CHAPTER I

INTRODUCTION

Background and Problematic Importance

Nature of the problems

Thailand is an agricultural country and majority of people have agricultural occupation for very long time from the ancient. Therefore, the increase of crop productivity is the major goal. To accomplish this, the mass production development of fertilizer and transferring some modern technology to use them in wide-spread are necessary. However, the productivity maybe decreased if there are troubles by many kinds of pests that sometime make the damage to agricultural areas.

Cramer, a German academic has estimated that the damage of plant which human uses as food all over the world caused by all kinds of pests, has total as much as 50 percent. This figure is very high. This means that all plants which have been cultivated put real outcome (productivity) only half.[1] In all kinds of plant enemies, it was found that insects are the pests which have highest direct damage to agricultural productivity. In addition, insect can indirectly cause plant pathogen and conduct or bring some disease to post-harvesting of receiving products.[2]

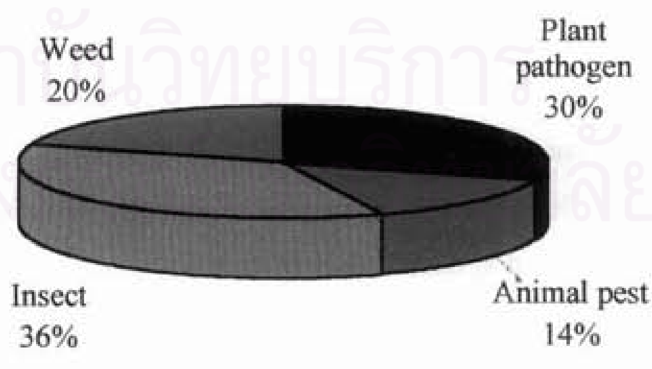

Figure 1.1 Percentage of damage caused by all pests from FAO's statistical data

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Therefore, when the advanced technology is progressing, there are a lot of effort to search for new chemicals. Most of them are synthetic organic chemicals being used to stop of the spread of insect that cause the damage to agricultural products. These chemicals are called insecticide. These of insecticides are very effective. However they may cause various serious side effects even if the chemicals from natural origin as insecticides were used. This phenomena stimulated scientists to find a new tendency to control insect pest not to spread. One method that received more increased interest in last decade was insect antifeedant, particularly by using substances derived from natural products.

Severety of the problems

It is well known that insects are the most species in the animal kingdom. The recent study has shown that there are approximately 1 million species of what and about half of them are phytophagous insects. In Thailand, it has not been collected significant insect species which make economic damage. There are two ways according to the damage of the production that results from destroying of insect: the direct damage of harvesting product as the giving damage to the product directly and the indirect damage of harvesting product as the giving productivity decrease or low quality. Last year, in Thailand it has estimated that only one species of grasshopper could make the damage to agricultural product in more than 200,000 rai all over the country. This figure is certainly not included the damage derived from other insect species.[3]

Scientists have estimated the damage of plants that caused by insect pest with the value of 400,000 million dollars, if account to the percent of damage caused by the destroying of insect pest. It is about 36 percent which is not account for the damage caused by insect from post harvesting.[1]

According to the above mentioned problem, it was essential to find a measure to eliminate and to control insect pest. Mostly, synthetic organic chemicals are employed to get rid of the mentioned insects from the crops. The use of insecticide is a favorite method for controlling insect pest because of its advantages of high efficiency, immediately appearance and simplicity to use. However, the insecticide can cause many serious problems such as it may be poisonous to human being and warm-blooded animals by accumulating in lipid layer causing illness. For example, although there was an order to stop using DDT more than 30 years but in the present it was still checked up in humans who have been used DDT. They have also affected to environment such as the contamination and the concentration in soil and water that makes the cycle feedback to human and other animals. Since those insecticides are difficult to degrade naturally insecticides derived from natural products which are thought to be biodegradable are used. However, the use of insecticide for a long period of time may cause many problems afterwards such as destroying equilibrium of the ecosystem and, destroying food chain relationship.[3]

As a consequence, during the last decade, there were may attempts searching for new perspectives to control insect population in order to reduce the damage of agricultural products, to economic threadhold including no environmental pollution and no interferring ecological equilibrium system. One modern approach to control insect pest is the usage of antifeedant compounds derived from natural products.

Review of Literature

Insect pest management

The prevention and controlling insect methods can be classified into 2 groups:

Natural control

This method goes on by nature without the participation of human beings. The major factor of natural pattern in insect control includes natural enemies (predator, parasite and disease), environment and geographical conditions. In addition, there are other important factors such as food (quantity and quality) and the competition between the same group and the others.

Applied control

This is a prevention and controlling method under human operating which can be divided into 2 subgroups: non chemical control and chemical control.[4]

a) Applied non-chemical controls are composed of many methods that do not make pollution to environment and interfere with natural equilibrium as

1. mechanical method such as hand-picking, sticky band and small holding etc.

2. shelter method such as a net house and a glasshouse.

3. physical control such as the methods of prevention and elimination insect involving the usage of physical factors including light, color, sound, heat and radiation.

4. cultural control

This is a method in agriculture which helps to minimize the destruction from insect. It is the prevention more than to kill insect directly. For instance, to adjust area or plant to make circumstance not suitable for reproduction or the survival of insect pest or to improve the plant to be of strong resistance from destroying and make circumstance suitable for natural enemies to control insect pests in low level quantity such as crop rotation, tillage, crop sanitation, grow perenial plant and giving weed to grow between plant plot and postpone the duration time of growing plant.

5. legal control

This method is the prevention the spread of insect pest from one country to the others by law.

6. breeding / genetic method

This method includes the growing insect resistance plant by breeding or using genetic manipulation to resist insects. The plant mechanism to resist insect from destroying can be classified into 4 types: pest avoidance, antixenosis or non-preference, antibosis and tolerance.

7. biological control (Biocontrol)

For example: natural enemies such as predator and insect parasite, organism such as bacteria, virus and protozoa *etc*, insect sterilization (autocide) and semiochemicals such as pheromone.[5]

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b) Applied chemical controls are the chemicals which use to prevent or eliminate the insect. These can be divided into 3 characteristics depending on the reaction to insect.

1. insecticides define as the chemicals used to prevent and eliminate insect including repel or decrease a number of insect.

2. insect growth regulators involve the chemicals which are used to inhibit the growth of insect that causes worm or larva cannot grow to adult.

