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LEAF ARCHITECTURE AS A PROMISING TOOL IN CONFIRMING IDENTITY OF CONFUSING PLANT TAXA

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ABSTRACT – The search for stable taxonomic characters has always been a subject of interest among taxonomists and systematists. Several studies had been using tools such as gross morphology, anatomy, biochemistry and molecular biology to address or somehow resolve taxonomic problems. However, these studies were limited, depending on the resources made available to a laboratory or office. This paper presents leaf architecture as another potential tool in taxonomy. Leaf architecture refers to patterns of venation in leaves, found to be genetically fixed. Published literature in selected journals were reviewed in detail and were synthesized. The review indicated that leaf architecture is currently used in 1) species identification, 2) species description, and 3) species classification. It has been quite effective in addressing nomenclatural issues between two morphologically similar species, very common in many cultivated plants, like *Hoya*, causing taxonomic confusion among gardeners, hobbyists and even taxonomists. Gaps include, 1) small number of specimens or small number of representative species (for family and generic studies), though leaf samples taken, were at least 30 per species, 2) samples were taken from a single source most of the times, disregarding possible variation when samples are from diverse localities, topographies, elevations and agroclimatic conditions, and 3) age of the leaf was not a consideration in collecting samples. Despite these gaps, though, leaf architecture has a very high potential to complement reproductive structure-based Linnean taxonomy and the modern day molecular taxonomy.

Keywords: leaf morphology, leaf venation, systematics, taxonomic evidences

INTRODUCTION

Plant taxonomy is beset with never-ending problems and controversies from species identification, nomenclature to classification (Baltazar and Buot 2019a, Christenhusz and Chase 2014, Aurigue 2013, Larano and Buot 2010, Wanntorp et al. 2006, Smith et al. 2006 and APG or Angiosperm Phylogeny Group, 1998 among others). Most of the times, this is due to differences in the use of evidences as basis for scientific decision making. Arguments arise because of differing views, limitations in methodology and the appropriateness in terms of number, variety and sources of specimens used. Hence, we have several botanical classification systems through the years (PPG or Pteridophyte Phylogeny Group 2016, APG 2016, 2009, 2003, 1998, Takhtajan 1996, Cronquist 1988). Plant taxonomists all over the world are continuously solving nomenclatural problems and use reliable and affordable methodologies within their reach.

Some researchers in well-facilitated laboratories use molecular methodologies. Others use morphological methods which are relatively cheaper. Among those who are using the morphological

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approach, a considerable number utilize reproductive structures, considered as more stable as main basis, while few others still utilize the vegetative characters with the leaf surface as the focus (Sack and Scoffoni 2013, Ellis et al 2009, Stace 1984, Dilcher 1974, Hickey 1973).

Taxonomists and systematists, however, often neglected leaf characters and other vegetative characters in the identification and classification of plant taxa due to the prevailing view that these characters have high phenotypic plasticity which could be true anyway to a certain extent (Medina et al., 2016). Nevertheless, according to Roth-Nebelsick, et al. (2001), leaf architectural pattern which is a morphological character, is genetically fixed. Numerous studies had been following and testing the initial results of Roth-Nebelsick, et al. (2001), and all reported the great potentials of leaf architecture as a tool in addressing taxonomic problems (Tan and Buot 2020, 2019, and 2018, Baltazar and Buot 2019b and 2019c, Masungsong et al. 2019a and 2019b, Paguntalan and Buot 2019, Huiet et al 2018, Conda and Buot 2018, Conda et al., 2017, Torrefiel and Buot 2017, Jumawan and Buot 2016, Villareal and Buot 2015, Pulan and Buot 2014, Barroga and Buot 2014, Salvana and Buot 2014, Larano and Buot 2010, Obico et al. 2007, Banaticla and Buot 2004). Results of these studies claimed that leaf architecture can be very relevant in solving the identity of taxonomically confusing taxon, in the absence of reproductive structures which are quite seasonal. In fact, one study on the phylogeny of *Adiantum* species (Huiet et al. 2018) found out that the phylogenetic tree generated from leaf architectural data, perfectly matched with the phylogenetic tree generated from molecular data, including the identification of biogeographic provinces of the species of *Adiantum* in the study.

