

CHAPTER 5

CONCLUSION

The conclusion of this study should be focused in four important points including the quantitative HPLC analysis of rhinacanthins, preparation of the rhinacanthin high-yielding *R. nasutus* extract, establishment of standard information of the extract, and stability of the extract. The following conclusions can be drawn:

1. A simple, specific, precise, accurate, rapid and reproducible HPLC method has been developed and validated to quantify the rhinacanthin-C, -D, and -N in *R. nasutus* leaf extract. The simultaneous quantitative determination of rhinacanthin-C, -D, and -N allows presence of the content of these compounds to provide useful markers information for the quality control of *R. nasutus* leaves extract.

2. Fractionation of the ethyl acetate extract of *R. nasutus* using Amberlite[®] IRA-67 column eluted with 10 % acetic acid in methanol afforded the rhinacanthin high-yielding *R. nasutus* leaf extract, which contained the total rhinacanthin content up to 83.61 %w/w.

3. The standard specification of the rhinacanthin high-yielding *R. nasutus* leaf extract was established as follow;

- The total rhinacanthin content is not less than 70 %w/w, calculated as rhinacanthin-C, rhinacanthin-D, and rhinacanthin-N.

- The moisture content (loss on drying) is not more than 0.2 %w/w.

- Total ash is not more than 2.3 % w/w.

- Acid insoluble ash: No acid insoluble ash.

- Microbial contamination: No contamination with aerobic bacteria, *E. coli* and fungi.

4. The rhinacanthin high-yielding *R. nasutus* leaf extract contains most likely moderate non-polar compounds therefore the suitable solvents for the leaf extract should be a moderate non-polar solvent such as chloroform, ethyl acetate, ethanol, and methanol.

5. The hydrophobic parameter (log K value) of the rhinacanthins in the rhinacanthin high-yielding *R. nasutus* leaf extract was 1.73.

6. Stability evaluations of the rhinacanthin high-yielding *R. nasutus* leaf extract in several conditions in the period of 4 months found that the extract possessed a satisfactory stability at room temperature (30 ± 2 °C). However, the extract should be stored in well-closed container, protected from light. The aqueous solutions of the extract are not stable either in acid or base conditions.