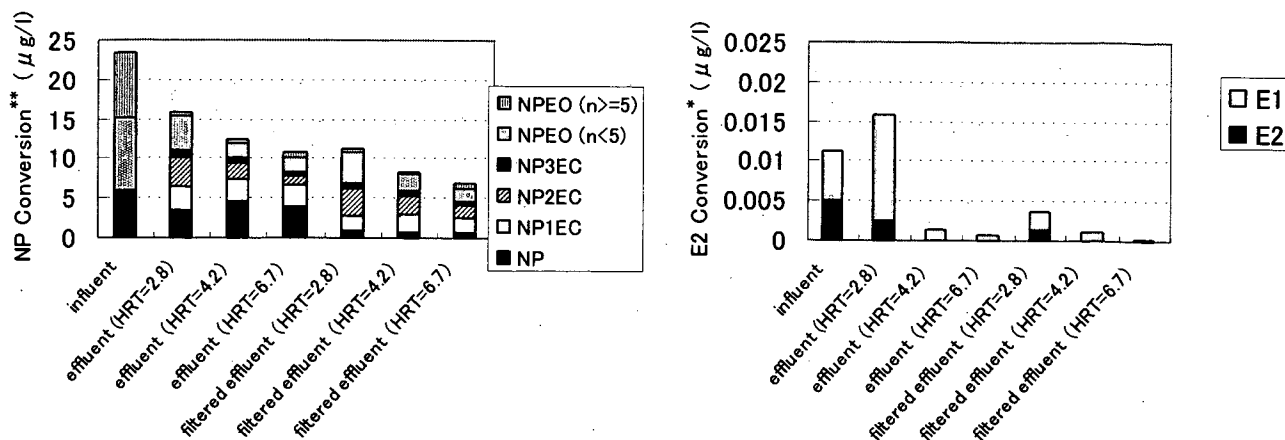
## RESULTS

### **(1) Effective removal of EDs by controlling SRT and HRT in standard activated sludge process pilot plants**

1) The optimum SRT for more effective removal of EDs was researched by comparing the concentration of EDs in effluent in three standard activated sludge process pilot plants with different SRT in the range of 6-13 days. Except SRT, the three plants had the same conditions such as HRT (4.1 hours) and volume (100 L). Measured EDs were nonylphenol (NP), nonylphenoethoxylate (NPEO), and  $17\beta$ -estradiol (E2). The results showed that NP, NPEO and E2 were effectively removed for varying SRT when the dissolved oxygen in aeration tanks was 5-10 mg/l.

2) The optimum HRT for more effective removal of EDs was researched by comparing the concentration of EDs in effluent in the three plants with different HRT in the range of 4.1-6.7 hours. Except HRT, the three plants had the same conditions such as SRT (10 days) and volume. Measured EDs were NP, NPEO, nonylphenoxyethoxylate (NPEC),  $17\beta$ -estradiol (E2) and estrone (E1). The results showed that NPEO ( $n \geq 5$ ) was effectively removed for varying HRT, but that NPEO ( $n \leq 4$ ) and NP1~3EC which are derivatives of NPEO ( $n \geq 5$ ) were not effectively removed for smaller HRT (2.7 hrs). E1 had as high an estrogenic activity as E2 in influent, and higher estrogenic activity than E2 in effluent (see the Figure).

3) When was HRT changed in the range of 2.6 to 6.7 hours, the conversion concentrations of NP, NPEO and NP in activated sludge in aeration tanks were 10 times larger than those in influent, but conversion concentrations of E2, E1 and E2 were smaller. When HRT became smaller, the conversion concentrations of NP, NPEO, NP, E2, E1 and E2 in activated sludge became larger than those in influent. In future, we will clarify the fate of these EDs in wastewater treatment processes by improving the recovery of EDs in sludge.



\*NP conversion: 1 mol of material is assumed to be converted finally to 1 mol of NP.  
 NP conversion concentration of X = (molecular weight of X)/(molecular weight of NP) × (weight of NP)  
 \*\*E2 conversion: estrogenic activity  
 E2 conversion of E1 = (weight of E1) × 0.3

**Figure NP conversion and E2 conversion concentration**  
 Comparison of influent, effluent and filtered effluent

4) In effluent, E2 hardly stuck to SS, whereas NP mainly stuck to SS. Therefore, it was suggested that the removal of SS through sand filtration and the coagulation sedimentation processes is effective for the removal of NP, but is not effective for the removal of E2.

## (2) High-rate removal of EDs by tertiary treatment such as ozonization, coagulation-filtration, activated carbon treatment and chlorination

1) By coagulation-filtration (10 mg/L of PAC), NP and NPEO were removed from wastewater as effectively as dissolved BOD, but estrogens were not removed so effectively.

2) By ozonization (5 mg/L of dosage), NP, NPEO and estrogens were removed effectively, and estrogenic activity was effectively reduced from wastewater (see the Table). Though TOC was not removed so well, the unsaturated combination of organics was degraded.

3) The activated carbon process effectively removed NP, NPEO and estrogens from wastewater in 90 days.

4) Chlorination caused the removal of NP, NPEO and E2 to be unstable, even if more chlorine was injected.

**Table Removal of EDs and TOC by ozonization of wastewater**

	Influent (biologically treated)	Effluent
NP (μg/L)	0.74	0.27
NPEO n≤4 (μg/L)	ND	ND
NPEO n≥5 (μg/L)	6.99	ND
E2 (ng/L)	2.52	0.11
ES* (ng/L)	24.87	tr (0.06)
TOC (mg/L)	5.5	5.0
UV260 (1cm cell)	0.064	0.011
Estrogenic activity (BEACON) (μgE2/L)	0.669	0.049
Estrogenic act (Sumpter method) (ngE2/L)	0.56	ND

\*ES: E1+E2+E3 (ELISA method)

# RESEARCH ON WASTEWATER REUSE SYSTEM AS AN URBAN INFRASTRUCTURE

Y. Suzuki, M. Ogoshi and H. Yamagata  
Advanced Wastewater Treatment Division

Project period: 2000 - 2003

## Objectives

In 1997, the multiple accidents happened in the water reuse system in Fukuoka city. They were the decrease of reclaimed water pressure at the end of the service pipes and the flowing out of red or muddy water at the flush toilets. The major reason of the pressure trouble was plugging of the service pipe by the rust. Though the reclaimed water system had these troubles, the water system had not any troubles at the same buildings. These two systems employ almost the same distribution facilities. So, the basic factor of the accident was thought to be in the reclaimed water quality.

The object of this study is to make a technical standard of the reclaimed water system, to guarantee the safe and comfortable reclaimed water supply. In FY2000, we studied following 4 themes.

- 1) Factors affecting metal corrosion in the reclaimed water system.
- 2) Control method of the biofilm growth on the inner surface of distribution pipes and storage tanks.
- 3) Data collection about the risks related to water reuse during recent 10 to 25 years.
- 4) Fundamental of the reclaimed water movement injected directly into the permeable sand layer.

## Results of the study in FY2000

On-site survey was conducted to understand the actual situation of service pipe plugging and corrosion. And a laboratory experiment was made to see the relationship between corrosion and reclaimed water quality. Numbers of service pipes, meters, valves, and joints were disassembled and checked in the actually operated reclaimed water reuse system. And we found that the service pipe plugging occurred mainly at bare metal surface, and the corrosion occurred mainly around the contact point of two different metals. The result of reclaimed water quality analysis showed that the dissolved ferrous ions turned into the ferric suspensions on the way of distribution. The total concentration of iron increased in the distribution pipe, which seemed to come out from the uncoated iron piping materials. The laboratory experiment supported these phenomena, and indicated that the oxidation of ferrous ion would be accelerated by residual chlorine and high temperature.

The experiment about the biofilm control was conducted using small plastic tube placed in the incubator which controlled at 25°C. Residual chlorine controlled the growth of adherent heterotrophic bacteria (Fig. 1). And the average adherent heterotrophic bacteria growth rate was well explained by residual chlorine and TOC (Fig. 2). So it is important to control not only residual chlorine but also TOC for the inhibition of reclaimed water quality deterioration in the distribution system.

According to the research of literatures about the risk on water reuse, the general concern was the direct infection of pathogens in the reclaimed water, and there were not any reports about the actual troubles of reuse systems including pipe works.

A model experiment about direct reclaimed water injection into the ground water showed that the recharged water moved differently with the base ground water flow, particularly after the injection had stopped. Also, each component in the recharged water indicated different movement, which depend on the mutual reaction with the particles of the stratum. The typical difference was observed between EC and HPC, and obviously HPC movement was delayed due to holding into narrow gap among the stratum particles.

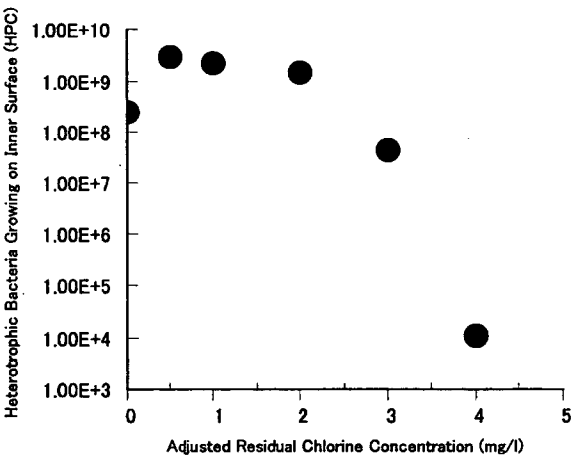


Fig. 1 Control of Adherent Heterotrophic Bacteria by Residual Chlorine

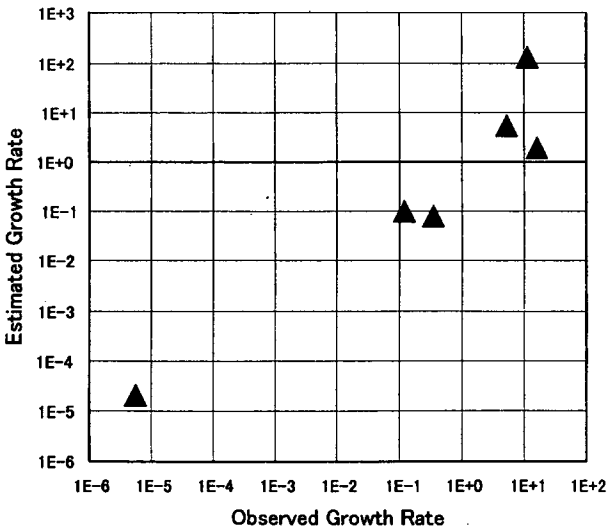


Fig. 2 Observed and Estimated Adherent Heterotrophic Bacteria Growth Rate by Multi-regression Analysis

# CONTROL OF PATHOGENIC MICROORGANISMS IN WASTEWATER SYSTEM

H. Morita, S. Ochi and T. Kitamura

Sludge Management Division

Project period: 1997-2000

## **OBJECTIVES**

An outbreak of cryptosporidiosis was caused through the water supply system in 1996 in Ogoe Town, Saitama, Prefecture and more than 8,000 people were infected. Although the initial cause of this outbreak is not known, in the water cycle consisting of the wastewater treatment plant (WWTP), river and the water supply plant, *Cryptosporidium* is supposed to have multiplied and caused this outbreak, because the WWTP is located upstream of the water supply plant intake.

As the construction of sewerage systems proceeds, WWTPs tend to be located in upstream areas. The ratio of treated water to environmental water has been increasing year by year. In order to decrease the risk of infection, *Cryptosporidium* must be removed or inactivated at the WWTP. Furthermore, the sanitary treatment of sewage sludge is important. This study investigates the control of pathogenic microorganisms in wastewater treatment systems.

## **METHOD**

### **(1) Examination of the applicability of flow cytometry to excystation assay**

The determination of excystation ratio with microscopy has defects in speed and objectivity, so we examined the applicability of flow cytometry to the measurement of excystation ratio.

### **(2) Effect of water temperature on the survival of *Cryptosporidium parvum* oocysts**

The survival of *Cryptosporidium* oocysts is affected by water temperature and storage time. We evaluated the survival of *Cryptosporidium parvum* oocysts under the conditions of controlled temperature from 4 to 60°C. The survival ratio of oocysts was measured using in vitro excystation-flow cytometry assay.

## **RESULTS**

### **(1) Examination of the applicability of flow cytometry to excystation assay**

Excystation ratios measured with flow cytometry and microscopy were almost the same. The time required for the flow cytometry method was shorter than that of the microscopy method.

### **(2) Effect of water temperature on the survival of *Cryptosporidium parvum* oocysts**

The survival ratio of the oocysts rapidly decreased with the increase of water temperature. The required time for inactivation of 90% of oocysts at 60, 55, 50, 45, 40, 30 and 20°C was 3 min, 42 min, 3 hours, 31 hours, 7 days, 31 days and 150 days, respectively. The inactivation ratio of the oocysts at 10 and 4°C was less than 10% after 150 days.

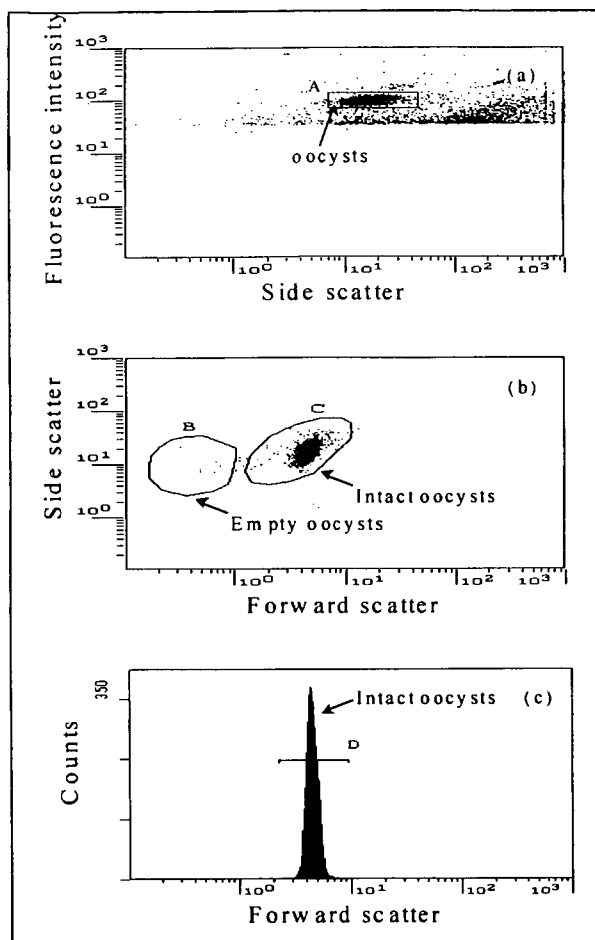


Fig.1 Flow cytometric analysis of fresh *C.parvum* oocysts before excystation assay . (a) The relationship between side scatter and fluorescence intensity. (b) The relationship between forward scatter and side scatter. (c) The relationship between forward scatter and counts.

