องค์ประกอบทางเคมีของกลิ่นหอมของดอกและอนุกรมวิธาน ของปาล์ม 47 ชนิด

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Floral scents of 47 palm species in relation to their systematics

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บทคัดย่อ

ทำการสกัดกลิ่นหอมจากดอกปาล์ม จำนวน 47 ชนิด (จัดอยู่ใน 30 สกุล, 8 เผ่า และ 4 อนุวงศ์) ด้วยเทคนิค ”dynamical headspace” จากนั้นนำไปวิเคราะห์หาองค์ประกอบทางเคมีด้วย GC-MS สารกลิ่นหอมที่ได้ถูกนำไปวิเคราะห์ทางสถิติ เพื่อหาคุณลักษณะเฉพาะทางเคมีของกลิ่นหอมของดอกปาล์ม และจากการวิเคราะห์เพิ่มเติมในเชิงปริมาณโดย PCA (Principal Component Analysis) แสดงให้เห็นว่า การจับกลุ่มของปาล์มทั้ง 47 ชนิด บนพื้นฐานขององค์ประกอบทางเคมีของกลิ่มหอมนั้นไม่สัมพันธ์กับกลุ่มทางอนุกรมวิธาน และไม่สอดคล้องกับความสัมพันธ์เชิงวิวัฒนาการของพืชในวงศ์ปาล์ม (Asmussen *et al.*, 2004, *in prep*. and C. Asmussen, *pers.comm.*) นอกจากนี้เมื่อพิจารณากลุ่มที่ได้จาก PCA เทียบกับชนิดของสัตว์ผสมเกสร ก็ไม่พบความสอดคล้องเช่นเดียวกัน การศึกษานี้แสดงให้เห็นว่ากลิ่นหอมของดอกปาล์มนั้นมีองค์ประกอบทางเคมีที่ความหลากหลายอย่างมาก

คำสำคัญ: ปาล์ม กลิ่นหอม การสกัดโดยเทคนิค ”dynamical headspace” GC-MS อนุกรมวิธาน ความสัมพันธ์เชิงวัฒนาการ สัตว์ผสมเกสร

Abstract

Floral volatile compounds from 47 palm species (belonging to 30 genera, 8 tribes and 4 subfamilies) were extracted by dynamical headspace and analysed by GC-MS. The volatile components were examined in terms of presence/absence in order to characterize the chemical profiles of the floral scents of palms. A PCA did not group the species in accordance with the known phylogenetic relationships among palm genus (Asmussen *et al.*, 2004, *in prep*. and C. Asmussen, *pers.comm.*) nor with the putative pollinator type of the species. A large spectrum of chemical compounds (fatty acid derivatives, aromatic compounds and terpenoids) was emitted from the palm flowers.

Keywords: palms; Palmae; floral bouquets; headspace extraction; GC-MS; phylogeny; pollination systems; monophyletic groups; calamoid clade; coryphoid clade; arecoid clade; Sabalinae group; Livistoninae group; borassodendroid-caryotoid group.

Introduction

Natural volatile compound is one of the well known categories of secondary metabolites contributing to essential ecological functions in plants. These compounds play significant roles in defence against herbivores, and can also function as attractants for pollinators. Throughout evolutionary history, under phylogenetic constraints and ecological selection pressures (e.g. plant-insect interaction), plants have developed a large, diverse spectrum of defensive/attractive bouquets.

During the last three decades, many studies have elucidated the variation of floral fragrance chemistry within various plant taxa. The trends in chemical profiles of floral scents of plants sharing similar pollination syndromes have been widely described, either among closely related plants, often members of the same genus (Jürgens *et al.*, 2002, Jürgens *et al.*, 2003), or to detect convergent evolution among taxonomically distant species, e.g. species belonging to different families (Knudsen & Tollsten 1995, Knudsen & Tollsten 1993, Andersson *et al.*, 2002). These studies indicate, that in general, the composition of the fragrance reflects to some degree phylogeny. For example, some volatile compounds characterise particular genus (Azuma *et al.*, 1997, Dobson *et al.* 1997, Williams & Whitten 1999, Dunlop *et al.*, 1999, Nogueira *et al.*, 2001).

Most studies carried out have focused on plant groups at a small taxonomic scale (i.e. generic level), but very few have considered groups as large as a whole plant family. We draw attention to the family level for the reason that many branches of biosynthetic pathways to secondary metabolites are particularly well developed in or restricted to, specific plant families. Undue emphasis on a single species can obscure the tremendous variation in biosynthetic facilities that exists in plants (Croteau *et al.*, 2000).

Palmae, the palms, is a distinctive family distributed in tropical and subtropical habitats throughout the world (Baker *et al.*, 1999). Impressively, despite the enormous diversity existing among the members, one can instantly recognize palms. The highly diverse systems of pollination, along with the large spectrum of typical floral fragrances exhibited by palms, exert great fascination. Several biologists have launched investigations of correlation between floral scents and, on the one hand, pollinator type or phylogenetic relationships of palm species, on the other. A number of seminal works have provided an important stock of information on chemistry of palm floral scents. (Ervik *et al.*, 1999, Knudsen 1999, Knudsen *et al.*, 2001).

Furthermore, during recent years, the molecular studies of palms have been extensively carried out (Uhl *et al.*, 1995, Asmussen *et al.*, 2000, Baker *et al.*, 2000, Baker & Dransfield 2000a,b, Asmussen & Chase 2001, Baker *et al.* 1999, Hahn 2002a,b, Lewis & Doyle 2001, Lewis *et al.*, 2001). Hence, the relationships among palm genera are well known and a well-resolved molecular phylogeny of the family is currently available (Asmussen *et al.*, 2004, *in prep.*, and C. Asmussen *pers.comm.*). Studies of floral fragrances of palms may further contribute to a comprehensive understanding of palm natural history.

Here, we describe **1)** the general aspect and the variations among the palms in floral volatiles. Additionally, we show **2)** whether or not the segregation of the species based on the volatiles dataset matches with the monophyletic groups recognized in the phylogeny and examine **3)** the level of analysis that is most relevant, individual compounds, chemical compound classes, or biosynthetic pathways. Even though pollination systems have not yet been exhaustively investigated among the palms, the available data on some species encourage us to **4)** preliminarily scrutinize the relationship between chemical composition of floral scents and pollinator affinities. Finally we discuss **5)** the relationship between floral scent composition and phylogeny, and the evolutionary forces that have driven the variation of odours.

