CHAPTER 5

DISCUSSIONS

5.1 Diversity of benthic diatoms and the relationship of benthic diatoms with water quality

In this study, a total of two hundred and twenty species of benthic diatoms were found from the Wang River of Thailand. These belonged to 3 classes, 5 subclasses, 12 orders, 25 families and 53 genera. Among these, 42 species were revealed to be a new record for Thailand and belonged to 2 classes, 3 subclasses, 6 orders, 14 families and 19 genera of benthic diatoms. The new record species of Thailand were compared with the checklist of freshwater algae, other publications and relevant books of Thailand (Lewmanomont *et al.*, 1995; Pekthong and Peerapornpisal, 2001; Pekthong, 2002; Kunpradid, 2005; Suphan, 2009; Inthasotti, 2006; Yana and Peerapornpisal, 2009; Leelahakriengkrai *et al*, 2009; Suphan and Peerapornpisal, 2010; Leelahakriengkrai, 2011 and Yana, 2014). The newly recorded species of benthic diatoms for Thailand in this study were found to be higher than in the earlier reports in Thailand because this study collected samples from various water locations both in running and standing water, such as waterfalls, reservoirs, agricultural, irrigation and urban areas. Moreover, from this study, a new species of benthic diatom could be found in the name of *Cymbella* cf. *bifurcumstigma* (under the publishing process). This species was classified in the genus *Cymbella*, which is described as being from the Wang Kaew Waterfall on the upper of the Wang River in Doi Luang National Park, Lampang Province. The valve shape of this species matches that of *Cymbopleura*; however, it has a stigma on the dorsal side of the central area. This stigma is uniquely bifurcated; the valve exterior has one circular opening, but the interior has two elongated openings. In addition, unlike *Cymbopleura*, this diatom has a papilla-type areola occlusion. The morphology of this diatom is documented by light and scanning electron microscopy and its taxonomic status is discussed along with other genera in Cymbellales.

In this study, *Navicula*, *Nitzschia* and *Gomphonema* play a role as the dominant genera, which is similar to that which was reported in Europe and Asia by Tien (2004), Atazadeh *et al*. (2007), Chatháin and Harrington (2008) and Kupe *et al*. (2008) all of whom found that the genera was dominant in the Erh-Jen River (China), the Gharasou River (Iran), the Deel River (Republic of Ireland), and the Töss River (Switzerland), respectively. *Navicula* was found to have the highest number of species (30 species) followed by *Nitzschia* spp. (29 species), *Gomphonema* spp. (15 species), *Sellaphora* (11 species), *Achnanthidium* (11 species), *Surirella* (11 species) and *Cymbella* (10 species), respectively. The most abundant species of the Wang River are *Nitzschia palea, Achnanthidium minutissimum, Seminavis strigosa*, *Achnanthidium exile* and *Planotidium frequentissimum*, respectively.

Moreover, most species found in this study could be considered as cosmopolitan. Nevertheless, some of them are recorded to have abundant for tropical regions (Duong, 2006). *Nitzschia palea* and *Achnanthidium minutissimum* were recorded from many countries in Europe, America, Asia and Africa (Nehar *et al*., 2014). *Planotidium frequentissimum* was also reported to be cosmopolitan often found in the tropical regions according to Krammer and Lange-Bertalot (1986). *Gomphonema parvulum* was regarded as tropical form and with higher frequency in tropical rivers (Benavides, 1996)

In running water, a total of two hundred and thirteen species of benthic diatoms were discovered from ten sites along the mainstream. The most abundant species of the running water was *Nitzschia palea*, *Achnanthidium minutissimum, Seminavis strigosa*, *Achnanthidium exile* and *Planotudium frequentissimum*, respectively. This was identical to that which was published in many previous studies conducted in Thailand and Lao PDR by Pekthong and Peerapornpisal (2001), Yana and Peerapornpisal (2009), Suphan and Peerapornpisal (2010), Leelahakriengkrai and Peerapornpisal (2010), and Yana (2014). These studies found that the highest numbers of those dominant species in the Mae Sa Stream (Chiang Mai Province), the Kok River (Chiang Rai Province), the Huerng River (Loei Province), the Songkram River (Nakhon Phanom Province), the Moon River (Ubonratchatani Province), the Nam Ngum River (Lao PDR), the Ping River (Northern Thailand), the Mekong River (in section of Thailand) and the Yom River, respectively.