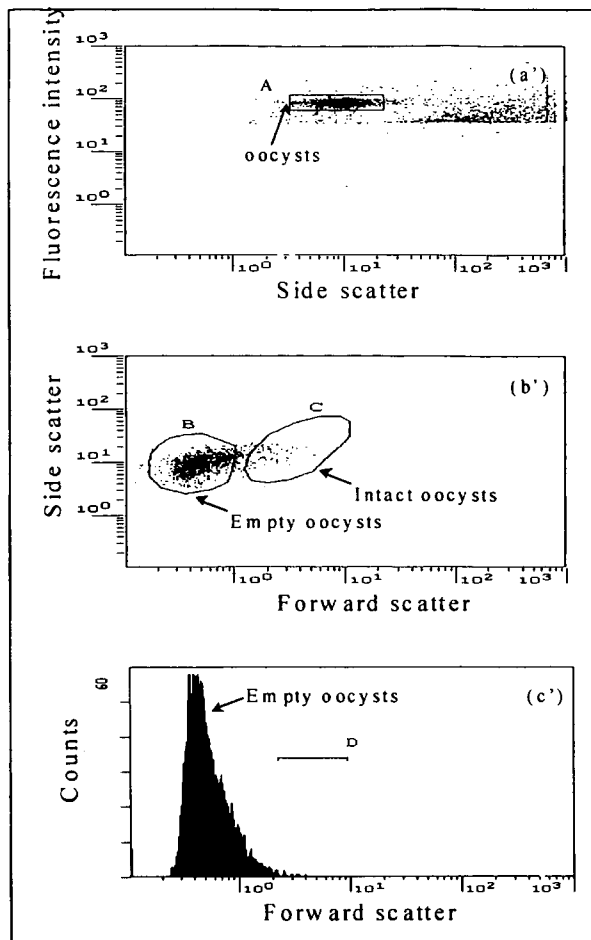


Fig.2 Flow cytometric analysis of fresh *C.parvum* oocysts after excystation assay. (a) The relationship between side scatter and fluorescence intensity. (b) The relationship between forward scatter and Side scatter. (c) The relationship between forward scatter and counts.

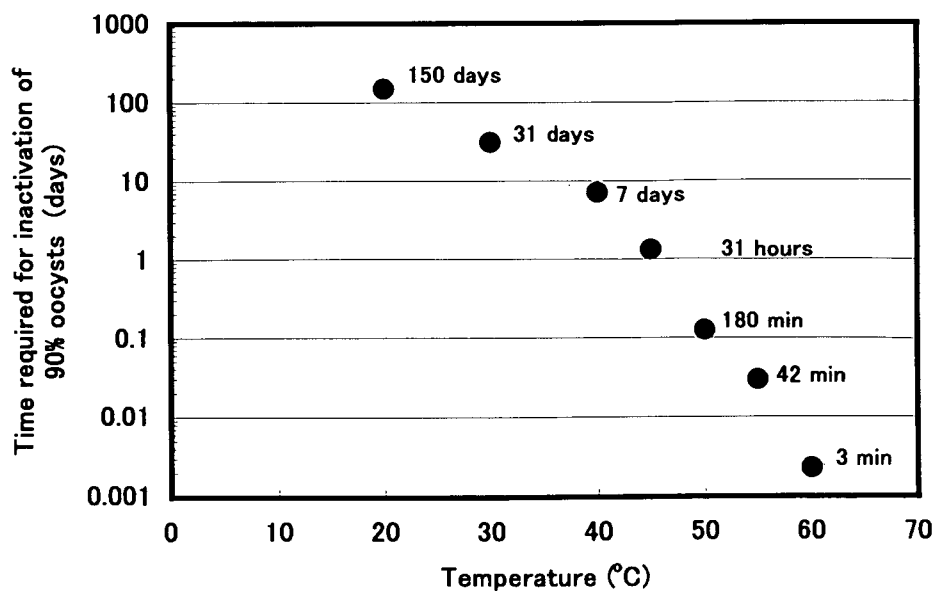


Fig.3 The relationship between temperature and the required time for 90%oocyst inactivation.



# CONTROL OF HEAVY METALS IN SLUDGE TREATMENT PROCESS

Hiroaki Morita, Yukinori Kawashima, and Yuichi Ikeda

Project period: 1998 - 2000

## OBJECTIVES

The recycling of sewage sludge is now required in Japan, since the amount of sludge is increasing in proportion to the sewered population and the sludge is useful as an energy resource and raw material. To promote the recycling, hazardous substances such as heavy metals contained in the sludge should be removed as much as possible. In this study, the behavior and chemical form of heavy metals in sewage sludge were investigated to develop a method to reduce them. In FY 2000, the behavior of arsenic (As) and selenium (Se) in the incineration process were investigated by a survey of incinerators and experiments using a pilot-scale incinerator. In addition, some leachate tests were conducted to estimate the quantity and rate of heavy metals that leach out from sewage ash.

## RESULTS

### SURVEY ON THE BEHAVIOR OF HEAVY METALS IN ACTUAL INCINERATORS

Our former survey on the fluidized bed incinerator revealed that the selenium in sludge, which is gasified in the incinerator, condenses or is adsorbed in the ash collected by bag-filter at about 200°C. In FY 2000, four other incinerators which adopted the bag-filter were added to the survey to clarify the relationship between the concentration of selenium in ash and the temperature of the ash collector. Those ash collectors were operated at about 200°C, and as much selenium was contained in it as in the previous survey. The result is shown in **Figure 1**.

Through these surveys and the bench-scale experiments, we found that the transfer of selenium to the ash depended mainly on the coagulant added in the dewatering process and the temperature in the ash collector. Approximately 50-100% of the selenium in the polymer-added sludge was transferred to the ash at about 200°C; and a trace of selenium was detected in the ash at over 300°C. When slaked lime was added in the dewatering process, selenium was not gasified and remained in the ash. On the other hand, the gasification of arsenic was not observed in our surveys and experiments.

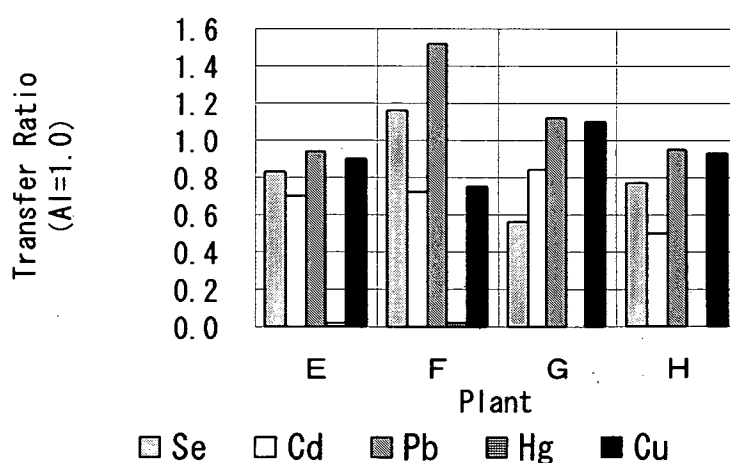


Fig. 1 Aluminum-based ratio of transfer to ash

### EXPERIMENT WITH PILOT-SCALE INCINERATOR

According to some existing reports, including studies on carbonization, arsenic in sludge is gasified in

reducing incineration, under the combustion air ratio of less than 1.0. In FY 2000, we examined the reducing incineration and ash collection by ceramic filter. It was expected that arsenic would be removed in the filter, which was operated at around 700°C at which the arsenic was still gaseous. Also, the behavior of selenium was observed. The air ratio was set at 0.8 according to the result of thermal equilibrium simulation, and the case of air ratio of 1.4 was also examined as a control. The behavior of selenium was almost the same as in the previous survey. The gasification of arsenic was, however, hardly observed. It was assumed that the air ratio might be larger; the measured ratio was larger than 0.9.

## ELUTION OF ARSENIC AND SELENIUM FROM ASH

Arsenic and selenium are easily eluted from the ash, and sometimes hinder the recycling and disposal of sewage sludge. We conducted leachate tests to estimate the elution of arsenic and selenium from the ash. The effects of pH of the solvent, the oxidation-reduction conditions in the incinerator, the temperature in the ash collector and the coagulant of dewatering were investigated in the test. Na(OH) and HCl were used to control pH. As shown in **Figures 2 and 3**, the elution of arsenic was affected by pH; it was larger in the range of low pH. The elution ratio of selenium was approximately 50% in the range of pH 4-12. The elution of arsenic and selenium from the ash to which slaked lime was added and whose pH was around 12, was extremely small. Considering the results of pH controlled tests, it is suggested that the arsenic and selenium formed some insoluble compounds with lime.

According to these studies, the results of the leachate tests on arsenic and selenium can be estimated in advance. In the case of polymer-added sludge, 50-80% of the selenium in the sludge will transfer to the ash, and 50% of the selenium on average will be eluted from the ash, if the ash is collected at about 200°C.

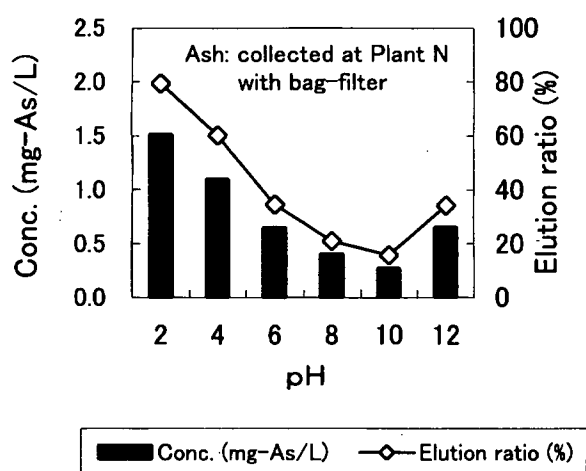


Fig. 2 Concentration and elution ratio of arsenic in pH-controlled leachate test

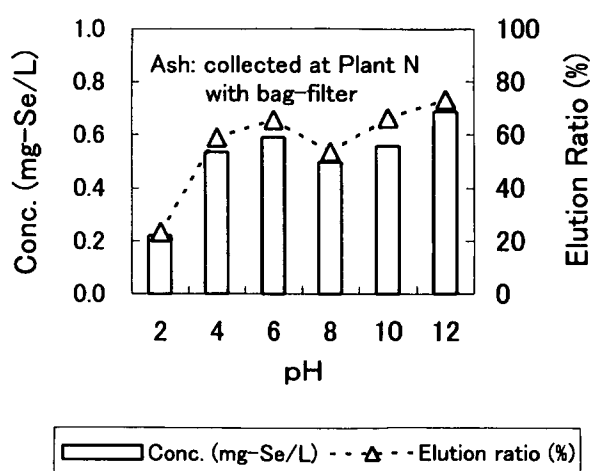


Fig. 3 Concentration and elution ratio of selenium in pH-controlled leachate test

# UNIT ENVIRONMENTAL LOAD FOR LCA OF SEWERAGE SYSTEM

H. Morita and Y. Kawashima

Sludge Management Division

Project period: 1998 - 2000

## **OBJECTIVES**

The sewerage system plays a vital role in improving the living environment, preventing floods, and preserving water quality, but it also has an impact on the environment: the operation of facilities consumes electric power and fuel, and generates greenhouse gases (such as carbon dioxide (CO<sub>2</sub>)). The construction of sewerage systems also consumes energy and resources. Therefore, the environmental impact of sewerage systems must be minimized. The first step to reducing the environmental impact is a quantitative and objective evaluation, for which life cycle assessment (LCA) is useful.

We conducted a methodological study on using LCA to evaluate sewerage systems in the planning stage. The study focused on modeling of the sewerage system and organizing the unit environmental load (UEL), which were used to estimate the environmental load in the life cycle inventory analysis (LCI). Among the environmental loads, we dealt with CO<sub>2</sub> emission and energy consumption.

## **RESULTS**

### **UNIT ENVIRONMENTAL LOAD**

In principle, the UELs for elements such as raw materials and components of the devices were taken from existing databases. The UELs for materials not available in the databases were defined in this study. In addition, the load caused by the construction of unit length (or area, volume, etc.) was also determined in advance to simplify the calculation. The UELs stated above were used as fundamental UELs in this study.

In the early stage of planning, however, the data for the LCI is restricted to these fundamental values, so the load for each unit process (e.g., pumping station, aeration tank, dewatering process, etc.) should be used. Another objective of this study was to assess whether it is possible to define such values, and some trials suggested that it is possible.

### **LCI MODEL**

The inventory of the LCI of this study was designed for process analysis, in which the environmental load is summed up from the elements. The sewerage system needed to be modeled to simplify the analysis. The model of the construction of sewer pipes, tanks (e.g., sedimentation tank, aeration tank, etc.) was composed based on the cost table of the detailed design stage. The table includes materials, electricity, fuel and other factors that should be included in the LCI.

On the other hand, machinery (e.g., pump, blower, dewatering machine, etc.) is mainly assembled at the factory and then installed in the plant by the manufacturer. The list for machinery does not give information about the load from raw materials and from manufacturing, so we surveyed the raw materials and manufacturing process.

The operation stage was modeled based on a survey of treatment plants conducted as case studies.

### CASE STUDY

Case studies were made at some plants to check the framework of the LCI model and to develop the method to calculate the load for unit processes. The overall loads of plants are shown in **Figure 1** as annual load per capacity; materials are included in the construction stage, and the retirement stage includes disposal and recycling. In terms of the CO<sub>2</sub> emission and energy consumption, the ratio of operation was significantly large, and the construction of pipes followed. Most of the load caused by operation in any unit process was from the consumption of electricity, suggesting that the model for electricity consumption should be constructed carefully.

