CHAPTER IV

CONCLUSION

In our search for agricultural-based bioactive compounds from *D. metel,* the ethyl acetate extract exhibited 100% plant growth inhibition on *Lactuca sativa* Linn. at concentration of 1.0 g and 0.5 g of weight corresponding to dried plant materials and revealed medium cytotoxicity against brine shrimp. Moreover the alkaloid fraction showed high molluscicidal activity against *P. canaliculata.* After fractionation and purification, five pure compounds and four mixtures were obtained. The structures of all isolated substances were elucidated by means of their physical properties, chemical reactions and spectroscopic evidences and are illustrated in Tables 4.1 and 4.2.

Table 4.1 Isolated substances from the ethyl acetate extract of *D. metel* flowers.

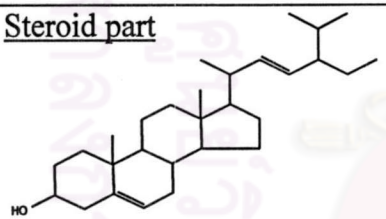
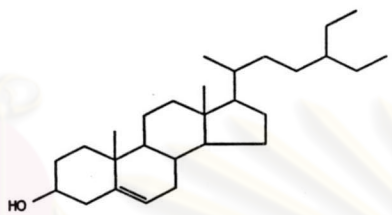
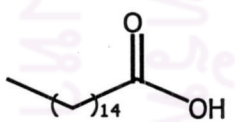
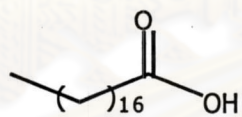
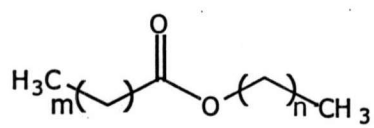
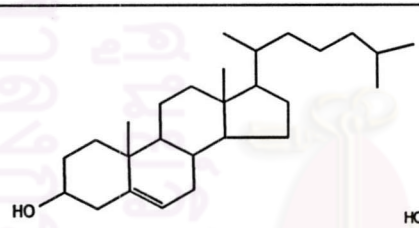
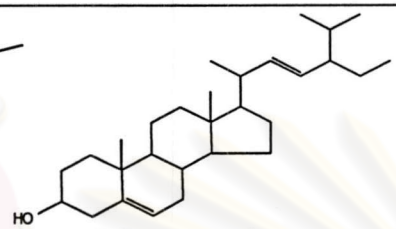
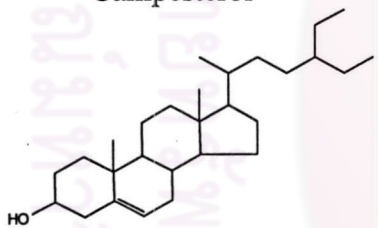
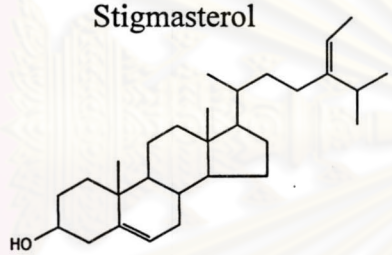
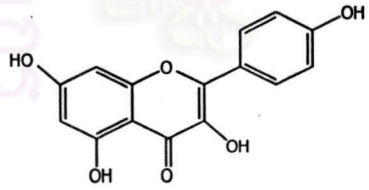
Substances	Structure	Weight (g)	% Yield*	Remarks
Mixture **1** Mixture of steroid ester	<u>Steroid part</u>  Stigmasterol  β-Sitosterol <u>Acidic part</u>  Hexadecanoic acid (palmitic acic)  Octadecanoic acid	0.49	0.22	White solid
Mixture **2** Mixture of long chain ester	 long chain ester	1.19	0.54	White amorphous solid

Table 4.1 (Cont)

Substances	Structure	Weight (g)	% Yield*	Remarks
Mixture **3** Mixture of steroid	 Campesterol  Stigmasterol  β-Sitosterol  Fucosterol	1.50	0.68	White powder
Compound **1** Kaempferol	 Kaempferol	0.03	0.014	Yellow small needle

* The percentage yield of isolated substances was calculated based on ethyl acetate crude extract (220.0 g)

Table 4.2 Isolated substances from alkaloid fraction of *D. metel* flowers.

