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Part I — Distribution and Community Structure of Aquatic Macroinvertebrate Analyses

Part II — 1. The Performance of Biotic Scores and Indices in Assessing Water Pollution: a Case Study in the Pong Catchment of Northeast Thailand

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2. Diversity of Ephemeropteran and Trichopteran Adults

3. Preliminary Keys to Order and to Family of Selected Order of Immature Aquatic Insects of Thailand

By

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การศึกษาความสัมพันธ์ระหว่างสัตว์ไม่มีกระดูกสันหลังหน้าดินในแหล่งน้ำจืดกับปัจจัยคุณภาพสิ่งแวดล้อมในลุ่มน้ำพอง

นฤมล แสงประดับ ธรรยงค์ อินทร์ม่วง จุติมา หาญจวณิช และจุไรวรรณ อินทร์ม่วง

บทคัดย่อ

ผลการศึกษาสัตว์ไม่มีกระดูกสันหลังหน้าดินด้วยวิธีการเก็บตัวอย่างเชิงปริมาณและตัวอย่างน้ำจากลุ่มน้ำพอง 24 สถานี แม่น้ำชี 3 สถานี จำนวนสถานีละ 6 ซ้ำ ทุก 2 เดือน เป็นระยะเวลา 1 ปี พบโครงสร้างชุมชนสัตว์ไม่มีกระดูกสันหลังหน้าดินในลำน้ำพองตอนบนซึ่งมีการปนเปื้อนของสารอินทรีย์น้อยมีความหลากชนิดของสัตว์มากกว่าลำน้ำพองตอนล่างที่มีการปนเปื้อนของสารอินทรีย์มากกว่า พบว่าฤดูกาลมีผลมากต่อชนิดและความหนาแน่นของสัตว์ คือในฤดูฝนจะพบจำนวนชนิดและจำนวนตัวของสัตว์ลดลงอย่างมากเนื่องจากเกิดน้ำหลากพัดพาสัตว์ไปกับกระแสน้ำ

การวิเคราะห์ข้อมูลด้วยสถิติวิเคราะห์หลายตัวแปร พบว่าสามารถจัดจำแนกสถานีต่าง ๆ ได้ด้วยข้อมูลองค์ประกอบทางชีวภาพในแต่ละสถานีนั้น และสามารถแสดงปัจจัยทางเคมีฟิสิกส์ที่มีความสัมพันธ์ต่อคุณภาพน้ำได้ พบตัวอ่อนแมลงชีปะขาวและตัวอ่อนแมลงหนอนปลอกน้ำมากในบริเวณแหล่งน้ำที่มีคุณภาพดี คือมีค่า DO สูง และ BOD ต่ำ พบตัวอ่อนแมลงสองปีก เช่น หนอนแดง (Chironomidae) มากในบริเวณที่มีสภาวะมีการปนเปื้อนของสารอินทรีย์สูง ตัวอ่อนริ้นน้ำกร่อย (*Chaoborus sp.*) พบมากในบริเวณที่มีค่าการนำไฟฟ้าสูง และพบไส้เดือนน้ำจืดซึ่งเป็นสัตว์ที่มีความทนทานมากที่สุดมีปริมาณมากในบริเวณที่มีการปนเปื้อนของสารอินทรีย์สูงมาก การเปรียบเทียบผลการวิเคราะห์ข้อมูลจากตัวอย่างสัตว์ที่เก็บด้วยวิธีเชิงปริมาณและวิธีเชิงคุณภาพ พบว่าการเก็บตัวอย่างด้วยวิธีเชิงปริมาณให้ผลการวิเคราะห์ข้อมูลที่น่าเชื่อถือทางสถิติมากกว่าการเก็บตัวอย่างด้วยวิธีเชิงคุณภาพ และเมื่อเก็บตัวอย่างด้วยวิธีเชิงปริมาณข้อมูลระดับวงศ์ให้ผลการวิเคราะห์เช่นเดียวกับข้อมูลระดับสกุลหรือชนิด

ระบบค่าคะแนน BMWP/ASPT และ/หรือดัชนี EPT และดัชนีความหลากหลาย Shannon-Weiner's index ล้วนเป็นวิธีที่ใช้ได้สำหรับประเมินการเปลี่ยนแปลงคุณภาพของน้ำจากการปนเปื้อนของสารอินทรีย์ แต่เนื่องจากค่าคะแนนระบบ BMWP/ASPT ยังไม่เหมาะสมกับข้อมูลสัตว์ไม่มีกระดูกสันหลังหน้าดินของประเทศไทยนัก ในการศึกษาครั้งนี้ได้พัฒนาดัชนีชีวภาพสำหรับจัดจำแนกคุณภาพแหล่งน้ำจืด 2 แบบ คือ (1) ดัชนีน้ำพอง โดยกำหนดค่าคะแนน 1-10 แก่สัตว์ไม่มีกระดูกสันหลังหน้าดินในระดับวงศ์ตามความทนต่อการเปลี่ยนแปลงสิ่งแวดล้อม โดยใช้ข้อมูลการกระจายและคุณภาพน้ำในลำน้ำพองแต่ละสถานีเป็นหลัก (คะแนนต่ำหมายถึงสัตว์มีความทนทานมากกว่า คะแนนสูงหมายถึงสัตว์มีความทนทานน้อยกว่า) และแบ่งระดับดัชนีในการจัดจำแนกคุณ

ภาพน้ำทางชีวภาพได้ 6 ระดับ และ (2) ดัชนี Q โดยแบ่งสัตว์ออกเป็น 5 กลุ่มตามระดับความทนทานต่อสารอินทรีย์แล้วพิจารณาสัดส่วนของสัตว์แต่ละกลุ่มที่ปรากฏในโครงสร้างของสัตว์ไม่มีกระดูกสันหลังหน้าดินในสถานีหนึ่ง ๆ และสามารถแบ่งระดับคุณภาพแหล่งน้ำได้ 5 ระดับ

นอกจากนี้ยังได้ศึกษาอนุกรมวิธานตัวเต็มวัยของแมลง 3 อันดับ คือ แมลงชีปะขาวพบ 6 วงศ์ 16 ชนิด แมลงสโตนฟลาย 1วงศ์ 1 ชนิด และแมลงหนอนปลอกน้ำ 8 วงศ์ 28 ชนิด ตามลำดับ และได้สร้างรูปวิธานเบื้องต้นระดับอันดับของตัวอ่อนแมลงน้ำ ระดับวงศ์ของตัวอ่อนแมลงชีปะขาว แมลงสโตนฟลาย และแมลงหนอนปลอกน้ำ

A Correlation Study between Freshwater Benthic Macroinvertebrate Fauna and Environmental Quality Factors in Nam Pong Basin Thailand

Narumon Sangpradub, Yanyong Inmuong, Chutima Hanjavanit and Uraiwan Inmuong

Abstract

Six replicates of benthic macroinvertebrate fauna and water chemistry samples were collected by quantitative method at each sampling site along the Pong river (24 sites) and Chi river (3 sites) bimonthly for one year. The results showed that the benthic community structure varies through time and space over the study sites, most benthic taxa were abundant in less impacted upstream waters but declined in downstream water disturbed by heavily organic pollution. The seasonal flooding caused a significant decrease in taxa and the abundance of most benthic groups decreased significantly during the rainy season.

The multivariate analyses were tested and the results revealed that classification of sites based on benthic fauna content and were well related to certain physico-chemical factors of water quality. Certain mayfly and caddisfly larvae were occurring very restricted to good environmental factors (i.e., high DO and low BOD). Dipteran larvae Chironomidae were found in water disturbed with organic pollution and *Chaoborus* sp. occurred in water with high conductivity. Oligochaetes were abundant and tolerated to heavily organic impact pollution. The statistical result of quantitative sampling methods was very significantly higher than the qualitative samples and data at family level provided a same result as generic and species levels.

BMWP/ASPT scores and/or EPT index and diversity indices, Shannon-Weiner's index were significantly correlated to organic water pollution. Due to BMWP/ASPT scores is not fitted to the benthic fauna of Thailand. Therefore, this study develops 2 biotic indices to classify the quality of freshwater as following: (1) Pong index based on family levels of benthic fauna distribution and water quality at each sampling site. The index scores assigned to 1-10 (low scores means strong tolerant groups, high scores means less tolerant groups) and there was 6 classes of biotic groups; (2) Q index based on general patterns of organism tolerance and ratio of presence/absence of organisms in benthic community at sampling sites and this index was classified water quality into 6 categories.

In addition, taxonomical study of adult insects of 3 orders was conducted. The results showed 6 families, 16 species of Order Ephemeroptera, 1 family, 1 species of Order Plecoptera and 8 families, 28 species of Order Trichoptera were recorded, respectively. The preliminary keys to order of aquatic insects and keys to families of Ephemeropteran, Plecopteran and Trichopteran larvae were constructed.

A Research Report on

A Correlation Study between Freshwater Benthic Macroinvertebrate Fauna and Environmental Quality Factors in Nam Pong Basin Thailand

Part I

Distribution and Community Structure of Aquatic Macroinvertebrate Analyses

by

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We also like to extend our grateful thanks to the senior and masters students of Department of Biology, Faculty of Science, Khon Kaen University, in helping us conducting a very hard field works. We are very appreciated to the staff from the Environmental Health Center Region 6, Department of Health, Ministry of Public Health, who also continuously help us in difficult tasks with sampling benthic samples and water chemistry.

Data analysis even with a very rather sophisticated techniques, we like to thank to Dr Peter McQuillan of the Centre for Environmental Studies, University of Tasmania, Australia, the one who simplifies such difficult analyses for us, and guiding us conducting our data analyses.

Many persons out there in the fields aid us finishing our works particularly at the remote countryside, including the staff from the Nam Cheon Conservation Unit, the Royal Thai Forestry Department, we are very thankful to these persons. We also like to thank the villagers at all around the sampling places, they are very pleased to help us handling all of the sampling gears, and kindly participating our sampling operation.

Narumon Sangpradub
Yanyong Inmuong
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Uraiwan Inmuong

14 October 1996

Abstract

The study is aimed to conduct a series of research of Thailand aquatic benthic macroinvertebrate fauna. This study result is the first series of research activities which is primarily indicating what findings about our native macroinvertebrate taxa in the northeast region of Thailand. The faunal abundance, community structure and their relationships to environmental factors both in space and time are firstly analyzed and recorded in Thailand. The Pong catchment of northeast Thailand is selected to be the region investigated. Distribution of benthic taxa at a basin wide scale with bimonthly samplings of 27 established locations is extensively examined.

The aquatic macroinvertebrates are sampled, of shallow waters by the US Standard Surber where at deeper waters the US Standard Ekman Grab is used. Six replicates are applied at each sampling site both with fauna and water chemistry samples. The benthic specimen is identified to possibly at lowest taxonomic level by available keys. All of the discovered species are recorded, and establishing them as a specimen collection at Department of Biology, Khon Kaen University, for later uses. Water physicochemical analyses follow the US Public Health Association standard methods. The univariate analyses are used to generalize the benthic taxa variations. Current multivariate analysis methods are applied, in which the benthic taxa are classified by the polythetic divisive clustering (TWINSPAN) and ordinated by the semi-strong hybrid multidimensional scaling (SSH). The relationships between benthic taxa assemblages and environmental factors use the correlation analyses.

The study explores a certain facts of our native benthic macroinvertebrate species which determined by some environmental variables. All species of Perlidae, some Ephemeroptera and Trichoptera species are occurring very restricted to good environmental quality. The physical factors, for examples, discharge, river dimensions, substrate types and turbidity have major influences on benthic taxa assemblages. The BOD, DO and nitrate, phosphate and dissolved chloride ion are chemically revealed to have a very critical effects on benthic taxa communities. The aquatic benthic taxa are temporally and spatially varying in species occurrence and community patterns. There is a clear distinct difference between benthic species and community structure of the impacted and less impacted sites. The benthic taxa, by using both indicator species and community composition, can be effectively applied in quantifying the degrees of environmental impact at a site through time and space.

The multivariate procedures, the TWINSPAN and SSH, are tested, and the results reveal that they can perform effectively in grouping the sites based on faunal content. Moreover, the reference sites can be evidently created by these two techniques. The grouped sites arranged by the TWINSPAN and SSH are well related to certain environmental factors. The intensity of each environmental factor due determine on the benthic community can be drawn from the results of the TWINSPAN and SSH. Critically, the multivariate procedures show the more pristine sites having more diverse species while the severe impacted sites occupying only minute species numbers. This is viewed that the multivariate procedure will be a very much useful tool in quantifying the degree of "biodiversity".

However, what is found by this study is only at a starting stage in this area of research in Thailand. There are yet to be a series of research needed to be done by this study

as to fulfill much understanding about our native macroinvertebrate taxa, which they have never been comprehensively studies before. It is expected that such the next series of this research would conduct a more detailed biological studies of a particular group of these taxa. Also, the appropriate investigations which may include the studies of biotic indices, qualitative sampling approach and even initiating the rapid bioassessment method. All of those need more supports on both by resource persons and research funds. As the country lacks knowledge, experiences and several available techniques in this area of research, overseas experts are also expected to actively institutionally participate and assist our studies.

