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Klasyfikacja i filogeneza rodzaju *Dendrobium* Sw. sekcja *Dendrobium* (Orchidaceae)

Classification and phylogeny of the genus *Dendrobium* Sw. section *Dendrobium* (Orchidaceae)

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Spis treści

Streszczenie

Tematem niniejszej rozprawy doktorskiej jest klasyfikacja i filogeneza nominalnej sekcji rodzaju *Dendrobium* Sw. (Orchidaceae).

Rodzaj *Dendrobium* został opisany ponad 220 lat temu (Swartz 1799) i do dzisiaj w jego obrębie znanych jest blisko 1600 gatunków. Zasięg ich występowania jest bardzo szeroki, rozciąga się od Korei i Japonii przez Azję Południowo-Wschodnią, na zachód do Himalajów, na wschód do Filipin i na południe przez Indonezję, Wyspy Pacyfiku, Nową Gwineę, Australię aż do Nowej Zelandii (Moudi i in. 2013). Przedstawiciele rodzaju posiadają boczny, najczęściej groniasty kwiatostan, mentum, cztery nagie pyłkowiny i nabrzmiały dziubek (łac. *rostellum*). Jednak pomimo tych wspólnych cech wykazują także silne zróżnicowanie, zarówno cech wegetatywnych, jak i kwiatowych.

Ze względu na dużą liczbę gatunków należących do *Dendrobium*, głównym celem pracy była rekonstrukcja filogenezy i klasyfikacja sekcji nominalnej, obejmującej ponad 50 gatunków (Wood, 2006). Prace badawcze wykonano wykorzystując technikę skaningowej mikroskopii elektronowej (analiza struktur mikromorfologicznych na warżkach), metody biologii molekularnej (PCR, sekwencjonowanie DNA), analizy filogenetyczne oraz fenetyczne (rekonstrukcja cech morfologicznych przodków, analizy UPGMA).

W oparciu o uzyskane i zgromadzone wyniki przygotowano opisy struktur mikromorfologicznych występujących na warżkach u wybranych przedstawicieli rodzaju, wskazano optymalny marker do oszacowania filogenezy sekcji typowej. Ponadto porównano użyteczność niskokopijnego genu *Xdh* z wielokopijnym fragmentem ITS (ITS1-5.8S-ITS2) oraz markerami plastydowymi. Zestawiono dane filogenetyczne i morfologiczne taksonów, co pozwoliło na weryfikację przynależności taksonomicznej wybranych gatunków – zwłaszcza należących do sekcji nominalnej. Szczegółowe wyniki i wnioski z dotychczasowej pracy zostały opublikowane w formie dwóch artykułów w czasopismach z otwartym dostępem. Ostatni przygotowany manuskrypt został złożony i wysłany do recenzji. Wszystkie prace zostały także zamieszczone w kolejnych częściach rozprawy.

English summary

The subject of this doctoral dissertation is: „Classification and phylogeny of the genus *Dendrobium* Sw. section *Dendrobium* (Orchidaceae)".

The genus *Dendrobium* was described more than 220 years ago (Swartz'a 1799), and to date, nearly 1,600 species have been described within it. Their range is wide, extending from Korea and Japan through Southeast Asia, west to the Himalayas, east to the Philippines, and south through Indonesia, the Pacific Islands, New Guinea, Australia, to New Zealand (Moudi et al. 2013). Representatives of the genus have a lateral, mostly clustered inflorescence, a mentum, four naked pollinia, and a swollen rostellum. However, despite these common features, they also show strong variation, both in vegetative and floral traits.

Due to the large number of species belonging to *Dendrobium*, the main goal of the study was to reconstruct the phylogeny and classification of the nominal section, which includes more than 50 taxa (Wood, 2006). The research work was carried out using the scanning electron microscopy technique (analysis of micromorphological structures on labellum), molecular biology methods (PCR, DNA sequencing), and phylogenetic and phenetic analyses (reconstruction of ancestral morphological features, UPGMA analyses).

Based on the obtained and collected results, descriptions of the micromorphological structures present on the labellum of selected representatives of the genus were prepared, and the optimal marker for estimating the phylogeny of the type section was indicated. In addition, the utility of the low-copy *Xdh* gene was compared with the multicopy ITS fragment (ITS1-5.8S-ITS2) and plastid markers. Phylogenetic and morphological data of taxa were compiled, which allowed us to verify the taxonomic affiliation of selected species - especially those belonging to the nominal section. The detailed results and conclusions of the work to date are either already published or prepared for publication in open access journals, and are included in the following sections of the dissertation.

CZĘŚĆ I

1. Wprowadzenie

Dendrobium należy do rodziny Orchidaceae Juss., jednej z najbardziej rozpowszechnionych, różnorodnych i wyjątkowych grup roślin. Obejmuje ona niemal 30 000 gatunków, zgrupowanych w blisko 700 rodzajach (Chase i in. 2015; Pérez-Escobar i in. 2024). Przedstawiciele rodziny występują na wszystkich kontynentach, z wyjątkiem Antarktydy (Lipińska i in. 2016, Pessoa i in. 2018). Ponadto wykazują silne zróżnicowanie cech wegetatywnych, jak i kwiatowych. Można również zauważyć tendencję, według której dalsza odległość od równika wpływa na mniejszą liczebność i różnorodność gatunkową. Zatem największe ich bogactwo obserwowane jest w tropikach (Szlachetko i Skakuj 1996; Lipińska i in. 2016). Dominującą formą życiową omawianych roślin są epifity, ale nierzadkie są też geofity, litofity, czy liany. Cechują się zarówno monopodialnym, jak i znacznie częściej sympodialnym typem wzrostu. Ponadto mogą wytwarzać różne organy przetrwalno-spichrzowe, takie jak kłącza, bulwy czy pseudobulwy. Korzenie powietrzne epifitów u Orchidaceae pokryte są specyficzną tkanką, welamenem, zbudowaną z kilku warstw martwych komórek. Mają one porowate ściany komórkowe, dzięki czemu wchłaniają parę wodną oraz wodę opadową z powietrza (Szlachetko 2001).

Storczykowate to rośliny, które zawsze wzbudzały ciekawość i zainteresowanie wśród badaczy. Pierwsze wzmianki możemy znaleźć już w pracach Teofrasta, z przełomu IV i III wieku p.n.e. (Szlachetko i Skakuj 1996). Początkowo były traktowane przede wszystkim jako surowce lecznicze lub silne afrodyzjaki. Nieco później zwrócono uwagę na ich piękne i wyjątkowe kwiaty przez co zaczęto traktować je również jako rośliny ozdobne. Poznanie szczegółowej budowy tych organów stało się też obiektem wielu badań.

Kwiaty Orchidaceae są niezmiernie różnorodne, zarówno pod względem kształtu, koloru czy zapachu. Mogą występować pojedynczo, ale zdecydowanie częściej skupione są w kwiatostany typu grono lub kłos, rzadziej typu wiecha czy koszyczek. Okwiat zbudowany jest z dwóch okółków, zewnętrznego (ang. *sepals*) i wewnętrznego (ang. *petals*), a każdy z nich z 3 listków. Poszczególne listki mogą różnić się od siebie zarówno kształtem, wielkością jak i kolorem. Jednak szczególnie wyróżniającym się jest środkowy listek okółka wewnętrznego okwiatu, czyli warżka (ang. *labellum, lip*), która odgrywa niezmiernie istotną i ważną rolę

w procesie zapylania tych roślin. U niektórych gatunków jest ona swego rodzaju pułapką dla owada zapylającego (*Cypripedium* L., *Paphiopedilum* Pfitzer). U innych na jej powierzchni znajdują się różnego typu struktury, które mogą wytwarzać i gromadzić nektar (*Gymnadenia* R. Br., *Satyrium* Sw.). Natomiast u przedstawicieli rodzaju *Ophrys* L., czy *Drakaea* Lindl. przypomina swoim wyglądem samicę owada zapylającego. Kwiaty są wówczas zapylane na drodze tzw. pseudokopulacji. Najczęściej jednak warżka jest lądowiskiem dla zapylacza, który dzięki niej ma prosty dostęp do struktur generatywnych. Budowa ich jest na tyle specyficzna, iż wyróżnia przedstawicieli Orchidaceae na tle pozostałych grup roślin. U większości storczykowatych nastąpiła redukcja liczby płodnych pręcików. U bardziej prymitywnych jednego, a u zaawansowanych dwóch. W rezultacie ulegają one przekształceniu w prątniczki, czyli *staminodia*. Natomiast płodny pręcik lub pręciki zrastają się z szyjką słupka tworząc charakterystyczną dla tej grupy roślin strukturę, prętosłup (łac. *gynostemium*). Ziarna pyłku połączone są ze sobą i tworzą masę pyłkową (łac. *pollinium*). Łatka środkowa znamienia słupka przekształcona jest w dziubek (łac. *rostellum*), a jego szczytowa część wytwarza uczepek (łac. *viscidium*), którego głównym zadaniem jest przyklejanie masy pyłkowej do odwiedzającego roślinę zapylacza. Dziubek może wytwarzać także inne, dodatkowe struktury, takie jak szypułki pollinium (łac. *tegula* czy *hamulus*). Odgrywają one istotną rolę w procesie zapylania, umożliwiając m.in. deponowanie masy pyłkowej na znamieniu. Owocem występującym u Orchidaceae jest torebka zawierająca nasiona, które charakteryzują się szczególną budową. Nie posiadają one bielma, przez co są mikroskopijne i należą do najmniejszych nasion w świecie roślin (Szlachetko i Skakuj 1996; Szlachetko 2001). Utrata bielma ułatwia transport na duże odległości. Brak bielma jest jednoznaczny z brakiem materiałów zapasowych, jednak dzięki zjawisku mikoryzy, czyli symbiozy z grzybem, nasienie zaopatrywane jest w wodę i sole mineralne.

Rozprawa doktorska poświęcona jest klasyfikacji i filogenezie rodzaju *Dendrobium* ze szczególnym uwzględnieniem sekcji nominalnej. Jest to liczna grupa, obejmująca około 1600 gatunków o szerokim zasięgu występowania (Liu i in. 2019; Zhao i in. 2019). Taksony spotykane są w Azji kontynentalnej, Australii, na wyspach Pacyfiku, jak również w Nowej Gwinei i Nowej Zelandii (Wang i in. 2009; Moudi i in. 2013). Przedstawicieli rodzaju charakteryzuje silny polimorfizm zarówno cech wegetatywnych, jak i kwiatowych. Może on być spowodowany zarówno przystosowaniem do zwierząt zapylających, jak i warunków siedliskowych. Przekłada

się on także na wiele problemów klasyfikacyjnych oraz trudności w konstruowaniu kluczy do identyfikacji gatunków. Mimo tych różnic wszystkie gatunki rodzaju cechuje posiadanie bocznego kwiatostanu powstającego w górnej części pędu, obecność mentum - struktury powstałej w wyniku zrośnięcia stopy kolumny z bocznymi listkami okółka zewnętrznego okwiatu (niekiedy przypominające ostrogę), czterech nagich pyłkowin oraz występowanie nabrzmiałego dziubka (*rostellum*). Zarówno obiekt badań, jak i historię jego klasyfikacji szczegółowo opisano w załączonych artykułach, które są efektem niniejszej pracy.

Brak jednego i szeroko akceptowanego systemu klasyfikacyjnego rodzaju *Dendrobium*, skłonił mnie do przeprowadzenia badań, w których głównym celem była rekonstrukcja filogenezy i systematyka sekcji nominalnej. Sekcja nominalna obejmuje gatunek-typ rodzaju wskazany przez autora (lub późniejszych badaczy), określa więc w sposób jednoznaczny oryginalną koncepcję całego rodzaju w rozumieniu jego odkrywcy. Determinuje poniekąd późniejsze ujęcia całego rodzaju. Stąd jej zakres gatunkowy jest niezwykle istotny.

Prace badawcze przeprowadzono wykorzystując skaningową mikroskopię elektronową, techniki biologii molekularnej (PCR, sekwencjonowanie DNA), analizy filogenetyczne oraz fenetyczne (rekonstrukcja cech morfologicznych przodków, analizy UPGMA). W kolejnej części rozprawy zaprezentowano wyniki realizowanych badań.

2. Cele pracy

Podstawowym celem niniejszej rozprawy doktorskiej była rekonstrukcja filogenezy i klasyfikacja rodzaju *Dendrobium* ze szczególnym uwzględnieniem sekcji nominalnej w oparciu o szerokie spektrum cech morfologicznych, ultramorfologicznych i genetycznych. Wykonanie tego zadania przebiegało w kilku etapach, poprzez realizację następujących celów pośrednich:

- przeanalizowanie dostępnych okazów utrwalonych w płynie konserwującym (Kew Mixture), reprezentujących gatunki sekcji nominalnej i innych badanych sekcji, pod kątem wykorzystania obecnych mikrostruktur do klasyfikacji wewnątrzrodzajowej,

- określenie zmienności rozmieszczenia struktur mikromorfologicznych w poszczególnych częściach warżki,

- określenie użyteczności niskokopijnego genu jądrowego *Xdh* w badaniach filogenetycznych na poziomie sekcji typowej,

- sporządzenie drzew filogenetycznych w oparciu o zmienność w obrębie sekwencji jądrowych, ITS1-5.8S-ITS2 i *Xdh* oraz fragmentów plastydowych, genu *mat*K i regionu *trn*L-*trn*F, a także porównanie przydatności wymienionych markerów w badaniach taksonomicznych,

- przeanalizowanie wybranych cech morfologicznych oraz porównanie i zestawienie ich z wynikami analiz filogenetycznych dla gatunków reprezentujących sekcje całego rodzaju,

- zgromadzenie i przedstawienie danych dotyczących rozmieszczenia geograficznego gatunków,

- ustalenie pozycji systematycznej wybranych gatunków w oparciu o kompleksowe informacje morfologiczne i molekularne.

3. Artykuły wchodzące w skład rozprawy doktorskiej

I. Burzacka-Hinz, A., Narajczyk, M., Dudek, M., & Szlachetko, D. L. (2022). **Micromorphology of Labellum in Selected *Dendrobium* Sw. (Orchidaceae, Dendrobieae).** *International Journal of Molecular Sciences*, *23*(17), 9578.

W pierwszym artykule „Micromorphology of Labellum in Selected *Dendrobium* Sw. (Orchidaceae, Dendrobieae)" przeanalizowano struktury mikromorfologiczne obecne na warżkach dla wybranych gatunków. Tego typu badania opierają się na wykorzystaniu skaningowej mikroskopii elektronowej (SEM). Daje ona możliwość dokładnego poznania struktur komórkowych występujących na powierzchni poszczególnych komórek. Wykorzystana metoda polega na skanowaniu badanej próbki za pomocą wiązki elektronów uformowanych przez układ elektrono-optyczny mikroskopu. Sygnał z powierzchni analizowanej struktury, najczęściej w postaci elektronów wtórnych lub odbitych, dociera do detektora. Jego główne części to scyntylator i fotopowielacz. Zadaniem scyntylatora jest przekształcenie energii elektronów w impulsy świetlne, które następnie wzmacniane są przez fotopowielacz. Wychodzący z detektora sygnał steruje jasnością obrazu widocznego na ekranie monitora. Obrazy SEM przedstawiają trójwymiarowy widok badanej powierzchni, co umożliwia wykonanie dokładnych pomiarów analizowanych obiektów, ocenę morfologii, grubości czy porowatości (Słowik 2012; Barbacki 2003). Szczegółowa procedura przygotowania materiału badawczego oraz dokładna metodyka zostały opisane we wspomnianym artykule. Uzyskane przeze mnie wyniki badań w połączeniu z badaniami terenowymi, w przyszłości z pewnością pozwolą na zrozumienie roli poszczególnych mikrostruktur w zwabianiu zapylaczy. Ponadto rezultaty przeprowadzonych analiz mikromorfologicznych mają również znaczenie dla klasyfikacji wewnątrzrodzajowej u *Dendrobium*. Często obecność lub brak mikrostruktur, a także ich charakter, mogą okazać się ważnym źródłem informacji taksonomicznej.

Wnioski: Badaniami mikromorfologicznymi objętych zostało 21 gatunków *Dendrobium* reprezentujących 13 sekcji, w tym 7 gatunków sekcji nominalnej. Na warżkach u wszystkich taksonów z sekcji *Dendrobium* zaobserwowano dwa typy struktur. Były to cylindryczne i stożkowate włoski oraz stożkowate i półkoliste brodawki. Większość gatunków z innych sekcji również miała stożkowate i cylindryczne włoski i brodawki, ale obecne były także włoski

elipsoidalne i brodawki półkoliste. Natomiast u pięciu gatunków nie zaobserwowano obecności żadnych analizowanych struktur. To badanie jest pierwszą analizą w rodzaju *Dendrobium* skupiającą się na mikromorfologii. Na podstawie uzyskanych wyników mikromorfologicznych i przeprowadzonych analiz filogenetycznych zasugerowano, że obecność i brak struktur na warżkach wynika prawdopodobnie z konwergencji, a nie pokrewieństwa. W związku z tym może być to ściśle związane z presją zapylaczy. Oczywistym jest, że potwierdzenie tej tezy i dokładne powiązanie funkcji z obecnością i rozmieszczeniem poszczególnych struktur wymaga dalszych prac. Przede wszystkim rozszerzenia grupy badawczej, dodatkowych badań terenowych i obserwacji procesu zapylania. Niemniej jednak niniejszy artykuł dostarcza nowych, zróżnicowanych danych i solidne podstawy do dalszych badań.

II. Burzacka-Hinz, A., Dudek, M., & Szlachetko, D. L. (2024). **Potential use of low-copy nuclear gene *Xdh* at lower taxonomic levels based on phylogenetic analysis of the nominal section of *Dendrobium*.** *Acta Societatis Botanicorum Poloniae*, *93*, 7922.

Współczesna taksonomia to nie tylko dziedzina rozpatrująca podobieństwo morfologiczno-anatomiczne pomiędzy organizmami. To przede wszystkim nauka, która czerpie i wykorzystuje wiele różnych metod, takich jak wspomniany i zastosowany przeze mnie SEM, czy techniki powszechnie stosowane w biologii molekularnej. W związku z tym jej założeniem nie jest tylko sklasyfikowanie organizmów, ale również stworzenie tzw. „drzewa życia", czyli odzwierciedlenie relacji filogenetycznych pomiędzy poszczególnymi taksonami. Jest to nie lada wyzwanie dla badaczy. Należy uwzględnić różne wyniki analiz, a przy tym wybrać do badań molekularnych odpowiednie fragmenty genomu.

Do tej pory próby zrekonstruowania filogenezy dla rodzaju *Dendrobium* opierały się głównie na kilku markerach plastydowych, takich jak *rbc*L, *mat*K czy region *trn*L-*trn*F oraz jądrowych sekwencjach ITS1 i ITS2 (ang. *Internal Transcribed Spacer*) znajdujących się w genomie pomiędzy genami kodującymi dużą i małą podjednostkę rybosomalną. Dlatego, w niniejszej pracy sprawdzono także możliwości zastosowania niskokopijnego genu jądrowego *Xdh*. Był on z powodzeniem wykorzystany u innych grup roślin, także u pozostałych przedstawicieli Orchidaceae, zwłaszcza tam, gdzie uniwersalne znaczniki molekularne nie były

w stanie wygenerować silnego sygnału filogenetycznego (Sang, 2002; Górniak i in. 2010). Natomiast w przypadku *Dendrobium* został on użyty tylko raz (Moudi i Go, 2015). Markery niskokopijne są dość często wykorzystywane w analizach filogenetycznych głównie ze względu na dużą ilość cech informatywnych oraz dwurodzicielskie dziedziczenie (Small i in. 2004). Użycie ich dawało zatem nadzieje na zaproponowanie optymalnego markera do przeprowadzenia analiz filogenetycznych, także na niższych poziomach taksonomicznych, takich jak rodzaj czy sekcja.

W artykule „Potential use of low-copy nuclear gene *Xdh* at lower taxonomic levels based on phylogenetic analysis of the nominal section of *Dendrobium*" sprawdzono użyteczność i zastosowanie do rekonstrukcji filogenezy wspomnianego niskokopijnego genu oraz porównano go z dotychczas wykorzystywanymi markerami. Sekwencje DNA uzyskane dla wszystkich analizowanych fragmentów najpierw zostały przyrównane (ang. *alignment*), a następnie w oparciu o zmienność w ich obrębie zostały wykonane analizy filogenetyczne. Do rekonstrukcji filogenezy użyto trzech metod, tj. największej oszczędności (ang. *maximum parsymony*), największej wiarygodności (ang. *maximum likelihood*) i wnioskowanie Bayesowskie (ang. *Bayesian inference*), w celu wykluczenia niezgodności topologii. Stworzone macierze danych zostały wykorzystane również do obliczenia cech stałych oraz zmiennych informatywnie. Ponadto wykonano analizy statystyczne porównujące ze sobą badane markery. W oparciu o uzyskane wyniki wskazano, że do badanej grupy storczykowatych najlepiej sprawdza się jądrowy marker ITS.

Wnioski: Należy podkreślić, że do rozwiązywania relacji filogenetycznych na różnych poziomach taksonomicznych oba markery jądrowe okazały się bardziej wartościowe niż fragmenty plastydowe. Rezultaty przeprowadzonych analiz, zarówno w przypadku macierzy dla genu *mat*K, jaki i regionu *trn*L-*trn*F nie wykazały zadowalającego efektu. Oznacza to, że zmienność w ich obrębie jest niewystarczająca do różnicowania gatunków rodzaju *Dendrobium*. Ponadto informacje uzyskane w oparciu o analizy danych jądrowych dowodzą, że do próby zrekonstruowania filogenezy dla rodzaju *Dendrobium* lepiej jest zastosować marker wielokopijny ITS1-5.8S-ITS2, niż gen o niskiej liczbie kopii *Xdh*.

III. Burzacka-Hinz, A., Dudek, M., Olędrzyńska, N., Naczk, A. M., & Szlachetko, D. L. **,,Evolution of morphological traits of *Dendrobium sensu lato* - an attempt to resolve phylogenetic relationships in nominal and morphologically convergent sections.''**

W złożonym do czasopisma BMC Plant Biology manuskrypcie ,,Evolution of morphological traits of *Dendrobium sensu lato* (Orchidaceae) - an attempt to resolve phylogenetic relationships in nominal and morphologically convergent sections'' skoncentrowano się głównie na dwóch aspektach – morfologii oraz filogenezie, a także przeprowadzono analizę cech przodka oraz zgromadzono dane geograficzne dla poszczególnych gatunków *Dendrobium*. Głównym założeniem prowadzonych badań było zestawienie i przedyskutowanie danych otrzymanych z różnorodnych analiz, tj. fenetycznej, filogenetycznej, cech przodka oraz geograficznej, na temat przedstawicieli rodzaju w kontekście klasyfikacji wewnątrzrodzajowej. Takie podejście umożliwia bardziej kompleksowe obserwacje oraz pozwala lepiej zrozumieć, że niektóre cechy morfologiczne mogły powstać kilkukrotnie w toku ewolucji, niezależnie od siebie. Ma to duże znaczenie, zwłaszcza w przypadku analiz dotyczących systematyki. Wyniki są wtedy bardziej złożone, wiarygodne i dokładne. W oparciu o rezultaty uzyskane w ramach badań prowadzonych do drugiego artykułu („Potential use of low-copy nuclear gene *Xdh* at lower taxonomic levels based on phylogenetic analysis of the nominal section of *Dendrobium*") rekonstrukcja filogenezy przeprowadzona została z wykorzystaniem markera ITS. Natomiast analizy fenetyczne wykonano dla zestawu 14 cech, zarówno wegetatywnych jak i kwiatowych, istotnych w taksonomii rodzaju. Jak wspomniano powyżej, dzięki powiązaniu i porównaniu uzyskanych informacji, w manuskrypcie przedyskutowano przynależność taksonomiczną wybranych gatunków, zwłaszcza z sekcji nominalnej. Niniejszy artykuł jest obecnie w recenzjach. Podobnie jak poprzednie prace dołączony jest również do kolejnej części rozprawy.

Wnioski: Wyniki naszych analiz pokazują, że w *Dendrobium sensu lato* mamy prawdopodobnie do czynienia z ewolucją konwergentną. Wiele cech kwiatowych ewoluowało wiele razy niezależnie. Prowadzi to do licznych problemów klasyfikacyjnych w obrębie rodzaju. Nie uzyskaliśmy jasnej odpowiedzi na pytanie, jak sklasyfikować poszczególne gatunki w obrębie całego taksonu. Z tego powodu w niniejszej pracy skupiliśmy się głównie na rozwiązaniu problemów w sekcji nominalnej. Określenie przynależności gatunków do sekcji typowej

zostało potraktowane priorytetowo. Warto zachować w niej tylko te gatunki, które na podstawie zestawu cech morfologicznych nie pozostawiają żadnych wątpliwości. Ze względu na tak dużą grupę, najlepiej pracować małymi partiami, tj. analizować poszczególne sekcje, a następnie zebrać wszystkie dane razem. W przeciwnym razie badanie całego rodzaju może przysporzyć wielu problemów i niejasności.

Konwergentna ewolucja może być wynikiem adaptacji do zapylaczy lub częściej zdaje się być wynikiem hybrydyzacji. Zjawisko to jest dość powszechne u roślin i niestety bardzo często utrudnia ustanowienie spójnego systemu klasyfikacji, zwłaszcza w przypadku taksonów licznych w gatunki i zróżnicowanych morfologicznie, a takim właśnie jest *Dendrobium*. W tym artykule żadna z przeprowadzonych analiz, jednoznacznie ani nie potwierdziła, ani nie obaliła tego założenia. Warto jednak zwrócić szczególną uwagę na tę kwestię w przyszłości.

4. Podsumowanie

Status taksonomiczny i pokrewieństwo pomiędzy niektórymi gatunkami, czy sekcjami są nadal niejasne, chociaż każdego roku publikowanych jest wiele prac poświęconych różnym aspektom *Dendrobium*. Ważne jest, aby stopniowo przybliżać się do rozwiązania problemów systematycznych całego rodzaju, dlatego jestem usatysfakcjonowana, że dzięki podjęciu próby poznania sekcji nominalnej mogłam przedstawić nowe informacje i podzielić się wnioskami z przeprowadzonych przeze mnie analiz. Dołożyłam także wszelkich starań, aby we wprowadzeniu unikać powielania wiadomości, które można znaleźć w dołączonych publikacjach. Z tego powodu dane dotyczące wykorzystanych materiałów oraz szczegółowe informacje odnoszące się do zastosowanych metod badawczych zostały omówione wyłącznie w artykułach.

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CZĘŚĆ II

1. Artykuł badawczy I

Burzacka-Hinz, A., Narajczyk, M., Dudek, M., & Szlachetko, D. L. (2022). Micromorphology of Labellum in Selected *Dendrobium* Sw.(Orchidaceae, Dendrobieae). *International Journal of Molecular Sciences*, *23*(17), 9578.

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Molecular Sciences

MDPI

Article

Micromorphology of Labellum in Selected *Dendrobium* Sw. (Orchidaceae, Dendrobieae)

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Abstract: *Dendrobium* is one of the most species-rich genera of the Paleotropical orchids. It embraces more than 1000 species, most of which are epiphytes. The strong variation in floral characters causes many identification difficulties within this genus. One of the key structures, often sufficient in identification on a species level, is the labellum, which in many species of *Dendrobium* possesses a thickened callus and various types of trichomes and papillae. The aim of this study is to identify and describe the structures present on the labellum surface of the analyzed species, determine their distribution and density, as well as to check whether the obtained data have taxonomic value. In this paper, we present the results of a micromorphological study on the labellum of 21 species of *Dendrobium*, representing 13 sections, using scanning electron microscopy (SEM). Our studies revealed the presence of both uni- and multicellular structures on the surface of the labellum. We observed three types of trichomes (conical, cylindrical, ellipsoidal) and three types of papillae (conical, cylindrical, semicircular). Neither trichomes nor papillae were recorded for five species. In addition, we made diagrams showing the distribution and density of structures on the labellum. Based on the micromorphological results combined with the phylogenetic tree performed, we suggest that the presence/absence of labellum structures does not necessarily reflect the phylogenetic relationship and might be misleading, as in some cases, they arise due to convergence.

Keywords: *Dendrobium*; labellum micromorphology; orchids; scanning electron microscopy; taxonomy

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1. Introduction

Dendrobium Sw. was described by Olof Swartz in 1799 [1]. Currently, it is one of the largest genera belonging to the Orchidaceae, with over 1000 species [2]. They can grow as terrestrial plants or lithophytes, although most of them are epiphytes. Their range extends from Sri Lanka and India throughout tropical Asia and Oceania, north to Japan, east to Tahiti, and south to New Zealand [3].

The representatives of this genus are characterized by the presence of a lateral inflorescence arising from the upper part of the pseudobulbs. The flowers can reach from 0.4 cm to 17 cm in diameter and vary in form and color [4]. Distinctive features are also the presence of a mentum formed by lateral sepals and the column foot, and the presence of four naked, laterally compressed pollinia [4,5]. The mentum is usually slim and forms a spur-like structure, sometimes relatively long. In most species, the labellum is three-lobed and consists of a middle lobe and two lateral lobes. Usually, the middle lobe is tufted and flat in front, tapering towards the rear. At the base of the labellum, there are usually protrusions called callus, hairs, and various types of lumps. These structures, arranged in characteristic patterns, can lure and guide visiting pollinators, most often bees. Unfortunately, there is still little information on pollination of the *Dendrobium* species. Many species of *Dendrobium* are morphologically very similar to each other, however, some species have overlapping

morphological differences that may result in misidentifications [6,7]. In this case, the classical methods of identification that rely on distinguishing characteristic features visible to the naked eye are not sufficient.

Probably the lack of exhaustive data may be due to the sparse studies on the micromorphology of the labellum within this genus; many works facing a similar problem have been published, but they concerned other groups of orchids [8–12]. Only the results obtained by Davies showed that labellum-trichomes and pseudopollen are presented on *Dendrobium unicum* Seidenf [13]. The term pseudopollen most often denotes a floury, yellow–white material, usually, but not always, containing nutrients and externally resembling pollen [10,13]. Often the presence of food is intended to reward visiting pollinators. However, a lack of nutrients does not equal a lack of attracting insects, and species can still do this thanks to mimicry.

The trichomes of *Dendrobium unicum* differ from the trichomes of other orchid representatives, such as *Maxillaria* or *Polystachya*. The differences concern the way they are formed or the presence of various nutrients. In *Dendrobium unicum*, the pseudopollen trichomes consist of a stalk cell, the head of component cells that separate at maturity, and their main food substance is starch. In contrast, in members of *Maxillaria*, pseudopollen is formed by the fragmentation of single-row hairs, resulting in the formation of single cells or short chains of cells [13]. We can observe a similar situation in species of the nominal section of *Polystachya*. However, the feeding hairs found in the species from other sections of this genus are usually one or four-celled [10,13].

In this paper, we analyzed and recognized structures of the labellum from the selected representatives of *Dendrobium* using the scanning electron microscope (SEM). The aim of our studies was to identify and define the micromorphological structures which are potentially present. Moreover, we determined the variability of their distribution in the particular parts of the labellum.

Due to the large number of species of *Dendrobium*, the wide range of their occurrence, and also their huge morphological diversity, there are a lot of problems with the identification of species, and thus with intra-generic classification. Therefore, the micromorphological studies of the labellum of the genus representatives can also be a valuable source of taxonomic information. In addition, they will constitute the foundation for further research on the pollination of *Dendrobium* flowers.

2. Results

2.1. SEM

While describing the labellum and determining the distribution of the analyzed structures, the terminology presented in Figure 1 was used. The following parts of the labellum were distinguished: the basal part, the hypochile; the apical part, the epichile; and the section between the two aforementioned, the mesochile.

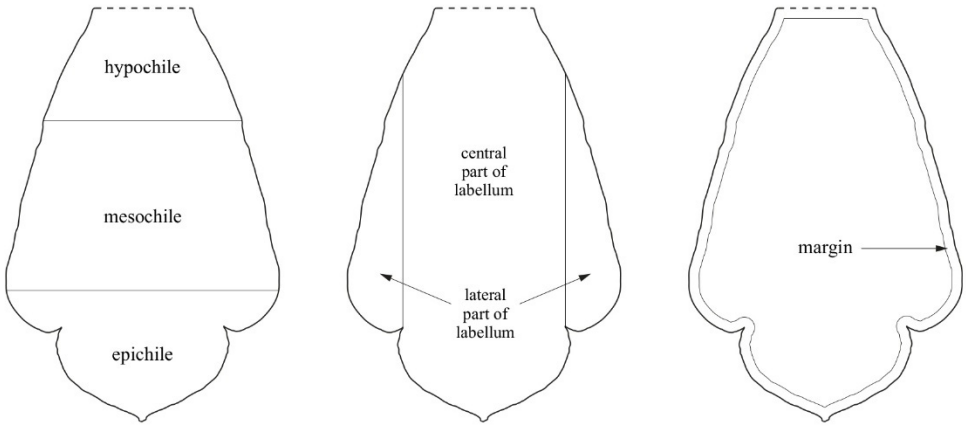

Figure 1. Terminology for parts of the labellum used in the Results section.

