CHAPTER 3 : METHODOLOGY

3.1 Site Description

10 sampling sites of which 4 types of water in different areas of Chiang Mai province (stream (ST), river (R), irrigation canal (IC), and sewage canal (SC) habitats) were chosen. Water samples were collected once a month starting from May to December 1996. The characteristics of the sampling sites were described as follow;

Streams

Four sampling sites were selected from 2 streams, three sites from Mae Sa stream and another from Huay Kaew stream. Mae Sa stream, a 5-10 m wide and approximately 19 km long stream, locates in Mae Rim District (14 km far from Chiang Mai city) at vary elevation of 600-800 m above sea level. This stream is assumed to receive organic discharge from human activities such as resorts, botanical garden, agriculture areas, elephant camps, orchid farms and household along the Mae Rim-Samueng road. In addition to the geology of this stream, stream line passes through those mentioned activities above before going to Mae Sa waterfall, one of popular tourist spots in Doi Suthep Pui National Park, and then discharge to Ping river. Therefore, tourists have easily been exposed to the pollutants. Huay Kaew stream, a small stream originated from Doi Suthep Pui National Park, passing through Chiang Mai Zoo. This stream is assumed to contains organic loading from the zoo and discharges to Ang Kaew Reservoir in Chiang Mai University.

ST1 : A sampling site of Mae Sa stream, located on latitude 18° 53 N and longitude 098° 49 E, Sri Muang Khun subdistrict, Mae Rim District. The elevation is 800 m. Average stream width is 8.25 m and depth is 0.54 m. The surroundings are dense of trees and grasses on both side banks. Water body is visually clear. This site might be impacted from the road near by and the garbage dumping on the bank (Fig. 2).

