

## CHAPTER 1

### INTRODUCTION

The tropical weeds are distributed mainly over the tropical area of the world. Every year many weed areas have been damaged and used in many ways, for example, they are burnt after the harvest season or are fed by animals. In the present day, the chemistry of natural products of tropical weeds has widely been studied. Several isolated compounds were identified as novel compounds and number of known compounds were reisolated and proved to possess biological activities.

The weeds in *Sphaeranthus* genera, belonging to the family Compositae<sup>1</sup> are widespread in the tropical and subtropical areas of Africa, Asia and Australia.<sup>1,2</sup> Few of its 40 species have been chemically studied. The importance of this genus stems from the wide use of its members in folk medicine in the treatment of skin infection, glandular swelling, bronchitis, jaundice and nervous depression<sup>3</sup>.

In Thailand, the plants belonging to *Sphaeranthus* genus are composed of two species, *Sphaeranthus africanus* (Yah khon klong) and *Sphaeranthus indicus* (Ka ra boon),<sup>4</sup> *Sphaeranthus africanus* was selected to investigate for the bioactive compounds because of the preliminary screening of ethanolic crude extract of the whole plant of *S. africanus* revealed high cytotoxic activity against brine shrimp *Artemia salina* and carcinoma cell lines.

*S. africanus* was found abundantly as a weed in rice fields. They were scattered over the central region of Thailand. Their life cycle was about 1 year and the botanical aspects of *S. africanus* can be summarized as follows<sup>5</sup>:

**Stem** : The height is 15-45 cm. The upper part of the stem is greenish and the lower part is yellowish, around the stem full of hair and branches.

**Leaves** : The leaves are oval shaped, the size is about 0.5-3.5 cm. wide by 2-11 cm. long.

**Inflorescence** : The small, violet, typically bisexual flowers.

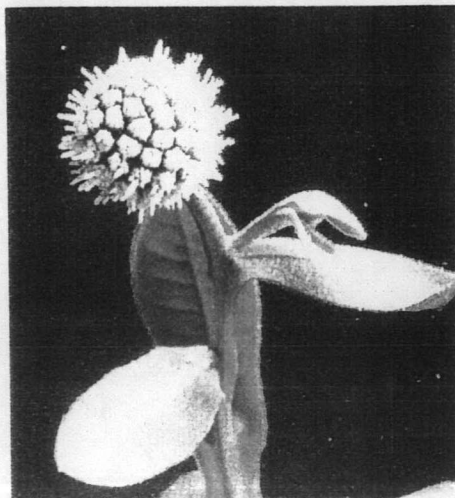
**Seed** : The dried seed is brown and cylinder-shaped, the size is about 1 mm.



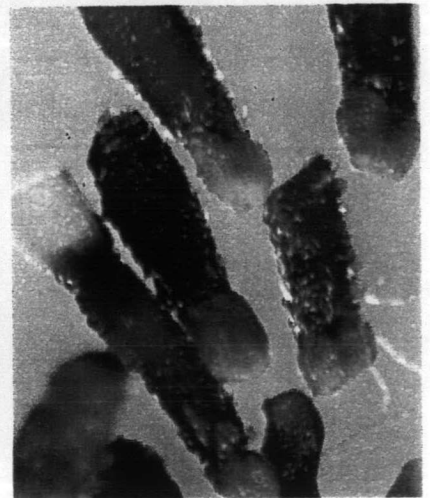
1. habitat



2. whole plant



3. inflorescence



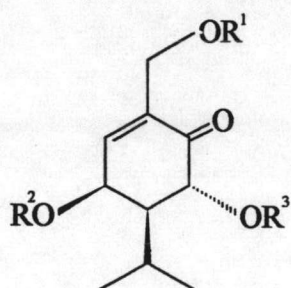
4. seed

**Figure 1.1** (1) habitat, (2) the whole plant, (3) inflorescence, (4) seed of *S. africanus*

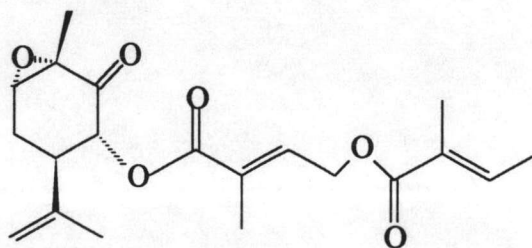
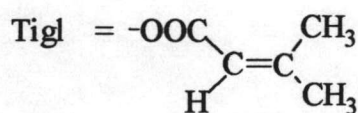
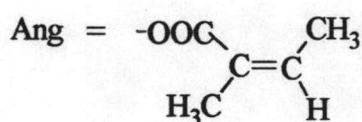
## 1.1 Chemical Constituents of Sphaeranthus Genus

Among 40 species of genus *Sphaeranthus*, six of them have been chemically studied<sup>5</sup>. The main isolated compounds could be classified into four groups: monoterpenes, sesquiterpenes, flavonoids and miscellaneous.

