Published by: Geological and Agricultural University of Science and Technology, Faculties of Science, Faculty of Science, Department of Chemistry, Faculty of Science, Department of Biology, College of Science, Chulalongkorn University.

Title: The Identities and Anti Herpes Simplex Virus Activity of Clinacanthus Nutans and Clinacanthus Siamesis.
Clinacanthus nutans and Clinacanthus siamensis (Thai name Phaya Yo and Lin Ngue Hao, respectively) are plants in genus Clinacanthus that can be found in Thailand. C. nutans has been used as a herbal drug to relieve skin rashes, herpes simplex and herpes zoster. Moreover, this plant was accepted to be a herbal drug in the list of herbal medicinal products A.D. 2006 by the national drug committee. Since some of the morphologies of C. siamensis are somewhat similar to C. nutans, distinguishing these related plants by pharmacognostic evaluation and biomochemical technique which are including of macroscopic, microscopic evaluation of stem, midrib cross section and components in leaf epidermis, measurement of stomatal number, stomatal index and palisade ratio, biomochemical technique by comparing the nucleotide sequence in internal transcribed spacer (ITS) region, the result revealed that stem, midrib cross section, components in leaf epidermis of C. nutans and C. siamensis are familiar. Stomatal number of C. nutans and C. siamensis are 168.32 ± 29.49 and 161.60 ± 18.04, respectively. Stomatal index of C. nutans and C. siamensis are 13.83 ± 0.86 and 11.93 ± 0.81, respectively. Palisade ratio of C. nutans and C. siamensis are 6.84 ± 0.66 and 3.37 ± 3.51, respectively. Comparison ITS region of all 6 samples of the two species (3 samples of each species), there are 33 polymorphisms which comprise of 23 nucleotide polymorphism and 11 indels which indicated 97-99% similarity. Moreover, testing for the anti herpes simplex virus type 1 and type 2 (HSV-1 and HSV-2) by plaque reduction assay of dried leaves that extracted with n-hexane, dichloromethane and methanol, respectively. The result showed the lowest extract concentration that inhibited HSV-1 by 50% (IC50) was 32.05 ± 3.63 µg/ml and selective index (SI) which was calculated from the extract concentration that causes death of tested cells by 50% divided by extract concentration that inhibited 50% of the virus was moreSkin 50/36 from C. nutans n-hexane extract, whereas the lowest IC50 that inhibited HSV-2 was 46.52 ± 4.08 µg/ml, SI was 34.53 from C. siamensis n-hexane extract. Cytotoxicity was tested by MTT assay. CC50 of n-hexane and methanol extract of both plants was more than 1,600 µg/ml. CC50 of dichloromethane extract of C. nutans and C. siamensis were 869 ± 141,93 and 193 ± 336, respectively. According to these evidences, it could be concluded that these plants are closely related to each other. Pharmacognostic evaluation can distinguish these plants especially, stomatal index and palisade ratio. However, evolution of nucleotide sequence in ITS region cannot distinguish these closely related plants due to their sequence variation in the ITS region. Furthermore, these medicinal plants can be developed or used as a source for isolation of anti-HSV compounds.