According to Jüttner *et al*. (2003) *Nitzschia palea* and *Achnanthidium minutissimum* were regular species in streams and in agricultural catchments of the Kathmandu Valley, Nepal. Similarly, Duong *et al*. (2007) reported on the impact of urban pollution from the Hanoi area on the diatom assemblages collected from the Red, Nhue and Tolich Rivers in Vietnam. These investigators reported that the diatom assemblages at the Tolich site consisted mainly of *Nitzschia palea*.

In the standing water, a total of one hundred and nineteen species of benthic diatoms were found from 2 sites including the Kiew Lom Dam and the Kiew Kor Ma Dam. The most abundant species in the standing water of the Wang River were *Achnanthidium minutissimum*, *Achnanthidium exile*, *Kobayasiella* sp.1, *Aulacoseira granulata*, respectively. This was dentical to that which was published in many previous studied conducted in Thailand by Peerapornpisal (1996), Hunpongkittikul (2003), Malaiwan (2010) From this study, the penate diatoms presented as the dominant species in standing water, which was similar to that which was reported by Chung *et al*. (1992), Lee and Kim (1996), Poulíčková *et al*. (2002), Lee and Yoon (2003) and Novais (2011). These studies found that the pennate diatoms dominated the centric diatoms in terms of the number of species observed and it could probably be assumed that this was due to the presence of the raphe structure of pennate diatoms (Lee and Yoon, 2003).

Normally, the Wang River area can be divided into 3 categories, including the upper, middle and lower parts. The upper part is the upstream and highland area, the middle part is made up of irrigation and agricultural areas and the lower part is made up of urban, agricultural and downstream areas. Each area had different distribution ratios of benthic diatoms because their spreading pattern responded to several of different factors, ranging from geographical to geochemical and human influences (Potapova and Charles, 2007).

At the upper part, in the upstream sites of this study, which had clean water quality, a high abundance of *Encyonopsis microcephala*, *Navicula suprinii*, *Navicula heimansioides*, *Navicula leistikowii*, *Achnanthidium minutissimum*, *Diploneis oblongella* and *Encyonopsis leei* were found, which was similar to the report by Besch *et al*. (1972) Chen and Wu (1999), Wan Maznah and Mansor (2002), Lobo *et al*. (2004), Sahun (2004), Stenger-Kovács *et al.* (2007), Wang *et al*. (2010), Leelahakriengkrai (2011) and Martin and Fernandez (2012). It appeared that the species in clean water quality could be used to indicate the oligotrophic species. In addition, at the Wang Kaew Waterfall, the upstream site, which was located on high slopes and at high altitude areas, induced greater current velocity and lower temperatures, which affected the dominant diatom species of this site. This was different from the other sites. Some diatoms species were found only at this site, such as *Cymbella* cf. *bifurcumstigma*, *Delicata* cf. *spartistriata*, *Encyonema malaysianum*, *Gomphonema pala* and *Encyonopsis leei*. The species of this site was in accordance with Jüttner *et al*. (2010a,b), who found the cymbelloid taxa, such as *Cymbopleura*, *Cymbella*, *Encyonema* and *Encyonopsis* in high altitude freshwater areas. Nevertheless, the gradient and temperature readings of the fast-flowing rivers of mountainous areas, which also correlated strongly with the latitude and altitude, explained most of the variations in the benthic diatom assemblages (Moore, 1977a, b; Vannote *et al*., 1980). Moreover, current velocity plays a significant role in affecting the heterogeneity and diversity pattern of the benthic diatom distribution (Stevenson *et al.*, 1997; Soininen 2004), while current increase drags on cells and affects immigration and export rates (Mc Intire, 1966; Peterson and Stevenson, 1989). In addition, some diatom species form the stalk attached to the surface while some species form short stalks, or mucilage pads that are horizontally positioned species, such as *Gomphonema* and *Cymbella* (Korte and Blinn, 1983; Ghosh and Gaur, 1988).

