

# CHAPTER I

## INTRODUCTION

### 1. Rationale and Background

Alzheimer's disease (AD) is the most common neurodegenerative disorder with dementia in the elderly. At present, the elderly is the most rapidly growing part of the population. In addition, the increase in life expectancy will inevitably lead to a further increase in the prevalence of AD. It was estimated that the number of AD patients worldwide was approximate 20–25 million. In 2004, the prevalence of AD in Thailand was estimated to be 953,904 cases from the total of 64,865,523 Thai populations (US Census Bureau, International Data Base, 2004).

Previous studies have demonstrated that AD accounted for more than 60% of dementia cases (Fratiglioni et al., 2000). It has been proposed that the main cardinal symptom of AD is memory impairment. However, all memory functions are not equally affected. It was found that episodic memory was markedly affected in the early course of the disease (Coyle et al., 1983). This cognitive decline could interfere with normal daily life activity, which in turn disturbed the quality of life of the patients. In addition, this condition also affected the quality of life of their families because the health care cost of AD patient was very high. Previous study demonstrated that the Medicare cost for demented patient was higher than other cases approximate 4,000\$ per case (Alzheimer Association, 2001). Therefore, AD is one of the important problems worldwide including in Thailand.

At present, the therapeutic strategies of Alzheimer's disease principally relies on nootropic agents, such as piracetam, aniracetam, fosracetam, nefiracetam, etc., and anticholinesterases, such as donepezil, metrifonate, rivastigmine, etc. (Ringman and Cummings, 1999; Sramek et al., 2000; Mashkovskii and Glushkov, 2001; Potkin et al., 2001; Balaraman and Shingala, 2002; Sugimoto et al., 2002; Gauthier et al., 2003). Unfortunately, it is currently reported that no effective treatment to halt the progression or prevent the onset of AD. Therefore, it is worthwhile to look for new directions, which would minimize the memory loss of patients with AD.

Inflammation and oxidative stress are proposed to be key components of the pathology of the chronic neurodegenerative conditions particularly in AD. Thus, numerous dietary substances and herbal extracts possess antioxidant could significantly modify the complex mechanisms of neurodegenerative diseases and exerted neuroprotective effect against these diseases including AD (Aruoma et al., 2003). Previous studies demonstrated that many anti-aging products and therapeutic agents against age-related neurodegenerative diseases particularly AD had been developed from Oriental herbal medicine (Chang, 2001; Bastianetto, 2002; Lei et al., 2003). The neuroprotective effects of the both diet and herbal extract were also reported to be associated with the polyphenolic compounds existed in those substances (Kang et al., 2005). Moreover, growing lines of evidence have suggested that flavonoids have neuroprotective effects against AD.

Quercetin is a member of an extensive class of polyphenolic flavonoid compounds that is widely distributed mainly as glycosides in components of the daily diet such as onions, apples, berries, tea and red wine (Hertog et al., 1993). It is nontoxic and displays a variety of biological actions such as antiviral (Ohnishi and Bannai, 1993), antiulcer (Alarcon de la Lastra et al., 1994), antiallergic (Murray, 1998), and anticancer (Lamson and Brignall, 2000), anti-inflammatory, antineoplastic and cardioprotective actives (Middleton et al., 2000). Moreover, it is known as a strong free radical scavenger (Takahama, 1985). Based on its antioxidant and anti-inflammation, quercetin may possibly be a potential candidate as therapeutic agent against Alzheimer's disease. However, until today, the scientific evidence to support this application is very limited.

At present, it was found that targeted drug delivery to the central nervous system (CNS) for the therapeutic advancement of Alzheimer's disease is complicated by restrictive mechanisms imposed at the blood-brain barrier (BBB). In order to overcome the impositions of the BBB, polymeric biocompatible drug carriers have been applied to the central nervous system. Most of the conventional delivery system of antioxidant therapy nowadays shows many disadvantages including the limitation in route of administration and first pass effect (Kreuter, 2001). Previous studies also demonstrated that a novel drug delivery system such as vesicle mediated carrier system particularly antioxidant liposomes were able to improve the efficacy of

antioxidant on the central nervous system. The antioxidant liposomes could prevent ischemia and reperfusion in the rat brain whereas the free forms of antioxidants failed or showed little effect (Sinha et al., 2001). Therefore, this raises the possibility that quercetin in a vesicle carrier system should be able to improve the therapeutic potential against AD. Unfortunately, the supported document about this issue is still very limited until now. Therefore, the current study is designed to determine the efficacy of liposomes encapsulation of quercetin on neuroprotective effect against AD.

