# CHAPTER 2

## LITERATURE REVIEW

### 2.1 Taxonomic History of the "*H. parasitica* complex"

The first study of this species was recorded at the end of the 18th century by Danish mission in S. E. India. Some of its employees had paid their attention on tropical plants, among them was J. G. Koenig. He collected some plants and sent them back home. These specimens were studied by Vahl in Copenhagen. Among these specimens, two of them were thought to represent a new genus, *Spelringia*. In 1810, Vahl described two species in this genus, namely *S. verticillata* Vahl and *S. opposita* Vahl. Then in 1837, G. Don transferred these two species to the genus *Hoya* (R. Br.), i.e. *Hoya verticillata* (Vahl) G. Don. These combinations are validly published, though the type specimens are not complete (Veldkamp et al, 1995).

Roxburgh (1814) named a climbing plant based on the material in the Botanic Gardens of Calcutta as *Asclepias parasitica* Roxb., which was validly described in 1832. In 1834, Wigth made a new combination of this species as *Hoya parasitica* (Roxb.) Wall. ex Wight and this name was adopted in Flora of the British India (Hooker, 1883), the Fl. Gen. Indo-Chinae (Costantin,1912), the Flora Siamensis Enumerratio (Kerr, 1951), A Flora of Malay Peninsula (Ridley, 1923) and the Malaysia Nature Journal (Rintz, 1978).

In addition, this species was originally introduced to the Royal Botanic Gardens, Kew in 1818 by Dr. Wallich from Calcutta. At Kew this plant species was distributed to the other gardens and some plant collectors in London, where it has been generally known under the name *H. lanceolata*. In 1819, Haworth had seen this plant growing without flowers at Kew Gardens, he coined a name, *H. acuta*. The plant blossomed for the first time at Sion House, Kew Gardens in 1825 and was registered under the name, *H. pallida* in 1826. It was noted that a plant which is sold under the name of *H. albens* in the nursery of Mr. John Miller of Bristol, is probably the same species (Traill, 1826).

The first variety of *Hoya parasitica* (Roxb.) Wall. ex Wigth was proposed in 1912 by Costantin. He described two new varieties from plants collected from Cambodia as *H. parasitica* var. *geoffrayi* and var. *spirei*. Then Rintz (1978) has reduced *Hoya citrina* Ridl. to a variety of *H. parasitica*. The variety *citrina* distributed in peninsular Thailand, Malaysia and Singapore. In 1995, Kiew described a new variety, *H. parasitica* var. *hendersonii* which is endemic to Malaysia.

Veldkamp and Hansen (1996) confirmed the treatments of Vahl and G. Don to reduced *Sperlingia* Vahl as synonym of *Hoya* R. Br. According to the International Code of Botanical Nomenclature (ICBN), the corrected name of a plant must be the first published name which has a description and a designed type specimen. Accordingly, *H. verticillata* (Vahl) G. Don. is a corrected name and *Hoya parasitica* (Roxb.)Wall. ex Wight is also regarded as a synonym of this species. So the two varieties from Cambodia described by Costantin (1912) were reduced to synonym under *H. verticillata* (Vahl) G. Don. Moreover, some previous distinct species were transferred to this species, i.e. *H. globiflora* Ridl., *H. acuta* Haw., *H. pallida* Lindl., *H. lanceolata* Lindl., *H. albens* J. Miller and *H. hookeriana* Wight. The other names may belong to *H. verticillata* as well, i.e. *H. rigida* Kerr, *H. obscurinervia* Merr. and *H. ridleyi* King & Gamble (Veldkamp et al., 1995). Hence, the new combinations were required for the previous varieties of the *Hoya parasitica*. Therefore, three varieties of *H. verticillata* (Vahl) G. Don were described, i.e. *H. verticillata* (Vahl) G. Don var. *verticillata*, *H. verticillata* (Vahl) G. Don var. *citrina* (Ridl.) Veldkamp and *H. verticillata* (Vahl) G. Don var. *hendersonii* (Kiew) Veldkamp.

## 2.2 Methodological review

The complex species are group of closely related taxa at and/or just below the species level (Stace, 1984). They usually have a great variation in morphological characters. So the status of the species complex is rather difficult to define. Morphological distinctions among these species are not always clear-cut and the problems in identification are still existed. This set up a research question of whether or not more than one species is included in the complex. So taxonomic studies in

complex species require intensive investigation and need more evidences to determine its taxonomic status. The followings are examples of methods (morphology, anatomy, palynology, morphometry and molecular biology) currently used to clarify the taxonomic status of the species complex.

