CHAPTER IV
CONCLUSION

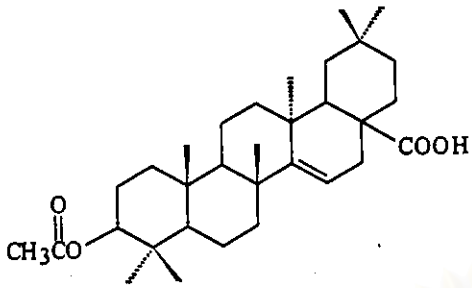
During the course of this research with the aim to search for active insect antifeedant from Thai medicinal plants and tropical weeds, it was found that the ethanolic extract of the roots of *Trigonostemon reidioides* Craib. belonging to Euphorbiaceae family showed highly active insect antifeedant against Greater Wax Moth *Galleria mellonella*. After fractional extraction following the polarity of solvent, the crudes hexane and dichloromethane revealed high insect antifeedant activity. These two fractions are selected for further investigation for their chemical constituents and searching for active principal possessing insect antifeedant activity. By means of physical properties and spectroscopic data, six components isolated from the crude hexane were characterized as a mixture of steroidal esters, a mixture of long chain acids, a mixture of β-sitosterol, stigmasterol and campesterol, acetyl alueritolic acid, Trigonostemone and 5α-stigmastane-3,6-dione. Three additional compounds were isolated from the crude dichloromethane and were identified as 5-hydroxy-6,7-dimethoxy coumarin, a mixture of long chain amides, a mixture of β-sitosteryl, stigmasteryl and campesteryl glycoside. Among isolated compounds, it was disclosed that acetyl alueritolic acid and a mixture of steroids exhibited highly active insect antifeedant.

All isolated substances from both active crude fractions of the roots of *Trigonostemon reidioides* Craib. could be summarized as shown in Table 4.1 and Fig. 4.1.

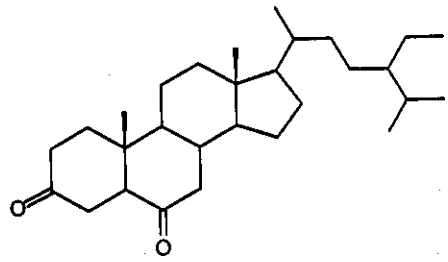
Table 4.1 All isolated substances from active crude fractions of the roots of *Trigonostemon reidioides* Craib.

Compound	Weight (g)	% Insect antifeedant activity (0.25 wt/wt)	Level of activity
acetyl aleuritolic acid (**A**)	2.10	83.15	high
mixture of steroid (**E**)	1.44	74.02	high
5α-stigmastane-3,6-dione (**B**)	0.64	53.54	medium
Trigonostemone (**F**)	0.54	53.36	medium
5-hydroxy-6,7-dimethoxy coumarin (**G**)	0.62	52.65	medium
mixture of long chain amide (**H**)	0.20	49.83	medium
mixture of steroidal glycoside (**I**)	0.98	48.25	medium
mixture of long chain acid (**D**)	1.39	46.38	medium
mixture of steroidal ester (**C**)	1.09	-33.52	attracted

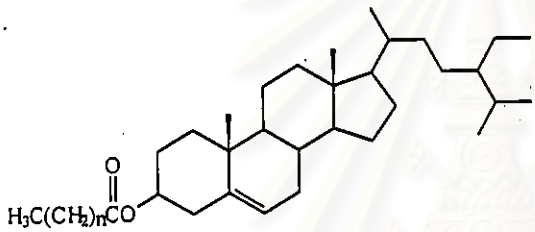
The major component and active ingredient of this plant was found to be acetyl aleuritolic acid and a mixture of steroid. From preliminary study on structure activity relationship, it could be hypothesized that pentacyclic triterpenoids containing an acetyl group at carbon position 3 could extremely promote the insect antifeedant activity.