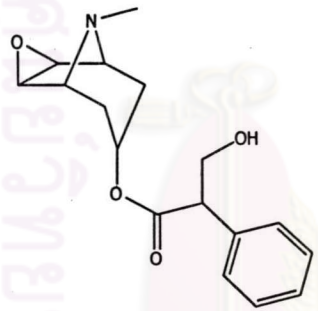
Substances	Structure	Weight (g)	% Yield*	Remarks
Compound **2** Scopolamine	 scopolamine	11.64	37.45	Yellow liquid
Compound **3** Tropine	 Tropine	0.063	0.02	Yellow liquid

Table 4.2 (Cont)

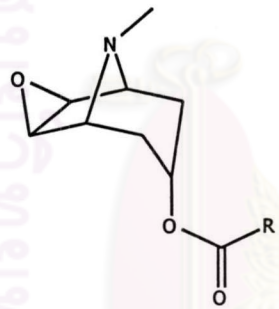
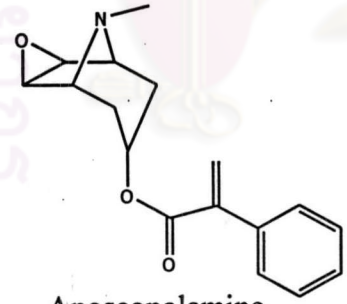
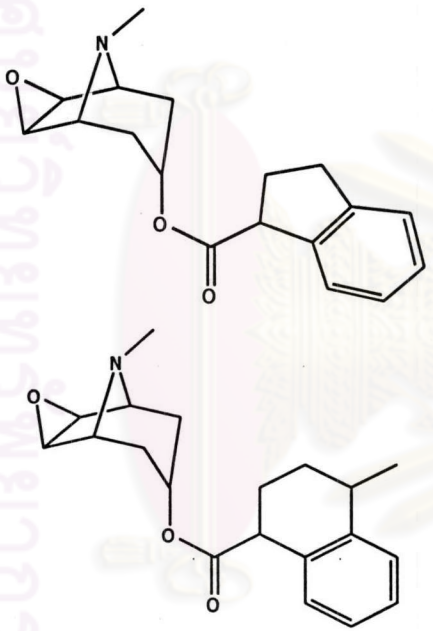
Substances	Structure	Weight (g)	% Yield*	Remarks
Compound **4**		0.002	0.006	Yellow liquid
Compound **5** Aposcopolamine	 Aposcopolamine	0.004	0.013	Yellow liquid

Table 4.2 (Cont)

Substances	Structure	Weight (g)	% Yield*	Remarks
Mixture **4**		0.0097	0.031	White solid

* The percentage yield of isolated substances was calculated based on alkaloid fraction (31.08 g)

All isolated substances were further bioassay. The bioactivity assay in this reseach involved molluscicidal activity against *P. canaliculata*, plant growth inhibition on *L. sativa*, cytotoxicity test against brine shrimp and insecticidal activity toward *S. litura*. The result of bioassay found that Compound **1** (kaempferol) revealed medium cytotoxic activity against brine shrimp *Artemia salina*, while Compound **2** displayed molluscicidal activity against *P. canaliculata* and root growth inhibition on *L. sativa*. Compound **3** showed highly active toward neonate larvae of *S. litura*.

According to the results of agricultural-based activity derived from the flowers of *D. metel,* scopolamine was a major compound in this plant that exhibited impressive result of molluscicidal activity and root growth inhibition. Moreover, tropine was found to reveal high insecticidal activity. Thus, it was manifestly seen that the utilization of *D. metel* was not only important in pharmaceutics, but also promising for agrochemical uses. In addition, from the results of cytotoxicity of alkaloid fraction, it did not exhibit toxicity to brine shrimp. This implied that the alkaloid fraction should not be toxic to mammal. The results obtained from this work strongly support the concept that naturally occurring compounds are still a good source providing numerous potential lead structures.

Proposal for future work

According to the bioassay results, scopolamine revealed satisfied molluscicidal activity toward *P. canaliculata*, as well as it exhibited highly active on the root growth inhibition of *L. sativa*. Furthermore tropine also disclosed as a highly active compound against *S. litura.* These informative data has not been reported in literature. Therefore, these compounds pointed out towards the possibly positive trend for utilizing these compounds in agriculture and might be further test for other activities such as antifeedant, insect attractant activity *etc.* The study on the structure activity relationship of scopolamine or tropine and their derivatives may provide an opportunity to understand what parts of the molecule have an influence for this activity. Moreover, the investigation of agrochemicals from *D. metel* could be used as a preliminary indicator for further study. For instance, the study on field test of scopolamine and derivatives as molluscicidal agents against *P. canaliculata* should be considered. The modification of scopolamine structures to water-soluble compounds *via* glycosylation or salt formation would be a great use in practical world. Various parts of *D. metel* should be comparatively investigated for both chemotaxanomy and

the amount of active ingredient. The outcome from these proposed studies would provide an excellent opportunity to reach the milestone for totally utilization of this particular plant species.