

INVESTIGATION OF COLIPHAGES AND FECAL COLIFORM IN A COMBINATION PROCESS OF ANAEROBIC FILTRATION AND AEROBIC BIOFILM FILTRATION SYSTEMS

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1. Introduction

Coliphages are bacteriophages that infect and replicate in coliform bacteria and appear to be present wherever fecal coliform are found. Coliphages₅ in domestic sewage were detected in the average range of 10^2 to 10^5 PFU/mL (Furuse K., 1987). A quick and reliable way of reducing their numbers is found in wastewater treatment. Since, under the very diverse in nature and different condition operation of wastewater treatment, the various degrees of coliphages removal are found in various sewage treatment systems. However, the significant removal of them in biological wastewater treatment process occurs mainly by association onto biomass particulates with the subsequent sedimentation or inactivated by biological antagonism. Here we report an investigation the fate of coliphages in aqueous phase (unadsorbed) and particulate-associated phase (adsorbed), and fecal coliform in a pilot domestic wastewater treatment system which is a combined process of anaerobic and aerobic systems.

2. Materials and Method

Pilot Wastewater Treatment System Wastewater samples were taken from a pilot domestic wastewater treatment system which is located at Shibayama area. This wastewater treatment system is a combined process of anaerobic filtration and aerobic biofilm filtration systems. The wastewater treatment unit with the total effective volume of 2.5 m^3 is used to treat domestic wastewater under a influent flow rate of $1.25 \text{ m}^3/\text{day}$. Circulation of treated water from storage chamber to anaerobic filtration chamber was done due to denitrification purpose. Back wash is also operated at night time with the flow rate of 200 liter/day. Detention times of anaerobic filtration chamber and aerobic biofilm filtration chamber are 7.2 h. and 1.1 h., respectively. Polyethylene ring was fixedly used as filter media in anaerobic filtration chamber. While sphere ceramic was used as supporting media in aerobic biofilm filtration chamber. Fig.1 shows the combination process of this system and the wastewater sampling points. Wastewater sample from aeration tank was collected both of wastewater and ceramic with the ratio of 50% by volume.

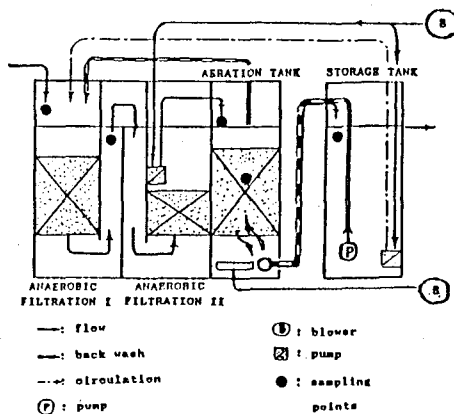


Fig.1. Flow diagram and sampling points of the wastewater treatment system at Shibayama area.

Bacteria Host Strains The bacteria host strains used in this investigation was Escherichia coli K-12, $F^+(A/\lambda)$.

Coliphages Concentration Measurement Coliphages concentration measurement was done by double-agar layer method. RNA-F-specific

coliphages concentration were determined by plating with and without RNase A, Sigma (200 µg/plate) on lawn of bacteria host cell. The difference of counts on plates with and without RNase showed the number of RNA-F-specific coliphages. Adsorbed and unadsorbed coliphages were detected by using the filtration-elution technique as explained by Ketratanakul and Ohgaki (1988). But, 10% buffered beef extract (10 g of paste beef extract, 1.79 g of $\text{Na}_2\text{HPO}_4 \cdot 12\text{H}_2\text{O}$ and 0.13 g of $\text{C}_6\text{H}_8\text{O}_7 \cdot \text{H}_2\text{O}$ in 100 ml. of distilled water) was used as eluent. Elution was performed at 4 °C.

Fecal Coliform Determination Fecal coliform concentration is measured by using isogrid membrane filtration method.