Forster and Liddle (1991) recognized five subspecies within *Hoya australis* complex using qualitative and quantitative characters of both vegetative and reproductive structures. *H. australis* subsp. *oramicola* was newly described and the new combinations *H. australis* subsp. *tenuipes* (*H. oligotricha* subsp. *tenuipes* K. Hill) and *H. australis* subsp. *rupicola* (*H. rupicola* K. Hill) are proposed.

Chatrou (1997) used cluster analysis to reveal the patterns of macro-morphological variation in a species complex of *Malmea* (Annonaceae). Of 53 characters, 24 were important for clustering 238 herbarium specimens into 12 clusters. A new subspecies, *M. depressa* subsp. *abscondita*, was described. Moreover, *M. gaumeri* and *M. leiophylla* are suggested as synonyms of *M. depressa*. While, cluster analysis and principal component analysis of 66 morphological characters from 103 populations of the *Lobelia cardinalis* complex failed to disclose groups of populations. The complex comprises a single species, *L. cardinalis*, and that this species should not be divided into infraspecific taxa (Thompson and Lammers, 1997). Likewise, 215 accessions of 30 taxa in the *Solanum brevicaule* complex and 42 accessions of six taxa outside the complex were determined using 53 morphological characters. Principal Component Analysis and Discriminant Analysis were used, however, the outcomes were unable to support 30 taxa, suggesting a single variable complex (van den Berg et al, 1998).

Aldasoro et al. (1998) carried out a multivariate morphometric study from 127 herbarium specimens and nine populations of the genus *Sorbus*. The principal component analysis, discriminant analysis and cluster analysis of morphological and anatomical data were carried out. The results showed that twelve species could be easily recognized in the area. It was reported that *Simarouba amara* was frequently confused with the other two continental species, *S. glauca* and *S. versicolor*. Cluster analysis and Principal Component analysis were applied to verify the distribution and variation of the diagnostic characters proposed in the preceding revision, i.e. anther

size, stamen appendage, indument, leaflet surface, and venation features. *S. glauca* and *S. versicolor* were found to be morphologically closer than *S. amara*. Overlapping of characters in boundary populations of the three species was also found (Franceschinelli et al.,1998).

In northern America, the cosmopolitan species, *Pteridium aquilinum* is represented mainly by var. *latiusculum* and var. *pseudocaudatum*. Twelve quantitative and qualitative morphological characters were examined in 262 specimens using PCA and cluster analysis to assess the taxonomic relationship between these two varieties. When the qualitative characters were used alone or in conjunction with some of the quantitative traits, the samples grouped into two distinct clusters corresponding to the two recognized varieties. The morphological study also supports a taxonomic treatment at the varietal level (Speer and Hilu, 1998). Furthermore, intraspecific morphological variation was investigated in *Eriastrum densifolium*. To assess the five currently recognized subspecies vegetative and floral characters were analyzed at the species and population level by using cluster analysis. The herbarium specimens, field collections, and common garden plants were used. The only exception was a group of plants distinguished from the remainder species by corolla tube length. This group of individuals matched the circumscription of *E. densifolium* subsp. *sanctorum*. The other four recognized subspecies failed to form distinct morphological groups in all analyses (Brunell and Whitkus, 1998).

Hess and Stoynoff (1998) used cluster analysis and discriminant analysis examined vegetative and reproductive characters in *Quercus shumardii* var. *acerifolia* and comparing with *Q. shumardii*, *Q. buckleyi*, *Q. texana*, and the maple-leaf oak. Cluster analyses segregated maple-leaf oak from *Q. shumardii* and the other two recognized taxa. Based upon these numerical analyses and the evaluation of descriptive characters, *Q. acerifolia* was shown to be a distinct species. Nelson and Elisens (1999) performed cluster analysis, principal components analysis and canonical variance analysis based on 16 morphological characters from 33 populations represent all taxa and ploidy levels of the genus *Chelone*. This work recognized three diploid species without infraspecific taxa in this complex. Kephart et al. (1999) used principal component analysis and discriminant analysis to determine whether quantitative morphology could effectively distinguish varieties, population,

and subpopulations of the polymorphic species, *Silene douglasii*. A phenetic analysis of 354 plants samples from 16 populations using vegetative characters (e.g., leaf width and pubescence) were the most effective characters to distinguish the variety *rupinae*, whereas reproductive characters (e.g. calyx width, petal dimensions) were more useful for var. *oraria* and var. *douglasii*. Recently, Boonkerd, Saengmanee and Baum (2002) examined 200 specimens of the *Bauhinia pottsii* complex using 43 quantitative characters. Cluster analysis and canonical discriminant analysis were performed. It was found that these characters collectively support the four varieties as defined by qualitative characters.