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1 An overview

This report is aimed, at this stage, to primarily report of what is found in the first year of benthic macroinvertebrate reconnaissance of northeast region Thailand. This research is in fact being one of the pioneer studies in freshwater fauna research of the country, as currently with a very few studies done in Thailand. The result by this report is expected to contribute significant knowledge principally associated with macroinvertebrate fauna existing of the country, particularly in northeastern region of Thailand. Further, faunal distribution pattern is analysed and any feasible correlations between faunal structure and environmental factors are examined.

Certain pattern correlation from data analysis is anticipated to characterise some forms of species being determined by environmental attributes. Some indicator species of Ephemeroptera, Plecoptera and Trichoptera (EPT) for examples are well understood by overseas nature conservation authorities that these species can reveal degrees of "healthy ecosystems". It is almost certainly that wherever any healthy ecosystems existed, these species are relatively much more abundant. Currently, most European and North American nations they adopted a routine programs assessing their water resources towards this end. In response to such notion, this study then needs to generate particular hypotheses in the following years whether we can assess our healthy environment via our native benthic fauna or *vice versa*. Prior to doing this exercise, our native benthic fauna communities have to be surveyed and recorded. Their correlations associated with environmental variables are likewise fundamentally to be understood.

Even a large numbers of document published by overseas researchers, who studying taxonomically about freshwater benthic fauna and their uses in environmental impact assessment, it is of a very limited published papers found to date within Thailand. Knowledge of benthic fauna taxonomy in Thailand is very limited, and thus this causes a hardship for anyone who likes to study benthic species distribution and their abundance at both local and regional scales. More recently, as this approach is internationally becoming one of common tools used in quantifying biological resources availability within a country, and that to a major extent namely "biodiversity", the studies associated with freshwater benthic animals in Thailand have been increasingly received much attention.

Applying benthic fauna both communities and species methods for biologically assessing water resources as practicing internationally, this would be viewed not easily possible application within Thailand as due to such limited preceding available studies. Not only Thailand which faces difficulty in studying this faunal group but also as to many Asian countries which recently Rundle *et al.* (1993) once concluded that knowledge of freshwater invertebrate fauna in Asia is still very scarce in terms of both community ecology and taxonomy.

Only taxonomy and biology of freshwater mollusc group is at present being well published and available for later study (see more details in Brandt 1974, Upratham *et al* 1995), aquatic insect larva, however, as a major group of benthic living fauna are very rarely documented. Thus, any attempt associated with studying aquatic insects within this country will be very important issue as such that exercise will generate significant knowledge of our native benthic fauna contributing to the country. Of among biological disciplines in Thailand Bimai (1995) reported that aquatic entomology is now at the top priority, and thus the country needs to develop quite a large number of scholars in order to placing them in entomological diversity investigation works.

This study is attempting to investigate distribution of macroinvertebrate fauna and also their taxonomy. Even such approach by this study is rather difficult but with an increasing need for nature conservation in Thailand, this type study is considerably very essential. Most recently Inmuong *et al.* (1996) noted that the biological method by using benthic macroinvertebrate fauna is proved to be an effective tool in assessing water pollution degrees in Thailand. However, what they concluded is from the data collected limited to only one of the waterways and they recommended that a catchment wide study is still needed. Unlike international practices, biological methods other than coliform bacterial counts have never been employed to assess water quality in Thailand. This study may be viewed as the first attempt of the country in investigating whether benthic macroinvertebrate communities would characterise a reliable information relating to various environmental stresses. Further, knowledge of benthic fauna taxonomy and community is anticipated to be initially subsequently achieved by this study, and that their applications to ecosystems and water resource conservations can later be applied.

The term "benthic macroinvertebrate fauna" used here, and as agreement to what stated elsewhere, is referred to aquatic faunal groups consisting mainly of insect larvae, molluscs, crustaceans and aquatic earthworms. All of these fauna some spend all of their life in waters, and others inhabit partly in waters while their adult stage living on terrestrials. These aquatic fauna may be grouped by orders i.e. Ephemeroptera, Plecoptera, Trichoptera, Odonata, Hemiptera, Diptera, Coleoptera, Megaloptera, Lepidoptera, Isopoda, Decapoda, Mollusca and Oligochaeta. Nevertheless, none of these fauna are well understood in Thailand, it is only a few number of papers currently available which relating to Trichopteran adults studied in Chiang Mai Thailand by some investigators (see for examples in Chantaramongkol and Malicky 1989, Malicky and Chantaramongkol 1993). At larval stage, largely there is only a few preliminary studies appeared recently as being the fourth-year student projects and some are master's theses (for examples in Rajchapakdee 1992, Thanee 1994, Panreansaen 1995).

A basin-wide scale study by looking at a distribution of aquatic fauna in Thailand has never been existed so far even current publications revealed that this approach can be applied as one of the major tool in measuring local and regional biodiversity. In addition to this, information from these faunal distributions can be used to assess degrees of biological (environmental) integrity (quality) of the lands, as similar to what are practicing in UK and USA (see for examples in Loeb and Spacies 1994, Bunce 1994). Moreover, benthic macroinvertebrate fauna method is currently included in the standardised methods in assessing water quality impacts by human and natural disturbances in North American and most European countries, or even develop them to be the biotic scores available for ecosystems quality classification, all of these approaches are lately summarized by Rosenberg and Resh (1993).

This study is then tailoring to meet what is lacking in this area of research within the country by firstly attempting to reconnaissance distribution of benthic fauna at a catchment scale. The Pong river catchment locating in northeast Thailand is selected to be the site investigated.

Secondly, state-of-the-art of data analysis techniques are also used to characterise data pattern which later can be applied for interpreting the research results. It should be noted that such available techniques are also worthy to test whether they would probably be working when applying in the Thai environment. It is also expected that the results from these spatial and temporal surveys by this study will considerably contribute the facts of the numbers and species of aquatic fauna inhabiting within this catchment. Also, most importantly, it will be significantly leading to the development of knowledge and understanding of our native aquatic macroinvertebrates.

2 Sampling site description

Twenty-seven sampling sites are established from upstream highlands and downstream lowlands, and all of the sites are locating spreading over the Pong catchment. Fig. 1 shows detailed locations of the sampling sites. All of the sampling sites lie between 16° 00′-17° 15′ N and 101° 15′-103° 15′ E with altitudes range from 600 to 100 M.S.L. Three main rivers are sampled namely the Pong, Cheon and Chi. Upstream sites are H01-H04 which of the Cheon river and P01-P03 of the Pong river, where stations N01-N05 and P04-P05 are locating in the intermediate zone of the Cheon and Pong rivers, respectively. Other sites (P06-P15 and C01-C03) are classified as downstream lowland sites.

Among all sites, the site H01 (Hin Lek Fai) is the only site which having minimal human perturbation as it locates in the Nam Nao National Park while the other sites are received various degrees of impacts through the year from both human and natural disturbances. The lower sites, particularly the sites which locating close to city boundary, are relatively much more disturbed than the upper sites.

The upstream lands are modified mainly for agricultural farms, while the lower downstream sites are of mixed extensively uses for rice and cassava fields, large to small industrial establishments and community residences.

River waters of the catchment are regulated and abstracted for agricultural, industrial and community uses, particularly of along the lower parts of the river length. Channel morphology of both the Pong and Cheon rivers are relatively narrow at upstream highlands with comparatively rapid current, while their channels become much wider at lowland downstream with somewhat slow speed. The river depths are shallower at upstream and relatively deeper at lowland sites.

Vegetation community along the river banks are relatively dense at upstream while downstream zones the riparian flora are growing rather sparsely. Except the site H01, all of the remaining sites the river banks are modified for planting crops and or partly being cruising ramp. At some river stretches, the banks are well protected by vegetation community and thus less soil erosion can be observed, for examples at sites P08 and P09. In contradictory, at upstream sites where the banks are heavily modified, for examples at sites H02 (Huay Chan), H03 (Tad Fa), P01 (Na Noi), P03 (Pa Nokao), surface run-off from the lands to river waters are immense.

The Pong river originates from the seepage spring which locating almost at the center of the Phu Kradueng National Park. The first site of the Pong river P01 locates approximately 30 km. from that origin. The subsequent sites of the Pong river locate all throughout the river length till the last station P15 (Kok Noi) which is 315 km. from the river origin. The Pong river drains southwesterly passing through agricultural lowlands, industrial areas and various communities, i.e. district towns, suburbs and city centers.

The Cheon river originates from the Nam Nao National Park, the first sampling station H01 locates 35 km. from the river origin in which the waters upflowing from underneath the Tham Yai cave. The Cheon river flows southwesterly through extensive corn fields at highlands and large lowland rice fields, and eventually draining into the Pong reservoir.

The water networks of the lower catchment where beyond the Ubolratana Dam forming into only one main channel also namely "the Pong". The Pong river is eventually joined by the Chi river at about the vicinity where locating immediately above the Mahasarakam Dam (see Fig.1).

The flowing waters of the lower Pong catchment are almost entirely regulated and influenced by three Dams operations, the Ubolratana Dam, the Nong Whai Weir and the Mahasarakam Dam. Thus, the fluctuation of the lower Pong water levels are mostly depending on the functions of those three Dams.

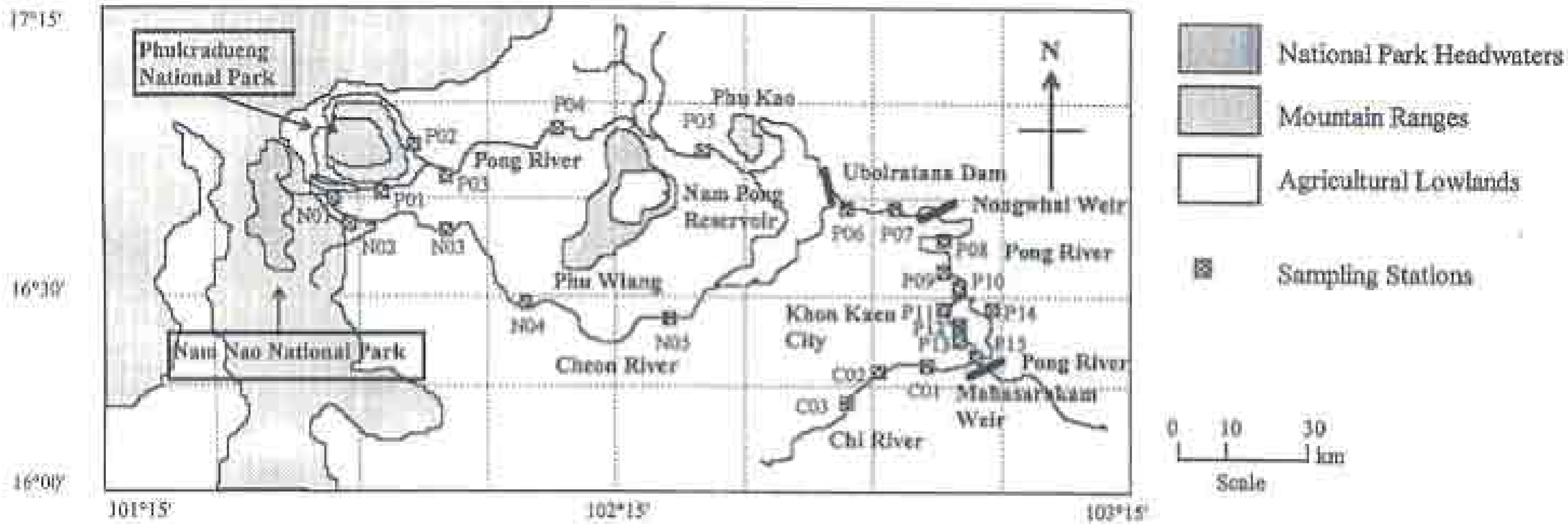

Figure 1 Location map of sampling sites across the Pong catchment.

3 Material and methods

Six replicates (sampling units) are sampled at each sampling sites following the replicate numbers caculation method recommended by Elliot (1978). A certain stretch of river waters at each sampling site with approximately 100 m. length is marked and that six replicates are sampled randomly at this stretch. Sampling both benthic fauna and water chemistry is conducted bimonthly, from October 1995 to August 1996.

Benthic fauna are sampled following the methods recommended by Davies (1994). Sampling equipment for recovering benthic fauna at upstream sites uses the US Standard Surber Sampler with 0.30×0.30 m. opening and 500 µm. mesh size. The US Standard Ekman Grab with 0.15×0.15 m. opening is used to sample benthic animals in deeper waters at downstream sites. The boat is also used to sample animals and water chemistry of that deeper waters. Sampling river waters uses the Van Don Bottle at mid-depth water column where the faunal sampling unit located. River flow-rate is also measured at mid-depth range of the river channel. All of water chemistry analyses follow the standard methods described in APHA (1992).

Benthic samples (fauna plus sediments) recovered are beforehand preserved with 90% ethyl alcohol and kept in polyethylene plastic bags which later brought to the laboratory. Sieving benthic fauna uses the US Standard Sieve with 500 µm. mesh size screen. Benthic samples are then placed on white trays and sorted by eyes using the forceps. The specimens are preserved with 70% ethyl alcohol contained in vials and eventually labeled. All of the specimens are tried to identify them till to possibly at lowest taxonomic level, and further keeping and establishing them as a specimen collections for later references at Department of Biology, Khon Kaen University.