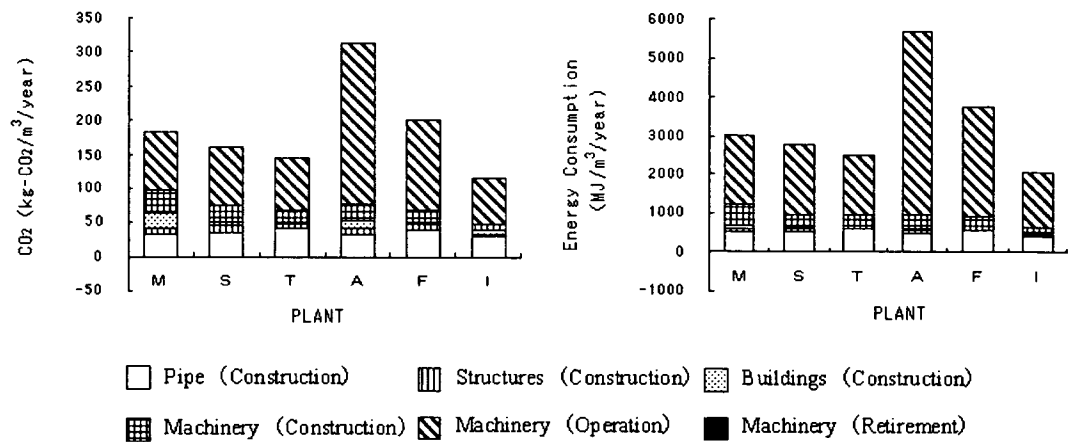


Fig. 1 Life cycle environmental load of sewage treatment plants

For structures such as the aeration tank, the load from concrete work and steel work accounted for about 80% of the overall load regardless of the capacity of the plant. (**Fig. 2**). The load from buildings in the construction stage was estimated as the load per unit floor area, and was categorized into two or three groups by type of building as shown in **Figure 3**. For the construction of structures and buildings, which did not account for such a large proportion of the overall load, the load from them could be roughly estimated using these findings.

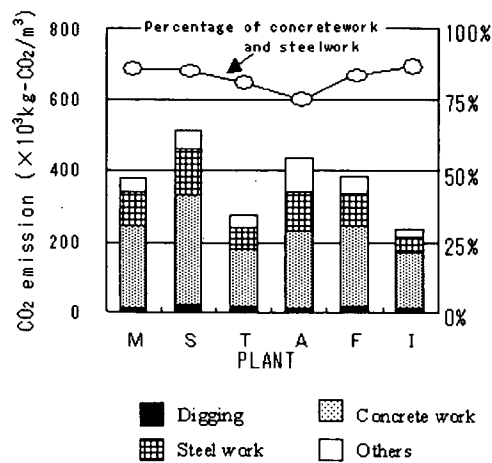


Fig. 2 Load of the structure and percentage of concrete work and steel work

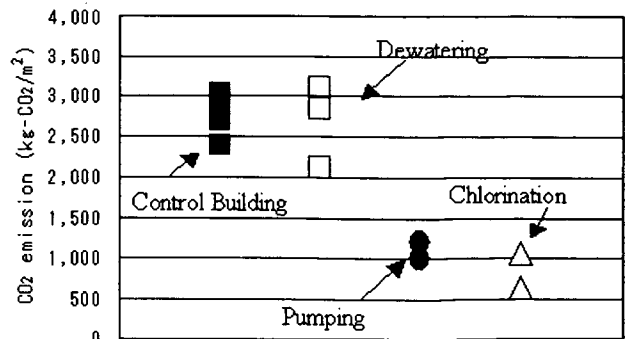


Fig. 3 Load of the buildings categorized by type of building

# IDENTIFICATION OF PATHOGENIC MICROORGANISMS IN WASTEWATER SYSTEM

H. Morita, S. Ochi and T. Kitamura

Sludge Management Division

Project period:1999 - 2002

## **OBJECTIVES**

It is important to quickly confirm the existence of pathogenic microorganisms in sewage, treated sewage and sewage sludge in order to evaluate the pathogenic safety of the water environment. This study aims at developing quick and convenient methods to detect pathogenic microorganisms. Recently, an outbreak of waterborne cryptosporidiosis occurred in Japan. The *Cryptosporidium* oocysts must be removed in the sewage treatment process to prevent further such outbreaks of cryptosporidiosis. *Cryptosporidium* oocysts are measured as follows. First, the oocysts in sewage or sewage sludge are concentrated by a filter or the sucrose floating technique. The concentrated oocysts are then stained by the fluorescent antibody technique. Finally, the oocysts are detected by observation using a microscope. Microscopy is a time-consuming way to detect oocysts, and a convenient and quick detection method is needed.

In FY 2000, a study on a rapid method of simultaneously detecting *Cryptosporidium* oocysts and *Giardia* cysts with a flow cytometry cell sorter (FCCS) was carried out. The pretreatment method for detecting *Cryptosporidium* oocysts by PCR was examined.

The following were examined in this fiscal year.

- (1) Is simultaneous detection of *Cryptosporidium* and *Giardia* possible with flow cytometry?
- (2) What method can be used to extract DNA from *Cryptosporidium*?
- (3) Which primer can detect *Cryptosporidium* with PCR?

## **RESULTS**

The following results were obtained:

- (1) Simultaneous detection of *Cryptosporidium* oocysts and *Giardia* cysts with flow cytometry was possible. The fluorescence intensity and forward scattering strength were different between oocyst and cyst: those of oocyst were lower than those of cyst.
- (2) DNA extraction from *Cryptosporidium* was possible with a commercial extraction kit. The most suitable treatment temperature and time of proteinase K were 55°C and 3 hours, respectively.

The most sensitive primer was SB012 of Wu et al. The SB012 primer enabled the detection of 10<sup>0</sup> order oocysts.

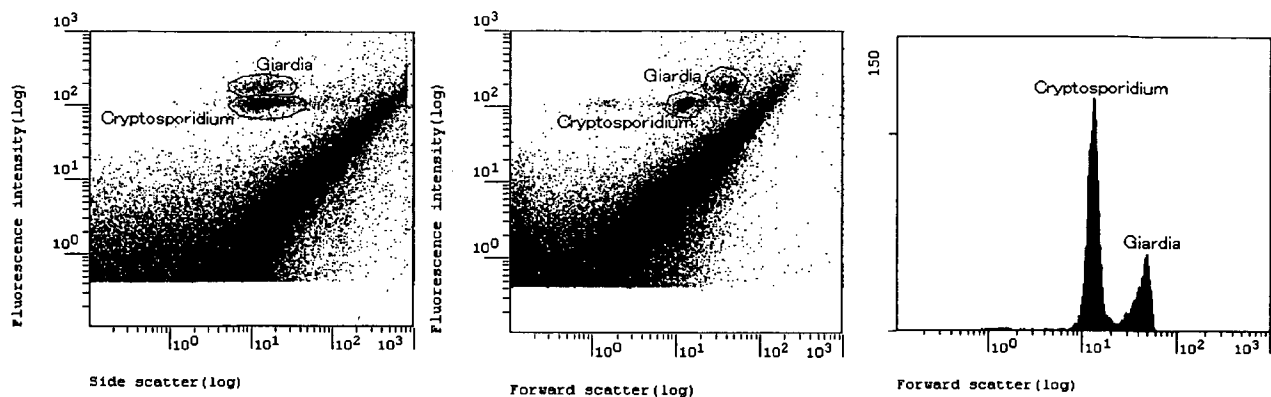
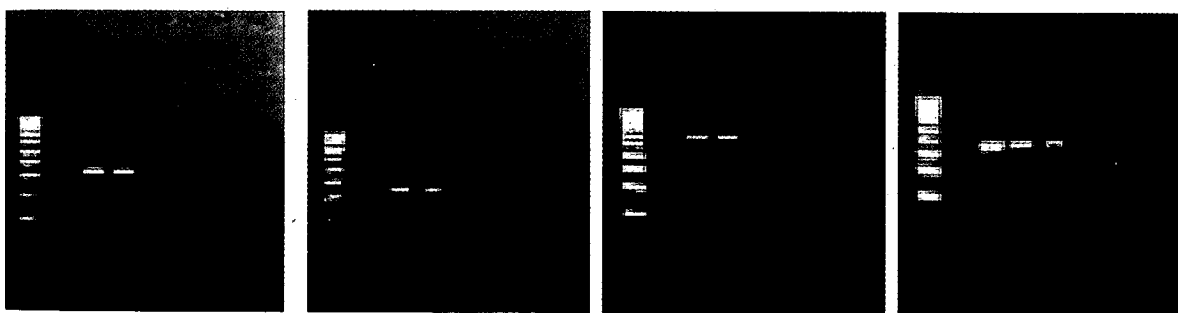


Fig.1 Result of analysis of *Cryptosporidium* oocysts and *Giardia* cysts with flow cytometry.

Table.1 Property of the primer used for the experiment.

Developer	Target gene	Primer name	Primer arrangement	Fragment size	Annealing temperature
Wu	RAPD	SB012F:	5'-CTCCGTTTCGATGATGCAGATG-3'	458	51°C
		SB012R	5'-CGGCCCTGTAGAAATAAGTCA-3'		
Laxer	Chromosomal gene	LAX469F	5'-CCGAGTTTGATCCAAAAAGTTACGAA-3'	451	52°C
		LAX869R	5'-TAGCTCCTCATATGCCTTATTGAGTA-3'		
Awad	18SrRNA gene	AWA772F	5'-AGTGCTTAAAGCAGGCAACTG-3'	556	50°C
		AWA1235R	5'-CGTTAACGGAATTAACCAGAC-3'		
Rochelle	Hsp70 gene	cphsp2423F	5'-AAATGGTGAGCAATCCTCTG-3'	361	55°C
		cphsp2764R	5'-CTTGCTGCTCTTACCAGTAC-3'		

Lane 1 2 3 4 5 6 7 8      1 2 3 4 5 6 7 8      1 2 3 4 5 6 7 8      1 2 3 4 5 6 7 8



(a)SB012,Wu

(b)LAX,Laxer

(c)AWA,Awad

(d)cphsp,Rochelle

Lane1:100-1000marker,      Lane2:negative control      Lane3: $2.4 \times 10^1$  oocysts,      Lane4: $2.4 \times 10^0$  oocysts  
Lane5: $2.4 \times 10^3$  oocysts,      Lane6: $2.4 \times 10^2$  oocysts,      Lane7: $2.4 \times 10^1$  oocysts,      Lane8: $2.4 \times 10^0$  oocysts

Fig.2 The relationship between type of the primer and PCR detection sensitivity.

# BEHAVIOR OF ENDOCRINE DISRUPTORS IN SEWAGE SLUDGE

H. Morita, S. Ochi, and M. Minamiyama

Sludge Management Division

Project period: 1999 - 2001

## OBJECTIVES

In recent years, there have been many reports on environmental problems caused by endocrine disruptors (EDs) discharged as trace chemicals in many countries and regions. Among the many manufactured chemicals necessary for daily lives and activities that are used and discharged, some can disrupt the endocrine system of humans and living things in the wild state. Detailed investigations on this problem have been done around the world.

The Ministry of Construction carried out a national investigation on the pollution of river waters and treated wastewater by EDs in Japan in FY1998; some trace chemicals suspected to be EDs were detected at almost all surveyed points. Research and investigation are needed to study countermeasures to deal with EDs in the sewerage system. This research aims to clarify the behavior of EDs in the unit processes performed at sewage sludge treatment facilities and in the soil that includes sewage sludge compost.

## RESULTS

In FY2000, we carried out experiments to develop an EDs analysis method for sewage sludge samples and to monitor the EDs in soil on which sludge compost had been spread. Nonylphenol (NP), nonylphenol ethoxylate (NPEO), and 17  $\beta$ -estradiol (E2) were chosen as the target EDs because these trace chemicals were detected in higher amounts and at many surveyed points during the national investigation.

## DEVELOPMENT OF ANALYSIS METHODS FOR SEWAGE SLUDGE SAMPLES

It is necessary to develop an appropriate method of analyzing EDs in the sewage sludge samples. Because the extraction process is an important step in the analysis method, extraction experiments were carried out. As a result, extraction conditions that yield a higher recovery rate than the conventional method were achieved by using pressurized fluid extraction.

## MONITORING OF BEHAVIOR OF EDs IN SOIL WITH SLUDGE COMPOST

It is necessary to clarify the behavior of EDs in soil that includes sewage sludge compost. The experiment was started with six lysimeters. Photo 1 and Figure 1 show the outline of the setting of lysimeters. The soil setting conditions of each lysimeter are shown in Table 1. The lysimeters were set for different soil conditions, with soil as the only control case, then EDs-containing sludge compost soil was added, and the soils were plowed.

The transition in the proportion of accumulated quantity of leached NP and E2 to the initial quantity of NP and E2 in the soil is shown in Figure 2. The horizontal axis shows the value of the accumulated leachate quantity divided by the soil bulk volume. For approximately six months of the outdoor investigation, 6.7% or less of NP and 4.7% or less of E2 leached out from each lysimeter by rainfall. According to the transition of NP and E2 retention rate in the soil, the existence of the decrease mechanism in the lysimeters except for the leaching out was clarified on NP.

In order to further clarify the behavior of EDs in the soil, it is necessary to understand the material balance including relating substances such as NPEO and estrone. However, there are many problems with the procedure for analyzing the content of EDs and relating substances in the soil and sludge, and an appropriate analysis method needs to be established.

