CHAPTER I

INTRODUCTION

Breast cancer is a disease which often happens with Thai female. From statistical data of National Cancer Institue of Thailand in 1993 reported that epitheloid cervix cancer was the first of all cancer disease in Thai people and breast cancer was the second estimated at 16.3% of cancer disease in Thai women (1). So, attemping to increase potential treatment for the patient with breast cancer disease is very interesting. At present there are 4 methods for cancer treatment which are by surgical management, radiotherapy, chemotherapy and hormone therapy. If the patients, with first stage, cancer development were treated with the right therapy over 90% of these patients will survive longer than 5 years. However, in most patients the symptom of breast cancer was detected when they were in the last stage of disease. The treatment will be difficult and the majority of patients in this stage will response to chemotherapy. Therefore, chemotherapy is the method of choice which is wildly taken to treat cancer now (2,3,4).

Chemotherapy is the method of treatment for patients with cancer by using medicines which are chemical agents for destruction or proliferous inhibition of cancer cell. However the unacceptable and undesired side effects of many antitumor agents are a major problem that needs to be improved, due to the need for the antitumor agents to be used for long durations. In recent years there were many researchs revealed and found antitumor agents for inhibition of breast cancer cell growth such as anacadic acid, cardols, methylcardols in the cashew (*Anacardium occidentale*) apple juice which exhibited significant cytotoxicity ($ED_{50} < 20$ μg/ml) in vitro cytotoxicity against BT-20 breast carcinoma cells (5). Indole-3-carbinol from Brassica vegetables has been shown to reduce the proliferation of breast cancer type MCF-7 (6) etc. Consuming of these fruits may reduce the risk of breast cancer. In addition, the National Cancer Institute discovered new chemotherapy agent Taxol (Paclitaxol), identifies as diterpene group

which was extracted from bark of *Taxus brevifolia.* Patients with metastatic breast cancer, advanecd lung cancer, and lymphomas have positive responded to Taxol. However Taxol had many side effects such as myelosuppression or neurotoxicity (7). The major problem for Thai people to use taxol are the expensive cost because drugs are imported from aboard inwhich one dose for cancer treatment by Taxol was estimated at 100,000 Bath. So medicinal plants are new alternative for Thai people because they are widly found, cheaper and have less side effects.

In 1993 Akira and co-worker studied the antitumor promoting properties of a total of 40 methanol extracts from Thai medicinal plants, including 5 non-edible species. They were tested in vitro by an inhibition test of tumor promoter - induced Epstein - Barr virus (EBV) activation. Fourteen species (35% of the total) were found to be strongly active (8). In addition, many Thai herbs show antifungal properties such as leaves and stems of *Rhinacanthus nasutus* are often taken to cure cutaneous eruptions due to ringworm, eczema. In 1988 root extract of this plant with methanol by Tian-Shung Wu and his group found two compounds called rhinacanthin-G and rhinacanthin-C which inhibited the growth of P-388 and HL-60 cancer cell line (9).

Kaempferia parviflora, Zingiberaceae family, is a indigenous plants in Thailand. A lot of plants in this family are edible plants and they are taken to be folk medicine as ginger (*Zingiber officinale*). Moreover many of these herbs contain potent antioxidant, antitumor, antimicrobial compound. As an example, *Curcuma longa* contains curcumin (diferuloylmethane), gingerols and diarylheptanoids which have antioxidant, antiinflammation and antitumor as metastatic breast cancer activities (10).

Kaempferia parviflora has various scientific name such as *Gastrochilus pandurata*, *Boesenbergia pandurata, Boesenbergia rotunda*. Many medicinal plants books classify *Kaemferia parviflora* into 3 types as black, red and yellow rhizomes *Kaemferia parviflora* but generally the people know yellow rhizome. They are used to

improve flavor and taste in traditional Thai food. Traditionally, rhizomes are used for the treatment of oral cavity diseases such as aphthous ulcer and dry mouth, diuretic, treatment of stomach pain and Leukorrhea antidysenteric (11). There are many reports that crude extract from yellow rhizome with boiling water, cold water, ether, petrolium ether show no antibacterial activity to *Bacillus subtilis, Escherichia coli* and *Pseudomonas aeruginosa.* However, when extracted with alcohol, chloroform it had medium antifungal activity on *Microsporium gypseum, Candida albicans, Cryptococcus reoformans* (12). Moreover, cardamonin from crude methanol extract could inhibit tumor promoter-induced Epstein-Barr Virus (EBV) over 90% at 25 μM concentration (8). Volatile oil from black rhizome could inhibit the growth of *B. subtilis, Staphylococcus aureus* and *P. aeruginosa.* Result of cytotoxic activity against breast cancer cell lines from crude methanol extract of 14 herbs revealed that black rhizome *Kaemferia parviflora* (**Fig. 1.1**) had the highest cytotoxic activity against breast cancer cell lines (13) . Therefore, detail investigation on cytotoxic activity of *Kaemferia parviflora* should be continued.

1.1 THE OBJECTIVES OF THIS RESEARCH

1. To extract, isolate and purify the chemical constituents of *K. parviflora.*
2. To identify the structure of the isolated compounds from *K. parviflora.*
3. To determine cytotoxic activity against breast cancer cell lines as well as other cell lines of isolated compounds from *K. parviflora.*

1.2 THE SCOPE OF THIS RESEARCH

The research began by screening for the cytotoxic activity against breast cancer cell line of 17 medicinal plants and select the one which had the highest activity. That plant was extracted with suitable solvents, then the compounds from those crude extracts were isolated by chromatographic techniques. Structural elucidation of the isolated compounds was deduced from spectroscopic evidences. Finally, biological tests of the isolated compounds were conducted for cytotoxic activity against breast cancer cell line.

1.3 THE ADVANTAGE OF THIS RESEARCH

1. To know the cytotoxic activity against breast cancer cell line of the crude extract from *K. parviflora.*
2. To know the structure of compounds which have cytotoxic activity.

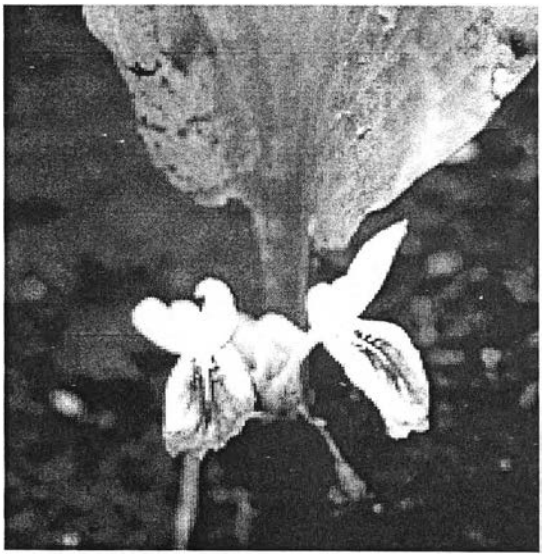

Figure 1.1 The picture of *Kaempferia parviflora.*