All available keys are used, these are of the North American Keys (Lehmkuhl 1979, McCafferty 1983, Merrit and Cummins 1984), European Keys (Edington and Hildrew 1981, Wallace *et al.* 1990), Chinese Keys (Morse *et al.* 1994), Malayan Keys (Fernando and Cheng 1963) and Thai Keys (available only mollusc) (Brandt 1974, Upratham *et al* 1995). Most of faunal specimens can be identified at best lowest taxonomy to generic level by using these keys.

4 Data analyses

As faunal data are from two capture equipment sources, the Surber and the Grab, thus the average organism density per square meter is calculated prior to applying any statistical analyses.

Data analyses use both univariate and multivariate techniques. All data are statistically examined, and transformation is applied when necessary as to improve normality. Water chemistry and faunal variables are generally described and compared by conventional univariate analyses, both parametric and non-parametric procedures (e.g. ANOVA, regression, Kruskal-Wallis etc.).

All multivariate data analyses use the software PATN (Belbin 1995). The sampling sites are classified using Polythetic Divisive Hierachical Clustering method, namely Two-Way INdicator SPecies ANalysis (TWINSPAN) in PATN, these classification results are based on faunal family composition at a site. Also, all of the sites are ordinated using Semi-Strong Hybrid Multidimensional Scaling Method (SSH).
Significant associations between the TWINSPAN grouped sites and environmental attributes are employed the Kruskal-Wallis test. Spatially and temporally correlation between the SSH axes and environmental variables at a site is analyzed using the Pearson-Product Moment method. The vectors of species (families) and environmental variables are created by the PCC option in PATN. These vectors will be featuring the influences of the attributes (faunal families or environmental variables) relating to the sites' position of the plot.

Initial assumption should be made here is that all of the faunal data used for multivariate analyses are limited to using only family level data (present and absent). However, some forms of analysis results will use faunal data at generic level. The "genus" here also means "species" where the voucher system is established to differentiate all specimens collected. The multivariate data analysis is aimed at seeking for a generation of data structure of macroinvertebrate fauna as which varying by time and space. Such faunal data results created by the multivariate procedures will be later analyzed for any relationships between taxa inhabiting versus environmental variables.

5 Results

5.1 Spatial and temporal variations of macroinvertebrate fauna

Sixty-four families with one hundred thirty species are explored and recorded by this study from five sampling occasions during October 1995 to June 1996. The numbers of families and species are higher at the upper sites and these numbers decrease significantly at downstream sites. Sampling benthic fauna in February 1996 apparently yields comparatively highest average numbers of both faunal species and individuals.

Abundance of macroinvertebrate fauna of all five sampling occasions is markedly varying by time and space. Of all 27 sites investigated, the density of individuals of macroinvertebrate fauna, on the average, ranges from 2334.16 to 4946.91 organism/m^2 which the maximum occurs in February and the minimum in April (Table 1). Fig.2 shows the abundance of macroinvertebrate fauna of all 27 sampling sites.

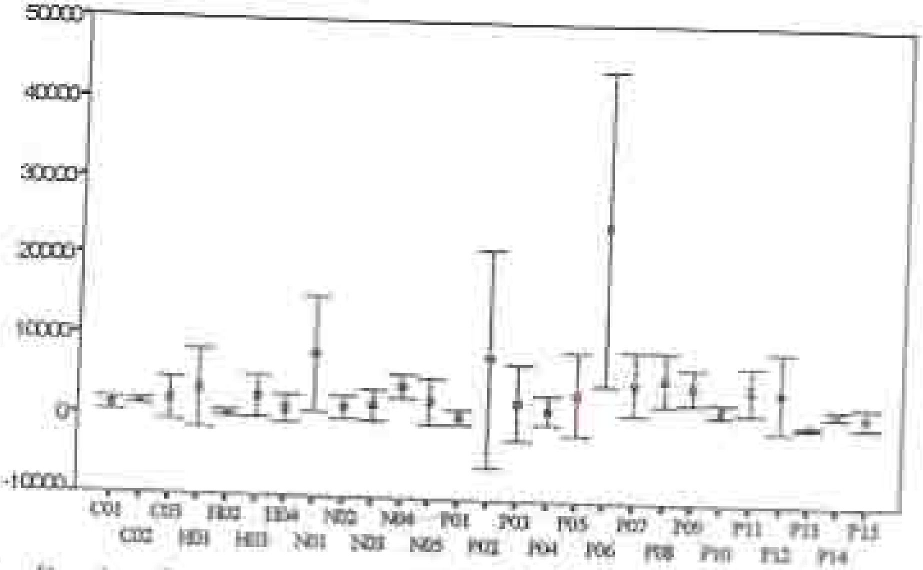

Figure 2 Density of macroinvertebrate organisms from all sampling occasions at all sites of the Pong catchment (±95% Confident Interval).

The minimum organism per m^2 belongs to the site P13 (Pra Kuae) in June 1996, 44.44 organism/m^2 , while the site P06 (Nong Tae) has the maximum density in the same month, 49333.33 organism/ m^2. There is a very much different in density values of individuals when comparing them by bimonthly samplings ($F_{(26,108)}$=4.4167, P<0.001). Table 1 shows more details the density of individuals and species of all five sampling occasions.

Table 1 Bimonthly average individual densities and species numbers by combining all of the sampling sites.

Month	Density of organisms (Mean ± SD)	Number of species (Mean ± SD)	Number of sites (N)
October, 1995	3111.52 ± 6059.26	7.77 ± 3.42	27
December, 1995	4331.68 ± 5879.22	9.70 ± 6.23	27
February, 1996	4946.91 ± 4541.81	10.15 ± 7.18	26
April, 1996	2334.15 ± 2617.66	7.38 ± 4.04	26
June, 1996	4230.86 ± 9297.67	8.29 ± 6.38	27

SD = Standard deviation

All of the faunal abundance measured by density shows very high deviations in values. The most deviated density level is in June sampling occasion (Table 1). This may be largely due to the effects of flooding during monsoon season. While in February, the late cool month, the faunal density indicates less variation. However, as all of the density values appeared in Table 1 are still highly deviations, thus it implies that the faunal density of all sites at each sampling month is very intensively varied.

Species abundance is also very diverse from upstream to downstream locations (Fig. 3). The ANOVA test shows a very significant bimonthly variation of species numbers ($F_{(26,130)}$=8.37, p<0.0001). The maximum species number occurs since ever sampling is 29 species, which is at site H03 in December 1995, and at H04 in June 1996. Of among 29 benthic species inhabited at H03 in December 1995, the major taxa are Trichoptera plus Ephemeroptera species which accounting for 12 species of the total species numbers. The site H04 in June, that similar highest taxa number, 29, also belongs to these groups, but they increase in numbers with each of 8 Trichoptera and Ephemeroptera species has been found.

The site H03 (Tad Fa) locates almost at the top of the south Nam Nao valley, 620 M.S.L., where most of the lands have been cleared for corn fields. Vegetation strips along the stream bank are relatively plenty and thus becoming a buffer zone for aquatic faunal reproduction. Still, patchy of trees can be observed locating sparsely on this land. The water of this site is relatively clear during December and February with comparatively low levels of biochemical oxygen demand (BOD), averaging 1.77 and 4.25 mg/L, and high dissolved oxygen (DO) levels, with the mean 7.8 and 7.65 mg/L. The waters also have rather steady low current speed at this time of the year with approximately 0.16 and 0.00 m/s (non detectable by the flow meter).

The site H03, like other sites, has a certain trend of species numbers fluctuated throughout the year. The species number increases from October with 14 species found till reaching the maximum number 29 species in December 1995 and lastly the species numbers begin to decrease with a minimum of 13 species found in April 1996. The last sampling month in June the number of species found is increased up to 23 species.

Once at a sampling time in December 1995, one organism of the trichopteran goerid *Goera sp.* is explored from six replicates in this site. This is the rare species which is not normally found in lowland areas. The other sensitive species found are of the mayflies groups, these are heptageniid *Heptagenia* sp., (7.41 specimen/m^2) leptophlebiid *Choroterpes* sp. (14.81 specimen/m^2) and two ephemerids *Lituobrancha* sp. (3.70 specimen/m^2) and *Ephemera* sp. (22.22 specimen/m^2)

It should be noted that the above assigned sensitive taxa may be troublesome to southeast Asian tropical environment. Grading sensitivity of those taxa groups are preliminarily following the ranges of tolerant values of each faunal family and the score are given to a group of families, thus finally leading to establishing several similar indices for categorizing these faunal families, for examples, the scores as appeared in Hilsenhoff (1988), BMWP (Spellerberg 1991) and Chessman (1995). Those scores are in fact similar in nature and they appear to be widely accepted by many countries, for instances, in continental Europe, South African countries, some states of U.S.A., (for examples, Maine, Oregon, Ohio, Idaho) and Australasian plus some south pacific countries. However, such a consensus of the scores of among those countries is indeed resulting from the agreement as those countries have extensively studies their native macroinvertebrates, and in particular the ecotoxicology have been tested against each of benthic faunal family.

This study, at this stage, is then considerably likely to lend the above categorized families by which using them for generalizing our macroinvertebrate taxa in relation to environmental factors. Major reason is that our discovery reveals a certain facts of findings the occurrence of those families are principally determined by different environmental qualities, and this is largely following the overseas notes. In other words, the sensitive taxa are frequently found in the pristine areas or at the good environmental quality sites, the more tolerant taxa are normally discovered widespread, and only a certain family found at the severe polluted sites.

We find that most of the scored taxa mentioned above are likely to be applied in our environment, but there seems to be a limitation of using that scores in some taxa. For examples, we are getting into the doubtful score of ephemerid taxa, where these groups are classified as sensitive species in the continental Europe, but rather they do not appear to be categorized as the sensitive species in the some countries, for examples, Australia (see Chessman 1995). By this study, we discover these taxa rather widespread in our catchment, especially in rainy season at some lowland sites. It may be worth to have a further study about this type of taxa whether they are very much sensitive or not such as in the tropical environment.

As mentioned earlier that we often find these taxa group in lowlands especially during flooding period, particularly at when relatively cleaner waters become available but not absolutely good quality as such. This may be the case in raising the point of recategorizion these taxa score into the rather moderate tolerant taxa as in the tropical zone, rather than putting them into the very pristine high quality environment, as appeared in other countries. However, many research needed to be done within tropical countries associated with this issue. What are doing here, it is just a tentative record for our country by lending some available scores from overseas for generalizing our results that we have been discovered.

When considering the variations of macroinvertebrate species across the catchment. A more widespread trichopteran species found from this site H03 to other lowland upstream sites is *Ecnomus* sp. (Ecnomidae) and *Polycentropus* sp. (Polycentropodidae) while Ephemeroptera species are *Ephemera* sp., (Ephemeride) *Baetis* spp. (Baetidae) and *Litobrancha* sp (Ephemeride). Hydropsychidae is another trichopteran family which inhabiting almost everywhere particularly at the site having not very much pollution.

The most abundant organism of trichopteran species at H04 in June 1996 is hydropsychid *Ceratopsyche* sp. with its density 20.37 specimen/m^2. Two well known sensitive trichopteran taxa are also discovered at this site, these are leptocerid, *Triaenodes* sp. (11.11 specimen/m^2) and molannid, *Molanna* sp.(1.85 specimen/m^2). The highest mayflies taxa density occurred at this site is ephemerid *Ephemera* sp., (88.89 specimen/m^2). Apart from ephemerid taxa itself which included in the high sensitive mayflies species, this site also has the fragile sensitive heptageniid, *Heptagenia* sp. (46.30 specimen/m^2).

This site, H04, is just returned from drought, particularly at the very dry period February to April, and the waters are only available about a month or so. Referring to the density values and sensitive taxa above, this site is rather more enhancing microhabitats for the mayflies species reproduction during this time of the year. This site also has very much shading, and the substrate is mainly bed rocks. Subsequently, certain mayflies larvae are greatly abundance.

The most severe impacted site is P13 which had highest BOD (see more details in Appendix 2) since it is greatly affected by organic pollution discharged from Khon Kaen city, it is much obvious that Diptera Chironomidae is the only major taxa dwelling in this site (see Appendix 1). Plafkin *et al.* (1989) documented that Chironomidae is the specific taxa can be used to characterise organic pollution.

The site H02 (Huay Chan) even receives markedly impacts by surface run-offs from nearby corn fields, it is almost always discovering *Sialis* sp. (Sialidae) inhabiting at this site of all sampling occasions. It is worthy to note that all of the three sampling sites where finding these megalopterans are rather locating at high altitude and the weather at these sites is quite cool all year round. The possible hypothesis may probably be raised here is that whether the climate and landscape are far conditioning flourishing this type of taxa rather than the water pollution *per se*.

The occurrences of other faunal groups except caddisflies, mayflies and stoneflies are rather widespread both at highlands and lowlands.. Hemiptera fauna, for examples, *Tenagobia* sp. (Corixidae) and *Rhyacobates* sp. (Gerridae) are abundant at edgewaters in almost all sites.

Odonatan Gomphidae family found mostly elsewhere, Coenagrionidae, is mainly discovered only limited to upstream places

Coleoptera groups especially riffle beetles Elmidae *Stenelmis* sp., *Hexacylloepus* sp. and *Cleptelmis* sp. are also plentiful at upstream sites where scarce at downstream reaches. Coleopteran Gyrinid especially the shiny *Dineutus* sp. and water-penny Psephenidae are only inhabiting at upstream clean waters.