Materials and Methods

Studied species

We extracted and identified the floral volatiles of 47 palm species[1]. These species belong to 4 subfamilies (Uhl & Dransfield 1987): Calamoideae (with 1 tribe; 2 genera), Coryphoideae (with 4 tribes, including Caryoteae; 3 subtribes; 20 genera), Ceroxyloideae (with 1 tribe; 1 genus) and Arecoideae (with 2 tribes; 7 genera). Sampling was most extensive within the most diverse subfamily Coryphoideae (Dransfield & Uhl 1987), within which phylogenetic relationships are quite well known. Whenever possible, we sampled several species from each genus. The phylogenetic relationships among the

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[1] The species have been identified by M. M.D. Ferrero, the curator of the palm collection of the 'Nong Nooch' botanical garden. The identification was verified, from photographs, by Dr. J. Dransfield and Dr. A. Barfod (for the genus *Licuala*). The species whose names were uncertain were put as 'cf.' or 'sp.' Herbarium specimens were not collected, since would have greatly damaged the plants, which are protected as living specimens in the garden.

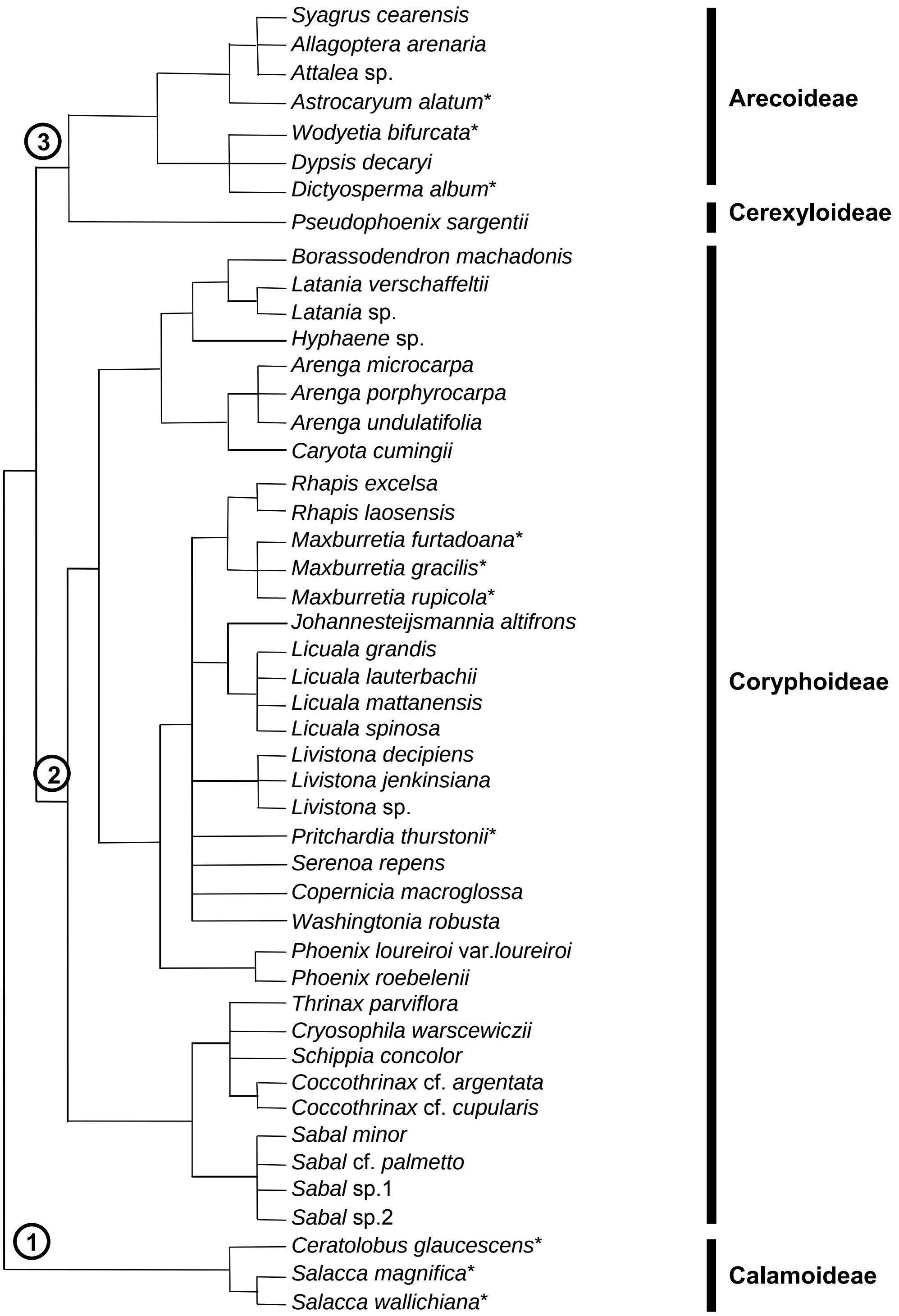


Figure 1: Phylogenetic tree of the 47 species of which floral odours were analysed. Phylogenetic relationships among the species studied were deduced from Asmussen & Chase (2001) and from further studies of Asmussen (pers.comm.).

* Genera not studied by Asmussen & Chase (2001) and Asmussen (pers.comm.).

(1) Calamoid group, **(2)** Coryphoid group, **(3)** Arecoid group (including Arecoideae and Cereroxyloideae).

species studied are those established by Asmussen & Chase (2001) and by further studies of C. Asmussen (*pers. comm.*).

The phylogenetic tree show in *Figure 1* includes 3 main monophyletic groups, the first corresponding to the subfamily Calamoideae, the second including members of Coryphoideae plus Caryotoid palms, and the third corresponding to the Arecoid line (*sensu* Moore 1973) comprising Arecoideae and Ceroxyloideae.