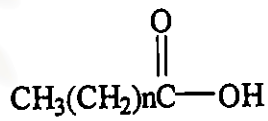
Compound A



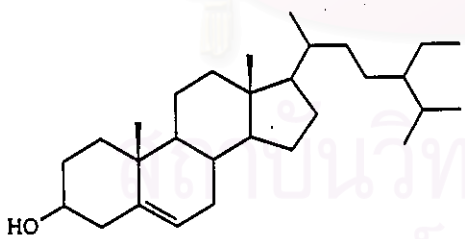
Compound B

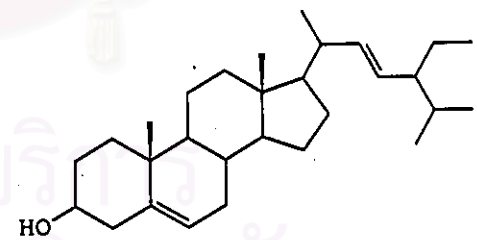


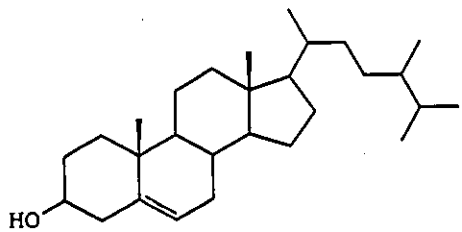
Mixture C



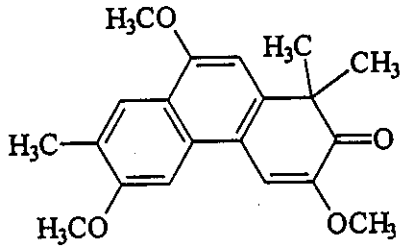
Mixture D



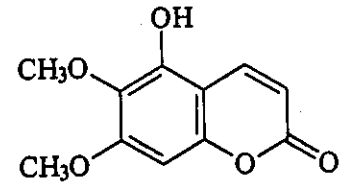




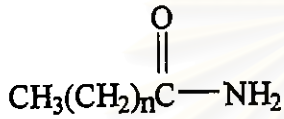
Mixture E



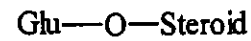
Compound **F**



Compound **G**

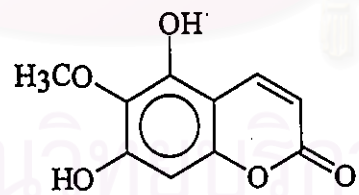


Mixture **H**



Mixture **I**

In addition, the crude ethyl acetate was studied for its chemical constituents for the first time of this plant, at least two compounds were elucidated their structural formulae as 5-hydroxy-6,7-dimethoxy coumarin and 5,7-dihydroxy-6-methoxy coumarin. The latter has not been previously reported in literature before.



5,7-dihydroxy-6-methoxy coumarin

In conclusion, it could obviously be seen from this work that the search for active principles by utilizing bioassay test as a guide would ultimately lead to the desired products. The combined knowledge of the chemistry and biology including SAR study would permit one to discover the useful compounds for specific purposes.

Proposal for the Future Work

For the next century trend, the insect antifeedant topic will certainly be still a hot issue and needed for more practical cooperation. The natural origin-derived compounds would be fundamentally important models for synthetic point of view. *Trigonostemon redioides* Craib. was proved to be among promising plants that exhibited this antifeedant activity. The outcome from this research provides many possibilities to carry on for further investigation. For instance, the ultization of active ingredients, acetyl aleuritolic acid and a mixture of steroids as an antifeedant agent against various lavae, particularly those that caused agricultural problems in Thailand should be seriously considered. The structure of an unknown coumarin should be fully confirmed, particularly by 2D-NMR spectroscopy. The close relationship of the new and isolated coumarins would provide another plausibility to carry on the structure and activity relationship study of naturally occurring and synthetic coumarins. The outcome from this proposed examination may lead to the disclosure of new and environmental friendly antifeeding agent. Another aspect that would make this research fulfill is the chemotaxonomic study on chemical constituents of other parts of *T. redioides*.