It is the objective of this paper to 1) preliminarily synthesize the significance of leaf architecture in taxonomy, 2) reflect on the gaps of these recent leaf architectural studies and 3) discuss insights on the potentials and prospects of leaf architecture as a taxonomic evidence.

METHODOLOGY

A review of published literature on the use of leaf architecture as basis in solving taxonomic problem was done. Journal articles (mostly in Web of Science and Scopus-indexed journals) on angiosperms and pteridophytes were reviewed. Then a synthesis was made on the use of leaf architecture in plant taxonomy, recognizing gaps and sharing insights on the potentials of the method.

RESULTS AND DISCUSSION

Significance of leaf architectural studies to plant taxonomy

The leaf is the most widely used vegetative characters in plant taxonomy, but still it ranks second only to reproductive structures (flowers and fruits). However, leaves have some advantages over floral structures as they are comparable to a wider taxonomic range and they persist longer during the life span of the plant (Stace 1984, Tomlinson 1984). One of the important taxonomic characters of the leaf surface is leaf architecture (Fig. 1 and 2). It is defined as the placement and form of those elements constituting the outward expression of leaf structure including shape, marginal configuration, venation, and gland position (Ellis et al., 2009, LAWG 1999, Hickey, 1973). According to Hickey (1973), this is the tendency of leaves to form definite, stable and unique structural patterns that can be described. Dilcher (1974) emphasized that gross leaf form alone cannot produce reliable information for the study of fossil plant remains. It must be coupled by the finer venation and/or cuticular characters to be proven useful in recognizing taxa of seed plants (Fig. 2).

Figure 1. The intricate leaf architectural pattern comprising primary, secondary, tertiary, and higher venation patterns and areolation, delineating species and taxonomic groups. *Photo by the author.*

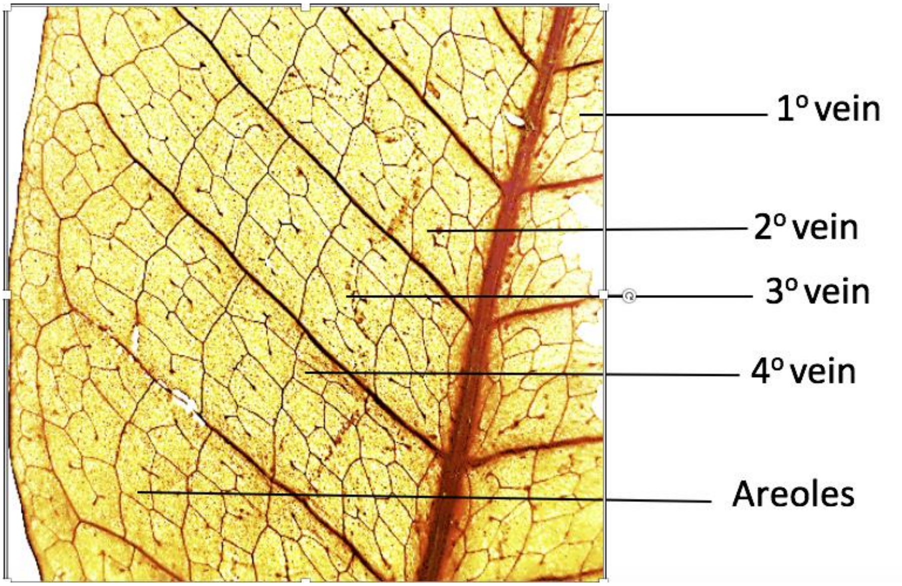

Figure 2. A photomicrograph showing the details of leaf venation pattern in a fern, *Microsorum heterocarpum*. After Tan and Buot (2020) and Tan (2019).