Collection of floral volatiles

Sampling was conducted in Nong Nooch Tropical Garden, one of the world's largest collections of palms, situated in the province of Chon Buri, southeastern Thailand. The floral volatiles were collected by dynamical headspace extraction. The inflorescences (still on the plants or cut, table 1) were enclosed in polyethylene terephtalate (Nalophan) bags (Etablissements Charles frères, Saint Etienne, France). Pure air (ambient air passed through two charcoal filters) was blown into the bag at a flow rate of 400 mL min^{-1}, using battery-operated pumps. The air containing floral volatiles was drawn at 300 mL min^{-1} through an adsorbent containing 30 mg of Alltech SuperQ (ARS Inc., Gainesville, FL, USA). The exceeding entering air flux compensated the unavoidable leaks and ensured that the extraction continued without contamination from the outside air. A blank was collected from an empty bag, in parallel with each collection of floral scents. All sampling was done under the same conditions. Extraction lasted for 3 h (4 h for some species whose odors were faint to the human nose; extraction details are given in table 1). The trapped volatiles were eluted with 150 µl of dichloromethane (CH_2Cl_2) and preserved at -20°C until analysis. The eluents were analyzed by coupled gas chromatography and mass spectrometry (GC-MS) on a Varian CP3800 gas chromatograph (GC) connected to a Varian Saturn 2000 mass spectrometer (MS). The MS (Varian) was equipped with a CP sil 8 CB column (length 30m, 0.25mm inner diameter, 0.25µm film thickness), with helium as carrier gas. The temperature of MS was programmed at 50°C for 3mn then increased 3°mn^{-1} to 100°C, 2.7°mn^{-1} to 140°C, 2.4°mn^{-1} to 180 and finally 6°mn^{-1} to 250°C.

Identification of molecules and biosynthetic pathways

The volatile compounds were identified by comparing mass spectra of the samples with those of NIST98, our personal library including more than 30 essential oils plus the databases of Wiley 6 (1994), Adams (1995), and Joulain & Konig (1998). When possible, we also checked with spectra of authentic compounds. In addition, we compared the retention indices obtained with those known for authentic compounds (Adams 2001, Jenning & Shibamoto 1980).

For quantification of chemical substances, the peak areas were integrated for the total ion current signal (chromatograms) using the MS data handling on a Saturn GC/MS workstation (Varian, Inc., 2001). The areas obtained were then used to determine the relative amount of a given chemical compound, the sum of areas of all identified compounds present in the sample analysed being equal to 100%. The volatile compounds were thus reported in amounts relative to the total amount of fragrance detected (%).

The chemical substances were classified in different hierarchical groups. First compounds were classified according to their **biosynthetic origins**: polyketide pathway, i.e. fatty acid derivatives; shikimic acid pathway, i.e. aromatic compounds; and mevalonic acid and methylerythritol phosphate (MEP; Lichtenthaler *et al*., 1997, Taiz & Zeiger 2002) pathways, i.e. terpenes or isoprenoids. The second classification is based on various **chemical classes**: isoprenoids, i.e. monoterpenes, sesquiter-penes and carotenoid derivatives; different kinds of fatty acid derivatives, i.e. alkanes, alkenes, alcohols, esters, aldehydes, ketones, lactones, and jasmonoid compounds; aromatic compounds sorted by their deviation from the basic phenylpropanoid (C_6-C_3) carbon skeleton to yield: C_6-C_3, C_6-C_2, or C_6-C1, N- and S-containing aromatic compounds.

Data analysis

We first treated the floral volatiles data qualitatively, i.e., in term of presence/ absence of compounds in the bouquets. General character and variation of floral bouquets in the palm family were then described. To further analyze possible grouping of species according to their bouquet, we then performed a principal component analysis, PCA on covariance matrices (SAS Institute Inc, 2001). Since it is not possible to know a *priori* at what level the information will be relevant, several PCA were performed on different variables: 1) relative amount (%) of the individual compounds; 2) proportion of compounds corresponding to each of the different chemical classes; 3) proportion of compounds from each of the three biosynthetic pathways. We then checked whether the PCA was grouping the species according to the three major monophyletic groups of palms and /or the different genera. Furthermore, based on the available pollination information, we tried to evaluate the relationship between pollination systems and floral bouquets.

Results

General character of palm floral bouquet

Among the bouquets of the 47 species studied, we detected a total of 398 compounds. The odour of a given species could include from 7 (*Pseudophoenix sargentii, Arenga porphyrocarpa* and *Pritchardia thurstonii*) to 100 (*Phoenix loureiroi*) substances, with a mean number of 34 (STDE=19) compounds per species (*Fig 2*).

These volatiles formed three groups according to their biosynthetic pathways: **fatty acid derivatives** originated from the polyketide pathway, **aromatic compounds** derived from the shikimic acid pathway, and **isoprenoids** from the mevalonic acid and the MEP pathways. Isoprenoid compounds represented

58% of the total number of compounds, fatty acid derivatives 21% and aromatic compounds 18% (*Fig 3*). Among isoprenoids, the most diverse class of compounds was sesquiterpenes.

palms, we performed a more quantitative analysis using principal component analysis (PCA). Since it was not possible to determine *a priori* what level of analysis would be most pertinent, we performed different PCA for the different compound classification ranks, i.e. pathway, different chemical classes and individual compounds.