Two characteristic forms can be observed on the surface of the examined lips: papillae and trichomes. The first ones are single-celled, with small protrusions of various lengths, usually wider at the base [10,14]. The trichomes are unicellular or multicellular structures of various shapes with a narrow point of insertion and often with a differently shaped apex [10].

Three different types of papillae and three different types of trichomes have been observed. All types that appeared in this review are presented below (Figures 2 and 3, Table 1). The descriptions also considered the shape of the epidermis, bearing, among others, trichomes and papillae, and the presence of callus-protuberances found in some species [14].

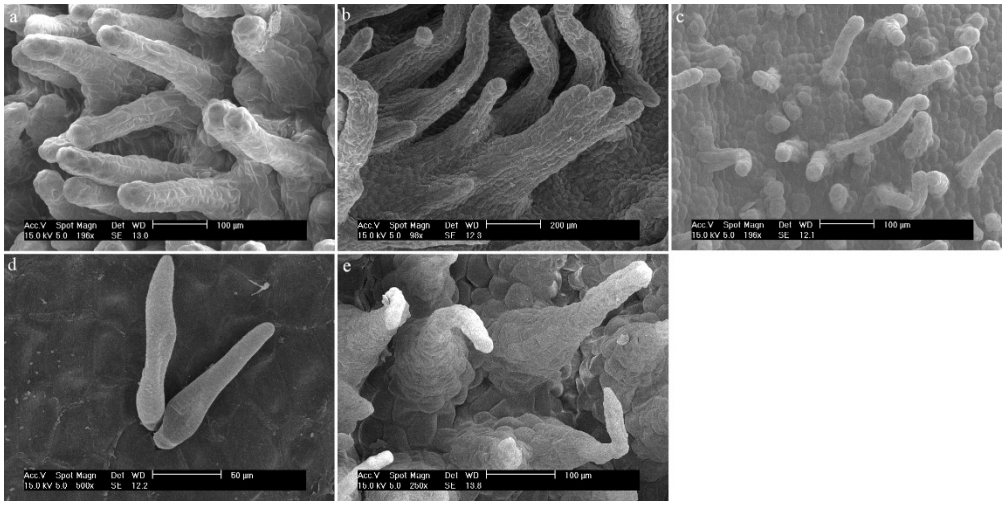

Figure 2. The types of trichomes found on the labellum of the *Dendrobium* species: (**a**) cylindrical; (**b**) cylindrical and branched; (**c**) cylindrical with a bulbous cell at the apex; (**d**) ellipsoidal; (**e**) conical.

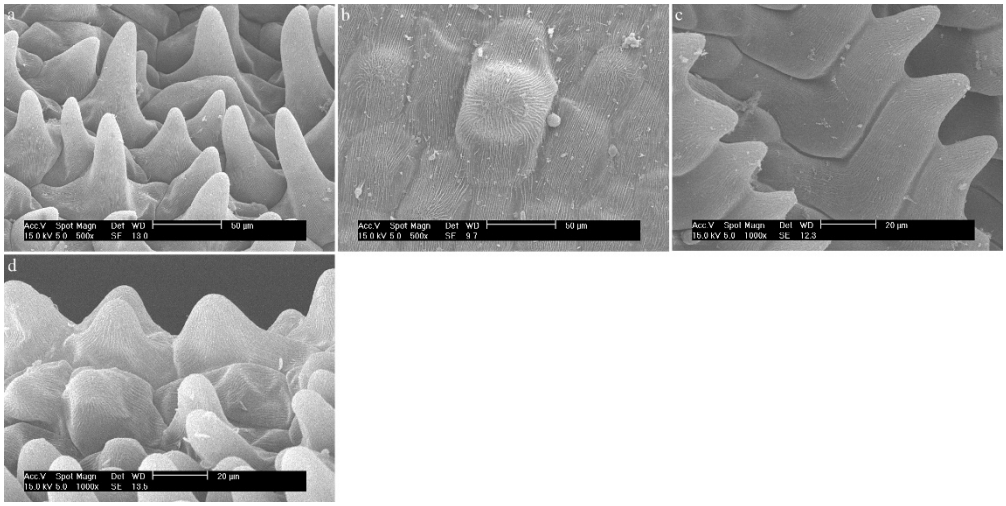

Figure 3. The types of papillae found on the labella of the *Dendrobium* species: (**a**) cylindrical and conical; (**b**) semicircular; (**c**) conical; (**d**) conical with rounded or pointed apices.

Due to the different organization of the papillae and trichomes, labella were drawn (with appropriate dimensions), which schematically show their distribution and density. The juxtaposed labella are shown in Figure 4, and the compaction intensity scale used in the drawings is presented in Table 2.

Table 1. Types of labellar structures found in all *Dendrobium* species researched in this paper, which have trichomes or papillae.

Section	Species	Trichomes	Papillae
Aporum Blume	*Dendrobium hainanense* Rolfe	conical, cylindrical (sometimes branched)	
Dendrobium Sw.	*Dendrobium anosmum* Lindl.	conical, cylindrical (sometimes branched)	
	Dendrobium brymerianum Rchb.f.		conical, semicircular
	Dendrobium chrysocrepis C.S.P.Parish & Rchb.f. ex Hook.f.	conical	
	Dendrobium nobile Lindl.	conical, cylindrical	
	Dendrobium parishii H.Low	cylindrical	
	Dendrobium regium Prain	cylindrical, cylindrical with a bulbous cell at the apex	
	Dendrobium scoriarum W.W.Sm.	conical, cylindrical	
Dendrocoryne Lindl. & Paxton	*Dendrobium* × *delicatum* (F.M.Bailey) F.M.Bailey		conical, cylindrical, semicircular
Formosae (Benth. And Hk.f.) Hk.f.	*Dendrobium christyanum* Rchb.f.	ellipsoidal	
Grastidium (Blume) Blume	*Dendrobium katherinae* A.D.Hawkes		conical with rounded or pointed apices
Monophyllaea Benth.	*Dendrobium monophylum* F.Muell.		conical
Oxyglossum Schltr.	*Dendrobium cuthbertsonii* F.Muell.		conical, semicircular
Rhizobium Lindl. & Paxton	*Dendrobium mortii* F.Muell.	conical, cylindrical	
Spatulata Lindl.	*Dendrobium schoeninum* Lindl. *Dendrobium discolor* Lindl.	conical	conical conical

Table 2. Relative density of trichomes and papillae on labella of *Dendrobium*.

Degree	Trichomes	Papillae	Characterization	Description as Used
4			The distal parts are not visible due to the density of trichomes/papillae	Very dense
3			5–15 trichomes/papillae, up to 3 pieces/100 μm^2	Dense
2			3–4 trichomes/papillae, up to 3 pieces/100 μm^2	Rare
1			Individual trichomes/papillae, up to 3 pieces/100 μm^2	Single/sparse
0			No trichomes/papillae	No trichomes/papillae

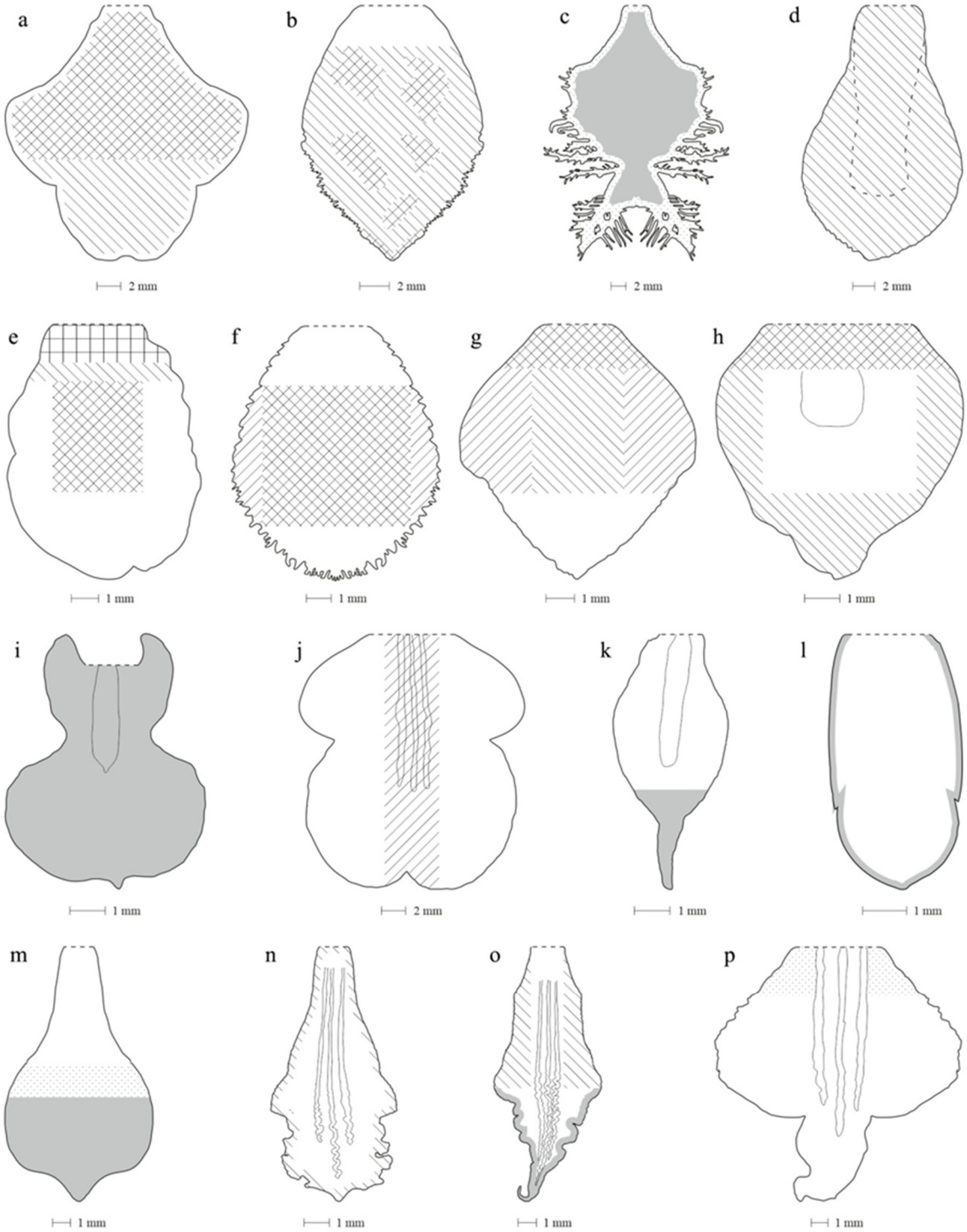

Figure 4. Relative density of trichomes and papillae on the labellum of *Dendrobium*: (**a**) *D. hainanense*; (**b**) *D. anosmum*; (**c**) *D. brymerianum*; (**d**) *D. chrysocrepis*; (**e**) *D. nobile*; (**f**) *D. parishii*; (**g**) *D. regium*; (**h**) *D. scoriarum*; (**i**) *D.* × *delicatum*; (**j**) *D. christyanum*; (**k**) *D. katherinae*; (**l**) *D. monophylum*; (**m**) *D. cuthbertsonii*; (**n**) *D. mortii*; (**o**) *D. schoeninum*; (**p**) *D. discolor*.

The descriptions of the investigated species according to sections are presented below. The first part includes species with labellar protrusions, and the second part includes species without trichomes and papillae.

2.2. Species with Labellar Protrusions

2.2.1. *Dendrobium* Sw. sect. *Aporum* Blume

1. *Dendrobium hainanense* Rolfe On the entire surface of the labellum, there are trichomes covered with a folded cuticle (Figure 5a). In the hypochile and the mesochile, the raised epidermal cells (column-shaped) are single and small. There are conical-shaped trichomes that are rare (Figure 5a). The epichile has much fewer individual, multicellular trichomes (epidermal cells) that vary in size (Figure 5b). Trichomes are branched and multicellular, most often cylindrical in shape and numerous (Figure 5b). The edges of the labellum are smooth, without trichomes.

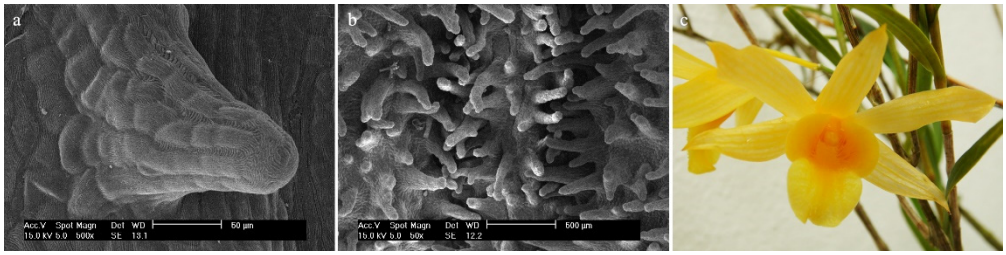

Figure 5. *Dendrobium hainanense* Rolfe.: trichomes (**a**) of the hypochile part of the labellum; (**b**) of the epichile part of the labellum; (**c**) flower (photo by Monika Lipińska).

2.2.2. *Dendrobium* Sw. sect. *Dendrobium*

1. *Dendrobium anosmum* Lindl. The hypochile consists of epidermal cells covered with a thick, folded cuticle. The folding is regular and extends over the entire surface of the labellum (Figure 6a). In the mesochile and the epichile, epidermal cells are regular and smooth, forming a jagged structure at the edges (Figure 6b) and multicellular conical and cylindrical trichomes, sometimes bifurcated at the ends (Figure 6c). In some parts of the surface, their occurrence can be described as rare, but there are also places where their density is higher. The cuticle is smooth.

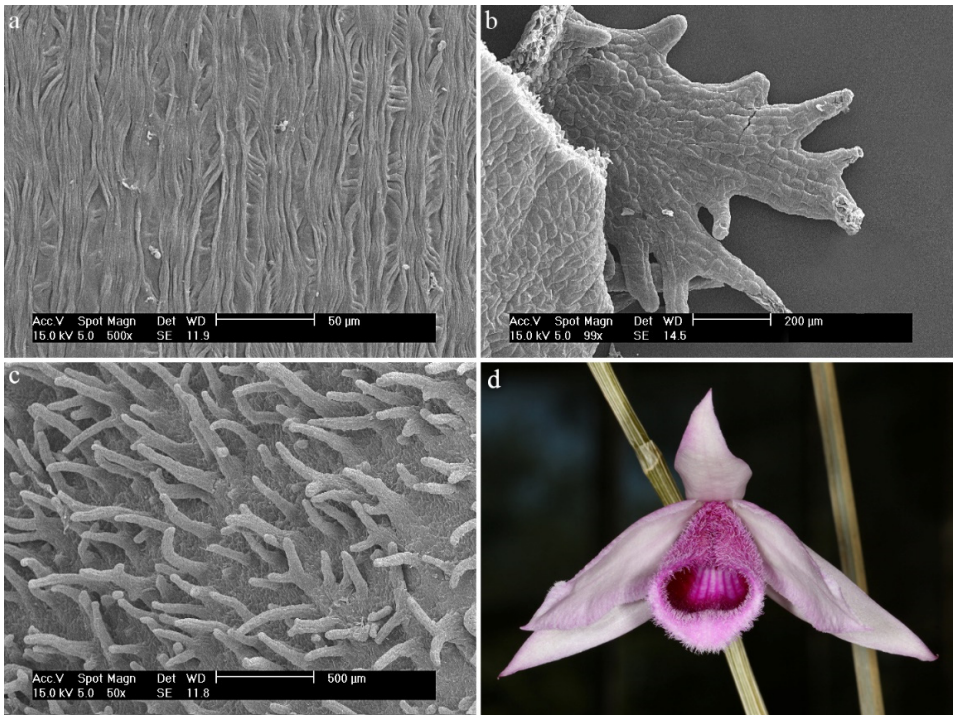

Figure 6. *Dendrobium anosmum* Lindl.: (**a**) the hypochile part of the labellum; (**b**) margin of the mesochile part of the labellum; (**c**) trichomes of the epichile part of the labellum; (**d**) flower (photo by John Varigos).

2. *Dendrobium brymerianum* Rchb.f. The entire surface of the labellum is filled with numerous conical and semicircular papillae. The hypochile has lower papillae than the mesochile and the epichile (Figure 7a). A cuticle is unseen. The edges of the labellum are jagged and branched (Figure 7b). They are made up of epidermal cells covered with a very strongly folded cuticle (Figure 7c). The secretion is possible at the very apex (Figure 7c, marked with an arrow).

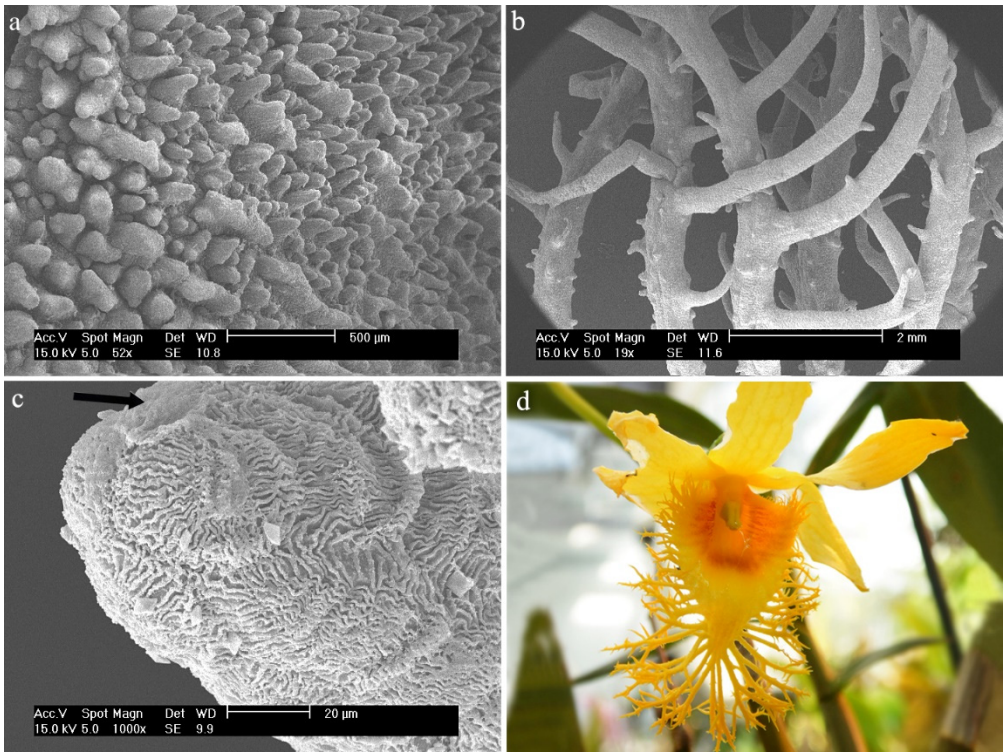

Figure 7. *Dendrobium brymerianum* Rchb.f.: trichomes (**a**) of the mesochile part of the labellum; (**b**) of the margin of the mesochile part of the labellum; (**c**) of the epichile part of the labellum; (**d**) flower (photo by Monika Lipińska).

3. *Dendrobium chrysocrepis* C.S.P.Parish & Rchb.f. ex Hook.f. The entire surface of the labellum is densely covered with conical trichomes (Figure 8a). The final part of the hypochile/initial part of the mesochile is formed by regular epidermal cells with elongated trichomes that are clustered in pairs or appear in a set of three (Figure 8b). In the middle part of mesochile, epidermal cells are slightly elongated, forming multicellular trichomes with a slightly folded cuticle (Figure 8c). The epichile is made up of epidermal cells, which also form elongated and multicellular trichomes (the longest on the entire labellum) (Figure 8d), although they are low at the edges (Figure 8e). A cuticle is unseen.

4. *Dendrobium nobile* Lindl. The hypochile is composed of elongated epidermal cells forming longitudinal, multicellular cylindrical and conical trichomes, which in some places are so densely distributed that their apical part is often not visible (Figure 9a,b). The surface of the mesochile is made up of regular epidermal cells, forming sparsely distributed, multicellular conical trichomes. Trichomes at the base are very wide, tapering towards the apex (Figure 9c). The sides of this part are filled with regular epidermal cells, covered with a folded cuticle (epidermal cells form hills) without any trichomes or papillae (Figure 9d). There are also no trichomes in the terminal epichile part (Figure 9e).

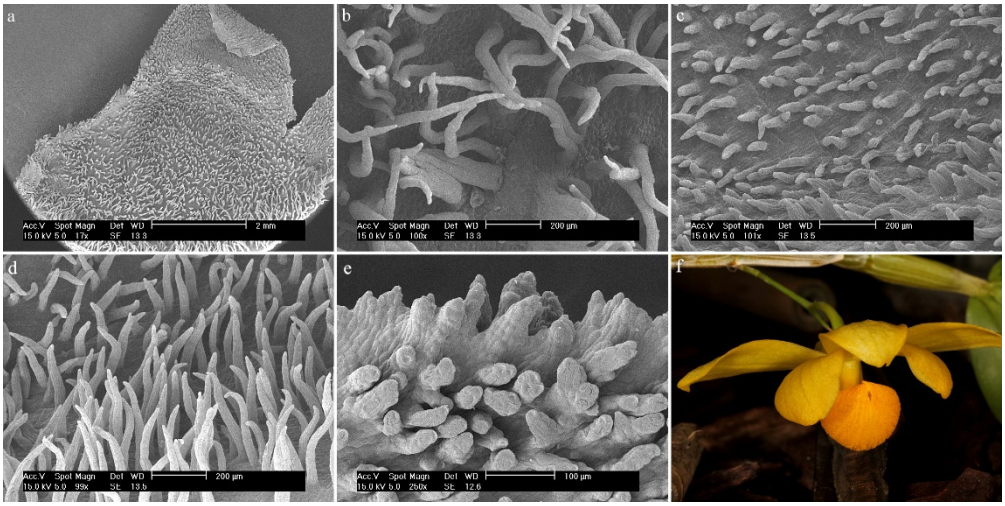

Figure 8. *Dendrobium chrysocrepis* C.S.P.Parish & Rchb.f. ex Hook.f.: trichomes (**a**) of the mesochile and the epichile part of the labellum; (**b**) of the final part of the hypochile of the labellum/initial part of the mesochile of the labellum; (**c**) of the mesochile part of the labellum; (**d**) of the epichile part of the labellum; (**e**) of the margin of the epichile part of the labellum; (**f**) flower (photo by John Varigos).

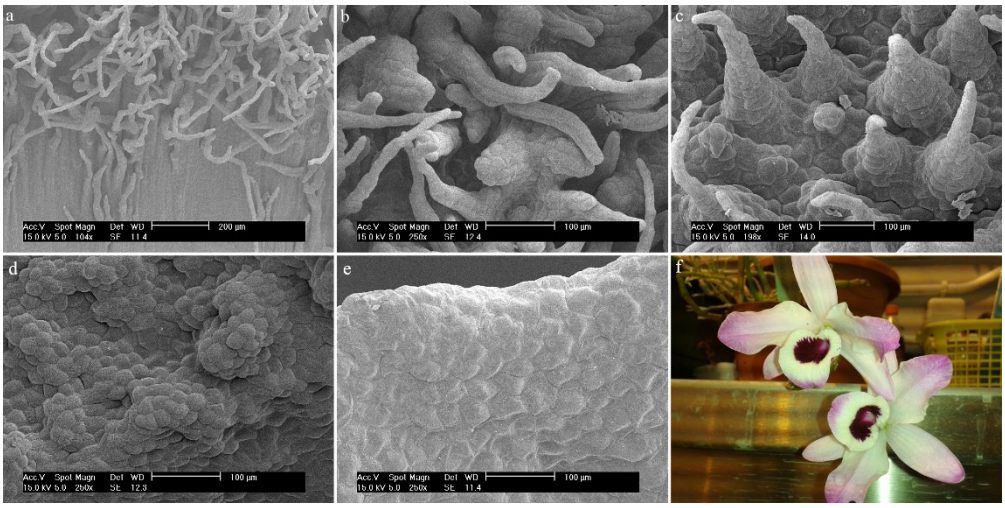

Figure 9. *Dendrobium nobile* Lindl: (**a**,**b**) trichomes of the hypochile part of the labellum; (**c**) trichomes of the mesochile part of the labellum; (**d**) margin of the mesochile part of the labellum; (**e**) margin of the epichile part of the labellum; (**f**) flower (photo by Monika Lipińska).

5. *Dendrobium parishii* H.Low The hypochile has regular epidermal cells without trichomes (Figure 10a). The mesochile also consists of regular epidermal cells, which in the central part form rarely arranged regular, multicellular cylindrical trichomes (Figure 10b). The cuticle is rather smooth. The margin of the mesochile part of the labellum is made up of regular epidermal cells (Figure 10c) that form irregular and jagged edges (Figure 10d). The cuticle here is very slightly folded (Figure 10c). There are single trichomes here, but in the apical part of the epichile, none of them are visible.

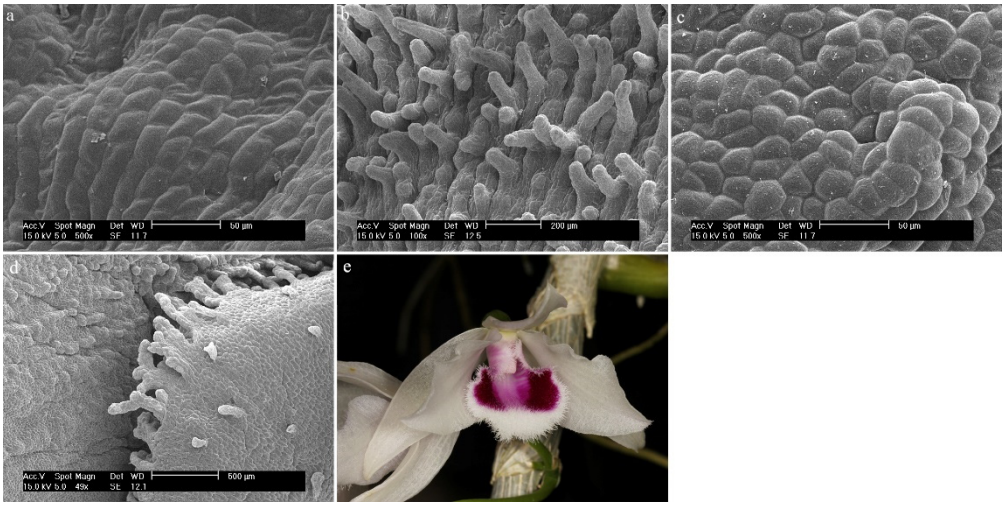

Figure 10. *Dendrobium parishii* H.Low: (**a**) the hypochile part of the labellum; (**b**) trichomes of the mesochile part of the labellum; (**c**) margin of the mesochile part of the labellum; (**d**) trichomes of the margin of the mesochile part of the labellum; (**e**) flower (photo by John Varigos).

6. *Dendrobium regium* Prain The hypochile is made up of regular epidermal cells. Rarely, there are elongated, multicellular cylindrical trichomes with a bulbous cell at the apex (Figure 11a). Further on, regular epidermal cells form lower, multicellular, and more cylindrical trichomes (Figure 11b). The mesochile, like the hypochile, consists of regular epidermal cells and has elongated cylindrical trichomes ending with a bulbous cell, which are abundant here (Figure 11c). In this part, both sides of the labellum are also composed of regular epidermal cells with multicellular trichomes, but lower than the trichomes in the middle part and cylindrical, occurring in a small number (Figure 11d). The epichile is composed of regular and smooth epidermal cells; it has no trichomes (Figure 11e).

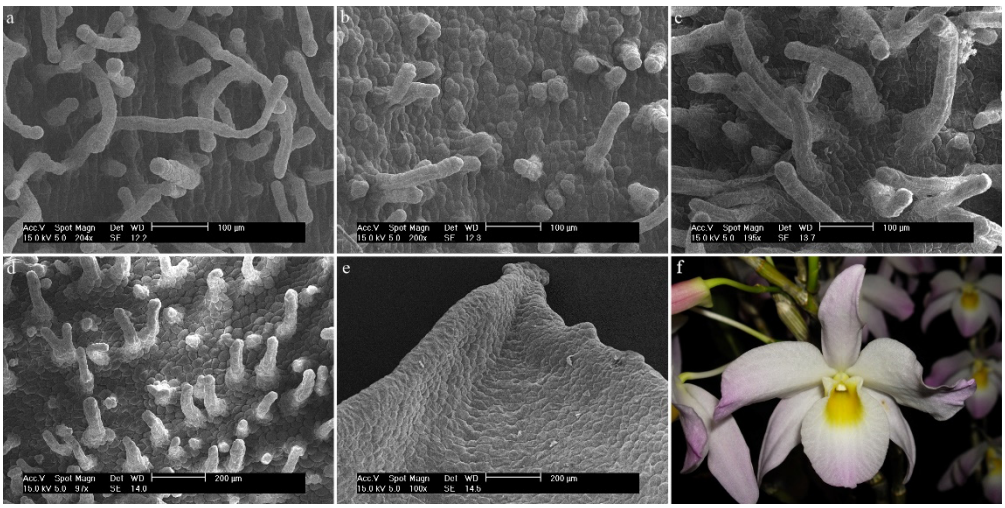

Figure 11. *Dendrobium regium* Prain: (**a**) trichomes of the hypochile part of the labellum; (**b**) trichomes of the final part of the hypochile of labellum/initial part of the mesochile of the labellum; (**c**) trichomes of the mesochile part of the labellum; (**d**) trichomes of the margin of the mesochile part of the labellum; (**e**) the epichile part of the labellum; (**f**) flower (photo by John Varigos).

7. *Dendrobium scoriarum* W.W.Sm. The hypochile is composed of elongated epidermal cells (Figure 12a) that form parallel, multicellular cylindrical trichomes covered with a folded cuticle (Figure 12b). They are rare in this part. Then, the visible callus is made up of regular epidermal cells with a cuticle, which is folded at the junctions between the cells (Figure 12c,d). In the mesochile part, the edges of the labellum are made up of regular epidermal cells, which are only elongated at the site of the formation of multicellular conical and cylindrical trichomes, which occur numerously there (Figure 12e). In the central part of the mesochile, the cuticle covering the epidermis is folded and there are no trichomes on it (Figure 12f). The entire epichile surface is densely covered with multicellular trichomes, mostly conical and sometimes cylindrical (Figure 12g,h). The epidermal cells are regular.

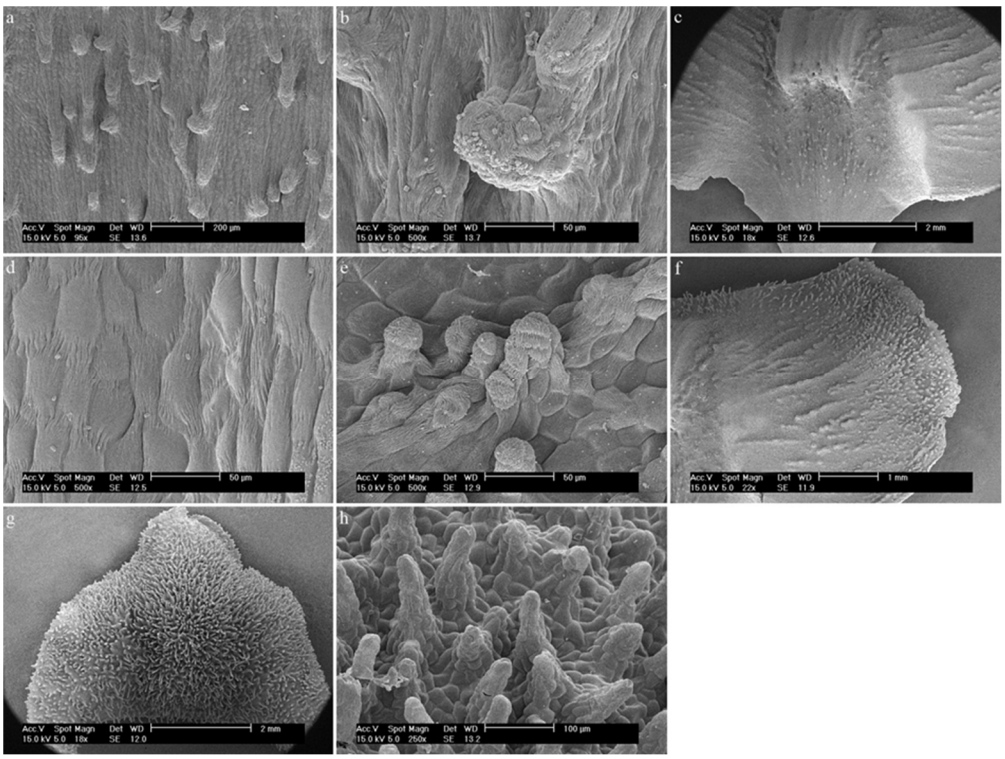

Figure 12. *Dendrobium scoriarum* W.W.Sm.: (**a**) the hypochile part of the labellum; (**b**) trichome of the hypochile part of the labellum; (**c**,**d**) the final part of the hypochile of the labellum/initial part of the mesochile of the labellum; (**e**) trichomes of the margin of the mesochile part of the labellum; (**f**) the mesochile part of the labellum; (**g**,**h**) trichomes of the epichile part of the labellum.