3. insect behavioral chemicals refer to the chemicals which affect to insect behavior as attracted insect or repelled insect.[4]

Insecticides

Insecticides derived from synthetic chemicals can be classified into 2 groups:

Inorganic chemical insecticide

This class of chemicals was probably the earliest used compounds for pest control; most are stomach poison and have to be ingested to be effective. They are mostly naturally occurring compounds and most of them were salts of heavy metals. The most important compounds were probably copper acetoarsenite, lead arsenate, cryolite (sodium aluminofluoride), mercurous chloride, sodium fluoride, cyhexatin and sulfur. Some are still being used in the present time.

Organic chemical insecticide

This group can be categorized according to chemical composition as :

1. Chlorinated hydrocarbon (Organochlorines)

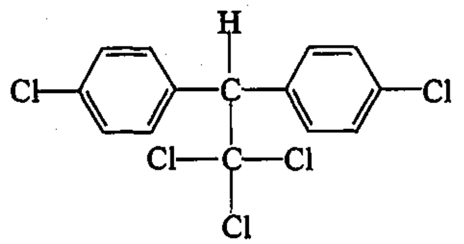
This was the first group among modern insecticides. Nowadays sometimes referred as "second-generation insecticide" whose development started with DDT in 1940. They are characterized by being long-lived (persistent), with a broad spectrum of action, as both contact and stomach poison. Insecticide in this group can be divided into 4 groups:[4, 6]

a) DDT and DDT analogues such as TDE and dicofol.

b) Hexachlorocyclohexane such as BHC and lindane.

c) Cyclodiene compound such as aldrin and heptachlor

d) Terpene compound such as toxaphene and endosulfan.



DDT structure

The destruction of natural enemies, including wild birds, environmental contamination and the development of resistance, led to the realization that these chemicals were not the universal panacea after all. For example, malaria is rapidly becoming just as wide spread a pest in the tropics as it was before the advent of DDT. The final blow of the organochlorines is that most are now banned both internationally and nationally by most countries.[7]

2. Phosphorus compound

They are very effective insecticides. Most are short-lived and have contacted and systemic-type actions. Some over 50 compounds have been withdrawn for general use because of toxicity dangers. Action appears to be *via* inhibition of acetylcholinesterase hence nerve poisons. Some of the newer compounds are more persistent and several are still very useful and widely used. Diazinon, dimethoate, fenitrothian, malathion, parathion, pirimiphos-methyl and tetrachloroinphos are some of the best known examples.[4, 6, 8]

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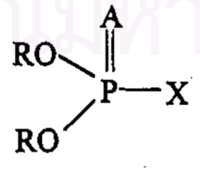


General structure of phosphorus compound

R = ethyl or methyl group

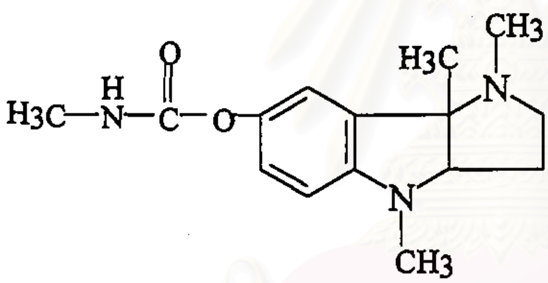
A = sulfur or oxygen atom

X = variable maybe are atoms or long/short chain group

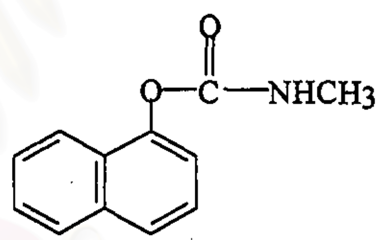
3. Carbamate compound

Several carbamates are used as insecticide at present (although most of them are fungicides and herbicides). They are generally not accumulated toxicity, degraded quickly, not have residual effect to environment. The toxicity to human and animal of these compounds is lower than that of organophosphorus. There are nevertheless still little use because it is too expensive and effective in prevention and elimination insect not equal to organophosphorus compounds. This type of chemicals can be divided into 3 groups:[4, 6, 8]

a) heterocyclic carbamate such as dimetilan and dimeton
b) phenyl carbamate such as carbaryl and carbofuran
c) oxime carbamate such as aldicurb and oxramyl



Physostismine

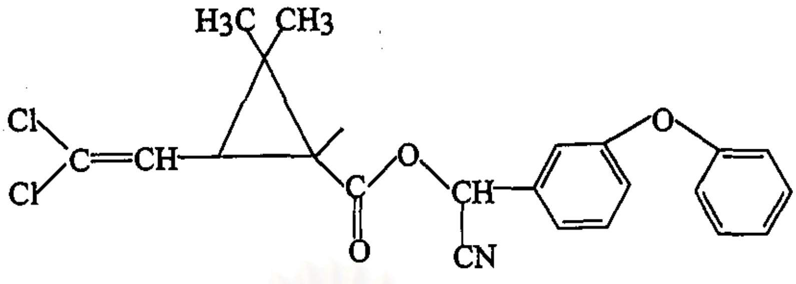


Carbaryl

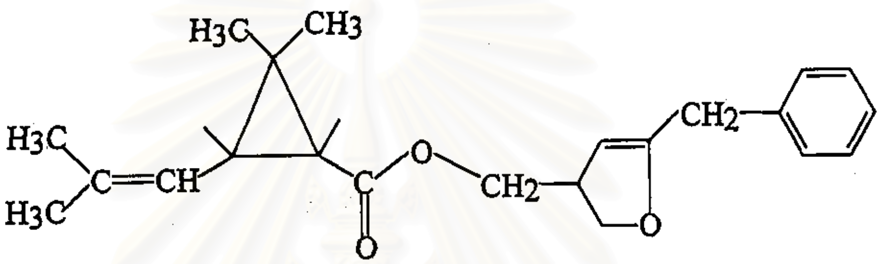
Some structures of carbamate compound

4. Synthetic pyrethroids

These are being developed as an alternative to the naturally occurring pyrethrums whose main drawback is its short life as an effective chemical. The modern chemical analogues are, however, more and more removed from the original pyrethrins. They do have the same spectacular "knock-down" effect and the broad action, but they are more stable and far more persistent. They also have a higher mamalian toxicity. These chemicals include cypermethrin, deltamethrin, resmethrin and fenvalerate, and the more recently developed bioallethrin, permethrin and tetramethrin.[4, 6, 9]



Cypermethrin



Bioresmethrin

Bioinsecticide

It was originally thought that these natural insecticides would not induce resistance in the target pest, but it is now known that this idea is not true: some resistance has been established, although less than that shown with the synthetic chemicals. The main source of these biological insecticides are categorized into 2 groups:[4]

