CHAPTER 3

MATERIAL AND METHOD

3.1 Sources of Wastewater and Soil

3.1.1 Wastewater

The domestic wastewater was collected from grit chamber at Huaykwang Sewage Treatment Plant every 7 days during 7.30-8.30 A. M. and was taken to the Department of General Science laboratory, Faculty of Science, Chulalongkorn University within 1 hour. Chemical and bacteriological characteristics of the samples were analyzed immediately. Another portion was applied to the soil columns. The remaining wastewater was stored at 4°C and was held until it reached room temperature before added to the columns again. Samples were obtained during November 1986 through March 1987.

3.1.2 Soil

Four soil series were used in this experiment. The location, where soil samples were chosen by soil map (77, 78, 79, 80). These soil were obtained during September 1986 through October 1986. All of them can generally be classified by 2 systems as shown in **Table 3.1.** The general characteristics of these soil can be described as follows:

Pak Chong series The taken soil is located about 6 kilometres northeast from Mitraparp Highway (at Km 100), Wat Mitraparb, Ban Klang Dong, Amphoe Pak Chong, Nakorn Ratchasima Province. It is an area of red clayer soil developed from shale

associated with Carboniferous and/or Permian limestone. Kaolinite is the dominant clay minerals and the percentage of free iron oxide is 7.5-7.6, and soluble aluminium is Ø.ØЗ-Ø.Ø5%. Permeability is estimated to be rapidly.

Khamphaeng Sean series The soil sample used in this experiment is located about 5ØØ meters east of Maraiman Road, Amphoe Khampaeng Saen, Nakorn Pathom province. This is a brown or strong brown clay loam, weakly developed, argillic B horizon. Permeability is morderate and runoff is slow.

Muak Lek series The sample examined is located about 12 kilometres northeast of Mitraparb Highway and Muak Lek railway station, at Wat Pakklong, Amphoe Muak Lek, Saraburi Province. Most of the area is occupied by clay loam with common fine faint reddish brown mottles and common very fine tabular pores. Permeability is moderate. Surface runoff is rapid.

Ban Bung series The soil sample is located about 1 kilometres east of Sukumvit Road, Ban Donhualou, Amphoe Panatnikom, Chonburi Province. This is sandy loam to loamy sand. Permeability is rapid. Surface runoff is slow.

Samples for the experiment were collected at 5Ø cm below the surface by driving 7.5 cm ID. and 6Ø cm length of PVC pipes by a jack in horizontal direction as shown in **Figure 3.1.** After the columns were filled, they were moved. The ends of soil columns were sealed and covered at the two ends with plastic. The sealed ends removed, when the experiment were set. Each soil series was collected for 3 columns. Two of them were treated with domestic wastewater while the remaining was cut into 7 sections. Sections A, B, C, D, E, F, and G were cut at 5, 1Ø, 2Ø, 3Ø, 4Ø, and 5Ø cm, respectively, far from the end of soil column which connected to the PVC pipe contained domestic wastewater.

Table 3.1 Classification of the soils under investigation

Soil Series	Soil Taxonomy[1]	[2]National	Location	Texture	Permeability
Pak Chong	clayey, kaolinitic Oxic [3]Paleustult	Reddish-Brown Lateritic	Nakorn Ratchasima	clay	rapid
Khamphaeng Saen	fine loamy, Udic [4]Haplustalf	Noncalcic Brown	Nakorn pathom	silty loam	moderate or slow
Muak Lek	loamy, Lithic Haplustalfs	Noncalcic Brown	Saraburi	silty loam	moderate
Ban Bung	sandy, Aquic Arenic [5]Eutrochrepts	Hydromorphic Regosolic Grey Podzolic	Chonburi	loamy sand	very rapid

[1]USDA = United Stated Department of Agriculture, Soil taxonomy

[2]Dudal and Moormann classification 1964

[3]Paleustults = Pale + Ust + Ult

[4]Haplustalfs = Hapl + Ust + alf

[5]Eutrochrepts = Eutr + Ochr + ept

Pale - Gk. hapeos, old, excessive delvelopement

Hapla - Gk. haplous, simple, minimum horizon

Eutr - Gr. eutrous, high bade saturation

Ust - L. ustus, burnt, dry climate, usually hot in summer, ustic moisture regime

Ult - L. Ultimus (last), ulitimate

Alf - aluminum & ferric of pedalfer

Ochr - ochric, pale color, low organic matter

Calc - calic horizon

Figure 3.1 Collection of soil samples

3.2 Experimental units

The experimental unit consisted of a 7.5 cm ID, and 100 cm, long polyvinyl chloride (PVC) pipe for filling with domestic wastewater and the same ID. but only 60 cm long PVC pipe for soil column. At the two ends of soil column were covered with 80-mesh nylon net in order to prevent the erosion of soil by domestic wastewater flow. The experimental unit was shown in **Figure 3.2-3.3.** At the lower end of each tee, a 20 cm long PVC pipe of same ID. is connected, and sealed with cement for blocking wastewater flow out. Therefore, wastewater confined through soil column in horizontal direction only and 1,000-ml, sterilized Erlenmeyer flask was used to collected the effluent and determine E.coli immediately, then used to collecting them for chemical analyzing. Five liters of domestic wastewater was poured into PVC pipe of each unit, and the initial level of each soil series was marked immediately. The wastewater was added to each unit as equal as the initial level in other next times, the volume of adding wastewater was recorded.