2.2.3. *Dendrobium* Sw. sect. *Dendrocoryne* Lindl. & Paxton

1. *Dendrobium* × *delicatum* (F.M.Bailey) F.M.Bailey Throughout the center of the hypochile and the mesochile parts, a fold-like callus extends, which is raised above the remaining cells. There are densely arranged semicircular papillae resembling a dragon's scale. They are lower than the papillae outside the fold. The papillae outside the fold appear in rows, one after the other (Figure 13a), also resembling the scales of a dragon (Figure 13b). The closer to the epichile, the longer the papillae are, covered with a slightly folded cuticle, and their shape is more conical (Figure 13c). There is possible secretion on the surface. The entire surface of the epichile also features densely arranged papillae with a slightly folded cuticle, conical in shape, and sometimes more cylindrical (Figure 13d).

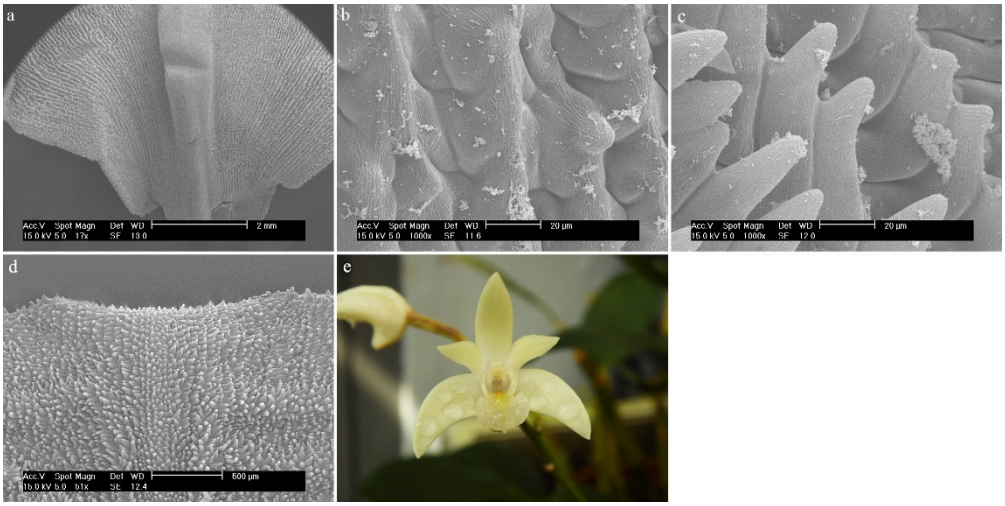

Figure 13. *Dendrobium* × *delicatum* (F.M.Bailey) F.M.Bailey: papillae (**a**) of the hypochile and the mesochile part of the labellum; (**b**) of the hypochile part of the labellum, (**c**) of the mesochile part of the labellum; (**d**) of the epichile part of the labellum; (**e**) flower (photo by Monika Lipińska).

2.2.4. *Dendrobium* Sw. sect. *Formosae* (Benth. & Hook.f.) Hook.f.

1. *Dendrobium christyanum* Rchb.f. Throughout the center of the labellum, there is a callus that consists of regular epidermal cells. The edges of the elevation are made up of epidermis cells resembling frills. In part of the hypochile, it is made up of typical, unextended epidermal cells covered with a rather delicately folded cuticle. The surface shows a few ellipsoidal single-celled trichomes with a smooth cuticle (Figure 14a,b), which are also present on the further parts of the elevation (Figure 14c).

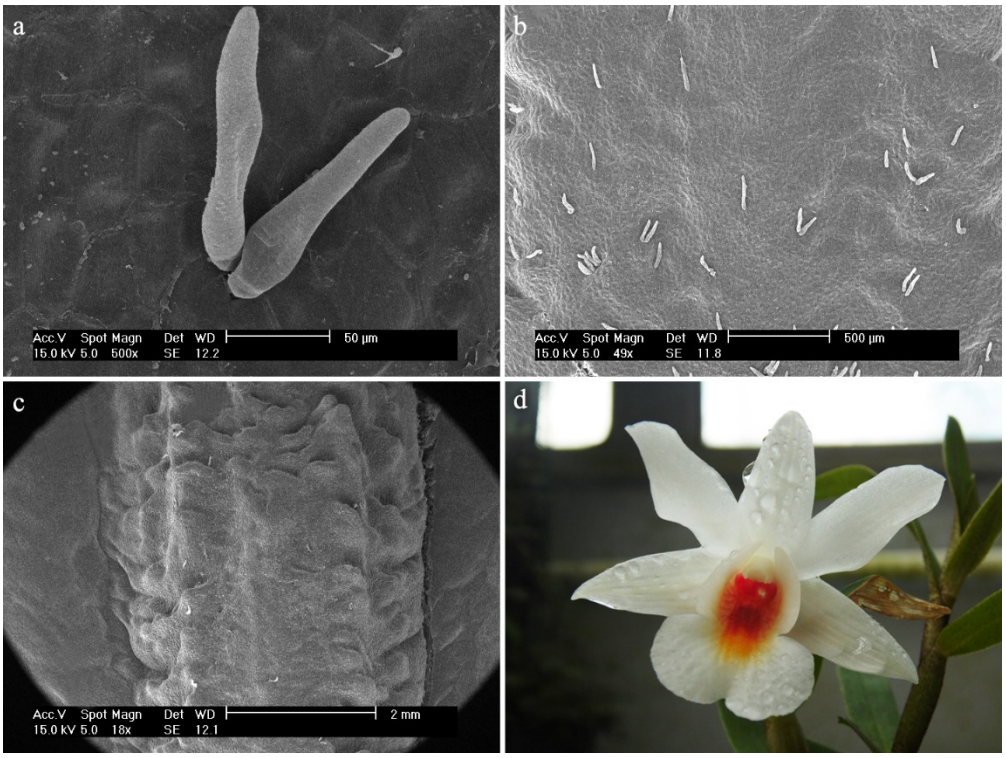

Figure 14. *Dendrobium christyanum* Rchb.f. Trichomes: (**a**,**b**) of the hypochile part of the labellum; (**c**) of the mesochile part of the labellum; (**d**) flower (photo by Monika Lipińska).

2.2.5. *Dendrobium* Sw. sect. *Grastidium* (Blume) Blume

1. *Dendrobium katherinae* A.D.Hawkes Smooth, regular epidermal cells forming a callus are present in the central part of the hypochile and the mesochile (Figure 15a,b). In the initial, lateral part of the mesochile, there are visible multicellular clusters of the epidermis elevated above the surface. Their apex is crowned with round cells (Figure 15c). There are also similar patterns further on, but they are larger (longer) than those described earlier (Figure 15d). The surface of the epichile is covered with tightly formed papillae with a slightly folded cuticle (Figure 15e). They have a conical shape with rounded or pointed apices and are densely arranged.

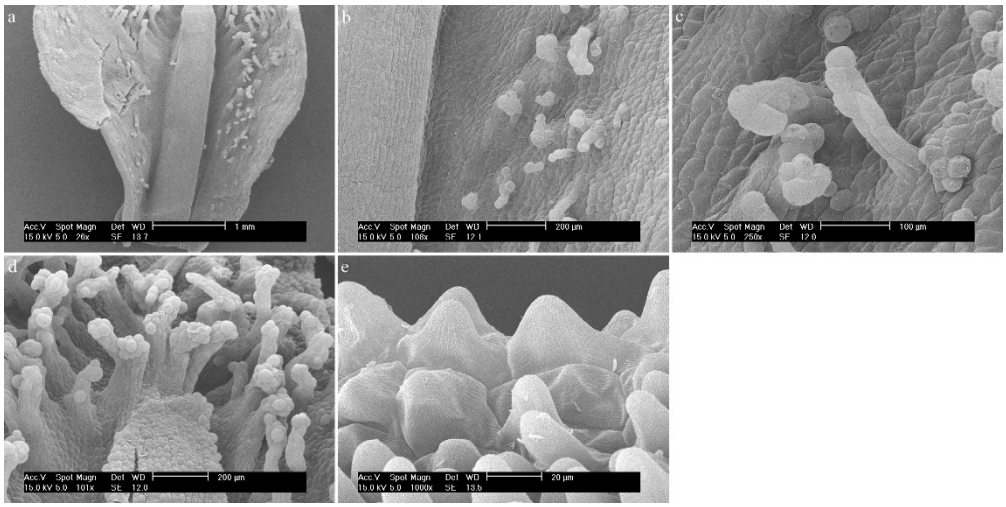

Figure 15. *Dendrobium katherinae* A.D.Hawkes: (**a**) the hypochile and the mesochile part of the labellum; (**b**) the hypochile part of the labellum; (**c**) the initial part of the mesochile of the labellum; (**d**) final part of the mesochile of the labellum; (**e**) papillae of the epichile part of the labellum.

2.2.6. *Dendrobium* Sw. sect. *Monophyllaea* Benth.

1. *Dendrobium monophyllum* F.Muell. The labellum is made up of regular, elongated epidermal cells with visible cuticle folds (Figure 16a,b). On its edges, there are raised epidermal cells, which are probably densely arranged conical papillae with a strongly folded cuticle (Figure 16c,d).

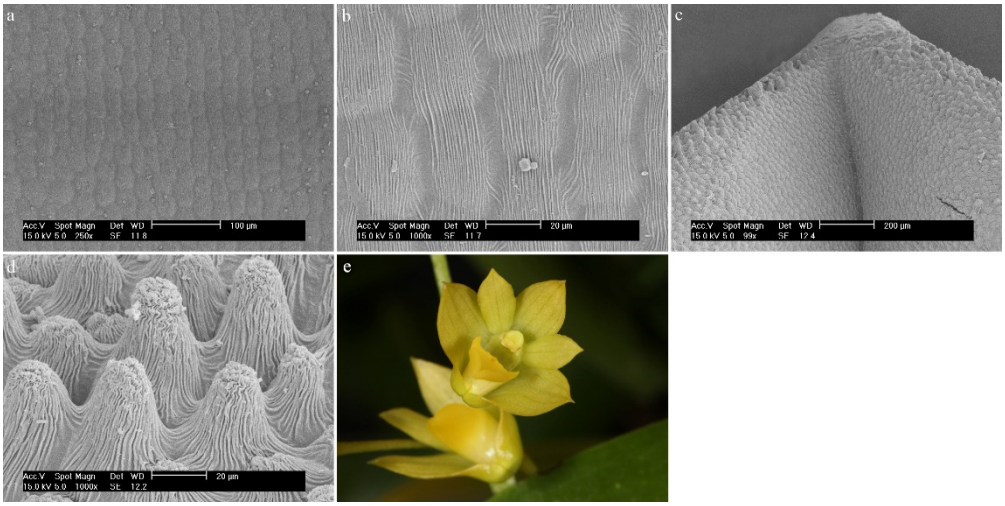

Figure 16. *Dendrobium monophyllum* F.Muell.: (**a**,**b**) the hypochile part of the labellum; (**c**) the mesochile and the epichile part of the labellum; (**d**) papillae of the margin of the epichile part of the labellum; (**e**) flower (photo by John Varigos).

2.2.7. *Dendrobium* Sw. sect. *Oxyglossum* Schltr.

1. *Dendrobium cuthbertsonii* F.Muell. The hypochile and the mesochile form epidermal smooth cells with a very delicately folded cuticle (Figure 17a,b). In the distal part of the mesochile, there are single semicircular papillae with a strongly corrugated cuticle (Figure 17c,d). The entire surface of the epichile is made up of densely arranged semicircular and conical papillae with a strongly folded cuticle (Figure 17e).

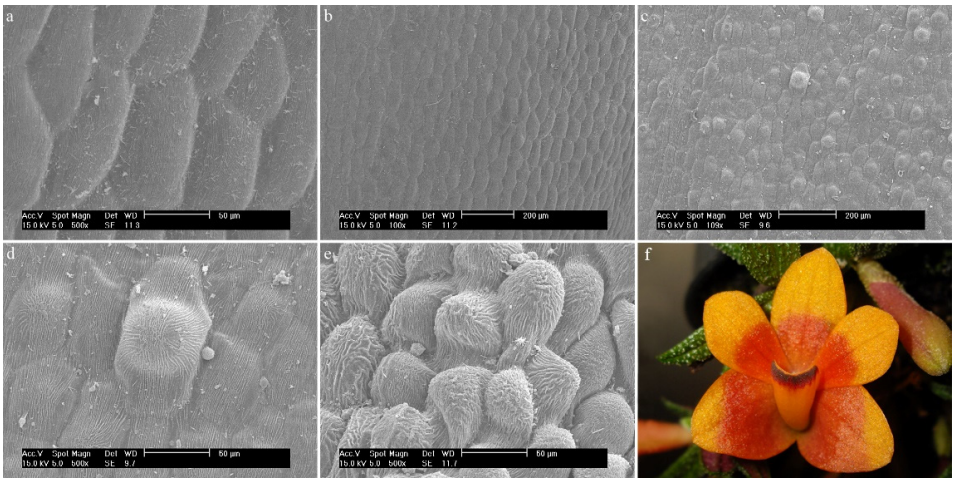

Figure 17. *Dendrobium cuthbertsonii* F.Muell.: (**a**,**b**) the hypochile part of the labellum; Papillae (**c**,**d**) of the mesochile part of the labellum; (**e**) of the epichile part of the labellum; (**f**) flower (photo by John Varigos).

2.2.8. *Dendrobium* Sw. sect. *Rhizobium* Lindl. & Paxton

1. *Dendrobium mortii* F.Muell. On the edge of the whole labellum, there are densely arranged conical and cylindrical trichomes covered with a corrugated cuticle (Figure 18a). In the central part, there is a callus made up of elongated epidermal cells covered with a delicately folded cuticle. It forms three major furrows, which are folded further on, but the cells remain the same (Figure 18b). The central bulge is the longest. The next parts of the labellum are made up of slightly convex epidermal cells with a folded cuticle (Figure 18c). It is possible that there is secretion on their surface (Figure 18d). In the epichile part, on the margin, around the labellum, there are densely arranged trichomes of the same form, as in the part below (Figure 18e).

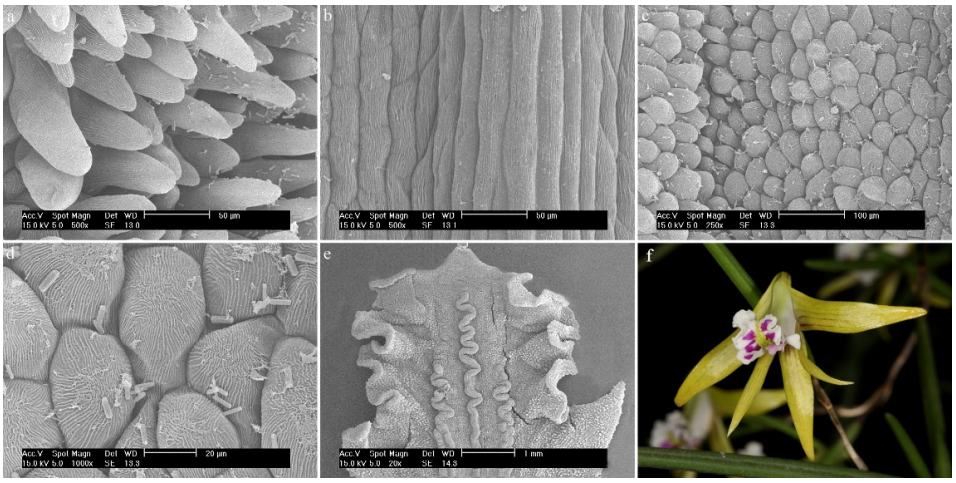

Figure 18. *Dendrobium mortii* F.Muell.: (**a**) trichomes of the hypochile part of the labellum; (**b**) the hypochile part of the labellum; (**c**,**d**) the mesochile part of the labellum; (**e**) the epichile part of the labellum with trichomes in the margin; (**f**) flower (photo by John Varigos).

2. *Dendrobium schoeninum* Lindl. In the hypochile and the mesochile, on the lateral parts of the labellum, there are dense, tightly arranged conical trichomes covered with a delicately folded cuticle (Figure 19a–c). In the center, there is a callus made up of regular and elongated epidermal cells covered with a delicately folded cuticle. It forms three furrows that are present along the entire length of the labellum, but in its distal parts, they take a wavy form (Figure 19b,d). Between the folded furrows there are convex, oval epidermal cells with a slightly folded cuticle (Figure 19d,e). In the epichile part, the lateral edges are composed of densely arranged conical papillae with a folded cuticle (Figure 19f).

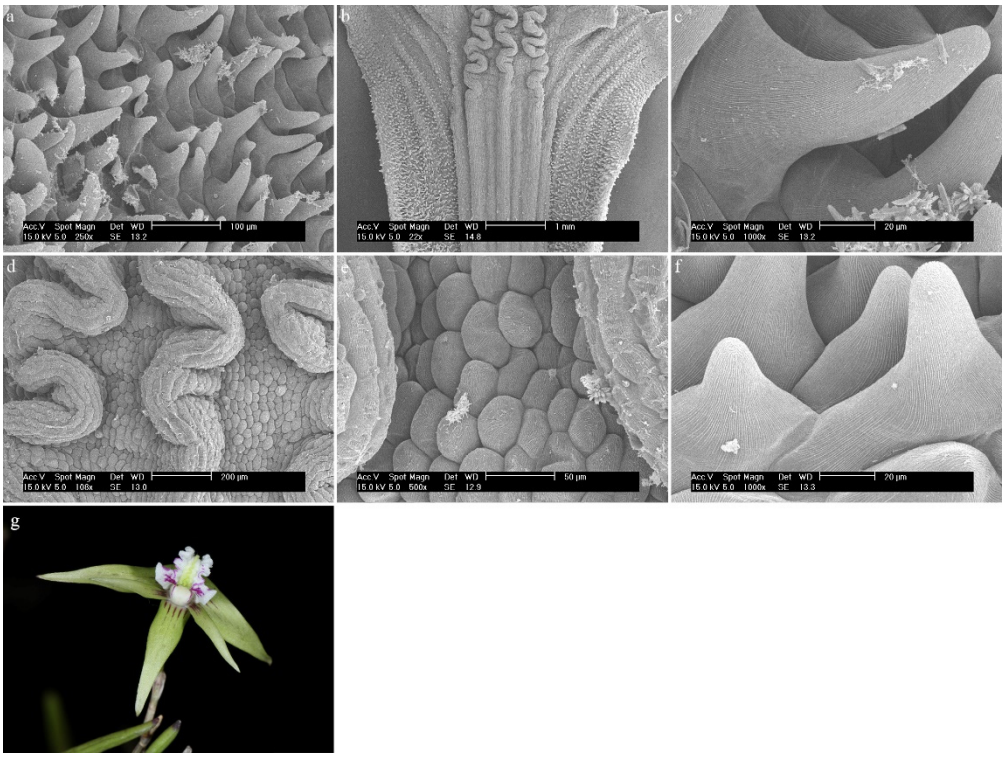

Figure 19. *Dendrobium schoeninum* Lindl.: (**a**) trichomes of the hypochile part of the labellum; (**b**,**c**) trichomes of the mesochile part of the labellum; (**d**,**e**) the mesochile part of the labellum; (**f**) papillae of the margin of the epichile part of the labellum; (**g**) flower (photo by John Varigos).

2.2.9. *Dendrobium* Sw. sect. *Spatulata* Lindl.

1. *Dendrobium discolor* Lindl. The entire surface of the labellum is composed of epidermal cells, covered with a strongly folded cuticle (Figure 20a). There are also protruding epidermal cells, forming a three-fold callus, extending from the hypochile to the epichile (Figure 20b). It is composed of elongated epidermal cells covered with a delicately folded cuticle (Figure 20c). In the hypochile part, there are a few small, conical-shaped papillae (Figure 20d).

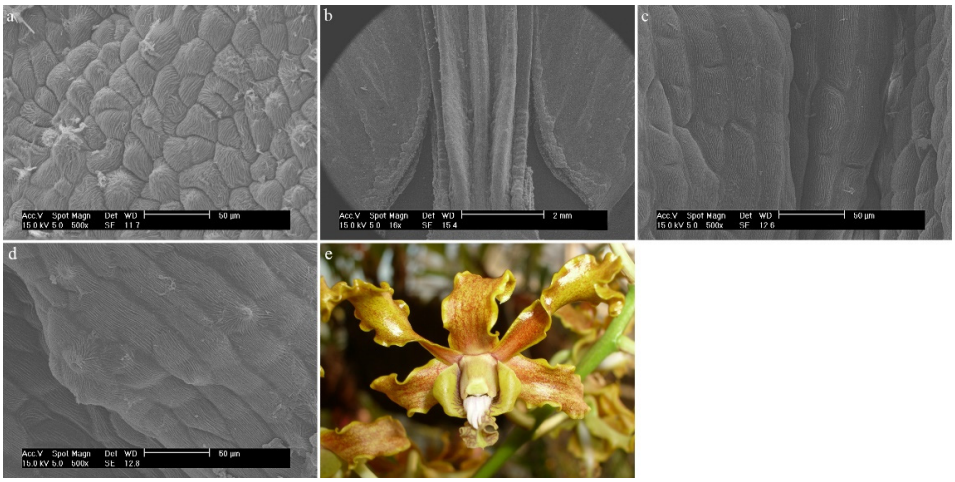

Figure 20. *Dendrobium discolor* Lindl.: (**a**) the epichile part of the labellum; (**b**) the hypochile and the mesochile part of the labellum; (**c**) the hypochile part of the labellum; (**d**) papillae of the hypochile part of the labellum; (**e**) flower (photo by John Varigos).

2.3. Species with No Labellar Protrusions

2.3.1. *Dendrobium* Sw. sect. *Conostalix* Kraenzl.

1. *Dendrobium attenuatum* Lindl. There are elongated epidermal cells with a folded cuticle on the entire labellum (Figure 21a). In the central part of the hypochile, the protruding portions of the labellum are visible, forming a callus reaching all the way to the tip (Figure 21b). The epichile may appear to have papillae, but they are elongated and convex epidermal cells (Figure 21c).

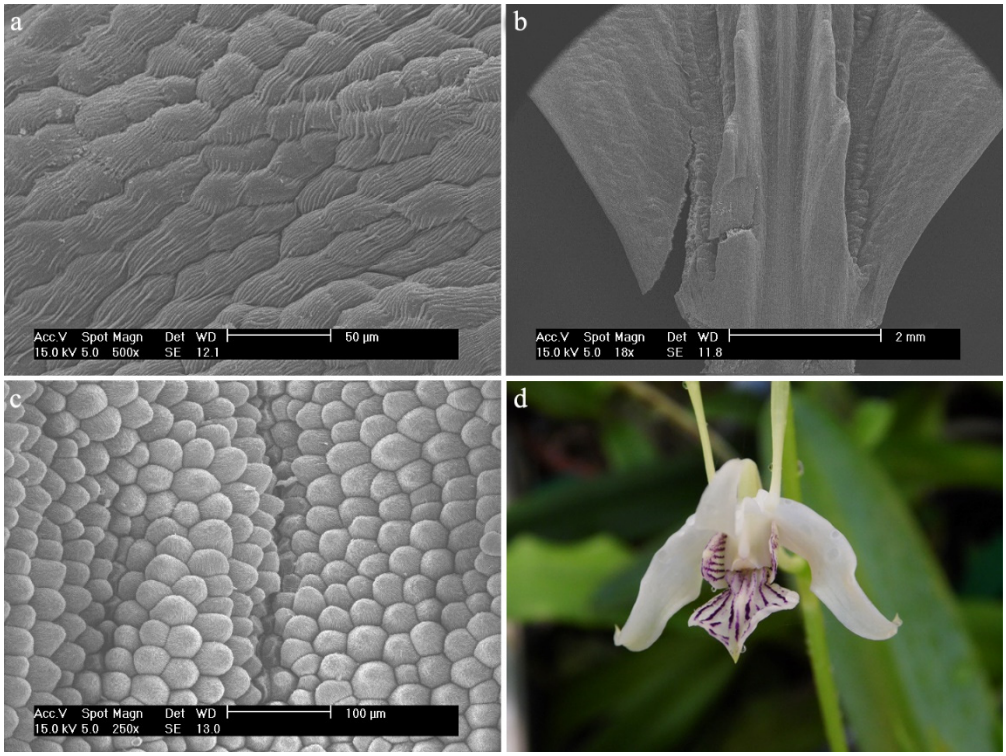

Figure 21. *Dendrobium attenuatum* Lindl.: (**a**) the mesochile part of the labellum; (**b**) the hypochile part of the labellum; (**c**) the epichile part of the labellum; (**d**) flower (photo by Monika Lipińska).

2.3.2. *Dendrobium* Sw. sect. *Distichophyllae* Hk.f.

1. *Dendrobium uniflorum* Griff. The entire surface of the labellum is free of trichomes and papillae. The hypochile is composed of elongated epidermal cells covered with a strongly folded cuticle (Figure 22a). The mesochile and the epichile are filled with delicately arched epidermal cells, covered with a very strongly folded cuticle (Figure 22b,c). (The fluff visible in the last part is not a secretion but dirt).

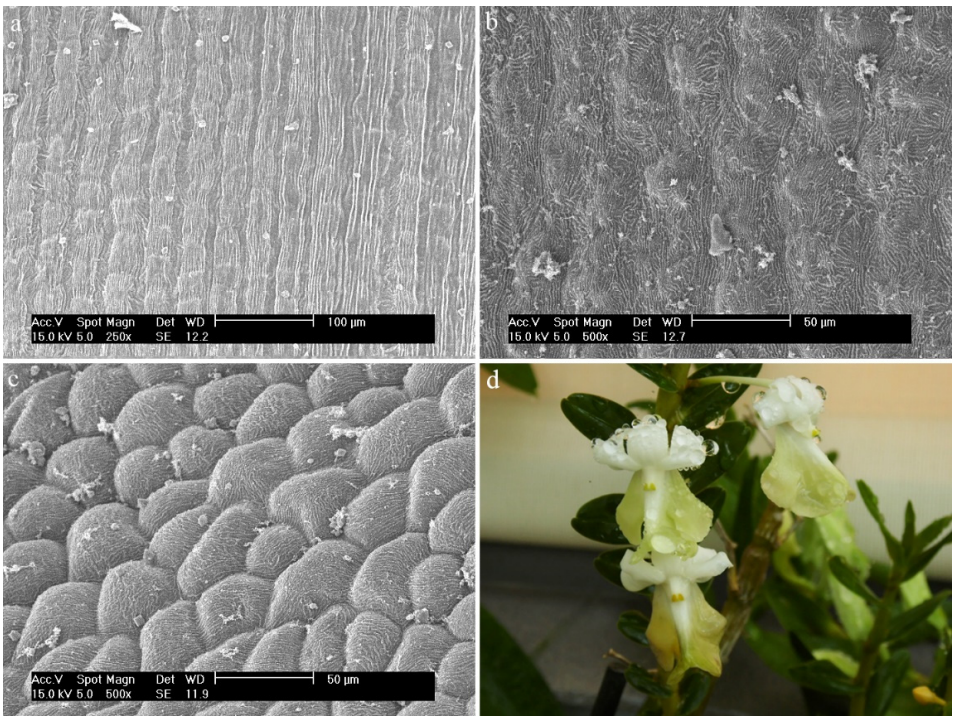

Figure 22. *Dendrobium uniflorum* Griff.: (**a**) the hypochile part of the labellum; (**b**) the mesochile part of the labellum; (**c**) the epichile part of the labellum; (**d**) flower (photo by Monika Lipińska).

2.3.3. *Dendrobium* Sw. sect. *Latouria* (Blume) Miquel

1. *Dendrobium bifalce* Lindl. In the central part of the labellum, there is a callus in the form of three combs, made up of elongated and smooth cells (Figure 23a). The sides of the labellum are composed of epidermal cells covered with a strongly folded cuticle (Figure 23b,c). The sticks visible on the surface are most likely to be bacteria.

2.3.4. *Dendrobium* Sw. sect. *Pedilonum* (Blume) Blume

1. *Dendrobium alaticaulinum* P.Royen The labellum is made up of smooth, regular epidermal cells (Figure 24a). On its edges, there are slightly raised cells covered with a folded cuticle (Figure 24b).

2.3.5. *Dendrobium* Sw. sect. *Spatulata* Lindl.

1. *Dendrobium canaliculatum* R.Br. The entire labellum is made up of typical epidermal cells, covered with a folded cuticle (Figure 25a) that is gathered at the top of each cell (Figure 25b). In the central part of the labellum, a protruding callus is visible, which in the last part takes on a more undulating surface (Figure 25c). In the mesochile, on the lateral parts of the labellum, epidermal cells with possible secretion are visible (Figure 25a).

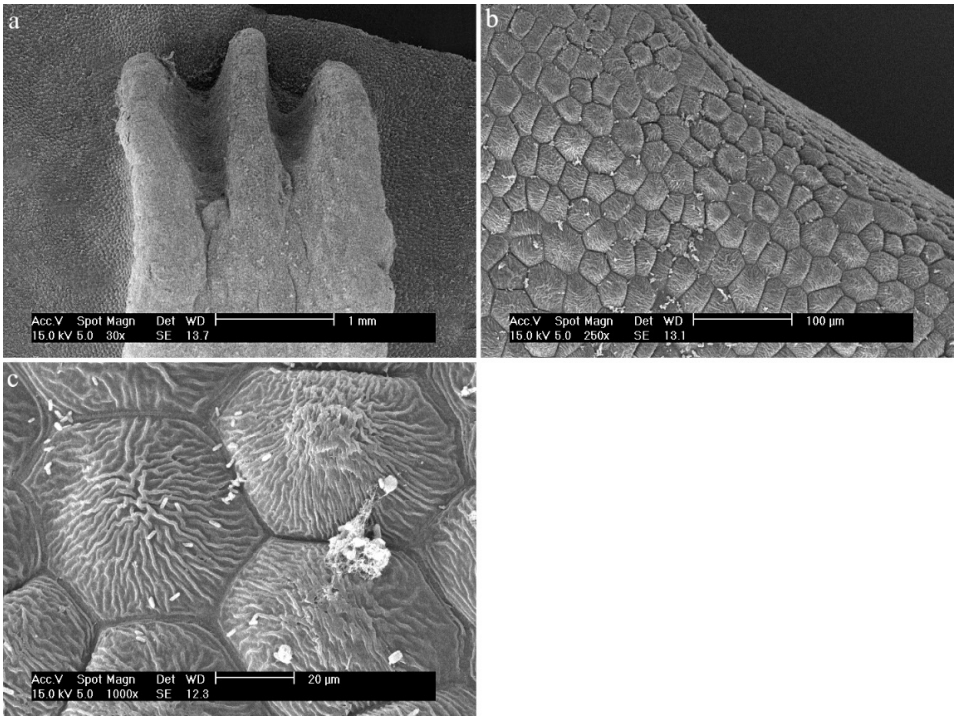

Figure 23. *Dendrobium bifalce* Lindl.: (**a**) the mesochile part of the labellum; (**b**,**c**) the epichile part of the labellum.

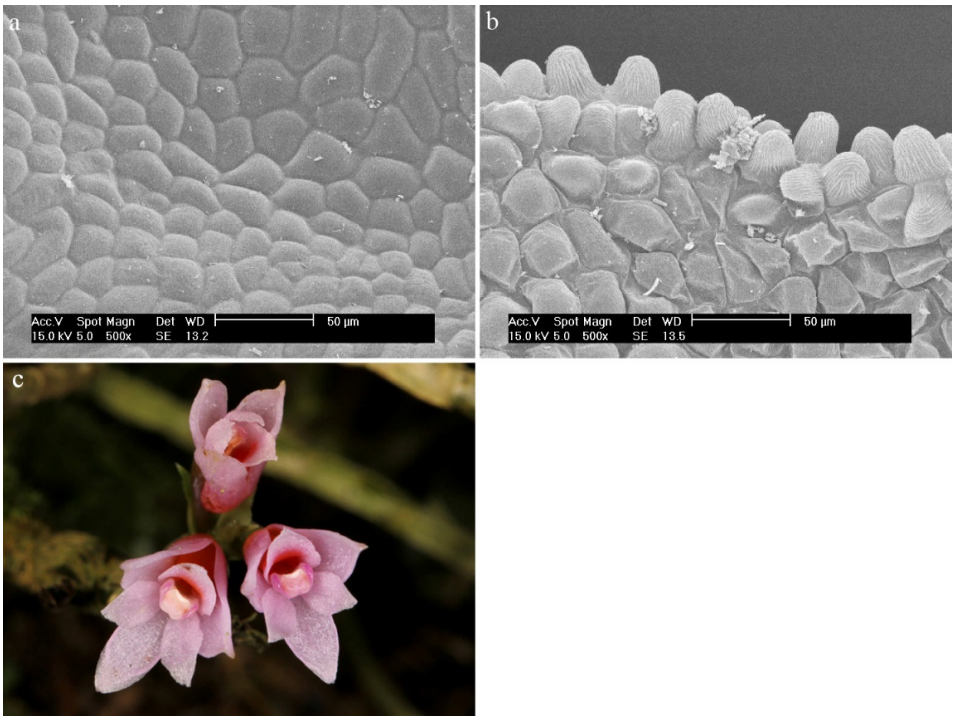

Figure 24. *Dendrobium alaticaulinum* P.Royen.: (**a**) the hypochile part of the labellum; (**b**) the mesochile part of the labellum; (**c**) flower (photo by John Varigos).