Another rarest aquatic insect taxa is Megaloptera. Two families of Megaloptera are explored here Sialidae alderflies and Corydalidae dobsonflies. Only three sites H02, H03, H04 are found Megaloptera fauna dwelling. Two species are recorded here *Sialis* sp. (Sialidae) and *Neochauliodes* sp. (Corydalidae), the first species is discovered frequently where inhabiting at sites H02 and H03 while the latter species is limited to site H04.

Unlike North American and European researchers, for examples as in Lehmkuhl (1979), who document that the blackflies dipteran Simuliidae larvae are very common and widespread in their waters, by this study, in contrast, this faunal group is found dwelling only at upstream clean water highlands. Only one Simulid species is discovered so far which is *Simulium* sp. This family is inhabiting in clean waters (Hellawell 1978). Common craneflies (Tipulidae) is another dipteran taxa explored where inhabiting only at upstream sites.

Another interesting dipteran taxa phenomenon is that the occurrence of the phantom midge *Chaoborus* sp.(Chaoboridae). This taxa is found being very tolerant to high level of BOD, conductivity and total dissolved solids (TDS). As a high level of both conductivity and TDS in this study is mainly from chloride content (see Appendix 2). In fact, the causes of chloride contamination in water bodies are from both domestic and natural induces.

The sites locating close to the city, for examples, P12 (Lueng Plei), P13 (Pra Kuea) and P14 (Ta Hin) are impacted by community sewage discharges, and these sites also have comparatively high BOD and TSS values.

High chloride levels of other sites, i.e. C01 (Don Bom), C02 (Ta Pra) and C03 (Ton), are influenced by natural cause which mainly resulting from rock weathering, as some parts of the land are geomorphologically bearing by rock salts. Even waters are affected by critical contaminants from human and natural induces, however, the *Chaoborus* sp can dwell tolerantly to high chloride, BOD and TSS contents, and thus being one of among indicator species associating with pollution found by this study.

Of all dipteran taxa, at this stage, it can be summarised that both Chironomidae and Chaoboridae are highly tolerant to gross organic pollution and heavy dissolved solids. Further, it is more interesting that from all of sampling occasions there have never been found any *Chaoborus* sp. existing in all upper catchment sites except N05 (Nong Pai) and P05 (Kok) which are the last upper sites locating immediately above the Ubolratana Dam. More specifically, in June 1996, the most impacted site P13 discovers only *Chaoborus* sp. dwelling.

5.2 Sites comparison

All of the sampling sites can be divided, according to their locations, into the upper and lower catchment sites, the upper sites are H's and N's of the Cheon river and P01-P05 of the Pong river, whereas the remaining sites (P06-P15 and C01-C03) are classified as the lower catchment sites. The statistical t-test shows a significant difference between the faunal density of the upper and lower catchment sites ($t_{(181)}$-5.38, $p<0.001$).

The upper watershed grouped sites have an average 10.94 species occupying whereas on the average only of 6.28 species inhibiting in the lowland sites. When considering the trend of the species deposited at all sites, it is obviously clear that the sites of the upper Cheon river particularly H01, H03 and H04 have far greater numbers of species than of the Pong (P's) and the Chi rivers (C's), Fig.3. Both sites of the Cheon river H03 and H04 have outnumbers of species more than any other sites.

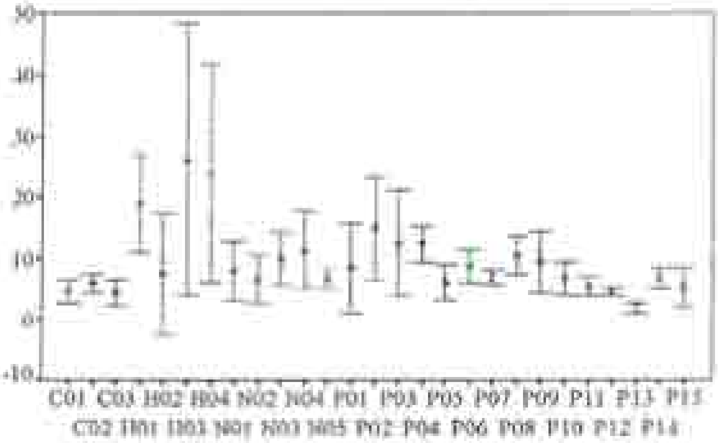

Figure 3 Average number of species abundance of all 27 sampling sites (±95% Confident Interval)

Even the two sites H03 and H04 have comparatively higher numbers of species assemblage but their temporal range of species of these two sites are distinctively large. The site H01, in contrast, has a rather less deviated species numbers, and also the mean species number is also comparatively high. The site H02 even locates in the same plain of the H's sites but this site is severely affected by land modifications for corn fields, thus the number of species appears markedly decreasing. The mean species of H02 is approximately similar to any lowland sites which having extensive impacts from human activities.

The statistical t-test shows no significant difference between the upper Pong river sites (P01-P05) and the Cheon river sites (H01-H04 and N01-N05)($t_{(86)}$ = 1.05, $P>0.05$

and $t_{(66)}$=-0.38, P>0.05). Even the mean species number of the Cheon sites (Mean=15.64) is comparatively higher than the Pong sites (Mean=10.92), but the number of species of the Cheon sites is fairly deviated with its higher standard deviation (SD=12.04) than that of the upper Pong sites (SD=5.88).

The site which having minimal deviation of species number is H01 (Fig.3). This site also reveals a rather consistent species numbers inhabiting through all sampling regimes (see also Appendix 1). The rarest benthic taxa plecopteran family Perlidae is very often found at this site. Two species wished to be recorded here, *Neoperla* sp. and *Phanoperla* sp. These "common stoneflies" are evidently known to occur limited to a high environmental quality. The occurrences of Perlidae taxa are also somewhat restricted to some sites, i.e. P01, P02, P03, P04, H01 and N01 (Wat Thamkayasit), where P04 (Huay Sainang) is the lowermost site which found this taxa inhabiting (see Fig.1 and also Appendix 1).

The Perlidae is found quite limited to a certain times and places of the year. It is also obvious that this taxa likes to dwell in cool and clean waters. Whenever such that environmental quality prevailing, there may be a high probability to find this taxa, and almost certainly that this type of condition will be available at the upper catchment sites. The weather at these upper sites are much cooler than any other sites, and the waters are mostly impacted by natural disturbance.

The most striking maximum number of the perlid taxa caught is at Wat Thamkayasit site in December 1995 when found a total of Perlidae *Neoperla* sp. 77 specimens from all of six replicates combined. The water at this site in December is quite clear and rather steady slowly flowing, and the waters are about just to recover from flooding storms. At P04 in October 1995, one specimen of *Neoperla* sp. is discovered at this site, the weather is relatively cool and again the water column is quite clean. There have never been any Perlidae existing at lower catchment sites as ever samplings.

The upper Cheon river sites (H01-H04) has no significant difference in faunal density when compared to the lower Cheon sites (N01-N05)($t_{(41)}$=-1.99, P>0.05), but the species number of the upper sites is significantly greater than the lower sites ($t_{(41)}$=2.84, P<0.01). The site H01 is relatively pristine conserved and administered by the Nam Nao National Park. The mean number of species in H01 is less than H03 and H04, which the latter two are classified as the National Reserve Lands managed by the Nam Cheon Upstream Conservation Unit. This part of the lands is largely used for growing crops, mainly corns and chili. The sites H01 to H04 are locating at the altitudes ranging from 610 to 780 M.S.L. As the site H01 has less species number than H03 and H04, this may be resulting from the stream orders. Ward (1992) finds that the very pristine sites of the first or second stream orders are often found less numbers of macroinvertebrate species than the sites of the later stream orders. In contradictory to the H's sites, taxa abundance decreases markedly in N's sites due to the fact that the lands have been cleared seasonally for growing crops and having very few percent of forest cover.

There has no significant difference in both density and species numbers between the upper Cheon river sites (H01-H04) and the uppermost Pong sites (P01-P03) ($t_{(17)}$=0.06, P.0.05 and $t_{(31)}$=1.16 P>0.05, respectively). The species composition of the upper Cheon river sites is similar to the sites P01-P03, but it differs from the lower Cheon river sites (N01-N05) as due to their different physical environment, for examples, depth and substrate types (see Appendix 2). Ward (1992) documented that one of the major factors determining the distribution of aquatic insects is the substrate types. The mayflies nymph *Heptagenia* sp., Trichoptera larvae Calamoceratidae, Goeridae, Psychomyiidae, Philopotamidae and Hydroptilidae are normally found in these upper sites. These taxa usually occur in clean water (Hellawell 1978, Ward 1992).

The lower Cheon river sites (N01-N05) and the upper Pong river sites (P01-P05), which locating in similar altitude, have no significant difference in both organism density and species numbers ($t_{(48)}$=1.76, P>0.05 and $t_{(48)}$=-1.23, P>0.05, respectively). However, the taxa composition is quite difference which many ephemeropterans and trichopterans disappear in the lower Cheon river sites. The hypothesis merged here may be that whether physical environmental conditions possibly play more important role than the chemical parameters.

The site P04 and P05, which locate in the intermediate zone of the Pong river, have taxa composition similar to the lower Cheon river sites, and the numbers of individuals in both sites are not different ($t_{(33)}$=1.2, P>0.05 and $t_{(33)}$=-0.22, P>0.05, respectively). They are mainly composed of mayflies nymphs Baetidae and Caenidae, caddisflies larvae Hydropsychidae, Ecnomidae and Polycentropodidae and also riffle beetle Elmidae, in which all of these taxa are widespread in all sampling sites.

The numbers of individuals in Chi river sampling sites are not different from the lower Pong river sites, but the diptera Chaoboridae is more abundant in the Chi river sites.

Among the lower Pong river catchment sites, the site P13 is the most severe impacted site. Bloodworms (Diptera, Chironomidae) and Oligocheate are frequently found in this site, this is featuring its severe pollution condition as confirming to the results stated by many researchers (for examples, Hellawell 1978, Pinder 1986, Plafkin *et al.* 1989, Ward 1992 and Williams and Feltmate 1992).

However the sites P08 (Kood Namsai) and P09 (Bueng Kae) show some improvements of water quality which characterized by the increases of both animal densities and species numbers. The sensitive mayflies *Heptagenia* sp. including other mayflies nymphs and caddisflies larvae, which found in the upper Pong river catchment sites, are also presented in these two sites. Less soil erosion in these sites can be observed, this is due to the existing riparian vegetation stretch along stream bank which acting as a buffer zone preventing the land despoliation. Williams and Feltmate (1992) document that many overseas research confirmed that it is necessary to left a 30 m. of riparian community along the river bank as being a buffer zone to prevent the soil erosion and sediment deposition into the stream, which consequently protecting stream macroinvertebrate community from that disturbance.

The density of macroinvertebrates in the lower Pong river sites (P06-P15) does not differ to the sites P08 and P09 ($t_{(38)}$=-1.43, P>0.05) but the number of fauna is significantly difference ($t_{(38)}$=-2.83, P<0.01). The common mayflies; Baetidae, Caenidae, trichopteran families; Hydropsychidae, Ecnomidae and Polycentropodidae, the riffle beetle Elmidae and the widespread dipteran, Chironomidae and Ceratopogonidae are inhabiting in these sites. Chaoboridae the phantom midges larva is widely distributed in all sites except the site P11. The fauna abundance increase markedly in site P08 and P09 as previously mentioned. The present of sensitive mayfly Heptageniidae and cranefly Tipulidae indicates that the environment of this grouped sites is better than the rest sites of the lower catchment part.

The lower Pong river sites and the Chi river sites (C01 to C03) have no significant difference in both organism density and species number ($t_{(63)}$=1.97, P>0.05 and $t_{(63)}$=1.62, P>0.05, respectively). Both individual and taxa numbers in the lower Pong river sites are more diverse than those of the Chi river.

Regarding the absence of common taxa in some sites, for examples, as clearly appeared in sites H02 and N's, such condition indicated that these sites are very vulnerable. It is also found that the higher levels of suspension solid, soil erosion and chemical disturbances all have critical effect on aquatic insect communities, by reducing species diversity and significantly changing species composition (Williams and Feltmate 1992).

5.3 Indicator taxa

An overview

Indicator species related to environmental pollution is widely applied elsewhere (Rosenberg and Resh 1993). The study discovers some facts about our native indicator taxa that generally agreed to such overseas application. This study shows that the aquatic macroinvertebrates which are possibly initially used as biotic indicators in the Pong catchment are some families of Ephemeroptera, Plecoptera, Trichoptera, Coleoptera, Odonata, Diptera, Oligochaeta and Decapoda. A brief description of what are found about our indicator taxa will be delineated. The occurrence of indicator taxa groups associating with various sites will be described.

Aquatic insects

Mayflies

From the study, sensitive organism of Ephemeroptera is Heptageniidae which is one of the most sensitive aquatic insects (Hellawell 1978). This taxa disappears from highland sites where disturbance occurs. Leptophlebiidae is widely spread in the upper catchment sites, and also in some highland sites. It is absent in the lower catchment sites, and in some highland sites which the lands are extensively modified for agricultural purposes, with a very few riparian strips exist. Among the widespread taxa, the Baetidae is less tolerance than other mayfly nymphs, i.e. Caenidae and Ephemeridae. The Baetidae is categorized as one of the most tolerant mayflies (Hellawell 1978, Brittain 1982).