3.3 Preparation of Wastewater and Soil Samples

3.3.1 Wastewater

After collecting from Haukwang Treatment Plant, the chemical and bacteriological properties of the influent were analyzed immediately. During the experiment was performed, the flask had been changed daily. Several parameters of the effluent, i.e., E.coli, pH, N, P, COD, Cl, SO_4 and cations, were analyzed on a daily basis. The result of each parameter was the average value of three analyzed samples of each duplicated column.

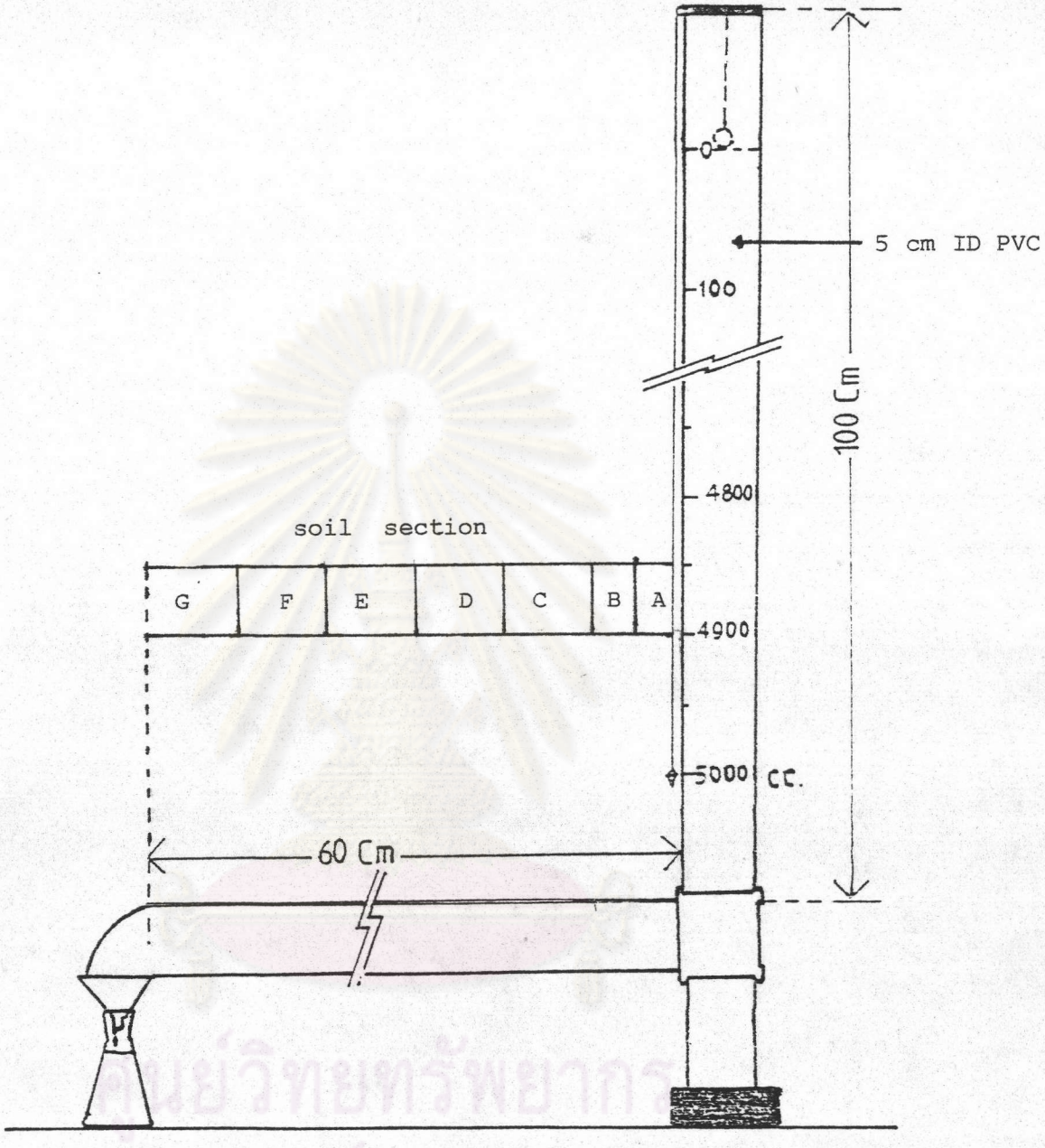

Figure 3.2 Typical soil column that used in this experiment

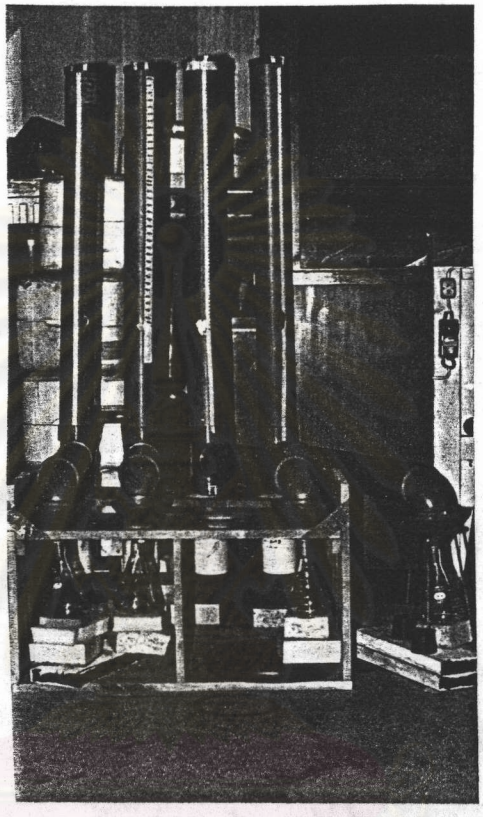

Figure 3.3 Soil column that was used in this experiment

3.3.2 Soil

When 20-week period passing with wastewater that soil was terminated, the experimental units were gradually dried for about one week. The soil column was removed from the experimental unit for viable count of E.coli. Soil samples were collected by making a hole on the top of soil column at 5, 10, 20, 30, 40 and 50 cm, long. After collecting of soil samples, the hole was covered with master tape. The soil columns were frozen for 2 days and divided into seven sections. Gravels and roots were removed from the soils of each section, and let them air-dried about one week. Then, they were crushed to a size small enough to pass 10 mesh (2mm) sieved, mixed well and stored in a plastic container. In this experiment, each air-dry section was carried out in triplicate of the duplicated columns and the results shown were the average.

3.4 Method

3.4.1 Wastewater

The influent and effluent were analyzed several parameters as in **Table 3.2** at the laboratory of the Department of General Science, Faculty of Science, Chulalongkorn University. Method of procedures were described in **Appendix A.**

Table 3.2 The characteristics and analytical method of wastewater (69).