Hickey (1973) devised a coherent and unambiguous classification of leaf architecture for dicotyledons. Later, the LAWG or Leaf Architecture Working Group (1999) broadened the scope of the classification by including the net-veined monocots. The works of LAWG (1999), Wolfe (1989), Dilcher (1974) and Hickey (1973), became instrumental in our current pursuit on the use of leaf architectural characters in the study of the taxonomy of Philippine taxa (Tan and Buot 2020, 2019, and 2018, Baltazar and Buot 2019b and 2019c, Masungsong et al. 2019a and 2019b, Paguntalan and Buot 2019, Conda and Buot 2018, Conda et al., 2017, Torrefiel and Buot 2017, Jumawan and Buot 2016, Villareal and Buot 2015, Barroga and Buot 2014, Kpadehyea and Buot 2014, Pulan and Buot 2014, Salvana and Buot 2014, Celadina et al., 2012, Larano and Buot 2010, Obico et al. 2007, Banaticla and Buot 2004). These leaf architectural characters include the study of both the general laminar and venation characters. Emphasis was on the finer venation patterns in the tertiary, quaternary, quintenary and up to areolation, where variation had been very significant (Fig. 2).

Preliminarily, the results of these studies pointed out the fact that leaf architectural patterns are very relevant in 1) *species identification and description*, 2) *addressing nomenclatural issues*, and 3) *confirming and reinforcing the classification or grouping of certain taxa like section or families*.

The most basic importance of leaf architecture had been in *species identification and description*. This had been illustrated in many studies. Among the related published works reviewed, were that of Barroga and Buot (2014) for around ten species of *Terminalia*, Kpadehyea and Buot (2014) for certain cultivars of *Mussaenda*, Pulan and Buot (2014) for selected species of *Shorea*, Celadina et al. (2012) for Philippine *Cinnamomum* species, Larano and Buot (2010) for some species of Malvaceae, and Banaticla and Buot 2004) for some species of *Psychotria*. Species were simply described using leaf architectural terminologies of LAWG (1999) and Hickey (1973). The second important relevance of leaf architecture as indicated in the reviewed papers, was in *addressing nomenclatural issues*. The field of botany has been experiencing a lot of confusion in identifying two almost similar species. The genus *Hoya* which has a number of confusing species had been represented in many papers reviewed here (Baltazar and Buot 2019c, Paguntalan and Buot 2019, Masungsong et al. 2019a and 2019b, Tan and Buot 2018, Torrefiel and Buot 2017, Jumawan and Buot 2016), Salvana and Buot 2014). Identity of confusing *Hoya* species had been addressed in these papers like the case of *H. benguetensis* and *H. ilagiorum* (Tan and Buot 2018). These two species are always misidentified and some people considered them as one species only, as they appear similar in outward appearance. Leaf architecture evidences, however, pointed out that there are really two species, *H. benguetensis* and *H. ilagiorum*. On the other hand, the leaf architecture study of several accessions of *Cucumis* in a gene bank (Masungsong et al. 2019a and 2019b), revealed many duplicate accessions. Their study was instrumental in remarkably reducing the cost of planting and replanting duplicate accessions in the gene bank.

Finally, this review found out that leaf architecture had been an aid in *confirming and reinforcing the classification or grouping of certain taxa like section or families* (Baltazar and Buot 2020, *submitted*, Tan and Buot 2020 and 2019, Larano and Buot 2010). Baltazar and Buot (2020, *submitted*) observed that leaf architecture had been reinforcing some currently recognized sections of the genus *Hoya*. Larano and Buot (2010), on the other hand, supported the lumping of Bombacaceae, Malvaceae, Sterculiaceae and Tiliaceae into one big family Malvaceae *sensu* APG I (1998) using leaf architectural evidences. Leaf architecture had also been utilized for ferns. In fact, it had also confirmed the classification of Eupolypods I families of ferns *sensu* PPG (2016) as reported by Tan and Buot (2020 and 2019). Interestingly, just like the case of Malvaceae, leaf architecture analysis of Eupolypods I, coincided with the findings of PPG (2016) which were largely based on molecular phylogeny. Indeed, leaf architecture as a taxonomic character, has a great potential for species identification, description and classification.