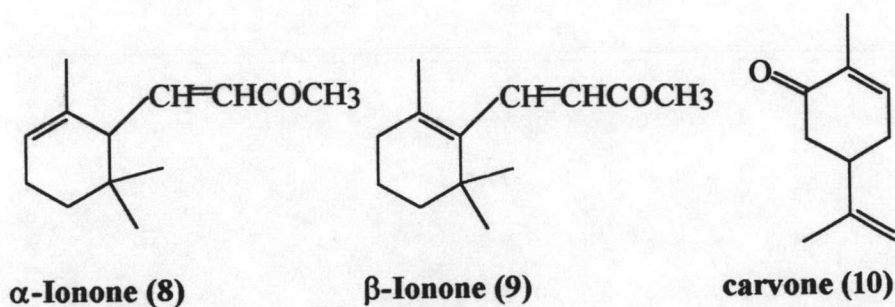
**1.1.1 monoterpenoids:** monoterpenoids were found in many species of *Sphaeranthus* genus such as *S. indicus*, *S. bullatus* and *S. suaveolens*. They are divided into two main groups, carvotacetone derivatives and simple cyclic monoterpenes. Several isolated carvotacetone derivatives (1-7) were identified as new compounds. The simple cyclic monoterpene, the main skeleton liked *p*-menthane, for example  $\alpha$ -ionone (8),  $\beta$ -ionone (9), and carvone (10) from *S. kirkii*<sup>6,7</sup>.



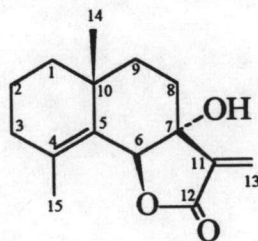
	(1)	(2)	(3)	(4)	(5)	(6)
R <sup>1</sup>	Tigl	H	Tigl	Ac	H	H
R <sup>2</sup>	Tigl	H	Tigl	Tigl	H	Ang
R <sup>3</sup>	H	H	Ac	H	Tigl	Tigl



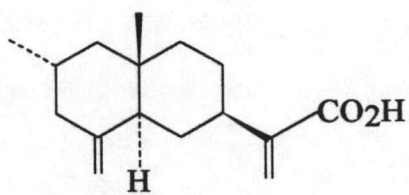
**1 $\alpha$ ,2 $\alpha$ -Epoxy-5 $\alpha$ [4-tigloyloxy-tigloyloxy]carvone (7)**



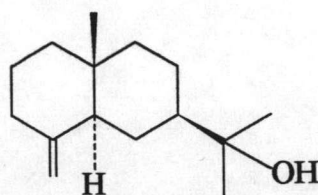
**1.1.2 Sesquiterpenoids:** According to previous reports,<sup>8</sup> sesquiterpenoids were likely to be predominant metabolite in this genus. Owing to the variety of their structures, they were usually discovered as new compounds. The major skeleton found amongst the eudesmanolides, which are sesquiterpene lactones.<sup>9</sup> Furthermore eudesmanolides isolated from this genus sharing the unusual 7-hydroxy substitution are very rare in nature. Such type of sesquiterpene lactone has been reported for the first time in 1986 from genus *Sphaeranthus* during the chemical investigation of *S. indicus*. has led to isolated a eudesmanolide and 7 $\alpha$ -hydroxyeudesm-4-en-6-12-olide(11).<sup>10,11</sup> In earlier studies, this compound was particularly interesting since it shown pronounced cytotoxicity and antitumor activity against a number of human cancer cell lines.<sup>12</sup> In addition, a new sesquiterpene acid, 2-hydroxycostic acid (12), along with the known compounds  $\beta$ -eudesmol (13) and ilicic acid (14) have been isolated from *S. indicus*. Later on two new eudesmanolides(15-16) were reported by Supada *et al*<sup>13</sup>



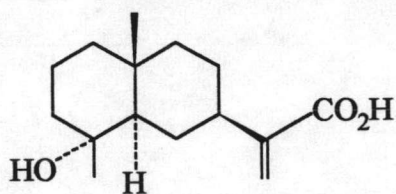
**7 $\alpha$ -hydroxyeudesm-4-en-6-12-olide (11)**