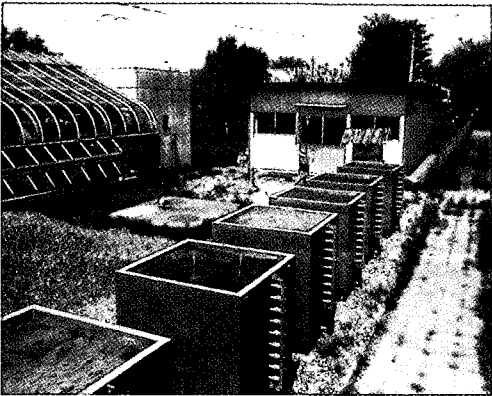


Photo 1 Lysimeter experiment field

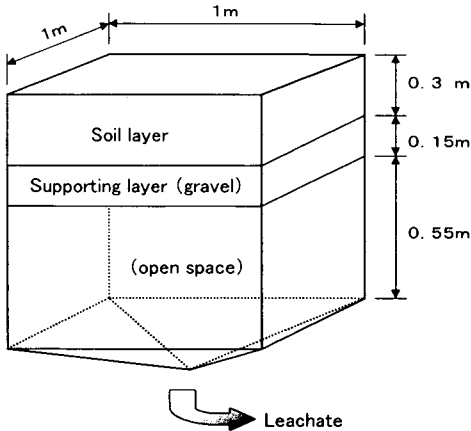


Fig. 1 Summary of inside of a lysimeter

Table 1 Soil conditions of the soil layers in lysimeters

Soil Condition			Key used in Fig. 2
Case 1	Soil	(control)	■
Case 2	Soil + Chemicals (NP, E2)		□
Case 3	Soil + Compost		▲
Case 4	Soil + Compost	(plowed after every soil sampling)	△
Case 5	Soil + Chemicals (NP, E2) + Compost		●
Case 6	Soil + Chemicals (NP, E2) + Compost	(plowed after every soil sampling)	○

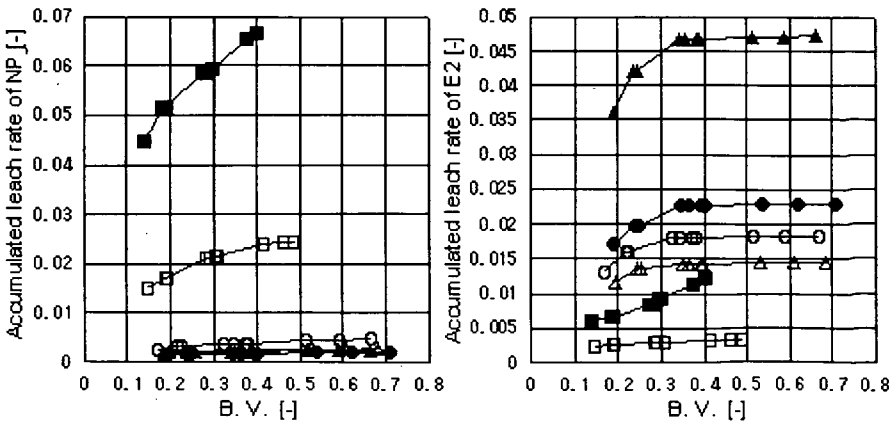


Fig. 2 Accumulated leach rate of NP and E2 from lysimeters



# REDUCING CH<sub>4</sub> AND N<sub>2</sub>O GAS EMISSION FROM WASTEWATER TREATMENT FACILITIES BY IMPROVING APPLICABILITY OF CORE CONTROLLING TECHNOLOGIES

M. Takahashi, Y. Suzuki, R. Hiraide, H. Morita and S. Ochi

Advanced Wastewater Treatment Division and Sludge Management Division

Project period: 2000 - 2002

## OBJECTIVES

Global warming is one of the most serious environmental problems, and so emissions of greenhouse gases (GHG) such as carbon dioxide (CO<sub>2</sub>), methane (CH<sub>4</sub>) and nitrous oxide (N<sub>2</sub>O) must be controlled. In wastewater treatment plants, these GHGs are emitted by biological wastewater treatment, the incineration of sludge and the use of electricity, fuels and chemicals. CH<sub>4</sub> and N<sub>2</sub>O are more problematic because these gases have a higher greenhouse effect than CO<sub>2</sub>. CH<sub>4</sub> is emitted from the biological wastewater treatment process and N<sub>2</sub>O is emitted from the biological nitrogen removal process and sludge incineration. CO<sub>2</sub> emitted from the treatment process is not considered because it is part of the global recycling system with vegetable matter.

It has already been shown that CH<sub>4</sub> emission can be suppressed by introducing anaerobic or anoxic zones in the wastewater treatment process, and that N<sub>2</sub>O emission from the sludge incineration process decreases with increase in furnace temperature. The purpose of this research is to improve the applicability of core technologies controlling the emission of GHG, and also to make the GHG emission inventory more accurate.

## METHOD

We carried out pilot plant experiments and field surveys. The pilot-plant scale experiments were carried out to evaluate the effect of decreasing CH<sub>4</sub> emission by introducing an anaerobic or anoxic zone to the head of the aeration tank, and also to measure N<sub>2</sub>O emission from the conventional process during the winter season. The pilot plant had an aeration tank of 10 m<sup>3</sup> which was equally separated into five blocks, and was operated with the conventional process, AO (Anaerobic-Oxic) process and RN/DeN (Recycled Nitrification/Denitrification) process, each of which had the same HRT (Hydraulic Retention Time) of 5.6 hr and SRT (Solids Retention Time) of 7 days.

Three field surveys were performed to continuously collect data for evaluating the effect of controlling furnace temperature on N<sub>2</sub>O emission from the fluidized bed incinerator when burning polymer-coagulated sludge cake. The capacities of surveyed incinerators were 50, 200 and 250 t sludge cake/day. Concentration of N<sub>2</sub>O in the furnace waste gas was analyzed for 7 days continuously, with an N<sub>2</sub>O monitor.

## RESULTS

The results of pilot plant experiments which measured the CH<sub>4</sub> balance in several treatment processes

are shown in Table 1. There was a consistent effect of introducing the anaerobic or anoxic zone on reducing CH<sub>4</sub> emission and the effect was significant when the process was RN/DeN which had an anoxic zone. The RN/DeN process showed that CH<sub>4</sub> emission could be halved compared with the conventional process. The emission of N<sub>2</sub>O gas from the conventional process in the winter season was smaller because the nitrification activity was low.

The results of field surveys on sludge cake incinerator are shown in Table 2. The concentration of N<sub>2</sub>O gas emitted from each incinerator changed with furnace temperature in the free board, and it seemed that the difficulty in keeping the temperature of the free board constant was caused by changes in the supply rate of sludge cake. N<sub>2</sub>O emission from incinerator C was higher than that of the other two incinerators, the cause of which is not clear.

Table 1 Results of pilot plant experiments on the CH<sub>4</sub> balance in wastewater treatment processes

Wastewater treatment process	CH <sub>4</sub> balance ((emission + outflow) / inflow)	CH <sub>4</sub> production in the aeration tank <sup>a)</sup> (mg-CH <sub>4</sub> /m <sup>3</sup> -treated water)					
		Total	1st. cell	2nd. cell	3rd. cell	4th. cell	5th. cell
Conventional process	1.4	162.6	168.5	-17.6	11.8	-2.1	2.0
AO process	1.2	64.7	133.7	-103.7	30.4	2.9	1.4
RN/DeN process	0.7	-153.9	47.0	-225.5	16.1	6.9	1.6

a) The conditions of 1st. cells were anaerobic for AO process and anoxic for RN/DeN process.  
The 1st. cell of conventional process and the all of other cells were operated with aerobic condition.

Table 2 Results of field surveys on the N<sub>2</sub>O emission from fluidized bed incinerator of sewage sludge cake coagulated with polymer

Surveyed incinerator (capacity)		Supplied sludge cake rate (t/hr)	moisture content (w/w %)	Furnace temperature in free board (°C)	N <sub>2</sub> O emission	
					concentration (v/v ppm(O <sub>2</sub> =6%))	emission ratio (g-N <sub>2</sub> O/t-cake)
A (50t/day)	average	2.29	80.3	849	118.5	359
	standard deviation	0.03	1.18	4.8	21.5	65
B (200t/day)	average	7.61	75.3	829	164.3	603
	standard deviation	0.61	0.34	7	40.9	152
C (250t/day)	average	8.52	78.6	838	327	1007
	standard deviation	0.49	0.81	7.4	73.3	243

# SYSTEMS FOR USING THE SELF-ENERGY OF SLUDGE

H. Morita and S. Ochi  
Sludge Management Division

Project period: 2000 - 2003

## OBJECTIVES

Sewage sludge possesses energy of about 20 kJ/g solids, so sludge treatment processes need a large amount of energy, while wastewater treatment processes need even more energy. This study aims to construct systems for using the self-energy of sludge to convert the sludge treatment process into an energy-producing process in the near future. In this research, the sludge incinerator which is normally located in the energy production process is converted to an generator. In addition, the anaerobic sludge digestion process is important for producing biogas (digestion gas) which is a high-quality source of energy containing about 60 vol% methane gas. We attempted to develop a gas storage process which has a higher capacity than the common process, for the sustainable, stable use of digestion gas.

## METHOD AND RESULTS

The energy potentially available from sewage sludge was quantitatively estimated, and the electric power available was found to be about 0.23 KWh/m<sup>3</sup> inflow of wastewater. This result suggests that inflow wastewater could produce 0.30 KWh/m<sup>3</sup>, which is the average of all wastewater and sludge treatment plants in Japan. A new power generation system using sewage sludge was devised, as outlined in Figure 1, in which the system consists of many generator devices.

In this study, it is important to determine the energy balance when developing systems, and to make an analytical model of energy balance. Therefore, we investigated the specifications of machinery and equipment which are used in the design of sewage treatment facilities, by conducting a questionnaire survey on power, measurements, weight and composition of materials. The results were compiled in the Technical Memorandum of PWRI, No.3827, March 2001.

In a joint research project by Tsuruoka City, the Japan Institute of Wastewater Engineering Technology and the PWRI, the gas storage system with gas adsorption process was developed, which has a larger gas capacity. A pilot plant experiment was carried out using actual digestion gas to investigate the gas capacity and to gather knowledge on the construction and operation of a gas storage system. The pilot plant consists of a pretreatment unit, compressor unit, dehumidification unit and adsorption unit, and the flow is as shown in Figure 2. The gas adsorption unit has a volume of 0.5 m<sup>3</sup>, and was packed with activated carbon as adsorbent. The temperature of the adsorbent rises as gas is adsorbed and drops as gas is desorbed. These phenomena influence the efficiency of gas storage and use, so the change in quantity with temperature is important. The results for the change in quantity with temperature in the pilot plant experiments are shown in Figure 3.

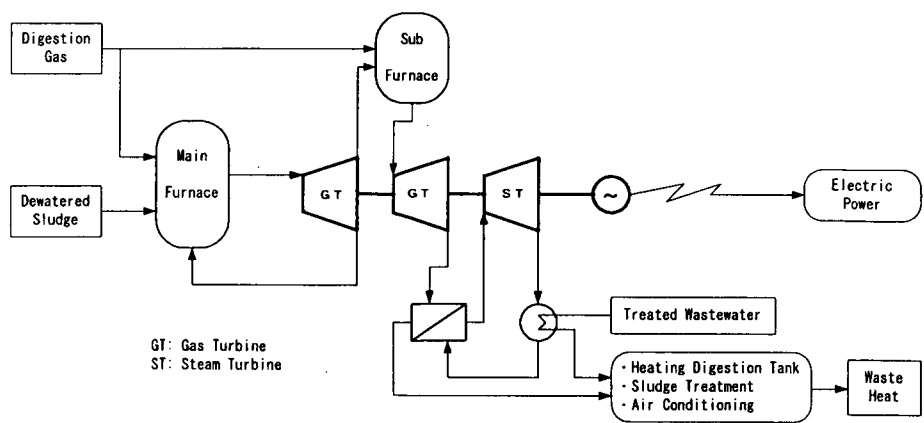


Fig. 1 Illustration of power generation system for using the energy of sewage sludge

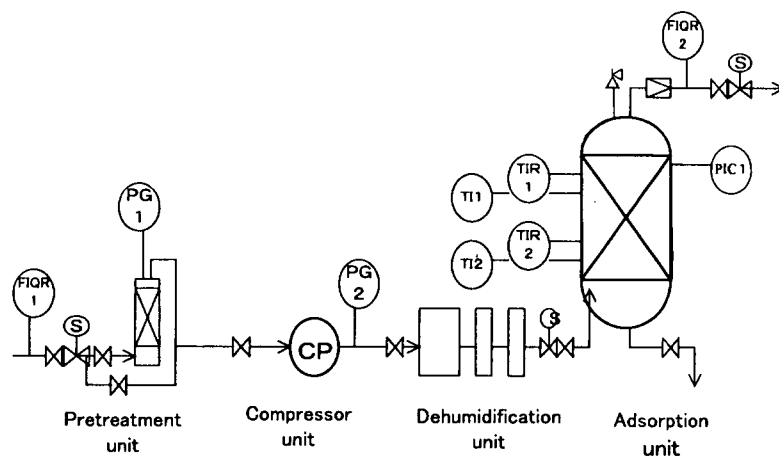


Fig. 2 Flow of pilot plant for gas storage system

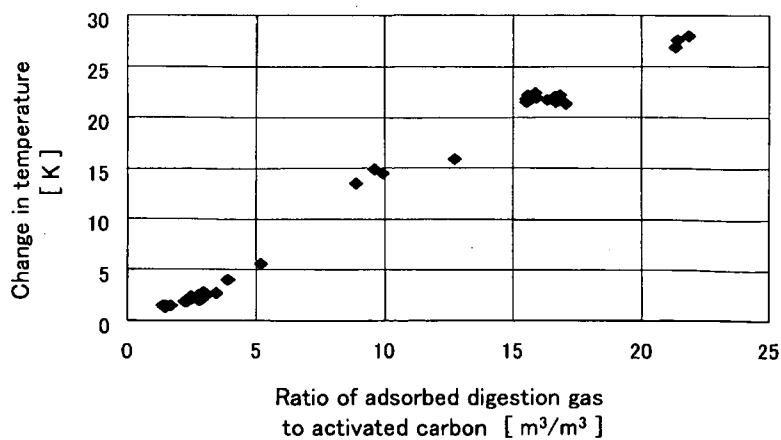


Fig. 3 Relationship between ratio of adsorbed digestion gas to activated carbon, and change in quantity with temperature

# SEWAGE SLUDGE TREATMENT WITH OTHER SLUDGE WHICH DIFFERS FROM SEWAGE SLUDGE

H. Morita and H. Saino  
Sludge Management Division

Project period: 2000 - 2003

## OBJECTIVES

Recently, sewage sludge and sludge generated from other treatment facilities are required to be treated together at sewage treatment plants to reduce the cost of public works, so some municipalities treat such sludge together with sewage sludge. However, when such sludge is treated at a sewage treatment plant, the sewage sludge treatment process will likely be affected because the sludge differs from sewage sludge. The quantitative effects are not clear. Therefore, this study set out to grasp the technological problems of co-treatment of sludge and to suggest the solutions.