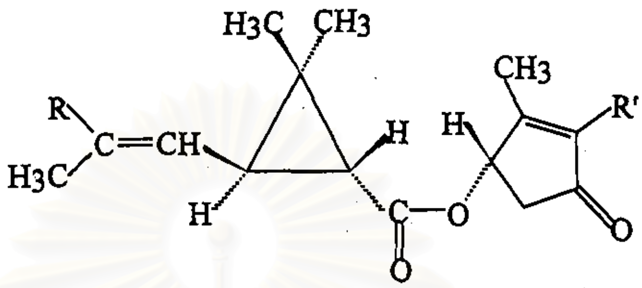
1. Insecticide from plant extracts

Plant extracts with toxic effects have been known since time immemorial in idea and other parts of tropical Asia. In the last couple of decades research interest in the use of plant extracts for plant and food protection has escalated, as well as the use of some of these chemicals to kill mosquitoes and other medical and veterinary pests. More than 2,000 species of plants are known to produce chemicals that have insecticidal properties.[5]

1.1 Pyrethrum

It is the most famous insecticide from plant. The crude extract from the flower head is called pyrethrum (which is also the trade name for retailing). There are 6 compounds from pyrethrum combinding pyrethrin I, II, cinenin I, II, jasmolin I and II.

This is a powerful contact insecticide which causes a rapid paralysis (knock-down), but is unstable to sunlight and rapidly hydrolyzed.[9]

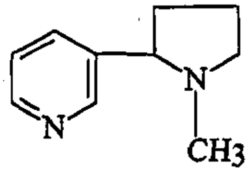


Basic structure

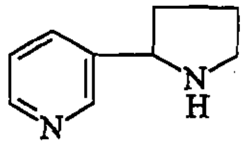
R	R′	
CH_3	$-CH_2-CH=CH-CH=CH_2$	pyrethrin I
$COOCH_3$	$-CH_2-CH=CH-CH=CH_2$	pyrethrin II
CH_3	$-CH_2-CH=CH-CH_3$	cinerin I
$COOCH_3$	$-CH_2-CH=CH-CH_3$	cinerin II
CH_3	$-CH_2-CH=CH-CH_2-CH_3$	jasmolin I
$COOCH_3$	$-CH_2-CH=CH-CH_2-CH_3$	jasmolin II

1.2 Tobacco

Nicotin that found in tobacco has been used as insecticide in Europe since 1790. The substances in nicotine group have been occurred in other plants that exhibited insecticide such as nornicotine extracted from *Duboisia hopwoodii* and *Nicotiana sylvestvis* with anabasine investigated in *Anabasia aphylla* and *Nicotiana glauca.*[4]



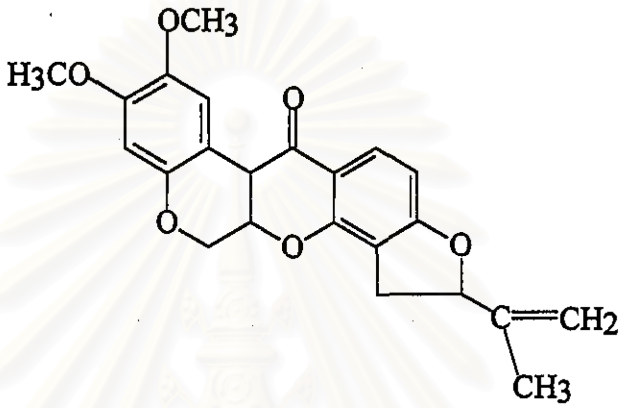
Nicotine



Nornicotine

1.3 Derris

Rotenone and rotenoid have insecticidal activities that found in roots of many genus plants in Papilioneae family, more particular in Derris genus which have much and wide spread in Southeast Asia.[4]



Rotenone

1.4 Neem

In India the multitodinous use of neem (*Azadirachta indica*) has long been known. In the last two decades the interest has become worldwide, with several international conferences on the subject of using extract of neem as insecticides. In the poorer regions of the tropics there is great interest in the use of indigenous plant materials as insecticides, and extensive research is now being conducted into these convenient and cheap alternatives to expensive synthetic chemicals. There are more than 30 compounds investigated which have prevention and control insect activities in different way including repellent, antifeedant and growth regulator. The most insecticidal activity compound is azadiractin (AZ) that found particularly in seed kernel of neem. The usage of neem to prevent and control insect has 3 forms combinding neem oil, neem extract and neem cake. The neem extracted compounds revealed 7 activities to prevent and control insect as antifeedant, repellent, growth regulator, antioviposition, inhibit chitin formation, disrupt reproduction and reproductive communication and cause the worm cannot swollow or ingest food.[4, 10, 11]

2. Insecticide from micro-organism

2.1 Bacterias

At least four species of *Baillus* are entomorphagous. *B. thuringiensis* (Bth) has long been known to be a killing agent against caterpillars, but several distinct varieties are now known to be effective against some Diptera and some Coleoptera. This bacterium is being further exploited by introducing into different plants so that the plant tissues contain the insecticidal toxin. It should perhaps be stressed that Bth does not harm other insects, spiders, or vertebrates; it is very host-specific. Research on entomorphagous bacteria has now become intensive and more than 50 different toxins have been isolated, effective against a wide range of insect and mite pests.[5]

2.2 Viruses

Viruses belonging to several different groups cause disease in insect, but only those from two families appear to be really suitable. The granulosis viruses (GV) and the nuclear polyhedrosis viruses (NPV) are both Baculoviridae, and the cytoplasmic viruses (CPV) belong to the family Reoviridae. Most entomorphagous viruses show very definite host-specificity and a number of them has been commercially developed for use against particularly serious crop pest-these include Codling Moth GV, Cabbage White Butterfly GV and *Neodiprion* PHV.[5]

2.3 Fungi

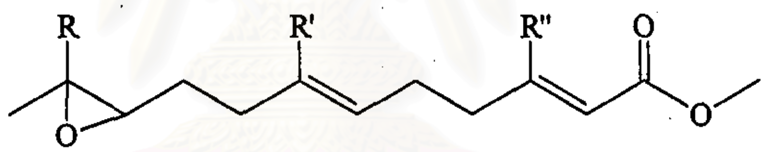
Some species of fungi produce natural antibiotics that have been used against plant diseases for a long time. It has been recently shown that entomorphagous fungi can be used for pest control. It is likely that the future may see great development in this field. One major problem, however, is that fungi need high humidity for survival and reproduction , which may prevent their exploitation on field crops.[5]

Insect growth regulator

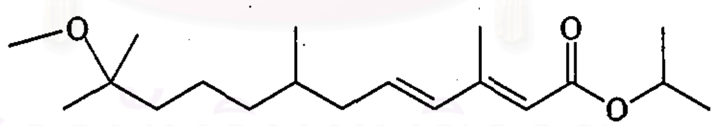
These insecticidal groups act as killing insect which are different from other general insecticides. This type of chemicals does not kill insect immediately, but affects in inhibiting the growth of insect particularly nymph or worm not to continue growing and insect will die eventually. These chemicals maybe called morphogenetic agent. These chemicals combinding:

1. Juvenile hormone mimic

These are the synthetic terpenoid type chemicals which have the same characteristic like juvenile hormone of insect. These compounds cause worm not grow to adult and still continue molting to be the worm. Some have abnormal growth and will die at the end. This group of chemicals has been sold as commercial products such as methoprene (Altosid).[4, 12]



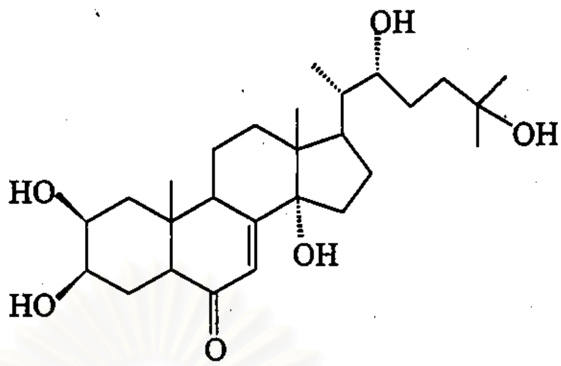
JH-OII



Methoprene

2. Ecdysone hormone mimic

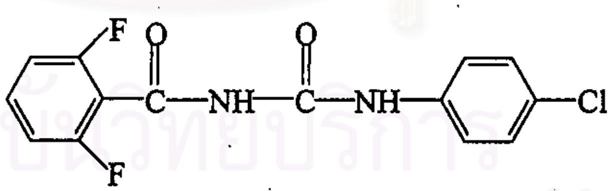
These chemicals have similar characteristic to ecdysone hormone of insect which causes insect growing abnormally to become adults before reproductive organs would have been developed. These mentioned chemical characteristic is derived from some plants. The synthesis of commercial products of this group are now operating.[4,13]



Ecdysone

3. Chitin synthesis inhibitor

This group of chemicals is urea analog (benzoyl phenyl urea). They can inhibit chitin formation by transferring digestion working of chitin synthase. By this mean, UPD-N-acetyl-glucosamine cannot bind to be long chain compound, chitin. Therefore, the worm cannot continue growing and will die while molting. These chemicals are produced in commercial in many aspects such as diflubenzuon, triflumuron and provided good properties to prevent and eliminate caterpillar, weevil and fly. However, according to the cost, they are rather expensive which caused the usage not very wide spread.[4, 14]

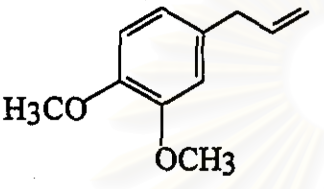


Diflubenzuon

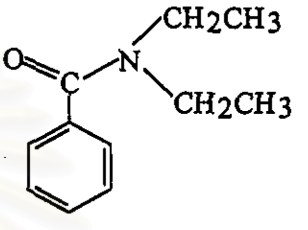
These chemicals are less toxic to humans, animals and environment which cause no problem regarding to accumulated toxicity or residual effect.

Insect behavioral chemicals

This refers to the chemicals that affect the behavioral control of insect, for example attractant or sex pheromone mimic such as methyl eugenol, repellent such as DEET or antifeedant such as cyhexatin which will illustrate further.[4, 15]



Methyl eugenol



DEET

From literature review, it could clearly see that most synthetic insecticides could be employed successfully; however many serious problems took place afterwards. A particular serious problem is that insecticides can cause eradication of any animals while all over the world calls for conservation of biodiversity. There is agreement that the eradication of any pest (any species) is a grave biological responsibility and that it should not be done unless really necessary, for the ecological consequences cannot always be predicted.[5, 16] There is increased public and government interest by means of managing population of pest insects with reduced amounts of insecticides in order to limit the impact on the environment and human health. One of the widest approaches to alternative insect controls would come from the use of the knowledge on natural products to influence insect chemosensory behavior as attractants, repellent and antifeedant.[16] Recently, antifeedant is the most interesting perspective to insect pest control approach and more increasing interest that showed to be successful method.

According to the above mentioned statement that there is a need to develop more ecologically acceptable methods of insect pest control. For this reason, plants which display behavior of modifying properties, such as antifeedant that deters insect from feeding, are becoming increasingly important. The practical use of natural product feeding inhibitors in insect control is rapidly becoming a reality.

Insect antifeedant chemistry

Recent advances in the isolation, structural determination and synthesis of active partial structures of antifeedants have allowed the scientists to perform some detailed studies of modes of action.[15]

The term antifeedant is defined as a chemical that inhibits feeding but does not kill the insect directly, the insect remaining near the treated leaves and dying through starvation. Gustatory repellent, feeding deterrent and rejectant are synonymous with antifeedant.[17] The feeding behavior of insect can be divided into four steps : (1) host plant recognition and orientation, (2) initiation of feeding, (3) maintenance of feeding and (4) cessation of feeding. Antifeedant is concerned with steps (2) and (3).

The compounds that would be acting as relative antifeedants are the compounds that inhibit feeding only for a defined time. Although a number of plant-derived compounds exhibit absolute antifeedant activity, the insects die from starvation rather than start eating treated foodstuffs. It is therefore desirable that a candidate antifeedant should have some toxic action when ingested. Studies of the natural phenomenon of antifeeding (no consumption of plant material by insect) could reveal new antifeeding substances from plants, and may provide correlations between chemical structures and antifeeding activities.[15, 18, 19, 20]

The insect pest specificity of antifeedant compounds maybe modifiable; that is, the range of species targeted maybe defined either by the chemical structure of the antifeedant or by the composition of the mixture of antifeedant if different feeding inhibitors are effective against species within the range. For example, polygodial is a selective aphid antifeedant. Therefore, the revealed antifeedant activity of substances depends on the insect species. The specifying antifeedant compounds have told what insect species is against. However, most antifeedant compounds can exhibit activity to affect more than one species but not in broad-spectrum. For example, even though azadiractin has broad-spectrum of antifeedant activity; effective against many insect species but not for all. For example, the Colorado potato beetle, *Leptinotarsa decemlineata*.[15, 18, 19]

The existence of potential antifeedant compounds, both natural and synthetic, in practical insect control has been known for some time. For example, Bordeaux mixture (copper sulfate, hydrated lime and water), known for over 100 years, acts as a feeding deterrent to flea beetles, leaf hoppers and the potato psyllid (*Paratrioza cockerelli*).[21] In 1930's, there are some field trial of antifeedant compounds, including some derived from plants but the success was limited and was not sufficient to compete with synthetic insecticides. Recently, the problems associated with total reliance on synthetic insecticide necessitate the re-evaluation of antifeedant compounds.[15]