2.4. Phylogenetic Analysis

Both applied methodologies (BI and MP) revealed the same results. The analyzed taxa grouped themselves into the same clades. We have noticed no differences in the topology of the trees obtained for the two methods used. Thus, we decided to present a 50% majority-rule consensus tree from the BI analysis (Figures 26 and 27). However, we also placed bootstrap support values on the nodes for each clade. Representatives of the *Dendrobium* sect. *Rhizobium* ranked on the tree with species of the *Dendrocoryne* section (clade 1b) and with taxa of the *Pedilonum* section (clade 11). As a result of the analyses, the representatives of the recently mentioned section also did not form a monophyletic group. *Dendrobium roseipes* (sect. *Pedilonum*) joined together with species from section *Oxyglossum* (clade 12). We can observe a similar situation with taxa from section *Distichophyllae*. Two species of this section, *D. ellipsophyllum* and *D. uniflorum* formed a strongly supported clade 7 (PP = 1, BS = 100) with representatives of section *Conostalix*. In contrast, *D. oligophyllum* was placed at the base of clade 5 as a polytomic branching. It is worth noting that *D. luzonense* (sect. *Grastidium*) did not merge together with other members of the section (clade 2) only created one group with species of the nominal section (clade 8). However, the species of section *Monophyllaea* (clade 1a), *Spatulata* (clade 3), *Latouria* (clade 4), *Formosae* (clade 6), and *Aporum* (clade 10) have formed a strongly supported, monophyletic groups.

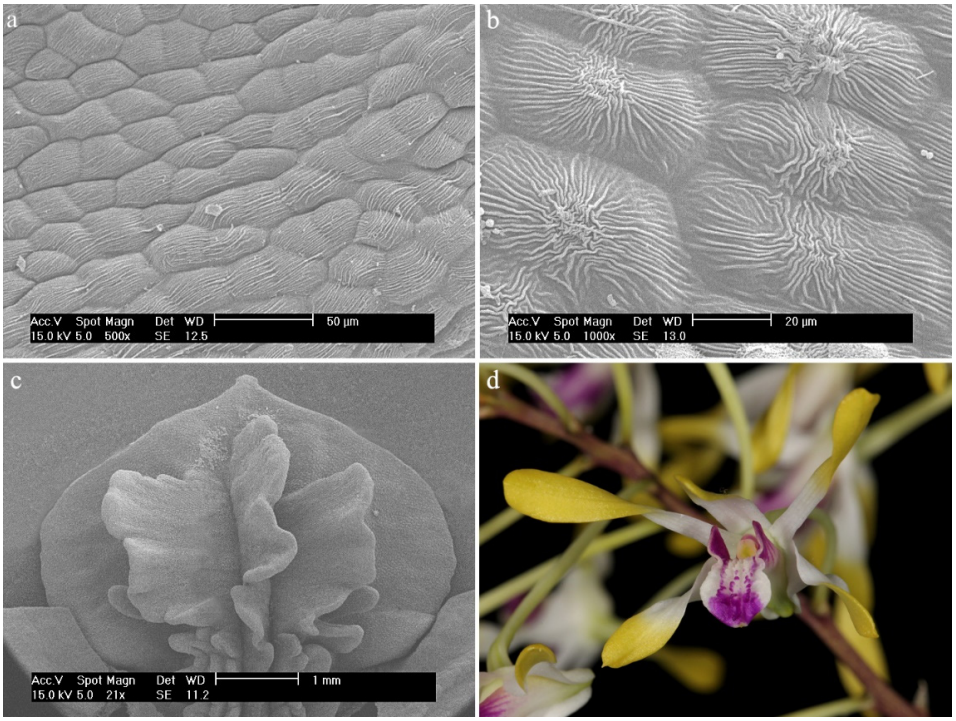

Figure 25. *Dendrobium canaliculatum* R.Br.: (**a**,**b**) the mesochile part of the labellum; (**c**) the epichile part of the labellum; (**d**) flower (photo by John Varigos).

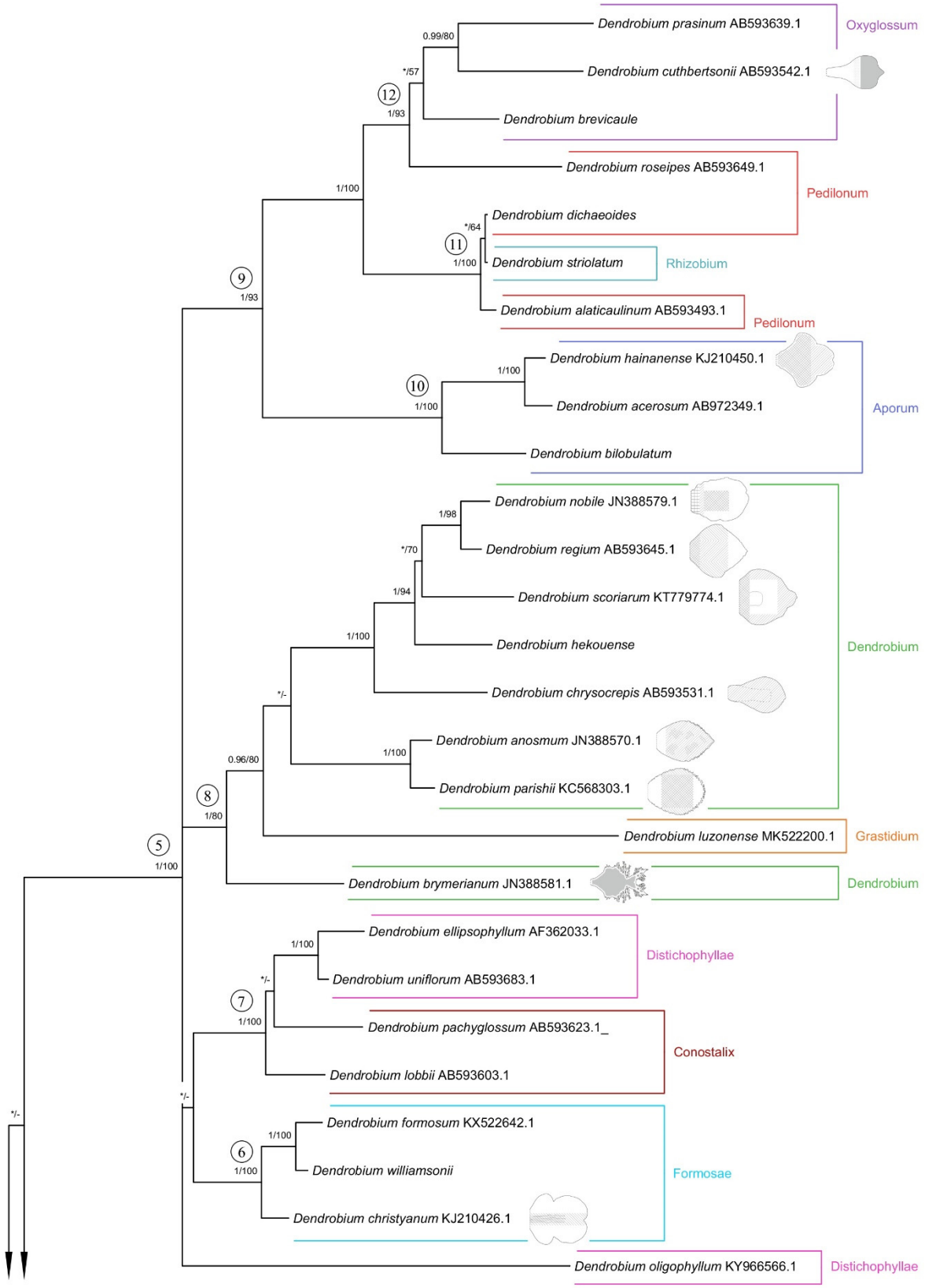

Figure 26. The 50% majority-rule consensus tree from the BI analysis of ITS marker. The numbers above the branches indicate posterior probability (PP) and bootstrap support (BS); PP < 0.95 were marked as an * and BS values of <50% were marked as -. Numbered black circles mark clades discussed in the text.

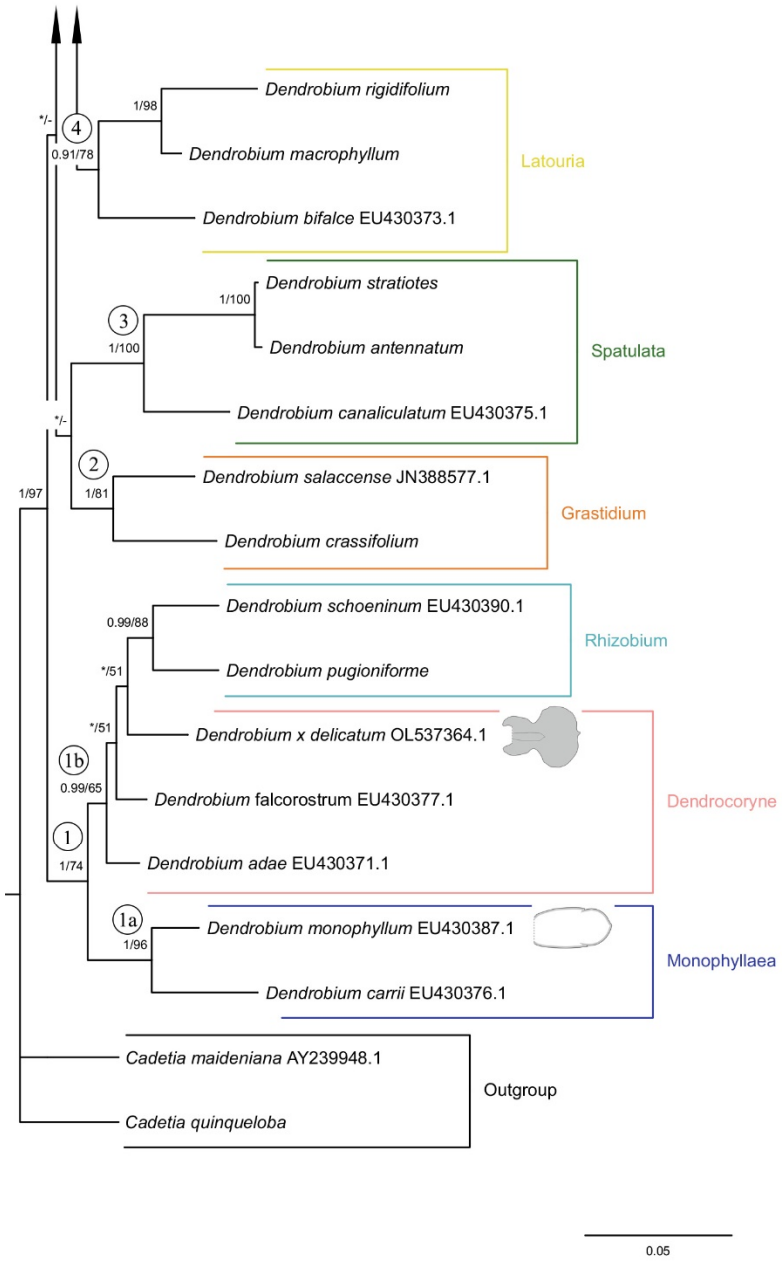

Figure 27. The 50% majority-rule consensus tree from the BI analysis of ITS marker (continuation of Figure 26). The numbers above branches indicate posterior probability (PP) and bootstrap support (BS); PP < 0.95 were marked as an * and BS values of <50% were marked as -. Numbered black circles mark clades discussed in the text.

3. Discussion

Previous studies on the micromorphology structures of the representatives of *Dendrobium* have included only a few species of this genus [13,15]. In 1987, Kjellsson analyzed the labellum of *Dendrobium unicum* and, as in most cases in this study, also noted the presence of trichomes [15]. Although he did not show nutrients in them, he suggested that they might function as a pseudopollen. After extending these analyses and conducting histochemical studies, this fact was also confirmed by Davies [13]. Additionally, in his study, the trichomes were compared to the trichomes from other genera such as *Polystachya* and *Maxillaria* and significant differences were noted—"in that the pseudopollen-forming trichomes consist of a stalk cell and a 'head' of component cells that separate at maturity, in contrast to *Maxillaria* and some *Polystachya* spp. (. . .) *Maxillaria* and *Polystachya* largely contains protein, whereas and

D. unicum the main food substances is starch" [13]. This work is of great value due to the fact that, until now, it is probably the only work analyzing the micromorphology of *Dendrobium*, and further demonstrates the morphological differences of pseudopollen compared to other orchid species. In this paper, there are the results for 21 species of *Dendrobium* presented, in particular from the nominal section.

Our studies embraced the analyses of micromorphology structures present in the labellum. Similar research was also performed for other groups of Orchidaceae, e.g., *Polystachya*, *Maxillaria*, and *Bulbophyllum*. Obviously, due to the large number of species in these genera, the topic is still unexplored. However, thanks to the results obtained by other authors, we can make a partial comparison. The analyses carried out by Teixeira et al. revealed that the most studied species of *Bulbophyllum* possess a callus, and the surface of their labellum has a papillary structure and unicellular trichomes [16]. The presence of secretions was also noted. It is worth mentioning that there are some differences in the structure of the labellum as far as the presence of osmophores and nectaries is concerned. This is related to pollination mechanisms [16]. Species of the genus *Maxillaria* have a variety of labellar structures. The hairs present on the surface may be rounded, elliptical, narrow, elongated, and fusiform, among others, depending on the species [17]. The papillae are most often cone-shaped with pointed and rounded tips, many of which are strongly pigmented [18,19]. As in *Bulbophyllum*, this is related to the diversity of visiting pollinators. Bees appear to be the main pollinators in this case, and pollination by hummingbirds has also been observed, but there is still no concrete evidence to support this [17,18]. Papillae and trichomes are also present on the surface of the labellum of *Polystachya*. The most common type of trichomes found in the analyzed species are uniseriate, two- to four-celled, with a clavate or subclavate terminal cell [8].

A recent paper examined the distribution and density of micromorphology structures on the surface of the labellum in representatives of *Polystachya* [10]. In our study, we also performed a similar analysis. In the future, based on their distribution, we can probably determine their function. Already at this stage, some fairly unambiguous conclusions can be made that the structure of the labellum and the presence of trichomes and papillae are related to pollinators. Of course, we are aware that in order to unambiguously confirm this fact and to concretely relate the distribution of structures to their role, field studies are necessary, which so far have been scarce both in *Dendrobium* and the other genera mentioned here.

Unfortunately, among *Dendrobium* species, still not much is known about pollination. Although there are a few studies on this topic, this thread remains unexplained for now. Of course, the huge number of species does not make it easier to deal with this issue. In 1988, Slater and Calder conducted a study on pollination in *Dendrobium speciosum* Sm. [20]. Unfortunately, there is a lack of micromorphological studies for this species and we do not know exactly whether, apart from typical pollinator-attracting features such as olfactory and visual exposure of flowers, whether the presence or absence of hairs and papillae on the labellum would also potentially be relevant. Based on available information, the species is known to be visited and pollinated by a variety of insects from the genera *Trigona*, *Homalictus*, *Lassioglossum*, and *Hylaeus* [20]. Studies during which species of the genus *Trigona* were determined to be pollinators were also conducted for the species *Dendrobium setifolium* Ridl. and *Dendrobium monophyllum* F.Muell. [21,22]. However, as in other cases previously, micromorphology was not analyzed. In the case of *D. monophyllum*, it is likely that bees are attracted by aromatic floral compounds. We also know that this species does not produce nectar [22]. We noted, based on studies from other groups, that often, nectar-secreting species were devoid of various ornaments, whereas, in the absence of nectar, structures on the epidermis appeared, thus suggesting the presence of a putative reward [23]. Our analyses showed that *D. monophyllum* has conical papillae. It is likely that in addition to floral compounds, these structures also contribute to attracting pollinators. Studies in the context of the pollination mechanism were also conducted for *Dendrobium infundibulum* Lindl. According to the analyses, it is mainly pollinated by *Bombus eximus*. *D. infundibulum*

does not attract by scent, has no nectar, or offers any other reward to the bumblebee. Its success is based on precise pollen placement on insects as a result of a high degree of adaptation of flower morphology to the pollinator [24,25]. As in other cases, little is known about the micromorphology of this species. *Dendrobium finisterrae* Schltr. is an interesting case because certain features, such as zygomorphic flower structure, creamy green color with nectar conductors, and pleasant odor, suggest that it may be pollinated by bees. However, it also has other features, such as massive nectary spur and collenchymatous secretory tissue, indicating pollination by birds. To date, there is no clarification on this issue [26]. Another known species associated with bird pollination is *Dendrobium secundum* (Blume) Lindl. ex Wall. [27]. A very interesting phenomenon, rarely occurring among *Dendrobium*, is the self-pollination observed in *Dendrobium biflorum* (G.Forst.) Sw. on the Society Islands. This may be due to a lack of suitable pollinators or stressful growing conditions in that area. However, it is not determined whether autogamy would also occur in these taxa outside the mentioned study area [28].

The fact that structures on the labellum can be attractive to pollinators was proved by Davies by analyzing the micromorphology of *D. unicum*, which has multicellular trichomes on its labellum, as mentioned earlier [13]. This species belongs to the nominal section. In our work, we analyzed the labella of as many as seven species from this group, and every sample had micromorphology structures on the labellum, papillae, or trichomes. This may indicate two important facts: first, species in this section can also be pollinated by bees, such as *D. unicum*; second, this is an important taxonomic issue. It seems to be no coincidence that all the species of the nominal section we studied have the analyzed structures, and additionally, another taxon of this group analyzed in earlier studies has such features. This may indicate that this information has useful taxonomic value for species identification at the section level. Moreover, in 2016 a new species *Dendrobium maguanense* Q.Xu & Z.J.Liu was discovered, which is closely related to *Dendrobium crepidatum* Lindl. & Paxton. It is sometimes treated as a synonym for the latter, but the authors of the publication showed differences between the two species. Molecular analysis indicates that they belong to the section *Dendrobium* and according to the authors describing *D. maguanense*, its labellum is also covered with hairs [29]. Another species relatively recently described and included by the authors in the nominal section is *Dendrobium wenshanense* Q.Xu, Y.B.Luo & Z.J.Liu, and it also has hairs on the labellum [30].

Moreover, we were also interested in the fact that *Dendrobium alaticaulinum* P.Royen belonging similarly to *D. secundum* to the sect. *Pedilonum* (Blume) Blume has no structures on the labellum. Due to the fact that they belong to the same section we wondered if the labellum of *D. secundum*, which we mentioned earlier, is also smooth and if the lack of trichomes and papillae could be related to pollination by birds. Furthermore, as we mentioned above, there is speculation that *D. finisterrae* belonging to section *Latouria* (Blume) Miquel can be pollinated by birds as well. In our study, we analyzed the species *Dendrobium bifalce* Lindl. from this section, and interestingly, this taxon was one of the few that did not have structures on the labellum.

Of course, our assumptions should be interpreted with caution. We know that because of such a large number of species in *Dendrobium*, our sample is limited and this study is only preliminary in this broad area. Based on previous studies of other genera, it is hard to conclusively assess whether we definitely have an important taxonomic trait for *Dendrobium*. Similar studies were conducted in 2021 by Mytnik for the genus *Polystachya*, and in that case, the presence and absence of structures were important for taxonomy [10]. We are also inclined to such conclusions for *Dendrobium*, but nevertheless, take into account that this is not always the case. This is evidenced, for example, by studies conducted on the genus *Maxillaria*, in which the overall conclusions were that the papillae were of little taxonomic value in this case [31].

In our studies, we also went a step further and compared how the presence of micromorphological structures for each section is distributed on the phylogenetic trees obtained by other authors [3,5]. It is interesting to note that there is an incongruence in the topology

of the tree based on the sequence of nuclear region ITS-5.8S-ITS2 and the classification proposed by Wood [4]. Numerous sections appear to be polyphyletic. We did not find correlations of the phylogenetic position of the species and the distribution of micromorphological features of the lip, either. The analyzed features' distribution seems to respond to pollinator pressure. Based on the resulting tree (Figures 26 and 27), we can confirm the above statement that the analyzed traits arose convergently. Here again, however, we must state that field studies are necessary to be certain of the above statement and also to relate our analyses of the distribution of structures on labella to their function. We realize that this is only the first step taken though, leading to very clear and interesting conclusions, which even at this stage of research on such a large genus seem highly probable.

4. Materials and Methods

4.1. Study Using Scanning Electron Microscope (SEM)

The materials used in this study came from the collections gathered at the Department of Plant Taxonomy and Nature Conservation at the University of Gdansk, Poland. All specimens were preserved in Kew Mixture consisting of 53% of ethyl alcohol, 37% of water, 5% of formaldehyde, and 5% of glycerol. They embraced 21 species of *Dendrobium*, representing 13 sections of this genus. The list of materials used is presented in Table 3. The classification is consistent with the one proposed by Wood [4].

Table 3. List of species of *Dendrobium* used in this study (classification of the taxa follows the Wood systematic [4].

Section	Species	Accession Number
Aporum Blume	*Dendrobium hainanense* Rolfe	UGDA.0051616
Conostalix Kraenzl.	*Dendrobium attenuatum* Lindl.	UGDA.0052546
Dendrobium Sw.	*Dendrobium anosmum* Lindl.	UGDA.0035621
	Dendrobium brymerianum Rchb.f.	UGDA.0035412
	Dendrobium chrysocrepis C.S.P.Parish & Rchb.f. ex Hook.f.	UGDA.0116182
	Dendrobium nobile Lindl.	UGDA.0116184
	Dendrobium parishii H.Low	UGDA.0034923
	Dendrobium regium Prain	UGDA.0116181
	Dendrobium scoriarum W.W.Sm.	UGDA.0116183
Dendrocoryne Lindl. & Paxton	*Dendrobium* × *delicatum* (F.M.Bailey) F.M.Bailey	UGDA.0051595
Disttichophyllae Hk.f.	*Dendrobium uniflorum* Griff.	UGDA.0051736
Formosae (Benth. And Hk.f.) Hk.f.	*Dendrobium christyanum* Rchb.f.	UGDA.0052513
Grastidium (Blume) Blume	*Dendrobium katherinae* A.D.Hawkes	UGDA.0045354
Latouria (Blume) Miquel	*Dendrobium bifalce* Lindl.	UGDA.0034715
Monophyllaea Benth.	*Dendrobium monophyllum* F.Muell.	UGDA.0034728
Oxyglossum Schltr.	*Dendrobium cuthbertsonii* F.Muell.	UGDA.0034867
Pedilonum (Blume) Blume	*Dendrobium alaticaulinum* P.Royen	UGDA.0045288
Rhizobium Lindl. & Paxton	*Dendrobium mortii* F.Muell.	UGDA.0034479
	Dendrobium schoeninum Lindl.	UGDA.0034714
Spatulata Lindl.	*Dendrobium canaliculatum* R.Br.	UGDA.0035422
	Dendrobium discolor Lindl.	UGDA.0034720

Obtaining the appropriate photos and then drawing the correct conclusions using a scanning electron microscope required several stages of preparation of the research material. In the first step, dehydration took place, which means treating the fixed material with aqueous solutions of ethyl alcohol of increasing concentration from 25% to 100%. The dehydration procedure was carried out at room temperature, and the treatment time of the material in each of the aqueous ethyl alcohol solutions ranged from 10 to 15 min [32]. The

next stage of preparations was drying the samples with the use of liquid carbon dioxide, using the occurrence of the so-called critical point using critical point dryer mark Emitech-model K850. As a result of this method, there are fewer unnecessary artifacts on the surface of the analyzed samples [33].

After that, the dried material was glued to microscope stubs, then coated with gold using Spi Module Sputter Coater. The material prepared in this way was analyzed using a Philips XL-30 (FEI) scanning electron microscope (Laboratory of Electron Microscopy, University of Gdańsk, Gdańsk, Poland) operating at an accelerating voltage of 5 or 15 kV [10].

4.2. Molecular Analyses

For phylogenetic reconstruction, we applied 44 sequences of the ITS region representing species of *Dendrobium* Sw. and *Cadetia* Gaudich. as outgroup taxa. Most of the sequences used in this article were downloaded from GenBank (http://www.ncbi.nlm.nih.gov). A list of the taxa with their accession numbers is included in Supplementary Materials Table S1. However, samples (leaf fragments) for 13 species of *Dendrobium* used for molecular analyses were collected in the greenhouse of the Faculty of Biology at the University of Gdansk. All sequences obtained from them were deposited in GenBank. The exact ID numbers for these samples and GenBank accession numbers are presented in Table 4.

Table 4. List of species of *Dendrobium* used in molecular study.

Section	Species	Accession Number	GenBank Accession Number
Aporum Blume	*Dendrobium bilobulatum* Seidenf.	UGDA.0076220	ON694111
Cadetia Gaudich.	*Cadetia quinqueloba* Schltr.	UGDA.0076230	ON694123
Dendrobium Sw.	*Dendrobium hekouense* Z.J.Liu & L.J.Chen	UGDA.0076247	ON694112
Formosae (Benth. And Hk.f.) Hk.f.	*Dendrobium williamsonii* Day & Rchb.f.	UGDA.0076204	ON694113
Grastidium (Blume) Blume	*Dendrobium crassifolium* Schltr.	UGDA.0076236	ON694114
Latouria (Blume) Miquel	*Dendrobium macrophyllum* A.Rich. *Dendrobium rigidifolium* Rolfe	UGDA.0076205 UGDA.0076226	ON694116 ON694115
Oxyglossum Schltr.	*Dendrobium brevicaule* Rolfe	UGDA.0076232	ON694117
Pedilonum (Blume) Blume	*Dendrobium dichaeoides* Schltr.	UGDA.0076215	ON694118
Rhizobium Lindl. & Paxton	*Dendrobium pugioniforme* A.Cunn. ex Lindl. *Dendrobium striolatum* Rchb.f.	UGDA.0076149 UGDA.0076231	ON694120 ON694119
Spatulata Lindl.	*Dendrobium antennatum* Lindl. *Dendrobium stratiotes* Rchb.f.	UGDA.0076155 UGDA.0076213	ON694122 ON694121

The DNA was extracted using the DNA Sherlock AX Kit (A&A Biotechnology, Gdańsk, Poland), following the manufacturer's protocol. Amplification and sequencing reactions were performed for nuclear region ITS1+5.8S+ITS2 using the same pair of primers, 101F and 102R [34]. The total volume of sample for PCRs was 25 µL containing 1 µL template DNA (~10–100 ng), 0.5 µL of 10 µM of each primer, 12.5 µL Start-Warm HS-PCR Mix (A&A Biotechnology, Gdańsk, Poland), and water. At the same time, the reaction parameters were taken, as Baranow [35]. The Clean-Up Concentrator Kit (A&A Biotechnology) was used to clean the PCR products following the manufacturer's protocol. Then, the sequencing reaction was prepared and performed by Macrogen (Seoul, South Korea; http://dna.macrogen.com/eng/), using the same primers as mentioned above. All DNA sequence chromatograms were examined/edited in FinchTV (https://finchtv.software.informer.com/1.4/).

The multiple sequence files were aligned with SeaView [36] using the "align" option according to the MUSCLE algorithm [37]. The best fit substitution model was calculated

with MrModeltest 2.2 and by both criteria [38], hLRTs (hierarchical likelihood ratio test) and AIC (Akaike information criterion) were selected GTR+G+I. For phylogenetic reconstruction, we used Bayesian inference (BI) with MrBayes v. 3.2.7a [39] and Maximum parsimony (MP) using PAUP* [40]. We used two different methods to test for possible topological incongruence.

In BI, we indicated two independent runs of four Markov-chain Monte Carlo (MCMC) chains were started from different random trees to ensure that individual runs had converged to the same result. We used 1 million generations per run with sampling every 100 generations. Convergence was assessed using the average standard deviation of split frequencies below 0.01. Thereafter, we discarded the initial 25% of the sampled generations of each chain as burn-in. Saved trees were summarized in a majority rule consensus tree. The nodal confidence was assessed by posterior probabilities (PP), which were considered strongly supported when equal to or higher than 0.95 [41].

MP analysis was undertaken with tree-bisection-reconnection (TBR) branch swapping and the MULTREES option in effect, simple addition, and ACCTRAN optimization. All characters were equally weighted [42], and missing data were coded as "?", and gaps as "-". All parsimonious trees (10,000) were used to obtain a strict consensus tree. We also performed bootstrap analysis using 1000 replicates in order to determine the internal support of clades (BS).

The BI tree was edited with FigTree v.1.4.4 (http://tree.bio.ed.ac.uk/software/figtree/) and Inkscape (https://inkscape.org/release/inkscape-1.0.2/).

5. Conclusions

In this study, 21 species of *Dendrobium* representing 13 sections were analyzed, including 7 species of the nominal section. The labella of all taxa in this group were covered with two different micromorphology structures. We observed cylindrical and conical trichomes, and small conical and semicircular papillae. Most species from the other sections also had conical and cylindrical trichomes and papillae, but ellipsoidal hairs and semicircular papillae were also present. In five species, the presence of the analyzed structures was not observed. This study is the first analysis in the genus *Dendrobium* focusing typically on micromorphology. Based on the micromorphological results obtained and the phylogenetic analyses performed, we suggest that the presence and absence of structures on the lips is due to convergence, and this is closely related to pollinator pressure. Of course, we recognize that confirming this thesis and accurately linking the function to the presence and distribution of structures requires an expanded study group, additional field studies, and pollinator observations. Nevertheless, the paper provides a wealth of data and a decent basis for expanding the study.

Supplementary Materials: The supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/ijms23179578/s1.

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Supplementary Materials

Section	Species	GenBank accession number
Aporum Blume	*Dendrobium acerosum* Lindl.	AB972349.1
	Dendrobium hainanense Rolfe	KJ210450.1
Cadetia Gaudich.	*Cadetia maideniana* (Schltr.) Schltr.	AY239948.1
Conostalix Kraenzl.	*Dendrobium lobbii* Teijsm. & Binn.	AB593603.1
	Dendrobium pachyglossum C.S.P.Parish & Rchb.f.	AB593623.1
Dendrobium Sw.	*Dendrobium anosmum* Lindl.	JN388570.1
	Dendrobium brymerianum Rchb.f.	JN388581.1
	Dendrobium chrysocrepis C.S.P.Parish & Rchb.f. ex Hook.f.	AB593531.1
	Dendrobium nobile Lindl.	JN388579.1
	Dendrobium parishii H.Low	KC568303.1
	Dendrobium regium Prain	AB593645.1
	Dendrobium scoriarum W.W.Sm.	KT779774.1
Dendrocoryne Lindl. & Paxton	*Dendrobium adae* F.M.Bailey	EU430371.1
	Dendrobium falcorostrum Fitzg.	EU430377.1
	Dendrobium × *delicatum* (F.M.Bailey) F.M.Bailey	OL537364.1
Distichophyllae Hk.f.	*Dendrobium ellipsophyllum* Tang & F.T.Wang	AF362033.1
	Dendrobium oligophyllum Gagnep.	KY966566.1
	Dendrobium uniflorum Griff.	AB593683.1
Formosae (Benth. And Hk.f.) Hk.f.	*Dendrobium christyanum* Rchb.f.	KJ210426.1
	Dendrobium formosum Roxb. ex Lindl.	KX522642.1
Grastidium (Blume) Blume	*Dendrobium luzonense* Lindl.	MK522200.1
	Dendrobium salaccense (Blume) Lindl.	JN388577.1
Latouria (Blume) Miquel	*Dendrobium bifalce* Lindl.	EU430373.1
Monophyllaea Bentham	*Dendrobium carrii* Rupp & C.T.White	EU430376.1
	Dendrobium monophyllum F.Muell.	EU430387.1
Oxyglossum Schltr.	*Dendrobium cuthbertsonii* F.Muell.	AB593542.1
	Dendrobium prasinum Lindl.	AB593639.1
Pedilonum (Blume) Blume	*Dendrobium alaticaulinum* P.Royen	AB593493.1
	Dendrobium roseipes Schltr.	AB593649.1
Rhizobium Lindl. & Paxton	*Dendrobium schoeninum* Lindl.	EU430390.1
Spatulata Lindl.	*Dendrobium canaliculatum* R.Br.	EU430375.1

2. Artykuł badawczy II

Burzacka-Hinz, A., Dudek, M., & Szlachetko, D. L. (2024). Potential use of low-copy nuclear gene *Xdh* at lower taxonomic levels based on phylogenetic analysis of the nominal section of *Dendrobium*. *Acta Societatis Botanicorum Poloniae*, *93*, 7922.