Stoneflies

Perlidae is the only one family of Plecoptera which is found limited to a cool and clean waters. It appears in the upper catchment sites H01, P01 to P04 and N01, but it is more abundant in the site H01. As the stoneflies like to present in a pristine areas, thus the stoneflies are then used to characterize the degree of environmental quality (Hellawell 1978).

Caddisflies

Trichoptera larvae of families Goeridae, Leptoceridae and Hydroptilidae are mainly found in rocky substrates. They are absent in the disturbed sites, for examples, the sites H02, P01, P05 and the N's sites. The net-spinning caddisflies Hydropsychidae and Polycentropodidae are in contradictory to those three mayflies species in which having quite a large tolerance range to water contamination. In some lowland sites, for examples, at P08, P14 and P15, some Hydropsychidae, Polycentropodidae and Ecnomidae species are explored. These sites are in fact classified as mild to moderate perturbation. Once when the sites are heavily organic impacted, none of these taxa are found. Mackay and Wiggins (1979) and Edington and Hildrew (1981) state that *Hydropsyche* and *Cheumatopsyche* of the family Hydropsychidae are the first caddisflies taxa to reappear when the water quality of the polluted sites improved. These two genera are also the last caddis taxa that will disappear from the polluted sites.

Water beetles

The whiriligig beetle Gyrinidae only inhabits in the upper clean highland sites and disappears from highland sites where disturbances occur. This beetle may be an indicator fauna of the highland. Riffle beetles Elmidae are widely distribution, but they do not inhabit in the severely polluted sites. Whenever the elmids reappear it could indicate that the environmental condition is markedly recovered.

Dragonflies

The clubtail dragonflies Gomphidae are widely distribution, but they do not inhabit in severely polluted sites. Whenever this fauna presents, it may imply the improvement of environmental condition.

True Flies

The cranefly Tipulidae inhabits in less polluted sites, this may show a possibility of using it as indicator species. The bloodworms Chironomidae can tolerate to very severe polluted site (P13). The chironomid can be used as a biotic indicator for organic content increasing (Pinder 1986, Ward 1992, Williams and Feltmate 1992). The bitting midges Ceratopogonidae appears in almost all sites except the poorest oxygenated site (P13). The phantom midge Chaoboridae is found abundant in lower catchment sites especially of the Chi sites. This taxa is found being very high tolerant to high level of BOD, conductivity and TDS. High levels of both conductivity and TDS in this study is mainly from both domestic and natural induces, the chloride content shares the major part of these two values.

Non-aquatic insects

Freshwater earthworms

The freshwater earthworms Oligocheata are found in all sampling sites, and more specifically they are greatly abundant in a very severe polluted site (P13). This fauna can be used as an indicator taxa for polluted aquatic environment.

Freshwater shrimp

The freshwater shrimps *Macrobrachium lanchesteri* has a wide range toleration to water impairment and it is also absent in many glossy polluted sites.

From the finding results of indicator taxa, certain associations with indicator taxa and aquatic environment can be concluded. The most sensitive organisms are Heptageniidae and Gyrinidae, which are only found in very clean (unpolluted) waters. Leptophlebiidae and Psephinidae inhabit in clean waters with less disturbed environment. Baetidae, Caenidae, Ephemeridae, Hydropsychidae, Ecnomidae, Polycentropodidae, Elmidae, Gomphidae, Tipulidae and Palaemonidae present in moderately polluted sites. The appearance abundant numbers of Ceratopogonidae and Chaoboridae shows that these taxa could tolerate to severe water pollution. While the presence of massive number of Chironomidae and Oligochaeta also indicate the grossly heavily organic water pollution.

5.4 Macroinvertebrate fauna variations implicated by multivariate data analyses

Prior to documenting all multivariate data analysis results explored by this study, some important preliminary premises associating with these data analysis methods should be addressed here. These are as followings.

Firstly, the macroinvertebrate faunal data of all sampling sites presented here, as the first year of investigation, use only presence (1) and absence (0) of family level data. Handling data analysis techniques through all multivariate analysis procedures are partly assisted by Dr Peter McQuillan, University of Tasmania, Australia. All of the methods used for these data analyses are conforming to whatever techniques currently applied in community ecology studies as appeared in most update international journals. Indeed, such methods are very much too new to the Thai academicians. Even all of that methods are proved very successful in analyzing biological data of both local and regional fauna and flora distributions at any parts of the world, nevertheless they have never been intensively tested within Thailand.

Secondly, the classification method employed here, the TWINSPAN (Two-Way INdicator SPecies ANalysis), is one of the classification methods originally created by Cornell University in 1979, and this effective method is currently being used by many biological investigators. The dendrograms from TWINSPAN are then drawn and further providing indicator families (species) that conditioning the dichotomy split levels. Not only the grouped sites produced by the TWINSPAN dendrogram are ecologically comparatively profitable, but those indicator species revealed by the TWINSPAN also show *a priori* species classification related to environmental variables.

Thirdly, the Semi-Strong Hybrid Multidimensional Scaling (SSH) which is claimed to be the most effective ordination method to date (Belbin 1995) applying here will reflect all of the sites based on their faunal composition. The ordination results produced by the SSH will then display these data patterns in multidimensional aspect. The plots between the axes will indicate the positions of the sites in the space biplot. The biplots created by the SSH will feature the proximities of the sites based on their similarity (or dissimilarity). For this instance, the paired sites with maximum dissimilarity are separated far apart in the plots, while the more similarity sites are located close to each other. Such proximity of the sites revealed by the SSH biplot will then allow us to group the sites in the biplot.

Fourthly, as those two methods both produce grouping of the sites, in this aspect the grouped sites created by the TWINSPAN can be seeked for their concordancy through a certain axes biplot of SSH. In this manner, the precise separation of the sites grouping can be reconfirmed *vice versa* through the two methods. This is thus leading high robustness of the sites separation which characterising a certain set of sites based on their similar faunal attributes.

Fifthly, the faunal families assemblage at a site of all sampling stations of each sampling month will also be ordinated, and leading to the positions of different families in those SSH biplots which relating to the site space. The vectors can then be drawn in such these biplots where showing the directions of the families relating to the sites' position in the biplot.

Sixthly, correlations between faunal data and physicochemical factors are explored by examining any merging relationships between the ordination axes and those environmental variables. Variations of physicochemical parameters according to the TWINSPAN grouped sites are also able to be delineated.

Lastly, the bimonthly physicochemical data of the sampling sites are ordinated by the SSH and their directions associated with the sites' position in the same plot are presented as the vectors. These vectors radiated via the centroid of the plot will show the directions and influences of various environmental factors relating to the sites' position in that plot.

All of the correlation analyses handling are tested for statistical significant by the Monte-Carlo Randomization, this test method is conducted through the available procedure in the PATN software.

5.5 Bimonthly sites and species classifications and ordinations

October 1995

The first data set is of October sampling month, the sites are classified and grouped by the TWINSPAN into four main grouped sites (Fig.4a). The grouped sites retained as shown in Fig.4a are the grouped sites divided by the TWINSPAN at level 2 which are very much comparatively ecological meaningful. Important indicator families which are used to split the sites grouping are also shown in Fig.4a. Among those indicator families, interestingly, on the positive side (n=16) of the first division indicator families are Oligochaete and Ceratopogonidae. On the other hand, the negative side (n=11) the indicator families are Elmidae, Gomphidae, Caenidae, Palaemonidae and Leptoceridae. Of the first split sites here, following the indicator families, it is more clearer that the sites locating on the negative side are much less impacted and having a relatively more numbers of environmental sensitive families than the positive side.

Table 2 indicates the average physicochemical profiles of the grouped sites A to D. The grouped sites A have comparatively less average values of all physicochemical parameters, this is excluded the DO value where the average DO shows a markedly higher level than the rest grouped sites. When referring to DO and BOD values of the two group sites (A plus B versus C plus D) there is a clear differentiation of BOD values between these two grouped sites, as the TWINSPAN negative end has the mean DO value 6.38 mg/L, whereas the positive end has 4.41 mg/L. The BOD of the negative side is averaging 1.18 mg/L, while the positive direction is 1.6 mg/L.

Table 2 Average values of selected physicochemical varibales in October 1995.

Environmental variables	Grouped sites			
	A	B	C	D
Air temp.(°C)	25.47	31.21	30.56	30.70
Water temp. (°C)	25.33	27.48	29.11	28.20
Velocity (m/sec)	0.50	0.85	0.73	0.43
Width (m)	5.67	29.38	56.67	32.57
Depth (m)	0.43	3.74	7.04	3.51
Discharge (cu.m/sec)	1.10	49.05	212.60	32.50
Alkalinity (mg/L)	77.33	103.50	84.44	89.43
Conductivity (microS/cm)	141.53	199.26	211.69	215.17
TDS (mg/L)	114.50	167.06	155.98	129.94
TSS (mg/L)	159.00	70.39	40.58	74.19
Ortho-P (mg/L)	0.02	0.06	0.05	0.05
Nitrate (mg/L)	0.13	0.19	0.19	0.44
DO (mg/L)	6.73	6.03	4.84	3.97
BOD (mg/L)	1.27	1.09	1.29	1.91

There have been a marked differences of certain environmental values between the TWINSPAN grouped sites (A to D) when testing them by the Kruskal-Wallis. These are river width ($H=11.43$, $P<0.01$), depth ($H=11.02$, $P<0.05$), discharge ($H=16.19$, $P<0.001$) and DO ($H=8.38$, $P<0.05$). This is indeed the major environmental variables that discriminating the TWINSPAN grouped sites. All of the first three variables are increasing from grouped sites A to B, and further to the other end. On the other hand, the DO is reducing from A to D. Such these variation results will later be tested for correlations between the ordination axes and all of the environmental variables. Moreover, if the axes of the ordination plot reveal any conform correlation results to such of the TWINSPAN, then it can be concluded that those certain environmental variables are commonly affirmatively relating to faunal groups of interest.

The second level is splitted into two grouped sites at each end. Baetidae and Leptophlebiidae (-) and Tipulidae (+) are indicator families are used to divide the first group sites (n=11). These families indeed reflect the more subtle environmental changes of among sensitive families. The Baetidae, with only limited to some species in this group, while the others have a large range of toleration, and Leptophlebiidae are very much sensitive to pollution, the Tipulidae, however, is relatively more tolerant when compared to the former groups.

The second level split of the other group (n=16) is divided by indicator families Polycentropodidae (-) and Hydropsychidae (-). Of these less sensitive sites, it can yet make a comparison between these sites, the negative end at this level (n=9) is consisting all of the sites which having family members moderate sensitivity to pollution, these sites also indicate quite a diverse macroinvertebrate fauna inhabiting than of the other end (n=7). This latter site, the major faunal group found is mainly Diptera which is more tolerant to water pollution. Preliminary conclusion according to sites classification by the TWINSPAN in October is that the sites are well successfully classified by the TWINSPAN as grouping them into less impacted to much impaired sites, and these grouped sites can be also simply correspondingly expressed by chemical variables.

Fig 4b shows certain positions of the sites arranged in the space and the most clearest grouping of the sites according to the TWINSPAN is the biplot from the SSH between axis1 and axis3. The sites can be grouped into four major grouped sites (Fig 4b). The most well separation is of the last grouped sites (D) where having relatively much water pollution than the others. Fig 4c also exhibits the families assemblage arrangement in relation to the sites' position (Fig 4b). The occurrence of all families in the biplot with respect to the site-pollution classification are clearly apparent, Fig. 4c. Most cleaner water macroinvertebrate fauna required occur limited to the grouped sites A and B. These are, for examples, Leptophlebiidae (LEP), Hydroptilidae (HYDT), Simuliidae (SIM), Sialidae (SIA) and Perlidae (PER). It is very obvious that these family groups never occur in the grouped sites C and D. This is also in contrast to Oligochaete (OLI) which is most abundant in the grouped site D and never exist in the grouped sites A and B.

Correlations between ordination axes and environmental variables by the Pearson-Product Moment reveal that axis1 negatively correlates to discharge ($r = -0.52$, $t_{(25)} = 3.06$, $P<0.01$) and positively relates to turbidity ($r = 0.49$, $t_{(25)} = 2.85$, $P<0.01$) and TSS ($r = 0.44$, $t_{(25)} = 2.48$, $P<0.05$). Axis3 is greatly significantly negatively related to DO ($r = -0.76$, $t_{(25)} = 5.92$, $P<0.001$) and positively associated with water temperature ($r = 0.53$, $t_{(25)} = 3.14$, $P<0.01$), width ($r = 0.47$, $t_{(25)} = 2.67$, $P<0.05$), depth ($r = 0.51$, $t_{(25)} = 3.01$, $P<0.01$) and discharge ($r = 0.43$, $t_{(25)} = 2.35$, $P<0.05$). There is no relationship between axis2 and any water quality variables.

As the results from Kruskal-Wallis shown earlier that four main environmental variables are contributed to the variation of the TWINSPAN grouped sites, the SSH axes also still reveal a consistent result to TWINSPAN output. It now can be concluded that in October these four environmental variables are primarily conditioning the existence of the faunal groups which dividing by the TWINSPAN analysis.

