CHAPTER 1

INTRODUCTION

The spirogyra is consumed in an uncooked form in the north and northeast Thailand. It has local name is tao, thao, or thaonam (Peerapornpisal, 2006). Its generic name is in the genus *Spirogyra* Link (1820). *Spirogyra* is the filamentous freshwater green algae most easily recognized in family Zygnemaceae due to its spirally coiled chloroplasts. It is comprises filamentous, unbranched algae that show a unique model of sexual reproduction, and there are more than 400 species in the world. It's reproduce asexually by cell division and fragmentation and sexually by conjugation. They form green scum on ponds, floated by the oxygen produced by photosynthesis. It is found in a wide range of habitats, included small stagnant water bodies, ditches as well as the littorals of lakes, rivers and streams. Interestingly, *Spirogyra* record remain limited to generic level in floristic checklists and biodiversity inventories because of identification problems nor species composition at a sampling site could be given. Taxonomy of *Spirogyra* at the vegetative growth can be classified for three characters: (i) type of cross walls (plane, replicated, semi-replicate or colligate), (ii) cell length and width and (iii) chloroplast numbers. Furthermore, the process of conjugation has to be included for species identification. Moreover, the sexually reproductive stage from field was rarely collected. On the other hand, stress induction such as temperature, drought and pH to *Spirogyra* can induce the formation of conjugation tube for the fertilization of male and female gametes. The morphology of thin conjugation tube and zygote are also be a significantly evidence for advanced

identification system. As little are known of its ecology and affected to morphologically distinct filamentous forms (Berry and Lembi, 2000; Hainz *et al.*, 2009).

Distribution of *Spirogyra* in Thailand is cosmopolitan which reaching abundance during hot dry and before entering rainy seasons. Thiamdao and Peerapornpisal (2011) was reported the morphology of *S. ellipsospora* from northern part of Thailand under light and transmission electron microscope but only few species have been identified due to a lack of scientific references. Moreover, another part of Thailand must be determined. Because of the different of geographical distribution, which may induce the emerging of variation at genetic level, including the effect of different preferred habitat and surrounding climatic features that seem to be the reliable causative variation factors (Shen, 2008).

This study is aimed to determine the morphology of *Spirogyra* distributed from some water resources of Thailand, and would be related to some ecological parameters, including to determine the genetic variation and ecological relevance of *Spirogyra* population in Thailand. For the specimen collection and ecological study, 36 sampling sites in some water resources of Thailand will be designed, biological parameters; cell length/width, pyrenoids and chloroplast spiral; will be measured. Some ecological parameters; water temperature, pH, DO, salinity and conductivity will also be taken to determine correlation between biological and ecological parameters using correlation coefficient in SPSS program. SEM will also be performed to investigate ultrastructural surface of *Spirogyra* species.

The molecular identification of *Spirogyra* which morphological difference in Thailand would be investigated the genotyping of ISSR marker, it has been

recognized as effective/reliable method for demonstrating of molecular identified and development of DNA markers. Furthermore, Riburose 1, 5 biphosphate caboxylase (*rbcL*) gene and internal transcribed spacer subunit 2 (ITS 2), a conserved region of ribosomal gene complex will also be introduced. Genetic relationships will be analyzed based on both ISSR profiles and DNA sequence. Phylogenetic tree (phylogram) will be constructed using CLUSTAL W and MEGA programs. All data from this study will be included and analyzed to perform the diversity and relationship of *Spirogyra* and its ecological habitat in Thailand. In the addition, the results of molecular identification in this study could be usefulness for *Spirogyra* classification in Thailand.

Research Objectives

1. To investigate some ecological, biological characters and morphological structure using light and scanning electron microscope of *Spirogyra* spp. found in some water resources of Thailand.

2. To evaluate the molecular identification and genetic relationships of *Spirogyra* spp. populations found in some water resources of Thailand using inter simple sequence repeats (ISSR) markers and internal transcribed spacer subunit 2 (ITS 2) sequences.

3. To develop DNA markers for identification of *Spirogyra* spp. found in Thailand.