## METHOD

In FY2000 we investigated sewage sludge composting with other sludge. We used sludge generated from a rural sewerage project and night soil treatment plant as sludge generated from other facilities, and conducted two main experiments. One was a separate fermentation experiment of each sludge on a laboratory scale, and the other was a mixed fermentation experiment of sewage sludge and other sludge on a full scale. Details are as follows.

- (1) The separate fermentation experiment was carried out to identify the characteristics of each sludge. Sewage sludge, three kinds of sludge generated from the rural sewerage project (contact aeration process [J-3], sequencing batch reactor process [J-11], and oxidation ditch process [OD]), and three kinds of sludge generated from the night soil treatment plant (standard loading denitrification process [SD], high loading denitrification process [HD], and anaerobic digestion activated sludge process [AD]) were put into each cylindrical composter of 4.7 liters in volume, and compulsory aeration and periodic mixing were performed. Grass was added as a bulking agent.
- (2) The mixed fermentation experiment was carried out to grasp the problems of actual composting of a mixture of different sludge and to solve the problems. Sewage sludge, sludge generated from the rural sewerage project (here, OD was used), and sludge generated from the night soil treatment plant (here, AA was used) were used. The mixing ratio of sewage sludge and other sludge was 1:0, 1:1, and 0:1 (dry solids basis). The wind-low type by a self-driving compost turning machine was applied. Grass was added as a bulking agent.

# RESULTS

Fundamental data concerning the composting of sewage sludge with other sludge were obtained through this study. The summary of the study in FY 2000 is as follows.

- (1) In the separate fermentation experiment, all cases except “AD” fermented actively (Fig. 1) and it is supposed that they ferment sufficiently even if sewage sludge is mixed with them.
- (2) According to the separate fermentation, fermentation showed the tendencies that a) the richer the sludge containing organics, the longer the fermentation time, and b) the higher the BOD of sludge, the earlier the fermentation ended.
- (3) In the mixed fermentation experiment, the fermentation temperature of mixed sludge was almost midway between the temperatures of the sewage sludge and other sludge single fermentation. Therefore, it is suggested that mixed sludge composting could be achieved by separately composting.
- (4) Harmful substances such as heavy metals appeared to be concentrated in sludge by long-term anaerobic digestion, so care is required regarding harmful substances when using “AD” sludge for compost. Although generally speaking sewage sludge contains many heavy metals, not only “AD” sludge but also other sludge generated from the night soil treatment plant or sludge generated from the rural sewerage project sometimes contains more heavy metals than sewage sludge, so care is required (Table 2).

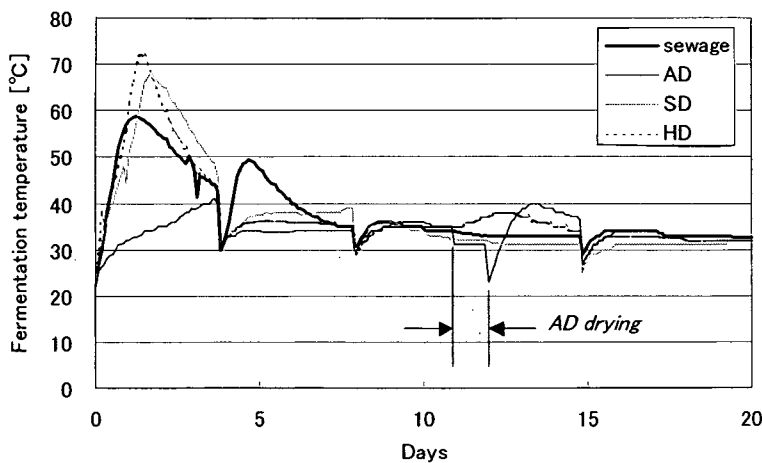


Fig. 1 Fermentation temperature of sewage sludge and night soil sludge

Table 1 Heavy metals contents of sludge

	Standard (mg/kg·DS)	Rural sewerage project				Night soil treatment plant			
		sewage	J-3	J-11	OD	sewage	AD	SD	HD
As	50	5.0	5.0	3.7	4.5	3.6	3.3	2.2	3.8
Cd	5	2.1	4.2	3.0	4.0	2.0	7.1	4.7	5.1
Hg	2	0.57	0.40	0.29	0.26	0.23	1.05	0.57	0.66
Ni	300	47	88	71	101	45	252	48	72
Cr	500	43	48	52	64	37	294	25	57
Pb	100	48	58	59	59	38	206	50	52
Cu	300	641	434	1,082	1,349	537	975	381	439
Zn	900	7,735	1,622	1,362	1,602	5,497	2,244	1,690	1,830

# DEVELOPMENT OF TECHNOLOGY FOR DIOXIN CONTAMINATED SOIL COUNTERMEASURES

H. Morita, S. Ochi, and M. Minamiyama

Sludge Management Division

H. Tanaka, K. Komori, Y. Okayasu

Water Quality Division

Project period: 2000 - 2002

## OBJECTIVES

In recent years, the pollution by dioxins including tetra- through octa-chlorodibenzo-*p*-dioxins (PCDDs), tetra- through octa-chlorodibenzofurans (PCDFs), and coplanar polychlorobiphenyls (Co-PCBs) has become a major nationwide problem, as dioxins have high toxicity even in trace concentrations.

The content of dioxins in the bottom sediment may greatly change with collection position. In order to carry out appropriate construction management and countermeasure evaluation, it is necessary to estimate the distribution of dioxins quickly and simply. The official method of measuring dioxins in bottom sediment and in water samples, from sampling to getting analysis results, is time consuming, so it is necessary to develop a quicker analysis method.

## RESULTS

### (1) Analysis Method for Dioxins in Sediment Samples

We collected a bottom sediment sample of a river. Using this sample, we examined the effect of differences of pre-treatment methods on the result of analysis. Regarding sampling and analysis of the sample, the official method was used. The target substances were PCDDs, PCDFs, and Co-PCBs.

Water content is high for the bottom sediment samples, so in many cases bottom sediment samples also contain large quantities of organic substances, so a long time is required in the official method of analyzing dioxins of bottom material for the drying process, extraction process, and clean-up process. Especially, the analysis could be speeded up by quicker drying and extraction. Therefore, various drying techniques and extraction techniques should be compared and examined. In FY2000, Sludge Management Division compared Soxhlet extraction method (Method A) with n-hexane extraction from the wet sediment method (Method B). The main procedures of both methods are shown in Table 1. Method A is generally used. The time required for Method B is shorter than that of Method A.

Table 1. Main procedures and required time of methods

	Drying process	Extraction process
Method A	Air-drying. (a few days)	Soxhlet extraction using toluene (more than 16 hours)
Method B	(not necessary)	Potassium hydroxide ethanol solution is put in and left for one evening at room temperature, followed by shaking extraction using n-hexane (10 minutes, 3 times).

The results of Methods A and B are compared in Figure 1. In result of analysis of individual isomers, many isomer contents derived by Method B were more than 80% of the values derived by Method A. The coefficient of variation of the result derived by Method B was bigger than that by Method A. The recovery rate of internal standard substances of Method B was lower than that of Method A; especially, the recovery rate of 7 chlorides and 8 chlorides was 50% or less on PCDDs and PCDFs.

Method B can be expected as quick method of extracting dioxins from bottom sediment samples, since the air-drying process is unnecessary. Comparing with Method A in many isomers, Method B gave a measured value of over 80% of that by Method A. However, there were some isomers whose recovery rate was 50% or less. Thus, when Method B is used for actual polluted site investigations, its applicability must be examined beforehand.

To speed up the analysis of dioxins for bottom sediment samples, it is necessary to speed up the extraction process including the drying process. It is also necessary to develop an analysis procedure which produces stable results. We will carry out comparisons of other types of drying technique and extraction technique and consider speeding up the whole analysis of bottom sediment samples.

### (2) Analysis Method for Dioxins in Water Samples

To develop a method that can be used for quick measurement of dioxins in water samples, Water Quality Division has, through joint research with private sectors, developed an analysis method using antigen antibodies. During FY2000, a new antigen was chemically synthesized, then injected into 19 rabbits as an immunogen to prepare new antibodies and established a measurement procedure by the ELISA method.

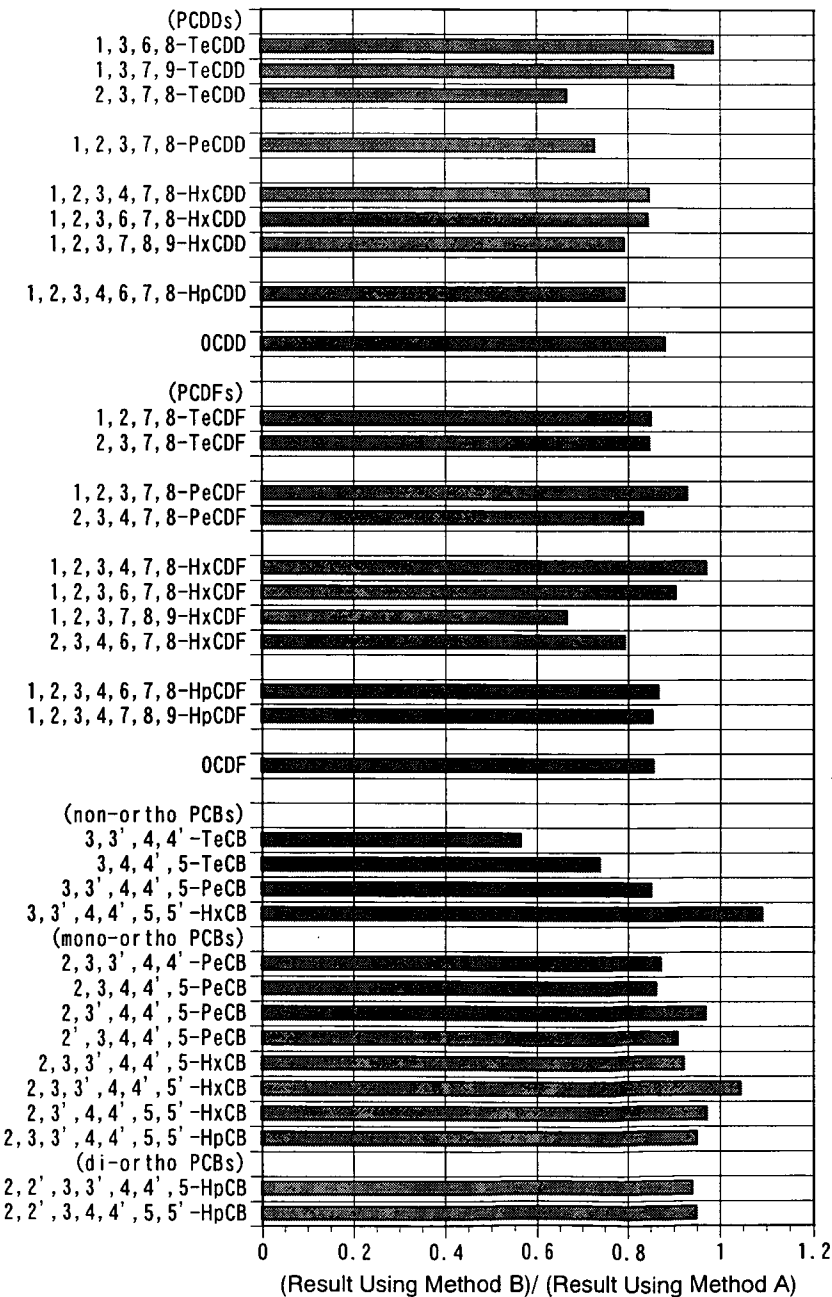


Fig. 1 Comparison of result between Method A and Method B



# EFFECTS OF SEWERAGE IN WATERSHED

H.Tanaka, K.Komori, M.Shirasaki, H.Ishii and K.Taketoshi

Water Quality Division

Project period: FY 1996-2000

## OBJECTIVES

In recent years, sewerage systems are being constructed steadily in Japan. Therefore, the water discharged through sewerage systems is increasing and has occupied the major part in water circulation in the watersheds, partially in urban areas. The greater water flow through the artificial water circulation including water supply and sewerage systems that is affected by human activities becomes, the more water quantity and quality of the rivers are affected by such artificial water circulation.

Considering this situation, we must comprehensively manage quantity and quality of water from the view of watershed in order to preserve the sound water environment. Although we need to grasp how and to what extent the artificial water circulation affects on the whole water circulation in the watersheds, we have not yet reached the answers to clarify the questions up to this present. Therefore, as a first step to materialize the comprehensive water management, we have investigated the effects of sewerage systems on the circulation of water and various pollutant loads in watersheds at present. Further, we will try to predict them under the development of sewerage systems in the future.

## METHODS

As a model of watershed that is significantly affected by artificial water circulation, we investigated the characteristics of discharge loads and flow rates in the Ohtsu River since 1996. In this year we consecutively investigated water pollution runoff every hour for a month. The Ohtsu River is one of the rivers flowing into the Lake Teganuma that is located in northwest of Chiba Prefecture

and the most polluted lake in Japan due to the advance of urbanization in its watershed. We set three observation points at the outlet of three sub watersheds that can almost cover whole the Ohtsu River basin. The backgrounds of this watershed are shown in Fig. 1. One was set up at the main stream, and the other points were set up at the tributaries

We observed flow rate and water quality at each observation point. Flow rates ( $Q_s$ ) were calculated from water levels ( $H_s$ ) observed by the flow gauge every five minutes and  $H$ - $Q$  curves obtained in this stream. Regarding the fluctuation of water quality, we sampled the river water every hour and then analyzed about BOD,  $COD_{Mn}$ ,  $NH_4-N$ ,  $NO_2-N$ ,  $NO_3-N$ ,  $T-N$ ,  $PO_4-P$  and  $T-P$  concentrations in laboratory. Discharge loads were calculated from the flow rate and the water quality.