To be much more clear about the correlation of family-site and environmental variables, this can be shown by grasping two extreme environmental variables, i.e. discharge and DO. The former is highly relating to axis1 ($P<0.01$), this can be seen in Fig 4b that the trend of discharge is increasing from the right to the left hand side of the plot. Fig 4c shows some facts of benthic animals with much susceptible to discharge positioning on the right end of the plot whereas the animals capable of resisting to high discharge are placed on the far left, for examples, the families Polycentropodidae and Ecnomidae. Such this explanation associating with environmental variables and SSH axes would then be applied to interpret the results onwards unless otherwise not mentioned.

The DO level is very much significantly related to axis3 ($P<0.001$). The DO value is apparently reducing from the bottom to the top along axis3 of the plot. Fig 4c reveals a certain fact associating with this DO profile by reflecting the distinctively different benthic families positioning along axis3, the families require high dissolved oxygen, for examples, Leptophlebiidae, Leptoceridae, Heptageniidae and Perlidae, as in Fig. 4c, are clearly placed towards the bottom end of the plot while the less sensitive families to DO level are positioned at the upper part of the plot. This is also showing how the environmental variables are relating to the particular fauna and the TWINSPAN grouped sites.

Fig.4d shows the vectors of environmental variables radiating from the centroid, and relating to the sites' position. The most outstanding features about physicochemical variables which selected to present here are BOD, width, depth and nitrate. These varaibles are pointing towards the impacted sites which having less number of animals deposited. In contrast, the sites with more animal numbers are located in the opposite end of these vectors. The DO, alkalinity and TDS vectors are obviously running across the sites with high numbers of animals assemblage, and they are all locating at upstream. Also, at this month the upstream sites show being influenced by the TSS, turbidity, velocity levels, and thus these variables never affect the downstream sites such as P12, P13 and P15

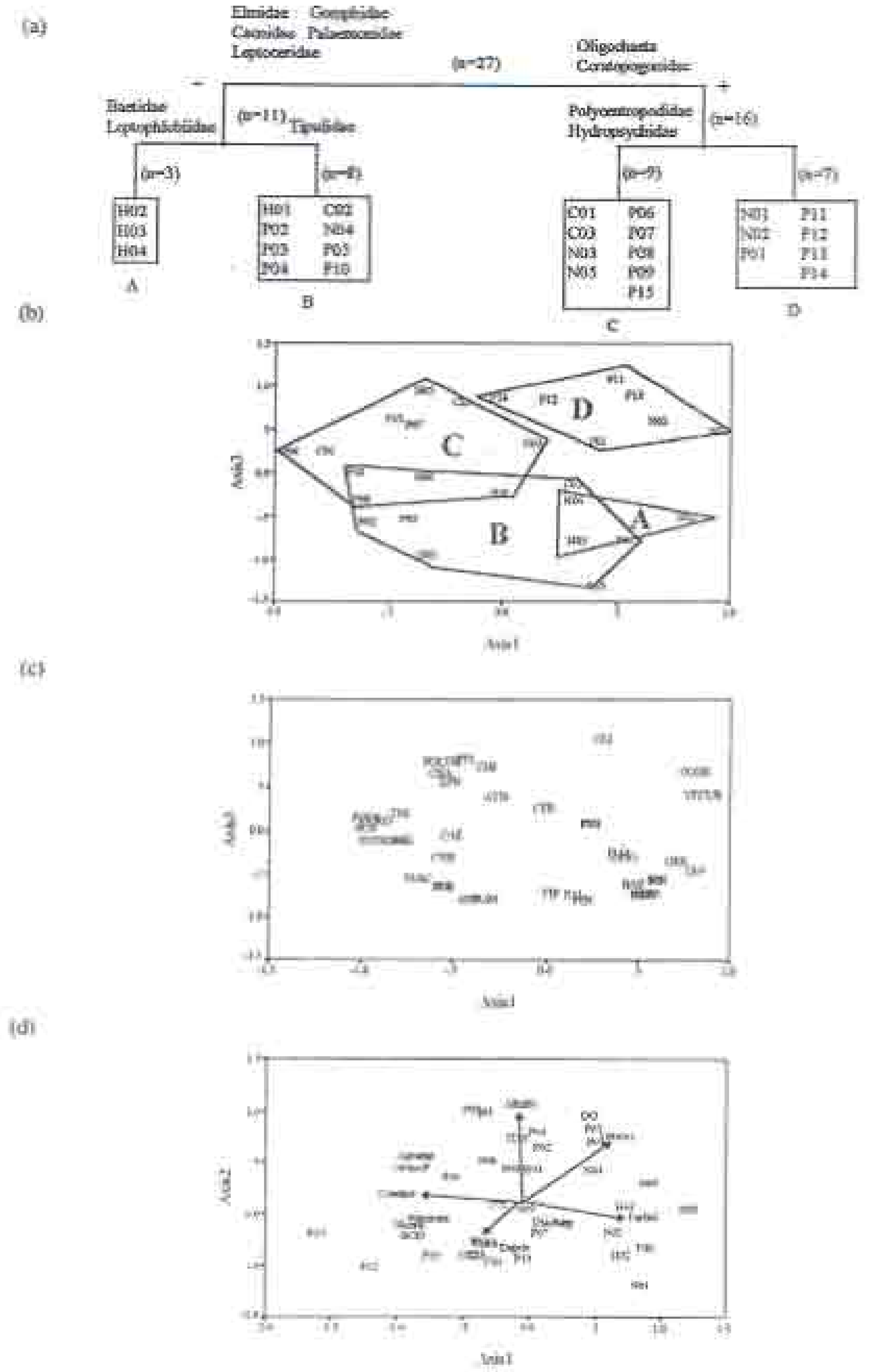

Figure 4 Classification and ordination analyses (October 1995).
(a) Results from TWINSPAN analysis.
(b) Axis1 vs. Axis3 biplot of sampling sites by SSH ordination.
(c) Axis1 vs. Axis3 biplot of macroinvertebrate fauna by SSH ordination.
(d) Axis1 vs. Axis2 biplot of physicochemical variable by SSH ordination.

December1995.

Unlike the October phenomena which features a late rainy condition and that still receiving a dramatic flooding effects, in December the water environment is considerably returning to rather static state. The TWINSPAN and SSH can both successfully classify the sites into five major groups A, B, C, D and E, as shown in Fig 5a and Fig 5b.

The indicator families conditioning the split of the first TWINSPAN level are Baetidae (-), Gomphidae (-), Leptophlebiidae (-) and Tipulidae (-) with no positive side family indication. The second level of the negative side (n=9) is then splited into two main grouped sites A (n=5) and B (n=4) with only family Elmidae (-) is an indicator, Fig 5a. Indicator family on the positive side (n=18) is Chaoboridae (+) and on the negative side is Polycentropodidae which dividing the sites into two grouped sites, C (n=10) and D plus E (n=8) sites. The third level is a split between the sites D (n=5) and E (n=3) which indicator species is Ephemeridae (-).

All of the sites divided by the TWINSPAN at the first level on the negative end are locating at upstream areas, except the site P09. These sites have more macroinvertebrate faunal numbers than other remaining sites. Since the site P09 is included within the uppermost highland sites, this is mainly from its family numbers content. This site has certain indicator families inhabited, i.e. Gomphidae and Elmidae. These families are in fact also used to characterise a relatively less polluted waters.

Table3 shows some distinctive physicochemical values comparing between the grouped sites A to E. The grouped sites A and B, referring to Table3, have less impacts from physical factors, i.e. velocity, depth, discharge, turbidity and TSS, while the remaining grouped sites are notably disturbed by these environmental factors. The average DO and BOD levels of the sites also reveal some facts of chemical variables associating with the TWINSPAN grouped sites, i.e. the grouped sites A and B have distinctively average high DO and low BOD values.

Table 3 Average values of selected physicochemical variables in December1995.

Environmental variables	Grouped sites				
	A	B	C	D	E
Velocity (m/sec)	0.86	0.40	0.22	0.27	0.25
Depth (m)	0.34	0.21	3.05	4.06	3.56
Discharge (cu.m/sec)	3.74	1.31	29.08	52.07	40.57
Alkalinity (mg/L)	160.8	128	135.4	114.4	4.782
Turbidity (NTU)	9.98	9.08	15.08	14.80	14.94
TSS (mg/L)	7.84	5.40	20.80	30.88	25.84
Nitrate (mg/L)	0.08	0.03	0.44	0.12	0.28
DO (mg/L)	8.00	7.96	6.10	7.25	6.67
BOD (mg/L)	2.06	1.78	3.60	2.18	2.89

Seven environmental variables reveal being major contribution to the variation of the TWINSPAN grouped sites. The result from Kruskal-Wallis test portrays these prime variables, they are water temperature ($H=11.49$, $P<0.05$), velocity ($H=10.19$, $P<0.05$), width ($H=12.13$, $P<0.05$), depth ($H=17.63$, $P<0.01$), discharge ($H=12.93$, $P<0.05$), nitrate ($H=11.16$, $P<0.05$) and DO ($H=12.86$, $P<0.05$). These seven environmental parameters are thus accounting for the differentiation of the grouped sites according to the TWINSPAN analysis, Fig.5a.

The best ordination result corresponding to the TWINSPAN groups is revealed by the biplot between axis2 and axis3. Five major groups can be drawn from the plot A, B, C, D and E, Fig.5b. The grouped sites A and B are the sites with less disturbed while the grouped sites C, D and E are rather much impacted.

There are six environmental attribures which relating to axis1, these are conductivity ($r = -0.44$, $t_{(25)} = 2.48$, $P<0.05$), water temperature ($r = -0.45$, $t_{(25)} = 2.53$, $P<0.05$), velocity ($r = 0.58$, $t_{(25)} = 3.54$, $P<0.01$), phosphate ($r = -0.40$, $t_{(25)} = 2.19$, $P<0.05$), nitrate ($r = -0.59$, $t_{(25)} = 3.72$, $P<0.01$) and DO ($r = 0.70$, $t_{(25)} = 4.92$, $P<0.001$). Axis2 is associated only with water temperature ($r = 0.44$, $t_{(25)} = 2.43$, $P<0.05$), and aixs3 is correlated with width ($r = 0.67$, $t_{(25)} = 4.52$, $P<0.001$), depth ($r = 0.78$, $t_{(25)} = 6.28$, $P<0.001$), discharge ($r = 0.59$, $t_{(25)} = 3.58$, $P<0.01$), TSS ($r = 0.53$, $t_{(25)} = 3.11$, $P<0.01$) and water temperature ($r = 0.53$, $t_{(25)} = 3.09$, $P<0.01$).

All of the seven variables used to differentiate the TWINSPAN grouped sites shown above are included in the SSH axes correlations. It is also worthy to note that the associations delineated by the SSH axes are obviously including more numbers of environmental factors than merely the ones which conforming to the TWINSPAN's variables. Thus, the SSH correlations are likely to show more robustness by embracing additional environmental variables other merely from that appearing in the TWINSPAN. In other words, the correlation between the SSH axes and environmental factors thus renders a more efficient detection of environmental changes along the space rather than by the TWINSPAN.

Fig.5c indicates the positions of macroinvertebrate families in the space, their places are indicated in correspondingly to the positions of the sites in Fig.5b. The grouped sites A and B reveal having more numbers of sensitive families than any other grouped sites. The directions of sensitive families of the grouped sites A and B can be apparently seen from the vectors of the plot in Fig.5c, and that most of sensitive families are extended to the bottom of the plot. These families lined to the bottom end are never found in the opposite direction sites, i.e. grouped sites D and E. In contrast, it is also very obvious that families Oligochaete (OLI) and Chaoboridae (CHA), which indicated by the vectors, only exist limited to the grouped site C where these sites are extensively polluted.

Correlations between environmental variables and the sites can be seen from the vectors radiated as shown in Fig.5d. Most of the polluted sites are influenced by high levels of BOD, turbidity, TSS and other physical morphological features of the river, i.e. discharge, width and depth. The grouped sites A and B locate in the opposite direction of these environmental vectors. The alkalinity and DO vectors however line across most of the sites in groups A and B. Such that influences of the alkalinity and DO vectors to the TWINSPAN grouped sites are also similar to the results revealed in October investigating month.