3. Results and Discussion

The chemical and physical characteristics of wastewater in this treatment plant are shown in Table 1. The characteristics of wastewater sample from aerobic chamber, such as suspended solids, coliphages and fecal coliform were determined by including suspended solids particulates and attached biofilm from ceramic.

Table.1 Chemical and Physical Characteristics of Wastewater in a Combination Process of Anaerobic Filtration and Aerobic Biofilm Filtration System at Shibayama Area.

| Sampling times | Soluble COD (mg/l) | | | | Suspended Solids (mg/l) | | | | pH | | | | DO (mg/l) | | | |
|----------------|--------------------|-----|------|-----|-------------------------|-----|------|-----|-----|-----|------|-----|-----------|-----|------|-----|
| | Inf | All | Aero | Str | Inf | All | Aero | Str | Inf | All | Aero | Str | Inf | All | Aero | Str |
| 13, Jun | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - |
| 23, Jul | 112 | 74 | 42 | 9 | 57 | 18 | 1106 | 5 | 7.4 | 7.6 | 7.3 | 7.3 | 2.3 | 1.9 | 4.1 | 2.1 |
| 4, Sep | 70 | 52 | 67 | 6 | 100 | 24 | 4379 | 7 | 7.4 | 7.3 | 7.1 | 7.1 | 0 | 0.6 | 4.6 | 3.6 |
| 30, Oct | 145 | 16 | 12 | 4 | 164 | 10 | 4106 | 10 | 7.9 | 7.4 | 7.4 | 7.3 | 0 | 1.0 | 4.3 | 2.5 |
| 13, Nov | 115 | 89 | 76 | 49 | 86 | 12 | 7050 | 4 | 7.4 | 7.3 | 7.2 | 7.1 | 1.5 | 0.4 | 3.1 | 3.4 |

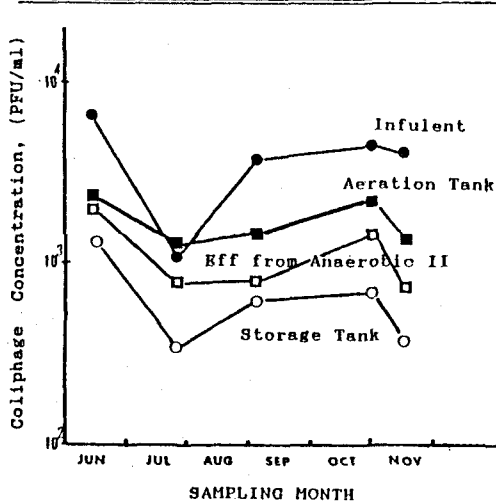


Fig.2 Total indigenous coliphages detected along the wastewater treatment system.

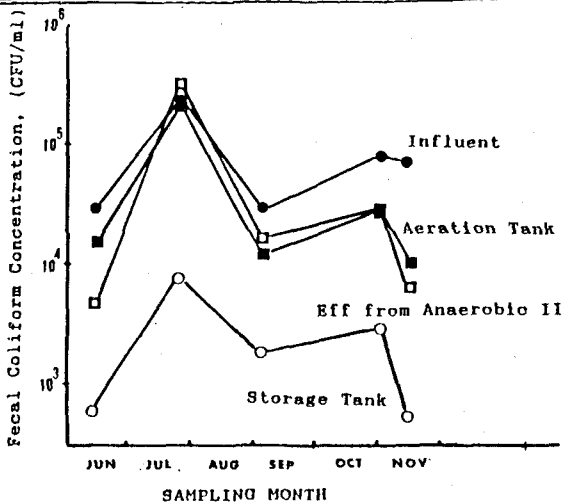


Fig.3 Fecal coliform detected along the wastewater treatment system.

