CHAPTER I

INTRODUCTION

Curcuma comosa Roxb. is a plant in family Zingiberaceae. It has common names in Thai as Waan chak mod look (เต็ม สมิตินันท์, 2543). In Thailand, it has been used widely in traditional medicine in several provinces along Cambodia border and North-eastern part of Thailand. Thai traditional practitioners have used its rhizome for the alleviation of postpartum uterine pain, enhancement of uterine involution and for reducing inflammation of the uterus after delivery (เสงี่ยม พงษ์บุญรอด, 2493; ประเสริฐ พรหมณี, 2516). It has been employed to relief pain in scrotal sac herniation (สมาคมโรงเรียนแพทย์แผนโบราณ สำนักวัดพระเชตุพนวิมลมังคลาราม, 2520).

C. comosa contains many chemical constituents such as diarylheptanoids (e.g. *trans*-1,7-diphenyl-5-hydroxy-1-heptene, *trans*-1,7-Diphenyl-6-hepten-3-one-5-ol, *trans*-1,7-diphenyl-3-acetoxy-6-heptene, *trans*-1,7-diphenyl-6-heptene-3-one, *trans,trans*-1,7-diphenyl-1,3-heptadien-5-ol, *trans,trans*-1,7-diphenyl-4,6-heptadien-3-one, 1,7-diphenyl-(*1E,3E,5E*)-heptatriene, 5-hydroxy-7-(4-hydroxyphenyl)-1-phenyl-(*1E*)-1-heptene), 7-(3,4-dihydroxyphenyl-5-hydroxy-1-phenyl-(*1E*)-1-heptene and acetophenones (e.g. 4,6-dihydroxy-2-O-(beta-D-glucopyranosyl)acetophenone.

Pharmacological investigations in animals demonstrated that *C. comosa* exerted uterotrophic activity (Piyachaturawat *et al.*, 1995a, 1995b; อนุกูล สวัสดิ์พาณิชย์, 2537), estrogenic activity (Piyachaturawat *et al.*, 1998a, 1999a), choleretic activity (Piyachaturawat *et al.*, 1996, 1998b, 2002a; Suksamran *et al.*, 1997), hypolipidemic effect (Piyachaturawat *et al.*, 1997, 1999b) and nematocidal effect (Jurgens *et al.*, 1994). Toxicities of *C. comosa* were investigated including acute & subacute toxicity studies (Piyachaturawat *et al.*, 2002b) and subchronic toxicity (Chivapat *et al.*, 2003).

Pharmacological properties of *C. comosa* have been reported. The uterotrophic activity of *C. comosa* was investigated *in vivo*. It was found that the hexane extract was most effective in increasing of uterine weight (Piyachaturawat *et al.*, 1995a) and vaginal mucosa thickness (Piyachaturawat *et al.*, 1995b). Effect of *C. comosa* ethanolic extract

was performed on the contraction of intact and isolated rat uterus. The results showed that the extract reduce the uterus contraction induced by oxytocin and some agents (อนุกูล สวัสดิ์พาณิชย์, 2537). Choleretic effect of *C. comosa* was exhibited in the ethyl acetate extract. Administration the extract to normal rats, was shown to significantly stimulate bile secretion with an increased biliary cholesterol secretion leading to a decrease in plasma cholesterol (Piyachaturawat *et al.*, 1996). The active compound isolated from the ethyl acetate extract was identified to be phloracetophenone glucoside (Suksamran *et al.*, 1997). Effects of synthetic phloracetophenone were investigated on bile flow and biliary lipid secretion. The results showed an increase of bile flow rate and biliary secretion of bile acid (Piyachaturawat *et al.*, 1998b). Ethyl acetate extract of *C. comosa* was shown to effectively reduce plasma triglycerides and cholesterol in diet-induced hypercholesterolaemic animals (Piyachaturawat *et al.*, 1997; 1999). Consistantly, phloracetophenone which was found in the ethyl acetate fraction was found to decrease plasma cholesterol, very low density lipoprotein (VLDL)-cholesterol and low density lipoprotein (LDL)-cholesterol (Piyachaturawat *et al.*, 2002).

C. comosa has been traditionally used for female abnormal symptoms of uterus and repeated administration is always suggested for good effectiveness. Generally, repeated exposure to some chemical may affect (either induce or inhibit) hepatic drug metabolizing enzymes. Thus, drug-drug interactions and possibilities to increase/decrease risks of chemical-induced toxicity, mutagenesis and/or carcinogenesis may occur following those chemicals administration. So far, no study was performed regarding the effects of *C. comosa* ethanolic extract on hepatic cytochrome P450 (CYP), the family of enzymes possessing the important role in drug metabolism and chemical-induced toxicity, mutagenesis and/or carcinogenesis. These enzymes included CYPs 1A1, 1A2, 2B1, 2B2, 2E1 and 3A (Soucek and Gut, 1992). Modulation of these CYP isoforms following *C. comosa* exposure, would provide an information of drug-drug interaction if *C. comosa* was administred concomitantly with other drugs which are metabolized by the particular CYP isoform. Inhibition of these CYP

isoforms are partly a key aspect explanation for anti-mutagenic and anti-carcinogenic potential of *C. comosa*. In contrast, induction of these CYP isoforms following repeated exposure of *C. comosa*, may increase risk of carcinogenesis from xenobiotics that are bioactivated by the CYP isoforms which are modulated. Therefore, the objectives of this study were to preliminarily investigate effects of *C. comosa* ethanolic extract on some CYP isoforms such as CYPs 1A1, 1A2, 2B1, 2B2, 2E1 and 3A, which were involved in drug metabolism and mutagen and/or carcinogen activation, using an *ex vivo* study in rats. Furthermore, effects of the *C. comosa* ethanolic extract on clinical blood chemistry and hematology were also determined to confirm the previous toxicity data of this plant in rats.

Hypothesis

C. comosa ethanolic extract demonstrated an induction and/or inhibition effects on hepatic microsomal CYP as well as caused changes of clinical blood chemistry and hematology in rats.

Study design and process

1. Preparation and identification of *C. comosa* ethanolic extract
2. Animal treatment: an *ex vivo* study
3. Blood collecting
4. Determination of clinical blood chemistry and hematology
5. Preparation of liver microsomes
6. Determination of total CYP contents and CYP activities
7. Data analysis

Anticipated benefits from the study

1. A preliminary data of *C. comosa* ethanolic extract whether it possessed an induction and/or inhibition effects on hepatic CYP, the isoforms involving in various bioactivation reactions of certain of drugs, chemicals as well as environmental toxicants resulting in reactive metabolites. This would be useful to estimate the possibility of *C. comosa* to increase and/or decrease risks of chemical-induced toxicities, mutagenesis and/or carcinogenesis.

2. The possibility of drug-drug interaction if this extract was taken simultaneously with other medicines metabolized by CYP isoform that was modulated by this extract.

3. The clinical blood chemistry and hematology data of *C. comosa* from this study, would be useful to confirm the previous toxicity data of *C. comosa* in rats.