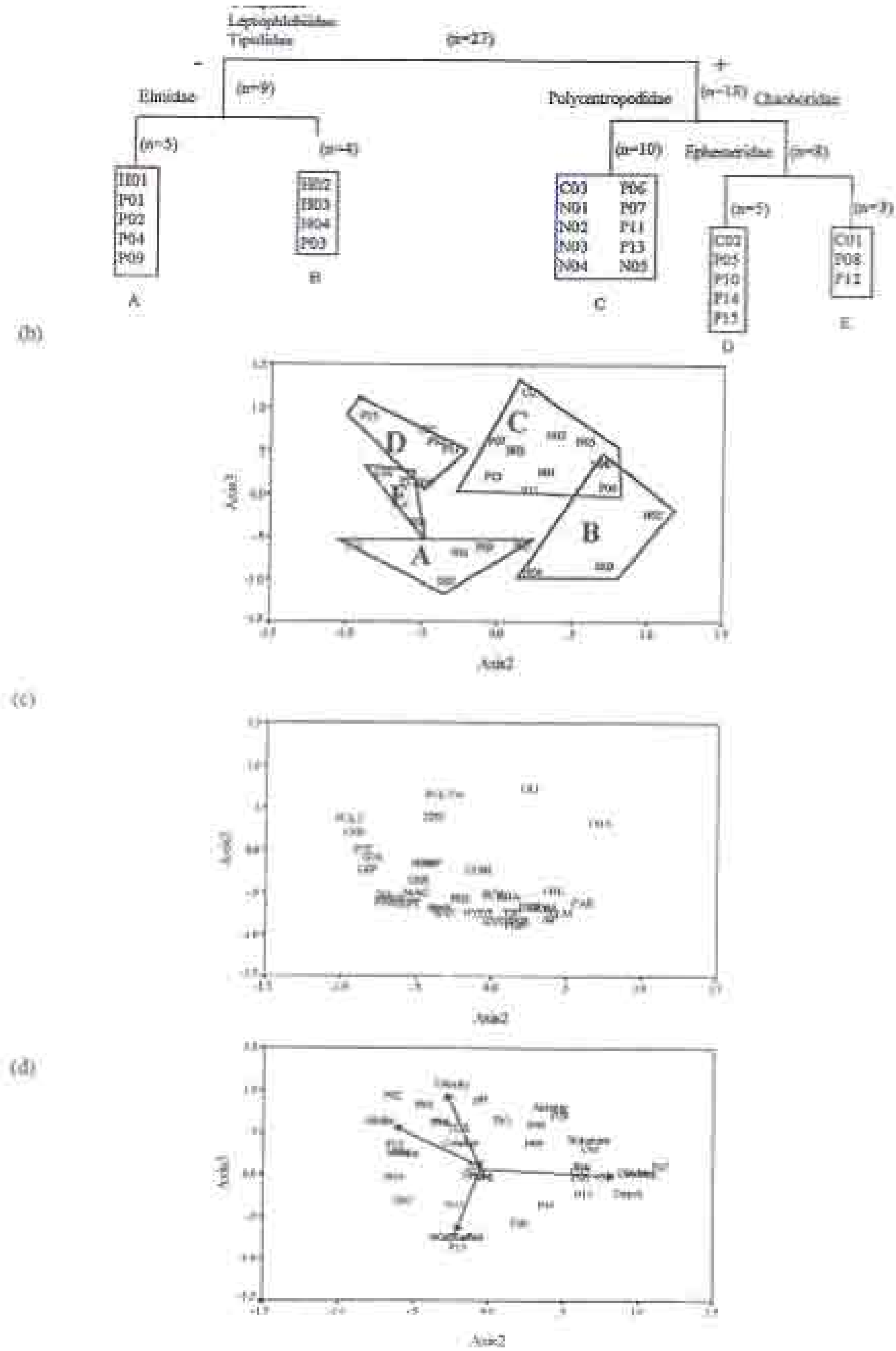

Figure 5 Classification and ordination analyses (December 1995).
(a) Results from TWINSPAN analysis.
(b) Axis2 vs. Axis3 biplot of sampling sites by SSH ordination.
(c) Axis2 vs. Axis3 biplot of macroinvertebrate fauna by SSH ordination.
(d) Axis2 vs. Axis3 biplot of physicochemical variables by SSH ordination.

February 1996

All of the investigated sites are divided into four major groups, A to D, according to the TWINSPAN analysis, Fig.6a. Only two families, i.e. Gomphidae (-) and Baetidae (-) are the indicator families used to split at the first level, with no positive family indicator. At the negative end the sites are splited into two main groups, A (n=5) and B (n=4), which indicator families are Elmidae (-), Psephenidae (-) and Ecnomidae (+).

On the positive direction (n=17), the sites are divided into two levels. The first level consists of two main grouped sites, i.e. C plus D (n=14) and E (n=3). The only indicator family used to split the sites at this first level is Ecnomidae (+).

Most of the sites in groups A and B locate at upstream lands, except two sites, P06 and P08, which locating downstream below the Ubolratana Dam. These two downstream sites at this month, unlike previous months, found having more numbers of faunal families dwelling. Gomphidae, Libellulidae, Ecnomidae, Caenidae and Polycentropodidae are common families discovered inhabiting at these two sites, whereas these families are not explored in any other lower sites.

The grouped sites C and D are rather disturbed sites, where the sites group C, which consisting of the maximum members of 14 sites, are to a larger extent impaired. Ceratopogonidae and Chironomidae families are extensively abundant here. The grouped sites D however is not very much disturbed, the faunal families being tolerant from mild to moderate pollutions can be found at these sites, for examples, Ecnomidae, Caenidae and Gyrinidae.

What is essential to note here is that the site P13 is not included in any specified groups and this site is far well separated from the remaining sites. This site in fact has only pollution preferential family deposited-the Chironomidae. During sampling occasion the eutrophication occurs profoundly at this site which can easily be seen by eyes with very much heavy abundance of unicellular algae *Anacystis* sp.

Table 4 shows some important environmental factors which relating to the TWINSPAN grouped sites. There is a certain differentiation of environmental variables between the grouped sites A plus B and C plus D. The two grouped sites obviously have different levels of temperature, turbidity and TSS. Also, the grouped site A, which containing the maximum numbers of benthic families, has minimal water depth and discharge when compared to the other groups, but this group has an average high value of alkalinity.

The most impacted sites in terms of BOD value is the grouped sites C which has the BOD value, averaging 2.30 mg/L, whereas the other sites have lower BOD levels. At this sampling month, the DO value of the grouped site A is at comparatively highest value, averaging 7.61 mg/L. Similarly, all of the sites in this sampling occasion also reveal high DO values, in which DO of each site is equal or greater than 7.00 mg/L.

The site P13, itself, is at this time, experiencing severe impacts from water pollution. The two nutrients eutrophic factors measured by orthophosphate (P) and nitrate (N) are rising up as highest levels at 1.4 mg/L and 20.0 mg/L, respectively. The BOD level is also reaching at highest value of 9.26 mg/L, this is almost maximum BOD level as compared to any sampling sites investigated in this month. Indeed, it is one of the most severe cases in light of water pollution consideration where this site exhibiting a clear evidence of only discovering the family Chironomidae dwelling with a very few numbers of specimens found (five individuals from six replicates). What is finding here in this case associating with the animals, it is also consistent to what the chemical variables exhibited.

Table 4 Average values of selected physicochemical variables in February 1996.

Environmental variables	Grouped sites			
	A	B	C	D
Water temp. (°C)	21.72	21.66	24.44	24.69
Depth (m)	0.21	2.60	3.77	1.54
Discharge (cu.m/sec)	0.24	52.23	37.01	16.72
Alkalinity (mg/L)	178.00	122.00	127.50	88.00
Turbidity (NTU)	9.03	3.91	19.12	104.26
TSS (mg/L)	6.72	3.55	14.51	29.47

To be more rigorous analysis rather than presenting the general trend as appeared in Table 4, there are five environmental factors being contributed to the discrimination of the TWINSPAN grouped sites. The results shown by the Kruskal-Wallis test are width ($H = 10.59$, $P<0.05$), depth ($H = 12.40$, $P<0.01$), discharge ($H = 11.23$, $P<0.05$), turbidity ($H = 9.24$, $P<0.05$) and phosphate ($H = 8.93$, $P<0.05$). This is apparently indicating that channel morphology is yet having a very much effects on the differentiation of the TWINSPAN grouped sites.

The biplot from the SSH, as in Fig. 6b, is also conforming to the result analysed by the TWINSPAN. A and B, the grouped sites with less disturbed, are also well separated from the grouped sites C and D. Disregarding the site P13, the grouped sites C at which relatively impacted are locating towards the bottom of the plot while the less impaired sites are placed at the upper part.

Significant associations when relating the SHH axes with environmental variables appear statistically only limited to two axes, i.e. axis 1 and axis 3 with no correlation occurred to axis 2. Axis 1 is significantly related to water temperature ($r = -0.58$, $t_{(24)} = 3.53$, $P<0.01$), width ($r = -0.72$, $t_{(24)} = 5.03$, $P<0.001$), depth ($r = -0.79$, $t_{(24)} = 6.20$, $P<0.001$), discharge ($r = -0.72$, $t_{(24)} = 4.92$, $P<0.001$), turbidity ($r = -0.41$, $t_{(24)} = 2.18$, $P<0.05$) and TSS ($r = -0.56$, $t_{(24)} = 3.32$, $P<0.01$). Axis 3 is markedly correlated with only one environmental variable, i.e. depth ($r = 0.43$, $t_{(24)} = 2.35$, $P<0.05$).

The correlation results of the SSH shown above is mostly compromising to the analysis produced by the Kruskal-Wallis of the TWINSPAN. It remains one of the environmental variables, i.e. phosphate, not being included in the SSH associations. This is in fact the phosphate variation between the TWINSPAN grouped sites analysed by the Kruskal-Wallis is rather statistically interval marginal with the H value = 8.92, and thus leading to no statistical significant in the SSH correlation output by the Pearson-Product Moment method. Additional advantage should be noted here is that even the environmental variables cannot be detected by the TWINSPAN grouped sites analysis through Kruskal-Wallis, the SSH results are able to fill this gap by embodying all of significant variables relating to the site-space.

Fig. 6c also indicates the family vectors corresponding to the sites arranged in Fig. 6b. It is clearly that Chaoboridae and Oligochaete have much influences on the grouped sites C which correspondingly shown by the SSH plot in Fig. 6b. Interestingly, these two taxa never exist in the grouped sites A. All of the faunal families occur in the upper part of the plot at where the grouped sites A and B locating, this indicates that the grouped sites A and B are likely relatively being less disturbed.

In contrast to the impacted sites, much more numbers of animals are clearly locating at the upper part of the plot in Fig. 6c. Psychomyiidae, Sialidae, Elmidae, Calamoceratidae, Philopotamidae, for examples, are inhabiting at the less polluted grouped sites A and B. The moderated impaired grouped sites D (N04, P09 and P11) are explored having some families dwelling, for examples, Gyrinidae, Palaemonidae, Caenidae and Ecnomidae.

Fig. 6d shows the vectors drawn from the centroid indicating the effects of environmental variables relating to the sites' position. At this late cooler month, February, dissolved mineral ions measured by conductivity and TDS values show relatively high concentrations at the upstream sites, particularly at P01, P02 and P03. This is mainly due to lower level of river waters. Apart from these mineral salts, alkalinity and to a lesser extent the DO have influential effects on the upper sites which can be seen from the upper left hand of the plot in Fig. 6d.

Still, at the lower sites the discharge and river channel dimensions expense the large effects on those sites which shown at the lower right hand side of the plot, Fig. 6d. This is thus leading to a higher levels of turbidity and TSS as previously shown by the SSH axes-environment correlations.

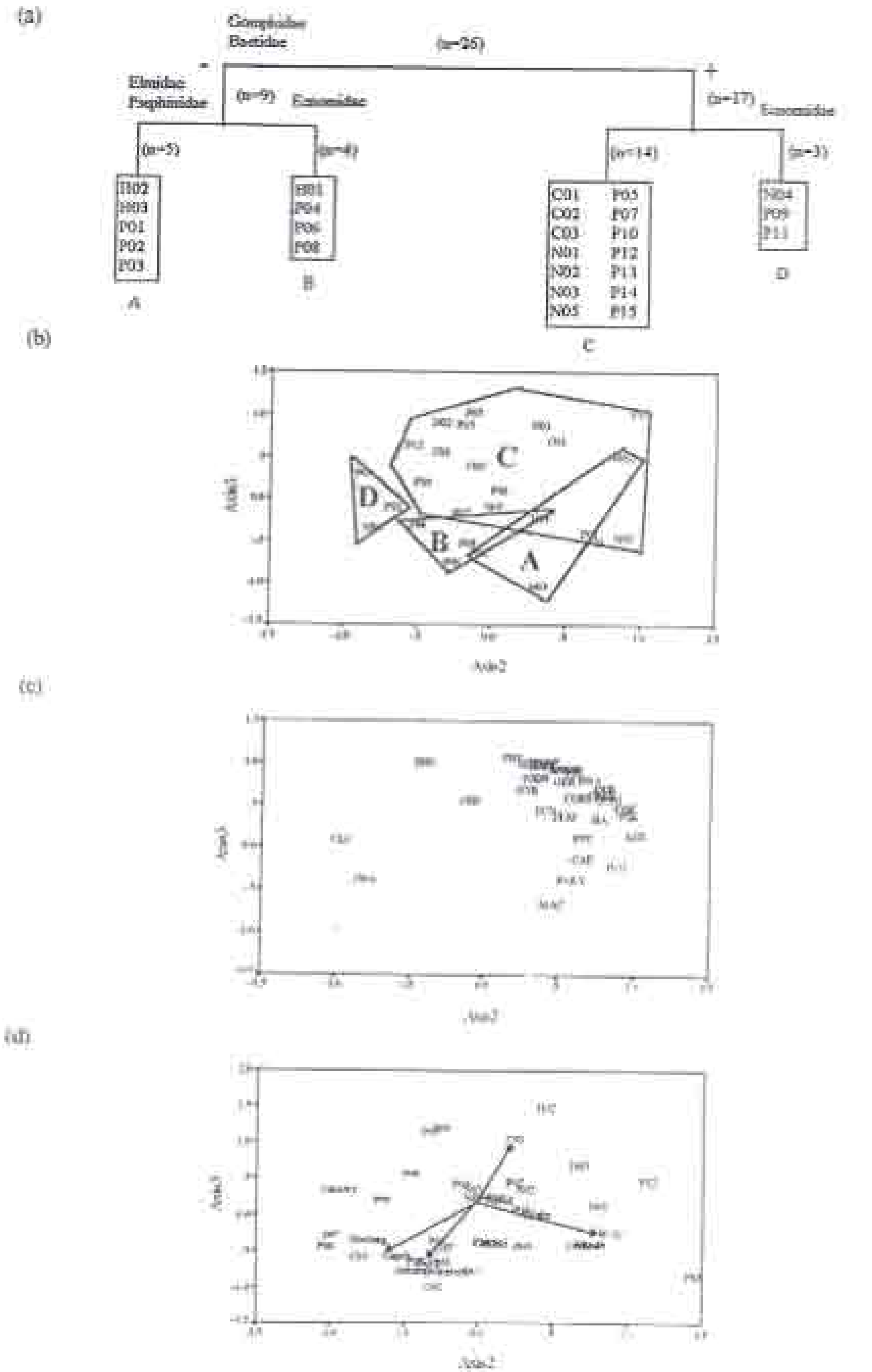

Figure 6 Classification and ordination analyses (February 1996).
(a) Results from TWINSPAN analysis.
(b) Axis2 vs. Axis3 biplot of sampling sites by SSH ordination.
(c) Axis2 vs. Axis3 biplot of macroinvertebrate fauna by SSH ordination.
(d) Axis2 vs. Axis3 biplot of physicochemical variable by SSH ordination.

