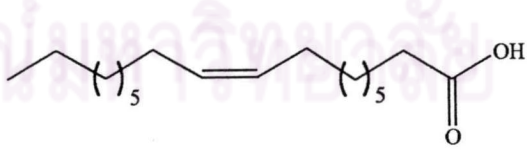
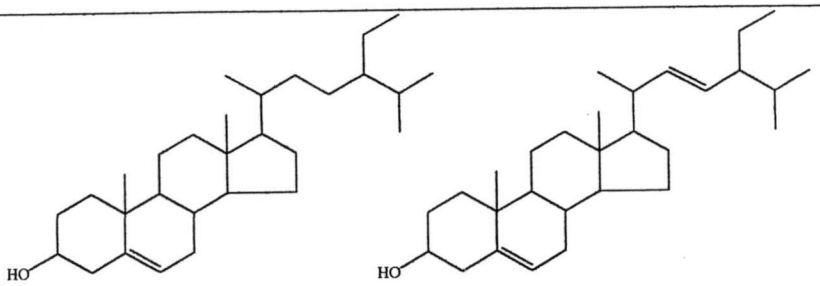
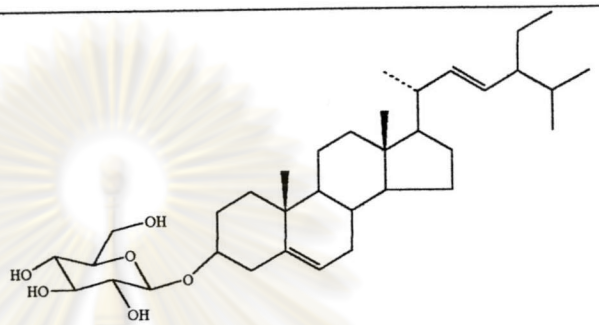
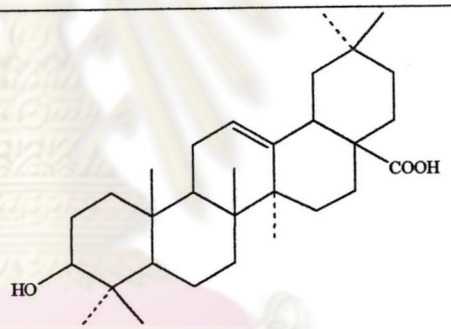
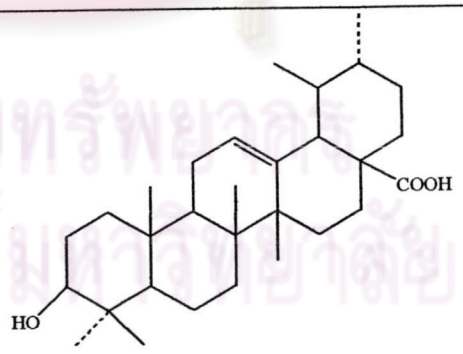
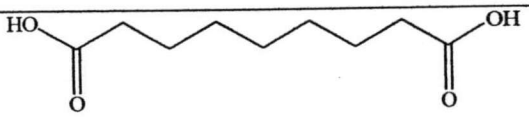
CHAPTER IV

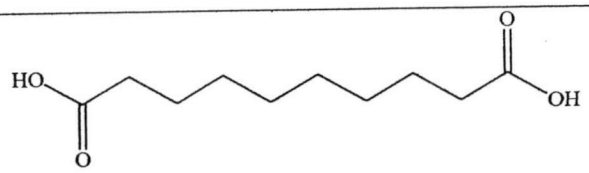
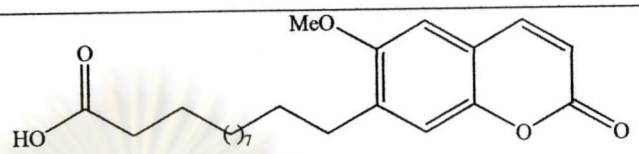
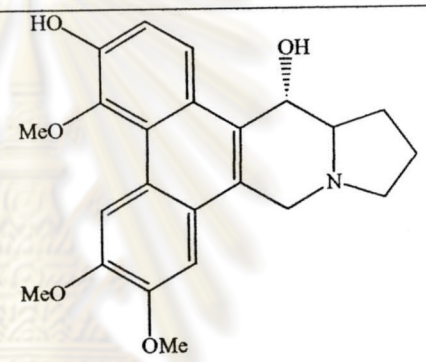
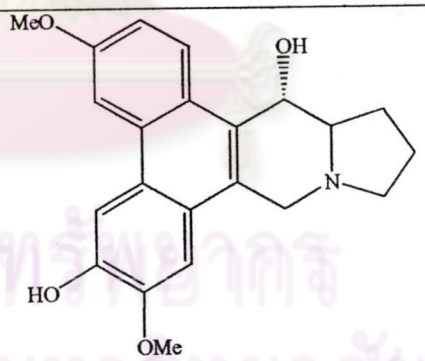
CONCLUSION