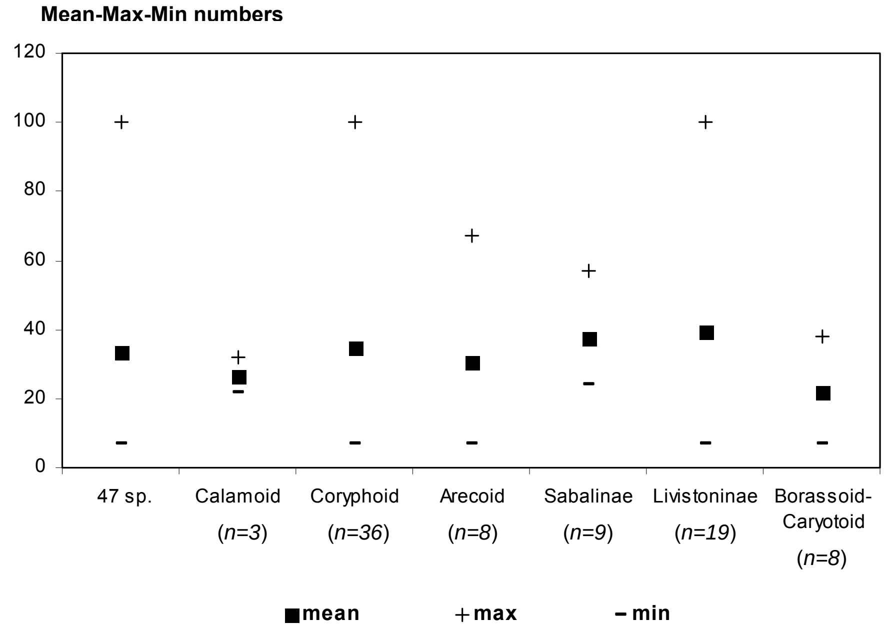


Figure 2: Mean, maximum, and minimum numbers of compounds comprised in floral bouquet of each clade examined as well as of 47 species studied

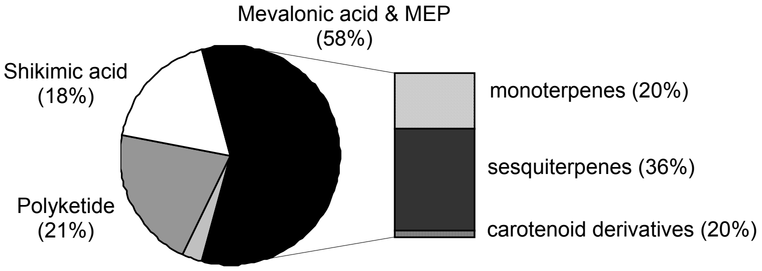


Figure 3: Proportion of number of compounds from different biosynthetic pathways. The main classes of compounds from the mevalonic acid and the MEP pathways are monoterpenes, sesquiterpenes and carotenoid derivatives

Principal component analysis (PCA)

To determine whether phylogenetic relationships affected the floral odours of

The PCA based on biosynthetic **pathways** and chemical **classes** did not reveal phylogenetic relationships among the species. Grouping of chemical substances at a finer scale, i.e. the dataset of all individual compounds, enhanced the revelation of phylogenetic relationships among the species (*Table 1a, b*). Hence, we will explain in detail only the PCA on all compounds which contained the most phylogenetic information.

Table 1: Summary of the results from PCA, showing the separation of species on the PCA initiated from different datasets, i.e. different levels of compound categorization. a) Considering the three main monophyletic groups, b) Considering the generic level. ✓ The members of the genus are grouped or closely segregate in the plot. ✗ The members of the genus are disperse far from each others in the plot.

Clade	Pathway *(n=3)*	Class *(n=5)*	Individual compounds *(n=398)*
Calamoid *(n=3)*	✗	✗	✓ **(1)*
Coryphoid *(n=36)*	✗	✗	✗
Arecoid *(n=8)*	✗	✗	✗

1a) The 3 main monophyletic

**(1)* Floral bouquet contain high relative amount of 'p-methylanisole'

Genus	Pathway *(n=3)*	Class *(n=5)*	Individual compounds *(n=398)*
Salacca *(n=2)*	✗	✗	✓ *nb=11, Σ =59-99%; *(9)*
Sabal *(n=4)*	✗	✗	✗ *nb=9, Σ =0.9-56%*
Coccothrinax *(n=2)*	✗	✗	✓ *nb=17, Σ =92-93%; *(9)*
Phoenix *(n=2)*	✓ **(1)*	✗	✗ *nb=26, Σ =0.3-29%*
Livistona *(n=3)*	✗	✓ **(2)*	✗ *nb=8, Σ =14-42%*
Licuala *(n=4)*	✗	✗	✗ *nb=1, Σ =0.1-7%*
Maxburretia *(n=3)*	✗	✗	✓ *nb=1, Σ =0.03-7%; *(9)*
Rhapis *(n=2)*	✓ **(1)*	✓ **(3)*	✓ *nb=10, Σ =12-21%; *(9)*
Arenga *(n=3)*	✗	✗	✗ *nb=0, Σ =0%*
Latania *(n=2)*	✗	✗	✗ *nb=7, Σ =28-52%*

1b) Genera

nb: represents total number of compounds that are common to all members of a given genus.

Σ: represents sum of the relative amounts (%) of the common compounds. The fist value is the lowest percentage found in a member of the genus and second one is the highest percentage.

**(1)* Floral bouquets contain high relative amount of compounds derived from 'Mevalonic acid pathway' (99% in *Phoenix* and 93-96% in *Rhapis* species)

**(2)* Floral bouquets contain high relative amount of 'aromatic compounds' (18-35%) and 'monoterpenes' (28-60%)

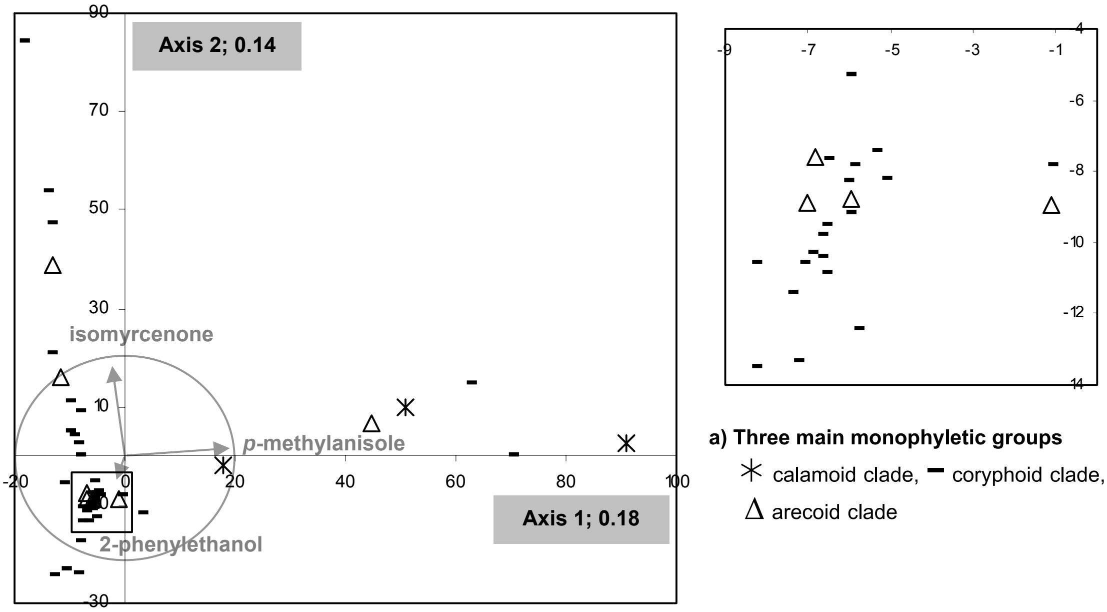
**(3)* Floral bouquets contain high relative amount of 'sesquiterpenes' (93-95%)