Gaps of recent leaf architectural studies

Current leaf architectural studies are focused more on controversial taxa. A closer examination of the methodology of the papers, revealed limited number of specimens used in the study. Leaf samples were usually obtained from few plant individuals only. Even those examining a taxonomic group (infrageneric section or family), the number of species and individuals was quite few (Tan and Buot 2020 and 2019, Conda and Buot 2018, Conda et al., 2017). Samples from many individuals growing in a variety of habitats and localities are preferred as these might illustrate some variations among leaf venation patterns. Leaf samples observed and measured from the reviewed studies, however, were around 30 in number per species, yet these were taken from few plant individuals only (Baltazar and Buot 2019c, Paguntalan and Buot 2019, Tan and Buot 2018, Torrefiel and Buot 2017, Jumawan and Buot 2016, Salvana and Buot 2014). Other studies were based on herbarium specimens (Tan and Buot 2020 and 2019, Conda and Buot 2018, Conda et al., 2017, Larano and Buot 2010). Incidentally, herbarium collections were not that extensive as well, as they are limited by the number and variety of deposited vouchers. Hence, samples were not able to address expected variations mentioned earlier. Therefore, though sample measurements were normally at least 30, but they were obtained from around 3-5 accessions only. The researchers had been assuming that leaf architectural patterns are fixed and stable across elevation and agroclimatic conditions as reported by Tan and Buot (2020 and 2019), Conda and Buot (2018), Conda et al. (2017).

Another gap seen in these studies is the absence of consideration that leaf architectural patterns may change across leaf age. As a norm and a matter of protocol, these studies cited above were using mature leaves. There was no attempt to examine the difference in leaf architecture pattern between the young and the older leaves. All of these studies were assuming that leaf architectural patterns are stable character.

One final gap observed in the preliminary works on leaf surface venation is the search for the most stable leaf architectural character state. That key character state which delineates a species. This is illustrated by Huiet et al. (2018) where they selected only around 6 key leaf characters of *Adiantum* which incidentally was very effective. This leaf architectural study utilizing only six character states resulted to a phylogenetic tree which perfectly matched with the one created using molecular data. This means that the selection of these 6 key characters were indeed principled. Studies along this line have to be looked into.

Insights on the potentials and prospects of leaf architecture as a taxonomic evidence

The Linnean taxonomy has been using the sexual system in plant identification. Hence, the reproductive structures are the main bases in the identification and classification of taxa. It has been very effective as well but reproductive structures (flowers, fruits, or spores for ferns) are not always available. Hence, some vegetative features are used in diagnosis of some major angiosperm families such as Dioscoreaceae (direction of stem twining), Bromeliaceae (marginal leaf spines, Cucurbitaceae (tendril morphology), Lauraceae (growth habit) and Myrtaceae (phyllotaxis) (Tomlinson, 1984).

In this paper, another very important character state is added to this very few options. Leaf architecture characters can be used as a tool in taxonomic studies. And it is very clear from the aforementioned studies, that leaf architecture as a taxonomic tool and evidence has a great potential. The work of Huiet et al. (2018) on *Adiantum* taxonomy proved that indeed leaf architecture patterns are genetically fixed, as first reported by Roth-Nebelsick et al. (2001). Hence, the use of leaf architectural data would refute the prevailing views that all leaf surface characters are plastic and not stable. Leaf architectural characters, particularly, the finer venation characters are stable and remain the same across environmental gradients (Tan and Buot 2020 and 2019, Conda and Buot 2018, Conda et al., 2017). Moreover, the results of the leaf architectural studies of Baltazar and Buot (2020, *submitted*) on the sections of the genus *Hoya* were also having similarities with the sections proposed by Wanntorp et al. (2006) based on molecular data. This has a particular reference to the Australian/New Guinean Clade of Wanntorp et al. (2006), where they lumped two existing morphology-based sections (sect. *Pterostelma-Physostelma*) into one clade. Similarly, the leaf architecture work of Baltazar and Buot (2020, submitted), provided additional evidence that they should be fused and in fact, with one more section, *Physostemma* which was not treated by Wanntorp et al. (2006). All of these were not mere coincidences, but simply pointed out the potential of leaf architecture as a reliable taxonomic evidence.

