CHAPTER 1

Introduction

1.1 Statement and significance of the problems

Echinostomes are intestinal parasitic trematodes that adult stage is infecting a wide range of vertebrate host species, including mammals and humans, and their larval stages are also be parasite of numerous invertebrate and vertebrate hosts. The zoonotic potential of echinostomes is associated with the ingestion of raw mollusks, fishes and amphibians that harbor the naturally infective stage of these parasites (Esteban and Antoli, 2009). Trematodes of the family Echinostomatidae (echinostomes) are food-borne parasitic zoonoses, which can cause a serious public health concern in many parts of the world. A total of 20 species belonging to 9 genera have been reported to cause human infections throughout the world and also constitute public health importance especially in the Southeast Asia and Far East (Haseb and Eveland, 2000; Chai, 2009). Echinostomiasis is relatively rare, yet the foci of transmission remain endemic owing to the local dietary preferences. Most of these endemic foci are localized in China, India, Indonesia, Korea, Malaysia, Philippines, Russia, Taiwan and Thailand (Haseb and Eveland, 2000; Chai, 2009). In Thailand, echinostomiasis is caused by *Echinostoma* spp. which commonly found in North and Northeastern parts of Thailand (Nithikathkul *et al*., 2008).

Several genera of Echinostomatidae show significant meanings in biological, medical, veterinary and experimental aspects and the genus of *Echinostoma* has frequently been introduced as experimental models of worldwide research including their biology, which has also been well studied (Maldonado and Lanfredi, 2009). Within the genus *Echinostoma*, the 37-collar spines group contains sibling species which resemble one another closely, so it's difficult to distinguish such parasites from one another by using only traditional morphological characteristics or conventional methods. From these, the systematics of *Echinostoma* have long been problematic due

to the inter-specific homogeneity of characters and the poor differential diagnoses of newly established taxa (Kostadinova and Gibson, 2000). Therefore, the specific and accurate detections are needed for better classification, identification and epidemiological control program.

Recently, molecular approaches provide new systematic evidences which are validated for echinostomes identification system (Toledo *et al*., 2009). Many researchers have applied the polymerase chain reaction (PCR) based technique for discriminating many parasites in taxonomically confusing status or specific diagnosis of some important parasites. Several previous studies have demonstrated that molecular genetic analysis may improve understanding on taxonomy.

In summary, due to the fact that systematics of the genus *Echinostoma* still remain in a confused state, to access the better definition and reliability of *Echinostoma* identification, consequent studies concentrating in multidiscipline via biological features; including the morphology, morphometrics, host-parasite relationships, geographic distribution and molecular biology are required to determine, identify and classify the actual species of this genus.

1.2 Research objectives

In general, the main objectives of this research were to investigate the epidemiology of echinostome metacercariae, biological characteristics and molecular characters of *Echinostoma* spp. which originated from the life history of this fluke in Chiang Mai Province. Therefore, this study was conducted with the specific objectives:

1. To observe the prevalence of echinostomes metacercariae in snails in some areas of Chiang Mai province.
2. To investigate the life history of *Echinostoma revolutum*.
3. To establish loop-mediated isothermal amplification (LAMP) based approach for the specificity, sensitive and rapid detection of *E. revolutum.*
4. To investigate the phylogenetic relationships of *E. revolutum* by using sequences of nuclear rDNA internal transcribed spacer subunit 2 (ITS2) and nicotinamide adenine dinucleotide dehydrogenase subunit 1 (ND1) gene.