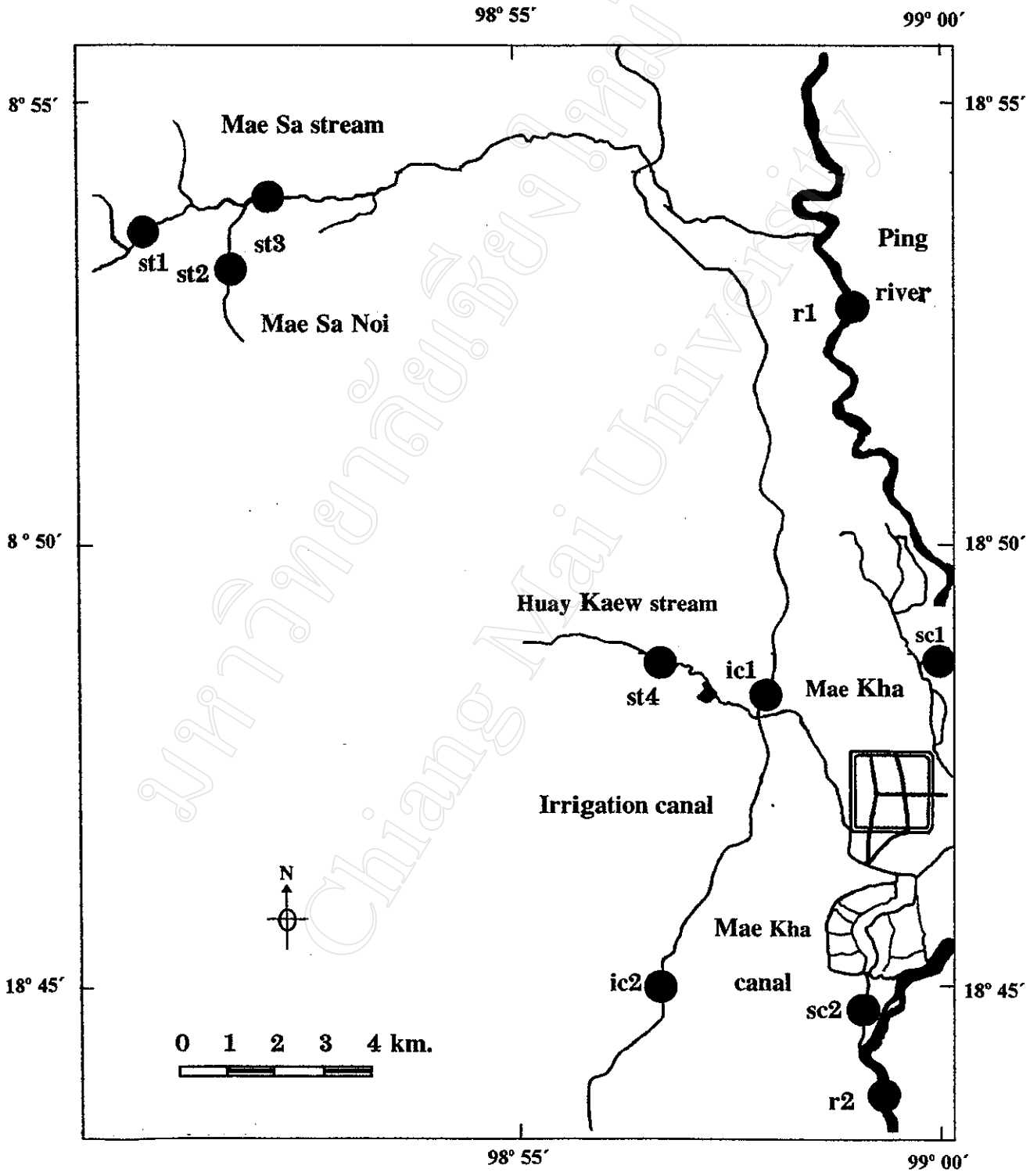

Fig. 1 : Map of the study areas

ST2 : A small stream called Mae Sa Noi, located in a Queen Sirikit Botanical Garden on Mae Rim-Samueng Road, Mae Rim District. It is the site which has been known very less pollution and discharges to Mae Sa stream. Land cover in this area is predominantly forest which provide shade condition to the ground. The water body was very turbid during the study period. The grid position is on latitude 18^{o} 53 N and longitude 098^{o} 51 E. Elevation is 720 m. Average stream width is 0.88 m and depth is 0.34 m (Fig. 3).

ST3 : Another sampling site of Mae Sa stream also locates in the Botanical Garden. The grid position of this stream is on latitude 18^{o} 53 N and longitude 098^{o} 51 E, elevation is 680 m. The surroundings are more open with fewer big trees but dense of grasses and bushes on both side banks. Water body is visually clear. The average stream width is 5.13 m and depth is 0.41 m (Fig. 4).

ST4 : A sampling site of Huay Kaew stream in vicinity of Chiang Mai zoo, public park and village. The grid position is on latitude 18^{o} 52 N and longitude 098^{o} 56 E. The area is relatively open with fewer trees near the site but dense of grasses along the banks. The elevation is 350 m and water body is visually clear. The average width is 2.98 m and depth is 0.25 m (Fig. 5).

River

Two sampling sites were selected on Ping River, one of the famous rivers in northern Thailand, running through Chiang Mai province before joining other three rivers Wang, Yom, and Nan. Ping River is a main natural water resource for almost all activities of Chiang Mai people such as for water supply, agriculture, industries, recreation, fishing and so on, and currently receives non-treated discharge from Chiang Mai city.

R1 : The sampling site of Ping River in Sob Rim District. This site was selected in order to study the water condition of Ping River before passing through the Chiang Mai city. Both sides of the banks are predominantly urban and orchards. The grid position of this site is on latitude 18^{o} 52 N and longitude 098^{o} 58 E. The elevation is 360 m. Water body is relatively turbid (Fig. 6).