The preliminary results for plant growth regulators activity revealed that, the crude extract of the roots of *Tylophora indica* Merr. displayed significant activity. This plant was selected for chemical constituent studies. Further solvent fractionation and biological test exhibited that dichloromethane and ethyl acetate crude extracts showed the strongest root and shoot length inhibition activity against seedling growth of *B. pekinensis* Rupr. at concentration 1000 ppm.

Two mixtures and eight compounds were isolated and purified from the roots of *T. indica* Merr. The chemical structures were characterized by means of spectroscopic studies, chemical reactions and physical properties. All the isolated substances are summarized as followed:

Mixture 1	$H_3C\text{-}(H_2C)_{13}\text{-}H_2C\text{-}C(=O)\text{-}OH$ hexadecanoic acid (palmitic acid) $H_3C\text{-}(H_2C)_{15}\text{-}H_2C\text{-}C(=O)\text{-}OH$ octadecanoic acid (stearic acid)  cis-9-octadecenoic acid (oleic acid)

Mixture 2	 β-sitosterol stigmasterol
Compound 1	 stigmasteryl-3-O-β-D-glucopyranoside
Compound 2	 oleanolic acid
Compound 3	 ursolic acid
Compound 4	 nonanedioic acid or azelaic acid

Compound 5	 decanedioic acid or sebacic acid
Compound 6	 12-(6′-methoxy-2′-chromenone-7′-yl) dodecanoic acid (new compound)
Compound 7	 3-demethyl-14α-hydroxyisotylocrebrine
Compound 8	 tylophorinidine

The chemical constituents of the roots of *T. indica* Merr. apart from mixture of two steroids, and tylophorinidine as described in this study was firstly reported in this plant.

The preliminary bioassay as plant growth regulators against *B. pekinensis* Rupr. seedling revealed that nonanedioic acid was highest inhibitory effect, it completely inhibited root length of lettuce seedling at concentration 1000 ppm. Decanedioic acid, 3-demethyl-14α-hydroxyisotylocrebrine and tylophorinidine inhibited root length of lettuce seedlings by 90.27, 86.27 and 70.38 % at concentration 1000 ppm, respectively. In addition, oleanolic acid and ursolic acid showed moderately inhibition of root and shoot length.

Furthermore, the bioassay towards *E. crus-galli* (L.) Beauv. for plant growth regulation activity was also conducted. Although at 1000 ppm, nonanedioic acid, decanedioic acid, 3-demethyl-14α-hydroxyisotylocrebrine and tylophorinidine completely inhibited root length of lettuce seedling while two phenanthroindolizidine alkaloids were still highly effective (96.05 and 89.01%) at 100 ppm. Thus, 3-demethyl-14α-hydroxyisotylocrebrine and tylophorinidine were more potent inhibitors of root growth than two dicarboxylic acids. Moreover, coumarin derivative showed moderately (66.54 %) effect against root length inhibition at concentration 1000 ppm. For shoot growth inhibition, decanedioic acid, 3-demethyl-14α-hydroxyisotylocrebrine, nonanedioic acid and tylophorinidine inhibited shoot length of *E. crus-galli* (L.) Beauv. seedlings by 100, 97.44, 90.53 and 85.58 % at concentration 1000 ppm, respectively. Some of these compounds might be candidate as a lead compound for new natural herbicides in the future.

Proposal for Future Work

From the result of plant growth regulators, many isolated substances showed significant inhibitory effect against both monocotyledon and dicotyledon plant. The possibly future work related to this research would be to further test for other monocotyledon and dicotyledon plant that caused problems in Thai agriculture. Moreover, the investigation of plant growth regulator activity was well known as a preliminary indicator that could be used for further study on other material methods such as pot test and field test. Two phenanthroindolizidine alkaloids showed high effect against monocotyledon and dicotyledon plants, which this is the first report for those compounds against this activity. The interesting SAR study of natural molecules such as phenanthroindolizidine type may be interesting to develop as natural herbicides.