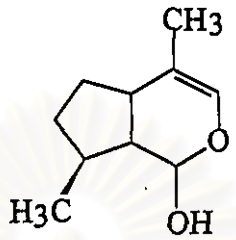
Feeding deterrents are usually nonvolatile and unstable molecules, and hence their action maybe transient both environmentally and within the pest. This offers wide latitude in the choice of delivery routes, as well as combinations with other compounds to develop pest selectivity while ensuring minimal environmental impacts. Some new approaches to understand the mode of action indicate that a great knowledge of the chemical and molecular basis of action can expand our control option.[15] Antifeedant compounds may affect some of the beneficial insects depending upon the compound or dose, requiring further development of its actual field use delivery system. Because many feeding inhibitors are transient chemicals acting rapidly on affected insect, their residual activity and environmental impact maybe very limited, although much remains to be done with formulations or mode of delivery.[16, 20]

Insect antifeedant compounds have various types of substances. They could be classified according to types of natural products as

1. Terpenoid

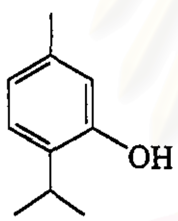
1.1 Monoterpenoid

Iridoids potentially act as deterrents or are toxic to a variety of generalist insect herbivores and have been shown to act as antifeedants to grasshoppers and lepiolopteran larvae. The deterrency or physiological toxicity is dosage dependent. Higher dose of iridoids inhibits insect growth.[22]

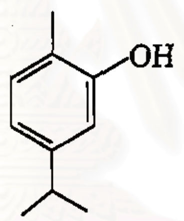


Iridoid

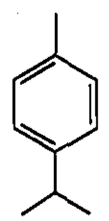
Other monoterpenoids such as thymol, carvacrol, *p*-cymene are insect repellent.



Thymol



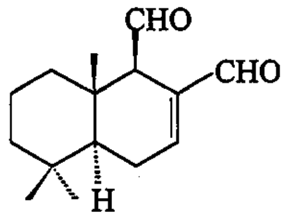
Carvacrol



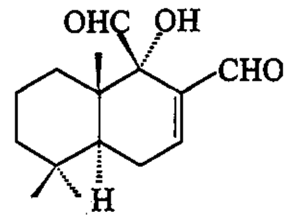
p-Cymene

1.2 Sesquiterpenoid

Drimane skeletal types of sesquiterpenes are the most potent insect-feeding deterrents, including warburganol and polygodial. These drimane dialdehydes inhibit the feeding of army worms *Spodoptera exempta* and *Leptinotarsa decemlineta* as well as the aphid *Myzus persicale*. The feeding inhibition activity of these sesquiterpenes against certain lepidoteran larvae seems to be due to the blocking of the stimulatory effects of glucose, sucrose and inosital on chemosensory receptor cell located in insect mouth parts.[13, 22]

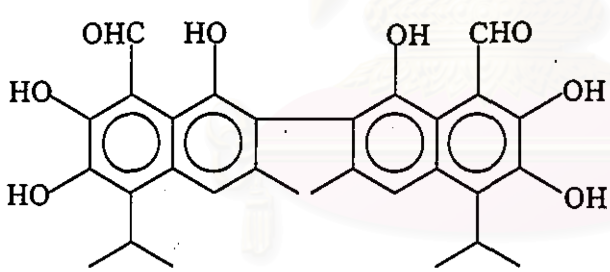


Polygodial

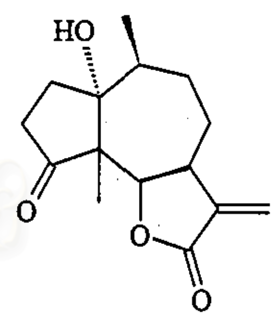


Warburganol

Other antifeedant sesquiterpenoids are sesquiterpene lactones including parthenin against *Dysdercus koenigi* (pyrrhocorid bug) and *Tribolium castaneum* (red flour beetle). Maximilin C was found to be an antifeedant compound against *Spodoptera eridania* (southern army worm), *Melanoplus sanguinipes* (Migratory grasshopper) and *Homoeusoma electellur* (sunflower moth). Gossypol (dimeric sesquiterpenoid) is against blister beetle and spring bollworm.[22]

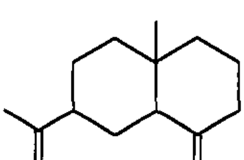


Gossypol

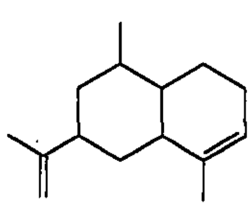


Parthenin

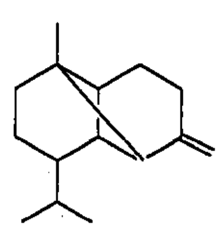
Several sesquiterpenes including caryophyllene, α-selinene, β-selinene and β-copaene are feeding deterency against *Spodoptera exigua* (beet army worm).



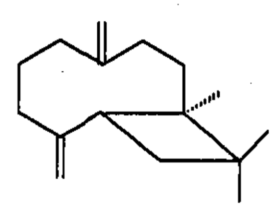
β-Selinene



α-Selinene



β-Copaene



Caryophyllene

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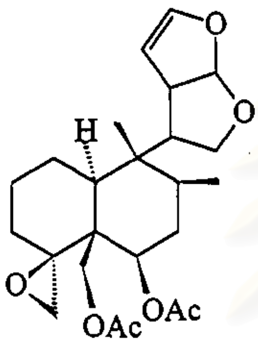
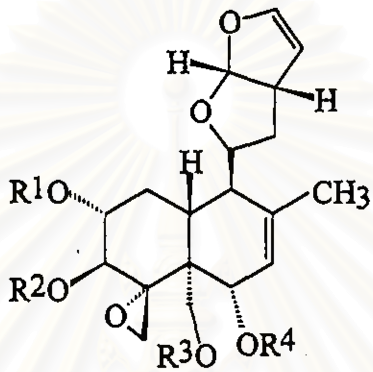
1.3 Diterpenoid