Along the middle part, most of the area includes the reservoir and irrigation area, and the water was settled in moderate water quality. The diatoms species that showed high abundance were *Achnanthidium exile, Branchysira neoexilis, Cymbella affinis, Gomphonema pumilum, Gomphonema auritum, Nitzschia recta, Rhopalodia musculus, Aulacoseira granulata, Cyclotella meneghiniana, Discostella stelligeroides* and *Ulnaria lanceolata*, which was similar to that which was reported by Chen and Wu (1999), Lobo *et al.* (2004), Sahun (2004), Stenger-Kovács *et al*. (2007) and Wang *et al.* (2010). The slower flowing water also could be the main factor that makes these diatom species different from other habitats. In dams or reservoirs, the very slow water current is usually present in accordance with that which was reported by Patrick (1997) and which found that some species of diatoms such as *Cyclotella*, *Discostella*, *Gomphonema* and *Fragilaria* were present in lakes and slower flowing bodies of water. Moreover, the shape and size of the diatoms also have a relationship with the water flow. Diatoms of a shorter and wider shape can grow well in slower moving bodies of water (Patrick, 1948; Soininen 2004)

At downstream sites, a high abundance of *Nitzschia palea, Gomphonema parvulum, Cocconeis placentula, Cymbella turgidula, Hippodonta pseudoacceptata, Navicula simulata,* and *Planotidium frequentissimum* were found, especially as the dominant species in urban area sites and this was related with moderate to polluted water quality, which was similarly reported by Palmer (1970), Whitmore (1989), Gomez (1998), Güttinger and Straub (1998), Pekthong, 2002, Jüttner *et al.* (2003), Kunpradid (2005), Stenger-Kovács *et al.* (2007), Duong *et al.* (2007) and García *et al.* (2008), Suphan, (2009), Leelahakriengkrai, (2011), Peeraporipisal (2013) and Yana (2014). They had all reported on the species present in moderate to polluted water quality, which indicated its tolerance to organic pollution. Moreover, in the urban or municipal area which were highly affected by human households resulting in the water quality at this location of the river being high in nutrient concentrations. This was related to the findings of Palmer (1980), Potapova and Charles (2004), Leira and Sabater (2005) who reported that these species were nutrient tolerant taxa and could be found in the area that was associated with nutrient enrichment. This was especially true for *Nitzschia palea*, which was usually used to indicate phosphate enrichment or organically polluted water (Force and Grafe, 2002; Pekthong, 2002; Kunpradid, 2005; Suphan, 2009; Leelahakriengkrai, 2011; Peeraporipisal, 2013; Yana, 2014).

However, from the results of this study, there were 102 diatoms species which were found to be present at the mainstream sites but were absent in reservoirs, such as *Fragilaria capucina, Eunotia curvata, Cymbella subleptoceros, Cymbella turgidula* , *Delicata delicatula*, *Encyonema malaysianum*, *Placoneis exigua*, *Gomphonema affine*, *Gomphonema minutum*, *Gomphonema obscurum*, *Gomphonema truncatum*, *Gomphonema productum*, *Reimeria uniseriata*, *Achnanthes inflata*, *Achnanthes pusilla*, *Achnanthidium jackii*, *Luticola terminata*, *Neidium dubium*, *Neidium gracile*, *Sellaphora obesa*, *Sellaphora pupula, Pinnularia biceps, Diploneis oculata, Caloneis bacillum, Hippodonta avittata*, and *Hippodonta pseudoacceptata* etc. On the contrary, there were 8 species of diatoms found in the reservoirs but that were absent at mainstream sites, such as *Placoneis elegans, Gomphonema bohemicum, Achnanthidium* sp.2*, Neidium affine, Sellaphora seminulum, Craticula ambigua, Craticula vixnegligenda* and *Epithemia cistula*. The main factor that results in differences in the diatom species between running and standing water is the flow or the velocity of the water current (Potapova and Charles, 2002). The changes in the flow conditions were found to induce responses in both the composition and physical structure of the benthic diatom communities (Biggs and Lowe, 1998; Passy, 2002), while the cells in the slow-current community tended to grow upright (Lamp and Lowe, 1991). Most species that grow in fast water are elongated and attached to the surface of stones or other plants and some diatoms stand erect on the stalk and are composed of mucilaginous material. However, diatoms thriving in slow currents tend to have shorter, bigger and thick plumose cell growth (Patrick, 1948). Moreover, each type of substratum, including rocks, mud, sand and silt, affects benthic behavior (Panha, 1998). The substrate type was related to current velocity and volume of water (Peerapornpisal, 2002).