## **2. Objectives of the Study**

### **General experiment objective**

This experiment was designed to investigate the potential neuroprotective effect of quercetin liposomes against Alzheimer's disease.

### **Specific objectives**

1. To determine the neuropharmacological activities of quercetin with oral conventional delivery system.
2. To determine the neuropharmacological activities of quercetin encapsulated liposomes via nasal administration.
3. To determine the neuroprotective effect of quercetin with oral conventional delivery system in both healthy and cognitive deficit conditions of Alzheimer's disease model.
4. To determine the neuroprotective effect of quercetin encapsulated liposomes via nasal administration in both healthy and cognitive deficit conditions of Alzheimer's disease model.
5. To determine the possible mechanisms underlying the cognitive enhancement and the neuroprotective effect of quercetin with oral conventional system against Alzheimer's disease.
6. To determine the possible mechanisms underlying the cognitive enhancement and the neuroprotective effect of quercetin encapsulated liposomes via nasal administration against Alzheimer's disease.

### **3. Scope and Limitation of the Study**

3.1 All animals used in this experiment were male only and the ages were approximately at 8 weeks old (young adult, weighing 180-220 grams).

3.2 All administration should be done at the same period to avoid influence of circadian rhythm.

3.3 All stress should be avoided to prevent the influence of stress.

3.4 In all experiments, control and experimental groups should be performed in parallel at the same period to avoid effect of seasonal changes.

### **4. Anticipated Outcomes**

1. To provide the information about neuropharmacological activities of quercetin and quercetin liposomes.

2. To provide the information about new therapeutic approach of antioxidant therapy of quercetin liposomes.

3. To provide the information about the efficacy of quercetin and quercetin liposomes administered via nasal administration on cognitive enhancement and neuroprotective effect in healthy condition.

4. To provide the information about the efficacy of quercetin and quercetin liposomes administered via nasal administration on cognitive enhancement and neuroprotective effect in memory deficit condition of Alzheimer's disease model.

5. To provide the information about the possible mechanism underlying the cognitive enhancement and neuroprotective effect of quercetin and quercetin liposomes administered via nasal administration against Alzheimer's disease.

### **5. Hypothesis**

1. If quercetin delivered via oral conventional form possesses the neuropharmacological activities, the animals that received quercetin should show more anxiolytic, anti-depression like activity and better cognitive performance than the vehicle treated group.

2. If nasal administration of quercetin liposomes possesses the neuropharmacological activities, the animals that received quercetin liposomes via

nasal administration should show more anxiolytic, anti-depression like activity and better cognitive performance than the free liposomes and vehicle treated group.

3. If quercetin with oral conventional delivery system possesses neuroprotective effect, the animals that received quercetin should show higher density of neurons and cholinergic neurons in cerebral cortex and hippocampus than the vehicle treated group in both healthy and cognitive deficit conditions of Alzheimer's disease model.

4. If nasal administration of quercetin liposomes possesses neuroprotective effect, the animals that received quercetin liposomes should show higher density of neurons and cholinergic neurons in cerebral cortex and hippocampus than the free liposomes and vehicle treated group in both healthy and cognitive deficit conditions of Alzheimer's disease model.

5. If quercetin with oral conventional delivery system exerts its cognitive enhancing effect via the increase in available acetylcholine, the animals that received quercetin plus AF64A, should show lower activities of acetylcholinesterase (AChE) than those of animals that received vehicle plus AF64A.

6. If nasal administration of quercetin liposomes exert its cognitive enhancing effect via the increase in available acetylcholine, the animals that received quercetin liposomes plus AF64A, should show lower activities of acetylcholinesterase (AChE) than those of animals that received liposomes plus vehicle and AF64A.

7. If quercetin with oral conventional delivery system exerts its neuroprotective effect via the increase in the activities of scavenging enzymes, the animal that received quercetin plus AF64A, should show higher activities of scavenging enzymes (superoxide dismutase, catalase and glutathione peroxidase) but lower lipid peroxidation product than those of animals that received vehicle plus AF64A.

8. If nasal administration of quercetin liposomes exerts its neuroprotective effect via the increase in the activities of scavenging enzymes, the animal that received quercetin liposomes plus AF64A, should show higher activities of scavenging enzymes (superoxide dismutase, catalase and glutathione peroxidase) but lower lipid peroxidation product than those of animals that received liposomes plus vehicle and AF64A.