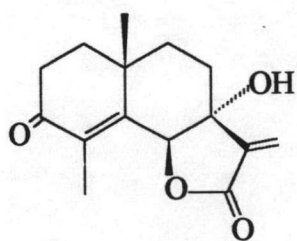
2-hydroxycostic acid (12)



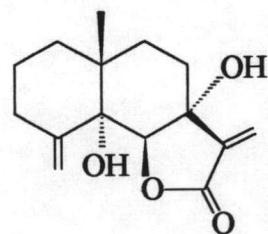
$\beta$ -eudesmol (13)



ilicic acid (14)

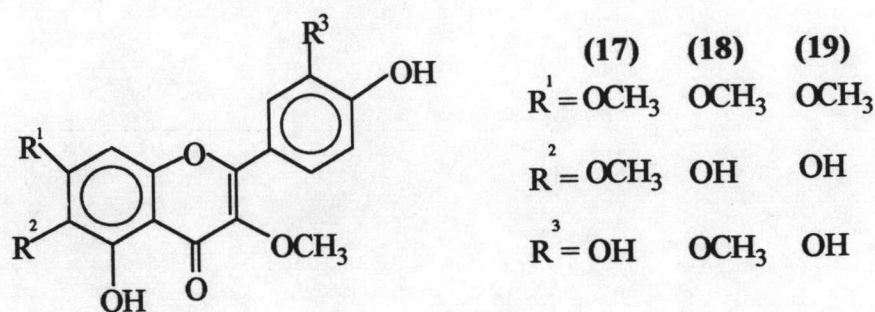


(15)



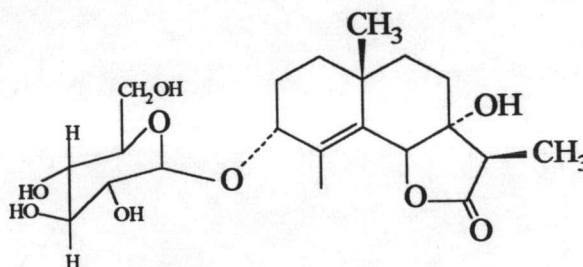
(16)

**1.1.3 Flavonoids:** Several plants in family Compositae usually possessed found flavonoids compounds as their chemical constituents, as well as *Sphaeranthus* genus. Three oxygenated flavonoids isolated from *S. africanus*<sup>15</sup>: quercetagenin-3,6,7-trimethyl ether (17), quercetagenin-3,3',7-trimethyl ether (18) and quercetagenin-3,7-dimethyl ether(19) are among these examples.

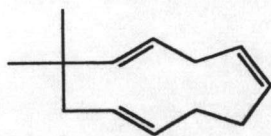


**flavonol**

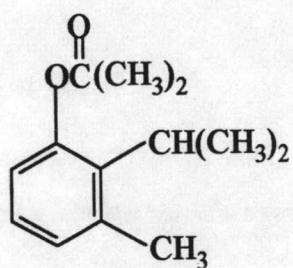
**1.1.4 Miscellaneous:** Although isolated compounds from *Sphaeranthus* genus were divided to three main groups other compounds were identified as new compounds and possess biological activity. For instance, a new sesquiterpene glycoside, sphaeranthanolides (20) has been isolated from the flowers of *S. indicus*. This compound exhibited immune stimulating activity.<sup>16</sup> Further investigation of *S. bullatus* and *S. suaveolens* revealed the presence of  $\alpha$ -humulene (21), thymolisutyrate (22), and complex mixture of inositol ester(23)<sup>17</sup> as their composition, respectively



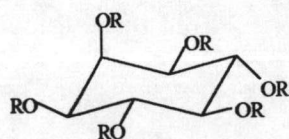
**sphaeranthanolides (20)**



$\alpha$ -humulene (21)



thymolisutyrate (22)



R = Ang, *i*-val, *i*-Bu, H

complex mixture of inositol ester (23)

## 1.2 The Goal of This Research

The preliminary screening test of the ethanolic extracts of the whole plants of *S. africanus*. showed high cytotoxic activity against brine shrimp *Artemia salina* and various cell lines. This is attractive to search for bioactive compounds. Therefore, the goal of this research can be summarized as follow:

1. To extract and to isolate the organic constituents from the whole plants of *Sphaeranthus africanus*.
2. To elucidate the chemical structures of the isolated compounds
3. To search for the bioactive compounds that can be used as cytotoxic agent against brine shrimp and/or otherwise by using bioassay result as a guide