CONCLUSION AND RECOMMENDATIONS

Current studies under review, illustrated that leaf architecture pattern, is stable and can be used to confirm identity of taxonomically confusing taxa with success, particularly in the absence of reproductive structures. It has a high potential as a taxonomic tool with cost-effective, practical and theoretical relevance and significance. It will be of help to expand leaf architectural studies to other taxa and try to explore on the possible variation of leaf venation between young and mature leaves, using as many plant species, individuals and leaf samples (following standard statistical principles), taken from different portions of the plant as possible.

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STATEMENT OF AUTHORSHIP

The author solely planned out and wrote this paper on leaf architecture to come up with a brief synthesis on the relevance of leaf architecture in addressing taxonomic problems, particularly, taxonomically confusing species.

REFERENCES

APG or Angiosperm Phylogeny Group. (1998). An Ordinal Classification for the Families of Flowering Plants. Annals of the Missouri Botanical Garden, Vol. 85, No. 4 (1998), pp. 531-553.

APG or Angiosperm Phylogeny Group. (2003). An update of the Angiosperm Phylogeny Group classification for the orders and families of flowering plants: APG II. Botanical Journal of the Linnean Society, 2003, 141, 399–436. With 1 figure.

APG or Angiosperm Phylogeny Group (APG III). (2009). An update of the Angiosperm Phylogeny Group classification for the orders and families of flowering plants. APG III. Botanical Journal of the Linnean Society 161: 105–121.

APG or Angiosperm Phylogeny Group. (2016). An update of the classification for the orders and families of flowering plants: APG IV. Botanical Journal of the Linnean Society, 2016, 181, 1–20. With 1 figure.

Aurigue, F.B. (2013). A collection of Philippine Hoyas and their culture. Philippine Council for Agriculture, Aquatic and Natural Resources Research and Development (PCAARRD), Department of Science and Technology (DOST): Los Banos, Laguna, Philippines.

Baltazar, A.M. and Buot I.E.,Jr. (2019a). Controversies on *Hoya* R. Br. Taxonomy. The Thailand Natural History Museum Journal 13(1): 59-68.

Baltazar, A.M. and Buot I.E.,Jr. (2019b). Leaf architectural analysis of taxonomic confusing coffee species: *Coffea liberica* and *Coffea liberica* var. *dewevrei*. Biodiversitas, 20: 1560-1567

Baltazar, A.M. and Buot, I.E.,Jr. (2019c). Resolving Taxonomic Confusion between *Hoya cumingiana* Decne. and *Hoya densifolia* Turcz. (Apocynaceae) using Leaf Architectural Analysis. The Thailand Natural History Museum Journal 13(2): 77-89.

Baltazar, A.M. and Buot, I.E.,Jr. (2020). A reinvestigation of the *Hoya* R.Br. Sections using leaf architectural analysis. *Submitted.*

Banaticla, M.C.N. and Buot, I.E.,Jr. (2004). Leaf architecture of ten Philippine *Psychotria* species (Rubiaceae). Philipp. Scient. 41:74-90.

Baroga, J. and Buot, I.E.,Jr. (2004). Leaf architecture of ten species of Philippine *Terminalia* Linn. (Combretaceae). International Research Journal of Biological Sciences. 3 (3): 83–88.

Celadina DA, Buot IE, Jr., Madulid, DA, Evangelista TT and Tandang DN. (2012). Leaf Architecture of Selected Philippine *Cinnamomum* Schaeff. (Lauraceae) Species. The Thailand Natural History Museum Journal 6(2): 89-111.

Christenhusz, M. and Chase M. (2014). Trends and concepts in fern classifcation. Annals of Botany. 113(4):571–594.

Conda, J., Buot, I.E. Jr. and Escobin R. 2017. Leaf architecture of selected Philippine *Diplazium* Swartz species (Athyriaceae). The Thailand Natural History Museum Journal 11(2):57–75.