5.2 Water Quality of the Wang River

The water quality investigation in the Wang River from October 2011 to September 2012 found that most of the sampling sites showed different trophic status during each season due to the fact that different activities occurred at different locations along the river. At the upstream sites, specifically the Wang Kaew Waterfall (site 1), there was usually oligotrophic water status, which indicated an undisturbed area, as it is located on Doi Luang National Park. This is the sampling site that revealed the lowest impact because of the reduced level of human activities. At the upstream section of the Wang River, low nutrient concentrations and low BOD levels were found. This was explained by Goldman and Horn (1983) who stated that it was a characteristic of upstream rivers having low nutrient levels. Nevertheless, this site showed high alkalinity due to the fact that this area is located in the limestone mountainous area. However, most sampling sites in the Wang River had higher alkalinity and could induce the conductivity in this river to a higher value as a result of carbonate fragmentation to bi-carbonate ions. These geological properties influenced the water resource in this area resulting in a high concentration of calcium carbonate (Wetzel and Gopal, 1999). In accordance with Crottava (2003), it was reported that if waters flow through limestone regions or bedrock containing carbonates, the water generally displays high alkalinity and conductivity. In this study, *Encyonopsis microcephala* was observed as a dominant species in site 1 which mostly was classified in the oligotrophic status, but the value showed a high concentration of alkalinity. The results were in accordance with those observed from the oligotrophic Alpine lake (Hustedt, 1930) and in the unpolluted streams and lakes of the Pyrences (Besch *et al.* 1972) where it was reported that this taxon are the main species of these areas. According to Krammer (1997), this species is composed of several ecotypes with different ecological requirements: in Northern Europe, it lives in oligotrophic waters with low electrolyte content, whereas, in Central Europe and North America it is frequent in lime-rich waters with middle electrolyte content (Gomà *et al*. 2005).

Conversely, alkalinity and conductivity values were slightly dropped at the irrigation sites, which were located along the middle part of the river due to the presence of suspended matter being trapped in the reservoir by the deposits from the water column to the bottom of the reservoir (Garnier *et al*., 1999; Harrison *et al*., 2009; Molisani *et al*., 2010). Although, the alkalinity and conductivity of the irrigation sites had trended to be lower than at other sites but most of sampling sites in this area showed the mesotrophic status and was affected by low concentrations of dissolve oxygen in the water, which normally occur less than in standing water that is influenced by the lower level of water flowing and lower water velocity rates (Smith, 1990).

Whereas the downstream areas were more contaminated by the waste from communities and agricultural activities, especially at sites 8 and 9 along the upper part of the downstream area, which was impacted by the urban area and revealed the meso-eutrophic status and showed high concentrations of ammonium-nitrogen and BOD. Most concentrations of these contaminants were released in the form of sewage from the local households, which were centered in this area. And this was similar to that which had been formerly reported by Neal *et al*., (2000). However, at sites 10, 11 and 12, along the lower part of the downstream area, the mesotrophic status was recorded. These areas were affected by the agricultural activity which showed high concentrations in nitrate-nitrogen and soluble reactive phosphorus. These sampling sites were mostly surrounded by agriculture crops that usually used fertilizers inducing the higher contamination of SRP and nitrate-nitrogen on these areas which was similar to the findings of Nakagaki *et al*. (2005) who reported that the nitrogen and phosphorus compounds are more easily applied in fertilizers. They can attach to the soil particles and more easily be transported through coarse-grained soil to the water by erosion.