Ohta and Takamiya (1999) performed the morphological investigations of the Japanese *Diplazium mettenianum* complex. An analysis of 374 plants utilizing 20 qualitative morphological characters showed that the complex could be divided into five forms and statistical analysis of 16 quantitative morphological characters supported the distinction of five forms which were regarded as independent species, viz. *D. mettenianum*, *D. fauriei, D. deciduum* nom. nov., *D. griffithii*, and *D. hayatamae* sp. nov.

Leaf anatomical data from 13 taxa of the *Muhlenbergia montana* complex were studied, and 18 important characters were found. Results obtained from the cluster analysis have given independent data to compare with the previously reported data in determining the relationships of the species. Leaf anatomy clearly supported the recognition of a close relationship taxa, i.e. *M. virescens* and *M. quadridentata*. Therefore, a change of taxonomic category was suggested (Herrera-Arrieta and Grant, 1994). Chamberland (1997) worked on the *Echinocactus polycephalus* complex, which included *E. parryi*, *E. polycephalus* var. *polycephalus*, and *E. polycephalus* var. *xeranthemoides*. Herbarium specimens and plant specimens collected in the field were examined using scanning electron microscope. Then phenetic analyses were carried out to support the distinction of *E. parryi* and *E. polycephalus* as separate species. The varietal ranking of *E. polycephalus* var. *xeranthemoides* was also confirm. It was found that the varieties in the complex species were distinguished by seed and hypanthium scale morphology. Recently, Saarela and Ford (2001) used a combination of macro- and micro- morphological, and anatomical data to support the recognition of three species in the *Carex backii* complex, namely *C. backii, C. saximontana*, and

*C. cordillerana.*

Form the last decade, the PCR-RFLP (polymerase chain reaction – restriction fragment length polymorphism) method is extensively used in plant systematics. Parducci and Szmidt (1999) studied chloroplast DNA (cpDNA) of the genus *Abies* (Pinaceae). They determined inter-specific variation in this genus and studied how the variation was distributed in different regions of the genome. The results showed that the chloroplast genome is highly variable in most of the investigated taxa and contains multiple variable regions that appear to be distributed throughout the whole genome. Species-diagnostic markers were found from four of the ten investigated species. Mohanty, Martin and Aguinagalde (2001) used the PCR-RFLP technique to detect cpDNA diversity in wild populations of sweet cherry (*Prunus avium*) from five European deciduous forests and some cultivars. The result proposed the possible utilisation of the technique for the characterisation of sweet cherry cultivars. Besnard and Berville (2002) worked on variation of olive cpDNA. Four chlorotypes were identified in the olive complex but the data was insufficient to distinguish the North African subspecies *europaea*, *maroccana*, *guanchica* and *laperrinei*. Then, Besnard, Khadari, Baradat and Bervillé (2002) studied cpDNA diversity in the olive (*Olea europaea* L.) complex. They distinguished fifteen chlorotypes and constructed a cpDNA phylogenetic tree in which five clades that located in distinct geographic areas were recognized. Holderegger, Stehlik and Abbott (2002) worked on cpDNA of *Saxifraga oppositifolia*. PCR-RFLP technique detected two common European haplotypes throughout the Alps and two additional rare haplotypes were found in three harboured populations.

Recently, a population genetic analysis of chloroplast DNA was performed covering nine wild populations of *Brassica oleracea* by Panda, Martin and Aguinagalde (2003). Three closely related species were also studied to detect their relationship with *B. oleracea*. The PCR-RFLP method, amplified product of each of twelve primer pairs, digested by three restriction enzymes, revealed no variation of cpDNA among the studied taxa. The author proposed that the populations may have the same chloroplast genotype. This study confirmed a close relationship between *B. alboglabra, B. bourgeaui* and *B. montana*, which is parallel to their morphological similarity.

From the aforementioned researches above it can be seen that macro-micromorphological, anatomical, palynological and molecular techniques were commonly applied to solve plant taxonomic problems, especially in determining the status of species complex and usually carry out in conjunction with numerical analysis. Therefore, these procedures can be effectively applied to elucidate the taxonomic problem within the "*Hoya parasitica* complex" in Thailand.