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Authors' Contributions
AB-H, MD, DLS: Research concept and design; AB-H: Collection and/or assembly of data; AB-H, MD, DLS: Data analysis and interpretation; AB-H, MD: Writing the article; MD, DLS: Critical revision of the article; DLS: Final approval of the article

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Competing Interests
No competing interests have been declared.

RESEARCH PAPER

Potential use of low-copy nuclear gene *Xdh* at lower taxonomic levels based on phylogenetic analysis of the nominal section of *Dendrobium*

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Abstract

Dendrobium is a large group of Orchidaceae, counting more than 1,100 species, whose classification, both within the genus and individual sections, is not straightforward. Therefore, the aim of our study was to perform phylogenetic analyses for representatives of a nominal section of *Dendrobium* and to test the usefulness of a low-copy nuclear gene encoding a xanthine dehydrogenase protein for reconstructing phylogeny. We also wish to compare the utility of two nuclear markers, *Xdh* and ITS. To realize this, we analyzed the diversity of the two markers and the reliability of the resulting trees. Our results indicate that the nuclear ITS region shows higher variability and clade credibility in trees, in closely related species, and between sections than the low-copy nuclear gene. *Xdh* may be more reliable at higher taxonomic levels, but confirmation of this requires further research.

Keywords

Dendrobium; *Xdh*; nrITS; utility; Orchidaceae; phylogeny; plastid markers

1. Introduction

Dendrobium is one of the orchid genera that is abundant in species occurring in Asia, Australasia, and Australia (Xiang et al., 2013). In total, it comprises about 1,100 species (Burke et al., 2008; Moudi et al., 2013; Takamiya et al., 2014).

Although it was described more than 200 years ago (Swartz, 1799), there is still a great desire to investigate it. Identification of *Dendrobium* species is very difficult. This is probably due to the high level of polymorphism, the large number of species, the wide range of occurrence, and their geographical distribution also contributes to identification problems within this taxon (Takamiya et al., 2014). Attempts to classify this genus have been made since the 19th century, first on the basis of morphological characters and later on phylogenetic analyses. To date, there has been no consistent classification of *Dendrobium*, and no unambiguous taxonomic treatment, either between species within the genus or within particular sections (Clements, 2006; Pridgeon et al., 2014; Takamiya et al., 2011; Wang et al., 2009; Xiang et al., 2013). Establishing a consistent classification of *Dendrobium* is one of the greatest challenges among orchidologists (Adams, 2011; Adams et al., 2006; Feng et al., 2015a, 2015b; Morris et al., 1996; Xiang et al., 2013; Yukawa et al., 1996; Yukawa & Uehara, 1996). A proposal for intraspecific classification based on morphology alone is almost impossible. For this purpose, it is also necessary to use information stored in DNA.

To reconstruct the phylogeny, the plastid DNA data was often used: i.e., markers such as *mat*K, *rbc*L, *ycf*1, the *trn*L-*trn*F fragment, *rpl*16, and *rpl*32 (Neubig et al., 2009; Niu et al., 2020; Shaw et al., 2014). Unfortunately, the phylogenetic trees obtained from these are often inconsistent. These markers are easy to align but are sometimes too conservative and lack sufficient variability. Consequently, the mentioned genes provide too little phylogenetic information and do not resolve intra- and interspecies relationships (Neubig et al., 2009; Zhao

et al., 2019). Studies of their heredity indicate only maternal transmission, with no evidence of paternal or two-parental inheritance (Cafasso et al., 2005). Consequently, cpDNA provides only half of the ancestry in hybrid and polyploid plants and thus does not identify phylogenetic conflicts arising from hybrid ancestry (Small et al., 2004). The selection of suitable markers is, therefore, one of the most important steps in phylogenetic research. This task has been made easier by the use of next-generation sequencing methods. In the last decade, we have come to know the sequences of complete chloroplast genomes, including many representatives of orchids (Zheng et al., 2018; Zhitao et al., 2017). However, information on the nuclear genome is still quite limited. There is a lack of data that unequivocally indicates the efficacy of a specific nuclear marker, so we decided to focus our analyses primarily on this area.

So far, among nuclear sequences, the most commonly used markers are ITS. These fragments are considered universal due to the presence of many genetic variations and a high copy number. A single copy is usually a combination of two ITS fragments and genes encoding a large and small ribosomal subunit, i.e. 18S-ITS1-5.8S-ITS2-26S. The main advantage of nuclear genes is the higher rate of sequence evolution compared to organelle genes. This is particularly important at low taxonomic levels (Soltis & Soltis, 1998). Furthermore, numerous molecular studies have been carried out using the above-mentioned markers due to the ease and relatively low cost of obtaining sequences, as well as their wide availability.

However, in recent decades, the use of nuclear low-copy genes seemed to be a breakthrough in reconstructing the course of phylogeny. Many of them contain large amounts of genetic information, i.e., informative sites. Moreover, like multicopy nuclear genes, they are also biparentally inherited and often show rapid rates of evolution (Small et al., 2004). Furthermore, they are potentially ideal markers for resolving polyploidy and hybridization (Górniak et al., 2010; Hazra et al., 2020; Peng & Wang, 2008; Russell et al., 2010; Small et al., 2004; Yin et al., 2020). In addition, low-copy-number markers can be used to reconstruct phylogeny at all taxonomic levels, especially where universal markers are unable to generate strong phylogenetic hypotheses (Bratzel et al., 2020; Sang, 2002). However, it should be remembered that they also have their disadvantages. They occur in low copy numbers and sometimes require additional laboratory work, such as cloning or designing their PCR primers (Li et al., 2019). The *Xdh* gene has been identified as one of the low-copy markers for use in phylogenetic analyses in the framework of Górniak et al. (2010), demonstrating the possibility of using the low-copy *Xdh* gene in representatives of the Orchidaceae (Górniak et al., 2010). The use of *Xdh* in determining phylogenetic relationships has occurred and continues to do so among different genera belonging to this family (*Bulbophyllum* Thouars, *Campylocentrum* Benth., *Paphiopedilum* Pfitz.) and at various phylogenetic levels, as well as among other plant groups (Chen et al., 2019; Górniak et al., 2021; Jin et al., 2017; Kikuchi et al., 2020; Nowak et al., 2023; Pessoa et al., 2018; Viruel et al., 2018). However, it is worth noting that often, due to the need to put more effort into laboratory analyses, the authors of the papers are less likely to decide to choose them.

This marker encodes a protein, xanthine dehydrogenase, which belongs to the molybdenum cofactor-dependent class of hydroxylase enzymes. *Xdh* plays an important role in the degradation of nucleic acids in bacteria, plants, and animals (Górniak et al., 2010; Rodríguez-Trelles et al., 2001). It takes part in the regulation of normal plant growth and aging processes (Han et al., 2020). It also participates in other important physiological processes, such as plant response to pathogen attack, acclimatization, and cell death associated with the hypersensitivity response (Taylor & Cowan, 2004; Watanabe et al., 2010). However, what is most important for taxonomists and this work is that the *Xdh* marker participates in phylogenetic studies of plants. In previous studies on the genus *Dendrobium*, the aforementioned gene has only been used once by Moudi and Go (2015), who managed to obtain sequences for 20 *Dendrobium* species from the Malaysian area. Therefore, we expected that it would also prove useful in studies on the phylogeny of the nominal section.

The aim of our work was to test the suitability of the low-copy *Xdh* gene for reconstructing section-level phylogeny based on the nominal section of *Dendrobium*. We performed phylogenetic trees using both the *Xdh* gene and ITS, as well as *trnL-trnF* and *mat*K. We collected quantitative data for the aforementioned markers and determined sequence length, number of fixed and variable sites, and number of parsimony-informative sites. In addition, our results also enriched the GenBank database with new sequences for the *Xdh* marker, which has so far been rarely represented in this database.

2. Material and methods

2.1. Material

2.1.1. Research object

In our studies, we used samples representing various species of *Dendrobium*. The members of this genus are mainly epiphytes with a sympodial type of growth. They are characterized by a lateral inflorescence emerging from the upper part of the stem, a mentum formed by lateral sepals and a prominent column foot, four naked pollinia, and swollen rostellum (Pridgeon et al., 2014). It should be noted that *Dendrobium* is one of the larger genera within the Orchidaceae. Therefore, focus primarily on the nominal section. It contains over 50 species, including the generitype *Dendrobium moniliforme* (L.) Sw. (Wood, 2006). The range of its representatives is almost as wide as that of the genus, except for the area of Micronesia and Melanesia (Takamiya et al., 2014). The main morphological characters that distinguish this section are a tall, leafy stem and multiple racemes carrying a few flowers with an expanded, tomentose, and velvety lip (entire in most species, without sidelobes). The color of the flowers is usually pink to purple or white. They bloom for about four weeks in the spring (Wood, 2006).

2.1.2. Plant material

The plant material used in these studies came from the gathering at the Department of Plant Taxonomy and Nature Conservation at the University of Gdansk, Poland. We have selected for testing all species that have ever been classified into the section *Dendrobium* (Clements, 2006; Feng et al.,

2014; Schuiteman, 2011; Takamiya et al., 2014; Wood, 2006; Xiang et al., 2013). For each marker, we tried to use sequences from the same species. In the case of the *Xdh* gene dataset, we also extended our analyses to species outside the nominal section. All DNA sequences obtained have been deposited in GenBank. The ID numbers for these samples and GenBank accession numbers are presented in Table S1. In addition, part of the sequences representing outgroup taxa and species of *Dendrobium* outside the nominal section for the *Xdh* marker were downloaded from GenBank (Table S1). Sequences were also taken for other utilized markers, and the list of these taxa is included in Table S2.

2.2. Molecular methodologies

For the phylogenetic reconstruction and to test the utility of the low-copy nuclear gene *Xdh*, we obtained 29 sequences representing species of the nominal section.

2.2.1. DNA isolation

Total genomic DNA was extracted from 20–100 mg of dried leaves stored in silica gel (Chase & Hills, 1991). The extraction was performed using a Sherlock AX kit (A&A Biotechnology, Poland), and the procedure was carried out with the attached protocol. The pellets of DNA were suspended in 50 µL of TE buffer.

2.2.2. Amplification and sequencing

Polymerase chain reaction (PCR) and sequencing reaction were performed using the same primers. The X551F and X1591R for the *Xdh* marker (Górniak et al., 2010), 101 F and 102 R for nrITS fragments (Douzery et al., 1999), and *trn*L-c and *trn*L-f for *trn*L-*trn*F markers (Taberlet et al., 1991). PCRs were carried out in a total volume of 25 µL containing 1 µL template DNA (~10–100 ng), 1 µL of 10 µM of each primer, 12.5 µL Hot-Start PCR Mix-Start Warm (A&A Biotechnology, Poland), and water. The amplification of the *Xdh* marker used a touchdown method, which involves lowering the attachment temperature of the primer by one degree for the first seven cycles. It was done to increase the efficiency of the reaction and is extremely important when amplifying low-copy genes. The parameters were the following: the initial denaturation 95 °C, 5 min and then by seven cycles denaturing in 94 °C, 45 s; primer annealing 59 °C, 45 s (reducing 1 °C per cycle) and extending in 72 °C, 90 s. The next 30 cycles proceeded in 94 °C, 45 s; 52 °C, 45 s; 72 °C, 90 s, and final elongate 72 °C, 10 min. All products of the PCR reaction were tested using electrophoresis in 1% agarose gel at 110 V for 40 minutes and then were purified using the PCR Clean-Up System (Promega, US) and DNA Clean-Up Concentrator Kit (A&A Biotechnology, Poland) following the manufacturer's protocol. The prepared samples were sequenced at Macrogen (Seoul, South Korea, http://dna.macrogen.com/eng/). The obtained sequence chromatograms were examined/edited using FinchTV (https://finchtv.software.informer.com/1.4/). The analyzed DNA regions (nrITS, *Xdh*, *mat*K, and *trn*L–*trn*F) were aligned separately using the 'align' option according to the MUSCLE algorithm (Edgar, 2004) with SeaView v.5.0 (Gouy et al., 2021).

2.3. Phylogenetic analyses

To test for possible incongruence in topologies, we performed phylogenetic analyses based on three methods: maximum parsimony (MP), maximum likelihood (ML), and Bayesian inference (BI). For the latter two, a nucleotide substitution model is required to be determined. It was calculated using the jModelTest 2 (Darriba et al., 2012), and based on the AIC criterion, we selected GTR+G+I for all datasets analyzed.

The MP analyses with PAUP v. 4.0 (Swofford, 2000) used a heuristic search strategy with tree-bisection-reconnection (TBR) branch swapping and the MULTREES option in effect, simple addition, and ACCTRAN optimization. The number of retained trees was 10,000. All characters were equally weighted (Fitch, 1971), while gaps were treated as missing values. In addition, we performed a bootstrap analysis with 500 replicates (Felsenstein, 1985). Whereas the maximum likelihood analyses were performed with RAxML-GUI 2.0 (Edler et al., 2021) by searching for the best-scoring ML tree, the branch support was calculated with 1000 replicates. We also carried out the Bayesian inference with MrBayes (Ronquist et al., 2012) using four Markov-chain Monte Carlo chains in two independent runs. Additionally, every run was started from different random trees to ensure that individual runs converged to the same result. For each dataset, different numbers of generations with sampling every 100 generations are inflicted. It depended on the achievement of convergence of split frequencies below 0.01. The initial 25% of the sampled generations of each chain were discarded as burn-in, and then all saved trees were summarized in a majority rule consensus tree.

3. Results

3.1. Statistical data showing the marker's usefulness

A matrix consisting of 53 sequences was obtained for the *Xdh* marker. Two species represented the outgroup, and the remaining taxa were representatives of *Dendrobium*. In the case of ITS, the dataset was obtained for 150 samples representing a nominal section of *Dendrobium* and two as an outgroup. At the same time, the matrices of the plastid regions (*mat*K and *trn*L-*trn*F, respectively) included 38 and 83 taxa of the *Dendrobium* sect. *Dendrobium* and 2 outgroup samples. For all the analyzed DNA fragments, the number of constant and variable features, as well as parsimony-informative and non-informative sites, were determined (Figure 1, Figure 2, Table 1). Based on these data, the variability within a given matrix was calculated as the ratio of the number of parsimony-informative sites to the number of all characters for each analyzed marker. The obtained results were plotted in a column diagram (Figure 3). In addition, in Table 2, we present the tree length, number of samples, value of consistency (CI), retention (RI), and homoplasy index (HI) for all analyzed matrices. We evaluated the efficacy of individual markers based on all the data presented above and their interpretation of them as a whole rather than only based on the analysis of a single piece of information. However, it is worth mentioning that the number of variable sites and parsimoniously informative sites seem to be most important here. The reliability of the obtained trees was determined based on calculations regarding the support of node, using bootstrap values for clades

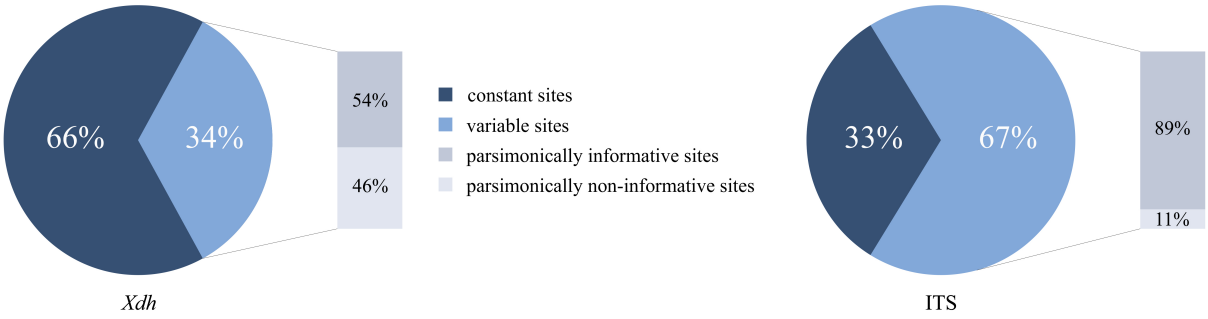

Figure 1 Chart showing the percentage of the number of constant and variable characters for *Xdh* and ITS markers.

Figure 2 Chart showing the percentage of the number of constant and variable characters for *trn*L-*trn*F and *mat*K markers.

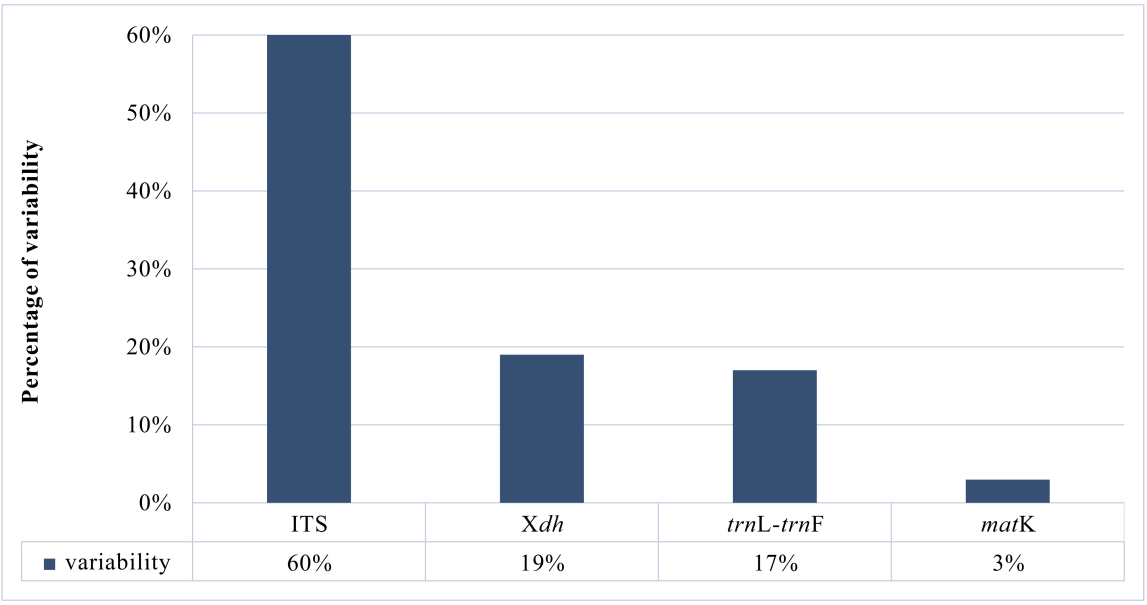

Figure 3 Graph showing the percentage of variation for each region.

Table 1 Summary data for all regions used in Figure 1 and Figure 2.

Region	Constant sites	Variable sites	Parsimony informative sites	Parsimony non-informative sites
Xdh	497 (66%)	259 (34%)	141 (54%)	118 (46%)
ITS	223 (33%)	461 (67%)	410 (89%)	51 (11%)
*trn*L-*trn*F	735 (69%)	336 (31%)	183 (54%)	153 (46%)
*mat*K	1149 (91%)	118 (9%)	44 (37%)	74 (63%)

Table 2 Statistical data for all regions used in the phylogenetic analyses (CI – Consistency Index; RI – Retention Index; HI – Homoplasy Index).

Region	Tree length	Number of Samples	CI	RI	HI
Xdh	379	53	0.794	0.876	0.206
ITS	1841	152	0.441	0.853	0.559
*trn*L-*trn*F	577	85	0.730	0.790	0.270
*mat*K	138	40	0.877	0.874	0.123

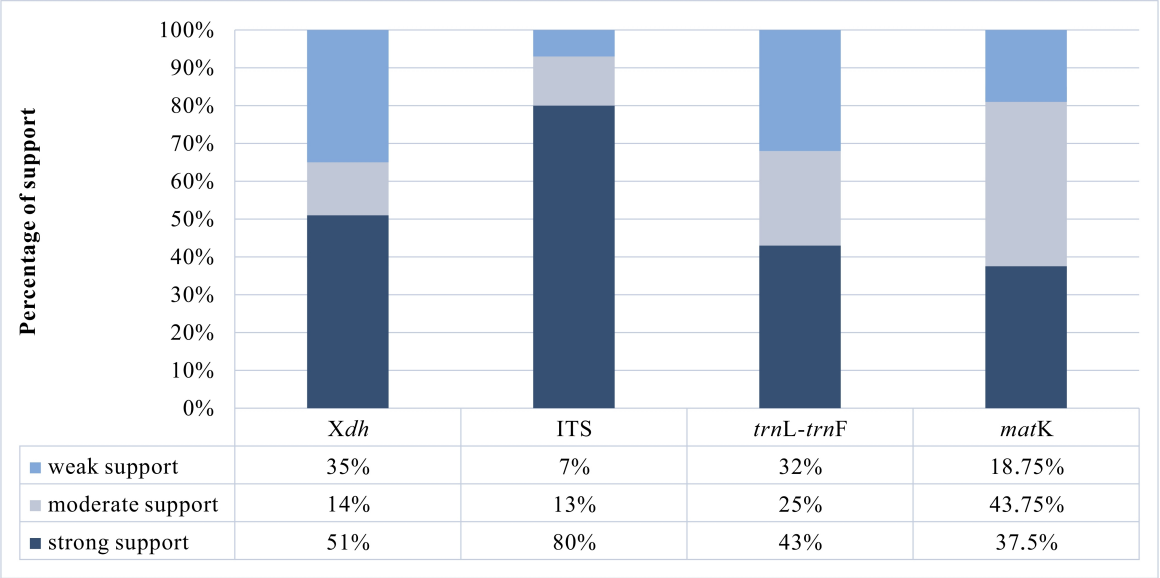

Figure 4 The reliability of ML trees for 4 markers (*Xdh*, ITS, *trn*L-*trn*F, *mat*K) was calculated using bootstrap support.

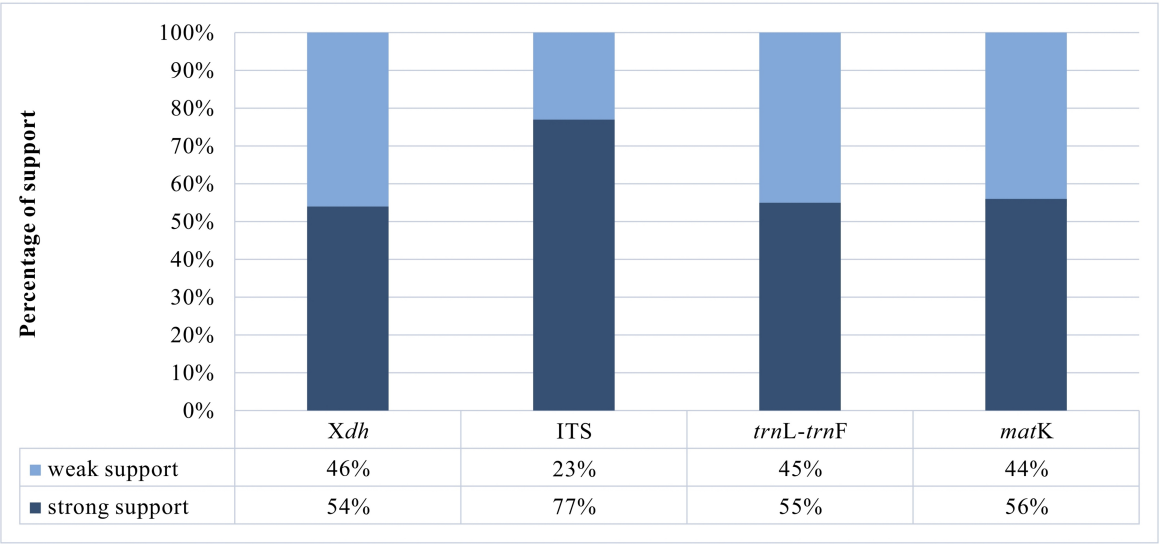

Figure 5 The reliability of BI trees for 4 markers (*Xdh*, ITS, *trn*L-*trn*F, *mat*K) was calculated using posterior probability.

generated by the ML method and posterior probability for BI analysis. As suggested by Kores et al. (2001), the level of bootstrap support above 85% is considered strong, between 85% and 71% as moderate, and below 70% as weak. In the case of a posterior probability, all values equal to or greater than 0.95 were considered strong, while the others were treated as weak (Cummings et al., 2003; Simmons et al., 2004). Our calculations for individual markers are summarized in two graphs – Figure 4 and Figure 5.

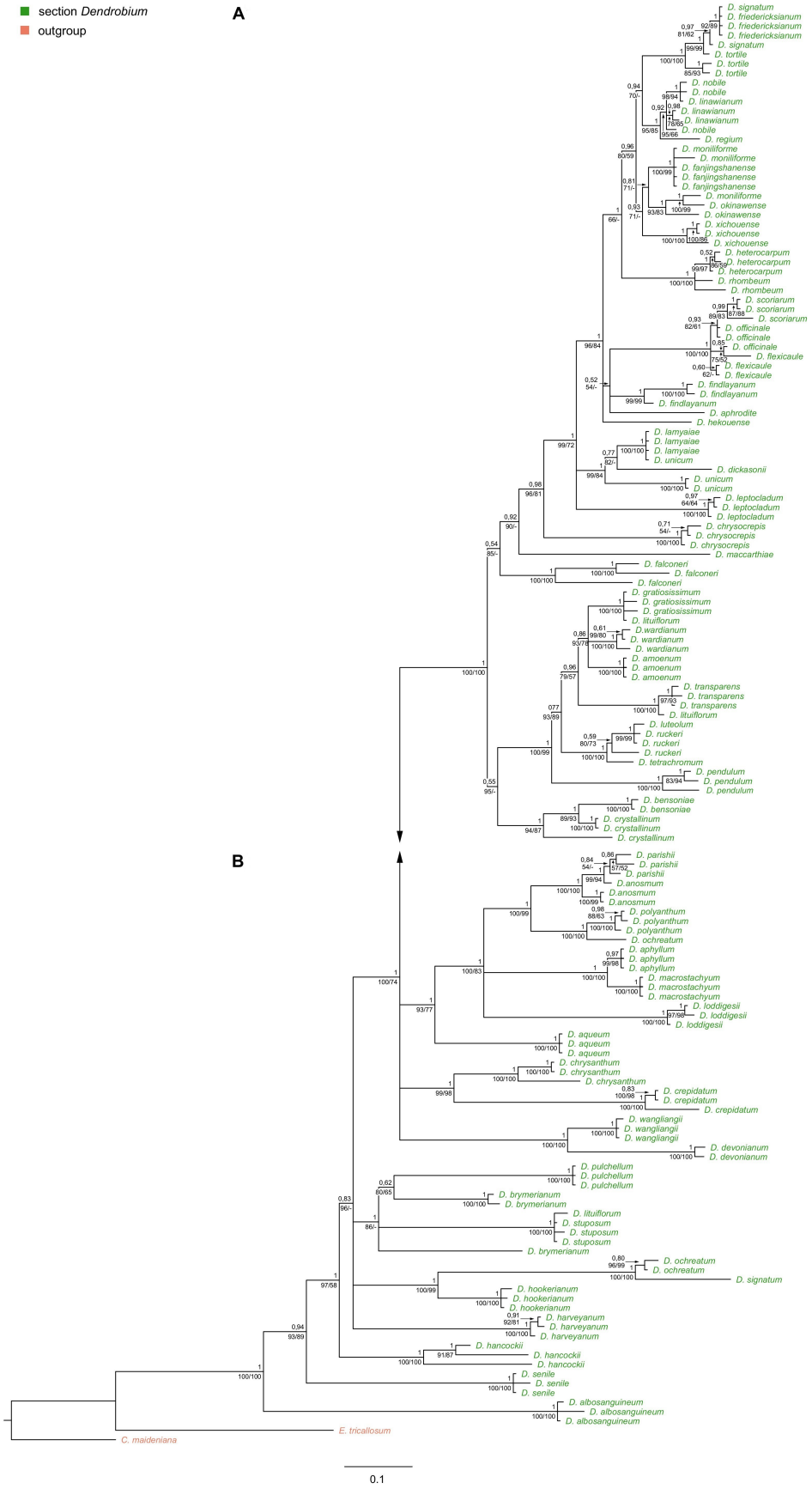

Figure 6 (**A**, **B**) The majority-rule consensus tree resulted in the Bayesian analysis for the nrITS marker of the *Dendrobium* sect. *Dendrobium*. The numbers above branches indicate posterior probability (PP) while the numbers below branches mean values of bootstrap support accordingly of maximum likelihood and maximum parsimony analyses. The BS values of less than 50% were marked as -.

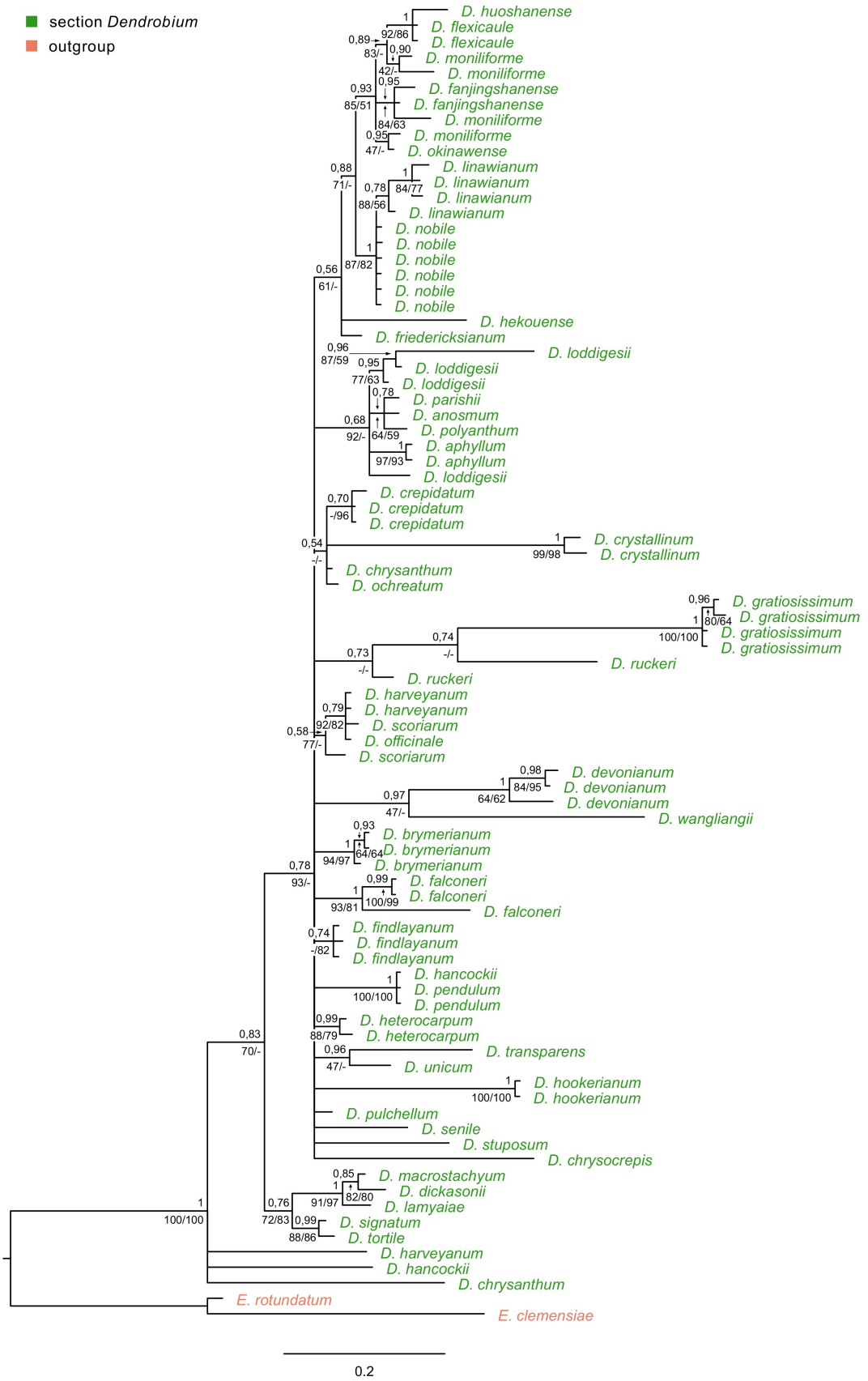

Figure 7 The majority-rule consensus tree resulted in the Bayesian analysis for the *trn*L-*trn*F marker of the *Dendrobium* sect. *Dendrobium*. The numbers above branches indicate posterior probability (PP) while the numbers below branches mean values of bootstrap support accordingly of maximum likelihood and maximum parsimony analyses. The BS values of less than 50% were marked as -.