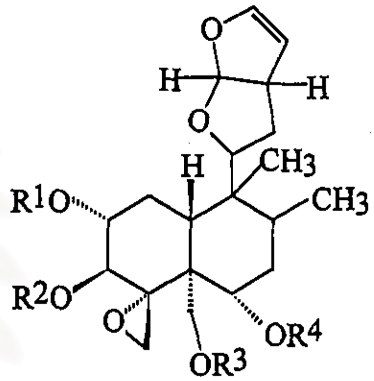
Clerodane diterpenes such as clerodin and ajugarin have been demonstrated to inhibit the feeding of a number of species of lepidoteran larvae. Clerodendrins A and B were reported to inhibit feeding of *Spodoptera litura*, *Euproctis subflava* and *Ostrinia nubilalis*.[17]



Clerodin



Clerodendrin A



Clerodendrin B

$R^1 = H$ $R^3 = R^4 = Ac$

$R^2 = C_2H_3-C(CH_3)(OAc)-CO-$

Kaurane diterpenes have previously been found to deter feeding of lepidopteran larvae and aphids and many of them have a rather broad spectrum of activities, and represent an interesting new class of active compounds.[23]

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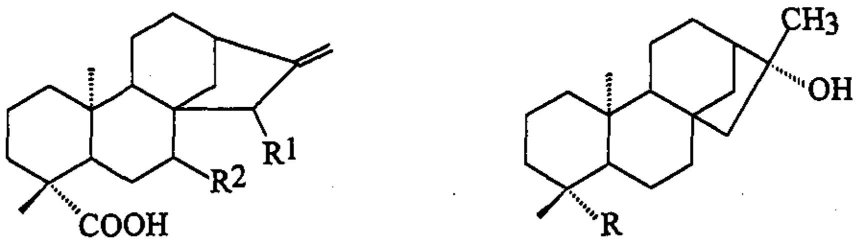


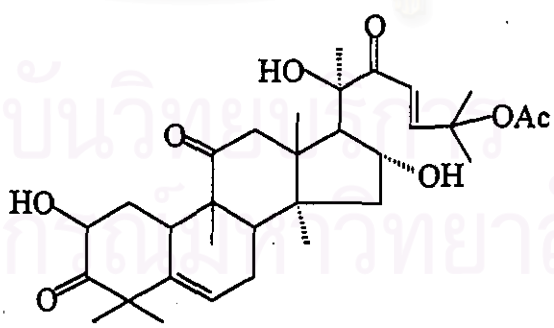
Kaurane diterpene

	R^1	R^2		R
A =	H	OAc	C =	CH_3
B =	H	H	H =	CH_2OH
E =	OAc	H		
G =	O	H		

1.4 Triterpenoid

Some triterpenes possessing antifeedant are cucurbitacins, limonoids and saponin.

Cucurbitacins are feeding deterrents for a number of arthropods including cucumber leaf beetles *Phyllotreta* spp., *Phaedon* spp. and *Cerotoma trifuaata*; stem borer *Margonia hyalinata*, and red spider mites. Cucurbitacin B is the predominant compound in nature and acts as powerful activity even at very low concentration.[22]



Cucurbitacin B

Limonoids are a major tetranortriterpenoid class; the best well known and highly effective activity is azadirachtin from the neem tree *Azadirachta indica* (Meliaceae). This compound, first isolated by Butter worth and Morgan, is effective

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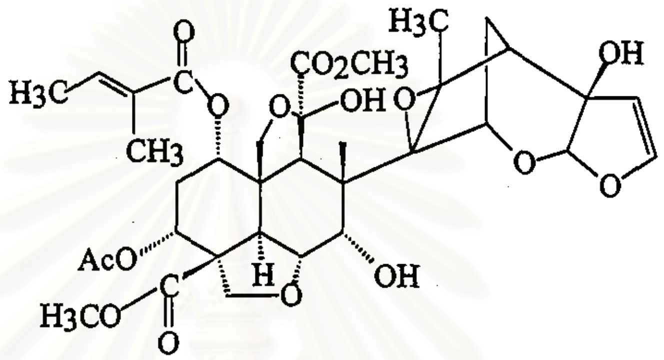
at dose as low as 0.1 ppm. In addition, it has been shown to act as feeding deterrent to more than 200 species of herbivore insects. Recent advances in azadirachtin research relate to a new field trial data, using commercial and semi-commercial preparations of neem, against phytophagous insect and their associated beneficial, investigations against stored product pest and also insect vectors of disease.[18, 19]



Azadirachtin

Azadirachtin exhibits activity in two ways (1) physiological pest control agent (growth inhibition) and (2) behavioral pest control agent (inhibit feeding and oviposition). The scientist has proposed that azadirachtin is composed with 2 sub-molecules combinding the section that shows antifeedant and oviposition function and the part that shows insect growth inhibition function.[10, 24] Both effects have been noted in numerous insect species. Antifeedant effects are more species-specific than growth inhibition. In other words, concentrations of azadirachtin required to prevent feeding vary widely among species, whereas concentrations that stop growth and disrupt molting are less variable. Antifeedant effects of azadirachtin are partly due to sensory detection and avoidance by insect.[20]

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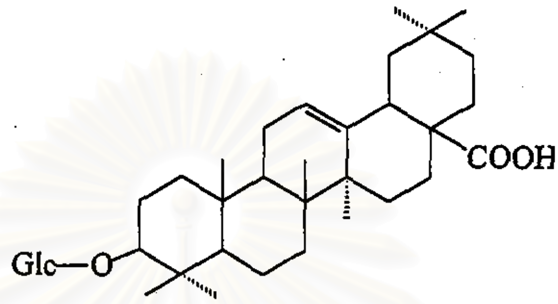
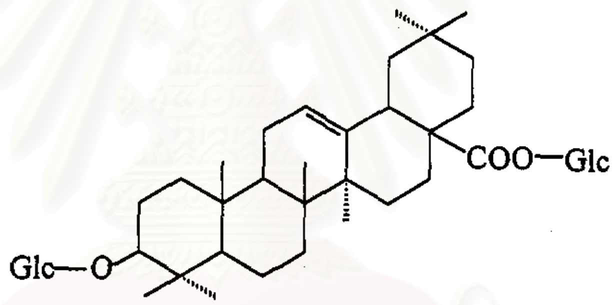
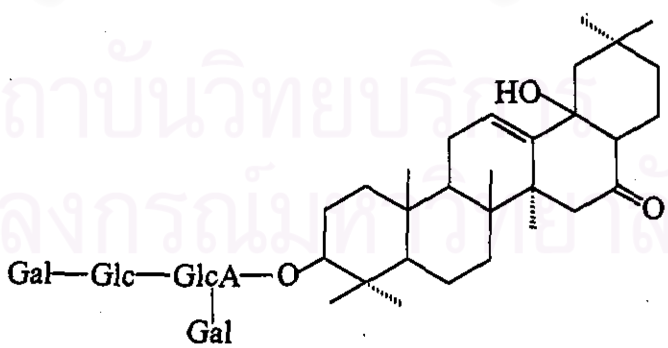
Saponins have been shown to act as antifeedant to species of mites, lepidopterans, beetle and many other insects. Mono- and bidesmosidic saponins repel termites, probably because of the presence of enzyme in the termite 's digestive tract which converts the bidesmoside into monodesmoside. Camellidin II has antifeedant activity against 5th instar larvae of the yellow butterfly Eurema hecade mandarina. Glycosides of medicagenic acid also appear to exhibit the highest degree of feeding inhibition. The steroidal alkaloid saponins are especially well known as insect antifeedants; solanine and chaconine, both steroidal alkaloids and their glycosides act as strong termite antifeedant.[25]