Conda, J. and Buot, I.E.Jr. (2018). Species delineation of the genus *Diplazium* Swartz (Athyriaceae) using leaf architecture characters. Bangladesh Journal of Plant Taxonomy, 25(2), 123-133.

Cronquist, A. (1988). The Evolution and Classification of Flowering Plants. 2nd ed. 1988. New York Botanical Garden Bronx MY USA, pp. 332.

Dilcher, D.L. (1974). Approaches to the identification of angiosperm leaf remains. Botanical Review 40 (1): 1–157.

Ellis B; D.C. Daly; Hicky, L.J.;Johnson. K.R.; Mitchell, J.D.;Wilf, P. and Wing, S.L. (2009). Manual of Leaf Architecture. New York Botanical Garden Press, Ithaca, New York.

Hickey, L.J. (1973). Classification of the architecture of dicotyledonous leaves. American Journal of Botany, 60(1):17–33.

Huiet, L.; Li, F.W.; Kao, T.T.; Prado, J.; Smith, A.R.; Schuettpelz, E. and Pryer, K.M. (2018). A worldwide phylogeny of *Adiantum* (Pteridaceae) reveals remarkable convergent evolution in leaf blade architecture. Taxon 67 (3): 488–502.

Jumawan, J. H. and Buot, I.E.Jr. (2016). Numerical taxonomic analysis in leaf architectural traits of some *Hoya* R. Br. species (Apocynaceae) from Philippines. Bangladesh Journal of Plant Taxonomy, 23(2), 199. doi:10.3329/bjpt.v23i2.30851

Kpadehyea, J.T. and Buot, I.E.,Jr. (2014). Leaf Architecture of two Species and nine Infraspecific Taxa of the Philippine Mussaenda Linn. (Rubiaceae): Conservation concerns. International Research Journal of Biological Sciences **3 (10):** 13-21.

Laraño, A.A. and Buot, I.E.,Jr. (2010). Leaf architecture of selected species of Malvaceae sensu APG and its taxonomic signi cance. Philippine Journal Systematic Biology 4: 21–54.

LAWG (Leaf Architecture Working Group) (1999). Manual of Leaf Architecture – morphological

description and categorization of dicotyledonous and net-veined monocotyledonous angiosperms. Washington DC: Smithsonian Institute. 65p

Masungsong, L.A.; Belarmino, M. and Buot, I.E.,Jr. (2019a). Delineation of the selected *Cucumis* L. species and accessions using leaf architecture characters**.** Biodiversitas 20: 629-635.

Masungsong, L.A.; Torreta, N.K.; Belarmino, M.M.; Borromeo, T.H. and Buot, I.E.,Jr. (2019b). Leaf Architectural Variations among Species and Accessions of Genus *Cucumis* L. The Thailand Natural History Museum Journal 13(2): 91-101.

Medina, M.; Amoroso, V. and Kloppenburg, R.D. (2016). Changes of leaf morphology of *Hoya amorosoae* from varying light exposure: Its implications to species description and taxonomy. Journal of Biodiversity and Environmental Sciences 8(6): 232–237.

Obico J.J.; Bagay,K.C.; Asencion, A.S. and Medecillo, M.M. (2007). Comparative Study of the Leaf Morphology of *Epipremnum* Schott and *Rhaphidophora* Hassk. (Araceae) in thePhilippines. Philipp. J. Sys. Biol. 1(1): 15-25.

Paguntalan, D.P. and Buot, I.E.,Jr. (2019). Short Communication: Investigation of leaf architectural patterns: Implications in delineating taxonomically controversial *Hoya merrillii* Schlechter and *Hoya quinquenervia* Warburg. Biodiversitas 20: 833-839. https://doi.org/10.13057/biodiv/d200329

PPG I or Pteridophyte Phylogeny Group. (2016). A community-derived classification for extant lycophytes and ferns. Journal of Systematics and Evolution, 54(6), 563-603.

