CHAPTER IV

CONCLUSION

During the course of this research, the fruits and the seeds of *X. granatum* are selected for investigation their chemical constituents and searching for biological active substances according to the attractive preliminarly bioassay results. By means of physical properties, chemical reactions and spectroscopic data, nine components isolated from the fruits were characterized as a mixture of saturated long chain aliphatic esters, a mixture of saturated long chain alcohols, a mixture of β-sitosterol, stigmasterol and campesterol, a mixture of saturated long chain carboxylic acids, 7-oxo-7-deacetoxy gedunin, xyloccensin K, a mixture of β-sitosteryl, stigmasteryl and campesteryl glycoside. Two additional compounds were isolated from the seeds. Unfortunately, their structures could not fully charachterize because of the limited amounts of samples. However, the spectroscopic data of both of them clearly revealed that they are limonoids. To our knowledge, there is no report of xyloccensin K in the chemical literature. Hence, this compound is a new naturally occuring limonoid.

All isolated substances from both fruits and seeds of *X. granatum* are summarized as shown in Table 4.1 and Fig 4.1.

Table 4.1 All isolated compounds from the fruits and the seeds of *X. granatum*.

plant parts	solvent extract	substances
fruits	dichloromethane	a mixture of saturated long chain aliphatic esters, a mixture of saturated long chain alcohols, a mixture of β-sitosterol, stigmasterol and campesterol, a mixture of saturated long chain alliphatic carboxylic acids, 7-oxo-7-deacetoxy gedunin, xyloccensin K
	butanol	a mixture of β-sitosterol, stigmasterol and campesterol, xyloccensin K, a mixture of β-sitosteryl, stigmasteryl and campesteryl glycoside
seeds	dichloromethane	the same compounds that were obtained from the fruits, two unidentified limonoids
	butanol	the same compounds as reported in the fruit

CH_3-$(CH_2)_n$-CH_2OH

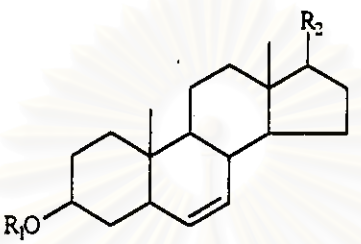
n=25, 27, 28, 29 and 30

Mixture **2**

CH_3-$(CH_2)_n$-CH_2-COOH

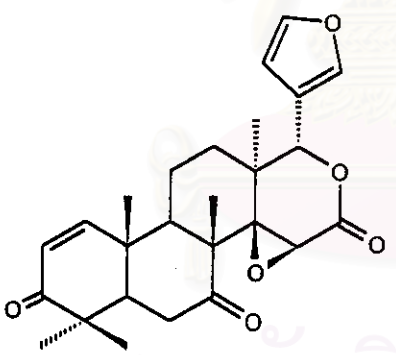
n=17, 18, 19, 20, 21, 22, 23, 24, 25, 27

Mixture **4**



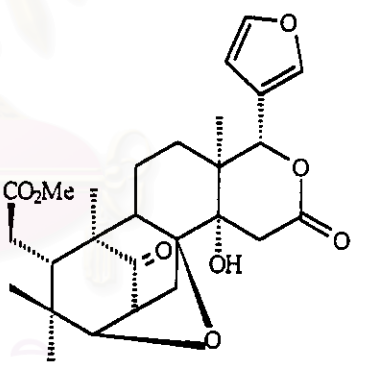
Mixture **3** mixture of R_1 = H, R_2 = $C_{10}H_{23}$, $C_{10}H_{21}$ and C_9H_{21}

Mixture **8** mixture of R_1= D-glucose, R_2 = $C_{10}H_{23}$, $C_{10}H_{21}$ and C_9H_{21}



Compound **5**

7-oxo-7-deacetoxy gedunin



Compound **6**

Xyloccensin K

Fig 4.1 All Isolated substances from the fruits and the seeds of *X. granatum*

Many limonoids have been isolated from Meliaceae plants.. The limonoids isolated from this research was found to be different from those previous reports particularly for a new limonoid xyloccensin K which was isolated from dichloromethane crude extract of both fruit and seed in a large quantity (0.07 % w/w of fruit and 0.10 % w/w of seed). This probably depends on the location where the plants were collected.

Concerning to the environmental impact of chemical insecticides, it is clearly a task for natural products chemists to try to search and develope new and environmental friendly insecticides. Many isolated compounds derived from plant origin were proved to exhibit these activities. Among them, limonoids, a group of chemically related bitter tetranortriterpenoids are mainly of insecticidal properties. The isolated compounds from *X. granatum* were also observed to possess the antifeedant activity against *Galleria mellonella*. The results indicate that only limonoids, 7-oxo-7-deacetoxy gedunin and xyloccensin K are active compounds in this respect with dosage 4 mg/box and gave the percentage antifeedant of 80.69 and 66.29, respectively.

The major component of this plant, xyloccensin K, was also investigated the proper concentration to inhibit feeding for wax moth larvae. The dosage required to inhibit of feeding approximately 80% is 20 mg/box. In order to compare the antifeedant effect of xyloccensin K and azadirachtin against *G. mellonella*, it was observed that the dosage of xyloccensin K at 4

mg/box give the near rate of percentage antifeedant activity to 0.05 mg/box of azadirachtin.

To extend the antifeedant activity study of xyloccensin K, the activity against Australian carpet beetle, *Anthrenoccerus australis* was investigated. The results revealed an antifeedant activity of xyloccensin K at the rate of 0.24 % w/w comparable with that by azadirachtin at the rate of 0.01 % w/w.

Although xyloccensin K is less active than azadirachtin, but it may be more stable. During the course of bioassay screening tests of the crude extracts of each part of *X. granatum* compared with neem crude, it was observed that neem crude did not show antifeeding activity after the crude was kept for several days at room temperature. It is possible that azadirachtin which is the most active compound in neem crude is easy to decompose during its storage. On the other hand, *X. granatum* crude extracts and the pure compound, xyloccensin K were still active even keeping for a long time (more than a few months) at room temperature.

In conclusion, it could be clearly seen that the fruits and the seeds of *X. granatum* could be used as a good source of limonoids. Moreover, the outcome of this research also supported a promising concept of fully utilization and agrochemical searching from ideal and potential resources of Thai mangrove plants. The study on chemical constituents accompanied with biological

activity study of mangrove plants is therefore one of the most worth considering for natural products chemists.

Propose for the Future Work

The occurance of xyloccensin K as a major component and one of an active ingredient provides many possibilities to carry on for further investigation. For instance, the utilization of xyloccensin K as an antifeedant agent against various larvae, particularly those that caused agricultural problems in Thailand should be seriously considered. The structure activity relationship of this limonoid and related compounds would provide an opportunity to understand what parts of the molecule have an influence for those interest activity. The outcome from this examination would lead to the finding of new and environmental friendly antifeeding agent. Another aspect that would make this research fulfill is the chemotaxonomy study on chemical constituents of other parts of *X. granatum* and also various parts of the other two *Xylocarpus* species in Thailand, *i.e.*, *X. molluccensis* and *X. gangeticus*.