R2 : Another sampling site of Ping River in Ban Chang Khlan subdistrict, Muang District. It was selected to represent the condition of Ping River after passing through Chiang Mai city and receiving the discharge from Mae Kha canal. Land use of this area consists of orchards and private houses. The grid position is on latitude 18^{o} 43 N and longitude 098^{o} 59 E. The elevation is 330 m. The bank covers with grasses only (Fig. 7).

Irrigation Canal

A concrete canal purposed to transfer water for water supply and agriculture of people living near by. The average width is 5 m but depth and flow rate are vary due to water discharge regulated by the Irrigation Department. The canal is assumed to receive impacts from road running on both side banks and some illegal discharge from hotels and villages near by. There were two sampling sites which are :

IC1 : A concrete irrigation canal near Chiang Mai Phucome Hotel, Cholapratan road, Muang District, located on latitude 18^{o} 48 N and longitude 098^{o} 56 E. The elevation is 340 m. Land use of this area is predominantly urban with heavy traffic (Fig. 8).

IC2 : The same concrete canal at Ton Kwane subdistrict, Muang District, located on latitude 18^{o} 44 N and longitude 098^{o} 56 E. Land use is a mixture of urban and agriculture. This site was chosen to represent the less impact than the previous site. The elevation is 330 m (Fig. 9).

Sewage Canal

Mae Kha canal, a canal started from Ban Tau subdistrict, passing through Chiang Mai city and discharge to Ping River at Ban Pa Dad subdistrict. This canal has been known to represent very high impact due to organic loading from human activities along the canal such as villages, restaurants, markets, hospitals, hotels, schools, temples etc. There were two sampling sites which are :

SC1 : This sampling site situated at small branch of Mae Kha canal near Lanna Hospital, Superhighway , Muang District. The position is on latitude 18^{o} 48 N and longitude 098^{o} 59 E . Both side banks are dense of grasses and bushes. Water body is relatively turbid and contains a lot of black sediment at the bottom (Fig. 10).

SC2 : A sampling site of Mae Kha canal at Pa Dad Subdistrict, Muang District. Water sample was collected at the site before it was drained into Ping River. Water body is quite similar to SC1 but more smelly and has garbage dumping on the banks. The grid position is on latitude 18^{o} 44 N and longitude 098^{o} 58 E. The elevation is 330 m (Fig. 11).

3.2 Sample Analysis

Sample Storage

Sample storage procedures, types of containers and container cleaning procedures were followed the Standard Methods (APHA and AWWA, 1989).

Table 1 : The holding time, type of container, sampling size, and preservation procedures for the sample condition storage.

Parameters	Holding Time	Type of container	Sampling size	Preservation procedures
Acidity Alkalinity hardness BOD_5	24 hours 24 hours 6 hours	plastic	1 l	kept in dark ice box during field trip, then transferred to cold room at 4^oC
Nitrate Ammonia	- -	plastic	1 l	added with 1 ml of conc. Sulfuric acid ,then transferred to cold room at 4^oC
Phosphate	-	glass	120 ml	kept in dark ice box during field trip, then transferred to be frizzed
Heavy Metals	-	plastic	1 l	added with 1 ml of conc. Nitric acid, then kept in cold room at 4^oC

Fig. 2 : Mae Sa stream site 1

Fig.3 : Mae Sa stream site 2

Fig. 4 : Mae Sa stream site 3

Fig.5 : Huay Kaew stream site 4

Fig. 6 : Ping river site 1

Fig. 7 : Ping river site 2

Fig. 8 : irrigation canal site 1

Fig. 9 : Irrigation canal site 2

Fig. 10 : Mae Kha canal site 1

Fig. 11 : Mae Kha canal site 2

Analyses of the samples

The following 15 parameters were measured :

a) Physical parameters : temperature, velocity, and conductivity , pH

b) Chemical parameters : acidity, alkalinity, hardness, biochemical oxygen demand (BOD_5), dissolved oxygen (DO), nitrate-nitrogen, ammonia-nitrogen, total phosphorus , iron, copper, manganese, and zinc.