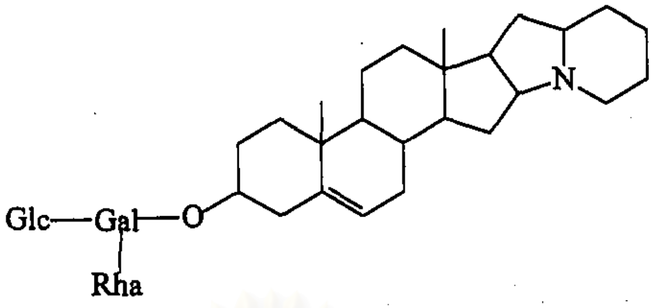
Monodesmosidic



Bidesmosidic



Camellidin II



Solanine

Rha-Glc-O- = Chaconine

Causing activity seems to be due to the binding of saponins to free sterols in the gut, thereby reducing the rate of sterol uptake into the hemolymph. In reducing the sterol supply, saponins could interfere with the insect molting process.[22]

2. Aromatic compounds

2.1 Phenolic compounds

Phenolic compounds such as aristolochic acid I and IV derived from *Aristolochia albida*, showing extremely strong feeding-deterrency against *S. litura*. Although aristolochic acids by themselves are known to be cytotoxic, they are also among the most potent antifeedant compounds known, approaching the efficiency of neem, against *Spodoptera* larvae. Anthranilic acid, *trans*-cinnamic acid and *trans*-cinnamaldehyde are antifeedant compounds against *Anthonomus grandis*, *Heliothis virescens* (tobacco bud worm). Capillin and methyl eugenol are showed antifeeding activity to cabbage butterfly, *Pieris rapae cruiuora*.[22, 23]

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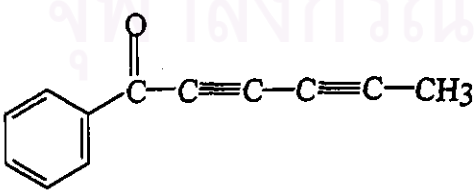
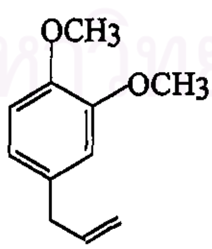
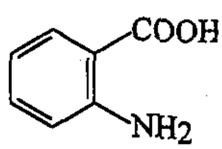


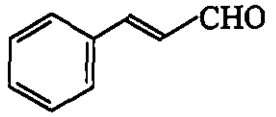
Capillin



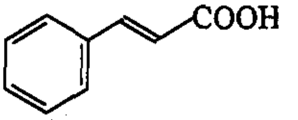
Methyl eugenol



Anthranilic acid



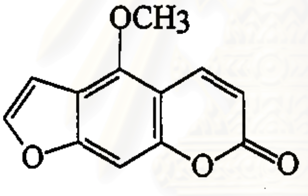
trans-Cinnamaldehyde



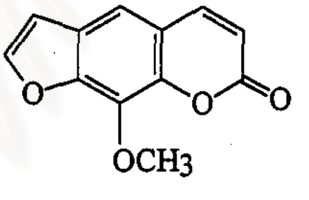
trans-Cinnamic acid

2.2 Coumarins

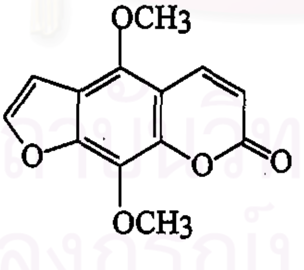
Coumarins have been also displayed antifeedant activity such as against vegetable weevil and 2-3% coumarin against alfafa weevil and sweet weevil. Dicoumarols have inhibited feeding pea aphid. Furanocoumarin is another type of coumarin and only linear furanocoumarin displayed potent antifeedant activity. Bergapten, xanthotoxin, isopimpinellin and oxypeucedanin four furanocoumarins with antifeedant activity against *S. litura* larvae (tobacco cutworm).[22, 23]



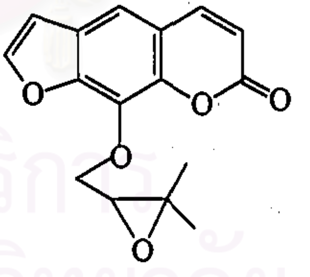
Bergapten



Xanthotoxin



Isopimpinellin



Oxypeucedanin

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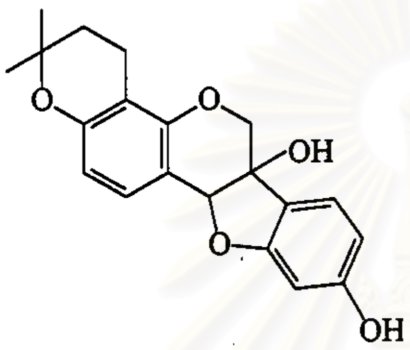
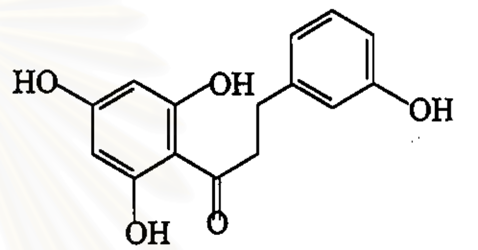
2.3 Flavonoids

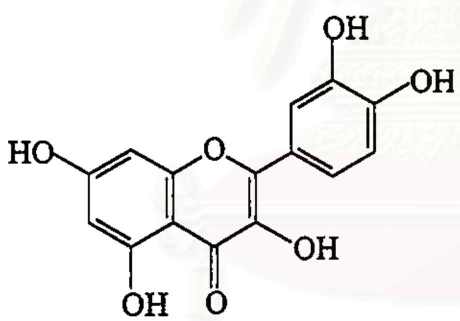
Some flavonoids have beeen shown antifeedant activity including quercetin and phloretin against *Scolytus multistriatus*, rutin against *Helicoverpa zea*. Glyceollin has strong antifeeding activity against the Mexican bean beetle (*Epilachna verivestis*).[22]