## RESULTS

### Consecutive Observation of Water Quality and Discharge Loads

We consecutively observed the discharge loads and flow rates for a month. The results of one week during this observation are shown in Fig.2. This week had no rainfall. So we could catch variation of the water quality in fine days. The water quality in the mainstream and Mashio Tributary fluctuated daily with human activity in these watersheds. In

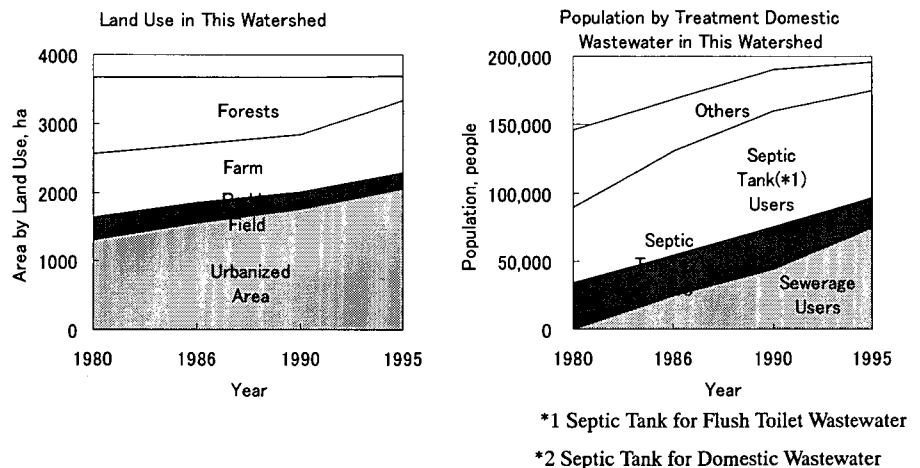


Fig.1 Background of the Observation Watershed

Natogaya Tributary fluctuation of the water quality was small, because sewerage system has already been developed in most of its watershed. But in all sites we observed unexpected variations of the water quality. One of the factors causing unexpected variation of the water quality seems increase in unexpected loading from industrial discharge. In these circumstances, we found that consecutive observation in fine days was necessary to the exact estimation of discharge pollutant loads.

### Influent Factors for Discharge Pollutant Loads in Rainy Days

We compared the difference of the rainy runoff and pollutant loads between the observation in the 1980s and the 1990s. We found the similar discharge trend of pollutant loads of SS and nutrients in both observations. However we could not observe the similar discharge trend of pollutant loads of BOD and COD in both observations.

So we analyzed relationships between rainy pollutant loads and other rainfall factors for the purpose of investigating the factor in variation of the rainy runoff and pollutant loads. Correlation coefficients between pollutant loads in rainy days and other rainfall factors of observation in the 1980s and the 1990s are shown in Table 1. As compared with the 1980s, in the 1990s correlation between rainy pollutant loads except for SS and TN and factors in rainfall scales became low. And in the 1990s correlation between rainy pollutant loads and factors in antecedent weather, in relation to the accumulations or remains of pollutant loads, were the opposite of the 1980s.

But the data of observation in the 1990s were limited. We did not know the trend of discharge pollutant loads for certain. So we must accumulate still more observation data.

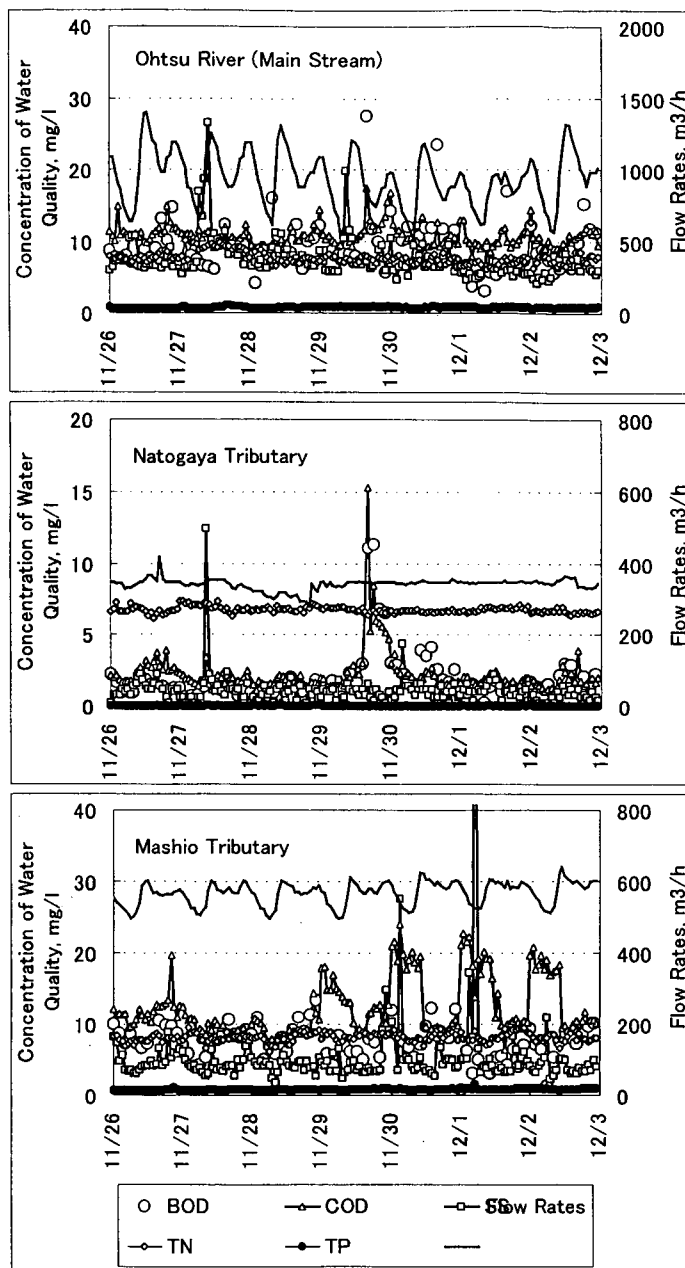


Fig.2 Results of the Close Observation

Table 1 Correlation Coefficients between Pollutant Loads in Rainy Days and Other Rainfall Factors of Observation in the 1980s and the 1990s

Pollutant Loads(kg/ha)	Observations in 1990s					Observations in 1980s				
	BOD	COD	TN	TP	SS	BOD	COD	TN	TP	SS
Specific Flow Rates(mm)	0.47	0.40	0.91	0.65	0.93	0.73	0.62	0.85	0.79	0.64
Rainfall(mm)	0.57	0.52	0.88	0.67	0.97	0.80	0.70	0.80	0.83	0.63
Maximum Rainfall Rate(mm/h)	0.65	0.69	0.53	0.50	0.83	0.71	0.85	0.74	0.80	0.71
Average Rainfall Rate(mm/h)	0.71	0.79	0.43	0.42	0.75	0.63	0.78	0.78	0.74	0.80
Rainfall Duration(h)	0.55	0.46	0.94	0.73	0.95	0.13	0.01	-0.08	0.10	-0.15
Antecedent Fine Days(day)	-0.46	-0.40	-0.46	-0.44	-0.48	0.42	0.65	0.66	0.70	0.42
Antecedent Precipitation Factor(mm)	0.26	0.24	0.80	0.47	0.77	-0.53	-0.61	-0.59	-0.72	-0.42

# EVALUATION METHODS OF CONTAMINATION DISCHARGED FROM URBAN WASTEWATER AFFECTING SOURCE WATER OF DRINKING WATER SUPPLY

H. Tanaka, K. Komori, A. Takahashi, Y. Yakou, M. Saito, T. Higashitani

Water Quality Division

Project period : FY 1998-2000

## **OBJECTIVES**

Recent fact-finding surveys on endocrine disrupters (EDs) have shown that many substances suspected as EDs and estrogen-like activities exist in water environment and wastewater systems in Japan. Our recombinant yeast studies revealed that several substances such as 17 $\beta$  estradiol (E2), estrone (E1), nonylphenol (NP) and bisphenol (BPA) are thought to be important in river water and discharge from sewage treatment plants (STPs) from the viewpoint of estrogen-like activity. But, still, it is less clear whether measured estrogen-like activity is attributed to natural estrogens or other anthropologic chemicals like NP and BPA. Thus, we attempted to fractionate the water samples to characterize and estimate major comes of the estrogenicity using fractionation technique and recombinant yeast assay.

## **METHODS**

We collected five water samples, consisted of discharge from two STPs, Plants A and B, waters of the Tama River and its tributary

Estrone (E1), 17 $\beta$  -estradiol (E2), Nonylphenol (NP) and Bisphenol A (BPA), were selected as typical environmental estrogens in the water samples because those have been reported to be important in the United Kingdom and Japan. Each chemical was injected into HPLC with a silica solid phase column and was succesively eluted from the column with the following solvents: dichloromethane:hexane/3:7 (Fraction No.1), dichloromethane (Fraction No.2), and methanol (Fraction No.3).

The DNA recombinant yeast used in this study was provided by Brunel University in the United Kingdom. The measurement was conducted with the partially modified basic protocol. For calculations of the estrogenic potential in water samples, the dose-response curve of 17 $\beta$  -estradiol was used. Each fraction was concentrated in 10000-fold and was assayed with the yeast.

## **RESULTS**

As shown in Table 1, E2, E1, NP and BPA were isolated and fractionated into Fractions No.1 to No.3, respectively.

Figure 2 and Table 2 present estrogen-like activity of each fraction of the water samples. Fraction No.1 in all the samples exhibited little or no estrogenicity except for that at downstream point of the STP whose estrogenicity was 6.3 ng-E2 equivalent/L and accounted for 32% of the total estrogenic activity.

Fraction No.2 dominated the estrogenicity in the discharge from both the STPs. The estrogenicity of Fraction No.2 occupied 82 % to 88 %, but that of Fraction No.3 did 7 % to 12 % in the discharge from both the STPs. This shows that there exist stronger estrogen-like substances in Fraction No.2 coincident with E1 than Fraction 1 coincident with E2 in the discharge.

Fraction No.3 dominated the estrogenicity of the water sample at the downstream point but did not in the other water samples. Because the estrogenicity of the river water at the upstream point from the STPs was not detected, the estrogenicity of the water at the downstream point was attributed to the discharge and/or the tributaries. Although the estrogenicities of the three fractions of both the discharges from Plants A and B

contributed similarly, they were different from those of the other waster samples. This suggests that major estrogen-like substances in the discharge from the STPs changed in the river.

Table 1 Recovery of fractionated standard chemicals

Standard chemicals	Dissolved in distilled water		Dissolved in river water		Fraction No.
	Added		Added		
	Recovery (%)	concentration n (i g/L)	Recovery (%)	concentration n (i g/L)	
17â-estradiol(E2)	68.3	0.02	99.7	0.02	3
Estron(E1)	72.0	0.02	99.2	0.02	2
Nonylphenol(NP)	94.7	0.5	76.2	2.5	1
Bisphenol A(BPA)	82.8	0.05	85.8	0.25	3

Table 2 Estrogen-like activity and elemental composition for three fraction in water samples

Standard chemicals	(A)			(B)	(C)	(C) / (B)	Elemental composition (%)		
	Estrogen-like activity * (E2 equivqlent ng/L)			Sum of elements	Unfractionated estrogen-like activity				
	Fr.1	Fr.2	Fr.3	(E2 equivqlent ng/L)		(%)	Fr.1	Fr.2	Fr.3
Upstream	nd**	nd**	nd**	—	0.5	—	—	—	—
Downstream	6.3	2.0	11.2	19.5	18.0	108	32	10	58
Tributary	0.1	2.5	1.4	4.0	8.0	50	3	62	45
STP(A)	nd	32.9	4.6	37.5	30.8	122	0	88	12
STP(B)	2.3	17.3	1.6	21.2	28.9	73	11	82	7

\* ; Quantity of 17 $\alpha$ -estradiol activities

\*\*; means lower than detection limit

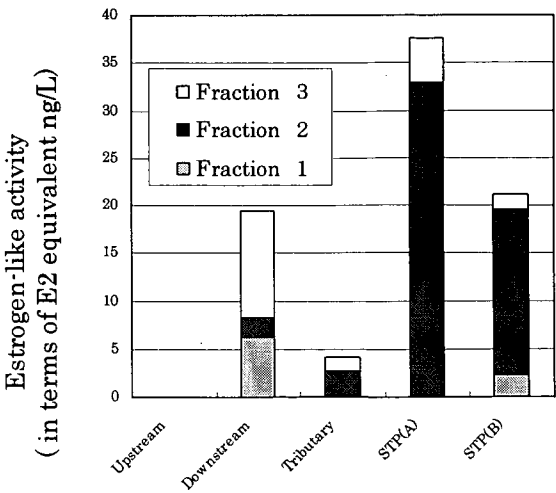


Figure 1

Estrogen-like activity of fractionated water samples

# RESEARCH ON THE ALTERATION OF ENDOCRINE DISRUPTORS IN WATER ENVIRONMENTS

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Water Quality Division

Project period: 1999 - 2002

## OBJECTIVES

The problem of chemical substances that cause endocrine disruption in plants and animals has been reported, and these substances include those such as nonylphenol (NP) that are formed by decomposition in a water environment. In order to be able to efficiently reduce their environmental load, it is, therefore, essential to clarify the behavior in water environments of, for example, nonylphenol ethoxylate (NPEO) that decomposes in water environments to produce nonylphenol.

## RESULTS

### (1) NPEO biodegradability test

Choosing polyethyleneglycol-mono-4-nonylphenol ether as a typical NPEO industrial product, we conducted a batch test to investigate NPEO's biodegradability and its by-products after biodegradation. In an airtight glass bottle, 30mg/L of activated sludge from a WWTP added with NPnEO as a carbon source, we measured the oxygen consumption, TOC and NPnEO concentration.

After a 10 day biodegradability test (Figure-1, 2), there was 12 mg/L of residual TOC even though the NPnEO concentration was reduced down to 0 mg/L, suggesting that by-products of the degradation of NPnEO were produced.

It is known that in NPnEO, the degree of polymerization of the ethylene oxide chains of the hydrophilic group is cut down under aerobic conditions, and eventually it is decomposed into nonylphenol monoethoxylate and nonylphenoxy acetic acid. However, in this 10 day period of the test, the NPnEO degradation could have occurred as far down to 2, 3, or 4 of ethylene oxide chains.