Physical Parameters

i) Temperature : The water temperature was measured at a depth of about 10 cm using the functional mode of the DO meter (OX1-86).

ii) Velocity : The water velocity was measured using a velocity meter (Current Meter, model 2100-1514, Swoffer Instrument, Inc. USA) Data were recorded in m/1.5 seconds.

iii) Conductivity : Conductivity of water was measured using a conductivity meter (LF-196). The meter must be calibrated before using.

iv) pH : pH was measured using a pH meter (pH-196T). The meter was calibrated with buffers before the field trip.

Chemical Parameters

v) Dissolved oxygen (DO) : DO was measured by two techniques, portable DO meter (OX1-86) and Azide Modification method. The instrument was calibrated before using. DO was recorded in mg/l or ppm.

Dissolved oxygen was converted to percentage of saturated oxygen based on a table of relationship between temperature and saturated oxygen (mg/l) at 760 mm Hg. For example, if we got DO = 7.0 mg/l at 28°C, and from the table at 28°C , oxygen is saturated at 7.75 mg/l.

$$\text{So, \% saturated oxygen} = \frac{100 \times 7.0}{7.75}$$

$$= 90.32\ \%$$

vi) Acidity : It was measured, in term of $CaCO_3$, by titration method using phenolphthalein as an indicator. Acidity was calculated from; (APHA and AWWA, 1989)

$$\text{Acidity (mg as } CaCO_3\text{/l)} = \frac{\text{ml titrant x N of NaOH x 50,000}}{\text{ml sample}}$$

vii) Alkalinity : It was measured, in term of $CaCO_3$, by titration method using Methyl Orange as an indicator (APHA and AWWA, 1989).

$$\text{Alkalinity (mg as } CaCO_3\text{/l)} = \frac{\text{ml titrant x N of } H_2SO_4 \text{ x 50,000}}{\text{ml sample}}$$

FIA procedure was also applied (The Standing Committee of Analysis, 1991).

viii) Nitrate-Nitrogen : Nitrate in water sample was directly measured using nitrate electrode. Before measurement, water samples was adjusted their pHs to 6.5-7.5.

ix) Ammonia-Nitrogen : Ammonia nitrogen in water sample was measured using calorimetric method, Phenate method (APHA and AWWA, 1989). Water samples were adjusted their pHs to 6.5-7.5 before the determination.

x) Total phosphate : Water samples were digested by using nitric-sulfuric acid digestion method to convert phosphate species to ortho-phosphate form. The digests were then analyzed by FIA procedure (Harirutseree, 1994).

xi) Biochemical oxygen demand (BOD_5) : An airtight BOD bottle was filled with water sample and then incubated in a dark room at 20°C for 5 days. BOD_5 was evaluated by the difference between the initial and final DO concentrations.

For the sites which were assumed to have BOD_5 concentration more than 7 mg/l, the water samples were diluted before measurement (APHA and AWWA, 1989).

xii) Heavy metals (iron, copper, manganese and zinc) : For heavy metals determination, water samples were digested by following the total metal digestion method (APHA and AWWA, 1989). The digests were then analyzed using atomic absorption spectrometry (AAS) at their own conditions.

3.3 Quality control of the experiment

Quality control was applied by using certified reference materials, standard addition method or % recovery. A blind sample was also used.