**(4)* Total number of the compounds which are common to all members of most grouped genera is relatively high, accompanied by the relatively high sum of percentages of those compounds.

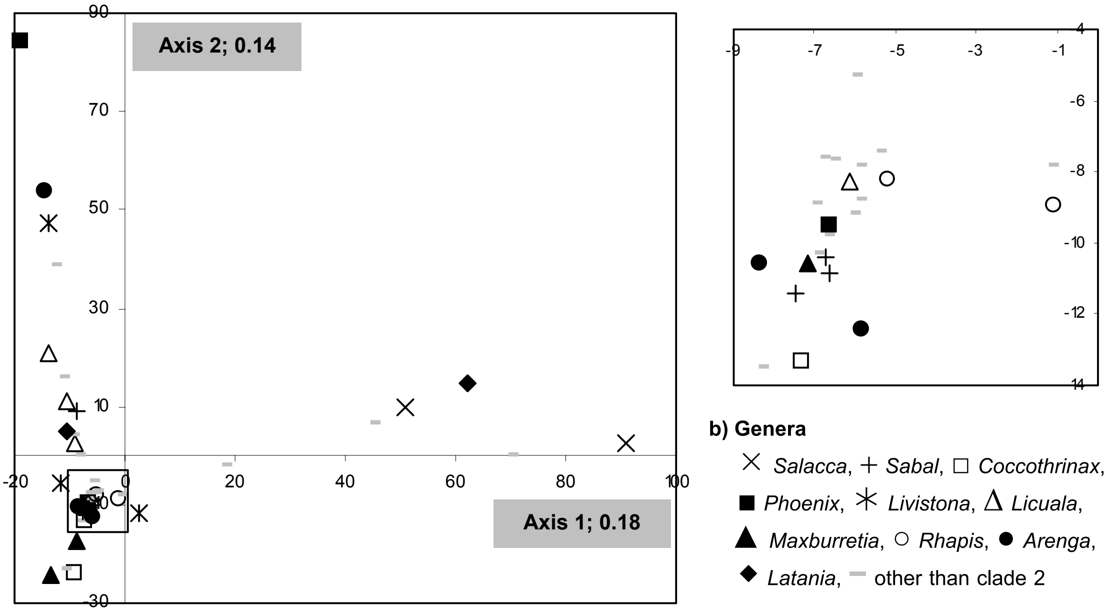
PCA on the percentages of individual compounds

The first and second axes explained respectively 18 and 14 % of the variation. They were strongly correlated with *p*-methylanisole and isomyrcenone (*Fig 4a*), which were present respectively in 13 and 21 species. The second axis was also correlated with 2-phenylethanol, which was present in 18 species. Moreover, this axis was slightly correlated with (*E*)-ocimene (present in 30 species), pentadecane (present in 15 species) and benzyl alcohol (present in 10 species).

The first axis separated few species, among them the three calamoids but also some coryphoids and arecoids. There was no clear grouping of the three major monophyletic



a) Three main monophyletic groups



b) Genera

Figure 4: ACP plot (covariance) originated from relative amount of individual compounds. The section in the frame was enlarged and placed at the right of the plot.