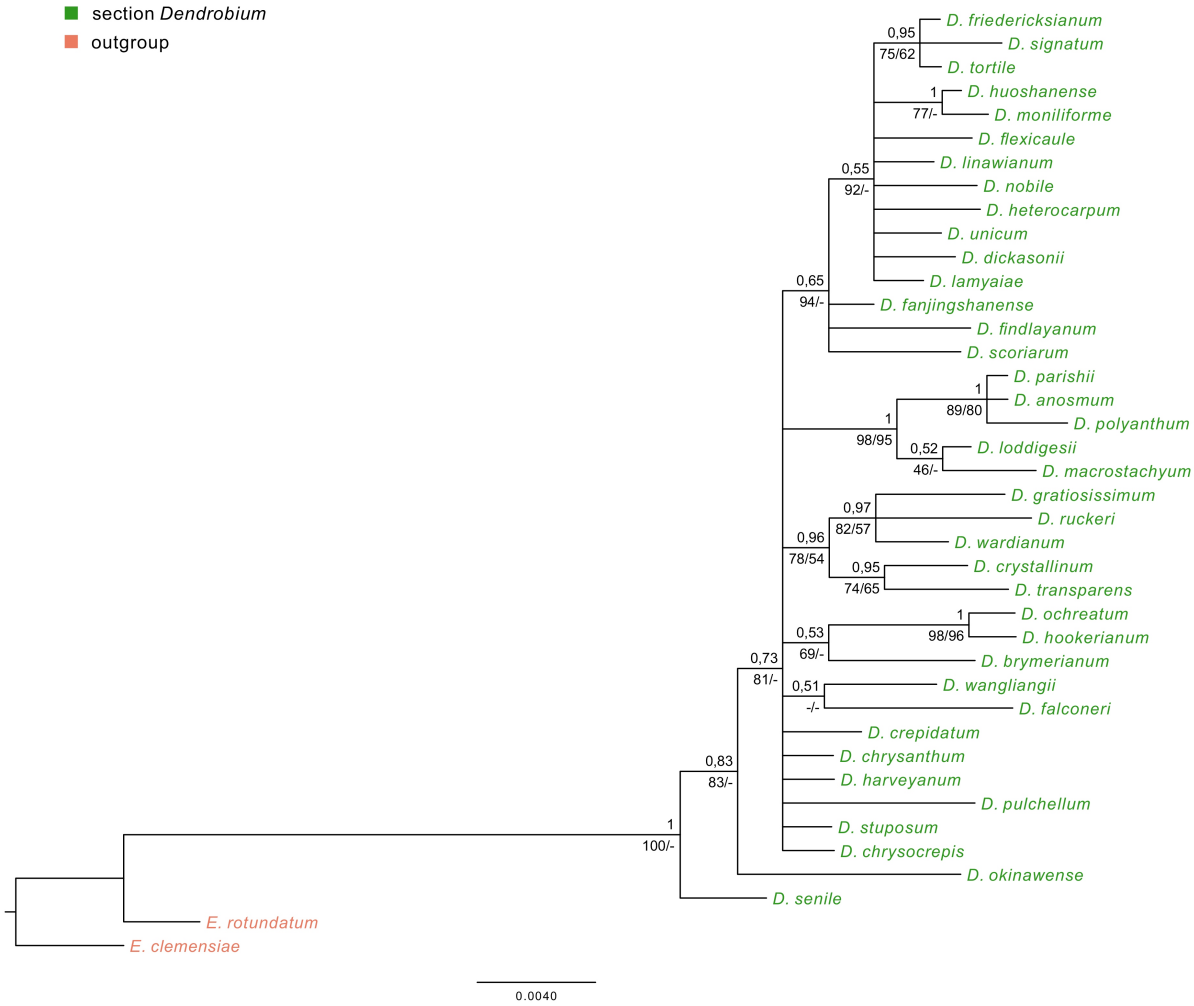

Figure 8 The majority-rule consensus tree resulted in the Bayesian analysis for the *mat*K marker of the *Dendrobium* sect. *Dendrobium*. The numbers above branches indicate posterior probability (PP) while the numbers below branches mean values of bootstrap support accordingly of maximum likelihood and maximum parsimony analyses. The BS values of less than 50% were marked as -.

3.2. Phylogenetic analyses

Regardless of the method used, for ITS, *trn*L-*trn*F, and *mat*K markers, tree topology turned out to be similar. Therefore, in this paper, we present only those obtained using the BI method, but we also add the nodes' values of bootstrap support (BS) obtained from both MP and ML analyses (Figure 6A–B, Figure 7, Figure 8). For the trees obtained based on the *Xdh* gene matrix, we decided to present the results from both ML and BI analysis (Figure 9). Admittedly, the differences between the two are not very glaring. However, we tested the usefulness of this marker and decided that two trees should be displayed in this case.

Phylogenetic relationships within the nominal section remain unresolved regardless of the method used (ML or BI). On both trees, species from this section form a strongly supported monophyletic clade (Figure 9A–B, PP=1, BS=96) with *Epigeneium suberectum* (Ridl.) Summerh. as a sister taxon. Clades, including representatives of each section, diverged as polytomic branches from a common ancestor. We obtained the maximum support of posterior probability (PP=1) and bootstrap (BS=100) at this node. Therefore, it can be inferred that they are closely related, but it is not possible to speculate on the relationship between them.

4. Discussion

Although *Dendrobium* is a widely studied genus, phylogenetic relationships, especially at lower taxonomic levels (e.g., section level), are often unclear and difficult to define. This is partly due to the high degree of morphological variation and partly to the selection of unjustified markers for phylogenetic reconstruction. In our research, we performed statistical analyses to demonstrate the utility of the novel low-copy marker *Xdh*.

Based on our phylogenetic analyses using the maximum parsimony, maximum likelihood, and Bayesian inference, this marker is believed to not perform well at all levels, at least for the genus *Dendrobium*. We obtained low support for both the value bootstrap and posterior probability at most nodes for clades embracing species from the same section (Figure 4, Figure 5). Moreover, we observed many polytomic branches.

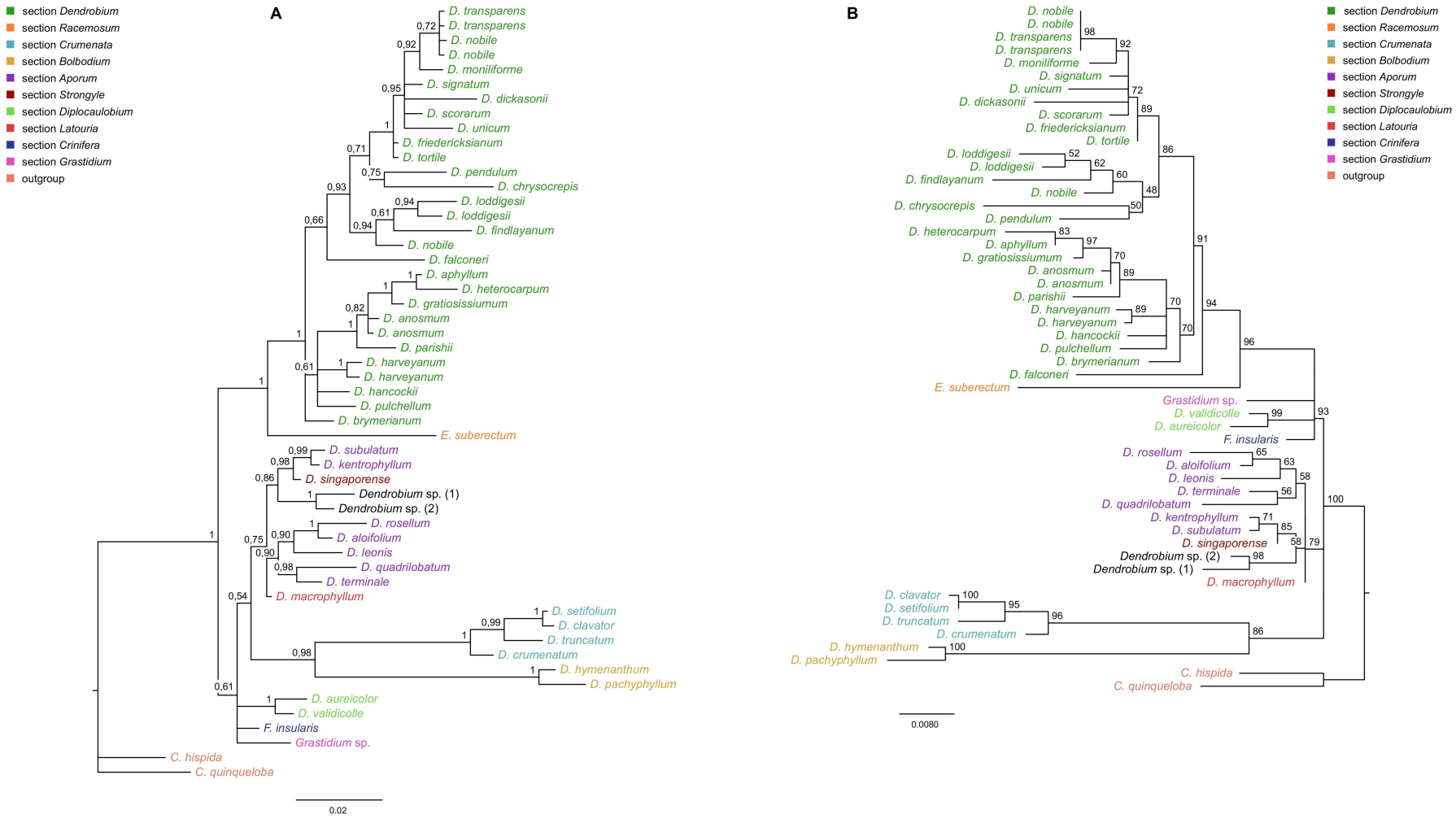

Figure 9 The results of phylogenetic analysis obtained for the *Dendrobium* species based on the *Xdh* marker. (**A**) The majority-consensus tree was obtained in the Bayesian analysis; the numbers above branches indicate posterior probability (PP). (**B**) The Maximum likelihood tree, the numbers above branches indicate bootstrap support (BS).

Therefore, we infer that the *Xdh* gene is unsuitable for solving phylogenetic relationships within closely related species (i.e., within a single section). Consequently, it also does not differentiate relationships within a nominal section (Figure 9A–B). In fact, all samples representing species of this section are grouped into a single coherent clade, with strong support for both posterior probability and bootstrap (PP=1, BS=96; Figure 9A). However, it is impossible to establish relationships between different species within this clade. Nevertheless, we want to emphasize that this does not mean it cannot be used in phylogenetic studies. Some authors have successfully used this marker, which means that at higher taxonomic levels, such as genus or intrafamily, it can give more reliable results (Górniak et al., 2010, 2014; Moudi & Go, 2015; Sang, 2002).

According to our analyses, another, more widespread nuclear marker, i.e., the ITS, performs better in this case. ITS is the most variable marker among the four analyzed regions (Figure 3). In addition, it is quite short and is easier to amplify, due to the significant number of copies in the genome. It is worth mentioning the work of Nguyen et al. (2020) at this point, in which this marker was used. Admittedly, in his study, it was used for species identification, as in the case of Duong et al. (2018) work, but among other markers such as *mat*K, or *rbc*L, it proved to be the most valuable. Likewise, Feng et al. (2015b) point to the ITS as a suitable barcode and indicate its potential for solving phylogenetic problems. As an effective tool for accurate identification and classification, the ITS regions were also recognized in the paper of Liu et al. (2019), which primarily analyzed species of two sections of *Dendrobium*: *Formosae* (Benth. & Hook. f.) Hook. f. and *Chrysotoxae* Kraenzl. A similar opinion was also expressed by Xiang et al. (2013), whose article additionally presents statistical analyses of the genes used. Not only has this marker enjoyed success recently, but earlier works, e.g., of Lau et al. (2001) or Tsai et al. (2004), have indicated that this marker is suitable and widely used in phylogenetic studies. The ITS region has been used not only to determine relations within *Dendrobium* but also for other taxa belonging to the Orchidaceae, e.g., *Bifrenaria* Lindl., *Cymbidium* Sw., *Dactylorhiza* Neck. (Burke et al., 2008). A direct comparison of the ITS and *Xdh* markers shows that the ITS is three times more variable than the other and contains more parsimony-informative sites (Figure 1, Figure 3). In this case, greater variability enables more reliable solutions to taxonomic problems also at lower levels, which is extremely important in the case of *Dendrobium*. Better-supported clades also occur on trees generated using the ITS rather than *Xdh*. This is clearly shown in the graph, where we present the percentage of well-supported tree clades for the analyzed markers (Figure 4, Figure 5).

5. Conclusions

It should be emphasized that both nuclear markers turned out to be more valuable than plastid ones to solve phylogenetic relationships on various taxonomic levels. In our analyses, neither matK nor trnL-trnF showed a satisfactory effect. The information obtained proves that the nuclear multi-copy marker is more valuable for studying the genus *Dendrobium* than the low-copy gene and plastid regions.

6. Supplementary material

The following supplementary material is available for this article:

Table S1. List of species of *Dendrobium* used in the molecular study for markers *Xdh*, ITS, *trn*L-*trn*F including Accession Number and GenBank Accession Number.

Table S2. List of species of *Dendrobium* used in the molecular study for markers ITS, *trn*L-*trn*F, *mat*K including GenBank Accession Number.

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Supplementary Materials

Table S1 List of species of *Dendrobium* used in the molecular study for markers *Xdh*, ITS, *trn*L-*trn*F including Accession Number and GenBank Accession Number.

Marker	Section	Species	Accession Number	GenBank Accession Number
Xdh	*Dendrobium*	*Dendrobium anosmum* Lindl.	UGDA.0073197	OR354767
		Dendrobium anosmum Lindl.	UGDA.0073096	OR354778
		Dendrobium aphyllum (Roxb.) C.E.C.Fisch.	UGDA.0076179	OR354757
		Dendrobium brymerianum Rchb.f.	UGDA.0073209	OR354775
		Dendrobium chrysocrepis C.S.P.Parish & Rchb.f. ex Hook.f.	UGDA.0073107	OR354761
		Dendrobium dickasonii L.O.Williams	UGDA.0073128	OR354776
		Dendrobium falconeri Hook.	UGDA.0076186	OR354754
		Dendrobium findlayanum C.S.P.Parish & Rchb.f.	UGDA.0073245	OR354768
		Dendrobium friedericksianum Rchb.f.	UGDA.0073240	OR354759
		Dendrobium gratiosissimum Rchb.f.	UGDA.0000247	OR354763
		Dendrobium hancockii Rolfe	UGDA.0073112	OR354772
		Dendrobium harveyanum Rchb.f.	UGDA.0073248	OR354770
		Dendrobium harveyanum Rchb.f.	UGDA.0073072	OR354758
		Dendrobium heterocarpum Wall. ex Lindl.	UGDA.0000243	OR354765
		Dendrobium loddigesii Rolfe	UGDA.0000246	OR354769
		Dendrobium loddigesii Rolfe	UGDA.0076178	OR354774
		Dendrobium moniliforme (L.) Sw.	UGDA.0073277	OR354764
		Dendrobium nobile Lindl.	UGDA.0076154	OR354751
		Dendrobium nobile Lindl.	UGDA.0073290	OR354752
		Dendrobium nobile Lindl.	UGDA.0073077	OR354779
		Dendrobium parishii H.Low	UGDA.0076175	OR354755
		Dendrobium pendulum Roxb.	UGDA.0073080	OR354773
		Dendrobium pulchellum Roxb. ex Lindl.	UGDA.0076156	OR354753
		Dendrobium scoriarum W.W.Sm.	UGDA.0073299	OR354760
		Dendrobium signatum Rchb.f.	UGDA.0076147	OR354762
		Dendrobium tortile Lindl.	UGDA.0000245	OR354756
		Dendrobium transparens Wall. ex Lindl.	UGDA.0073320	OR354777
		Dendrobium transparens Wall. ex Lindl.	UGDA.0076135	OR354766
		Dendrobium unicum Seidenf.	UGDA.0000244	OR354771
	Aporum	*Dendrobium aloifolium* (Blume) Rchb.f.		KC709960.1
		Dendrobium kentrophyllum Hook.f.		KC709972.1
		Dendrobium leonis (Lindl.) Rchb.f.		KC709959.1
		Dendrobium quadrilobatum Carr.		KC709963.1
		Dendrobium rosellum Ridl.		KC709961.1
		Dendrobium subulatum (Blume) Lindl.		KC709974.1
		Dendrobium terminale C.S.P.Parish & Rchb.f.		KC709962.1
	Bolbodium	*Dendrobium hymenanthum* Rchb.f.		KC709965.1
		Dendrobium pachyphyllum (Kuntze) Bakh.f.		KC709964.1
	Cadetia	*Cadetia hispida* (A. Rich.) Schltr.		GU004442.1
		Cadetia quinqueloba Schltr.		GU004443.1
	Crinifera	*Flickingeria insularis* Seidenf.		GU004446.1
	Crumenata	*Dendrobium clavator* Ridl.		KC709970.1
		Dendrobium crumenatum Sw.		KC701378.1
		Dendrobium setifolium Ridl.		KC709971.1
		Dendrobium truncatum Lindl.		KC709969.1
	Diplocaulobium	*Diplocaulobium aureicolor* (J.J. Sm.) A.D. Hawkes		GU004447.1
		Diplocaulobium validicolle (J.J. Sm.) Kraenzl.		GU004445.1
	Grastidium	*Grastidium* sp.		GU004444.1
	Latouria	*Dendrobium macrophyllum* A.Rich.		GU004440.1
	Racemosum	*Epigeneium suberectum* (Ridl.) Summerh.		GU004441.1
	Strongyle	*Dendrobium singaporense* A.D.Hawkes & A.H.Heller		KC709973.1
		Dendrobium sp. (1)		KC709968.1
		Dendrobium sp. (2)		KC709967.1
ITS	*Dendrobium*	*Dendrobium hekouense* Z.J.Liu & L.J.Chen	UGDA.0076247	OP783891
***trn*L-*trn*F**	*Dendrobium*	*Dendrobium anosmum* Lindl. Kraków	UGDA.0073096	OP829221
		Dendrobium chrysocrepis C.S.P.Parish & Rchb.f. ex Hook.f.	UGDA.0073107	OP829219
		Dendrobium dickasonii L.O.Williams	UGDA.0073128	OP829228
		Dendrobium friedericksianum Rchb.f.	UGDA.0073240	OP829226
		Dendrobium hekouense Z.J.Liu & L.J.Chen	UGDA.0076247	OP829223
		Dendrobium lamyaiae Seidenf.	UGDA.0076142	OP829229
		Dendrobium macrostachyum Lindl.	UGDA.0076065	OP829227
		Dendrobium ochreatum Lindl.	UGDA.0076235	OP829224
		Dendrobium polyanthum Wall. ex Lindl.	UGDA.0073073	OP829225
		Dendrobium signatum Rchb.f.	UGDA.0076147	OP829222
		Dendrobium tortile Lindl.	UGDA.0000245	OP829230

Table S2 List of species of *Dendrobium* used in the molecular study for markers ITS, *trn*L-*trn*F, *mat*K including GenBank Accession Number.

Marker	Section	Species	GenBank Accession Number
	Dendrobium	*Dendrobium albosanguineum* Lindl. & Paxton	AB593491.1
		Dendrobium albosanguineum Lindl. & Paxton	EU477498.1
		Dendrobium albosanguineum Lindl. & Paxton	AB972342.1
		Dendrobium amoenum Wall. ex Lindl.	KX600515.5
		Dendrobium amoenum Wall. ex Lindl.	HM054546.1
		Dendrobium amoenum Wall. ex Lindl.	HM054545.1
		Dendrobium anosmum Lindl.	EU477499.1
		Dendrobium anosmum Lindl.	JN388570.1
		Dendrobium anosmum Lindl.	KJ672650.1
		Dendrobium aphrodite Rchb.f.	AB593501.1
		Dendrobium aphyllum (Roxb.) C.E.C.Fisch.	JN388571.1
		Dendrobium aphyllum (Roxb.) C.E.C.Fisch.	EU840691.1
		Dendrobium aphyllum (Roxb.) C.E.C.Fisch.	FJ428219.1
		Dendrobium aqueum Lindl.	JF713085.1
		Dendrobium aqueum Lindl.	HM054571.1
		Dendrobium aqueum Lindl.	JN388584.1
		Dendrobium bensoniae Rchb.f.	HM054580.1
		Dendrobium bensoniae Rchb.f.	HM054579.1
		Dendrobium brymerianum Rchb.f.	JN388581.1
		Dendrobium brymerianum Rchb.f.	AF362036.1
		Dendrobium brymerianum Rchb.f.	FJ428221.1
		Dendrobium chrysanthum Wall. ex Lindl.	JN388584.1
		Dendrobium chrysanthum Wall. ex Lindl.	FJ384738.1
		Dendrobium chrysanthum Wall. ex Lindl.	EU003119.1
		Dendrobium chrysocrepis C.S.P.Parish & Rchb.f. ex Hook.f.	AB593531.1
		Dendrobium chrysocrepis C.S.P.Parish & Rchb.f. ex Hook.f.	KY966524.1
		Dendrobium chrysocrepis C.S.P.Parish & Rchb.f. ex Hook.f.	KY966525.1
		Dendrobium crepidatum Lindl. & Paxton	AY842035.1
		Dendrobium crepidatum Lindl. & Paxton	JN388586.1
		Dendrobium crepidatum Lindl. & Paxton	AF355574.2
		Dendrobium crystallinum Rchb.f.	GU339116.1
		Dendrobium crystallinum Rchb.f.	KY499225.1
		Dendrobium crystallinum Rchb.f.	AF363023.1
		Dendrobium devonianum Paxton	EU477502.1
		Dendrobium devonianum Paxton	FJ384735.1
		Dendrobium dickasonii L.O.Williams	KY966534.1
		Dendrobium falconeri Hook.	AF420246.1
		Dendrobium falconeri Hook.	AF521610.1
		Dendrobium falconeri Hook.	FJ384734.1
		Dendrobium fanjingshanense Z.H.Tsi ex X.H.Jin & Y.W.Zhang,	KJ210449.1
		Dendrobium fanjingshanense Z.H.Tsi ex X.H.Jin & Y.W.Zhang,	KJ210448.1
		Dendrobium fanjingshanense Z.H.Tsi ex X.H.Jin & Y.W.Zhang,	KF143460.1
		Dendrobium findlayanum C.S.P.Parish & Rchb.f.	JN388589.1
		Dendrobium findlayanum C.S.P.Parish & Rchb.f.	KF143462.1
		Dendrobium findlayanum C.S.P.Parish & Rchb.f.	KJ672620.1
		Dendrobium flexicaule Z.H.Tsi, S.C.Sun & L.G.Xu	AF355570.2
		Dendrobium flexicaule Z.H.Tsi, S.C.Sun & L.G.Xu	FJ384743.1
		Dendrobium flexicaule Z.H.Tsi, S.C.Sun & L.G.Xu	KT779770.1
		Dendrobium friedericksianum Rchb.f.	KY966545.1
		Dendrobium friedericksianum Rchb.f.	AB972331.1
		Dendrobium friedericksianum Rchb.f.	AB593565.1
		Dendrobium gratiosissimum Rchb.f.	JN388590.1
		Dendrobium gratiosissimum Rchb.f.	FJ384737.1
		Dendrobium gratiosissimum Rchb.f.	AF311780.1
		Dendrobium hancockii Rolfe	JN388591.1
		Dendrobium hancockii Rolfe	EU003120.1
		Dendrobium hancockii Rolfe	FJ384726.1
		Dendrobium harveyanum Rchb.f.	JN388594.1
		Dendrobium harveyanum Rchb.f.	KC568299.1
		Dendrobium harveyanum Rchb.f.	KJ210452.1
		Dendrobium heterocarpum Wall. ex Lindl.	JN388593.1
		Dendrobium heterocarpum Wall. ex Lindl.	JN388592.1
		Dendrobium heterocarpum Wall. ex Lindl.	GU339101.1
		Dendrobium hookerianum Lindl.	KJ210458.1
		Dendrobium hookerianum Lindl.	KF143475.1
		Dendrobium hookerianum Lindl.	KF143474.1
		Dendrobium lamyaiae Seidenf.	KY966555.1
		Dendrobium lamyaiae Seidenf.	KY966554.1
		Dendrobium lamyaiae Seidenf.	AB593595.1
		Dendrobium leptocladum Hayata	AF521612.1
		Dendrobium leptocladum Hayata	KJ672681.1
		Dendrobium leptocladum Hayata	EU840697.1
		Dendrobium linawianum Rchb.f.	JN388573.1
		Dendrobium linawianum Rchb.f.	EU003117.1
		Dendrobium linawianum Rchb.f.	EU003115.1
ITS		*Dendrobium lituiflorum* Lindl.	HQ114258.1
		Dendrobium lituiflorum Lindl.	AF355571.1
		Dendrobium lituiflorum Lindl.	KX792015.1
		Dendrobium loddigesii Rolfe	EU121418.1
		Dendrobium loddigesii Rolfe	AF311778.1
		Dendrobium loddigesii Rolfe	JN388569.1
		Dendrobium luteolum Bateman	AB593607.1
		Dendrobium maccarthiae Thwaites	AB593608.1
		Dendrobium macrostachyum Lindl.	HM054699.1
		Dendrobium macrostachyum Lindl.	HM054698.1
		Dendrobium macrostachyum Lindl.	HM054697.1
		Dendrobium moniliforme (L.) Sw.	GU339111.1
		Dendrobium moniliforme (L.) Sw.	EU003114.1
		Dendrobium moniliforme (L.) Sw.	AF521615.1
		Dendrobium nobile Lindl.	JN388579.1
		Dendrobium nobile Lindl.	EF618732.1

		Dendrobium nobile Lindl.	EU003118.1
		Dendrobium ochreatum Lindl.	KX522637.1
		Dendrobium ochreatum Lindl.	JF713121.1
		Dendrobium ochreatum Lindl.	HM054720.1
		Dendrobium officinale Kimura & Migo	GU339109.1
		Dendrobium officinale Kimura & Migo	FJ384724.1
		Dendrobium officinale Kimura & Migo	AF311776.1
		Dendrobium okinawense Hatus. & Ida	KF143495.1
		Dendrobium okinawense Hatus. & Ida	AB593622.1
		Dendrobium parishii H.Low	EU121417.1
		Dendrobium parishii H.Low	KC568303.1
		Dendrobium parishii H.Low	KX522639.1
		Dendrobium pendulum Roxb.	JN388596.1
		Dendrobium pendulum Roxb.	GU339115.1
		Dendrobium pendulum Roxb.	AF362912.1
		Dendrobium polyanthum Wall. ex Lindl.	KY966576.1
		Dendrobium polyanthum Wall. ex Lindl.	KY966574.1
		Dendrobium polyanthum Wall. ex Lindl.	KY966575.1
		Dendrobium pulchellum Roxb. ex Lindl.	KJ210492.1
		Dendrobium pulchellum Roxb. ex Lindl.	KF143503.1
		Dendrobium pulchellum Roxb. ex Lindl.	KY966577.1
		Dendrobium regium Prain	AB593645.1
		Dendrobium rhombeum Lindl.	AB593647.1
		Dendrobium rhombeum Lindl.	AB593646.1
		Dendrobium ruckeri Lindl.	KF143504.1
		Dendrobium ruckeri Lindl.	KX600507.1
		Dendrobium ruckeri Lindl.	KF143505.1
		Dendrobium scoriarum W.W.Sm.	KF143508.1
		Dendrobium scoriarum W.W.Sm.	KT779774.1
		Dendrobium scoriarum W.W.Sm.	KC568298.1
		Dendrobium senile C.S.P.Parish & Rchb.f.	KF143509.1
		Dendrobium senile C.S.P.Parish & Rchb.f.	EU477509.1
		Dendrobium senile C.S.P.Parish & Rchb.f.	KY966582.1
		Dendrobium signatum Rchb.f.	MT004895.1
		Dendrobium signatum Rchb.f.	MT004896.1
		Dendrobium signatum Rchb.f.	MK522218.1
		Dendrobium stuposum Lindl.	JN388599.1
		Dendrobium stuposum Lindl.	GU339104.1
		Dendrobium stuposum Lindl.	KY499214.1
		Dendrobium tetrachromum Rchb.f.	AB847672.1
		Dendrobium tortile Lindl.	MK522211.1
		Dendrobium tortile Lindl.	EU477511.1
		Dendrobium tortile Lindl.	KY966585.1
		Dendrobium transparens Wall. ex Lindl.	KX600508.1
		Dendrobium transparens Wall. ex Lindl.	MK522220.1
		Dendrobium transparens Wall. ex Lindl.	KF143520.1
		Dendrobium unicum Seidenf.	KF143523.1
		Dendrobium unicum Seidenf.	MK522213.1
		Dendrobium unicum Seidenf.	AB972348.1
		Dendrobium wangliangii G.W.Hu, C.L.Long & X.H.Jin	KJ210507.1
		Dendrobium wangliangii G.W.Hu, C.L.Long & X.H.Jin	KJ210506.1
		Dendrobium wangliangii G.W.Hu, C.L.Long & X.H.Jin	KF143524.1
		Dendrobium wardianum R.Warner	JN388600.1
		Dendrobium wardianum R.Warner	KY499218.1
		Dendrobium wardianum R.Warner	AF420245.1
		Dendrobium xichouense S.J.Cheng & Z.Z.Tang	KJ210514.1
		Dendrobium xichouense S.J.Cheng & Z.Z.Tang	KF143527.1
		Dendrobium xichouense S.J.Cheng & Z.Z.Tang	KC568304.1
	Cadetia	*Cadetia maideniana* (Schltr.) Schltr.	AY239948.1
	Epigeneium	*Epigeneium tricallosum* (Ames & C.Schweinf.) J.J.Wood	JF706721.1
	Dendrobium	*Dendrobium aphyllum* (Roxb.) C.E.C.Fisch.	KF143536.1
		Dendrobium aphyllum (Roxb.) C.E.C.Fisch.	EF397910.1
		Dendrobium brymerianum Rchb.f.	KF143538.1
		Dendrobium brymerianum Rchb.f.	KP749355.1
		Dendrobium brymerianum Rchb.f.	EF397911.1
		Dendrobium chrysanthum Wall. ex Lindl.	KF143549.1
		Dendrobium chrysanthum Wall. ex Lindl.	EF397914.1
		Dendrobium crepidatum Lindl. & Paxton	EF397916.1
		Dendrobium crepidatum Lindl. & Paxton	KF143552.1
		Dendrobium crepidatum Lindl. & Paxton	KP749356.1
		Dendrobium crystallinum Rchb.f.	EF397917.1
		Dendrobium crystallinum Rchb.f.	KF143553.1
		Dendrobium devonianum Paxton	KP749354.1
		Dendrobium devonianum Paxton	EF397919.1
		Dendrobium devonianum Paxton	KF143559.1
		Dendrobium falconeri Hook.	KP749351.1
		Dendrobium falconeri Hook.	KF143564.1
		Dendrobium falconeri Hook.	EF397920.1
		Dendrobium fanjingshanense Z.H.Tsi ex X.H.Jin & Y.W.Zhang,	KF143566.1
		Dendrobium fanjingshanense Z.H.Tsi ex X.H.Jin & Y.W.Zhang,	KF143565.1
		Dendrobium findlayanum C.S.P.Parish & Rchb.f.	KC568312.1
		Dendrobium findlayanum C.S.P.Parish & Rchb.f.	KP749361.1
		Dendrobium findlayanum C.S.P.Parish & Rchb.f.	KF143568.1
		Dendrobium flexicaule Z.H.Tsi, S.C.Sun & L.G.Xu	KP749358.1
		Dendrobium flexicaule Z.H.Tsi, S.C.Sun & L.G.Xu	KC568313.1
		Dendrobium gratiosissimum Rchb.f.	KF143571.1
		Dendrobium gratiosissimum Rchb.f.	KF143570.1
		Dendrobium gratiosissimum Rchb.f.	KP793697.1
		Dendrobium gratiosissimum Rchb.f.	KP749367.1
		Dendrobium hancockii Rolfe	KF143573.1
		Dendrobium hancockii Rolfe	EF397923.1
		Dendrobium harveyanum Rchb.f.	KF143574.1
		Dendrobium harveyanum Rchb.f.	KC568316.1
		Dendrobium harveyanum Rchb.f.	KP749363.1
		Dendrobium heterocarpum Wall. ex Lindl.	EF397924.1
		Dendrobium heterocarpum Wall. ex Lindl.	KF143579.1
***trn* L-*trn* F**		*Dendrobium hookerianum* Lindl.	KF143581.1
		Dendrobium hookerianum Lindl.	KF143580.1