As mentioned above, the water quality of the Wang River was mainly affected by agriculture, irrigation, household activities from the municipality and geological characteristics along the area. These activities directly affect the water quality at each sampling site. It was found that the water quality of this river was classified in the oligo-mesotrophic status to meso-eutrophic status. The upstream area held the waterfall site and was slightly affected from human activities and was classified into the oligo-mesotrophic status or as having clean-moderate water quality. The middle part of the river was comprised of reservoirs and the irrigation area. This area was more affected than the upstream area by the influence of the water flow from the reservoirs and some areas were affected by the agricultural activities, resulting in this area being classified in the oligo-mesotrophic to mesotrophic status or as clean-moderate to moderate water quality. The upper part of this downstream area was affected by the urban and municipal communities. The water in this area was classified in the mesotrophic to meso-eutrophic status or as having moderate to moderate-polluted water quality. Finally, the lower part of the downstream area was affected by the small community and agricultural activities. The water was classified in the mesotrophic status or as of moderate water quality. However, the overall water quality in the Wang River was classified in oligo-mesotrophic to mesotrophic status or clean-moderate to moderate water quality and this was similar to the reports by Petipong (2004) and was based on the standard surface water quality of Thailand in Notification of the National Environment Board, No. 8 (1994). The water quality of the Wang River was classified into class 3 and recommendations about beneficial usage of the water were made for this river. The water was deemed appropriate to be used for agriculture, industrial communication and consumption, but the water had to undergo special treatment before it was ready for consumption.

5.3 The establishment of Wang Diatom Index

The establishment of the Wang Diatom Index was done by the indicator value and weighted averages approach (WAs). A total of 100 species of benthic diatoms was found as having high relative abundance and were used to evaluate the index. The researchers in this study selected the species with a high relative abundance (>1%) at each site, which followed the reports of Kelly and Whitton (1995) and Kelly (2000). In that study, 23 species were found in the other Thailand index with no more differences found in indicator values. The ranges of indicator values were between 2.5-4.4, and 33 diatoms species in this study were found in the other Thailand index. The comparison among them was slightly different. (Pekthong, 1998; Kunpradid, 2005; Suphan, 2009; Leelahakriengkrai, 2011; Yana, 2014). Diatoms species of *Gomphonema lanceolatum, Geissleria decussis, Gomphonema pseudoaugur, Luticola* sp1*, Nitzschia gracilis, Nitzschia palea, Nitzschia persuadens, Nitzschia reversa* and *Nitzschia ruttneri* were shown in high values which was similar to that which was reported by Palmer (1970), Whitmore (1989), Jüttner *et al*. (2003), Stenger-Kovács *et al*. (2007), Duong *et al*. (2007) and García *et al*. (2008). They all reported the findings of these species indicating that they were tolerant to organic pollution. While the diatoms species of *Craticula molestiformis, Cymbella* cf. *bifurcumstigma, Cymbella* cf. *geddiana, Encyonopsis leei, Encyonopsis microcephala, Navicula capitatoradiata, Navicula heimansioides, Navicula hintzii, Nitzschia linearis* and *Nitzschia frequens* were found at low values and could be the oligo-mesotrophic indicator species. This result was similar to that which was reported by Chen and Wu (1999), Wan Maznah and Mansor (2002), Pekthong (2002), Lobo *et al*. (2004), Sahun (2004), Kunpradid (2005), Stenger-Kovács *et al*. (2007), Suphan (2009), Wang *et al*. (2010) and Jüttner *et al*. (2010), Leelahakriengkrai (2011) and Yana (2014).

In addition, the 17 benthic diatoms species reported in this studied were found to compare with the international index (Van Dam, 1994; Rott, 1997). The index values of some species of this study revealed differences when compared with the international index, such as *Achnanthidium exiguum, Achnanthidium minutissimum, Hantzschia amphioxys, Navicula cryptotenella*. This finding was similar to those of Soininen *et al*.