Total indigenous coliphages in raw sewage was detected in the range of 1.0×10^3 to 6.2×10^3 PFU/mL (Fig.2). Total RNA-F-specific coliphages and DNA-F-specific coliphages concentration in raw sewage were in the range of 8.6×10^2 to 3.3×10^3 PFU/mL and 1.6×10^2 to 4.6×10^3 PFU/mL, respectively. (raw data not present here). Fecal coliform in raw sewage was found to be in the range of 2.8×10^4 to 2.6×10^5 CFU/mL. (Fig.3). After anaerobic filtration treatment, the reducing numbers of indigenous coliphages and fecal coliform was found to be 25-82% and 45-90% respectively. But, after the final stage of wastewater treatment, the percent removal of indigenous coliphages and fecal

coliform was highly observed to be 67-91% and 93-99%, respectively. However, the concentration of fecal coliform and coliphages at the final stage of wastewater treatment were found to be quite high concentration. Fig.2 and Fig.3 show the concentration of coliphages and fecal coliform in storage tank were in the range of 3.4×10^2 to 1.3×10^3 PFU/mL and 5.2×10^2 to 8.4×10^3 CFU/mL, respectively.

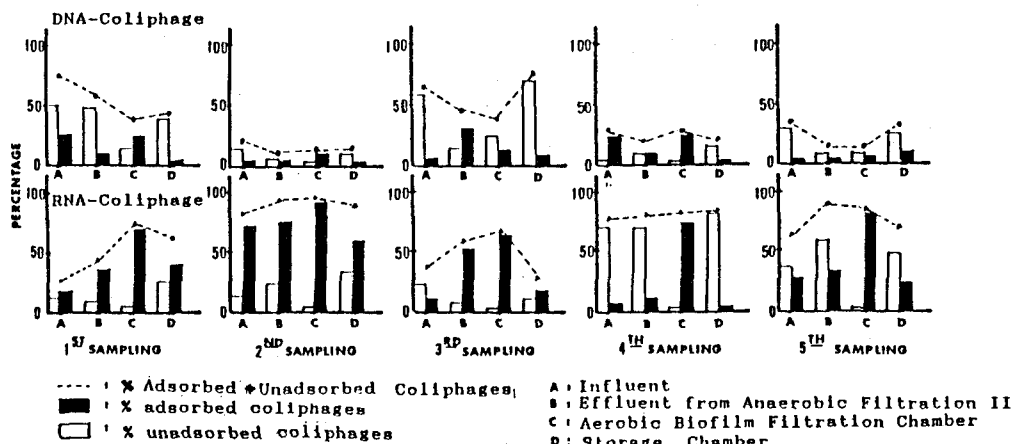


Fig.4 Percentages of adsorbed and unadsorbed coliphages in raw sewage, effluent from anaerobic filtration II, aerobic biofilm filtration and storage tank.

Fig.4 shows the percentage of adsorbed and unadsorbed of RNA-F-specific coliphages and DNA-F-specific coliphages present along this wastewater treatment. RNA-F-specific coliphage was found significantly in the wastewater. In raw sewage, RNA-F-specific coliphages were measured to be in the range of 25-84% of total coliphages. In anaerobic and aerobic chambers, RNA-F-specific coliphages were found in the range of 43-94% and 65-92% of total coliphages, respectively. A dominant type of particulate-associated RNA-F-specific coliphages was mainly found in aerobic biofilm filtration chamber (64-91% of total coliphages). This result is consistent with Kettratanakul's study (1988) that coliphages in the aeration tank of activated sludge system were mainly associated to suspended solids and most of them were RNA-F-specific coliphages.

DNA-F-specific coliphages were observed to be removed from this wastewater treatment system with higher percentage than RNA-F-specific coliphages. (79-93% for DNA-F-specific coliphages and 48-90% for RNA-F-specific coliphages) This indicates that RNA-F-specific coliphages are more tolerant in this wastewater treatment than DNA-F-specific coliphages. Therefore, Fig.4 shows that the percentage of RNA-F-specific coliphages increased in aerobic and anaerobic chambers while, the percentage of DNA-F-specific coliphages decreased.

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