CHAPTER V

CONCLUSIONS

The medicinal plant, *A. racemosus* is well-known for its use for female reproductive system. However, there is not much study on its application as antimicrobial. In order to investigate for the possibility of using this medicinal plant as an antifungal agent, several studies were performed.

The roots of *A. racemosus* were collected from different areas in Thailand. They were evaluated for saponin contents and antifungal activity. *A. racemosus* collected from Rayong possessed the highest saponin content. It also had the advantage over the plants from other sources as it could be obtained by cultivation while the other sources were from wild. This makes *A. racemosus* from Rayong more suitable to be used as raw material for industrial application.

A. racemosus roots were successively extracted with the series of solvents i.e. hexane (AR-H), ethanol (AR-E) and water (AR-A, AR-B and AR-S). To enrich saponin content in the extract, the dried A. racemosus roots were extracted with methanol, precipitated with acetone and partition with n-butanol. The extract with more than 35% of saponin was obtained (AR-En).

The chemical constituent profiles of *A. racemosus* roots were recorded using TLC. However, for quantitative analyses of saponin content in the extracts or the plant material, general method such as HPLC-UVvis could not be used as saponin glycosides in *A. racemosus* are lack of chromophore. Therefore, ELISA using MAb antibody against shatavarin IV was used in our studies.

For the antimicrobial activity of *A. racemosus* extracts, AR-E and AR-En showed an antifungal activity against *C. albicans*, *M. furfur* and *M. globasa* at the concentration of 1 mg/disc while the extracts with other solvents showed no inhibitory effects. No antibacterial activity was observed. MICs of AR-E against *C. albicans*, *M. furfur* and *M. globosa* were 2-25 mg/ml. Interestingly, with the saponin enrichment extraction method, the extract (AR-En) showed much lower MICs (0.10 for *C. albicans* 0.40 mg/ml for *M. furfur* and 0.20 mg/ml for *M. globosa*). Therefore, saponins might be responsible for antifungal activity in *A. racemosus*.

The mechanisms of action for antifungal activity *A. racemosus* extracts might be different than the antifungal agents, ketoconazole or zinc pyrithyone. Thus, synergistic effects between the extracts and the drugs might occur. The checkerboard synergy tests between AR-E or AR-EN and both antifungal agents against *M. furfur* and *M. globosa* were performed. However, the results showed that there were not synergistic effects of AR-E or AR-EN and antifungal agents tested.

For stability study, AR-E was kept at the accelerated condition (50 °C) for 30 days. Even though, the appearance of the extract slightly changed, the antifungal activity the chemical property of extract remained the same after the test.

In conclusion, these studies revealed the antifungal activity of A. racemosus extracts and also the method to prepare and to standardize the extracts. The extract showed satisfactory stability through the accelerate condition. The studies suggested potential of A. racemosus to be developed as antifungal products such as antidandruff and vaginal cleansing products.