April 1996

As this month is the hottest month, the TWINSPAN has much ability to classify the sites into more groups, i.e. up to five groups A to F as shown in Fig. 7a. The first level is splitted into two grouped sites A plus B plus C (n=16) and D plus E (n=10) by four indicator families Polycentropodidae (-), Elmidae (-), Gomphidae (-) and Chaoboridae (+). The sites on the negative end are relatively less impaired whereas the other end receives considerable organic pollution impacted.

On the negative side, the second level is further splitted into two main groups A (n=6) and B plus C (n=10) at which the indicator families are Elimidae (-), Ceratopogonidae (+) and Polycentropodidae (+). The third level split is a separation between the grouped sites (n=5) and C (n=5), at this division the only indicator family is Ecnomidae (-).

Of the positive end (n=10), two main groups are created D (n=7) and E (n=3) by which using two indicator families, Ceratopogonidae (-) and Chaoboridae (+). These grouped sites are extensively polluted which are mainly from human activities. There is a very few benthic animals found at these sites. For examples, the sites P15, H02 and C01 all are discovering only two families of fauna inhabiting which are mainly dipterans. The most diverse faunal families found, in contrast, are of the grouped sites A, and to a minor extent of the grouped sites B and C.

Table 5 shows some discrete important environmental trends. Much interestingly, such trends can be seen a very far differentiation of the sites grouped A and the remaining grouped sites. Effects of river channel morphology are still conditioning the upper sites condition as similar to other previous investigating months. The water temperature of the sites grouped A is, on the average, lower than the lower grouped sites. Occasional storms occurring during this month lead to relatively high levels of turbidity and TSS.

However, this situation cannot be referring to organic pollution origin, but rather it is very much involved mainly from siltation caused by natural induce. The grouped sites D and E, on the other hand, even feature low values of turbidity and TSS they both have high BOD levels when compared to the grouped sites A. Alkalinity is also found comparatively reaching at higher levels at the grouped sites A, this is similar to the relative high trends of alkalinity of this grouped sites in previous sampling months.

Table 5 Average values of selected physicochemical variables in April 1996.

Environmental variables	Grouped sites				
	A	B	C	D	E
Water temp. (°C)	27.88	28.80	26.71	30.75	30.41
Velocity (m/sec)	0.46	0.21	0.34	0.12	0.12
Width (m)	9.32	45.20	33.80	40.60	46.67
Depth (m)	0.45	3.31	2.06	4.22	2.91
Discharge (cu.m/sec)	1.80	29.67	15.92	18.20	14.97
Alkalinity (mg/L)	110.67	86.40	79.20	92.80	94.00
Conductivity (microS/cm)	226.27	210.78	175.62	785.61	611.53
TDS (mg/L)	149.87	138.67	117.06	611.01	407.10
Turbidity (NTU)	120.70	30.10	30.04	62.56	96.35
TSS (mg/L)	166.17	26.50	29.40	38.57	53.50
BOD (mg/L)	1.38	1.72	1.34	2.91	2.46

Significant variations of environmental factors across the TWINSPAN groups show four variables which are distinctively different by the Kruskal-Wallis test. Velocity is significantly highly variated ($H = 14.91$, $P<0.01$), the other three diverse factors are width ($H = 11.37$, $P<0.05$), depth ($H = 10.50$, $P<0.01$) and discharge ($H = 11.76$, $P<0.01$).

Even Table 5 reveals a markedly different values of several physicochemical variables, but the variation of within TWINSPAN groups is evidently greater than of the between groups. In this instance, it is thus leading to no significantly statistical variation occurred from among the groups tested. Such this phenomenon occurs in most of the sampling months tested as presented so far, where the general physicochemical trend is varying obviously but the test shows only some values being statistically significantly different, due explanations associated with that case can be made similar to this case by determining the extent to which whether the off-set variation within or between groups which one is greater than the other.

The result from the SSH plot is also clearly exhibiting a well separation of the sites in concordance with the grouped sites shown by the TWINSPAN, Fig. 7b. The grouped sites A, B and C are locating towards the bottom of the plot while the other two groups D and E being extended to the top of the plot. The farthest group, E, with severely impacted is located at the uppermost of the plot. It is essential to note here that as the site H02 is indeed apparently separated from the other sites on the SSH plot, and also it is well splitted from the grouped D at the third level (+) of the TWINSPAN analysis. What logical intention in this aspect is to retain the grouped sites at the second split level of the positive side of the dendrogram, as this for lending them to implying a more ecological meaningful sense.

All three axes are related to environmental factors. Axis 1 is correlated with velocity ($r = 0.55$, $t_{(25)} = 3.23$, $P<0.01$), depth ($r = -0.43$, $t_{(25)} = 2.21$, $P<0.05$), discharge ($r = -0.47$, $t_{(25)} = 2.54$, $P<0.05$), alkalinity ($r = 0.59$, $t_{(25)} = 3.60$, $P<0.01$) and nitrate ($r = 0.64$, $t_{(25)} = 4.212$ $P<0.001$). Axis 2 is associated with velocity ($r = -0.41$, $t_{(25)} = 2.22$, $P<0.05$), width ($r = 0.58$, $t_{(23)} = 3.56$, $P<0.01$), depth ($r = 0.59$, $t_{(23)} = 3.62$, $P<0.01$) and discharge ($r = 0.55$, $t_{(25)} = 3.12$, $P<0.01$). Only two variables are correlated to axis 3, i.e. conductivity ($r = 0.43$, $t_{(25)} = 2.37$, $P<0.05$) and TDS ($r = 0.44$, $t_{(25)} = 2.36$, $P<0.05$). Such these correlations reveal physical environments, for examples, velocity, depth and discharge, still being major factors disturbing the sites.

Families distributed along the sites in the space, Fig. 7b, are very much evident according to their directions radiated. Important families show certain directions extending to the left hand side of the plot, which is very much indeed corresponding to the grouped sites in Fig. 7b. All of the families in that direction are never found existing in any sites of the different ends. These families are, for examples, Elmidae (ELM), Philopotamidae (PHI), Leptophlebiidae (LEPT), and Heptageniidae (HEP). Chaoboridae (CHA) and Chironomidae (CHI), even the former family is discovered inhabiting at almost all sites but its direction is moving towards the grouped sites E, and the Chaoboridae is forwarded to the upper part of the plot where the grouped sites E located.

Of among the sites which considering having moderate impacts, the grouped sites B and C, the Polycentropodidae (POLY), Dytiscidae (DYT), Potamantidae (POT) and Oligochaete (OLI) are the common families showing apparently a distinctive location in relation to these grouped sites.

Regarding the variations of environmental variables across the sites, it is noticeably that the grouped sites A are less influenced by any physicochemical variables. Nevertheless, it is just only the amount of nutrients, phosphate and nitrate, that might be the case as can be seen from the right hand side of the plot, Fig. 7d. This grouped sites have average values of both nutrient variables, 0.38 mg/L and 1.11 mg/L, respectively. It is expected that such high nutrient levels will be resulting from first occasional run-offs diffused from the agricultural lands in this area.

The BOD vector runs across the sites N02, PO9 and moving forward to the directions where the sites P12 and P13 located. This can show the direction of organic pollution occurred across the sites in the space. The sites locating at the upper right hand of the plot also indicate encountering much disturbances from natural cause, which can be seen by the TSS and turbidity vectors lining upon them.

A very intense levels of conductivity is also occurring in this sampling month. The sites C01, C02 and C03 have highest ranges of conductivity from 1218.50 microS/cm to 1690.33 microS/cm. In fact, these sites belong to the Chi river and when the water level decreasing, particularly in April, the high levels of conductivity and TDS would be expected. Of the very high conductivity values prevailing at these sites, only four families found being able to inhabit, these are; Chaoboridae, Ceratopogonidae, Oligochaete and Chironomidae. Chloride from rock weathering is the main cause of this chemical pollutant. It is observed that of among other taxa, Chaoboridae is the one that is much tolerant to high levels of chloride.

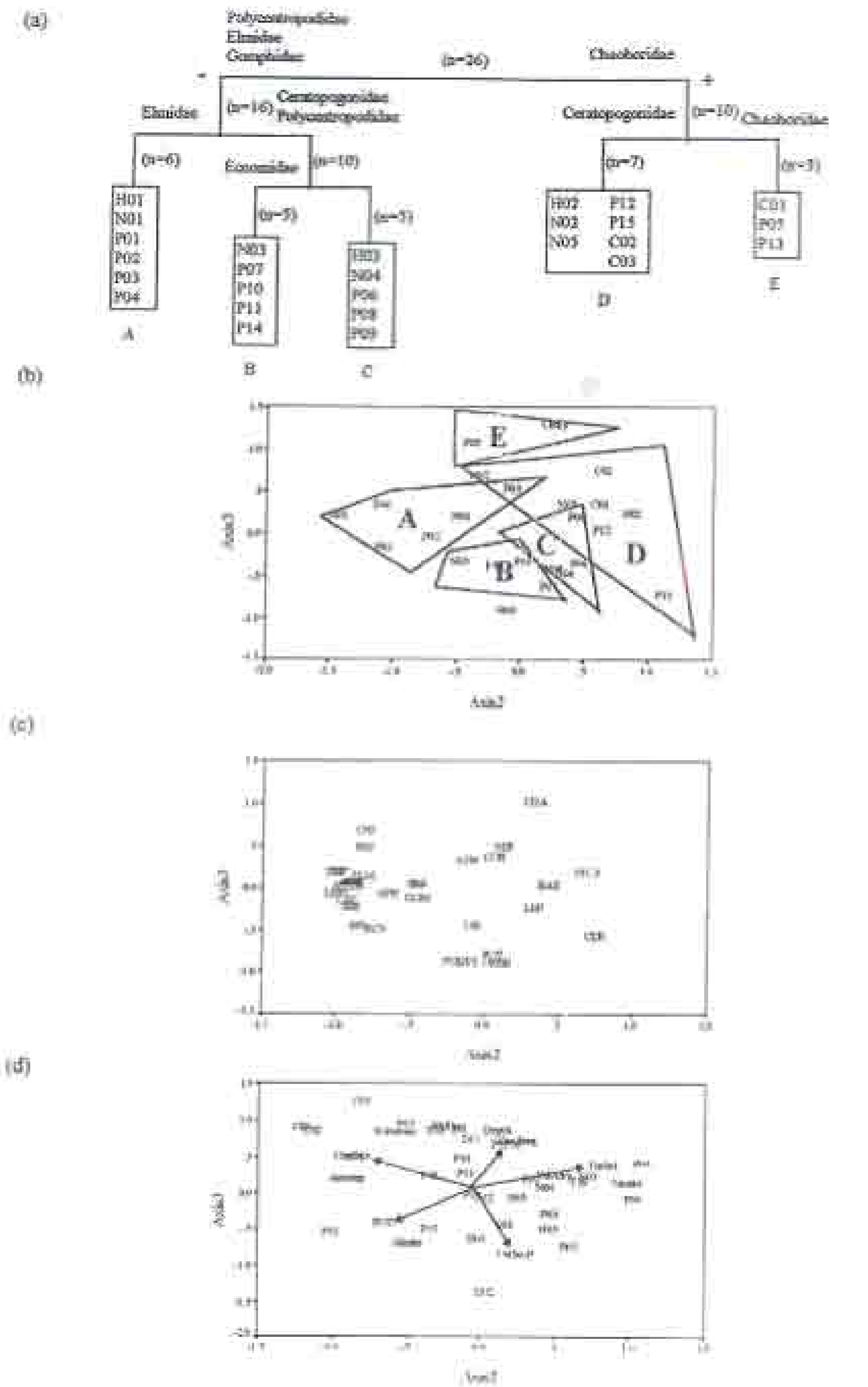

Figure 7 Classification and ordination analyses (April, 1996).
(a) Results from TWINSPAN analysis.
(b) Axis2 vs. Axis3 biplot of sampling sites by SSH ordination.
(c) Axis2 vs. Axis3 biplot of macroinvertebrate fauna by SSH ordination.
(d) Axis2 vs. Axis3 biplot of physicochemical variable by SSH ordination.