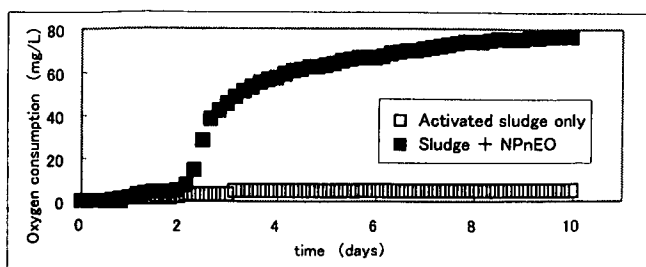


Figure-1 Oxygen consumption at NPEO biodegradability test

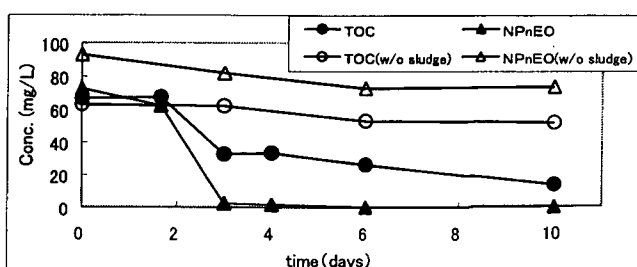


Figure-2 TOC and NPnEO concentration at biodegradability test

## (2) Experimental leaching test of NP, NPnEO, and NPEC from bottom sediment

It was hypothesized that NP is decomposed and leached from NPnEO in bottom sediment under both aerobic and anaerobic conditions. It is said that the formation of NPEC from NPnEO occurs under aerobic conditions and the production of NP under anaerobic conditions, but during this experiment, more NP was leached under anaerobic than under aerobic conditions. NPnEO,  $n \geq 5$  concentration suggested that the amount of degradation occurring in the water is greater than the amount of leaching from the bottom sediment. NPEC, which is a product of the degradation of NPnEO and an intermediate subject in the process of the formation of NP, is influenced by both the rate of degradation of NPnEO and by the rate of production of NP, and if the microorganisms that contribute to both of these factors differ, the concentration of NPEC may change in complicated ways.

The measurements of NP, NPnEO, and NPEC before and after the leaching test reveal that the concentration of NP increased slightly in the bottom sediment from its value at the beginning of the experiment under both aerobic and anaerobic conditions (Figure-3, 4). It was assumed that conditions are anaerobic inside the bottom sediment, NPnEO and NPEC are formed by anaerobic degradation. As NPnEO,  $n=1-4$ , while decreased under aerobic conditions, increased under anaerobic conditions. NPnEO,  $n \geq 5$  reveals that the concentrations were almost identical before and after the experiment at depths from 0 to 15 cm under both aerobic and anaerobic conditions, but at depths from 15 to 20 cm, the concentrations were higher when the experiment was completed. The trends for NPEC varied under aerobic and under anaerobic conditions. Under aerobic conditions, the concentration were higher in deep levels than at the surface, and under anaerobic conditions, the values were higher at the surface than at deep levels.

It is necessary to conduct more detailed study of the leaching of NP, NPnEO, and NPEC from the bottom sediment in order to comprehensively clarify both the leaching of these substances from the bottom sediment and their degradation in the water.

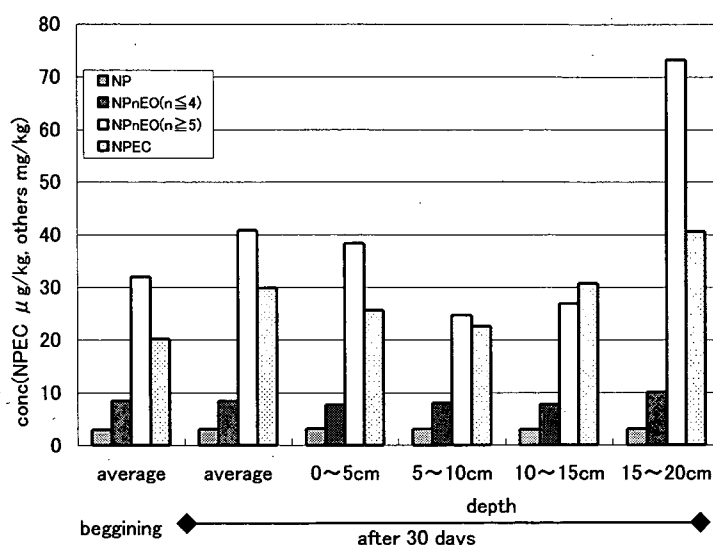


Figure-3 NPs concentration in sediments at aerobic test

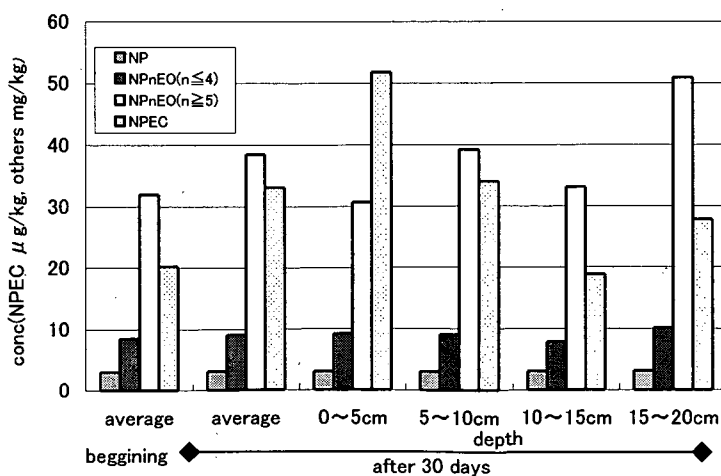


Figure-4 NPs concentration in sediments at anaerobic test

# NOVEL TECHNOLOGY FOR DETECTION OF MICROPOLLUTANTS IN WASTEWATER

H. Tanaka, K. Komori, A. Takahashi, Y. Okayasu and Y. Yakou  
Water Quality Division

Project period: FY1999 - 2000

## OBJECTIVES

### 1) Bioassay using nitrifying bacteria

It is very important to keep the condition of microorganisms stable in wastewater treatment process. However, there is possibility that these microorganisms are damaged with chemicals discharged into sewerage continuously or suddenly. Therefore, it is necessary to establish a method to estimate toxicity of chemicals against wastewater treatment ability. Moreover, it is also necessary to develop a technology to monitor toxicity in wastewater continuously. It is well known that nitrifying bacteria is affected with chemicals easily. In this research, bioassay methods using nitrifying bacteria have been examined.

In the fiscal year of 2000, relationship between dose and effect was examined about typical chemicals. Moreover, application of the toxicity monitor using nitrifying bacteria to real sewerage facilities was examined.

### 2) ELISA method

It has been noticed recently that some chemical disrupts an endocrine system in human or aquatic organism. And it has been feared that water environment is polluted by these endocrine disrupting chemicals (EDCs). It was reported that 17 $\beta$ -estradiol (E2) is the strongest EDCs in the substances that were being researched in sewerage. And some report concluded that capability of endocrine disrupting in wastewater was mainly caused by E2. However, there is possibility that E2 was overestimated in the mentioned research because an ELISA method without complicated pretreatment process of sample was used for measuring.

In the fiscal year of 2000, the reliability of ELISA method was examined comparing to the analytical LC/MS/MS method.

## RESULTS

### 1) Bioassay using nitrifying bacteria

Sensitivity of the bioassay against each chemical was different. A value of EC<sub>50</sub> for thiourea was the smallest in target substances. Values of EC<sub>50</sub> for other target substances are shown in the table-1. And, these values of EC<sub>50</sub> in the bioassay using nitrifying bacteria were about 1/20 times as large as the values of other bioassay system whose condition is near to real activate sludge process. It seems that the bioassay using nitrifying bacteria can detect chemical before activate sludge process is seriously damaged. On the other hand, the toxicity monitor, which had been developed for monitoring toxicity in influent, could work continuously for about a months.

Table-1 Values of EC<sub>50</sub> for target chemicals  
in the bioassay using the nitrifying bacteria (mg/L)

Target Chemicals	EC <sub>50</sub>	Target Chemicals	EC <sub>50</sub>
Phenol	0.701	Thiourea	0.018
2-Chlorophenol	0.11	m-Cresol	0.1
3-Chlorophenol	0.046	m-Aminophenol	0.94
4-Chlorophenol	0.009	Aniline	2.3
2,3-Dichlorophenol	0.054	o-Phenylenediamine	1.4
2,4-Dichlorophenol	0.15	m-Phenylenediamine	0.23
2,5-Dichlorophenol	0.041	p-Chloroaniline	0.91
2,6-Dichlorophenol	1.1	Formaldehyde	26
		Acetaldehyde	41

## 2) ELISA method

The measurement values of E2 in influent with the ELISA method were 2.0-8.3 times as large as the measurement values with LC/MS/MS method. Therefore, it seems that the ELISA method overestimate E2 in influent because wrong target substances were detected. There is no correlation between the measurement values with the ELISA method and the ones with the LC/MS/MS method. Moreover, there are no obvious trend in terms of wastewater treatment plant and sampling timing. Most of the measurement values with the LC/MS/MS of E2 in effluent were under the minimum determination limit and much smaller than the ones with the ELISA method. It seems that the ELISA method overestimate E2 in secondary effluent as in case of influent because wrong target substances were detected.

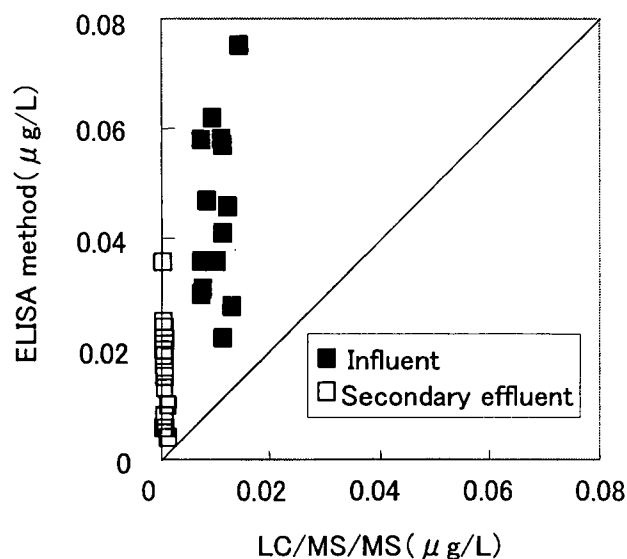


Fig.-1 Comparison between the measurement values  
with the ELISA method and with the LC/MS/MS



# TECHNOLOGICAL DEVELOPMENT FOR THE PREVENTION OF HAZARDOUS SUBSTANCES IN SEWERAGE

H. Tanaka, K. Komori, Y. Okayasu, K. Taketoshi, Y. Yakou and T. Higashitani  
Water Quality Division

Project period: FY2000-2000

## OBJECTIVES

Many chemical substances are indispensable for making human life comfortable and convenient, but some of them are suspected to have large toxicity to human and ecological health. At present, only a small number of them are regulated for discharge to public waters on the basis of Water Pollution Control Law and Sewerage Law in Japan.

Management of such chemical substances is quite important in sewerage systems, which play a role of returning treated wastewater to public waters. It is necessary to get information about identifying and quantifying chemical substances in untreated and treated wastewater, predicting their fates in wastewater treatment plants, and estimating their risk to human and ecological health, in establishing managing hazardous chemical substances in sewerage system.

In FY 2000, we studied on simulation model for predicting the fate of volatile organic compounds (VOCs) in activated sludge process. In addition, we developed a novel analysis method of estrogens, 17  $\beta$  estradiol (E2), estrone (E1) and ethynylestradiol (EE2), and nonylphenoxy acetic acids (NPECs) which are biodegradation product of nonionic surfactant, nonylphenol ethoxylates compounds (NPEO), in wastewater.

## RESULTS

### (1) A NEW SIMULATION MODEL FOR PREDICTING THE FATE OF VOCs IN ACTIVATED SLUDGE PROCESS

A new simulation model was developed for predicting the fate of VOCs in activated sludge process. This model considers adsorption, surface and bubble volatilization, and biodegradation from the dissolved phase as removal mechanisms. This model has the following characteristics compared with existing VOCs model: 1) model can reflect hydraulic influent and operational conditions in actual treatment process. 2) this model considers specific heterotrophic bacteria that degrade the target compound and autotrophic nitrifying bacteria in active biomass. 3) this model considers mass balance about biochemical reactions. 4) parameters on biodegradation by specific active bacteria can be estimated by biodegradation tests and simulation validating the tests. The model was evaluated with experimental data using pilot plant. Target compounds were benzene, dichloromethane, toluene and m,p-xylene. As a result, the model fairly simulated the fate of the target compounds in activated sludge process. However, further improvements were necessary. (see Fig.1)

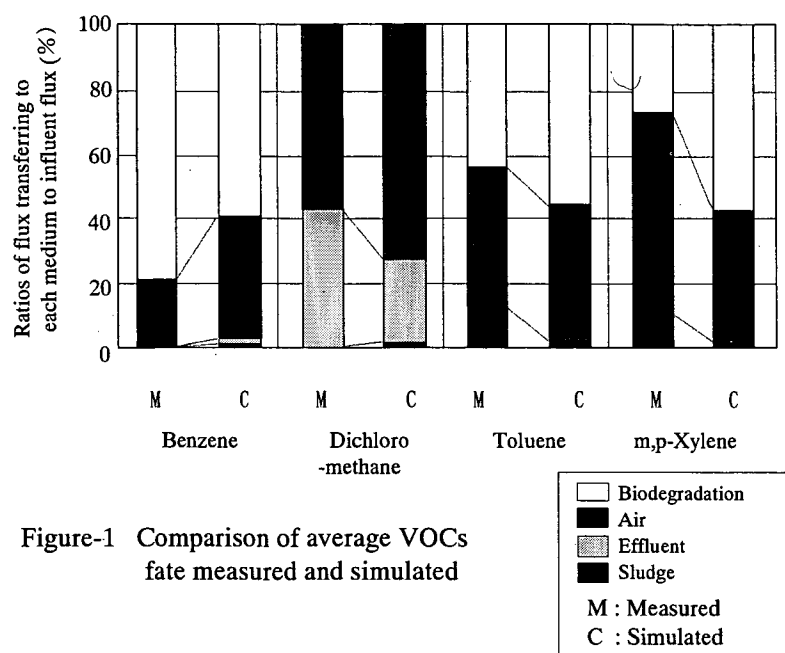


Figure-1 Comparison of average VOCs fate measured and simulated

## (2) DEVELOPMENT A NOVEL ANALYSIS METHOD OF ESTROGENS, E2, E1, EE2, AND NPEC IN WASTEWATER BY LC/MS/MS

Estrogens are deemed to be important substances that give estrogenic effects on fish in the water environment because they have higher estrogenic potentials than other industrial chemicals such as nonylphenol, bisphenol A and diethyl phthalate and are frequently found in river water and wastewater at significant levels. Therefore, we developed a novel analysis method for estrogens, E2, E1 and EE2 in wastewater.