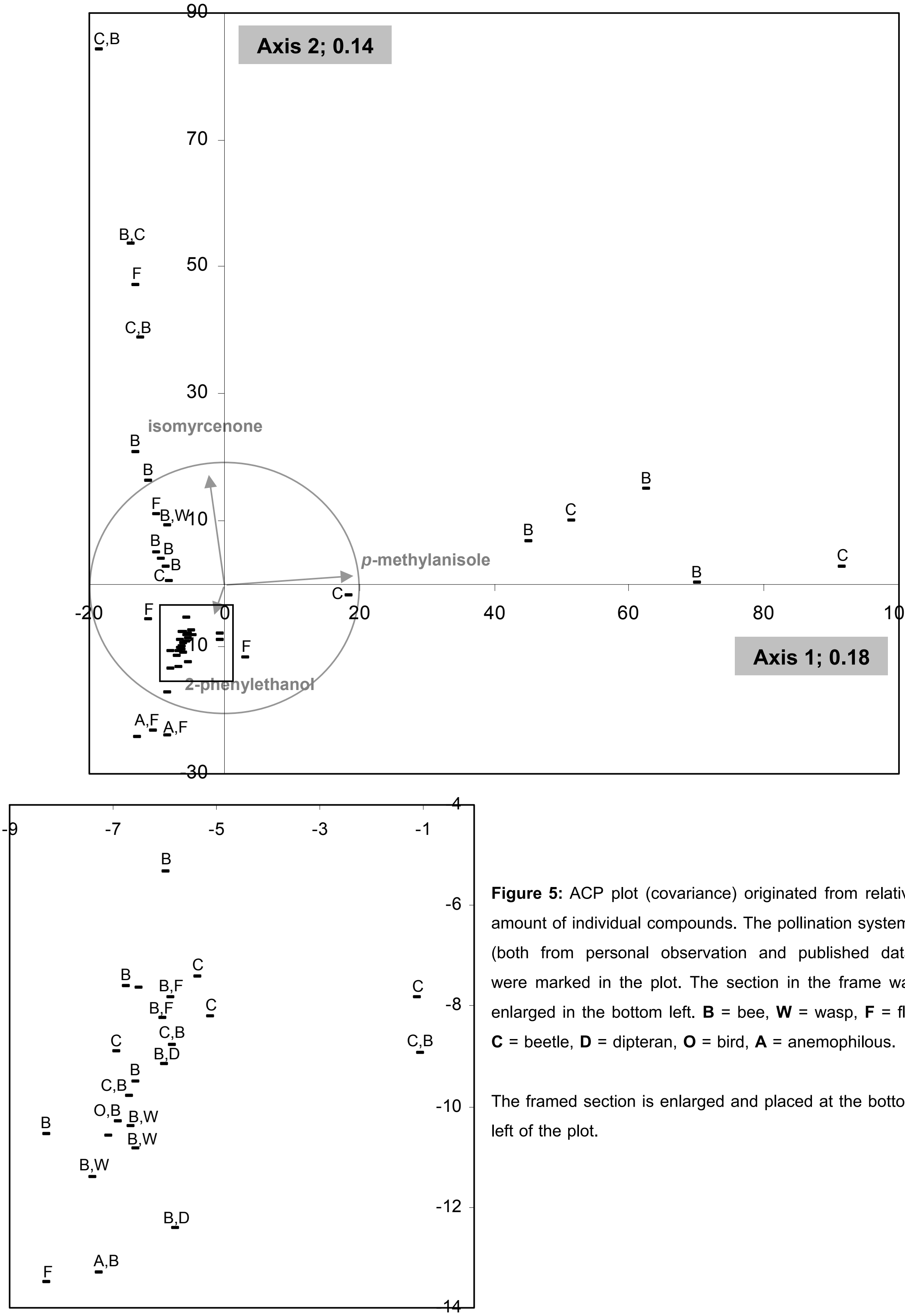


Figure 5: ACP plot (covariance) originated from relative amount of individual compounds. The pollination systems (both from personal observation and published data) were marked in the plot. The section in the frame was enlarged in the bottom left. **B** = bee, **W** = wasp, **F** = fly, **C** = beetle, **D** = dipteran, **O** = bird, **A** = anemophilous.

The framed section is enlarged and placed at the bottom left of the plot.

groups within palms (*Fig 4a*). For some genera, all members were found in proximity on the PCA plot (*Fig 4b*), i.e. *Salacca* (at the right of the plot; n=2), *Coccothrinax* (n=2), *Maxburretia* (n=3) and *Rhapis* (n=2; the last three genera were all at the bottom left). Moreover, 3 out of 4 species of *Sabal* grouped together (at the bottom left, the ungrouped species is *Sabal* sp.1; see the enlarged frame of *Fig 4b*), as did all but one species of *Licuala* (at the top left, the set apart species is *L. mattanensis*; *Fig 4b*). Species from a genus tended to be close, but did not form clear groups. Analysis at this level showed no apparent effect of phylogeny on the composition of floral odours of palm.

Relationship with different pollination systems

Based on the data available (i.e., the combination of personal observation during fieldwork and study of the literature), the pollination modes of most palm species were applied on the PCA plots to see if particular blends of volatiles reflect the identity of pollinators. Our preliminary results showed that, none of the PCA realized at different levels (i.e., on datasets of pathways, compound classes, compound subclasses, compound subclasses-bis and all individual compounds) revealed such relationships. The species pollinated by the same kind of pollinators were not discernably separated in any of the plots. For example, on the PCA of individual compounds (*Fig 5*), the species separated at the right of the plot are pollinated by bees or by beetles. *Wodyetia bifurcata*, a bee-pollinated palm (Dowe 1993) for example was in proximity with beetle-pollinated *Salacca* species (Uhl & Dransfield 1987). The species clustered together in the framed section are wind-pollinated, *Coccothrinax cupularis*, bee/wasp-pollinated, e.g., *Sabal palmetto* and *S. minor* (Henderson, 2002), beetle-pollinated, e.g., the *Rhapis* species (Henderson 1986), and *Astrocaryum alatum* (Henderson 1986, Henderon 2002), or fly-pollinated, e.g., *Schippia concolor* (per. obs.). The species segregated in the top left of the plot belong to a similarly diverse mix of pollination systems.

Discussion

An enormous assortment of fragrant substances from three chemically distinct biosynthetic pathways (fatty acid derivatives, aromatic compounds, and terpenoids) is emitted from palm flowers. The diversity is related not only by the high number of compounds (a total of 398 compounds found in the 47 species investigated) but also in their assemblage in bouquets comprising from 7 to 100 substances. However, in many palms, one or a few compounds dominate the bouquet (Table 1). About one-third of the fragrant compounds are restricted to a single species but others are shared by many species. Floral scents of the palms are dominated by terpenoids, the largest class of plant secondary metabolites (Croteau *et al*., 2000, Taiz & Zeiger 2002), and especially sesquiterpenes.

We will discuss to what extent the composition of these odours can reveal on the one hand the phylogenetic relationships among species and on the other hand the identity of their pollinators.

Compounds of the different chemical classes are present in somewhat similar proportions, in term of number of compounds, in all the three major monophyletic groups of palms studied. Their bouquets are constantly dominated by sesquiterpenes and, in a lesser proportion, monoterpenes.

The PCA failed to reveal clear separations between species according to their phylogenetic relationships, even though species of the same genus are sometimes grouped (e.g. *Salacca*, *Coccothrinax*, *Rhapis* or *Maxburretia*). Taken as a whole, fragrance characters were not reliable indicators of phylogeny and are therefore likely to be, mainly if not entirely, analogous characters, in which similarity are attained by convergent evolution. A further analysis of character evolution according to phylogeny will be done once the new phylogeny of Assmussen *et al.*, (*in prep.*) is finished.

Few data are available on palm pollinators. For many species, we had to assume that the pollinator group was the same as that described for other species of the same genus. Hence, our analysis can only be regarded as preliminary. The different PCA did not reveal any grouping of the species according to their putative pollinator group. Thus no general rules about the relationships between odours and pollinator identity appear to exist. Furthermore, though plants attract similar pollinator groups, the major components in their floral bouquets may be of different biochemical origins, as in the case of six Annonaceae species that attract small beetles and thrips (Jürgens *et al.*, 2000).