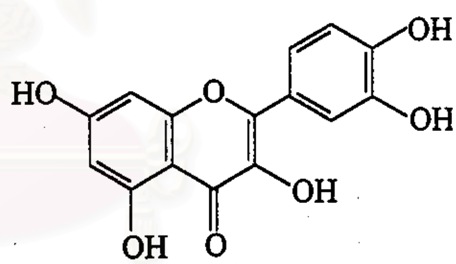
Glyceollin



Phloretin



Quercetin



Rutin

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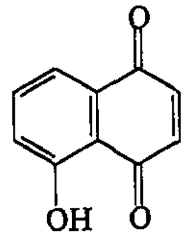
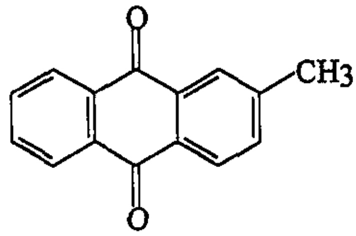
2.4 Quinones

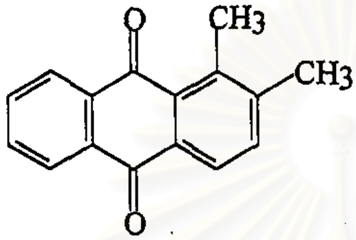
The simplest naphthoquinone, juglone (5-hydroxy-1,4-napthoquinone) is antifeedant to small Europe elmbark beetle, and β-benzoquinone inhibits feeding of *Scolytus multistriatus*. Anthraquinone such as 2-methyl anthraquinone and 2-formyl anthraquinone are antifeedant to termites.[22]



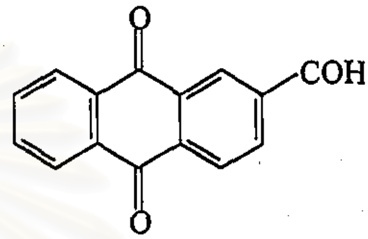
Juglone



2-Methyl anthraquinone



2-Hydroxy methyl anthraquinone



2-Formyl anthraquinone

Some scientists have argued that deterrency is not enough to prevent insect feeding reliably under field conditions, and antifeedants would need to be toxic to be practical and some literatures reported that some insecticides at low concentration can act as antifeedant. In mode of action of antifeedant has various routes of mechanism; in majority including smell and taste but not exactly, sometime involved their toxicity to physiology that causes insect to avoid and feeding is inhibited.[16, 20]

Finally, use of feeding inhibitors in pest management enables to exploit natural defense evolved by plants and other organisms in novel ways and thereby reduce the risks associated with synthetic insecticides.

1.2.2 Information about *Trigonostemon reidioides* Craib.

Trigonostemon reidioides Craib. belongs to Euphorbiaceae family and found predominantly in north eastern of Thailand. In Thai name this plant is generally called Lot thanong daeng, and has many local names according to local areas such as Lot thanong (Ratchaburi, Prachinburi and Trad), Khaao yen noen (Ratchaburi and Prachuap khirikhan), Duu bia and Duu tia (Phetchaburi), Thanong and Rak thanong (Nakhon ratchasima), Thanong daeng (Prachuap khirikhan), Naang saeng (Ubon ratchathani) and Naat kham (Northern). This plant was believed as one of medicinal plants and was used as drug from the ancient.[26]

Botanical description

Trigonostemon reidioides Craib. is a small undershrub 0.5-1.5 m high; all parts pubescent. Leaf: simple, alternate, oblong or oblonglanceolate, 2-4 cm wide, 7-12 cm long; finely, pubescent on both surfaces. Inflorescence in auxillary or ramiflorous panicle, unisexual, monoecious flowers: white, pink or purple. Fruit: capsule, 3 lobed subglobose.[27]

Ethnomedical uses

Root: grind with water, taken as laxative, antiasthmatic, emetic for food poisoning; especially from mushroom and shells; treatment of bloody and mucous sputum or stools; topically apply to abscesses, sprains, swelling, bruises, snake bites, especially to treat snake neurotoxin.[27]

From literature review, it was found that Udom Kokpol *et al.* were only the group who has studied by focusing on only chemical constituents in the root of this plant, particularly in non-polar and slightly polar parts. The chemical constituents are concluded in Table 1.1.

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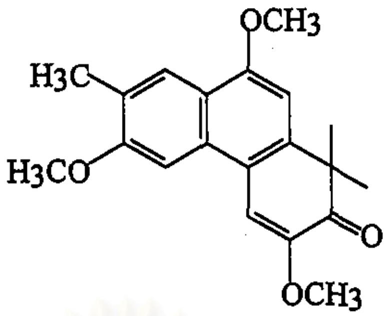
Table 1.1 The chemical constituents derived from *Trigonostemon reidioides* Craib.

Solvent	Chemicals	Reference
Hexane	The mixture of steroid palmitate (β-sitosteryl palmitate, stigmasteryl palmitate, campesteryl palmitate and cholesteryl palmitate) The mixture of long chain acid (C_{16}-C_{35}) The mixture of steroid (β-sitosterol, stigmasterol and campesterol) acetyl aleuritolic acid 1,1,7-trimethyl-3,6,9-trimethoxy-2-phenanthrenone (Trigonostemone) 5α-stigmastane-3,6-dione	28
CH_2Cl_2/ $CHCl_3$	acetyl aleuritolic acid Trigonostemone 5-hydroxy-6,7-dimethoxy coumarin The mixture of long chain amide (C_{44}-C_{48}) The mixture of steroid glycoside (β-sitosteryl-3-O-glucopyranoside, stigmasteryl-3-O-glucopyranoside and campesteryl-3-O-glucopyranoside)	28
Water	K, Ca, Cl, S, P, glycine, threonine, alanine, isoleucine, γ-aminobutyric acid, argenine, glucose, fructose, arabinose, rhamnose	28



1,1,7-trimethyl-3,6,9-trimethoxy-2-phenanthrenone

(Trigonostemone)

From the collaboration program in searching for biologically active compound of natural product research unit, Department of Chemistry, Faculty of Science, Chulalongkorn University and Department of Biology, Faculty of Science, Chulalongkorn University to preliminary screening test of many Thai medicinal plants and Thai tropical weeds, it was discovered that the ethanol extract from *Trigonostemon reidioides* Craib. exhibited high degree of antifeedant activity against Greater wax moth (*Galleria mellonella*).

Objective of this research

To extract, isolate and investigate chemical constituents from the root of *Trigonostemon reidioides* Craib., particularly in more polar part which there was no report concerning with this part before and to search for insect antifeedant compounds.