(2004), Soininen and Könönen (2004), Townsend and Gell (2004), and Poulíčková *et al*. (2010), each of whom reported that these species seemed to be tolerant in a wide range of stream water qualities and are common habitats for cosmopolitan freshwater pennate diatoms. The main factors related to the different index values were a result of the various kinds of specific characteristics found in each area, such as in factors associated with climate, geological, altitude and substrate etc. (Tison *et al.*, 2005). While some species were slightly different in terms of indicator values, such as *Gomphonema gracile, Gomphonema micropus, Navicula rostellata*, *Nitzschia palea* and *Nitzschia dissipata.*

5.4 Comparison of Wang Diatom Index with other indexes

The Wang Diatom Index was used to calculate the trophic status and water quality of the Wang River. This revealed a range of water quality from oligo-mesotrophic to meso-eutrophic status or clean-moderate water quality to moderate-polluted water quality. Different classifications of status were indicated by a comparison of the trophic status of the Wang Diatom Index with the AARL-PC score from each sampling site. The trophic status of the AARL-PC score was classified as oligotrophic to meso-trophic status, while most of the results of the Wang Diatom Index revealed a trophic status that was higher than the AARL-PC score. At a majority of the urban sites, a large percentage of the AARL-PC scores were less than those acquired from the trophic index and revealed oligotrophic to oligo-mesotrophic status, while the Wang River Index presented the trophic as being from mesotrophic to meso-eutrophic status. The reason for the differences in these index results was due to the fact that the AARL-PC score was acquired from certain physical and chemical properties that were directly analyzed in the field at that specific time. Nevertheless, using the Wang Diatom Index that was dependent on the diatom assemblage and was also directly related to water quality changes. Diatom index appropriate to indicate the water quality during the long period but AARL-PC score should be used to indicate at that time only. Diatom index values were calculated for optimum value per taxon with an average value of the main parameter at each site. This revealed various water properties (Rollins *et al.*, 2006). In addition, the water quality assessment that was made using diatoms is considerably more useful due to the fact that it is considered more precise when applied over a long time period of monitoring. It also takes into account the results occurring from large-scale pollution sources that change over time (Lowe and Pan, 1996). Whereas, the water assessment that is achieved by using chemical and physical methods reveals data on the conditions at the time that the measurements were taken (Kelly, 2003). These results are in agreement with those found in the study by Trivedi *et al.* (2008), who found that the biological indicator methods were practical in assessing the broader impacts of certain pollutants on the ecosystem, as well as revealing the chemical methods that are useful in assessing the present situation at the time of monitoring.

The comparison of the trophic status, between the Wang Diatom Index with other indexes such as the Rott Index, (1997), the Van Dam Index (1994), the Mea Sa Diatom Index (Pekthong, 2002), the Mekong Diatom Index (Suphan, 2009) as well as with regard to the saprobic status, such as in the Rott Index (Rott *et al*., 1997), the Ping and Nan Diatom Index (Kunpradid, 2005), the Thailand Diatom Index (Leelahakriengkrai, 2011) and the Yom Diatom Index (Yana, 2014) revealed the specific differences from each index. The calculation of the saprobic and trophic status by Rott *et al*. (1997) and the Van Dam Index (1994), which is used in Europe, showed a significant number of differences when compared with the Wang Diatom Index. Especially, interest was the Van Dam Index, which indicated higher values in the eutrophic status at all sampling sites because the distribution of diatoms species and water quality in Thailand were different with those from Europe. This result was similar to the findings published in the previous such as Pekthong(2002), Kunpradid (2005), Suphan (2009), Leelahakriengkrai (2011) and Yana (2014). However, the trophic status that were recorded using other indexes from Thailand, were slightly different from the status recorded with the Wang Diatom Index. According to the different values, the diatom list in each index was different, as each index was established from different diatom distribution locations in each area of study. Moreover, each index utility factor was different within the category-range. Therefore, for optimum convenience of using a diatom index, it should be developed based on the organisms that are naturally present in that area (Yusano *et al*., 1991; Koster and Hübener, 2001). Jüttner *et al*. (2003) reported that some diatom indices could not precisely reflect the water quality because the responses of the particular taxa to the water chemistry might vary between geographical regions, or taxa indicating different ecological conditions, which were being combined under a single name. Nevertheless, the use of diatom indices offers a good compromise between the exact estimation of the trophic parameters and the need for a simplified practical resource.