Parameters	Analytical Method
pH	pH meter
COD (mg/l)	Dichromate reflux method
NH_4-N (mg/l)	Kjedahl method for the influent and direct nesslerization for the effluent
TKN (mg/l)	Kjedahl method
NO_3-N (mg/l)	Nitrate electrode method
Orthophosphate (mg/l)	Vanadomolybdophosphoric acid colorimetric method
Total phosphate (mg/l)	Persulfate digestion
Na and K (ppm)	Flame photometer
Ca and Mg (ppm)	Atomic absorption spectophotometer
Cl (ppm)	Mohr method
SO_4 (ppm)	Turbidimetric method
E. coli (col/ml)	Direct plate count and IMViC test

3.4.2 Soil

E.coli content in soil samples was analyzed at the Department of General Science, Faculty of Science, Chulalongkorn University. Analysis of physical and chemical properties of soil samples were performed at Ecology Section, Chemical Soil Analysis Division, Land Developement Department, the Ministry of Agriculture and Coopertives. Parameters of soil samples are shown in **Table 3.3**and procedures were described in **Appendix B.**

Table 3.3 The characteristics and analytical method of soil (81)

Parameters	Analytical Method
Texture	Pipet method
pH	pH meter
CEC (meq/100g)	Sodium saturation
Organic carbon (%)	Walkey black method
NH_4-N (%)	Kjeldahl method
TKN (%)	Kjeldahl method
NO_3-N (%)	Nitrate electrode method
Extractable phosphorus (ppm)	Extract with dil. $HCl+H_2SO_4$ and detect by ascobic acid method
Total phosphorus (ppm)	Extract with fusion Na_2CO_3 and detect by ascobic acid method
Exchangeable Na, K (meq/100g)	Extract with 1 N NH_4OAc at pH 7 and detect by flame photometer
Exchangeable Ca, Mg (meq/100g)	Extract with 1 N NH_4OAc at pH 7 and detect by atomic absorption spectrophotometer
Cl (meq/100g)	Extract with distilled water 1:2 and detect by Mohr method

Parameters	Analytical Method
SO_4 (meq/100g)	1:2 soil/water and detect by turbidimetric method
E. coli (col/g)	Direct plate count and IMViC test