		Dendrobium huoshanense Z.Z.Tang & S.J.Cheng	KP749357.1
		Dendrobium linawianum Rchb.f.	KP749345.1
		Dendrobium linawianum Rchb.f.	KP749346.1
		Dendrobium linawianum Rchb.f.	KC568318.1
		Dendrobium linawianum Rchb.f.	KP749347.1
		Dendrobium loddigesii Rolfe	EF397927.1
		Dendrobium loddigesii Rolfe	KP749349.1
		Dendrobium loddigesii Rolfe	KP749348.1
		Dendrobium loddigesii Rolfe	KF143587.1
		Dendrobium moniliforme (L.) Sw.	KF143597.1
		Dendrobium moniliforme (L.) Sw.	KF143595.1
		Dendrobium moniliforme (L.) Sw.	KF143596.1
		Dendrobium moniliforme (L.) Sw.	EF397935.1
		Dendrobium nobile Lindl.	KP749340.1
		Dendrobium nobile Lindl.	KP749341.1
		Dendrobium nobile Lindl.	KF143600.1
		Dendrobium nobile Lindl.	KP749339.1
		Dendrobium nobile Lindl.	KF143599.1
		Dendrobium nobile Lindl.	EF397936.1
		Dendrobium officinale Kimura & Migo	EF397937.1
		Dendrobium okinawense Hatus. & Ida	KF143601.1
		Dendrobium parishii H.Low	KC568322.1
		Dendrobium pendulum Roxb.	KF143602.1
		Dendrobium pendulum Roxb.	KP749359.1
		Dendrobium pulchellum Roxb. ex Lindl.	KF143609.1
		Dendrobium ruckeri Lindl.	KF143611.1
		Dendrobium ruckeri Lindl.	KF143610.1
		Dendrobium scoriarum W.W.Sm.	KC568315.1
		Dendrobium scoriarum W.W.Sm.	KF143614.1
		Dendrobium senile C.S.P.Parish & Rchb.f.	KF143615.1
		Dendrobium stuposum Lindl.	KF143622.1
		Dendrobium transparens Wall. ex Lindl.	KF143626.1
		Dendrobium unicum Seidenf.	KF143629.1
		Dendrobium wangliangii G.W.Hu, C.L.Long & X.H.Jin	KF143630.1
	Epigeneium	*Epigeneium clemensiae* Gagnep.	KF143636.1
		Epigeneium rotundatum (Lindl.) Summerh.	KF143637.1
***mat*K**	*Dendrobium*	*Dendrobium anosmum* Lindl. Kraków	AB972311.1
		Dendrobium brymerianum Rchb.f.	MG490257.1
		Dendrobium chrysanthum Wall. ex Lindl.	AB972304.1
		Dendrobium chrysocrepis C.S.P.Parish & Rchb.f. ex Hook.f.	KY966815.1
		Dendrobium crepidatum Lindl. & Paxton	KY966817.1
		Dendrobium crystallinum Rchb.f.	MG490248.1
		Dendrobium dickasonii L.O.Williams	KY966824.1
		Dendrobium falconeri Hook.	MG490236.1
		Dendrobium fanjingshanense Z.H.Tsi ex X.H.Jin & Y.W.Zhang	KF143670.1
		Dendrobium findlayanum C.S.P.Parish & Rchb.f.	MG490262.1
		Dendrobium flexicaule Z.H.Tsi, S.C.Sun & L.G.Xu	FJ794061.1
		Dendrobium friedericksianum Rchb.f.	KY966835.1
		Dendrobium gratiosissimum Rchb.f.	AB847767.1
		Dendrobium harveyanum Rchb.f.	KY966839.1
		Dendrobium heterocarpum Wall. ex Lindl.	KY966840.1
		Dendrobium hookerianum Lindl.	AB847780.1
		Dendrobium huoshanense Z.Z.Tang & S.J.Cheng	LC086430.1
		Dendrobium lamyaiae Seidenf.	KY966845.1
		Dendrobium linawianum Rchb.f.	AB847798.1
		Dendrobium loddigesii Rolfe	KY966852.1
		Dendrobium macrostachyum Lindl.	AB847810.1
		Dendrobium moniliforme (L.) Sw.	AB519775.1
		Dendrobium nobile Lindl.	AB519772.1
		Dendrobium ochreatum Lindl.	AB847822.1
		Dendrobium okinawense Hatus. & Ida	AB847824.1
		Dendrobium parishii H.Low	AB972316.1
		Dendrobium polyanthum Wall. ex Lindl.	KY966866.1
		Dendrobium pulchellum Roxb. ex Lindl.	KY966867.1
		Dendrobium ruckeri Lindl.	AB847855.1
		Dendrobium scoriarum W.W.Sm.	AB847860.1
		Dendrobium senile C.S.P.Parish & Rchb.f.	KY966692.1
		Dendrobium signatum Rchb.f.	AB972302.1
		Dendrobium stuposum Lindl.	AB847871.1
		Dendrobium tortile Lindl.	KY966874.1
		Dendrobium transparens Wall. ex Lindl.	MW862195.1
		Dendrobium unicum Seidenf.	AB972320.1
		Dendrobium wangliangii G.W.Hu, C.L.Long & X.H.Jin	KF143733.1
		Dendrobium wardianum R.Warner	AB847889.1
	Epigeneium	*Epigeneium clemensia* e Gagnep.	KF143739.1
		Epigeneium rotundatum (Lindl.) Summerh.	KF143740.1

3. Manuskrypt przygotowany do publikacji

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Evolution of morphological traits of *Dendrobium sensu lato* (Orchidaceae) - an attempt to resolve phylogenetic relationships in nominal and morphologically convergent sections.

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ABSTRACT

Background: *Dendrobium* Sw. is a large genus of the family Orchidaceae, with more than 1,600 species. Most of them are epiphytes, growing sympodially. Species occur in regions of southern, eastern, and southeastern Asia, the Pacific Islands, Australia, New Guinea, and New Zealand. There are problems with the infrageneric classification of this group due to, among other things, the large number of taxa, the wide geographic distribution, and the morphological diversity. Therefore, the main goal of our research was to analyze the genus *Dendrobium* (mainly nominal section) using DNA sequences and to integrate the results with morphological data.
Results: Our results indicate that the nominal section is not monophyletic and is not entirely consistent with morphology. The taxonomy of this group and the entire genus is complex. *Dendrobium sensu lato* is a genus in which it is possible to observe different rates of evolution, the loss of certain characteristics, and the appearance of others in the migration process and under the influence of adaptation to pollinators.
Conclusion: Based on our research and ongoing analyses, we have discussed about species that should be included or excluded from the section *Dendrobium*. We believe it is reasonable to focus on the nominal section first, and in subsequent steps undertake research on species from other sections.

Keywords: Dendrobiinae, ITS, molecular analyses, morphology, plastid markers, taxonomy

Introduction

With about 1,600 species and a wide geographical distribution *Dendrobium* Sw. is one of the largest genera in the Orchidaceae [1, 2, 3, 4, 5, 6]. Species in this group are found in regions of southern, eastern, and southeastern Asia, the Pacific Islands, Australia, New Guinea, and New Zealand [7, 8]. Thus, this group represents one of the most complicated taxonomic challenges in the orchid family. In addition, there is a problem of correct identification due to the strong polymorphism observed among *Dendrobium* species [4, 9, 10, 11], lack of comprehensive taxonomic revisions and frequent description of species without verification of reference materials, which, *nota bene*, has been lost in many cases.

Species in this group are mainly epiphytes with a sympodial type of growth. However, lithophytes and terrestrial plants are quite frequent [7, 8]. Pseudobulbs are highly diversified. There are both green and succulent forms, as well as thin, long, and cane-like ones [8, 12], and any intermediate types. Leaves are conduplicate and arranged in two rows. They can be dorsiventrally flattened, bilaterally flattened, or terete as well. Flowers are generally small to medium-sized, occurring either

singly or more commonly in clusters with inflorescences placed apically or subapically on the pseudobulbs. Most often (but not exclusively), they come in colors such as white, purple, or yellow or a combination thereof. They vary in size, shape, persistence, and growth rate [9]. The lip may be 3-lobed, with a characteristic claw and striations or ridges present [8, 9]. In addition, there are other structures existed in some representatives, such as trichomes and papillae, which most probably are related to pollination [13, 14]. The gynostemium or column is cylindrical-conical. The column foot is typically very long and substantial [12, 15] Together with the lateral sepals and lip claw, they can be joined in various ways to form the spur-like mentum, a characteristic structure of this group [9]. The rostellum is swollen, and when touched, it releases an opaque liquid that attaches the pollen to the pollinator [9, 15]. Also present are 4 naked, laterally compressed pollinia, arranged within the anther in two pairs side by side [15]. The fruit is a dry, hairy capsule in which small seeds containing a spherical or ovoid embryo are usually present [8, 9, 16].

Unfortunately, few studies have been done on pollination, a phenomenon playing a key role in the evolution and continuity of the species. It is likely that some species, such as *Dendrobium unicum* Seidenf. from the nominal section, are pollinated by bees or wasps carrying pollen on their backs [9, 14]. Representatives of the sections *Dendrocoryne* Lindl. & Paxton, *Rhizobium* Lindl. & Paxton, and *Monophyllaea* Benth. mainly use pollinators from the genus *Trigona* Jurine, but also *Homalictus* Cockerell, *Lassioglossum* Curtis, or *Hylaeus* Fabricius [9, 17]. *Dendrobium setifolium* Ridl., which belongs to the section *Crumenata* Pfitzer, is also pollinated by the stingless bee of the genus *Trigona* [18]. *Amegilla* Friese probably can also pollinate species of this genus, mainly from the sections *Spatulata* Lindl. and *Calcarifera* J.J. Sm., while *Xylocopa* Latreille from the section *Latouria* Miq. [9]. Birds can also serve in pollination by species belonging to the sections *Pedilonum* Blume, *Oxyglossum* Schltr. and *Calyptrochilus* Schltr.. In this case, pollen adheres to their beak [9, 19]. It is known that most representatives of this genus do not produce nectar, so pollinators are often deceived. In addition to bees and birds, other animals can pollinate *Dendrobium*. *Bombus* Latreille probably pollinates species in the sections *Formosae* (Benth. & Hk.f.) Hk.f. and *Epigeneium* Gagnepain [9]. A study by Kjellsson and co-authors confirms these data. In this research, *Bombus eximus* Smith was observed pollinating *Dendrobium infundibulum* Lindl [20]. Self-pollination was also reported, but rarely, for example in *Dendrobium striolatum* Rchb.f (section *Rhizobium*). It is most often caused by a lack of suitable pollinators [9].

Background and goals

The history of *Dendrobium* is quite complicated and complex. In 1799, Swartz described this genus based on the presence of 5 spreading and upright petals with lateral sepals attached to the lip forming a "horn" [21]. The last characteristic was only mentioned, but it was not clearly defined. It was not until 1810 that Brown defined and named it as the mentum and a key feature of this genus. Later researchers also treated this character as taxonomically important [9]. It is worth noting that mentum occurs in several other groups of orchids, such as the Asian Vandeae and the Neotropical Maxillariinae, so it is not a feature unique only to *Dendrobium*. Since the description of the genus, different concepts have been proposed regarding its taxonomy and classification, which have been based on both morphological and molecular analyses.

Initially, Lindley (1830) placed *Dendrobium* with 19 other genera in Dendrobiinae G. Don. In *Dendrobium*, however, he distinguished four sections [22]. A few years later, the number of sections was increased to 10 [23]. This taxonomic concept focused primarily on floral characters, especially pollen, and less on vegetative traits. Whereas Schlechter divided *Dendrobium* into 4 subgenera and 41 sections based on the presence or absence of a sheath leaf base [24]. This division was considered completely artificial by Schuiteman [25]. However, some researchers, after making several modifications, adopted Schlechter’s system. For example, Brieger (1981) classified 28 genera in Dendrobiinae, most of which correspond to Schlechter's sections [4]. The other division was presented by Dressler (1993) and Wood (2006). They distinguished 6 and 5 genera, respectively, in the subtribe [9, 19]. At the end of the 20th century, most authors distinguished the following genera in Dendrobiinae: *Dendrobium*, *Diplocaulobium*, *Cadetia*, *Flickingeria*, *Epigeneium*, and *Pseuderia* [26].

During this period, the first results of molecular analyses of representatives of the Dendrobiinae have also been presented, primarily based on plastid markers sequences. Taxa of this subtribe were grouped on the three major clades. The first one included *Dendrobium* species found in continental Asia and western Malaysia (including the generitype *D*. *moniliforme* (L.) Sw.). The second one included *Dendrobium* along with the following genera: *Cadetia*, *Diplocaulobium*, and *Flickingeria*, found mainly in Australasia and the Pacific Islands. While the last clade included species of *Epigeneium* [2, 26, 27, 28, 29, 30]. Subsequent molecular studies based on additional nuclear markers correlated with the initial results. Tsai et al. presented the phylogeny of *Dendrobium* species from Taiwan [31]. In 2008, Burke et al. performed molecular analyses on the Australian section

Dendrocoryne [32]. However, as in the aforementioned study, molecular analyses do not always agree with species morphology, and often the sections analyzed are still not monophyletic. Also worth mentioning are the works of Clements and Jones (2002) and Clements (2003), in which *Dendrobium sensu l*ato was divided into 3 subtribes: Dendrobinae, Grastidiinae and Epigeneiinae with about 50 genera to obtain monophyly. Their results are based on analyses using the ITS marker [33, 34]. In 2011, Schuiteman and Adams showed a different concept for classifying the representatives of *Dendrobium sensu lato*. They proposed to include several genera: *Cadetia, Diplocaulobium, Epigeneium, Euphlebium, Flickingeria* and *Grastidium* in *Dendrobium sensu latissimo* [25]. In 2013, Xiang et al. conducted analyses of the genus by interpreting various molecular markers and based on extensive sampling. The results of the phylogenetic analyses indicate two major clades in the studied group. In these clades, smaller subclades are distinguished, with sections that often exhibit a polyphyletic and paraphyletic character [4].

The taxonomy of this genus is more complicated than previously thought [4, 9, 30]. In the present study, we have concentrated our attention on the nominal section of *Dendrobium*, which of course includes type species of the whole genus. This section includes species that show clear similarity in morphological structure to the generitype. The boundaries between sections are discretionary and depend on the author's conception. In our opinion, individual sections should show a kind of discontinuity in morphological structure. Therefore, it is important to define the scope of a nominal section, both in terms of morphological structure and its phylogenetic content.

Despite numerous attempts to establish a clear classification, there are still many uncertainties and the question of the taxonomy of the group remains unresolved. The aim of our paper is to analyze the genus *Dendrobium* using all available DNA sequences and to integrate the results with morphological data. From our perspective, floral traits are of paramount importance when conducting such studies. So, we focused primarily, though not exclusively, on them. The choice of appropriate markers is also very important. Based on the results of other scientists [5, 35, 36], the most reliable marker is nuclear ITS, therefore it is primarily used in this paper to discuss phylogenetic relationships.

Materials and methods

Materials. DNA sequences representing taxa of *Dendrobium sensu lato* and the outgroup were downloaded from the NCBI database (https://www.ncbi.nlm.nih.gov/genbank/). First, we used all the

sequences available in this database and performed preliminary analyses. Because of the need to minimize the resulting tree, we selected taxa that represented particular sections and then used them to generate the final trees. For some species, we obtained sequences from plant material presented in Burzacka-Hinz et al. (2022) [13]. The GenBank accession numbers of the sequences used are given in Additional file 1 (Table S1). The nominal section has been treated as *sensu lato*, with all the species that have ever been part of it.

Molecular analyses. We analyzed most of the markers available in GenBank (nuclear ITS and three plastid markers: *rbc*L, *mat*K and *trn*L-*trn*F). In addition, we performed an analysis for the low copy gene *Xdh*. First, the sequences for each marker were aligned using Mafft v. 7 [37]. The minor errors were corrected in SeaView v. 5.0 [38]. On the *trn*L-*trn*F matrix, we observed highly variable and ambiguously aligned characters. These were excluded from the analysis. In accordance with Givnish et al. (2015), a selection of *Bulbophyllum* species were used as an outgroup [39]. Substitution models were calculated using the following website http://www.atgc-montpellier.fr and based on the AIC criterion, GTR+G+I was selected for the ITS and plastid matrices, and HKY+G+I for the *Xdh* dataset. Then, to eliminate possible incompatibilities in the topologies, we performed the Bayesian inference (BI) for each individual dataset: nrITS (820 bp), *Xdh* (756 bp), *mat*K (1624 bp), *rbc*L (1229 bp), *trnL-trn*F (1065 bp). On the CIPRES Science Gateway [40], we used MrBayes 3.2.7a with Markov Chain Monte Carlo (MCMC). Two simultaneous runs of four chains for 3 000 000 generations, sampling one of every 100 trees, were performed until the average standard deviation of the split ranges reached a value below 0.01. The first 25% of the trees were deleted as a burn-in, and the rest of the saved trees were collated into a majority consensus tree.

The plastid marker matrices proved to be uninformative and the resulting trees were polytomous. These are available in a corresponding author. Despite the fact that the topology of the trees obtained for the nuclear datasets is similar, for further analyses we have chosen the tree of the ITS matrix. The main reason was the larger number of samples. In addition, the *Xdh* tree was presented in Burzacka-Hinz et al. (2024) [41].

For the ITS dataset, we also performed maximum likelihood analysis (ML) using raxmlGUI 2.0 [42]. 1000 bootstrap replicates were used. The topology of the two trees, the BI analysis and the ML analysis were similar. Therefore, we edited the majority consensus tree with FigTree v.1.4.4 (http://tree.bio.ed.ac.uk/software/figtree/) and InkScape, but the support on the nodes was presented

by both posterior probability (PP) and bootstrap (BS). We considered values of 0.95 or higher for PP and higher than 75 for BS to be strongly supported [43, 44].

Ancestral state reconstruction (ASR) of morphological features. The ancestral states of morphological characters for *Dendrobium* species were reconstructed for 14 morphological features, using Mesquite v. 3.70 and the parsimony ancestral state reconstruction method [45]. The phylogenetic tree, obtained from Bayesian analysis was used as an input for the analysis. Numbers above the branches indicate the PP/BS support values. Taxa characters were coded for the presence (1,2 – yes) or absence (0 – no) of a feature. All character states are presented in Table S2 (Additional file 2).

Analysis of morphological variation. To summarize the morphological variation in *Dendrobium sensu lato*, as well as between species belonging to 39 sections, we described 318 taxa using 14 well-distinguished characters (Table S3 and S4 in Additional file 3). The selected traits referred to apparent external differences for pseudobulbs, leaves and inflorescences (Ch1-9), as well as floral characters (Ch10-14). To examine morphological variation, the listed traits were binary coded <0,1>, and were often of opposite natures, with the exception of the trait describing leaf cross-section - Ch5, which was included in three states of this trait <0,1,2> (Table S3 in Additional file 3). Similarity between taxa were calculated using the Jaccard similarity coefficient. Based on this data set (Table S4 in Additional file 3), a cluster analysis was performed, and then a dendrogram was constructed using the UPGMA method. In addition, to describe the extent of morphological variation among *Dendrobium* sections and the interrelationships between them, we conducted a principal coordinate analysis (PCoA) and a non-metric multidimensional scaling analysis (NMDS), based on the Jaccard similarity matrix in both cases. To identify the morphological characters that contributed most to the observed differences between *Dendrobium* taxa, we carried out a SIMPER (SIMilarity PERcentage) analysis, which attempts to assess the average percent dissimilarity of each variable between specimens in the Bray-Curtis similarity matrix. The average percentage changes of each *Dendrobium* section in the differences in the Bray-Curtis similarity matrix are also summarized in Table S5 (Additional file 3).

Additional analyses but based on only 5 floral traits (Ch10-Ch14), were conducted in the same manner as described above. The difference was that the cluster analysis was performed using Ward's method, which allows clusters to be combined in such a way as to minimize the increase in within-

group variance. This method is very effective, although it aims to create clusters of small size, but represents the most essential clustering of individuals [46].

All multivariate analyses and tests were performed using software packages: STATISTICA v. 13 and PAST v. 4.14 [47, 48].

Results

Phylogenetic analysis. *Dendrobium sensu lato* is a monophyletic group that evolved from a common ancestor, according to the molecular analysis of the nuclear marker ITS (Fig. 1). Representatives of e.g. *Epigeneium*, *Diplocaulobium* or *Flickingeria* were placed in the basal branches of analyzed taxa with other species of *Dendrobium sensu stricto* and did not form with a separate clade. The most basal group on the ITS tree, clade **A1** (Fig. 1), includes species of the section *Oxystophyllum*, while the remaining *Dendrobium species* form clade **A2** (Fig. 1), but with only average bootstrap support (BS=82). Next, we observed divergence into two evolutionary lineages, **B1** (Fig. 1, PP=1, BS=98) and **B2** (Fig. 2, PP=0.92, BS=97). The first of these includes species that have been divided into 3 taxa: *Racemosum* and *Katherinea*, usually treated as sections of *Dendrobium*, and *Epigeneium*, which most often has the status of a separate genus. Clade **B2** also splits into two lineages, **C1** (Fig. 1, PP=0.99, BS=82) and **C2** (Fig. 2, PP=1, BS=100). The group **C1** contains species belonging to different sections. Within this group we can distinguish two subclades, **c1** (Fig. 1, PP=0.99, BS=82), which includes two species of section *Macrocladium*, *D*. *oppositifolium* and *D*. *muricatum*, and **c2** (Fig. 1, PP=1, BS=80). The subclade **c2** is significantly larger, and within it we did not get enough support at some nodes, so we observe six polytomous lineages: **c2.1**, **c2.2**, **c2.3**, **c2.4**, **c2.5**, **c2.6** (Figs 1-2). In addition, *D*. *spectabile* and *D*. *amboinense* were not placed with any of the above subclades. Therefore, we cannot discuss the relationship between the taxa in this subclade. However, they are certainly closely related because they evolved from a common ancestor (node **c2**).

Clade **C2** (Fig. 2) is also divided into two groups. The most basal group within **C2** includes representatives of the section *Fytchianthe*, clade **D1** (Fig. 2), with strong support on the node (PP=1, BS=100). In contrast, the second group, clade **D2** (Fig. 3), splits dichotomously into two lineages without sufficient node support. Therefore, we observed seven polytomous subclades: d1, d2, d3 (Fig. 2) and d4, d5, d6, d7 (Fig. 4). We also suggest that each of them may have evolved as an independent evolutionary lineage. Subclade **d1** (PP=1, BS=100) contains only two species, both

belonging to the section *Herbacea*, while **d2** (PP=1, BS=100) is divided into two further groups. First, **d2.1** (Fig. 2, PP=1, BS=100), includes members of the section *Distichophyllae*, and **d2.2** (Fig. 2, PP=1, BS=95) is represented by a larger number of taxa, all belonging to the section *Stachyobium*. Taxa representing the nominal section were grouped in the largest subclade in our analyses - **d3** (Figs 2-3, PP=1, BS=91). However, our results show that this is not a monophyletic group. This subclade also contains species from sections *Holochrysa* (10 species), *Breviflores* (8 species) and *Stuposa* (3 species). In addition, *D*. *braianense* (sect. *Holochrysa*) is grouped with *D*. *albosanguineum* and *D. capillipes* (sect. *Dendrobium*) in subclade **d6** (Fig. 4, PP=1, BS=100). However, *D*. *somae*, *D*. *luzonense* and *D*. *panduratum*, previously classified to the sections *Stachyobium* and *Grastidium*, were placed in subclade **d3** (Figs 2-3) with most species of the nominal section. In addition, *D*. *epidendropsis* (sect. *Calcarifera*), *D*. *khasianum, D. calocephalum* and *Fickingeria bicolor* are also placed in the subclade **d3**. On the other hand, *D*. *gibsonii*,*D*. *senile*, *D. capillipes* and *D. albosanguineum* representing the nominal section, were outside of subclade **d3**. Within this group, we also observe numerous polytomy. Therefore, it is difficult to speculate about the relationships between the individual species in this group. The other group, subclade **d4** (Fig. 4, PP=1, BS=100), is very small and contains only two species, both with the section *Densiflora*. In subclade **d5** (Fig. 4), with medium support (PP=0.96, BS=86), we can distinguish four polytomous lineages: **d5.1**, **d5.2**, **d5.3** and **d5.4** (Fig. 4), all strongly supported (PP=1, BS=100). Groups marked as **d5.1, d5.2** and **d5.4** include representatives of the *Formose* section, while **d5.3** include taxa of the sections *Conostalix* and *Distichophyllae*.

Subclade **d7** (Figs 4-5, PP=0.98, BS=89) is divided into two smaller groups, one of these, **d7.1** (Fig. 4, PP=1, BS=93), includes species representing the following sections: *Calcarifera*, *Platycaulon* and two species of *Pedilonum*. It is worth noting that some representatives of the *Calcarifera* were also placed in the **f2.1** and **f2.2** subclades (Fig. 5) together with species from the sections *Dolichocentrum* and *Pedilonum*. Furthermore, two other clades, **e1** (Fig. 4) and **e2** (Fig. 5), can be distinguished within **d7.2** (Fig. 5), but only with strong bootstrap support (BS=93). Clade **e1** (Fig. 4, PP=0.98, BS=89) was formed by species from the section *Densiflora*, with the exception of *D*. *jenkinsii* and *D*. *lindleyi*, which were also classified to this section, but created a separate clade **d4** (Fig. 4). Clade **e1** (Fig. 4) also includes *D*. *melanosticum*, *D*. *stocari* and *D*. *microglaphys*, representing the section *Amblyanthus*. While clade **e2** (Fig. 5, PP=1, BS=100) splits into two other groups marked as **f1** and **f2** (Fig. 5). Within

f1 (PP=1, BS=100) were placed representatives of the sections *Crumenata* and *Aporum*. The only exceptions are *D*. *pachyphyllum* and D. *hymenanthum*, both of which are part of the section *Bolbidium* and *D. confusum* from section *Pedilonum*. From node **f2** (PP=1, BS=100), probably three independent polytomic groups evolved. We marked them as **f2.1**, **f2.2,** and **f2.3** (Fig. 5). However, each of these included taxa previously classified in different sections, so the **f2.1** group supported only by posterior probability (PP=0.95) is *D*. *furcatum* (sect. *Dolichocentrum*) and *D*. *lancifolium* (sect. *Calcarifera*). Subclade **f2.2** (PP=1, BS=99) includes species from the sections *Calcarifera*, *Dolichocentrum*, and *Pedilonum*, and the latter, **f2.3** (PP=0.99), includes representatives of *Calyptrochilus* and other species from the sections *Pedilonum, Rhizobium* and *Oxyglossum*.

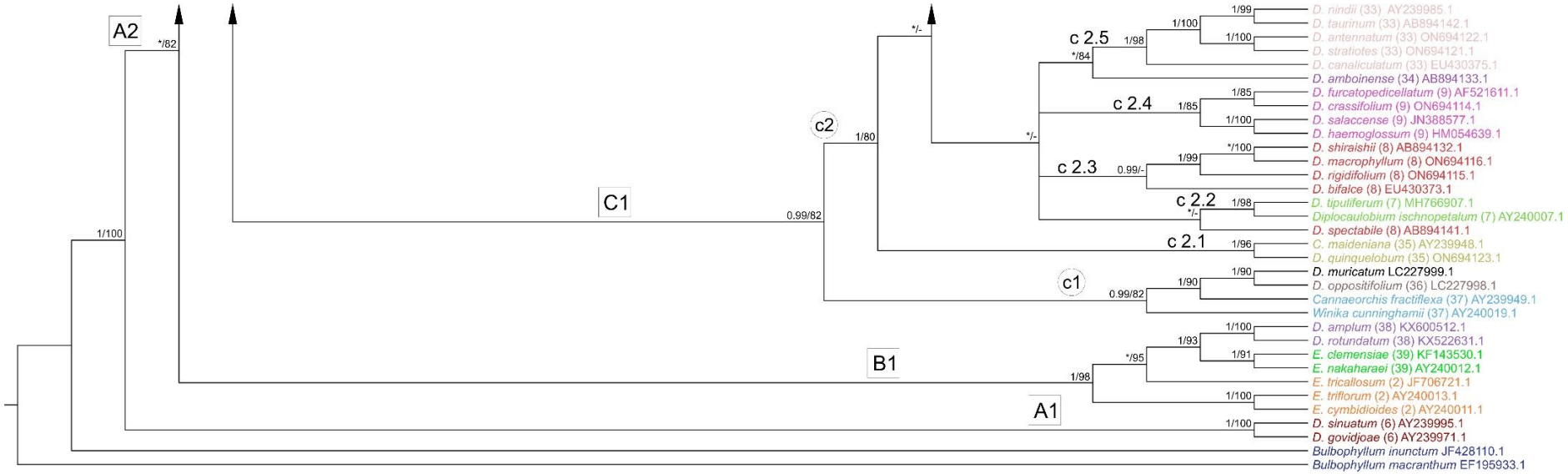

Fig. 1 The first part of the 50% majority rule consensus tree for *Dendrobium sensu lato*, obtained for the nrITS dataset using Bayesian inference. Numbers above branches indicate posterior probability and bootstrap support values from maximum likelihood analysis (PP/BS); PP values <0.95 are marked with an asterisk, and BS values <75% are marked with a -. Letters with numbers above branches mark clades discussed in the text. Numbers in parentheses next to taxon names indicate section affiliation. In addition, each section is highlighted in a different color and the species of nominal section are bolded.

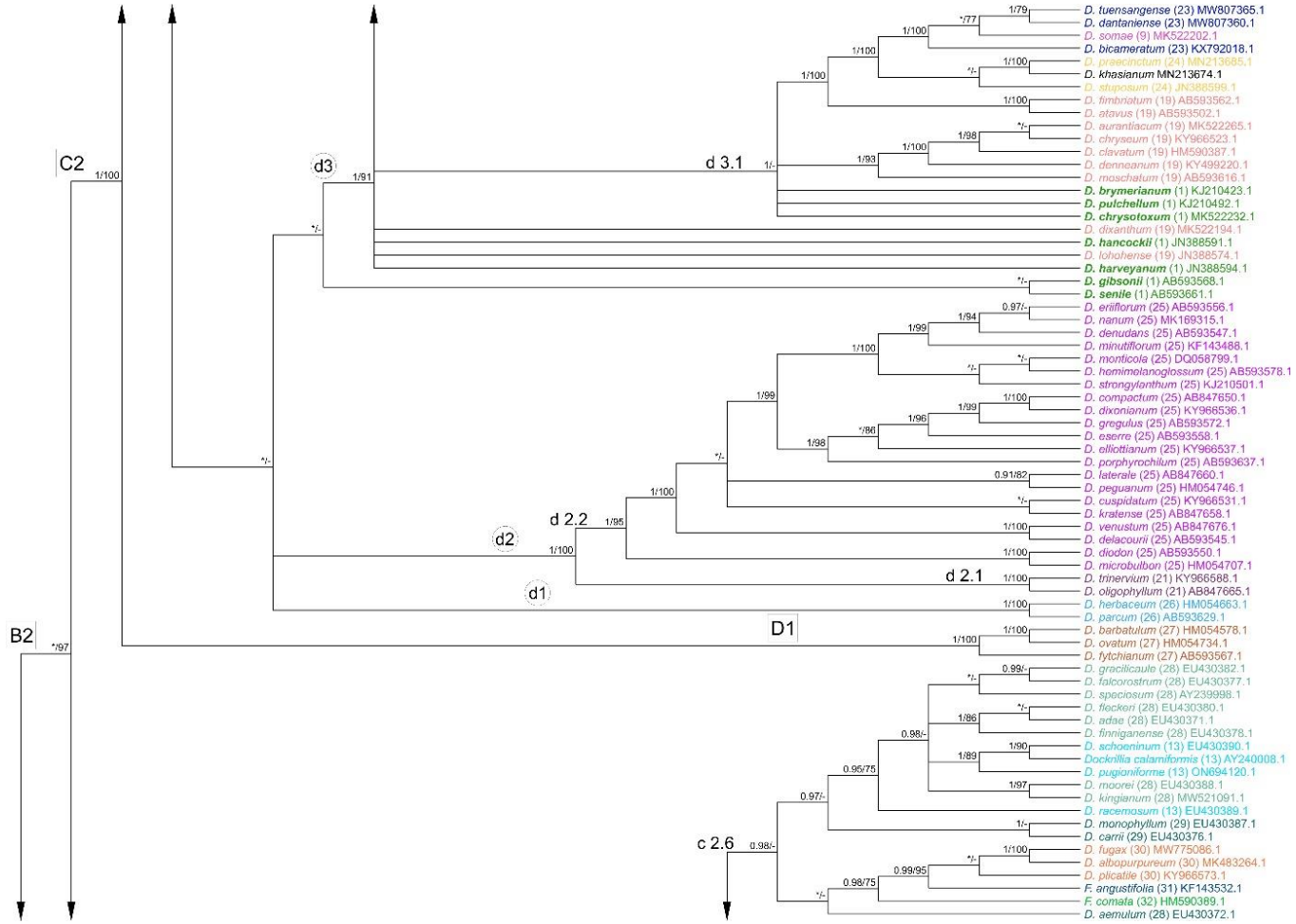

Fig. 2 50% majority rule consensus tree for *Dendrobium sensu lato*, obtained for the nrITS dataset using Bayesian inference (continuation of Fig. 1). Numbers above branches indicate posterior probability and bootstrap support values from maximum likelihood analysis (PP/BS); PP values <0.95 are marked with an asterisk, and BS values <75% are marked with a dash. Letters with numbers above branches mark clades discussed in the text. Numbers in parentheses next to taxon names indicate section affiliation. In addition, each section is highlighted in a different color and the species of nominal section are bolded.