3.4 Data Analysis

Statistical Methods

Statistic Package for Social Science for windows (SPSSWIN) was used for data analysis in this research. SPSSWIN consists of various statistical procedures.

a) Multiple Analysis of Variance (MANOVA)

It is used to compare groups of data containing multiple dependence and independence. In case of Chiang Mai water monitoring, types of water were classified into groups as the following :

Group 1 consists of ST1, ST2, ST3 and ST4

Group 2 consists of R1 and R2

Group 3 consists of IC1 and IC2

Group 4 consists of SC1 and SC2

MANOVA can be used both in multivariate and univariate form. The significant or non-significant differences among groups can be indicated through the probability of F. However, MANOVA can not differentiate one water sample from the others. Therefore, least-significant difference test (LSD) was applied (SPSS^x Inc., 1983).

b) Least significant difference test

In the analysis of LSD, all parameters were involved except those showing non-significant differences in MANOVA. Fact1_1 score, obtained from factor analysis, was used as dependent (SPSS^x Inc., 1983).

c) Hierarchical Cluster Analysis

c/1 Cluster analysis for season test

In this study, CLUSTER was used to differentiate the seasons (dry and wet seasons), on the other hand, group all parameters and arrange to class in order to study the pattern of similarity and dissimilarity. The matrix of months and physico-chemical variables were input (per site). The analysis was done by selecting all parameters as variables and analyzing for cases (months).

c/2 Cluster analysis for determinants selection

The matrix of sites and physico-chemical variables were input. The analysis was done by selecting all parameters as variables and analyzing for variables (parameters).

c/3 Cluster analysis for sites classification

The matrix of sites and physico-chemical variables of May and October were input. The analysis was done by selecting all parameters as variables and analyzing for cases (sites).

For a better interpretation, at least three clustering methods were selected as to confirm the results (SPSS^x Inc., 1983).

d) Factor Analysis

In factor analysis, membership of three clustering groups were involved as variables inputting in variable box. KMO value measurement, varimax rotation were selected. Factor analysis was used to study the relations and correlations of parameters affecting the water quality. The relations are considered strong when the correlation coefficient (r) = 0.7-1.0, and moderate when r = 0.4-0.7. Values $r < 0.4$ indicate weak relations or their absence. In this study, the factor matrix which has the highest percentage variance was considered and the parameters which have r values more than 0.7 were chosen to use as indicators or indices for indexing system (SPSS^x Inc., 1983).

e) Detrended Correspondence Analysis (DECORANA)

The matrix of sites and number of found macroinvertebrates in May and October were input. The DECORANA was done using MVSP. The option of downweighted rare species and detrended method were involved. DECORANA produced both species scores and sample scores and then plotted them onto the two-dimensional graph where similar entities were close together while the dissimilar entities were far apart (MVSP shareware manual guide, 1993).

f) Two-Way Indicator Species Analysis (TWINSPAN)

The matrix of Species and number of species found (family level) was input in fixed form and modified to DECORANA/TWINSPAN form before analyzing. The default cut-level was involved (PCORD manual guide, 1991).

In this study, TWINSPAN was used to classify the study sites on the basis of their macroinvertebrate faunas (family level). It also produces a dichotomous indicator key by which previously unclassified sites can be allocated to the groups on the basis of presence/absence or abundance of indicators taxa.

g) Product-moment correlation coefficient

The product-moment correlation of physico-chemical variables of May and October and the DECORANA axes, was done using MVSP. Pearson correlation and transpose method were involved. Such parameters must be transformed to be approximately normal distribution before analysis (MVSP shareware manual guide, 1993).

h) Multiple Discriminant Analysis (MDA)

In the study, parameters entered to each MDA were selected to minimize multicolinearity, previously identified by correlation and MANOVA. An enter method was undertaken, and the number of Discriminant functions which had a statistically significant input was determined by testing against Wilk's lambda, transformed into Chi^2 ($SPSS^x$ Inc., 1983).

Water Quality Indices (Chemical indices)

Attempts were made to derive a simple and rapid assessment of water quality , water quality indexing system was applied. For the operational process of a water quality index, the following topics were considered :

1. Selection of determinants
2. Rating curves development
3. Weighting of selected determinants
4. The aggregation process.

After the development of process, the proposed model was testified by using different water samples. 16 samples from Pong river were used in order to test the performance of the developed index system by refereeing with the Thai existing surface water quality standard and classification.