Sample preparation in this method includes solid-phase extraction with Sep-Pak Plus C18 for the filtrate, supersonic liquid extraction for suspended matter and clean-up with Sep-Pak Plus florisil. A prepared sample is analyzed by LC/MS/MS. Detection limit and recovery ratios of estrogens in this method are  $0.0005 \mu\text{g/l}$  and  $92 \sim 113\%$  respectively. Significant differences were found in E2 concentrations between by LC/MS/MS and by ELISA that were commonly applied in field surveys in Japanese river and sewerage. E2 concentrations by ELISA showed much higher values than those by LC/MS/MS for wastewater samples and river water samples.

NPECs are deemed to be important substances in understanding the decomposition behavior of nonylphenol that is one of the key estrogenic substances in water environment. Therefore, we developed a novel analysis method for NPECs in wastewater. Samples were filtered using a glass fiber filter with  $1 \mu\text{m}$  pore. The remaining sample on the filter was extracted with supersonic extraction by 5 ml acetone. NPECs were extracted from filtrate and supersonic extraction solution using Sep-Pak Plus PS-2. NPECs were eluted from the PS-2 cartridge with 5 ml of methanol. Elute solution was analyzed by LC/MS/MS. Detection limit and quantification limit of NPECs were 0.2 and  $0.6 \mu\text{g/l}$ , respectively. Detection limit and quantification limits were defined as three times and ten times of the calculated standard deviation from the measurement of actual effluent water. Recoveries of effluent water were about 80 %.

# EVALUATION OF AQUATIC ECOSYSTEMS AFFECTED BY SEWERAGE SYSTEMS

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Water Quality Division

Project period : FY 1998-2000

## **OBJECTIVES**

There are two major routes through which fish are exposed to endocrine disrupters (EDs); one route is through water that is surrounding environment; the other is through aquatic food such as algae and benthos. Few studies on the bioaccumulation of EDs in aquatic food have been conducted. Therefore, we evaluated the concentrations in the food of nonylphenol (NP), bisphenol A (BPA) and 17 $\beta$ -estradiol (E<sub>2</sub>) using GC/MS. These EDs were frequently detected in river water in fact finding surveys conducted by Ministry of Land, Infrastructure and Transport (MLIT). In this year, we focuses on the middle reaches of the Tama River in Tokyo, it is one of the most urbanized rivers in Japan, where the feminization of carp has been reported.

## **METHODS**

We collected river water, periphytons and benthos at the mainstream (St.1 to St.3) and its tributaries (St.4 to St.8) and analyzed NP, BPA and E<sub>2</sub> in them. Because only a limited number of chemicals are known as EDs at present, total estrogenic activities are not thought to be covered by the EDs that were measured in this study. Thus, we measured the estrogenicity of the samples by recombinant yeasts involving human estrogen receptor.

## **RESULTS**

The identified species names and weights of the benthos are shown in Table 1. The species of the benthos in the mainstream differ from those in the tributaries. The benthos consisted mainly of crayfish in the tributaries while river shrimps in the mainstream. The concentrations of EDs and the estrogenicity are shown in Table 2. NP concentrations ranged 0.1- 0.4  $\mu$  g/L in the river water, while they ranged from 8- 130  $\mu$  g/kg-wet in the algae and 8- 140  $\mu$  g/kg-wet in the benthos. BPA concentrations ranged 0.02- 0.15  $\mu$  g/L in the river water, while they ranged from 2- 8.8  $\mu$  g/kg-wet in the algae and 0.3- 12  $\mu$  g/kg-wet in the benthos. E<sub>2</sub> concentrations ranged 0.0001- 0.0076  $\mu$  g/L in the river water, while they ranged from 0.09- 2.26  $\mu$  g/kg-wet in the algae and <0.01- 0.22  $\mu$  g/kg-wet in the benthos. E<sub>2</sub> is more concentrated than NP and BPA in the algae, but less in the benthos than NP and BPA.

The estrogenicity ranged 0.0001- 0.0464  $\mu$  g/L in the river water, while it ranged from 3.4- 66.8  $\mu$  g/kg-wet in the algae and 7.4- 5458  $\mu$  g/kg-wet in the benthos. The estrogenicity is more concentrated than individual EDs concentration in the benthos.

E<sub>2</sub> in the benthos concentrations from their estrogenicity are quite different. It suggests that E<sub>2</sub> is more metabolized than NP and BPA or less concentrated via the algae in the benthos. The estrogenicity of the eriphyton

was larger than that of in the water, but the periphytons may synthesize estrogenic chemicals like phytoestrogens.

As shown in Table 3, bioaccumulation factors of NP are estimated 160-650 for the periphytons, 63-990 for the benthos, and 8-170 for the benthos, respectively. The factor of BPA and E<sub>2</sub> showed 18-650 and 64-1200 for the periphytons, 8-170 and 100-160 for benthos, respectively. Although some benthos are grazers of periphytons, the bioaccumulation factors of NP and BPA in the benthos are not always larger than those in the periphytons. Because the benthos included aquatic insects, crustacean and others, some portion of NP and BPA may be metabolized in the benthos.

The results suggest that food may be a more important route for fish exposed to NP, BPA and E<sub>2</sub> than via water. Relative estrogenic potential of NP was estimated 0.0001 and these of BPA was 0.00007 by using recombinant yeast in our previous study. This suggests that E<sub>2</sub> may have larger contribution to fish in the Tama River than NP and BPA.

Table-1 Species and Weight of Benthos

		Stonefly	Dragonfly larva	Crayfish	River shrimp	River crab	Others	Total
Main stream	St.1	15.0	-	-	-	-	35.0	50.0
	St.2	15.2	1.9	-	-	-	38.1	55.2
	St.3	-	0.3	-	40.0	-	12.1	52.4
Tributary	St.4	3.5	0.8	-	4.3	5.0	30.5	44.1
	St.5	-	1.1	44.8	0.3	-	14.5	60.7
	St.6	-	-	61.1	4.0	-	5.0	70.1
	St.7	-	0.1	45.2	-	-	18.4	63.7
	St.8	-	-	47.1	4.6	-	3.0	54.7

Weight: g-wet

Table-2 Concentration of EDs and Estrogenicity

	NP			BPA			E <sub>2</sub>			Estrogenicity		
	periphytons <sup>*</sup>	benthos <sup>*</sup>	water <sup>**</sup>	periphytons <sup>*</sup>	benthos <sup>*</sup>	water <sup>**</sup>	periphytons <sup>*</sup>	benthos <sup>*</sup>	water <sup>**</sup>	periphytons <sup>**</sup>	benthos <sup>***</sup>	water <sup>****</sup>
St.1	8	8	<0.1	2.9	1.2	<0.01	0.12	<0.01	0.0001	8.1	ND	0.0001
St.2	58	140	0.2	6	12	0.07	2.26	<0.01	0.0027	67.0	1600	0.0120
St.3	31	99	0.1	4.2	6	0.05	1.24	<0.01	0.0038	63.0	47.0	0.0095
St.4	64	25	0.4	8.8	1.7	0.14	0.26	<0.01	0.0012	10.0	7.4	0.0022
St.5	30	31	0.1	6.7	1.2	0.15	0.22	0.03	0.0003	3.4	11.0	0.0006
St.6	94	14	0.2	2.2	6.8	0.12	1.76	<0.01	0.0076	23.0	5500	0.0460
St.7	68	<3	0.2	2	0.4	0.03	0.13	<0.01	0.0019	5.0	ND	0.0073
St.8	130	<3	0.2	13	0.3	0.02	0.09	0.22	0.0014	3.5	10.0	0.0047

g/kg-wet, g/L, g-E2equivalent/kg-wet, g-E2equivalent/L ND: not detect

Table-3 Bioaccumulation factors of EDs and Estrogenicity

NP		BPA		E <sub>2</sub>		Estrogenicity	
periphytons	benthos	periphytons	benthos	periphytons	benthos	periphytons	benthos
St.1	-	-	-	1,200	-	81,000	-
St.2	290	700	86	170	840	5,800	140,000
St.3	310	990	84	120	330	6,600	4,900
St.4	160	63	63	12	220	4,600	3,400
St.5	300	310	45	8	730	5,400	17,000
St.6	470	70	18	57	230	500	120,000
St.7	340	-	67	13	68	680	-
St.8	650	-	650	15	64	740	2,200
Average	360	430	140	56	460	13,000	48,000

-: not calculated

# SYSTEMATIC WAY OF SURVAYING WATER QUALITY OF RIVERS

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Water Quality Division

Project period: 2000 - 2002

## OBJECTIVES

Water quality surveys are now performed in conformity with the River Erosion Control Technical Standard, but each river administrative official is allowed to independently decide the detailed survey locations and survey frequency. It is necessary to clarify the significance of the measurements and set rational survey conditions for the river water quality survey to perform more efficient water surveys, but at this time, river water quality surveys are not necessarily systematized.

This study, has been therefore, carried out cooperatively with a couple of technical offices in order to systematize water quality surveys and finalize the water quality survey regulation. The Public Works Research Institute will study the basic theory of water quality surveys performed by river administrative officials and in particular, the bottom sediment survey method that is one specific category of survey method that has not been sufficiently systematized. In FY2000, both foreign and domestic water quality survey planning conceptions are studied and existing documents classified.

## RESULTS

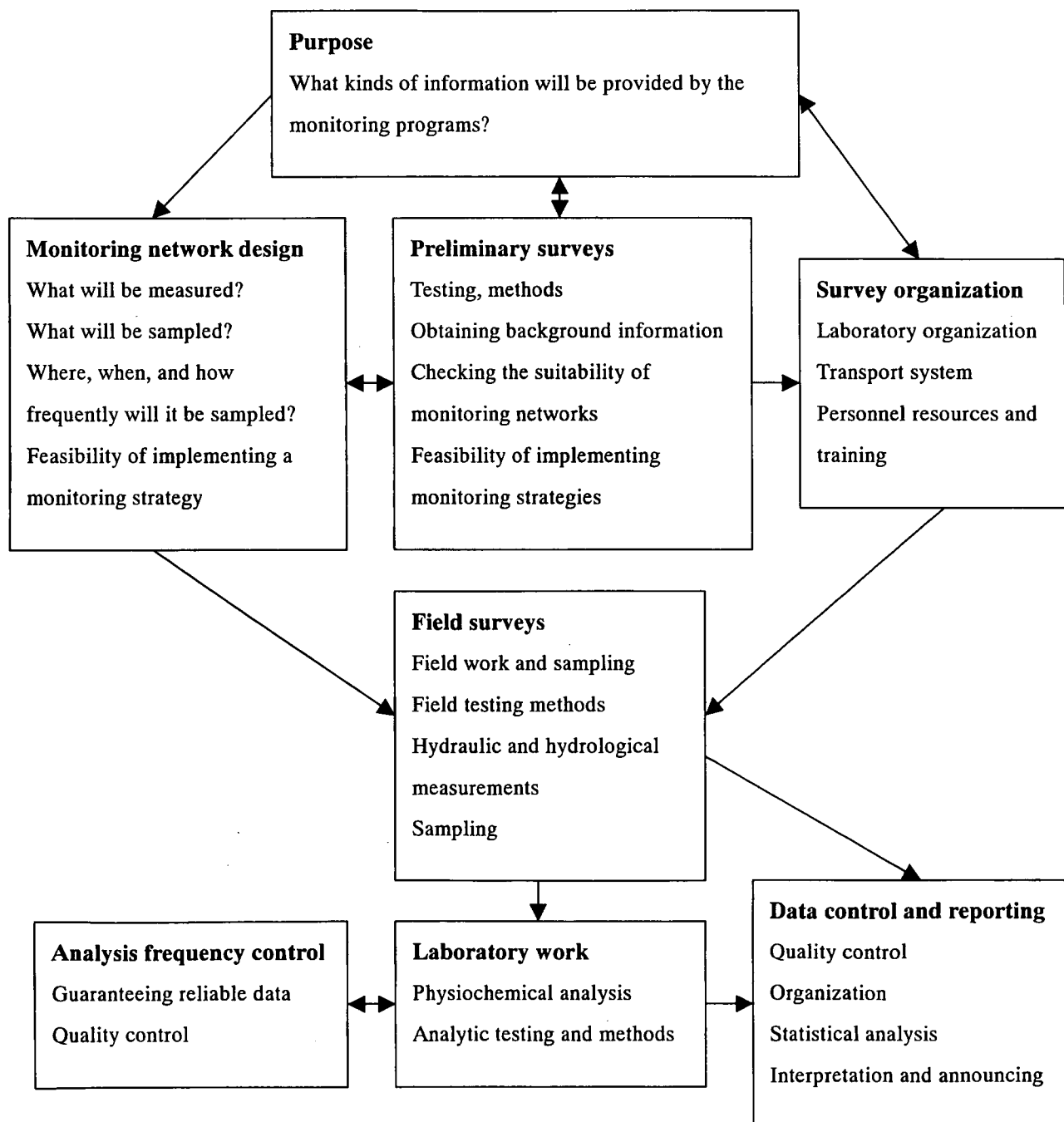
Survey regulation we are working on is applied to river, lake, and marsh water quality and bottom sediment surveys performed as part of river water quality control (below, referred to as, "River water quality surveys"). As the scope of environmental standards has expanded in recent years, river administrative officials have surveyed more river water quality characteristics at greater frequency than before. But decisions concerning survey locations are not necessary made rationally. For the above reasons, this survey regulation classifies river water quality surveys by purpose, and describes rational methods of selecting survey locations and methods of setting survey frequency in accordance with each purpose. The bodies of water covered by this survey regulation are rivers (but in principle, not including dam reservoirs and land inundated by weirs), lakes and marshes, and the groundwater in their surroundings.

"River water quality surveys" will cover river water quality surveys carried out to achieve the following four purposes.

- (1) Water quality surveys to execute and evaluate projects
- (2) Water quality surveys to monitor public waters
- (3) Water quality surveys for water utilization
- (4) Water quality surveys to conserve ecosystems

And this "River water quality surveys" will also deal with surveys necessary for water pollution

surveys and analysis by automatic water quality monitoring systems which are usually carried out to supplement the above water quality surveys. Since we have some guidelines for surveys following water quality accidents written in the Water Quality Accident Countermeasure Technology 2001 Edition (Water Quality Cooperation Committee of the Ministry of Land, Infrastructure and Transport), “River water quality surveys” will not be applied to those surveys follow water quality accidents.



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