Phylogenetic constraint and ecological selection pressures are likely to affect the evolution of floral scents despite the failure of our analysis to detect these effects. Regarding the phylogenetic constraint, though there present a large, various arrays of chemical compounds, the number of possible metabolic routes in plants is limited, to some extent, by phylogenetic history. Hence, ancestry settles on, at least in part, the biosynthetic apparatus available for selection (Berenbaum & Seigler 1992). Likewise, there exists so much evidence on insect-chemical compounds relationship, either by means of attraction or defence, having been far and wide attributed since Darwin’s time, that one cannot ignore the importance of pollinators as selective pressure for the composition of floral odours (Berenbaum & Seigler 1992, Dobson 1994). All the compounds produced by the flowers are not necessarily attractive to pollinators. Some compounds can be either simple noise, i.e. biosynthetic artefacts (Raguso *et al.*, 2003), or be integrated in the blends for other functions, e.g., repelling insects other than the desired pollinator (Faegri & van der Pijl 1979), or defending against herbivores (Mullin *et al.*, 1991, Ghazoul 2001). The presence of such compounds in the bouquet may obscure our analysis of relationship between odours and pollinator affinities. Likewise, concerning phylogenetic history, only certain ranges of compounds serve as applicable phylogenetic markers. We should therefore not use the entire set of substances contained in the blend (as in case of this study), but should focus on a 'tracer'. Perhaps some particular compounds, i.e. those known to exhibit phylogenetic inertia among plant taxa in general or those attractive to the effective pollinators of the species studied, could provide more revealing information on phylognetetic history or on pollinator affinities. This requires more intensively investigation. Unfortunately, while analyses of floral scents have been conducted on various plant taxa, the behavioural tests for clarifying which chemical volatiles are active in flower-insect interactions are often lacking (Dobson 1994).

Also, the variability of volatile compounds emitted by plant is the result of complex interactions between the organism and its environment, and which may be affect by developmental stage, by factors controlling emission, by stress such as injuries and air pollution, and even short-term influences such as light and temperature (Kesselmeier & Staudt 1999). Therefore, due to the complex and multifaceted nature of the interactions, phylogenetic inertia and/or pollen dispersal agents seem to be only a tiny part of the vast and tangled evolutionary forces that have driven such great variation.

Acknowledgements

The authors express their gratitude to Mr. Kumpon Tansacha for his generous welcome and his permission to sample odors and plant materials, Mr. M. D. Ferrero, the curator of palm and all staffs of Nong Nooch Tropical Garden for all their precious help. We are also very grateful to Dr. Dransfield and Dr. Barfod for invaluable helps in identification of palm species and to Dr. Asmussen for providing us the phylogenetic data indispensable for our study. We thank Bruno Buatois for help in MS works and D. McKey for scientific and linguistic comments on the manuscript. The authors are thankful to Mr. Pornchai Chutamas and the colleagues from the 'Plant Genetics Conservation Project as The Royal Initiation of HRH Princess Maha Chakri Sirindhorn', Bangkok,Thailand for the support that made this study possible.

References

Adams, R.P. 2001. Identification of Essential Oil Components by Gas Chromatography/ Quadrupole Mass Spectrometry. Allured Publishing Company, Carol Stream, USA, pp.1-456.

Andersson, S., Nilsson, L.A., Groth, I., and Bergström, G. 2002. Floral scents in butterfly-pollinated plants: possible convergence in chemical composition. Botanical Journal of the Linnean Society. 140: 129-153.

Asmussen, C.B. and Chase, M.W. 2001. Coding and noncoding plastid DNA in palm systematics. American Journal of Botany. 88: 1103-1117.

Asmussen, C.B., Baker, W.J., and Dransfield, J. 2000. Phylogeny of the palm family (Arecaceae) based on rps16 intron and trnL-trnF plastid DNA sequences. In: Monocots: Systematics and Evolution. (Eds. Wilson, K. L. & Morrison, D. A.). CSIRO, Melbourne, pp. 525-544.

Azuma, H., Toyota, M., Asakawa, Y., Yamaoka, R., Garcia-Franco, J. G., Dieringer, G., Thien, L. B., and Kawano, S. 1997. Chemical divergence in floral scents of Magnolia and allied genera (Magnoliaceae). Plant Species Biology 12, 69-83.

Baker, W. J., Asmussen, C. B., Barrow, S. C., Dransfield, J., and Hedderson, T. A. 1999. A phylogenetic study of the palm family (Palmae) based on chloroplast DNA sequences from the trnL-trnF region. Plant Syst.Evol. 219, 111-126.

Baker, W.J. and Dransfield, J. 2000a. Phylogeny, character evolution, and a new classification of the calamoid palms. Systematic Botany. 25: 297-322.

Baker, W.J. and Dransfield, J. 2000b. Towards a biogeographic explanation of the Calamoid palms. In: Monocots: Systematics and Evolution. (Eds. Wilson, K.L. & Morrison, D.A.) CSIRO, Melbourne, pp. 545-553.

Baker, W.J., Asmussen, C.B., Barrow, S.C., Dransfield, J., and Hedderson, T.A. 1999. A phylogenetic study of the palm family (Palmae) based on chloroplast DNA sequences from the trnL-trnF region. Plant Syst.Evol. 219: 111-126.

Baker, W.J., Hedderson, T.A., and Dransfield, J. 2000. Molecular phyloenetics of subfamily Calamoideae (Palmae) based on nrDNA ITS and cpDNA rps16 intron sequence data. Molecular Phylogenetics and Evolution. 14: 195-217.

Berenbaum, M. and Seigler, D. 1992. Biochemicals: engineering problems for natural selection. In: Insect Chemical Ecology: An Evolutionary Approach. (Eds. Roitberg, B.D. & Isman, M.B.). Chapman & Hall, New York, London, pp. 89-121.