Pulan, DE. and Buot, IE Jr. (2014). Leaf Architecture of Philippine *Shorea* species (Dipterocarpaceae). International Research Journal of Biological Sciences, **3:**19- 26**.**

Roth-Nebelsick A, Uhl D., Mosbbrugger V. and Kerp H. (2001). Evolution and Function of Leaf Venation Architecture: A Review. Ann Bot 87 (5): 553-566. DOI:10.1006/anbo.2001.1391.

Sack, L., and Scoffoni, C. 2013. Leaf Venation: Structure, Function, Development, Evolution, Ecology and Applications. In: The Past, Present and Future. New Phytologist, 198(4), 983–1000. doi:10.1111/nph.12253

Salvana, F.R.P. and Buot, I.E.,Jr. (2014). Leaf Architectural Study of *Hoya coriacea, Hoya halconensis* and *Hoya buotii* (Apocynaceae), International Research Journal of Biological Sciences,3(3), 37-44. Retrieved on March 1, 2015 from http://goo.gl/Fi9QPG

Smith, A.R.; Pryer, K.M.; Schuettpelz, E.; Korall, P.; Schneider, H.; and Wolf, P.G. (2006). A classification for extant ferns. Taxon, 55(3), 705-731.

Stace, C.A. (1984). The Taxonomic Importance of the Leaf Surface. Systematics Association Special Volume No.25 "Current Concepts in Plant Taxonomy" ed. by VH Heywood and DM Moore. London and Orlando: Academic Press.

Takhtajan, A. (1996). Diversity and Classification of Flowering Plants. New York: Columbia University Press. Pp.231-234.

Tan, JMP. (2019). Leaf Architecture and Petiole Anatomy of Eupolypods I (Polypodiaceae) *sensu* PPG. MSc Thesis submitted to the University of the Philippines Los Banos. College, Laguna, Philippines. 158p.

Tan, JMP and Buot, I.E.,Jr.(2018). Delineating Two Species *Hoya benguetensis* Schltr. and *Hoya ilagiorum* Kloppenb., Siar & Cajano (Apocynaceae): A Leaf Architectural Approach. The Thailand Natural History Museum Journal 12(2): 103-109.

Tan, JMP. and Buot, I.E.Jr. (2019). Cluster and ordination analyses of leaf architectural characters in classifying Polypodiaceae *sensu* PPG. The Thailand Natural History Museum Journal, 13(1):27-42.

Tan, JMP and Buot, I.E.,Jr. (2020). Investigating the leaf architecture of Eupolypods I (Polypodiales): implications to taxonomy. Journal on New Biological Reports 9 (1): 1-22.

Tomlinson, P.B. (1984). Vegetative Morphology- Some Enigmas in the Relation to Plant Systematics. Systematics Association Special Volume No.25 “Current Concepts in Plant Taxonomy” ed. by VH Heywood and DM Moore. London and Orlando: Academic Press.

Torrefiel, J. and Buot, I.E.,Jr. (2017). *Hoya carandangiana*, *Hoya bicolensis* and *Hoya camphorifolia* (Apocynaceae) species delineation: insights from leaf architecture. The Thailand Natural History Museum Journal 11(1): 35–41.

Villareal, A.M.M. and Buot, I.E.,Jr. (2015). Leaf architecture of *Hoya incrassata* Warb. and *Hoya crassicaulis* Elmer x Kloppenb. (Apocynaceae): Taxonomic identification and conservation concerns. International Journal of Ecology and Conservation 15: 203-213.

Wanntorp, L. A.; Kocyan, R.van Donkelaar and Renner, S.S. (2006). Towards a monophyletic *Hoya* (Marsdenieae, Apocynaceae): inferences from the chloroplast trnL region and the atpB-rbcL spacer, *Syst. Bot*., 31(3) 586-596.

Wolfe, J.A. (1989). Leaf architectural analysis of the Hamamelidae. In: Crane RR and Blackmore S, eds. Evolution, Systematics and Fossil History of Hamamalidae. Clarendon Press, Oxford.