Croteau, R., Kutchan, T.M., and Lewis, N.G. 2000. Natural Products (Secondary Metabolites). In: Biochemistry & Molecular Biology of Plants. (Eds. Buchanan, B., Gruissem, W., & Jones, R.). American Society of Plant Physiologists, pp. 1250-1318.

Nogueira, P.C.L., Bittrich, V., Shepherd, G.J., Lopes, A.V., and Marsaioli, A.J. 2001. The ecological and taxonomic importance of flower volatiles of Clusia species (Guttiferae). Phytochemistry. 56: 443-452.

Dobson, H.E.M. 1994. Floral volatiles in insect biology. In: Insect-Plant Interactions. (Ed. Bernays, E. A.) CRC Press, Arizona, pp. 48-81.

Dobson, H.E.M., Arroyo, J., Bergström, G., and Groth, I. 1997. Interspecific variation in floral fragrances within the genus Narcissus (Amaryllidaceae). Biochemical Systematics and Ecology. 25: 685-706.

Dunlop, P.J., Bignell, C.M., Brooker, M.I.H., Brophy, J.J., and Hibbert, D.B. 1999. Use of gas chromatograms of essential leaf oils to compare eight taxa of genus Angophora (Myrtaceae): possible relationship to the genus Eucalyptus. Biochemical Systematics and Ecology. 27: 815-830.

Ervik, F., Tollsten, L., and Knudsen, J.T. 1999. Floral scent chemistry and pollination ecology in phytelephantoid palms (Arecaceae). Plant Systematics and Evolution. 217: 279-297.

Faegri, K. and Van der Pijl, L. 1979. The principles of pollination biology. Pergamon Press, Oxford, pp. 1-244.

Ghazoul, J. 2001. Can floral repellents pre-empt potential ant-plant conflicts? Ecology Letters. 4: 295-299.

Hahn, W.J. 2002a. A molecular phylogenetic study of the Palmae (Arecaceae) based on atpB, rbcL, and 18S nrDNA sequences. Systematic Biology. 51: 92-112.

Hahn, W.J. 2002b. A phylogenetic analysis of the Arecoid line of palms based on plastid DNA sequence data. Molecular Phylogenetics and Evolution. 23: 189-204.

Henderson, A. 1986. A review of pollination studies in the Palmae. The Botanical Review. 52 221-259.

Henderson, A. 2002. Evolution and Ecology of Palms. The New York Botanical Garden Press, New York, pp. 1-259.

Joulain D. and Konig W.A. 1998. The Atlas of Spectral Data of sesquiterpene hydrocarbons. E.B. Verlag, Hamburg.

Jürgens, A., Webber, A.C., and Gottsberger, G. 2000. Floral scent compounds of Amazonian Annonaceae species pollinated by small beetles and thrips. Phytochemistry. 55: 551-558.

Jürgens, A., Witt, T., and Gottsberger, G. 2002. Flower scent composition in night-flowering Silene species (Caryophyllaceae). Biochemical Systematics and Ecology 30: 383-397.

Jürgens, A., Witt, T., and Gottsberger, G. 2003. Flower scent composition in Dianthus and Saponaria species (Caryophyllaceae) and its relevance for pollination biology and taxonomy. Biochemical Systematics and Ecology. 31: 345-357.

Kesselmeier, J. and Staudt, M. 1999. Biogenic volatile organic compounds (VOC): An overview on emission, physiology and ecology. Journal of Atmospheric Chemistry. 33: 23-88.

Knudsen, J.T. and Tollsten, L. 1993. Trends in floral scent chemistry in pollination syndromes: floral scent composition in moth-pollinated taxa. Botanical Journal of the Linnean Society. 113: 263-284.

Knudsen, J.T. and Tollsten, L. 1995. Floral scent in bat-pollinated plants: a case of convergent evolution. Botanical Journal of the Linnean Society. 119: 45-57.

Knudsen, J.T. 1999. Floral scent chemistry in Geonomoid palms (Palmae: Geonomeae) and its importance in maintaining reproductive isolation. Memoirs of the New York Botanical Garden. 83: 141-168.

Knudsen, J.T., Tollsten, L., & Ervik, F. 2001. Flower scent and pollination in selected neotropical palms. Plant Biology. 3: 642-653.

Lewis, C.E. and Doyle, J.J. 2001. Phylogenetic utility of the nuclear gene malate synthase in the palm family (Arecaceae). Molecular Phylogenetics and Evolution. 19: 409-420.

Lewis, C.E., Baker, W.J., and Asmussen, C.B. 2000. DNA and palm evolution. Palms. 44: 19-24.

Mullin, C.A., Alfatafta, A.A., Harman, J.L., Serino, A.A., and Everett, S.L. 1991. Corn rootworm feeding on sunflower and other Compositae: influence of floral terpenoid and phenolic factors. In: Naturally Occurring Pest Bioregulators. (Ed. Hedin, P.A.). American Chemical Society, Washington, DC, pp. 278-292.

Raguso, R.A., Levin, D.A., Foose, S.E., Holmberg, M.W., and McDade, L.A. 2003. Fragrance chemistry, nocturnal rhythms and pollination "syndromes" in Nicotiana. Phytochemistry. 63: 265-284.

Taiz, L. and Zeiger, E. 2002. Plant Physiology. Sinauer Associates, Inc., Massachusetts, pp. 1-690.

Uhl, N.W. and Dransfield, J. 1987. Genera Palmarum. Allen Press, Kansas, pp. 1-610.

Uhl, N.W., Dransfield, J., Davis, J.I., Luckow, M.A., Hansen, K.S., and Doyle, J. J. 1995. Phylogenetic relationships among palm: cladistic analyses of morphological and chloroplast DNA restriction site variation. In: Monocotyledons: systematics and evolution. (Eds. Rudall, P.J., Cribb, P.J., Cutler, D.F., & Humphries, C.J.). Royal Botanic Garden, Kew, London, pp. 623-661.

Varian Inc, 2001. Saturn GC/MS Workstation, Version 5.52. Varian Inc, USA.

Williams, N.H. and Whitten, W.M. 1999. Molecular phylogeny and floral fragrances of male euglossine bee-pollinated orchids: A study of Stanhopea (Orchidaceae). Plant Species Biology. 14: 129-136.