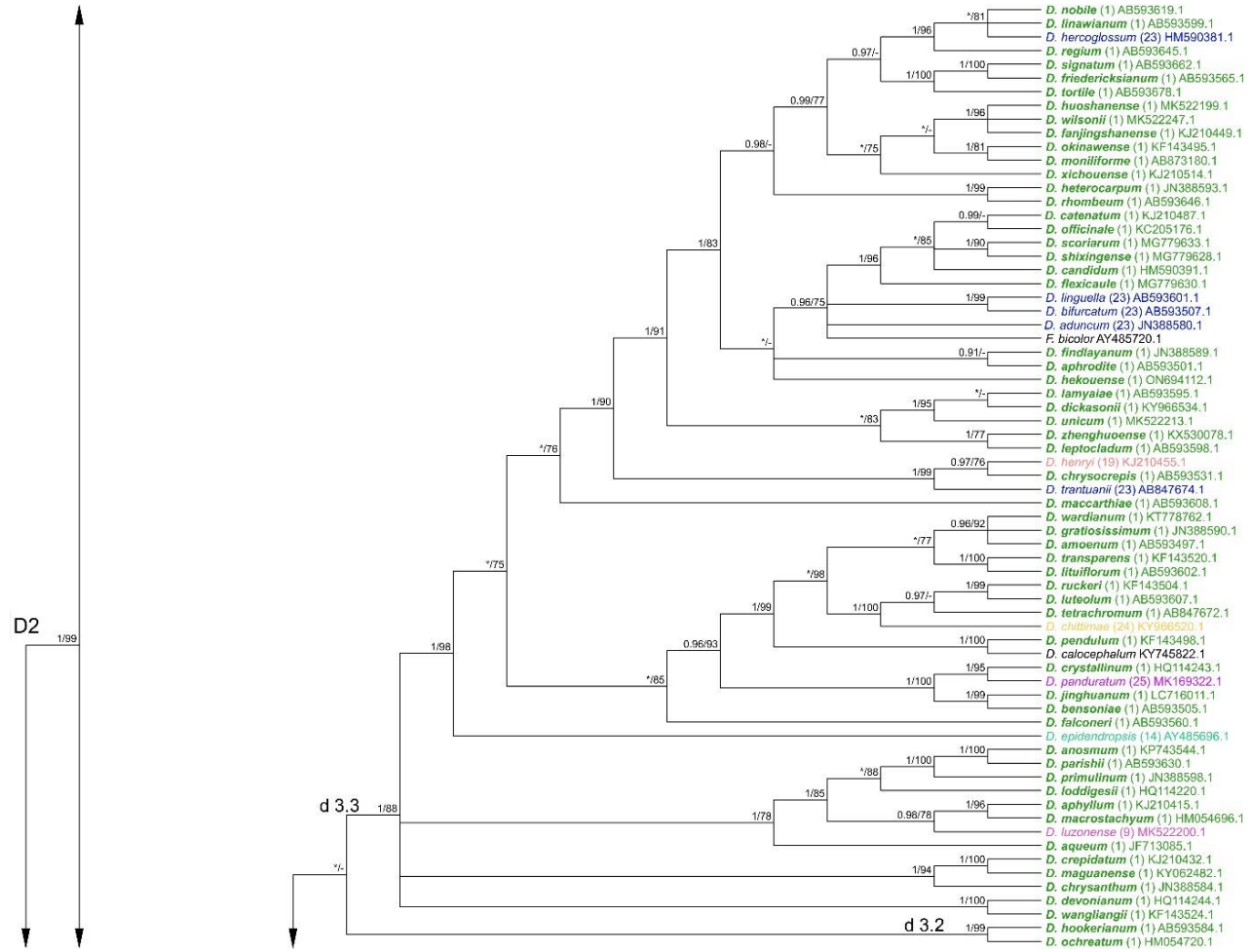

Fig. 3 50% majority rule consensus tree for *Dendrobium sensu lato*, obtained for the nrITS dataset using Bayesian inference (continuation of Fig. 2). Numbers above branches indicate posterior probability and bootstrap support values from maximum likelihood analysis (PP/BS); PP values <0.95 are marked with an asterisk, and BS values <75% are marked with a dash. Letters with numbers above branches mark clades discussed in the text. Numbers in parentheses next to taxon names indicate section affiliation. In addition, each section is highlighted in a different color and the species of nominal section are bolded.

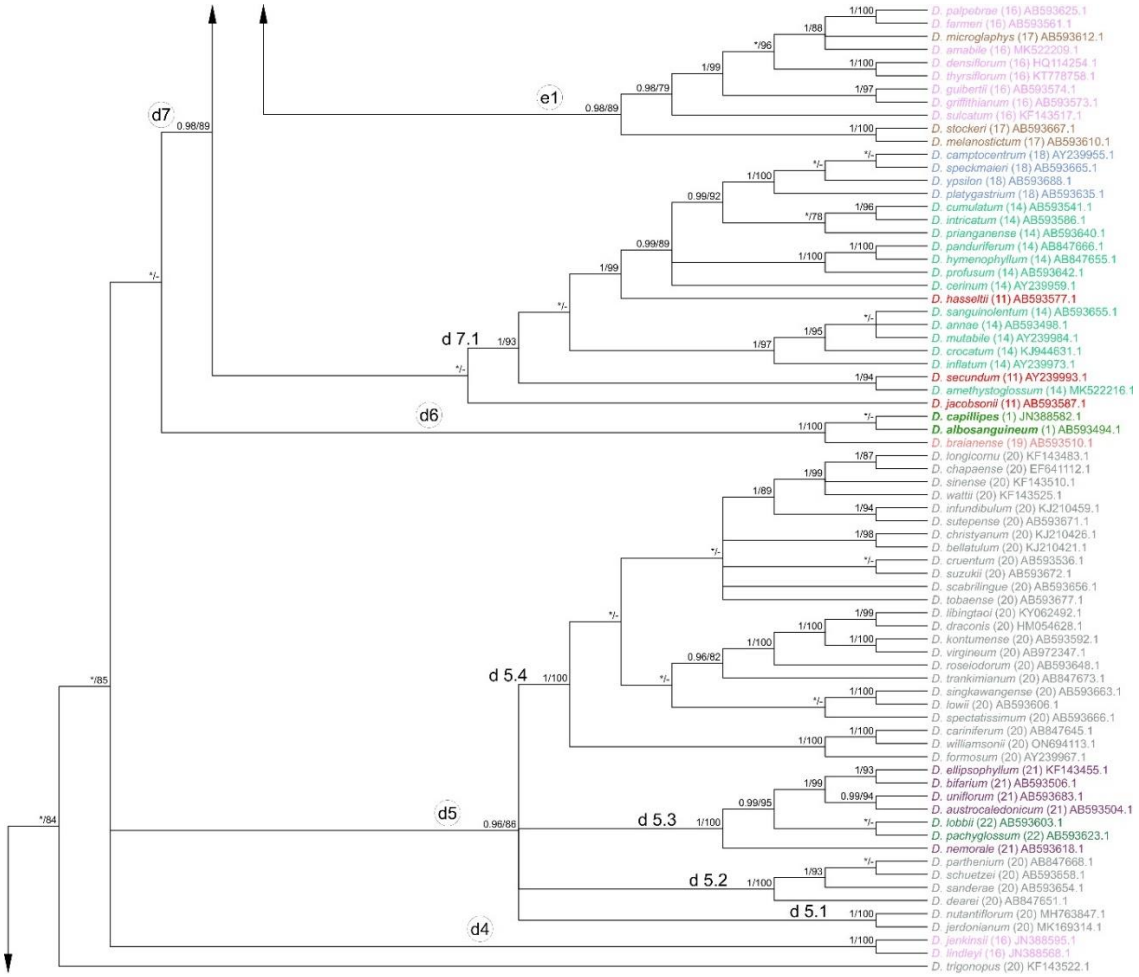

Fig. 4 50% majority rule consensus tree for *Dendrobium sensu lato*, obtained for the nrITS dataset using Bayesian inference (continuation of Fig. 3). Numbers above branches indicate posterior probability and bootstrap support values from maximum likelihood analysis (PP/BS); PP values <0.95 are marked with an asterisk, and BS values <75% are marked with a dash. Letters with numbers above branches mark clades discussed in the text. Numbers in parentheses next to taxon names indicate section affiliation. In addition, each section is highlighted in a different color and the species of nominal section are bolded.

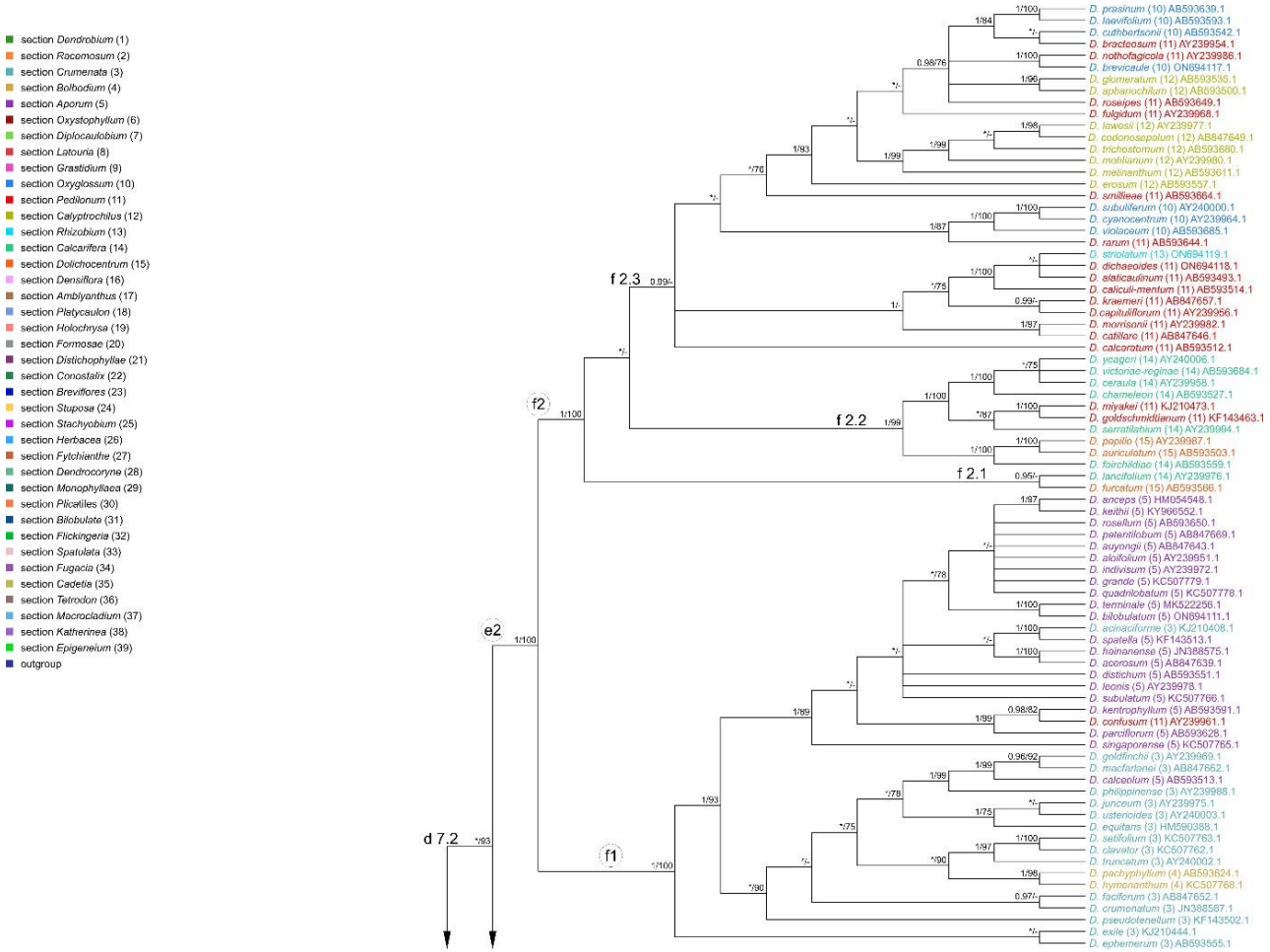

Fig. 5 50% majority rule consensus tree for *Dendrobium sensu lato*, obtained for the nrITS dataset using Bayesian inference (continuation of Fig. 4). Numbers above branches indicate posterior probability and bootstrap support values from maximum likelihood analysis (PP/BS); PP values <0.95 are marked with an asterisk, and BS values <75% are marked with a dash. Letters with numbers above branches mark clades discussed in the text. Numbers in parentheses next to taxon names indicate section affiliation. In addition, each section is highlighted in a different color and the species of nominal section are bolded.

Ancestral state reconstruction (ASR) of morphological features. The reconstruction of the ancestral states of *Dendrobium* features is presented in 4 different phylogenetic trees (Figs S1-S4, Additional file 2). States for characters: 1. Pseudobulbs separated (green color)/approximate; 2. Pseudobulbs and leaves hairy (red color)/glabrous; 3. Pseudobulbs leafy at the apex (light blue color)/leafy througout; 4. Pseudobulbs swollen (dark blue color)/reedlike stem present in the Fig. S1 (Additional file 2). The reconstruction of following features are given at the Fig. S2 (Additional file 2): 5. Leaves laterally (green color)/dorsiventraly (red color) compressed; terete or subterete (dark blue color); 6. Leaves thick, succulent (light blue color) are reconstructed at Fig. S2 (Additional file 2). Fig. S3 (Additional file 2) presents the reconstruction for characters: 7. Inflorescence elongate (green color)/short; 8. Inflorescence produced on last year' (red color)/current pseudobulbs; 9. Inflorescence placed laterally (dark blue color)/apically on/along pseudobulbs. Finally, the reconstruction of: 10. Lip 3-lobed (red color)/unlobed, 11. Lip surface tomentose, papillose, glandular to hairy (green color)/glabrous, papillate or wrinkled; 12. Lip callus absent (dark blue color)/prominent; 13. Mentum as long as or longer (light blue color)/shorter than dorsal sepal is shown in Fig. S4 (Additional file 2).

The results of the analysis suggest that the last common ancestor of *Dendrobium sensu lattisimo* was characterized by approximate, glabrous, leafy (throughout) and reed-like pseudobulbs, dorsiventrally compressed leaves, inflorescence placed apically on current pseudobulbs and glabrous, papillate or wrinkled, unlobed lip with prominent callus and mentum shorter than dorsal sepal. In addition, most of the characters examined arose independently several times during evolution and were then lost in different lineages. The only opposite to this is dorsiventrally compressed leaves, a character that arose only once, but has also been lost in species of different *Dendrobium* sections.

Morphological similarities. To describe mutual resemblance in *Dendrobium sensu lato*, a similarity matrix for the 14 morphological characters studied was used in a hierarchical cluster analysis. The morphological clustering cophenetic coefficient with the Jaccard similarity matrix was 0.786. The resulting UPGMA dendrogram separated specimens into two main clusters, where *D. ypsilon* remained distinct in the upper part of the dendrogram (Fig. S5 in Additional file 3). The two clusters differed primarily in traits describing: the appearance of inflorescences on current vs. last year's psedudobulbs (Ch8); the position of inflorescences on psedudobulbs (apically vs. laterally) (Ch9); lip shape (unlobed vs. three-lobed) (Ch11); lip surface (Ch12); and the presence of lip callus (Ch13) (Table S3 in Additional file 3). The first cluster (**C1**) included specimens from *D. cariniferum* to

D. antennatum and corresponded to species in which the inflorescences are primarily produced on the current pseudobulbs and are apically placed on them, the lips are mostly divided into 3-lobed structure, with glabrous, papillate or wrinkled surface, and with prominent and present callus. In contrast, the second cluster (**C2**) included the remaining species from *D. herbaceum* to *D. cerinum* and the species grouped here have inflorescences mostly laterally placed on last year's pseudobulbs, the lips are unlobed, with a usually tomentose, papillose, glandular or hairy surface, and no callus in most species in this group.

Cluster **C1** revealed three smaller subclusters (**a, b** and **c**) (Fig. 6). Subcluster **a** contained species from *D. cariniferum* to *D. keithii* and was further divided into two subgroups (**a1** and **a2**). The first of these, **a1** (from *D. cariniferum* to *D. sanderae*) included species characterized by hairy pseudobulbs and leaves (Ch2), where the leaves are dorsiventrally compressed and thin (Ch5 and Ch6). Representatives of section *Formosae* have been grouped here. In contrast, the second subgroup, **a2** (from *D. acinaciforme* to *D. keithii*) included species characterized by glabrous pseudobulbs and leaves (Ch2), where the leaves are laterally compressed, terete or subterete and thick (Ch5 and Ch6). Representatives of the sections *Aporum* and *Oxystophyllum* belong here. Distinguished below is subcluster **b**, which is further subdivided into the separate, single species *D. microglaphys*, and the extensive and highly diverse subcluster **c**, from *D. bicameratum* to *D. antennatum*. This subcluster was divided into two groups (**c1** and **c2**), and within the **c1** group, three subgroups were distinguished (**c1.1**, **c1.2**, **c1.3**). Group **c1** (from *D. bicameratum* to *D. densiflorum*) from group **c2** (from *D. minutiflorum* to *D. antennatum*) was separated on the basis of Ch3 and Ch4 characters, where species in group **c2** have pseudobulbs with full-length foliage, and the leaves are reed-like. This group included species from sections *Spatulata* and *Fytchianthe*. A smaller subgroup **c1.1** (from *D. bicameratum* to *D. bifalce*) was consistent with species with pseudobulbs leafy at the apices and swollen (Ch3 and Ch4), where the lips are 3-lobed with present and prominent callus (Ch11 and Ch13); another subgroup **c1.2** (from *D. brymerianum* to *D. laevifolium*) has quite often thick leaves (Ch6); and subgroup **c1.3** (from *D. senile* to *D. densiflorum*) was determined by the traits associated with unlobed lip and lack of callus (Ch11 and Ch13). Species belonging to the sections *Latouria*, *Dendrocoryne*, *Monophyllaea*, *Epigeneium*, *Plicatiles*, *Katherinea*, *Racemosum* and *Diplocaulobium* were represented in subgroup **c1.1**. Subsequently, specimens from the sections *Rhizobium*, *Cadetia*, *Bolbodium* and *Oxyglossum* were grouped in subgroup **c1.2**, while subgroup

c1.3 was designated by section *Densiflora*.

The second cluster (**C2**) appears to be more homogeneous and less diverse, and the species belonging here are characterized by inflorescences placed laterally along pseudobulbs that are of last year (Ch8 and Ch9), and the lip surface is tomentose, papillose, glandular to hairy (Ch12), with a few exceptions (Fig. 7). This cluster was divided into two subclusters - **d**, extensive but uniform, and a smaller one with two species, *D. herbaceum* and *D. auriculatum*, at the top. Subcluster **d** was further divided into two groups - **e** and **f**, differing primarily in the Ch11 trait. Group **f** includes species having a 3-lobed lip for this trait and consists of specimens from *D. cuspidatum* to *D. cerinum*, mainly from the sections *Distichophyllae*, *Conostalix* and *Breviflores*. In turn, group **e** was subdivided into two subgroups (**e1** and **e2**), while the **e1** subgroup was split into four smaller subgroups (**e1.1**, **e1.2**, **e1.3**, **e1.4**). To subgroup **e1** belongs species from *D. hemimelanoglossum* to *D. furcatum*, while subgroup **e2** aggregates species from *D. salaccense* to *D. dickasonii*. Identification of subgroup **e1.1** (from *D. hemimelanoglossum* to *D. platygastrium*) was based on features associated with an elongated inflorescence (Ch7) and a glabrous, papillate or wrinkled lip surface (Ch12). Some species of sect. *Pedilonum* belong here. Another subgroup **e1.2** (from *D. melanostictum* to *D. prianganense*) was distinguished on the basis of the short inflorescence (Ch7), which is placed laterally along pseudobulbs (Ch9) and the different lip surface (Ch12) than in the case of **e1.1**. Most species belonging to this subgroup are assigned to the broad *Dendrobium* section. In the smaller subgroup **e1.3** (from *D. lohohense* to *D. lituiflorum*), species have an elongated inflorescence (Ch7) which in turn is in most located apically on pseudobulbs (Ch9), and lip callus is absent (Ch13). Mainly representatives from section *Holochrysa* are characterized by such morphology. The last subgroup - **e1.4** within the **e1** group, included species from *D. victoriae-reginae* to *D. furcatum*, which have a short inflorescence (Ch7), a glabrous, papillate or wrinkled lip surface (Ch12), and above all, this subgroup is distinguished by having a mentum as long or longer than the dorsal sepal (Ch14).

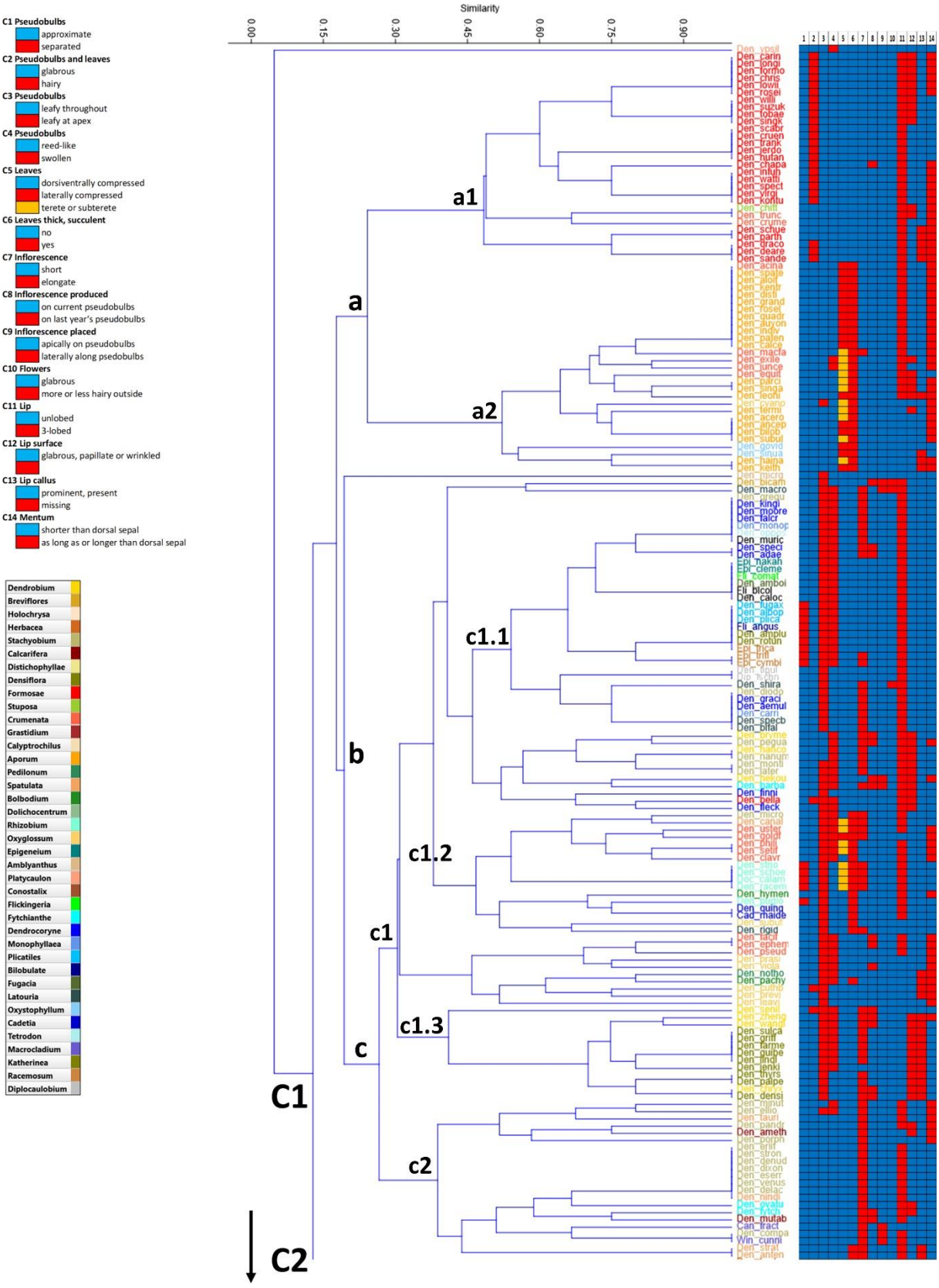

Fig. 6 The first part of the UPGMA analysis based on Jaccard similarity coefficients, showing morphological resemblance for *Dendrobium sensu lato* specimens within cluster **C1**. The discussed subclusters are denoted by lowercase letters and numbers. A detailed description of the morphological characters can be found in Table S3 in Additional file 3.

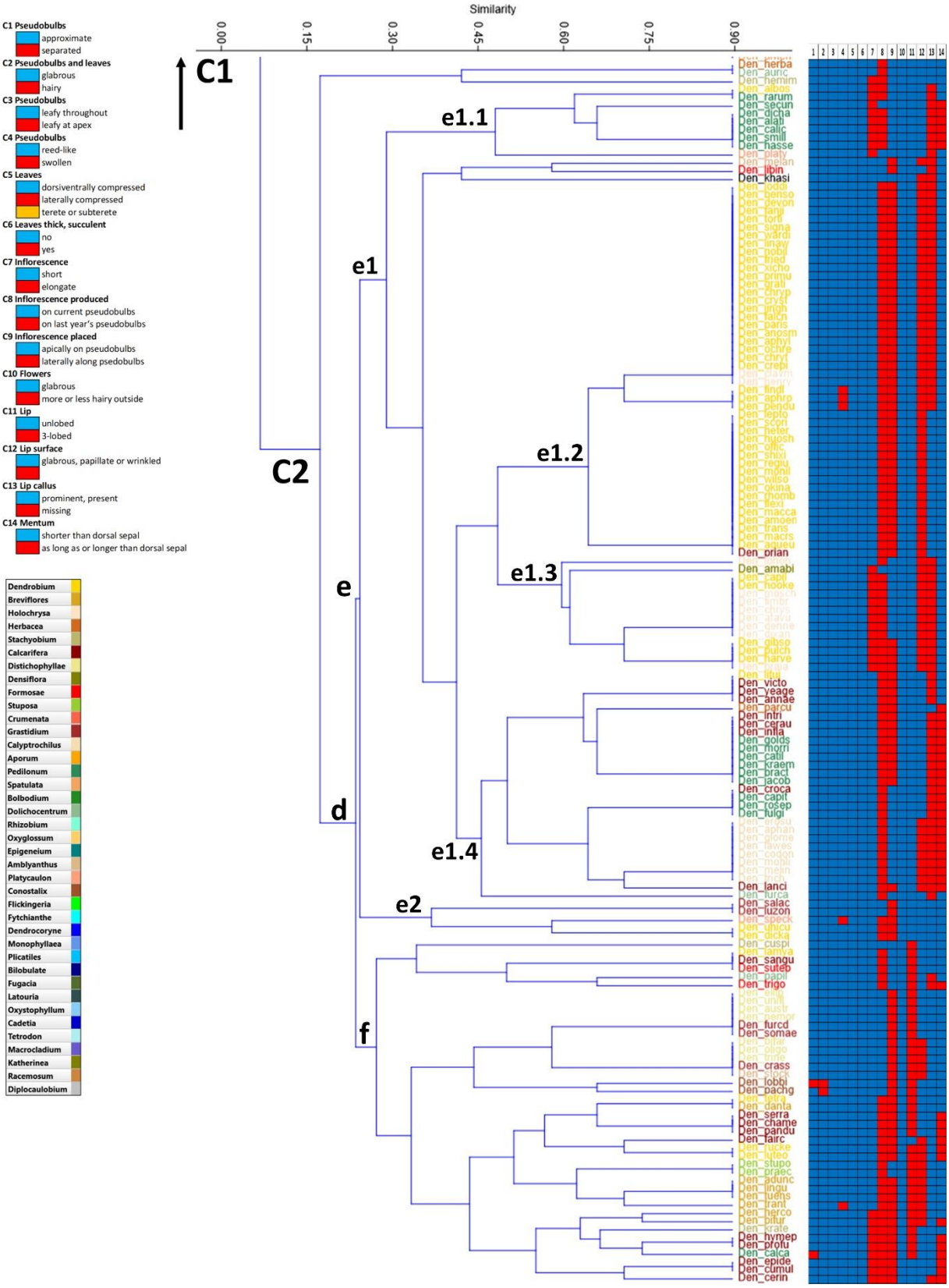

Fig. 7 The second part of the UPGMA analysis based on Jaccard similarity coefficients, showing morphological resemblance for *Dendrobium sensu lato* specimens within cluster **C2**. The discussed subclusters are indicated by lowercase letters and numbers. A detailed description of the morphological characters can be found in Table S3 in Additional file 3.

Additional cluster analysis was carried out to describe patterns of variation in *Dendrobium sensu lato* but based on 5 floral characters (Ch10-Ch14; Table S3 in Additional file 3). In this case, the cophenetic correlation coefficient was 0.774, and this is a good value for how accurately the resulting dendrogram reflects the pairwise distances between the original unmodeled data. Dendrogram obtained using Ward's method divided the specimens into two main clusters (**Cf1** and **Cf2**), in which six predominant patterns of floral variation can be observed (Fig. S6 in Additional file 3). The first floral cluster (**Cf1**) included species from *D. densiflorum* to *D. yeageri* and corresponded to species in which the flowers are glabrous with unlobed lips, where the surface is mostly tomentose or papillose, glandular to hairy; the callus is absent, and the mentum is shorter than the dorsal sepal. All species of the broad section *Dendrobium* are grouped here. In turn, the second floral group (**Cf2**) is more diverse and includes species from *D. shiraishii* to *D. fulgidum*, and the species grouped here also have glabrous flowers, but the lips are trilobed. However, in terms of the other three traits, this group is more variable, and it is the type of lip surface, the presence or absence of callus, and the length of the mentum in relation to the dorsal sepal that distinguished the smaller subgroups (**cf2.2-cf2.4**). The exceptions are three species in the upper part of this cluster (*D. shiraishii*, *D. bicameratum*, *D. macrophyllum*), which have flowers that are more or less hairy on the outside, unlike the other representatives of this group.

The first of the floral clusters, **Cf1** revealed two subclusters: **cf1.1** and **cf1.2**, where the latter is divided into three smaller groups (**cf1.2a, b, c**) (Fig. 8). Subcluster **cf1.1** was mainly dominated by three sections: *Densiflora*, *Dendrobium* and *Holochrysa*. The flowers of the species grouped here are glabrous, the lip is unlobed, with a tomentose, papillose or glandular to hairy surface of the lip, where callus is missing, and mentum is shorter than dorsal sepal. Subcluster **cf1.2**, on the other hand, was also dominated by other representatives of the section *Dendrobium* in the upper part, designating group **cf1.2a**, while below it placed the other two groups (**cf1.2b** and **cf1.2c**), which were already more diverse in terms of taxonomic affiliation. The **cf1.2a** group was distinguished by tomentose, papillose or glandular lip surface and the presence of callus (Ch12 and Ch13). In contrast, group **cf1.2b** included species from *C. fractiflexa* to *D. govidjoae*, which have a different lip surface and callus is present, in contrast to the **cf1.2c** group (from *D. annae* to *D. yeageri*), where callus is absent.

The second floral cluster, **Cf2** presented four main floral patterns (**cf2.1-cf2.4**) (Fig. 9). The first of these, subcluster **cf2.1**, is the most uniform and includes species from *D. mutabile* to *D. diodon*.

Species mainly from the sections *Dendrocoryne*, *Rhizobium*, *Epigeneium*, *Plicatiles*, *Cadetia*, *Katherinea* and *Racemosum* are grouped here. This is the group containing glabrous flowers with a three-lobed lip, where the callus is prominent and the mentum is shortened in relation to the dorsal sepal. The next subcluster **cf2.2** is divided into two groups, where the lip is trilobed with a tomentose, papillose or glandular surface, but the distinguishing feature between the two groups is the length of the mentum relative to dorsal sepal (Ch14): for **cf2.2a** the mentum is equal to or longer than dorsal sepal (from *D. luteolum* to *D. ruckeri*), while for **cf2.2b** the mentum is shorter than dorsal sepal (from *D. barbatulum* to *D. ovatum*). Another subcluster **cf2.3** is also divided into two groups (from *D. prasinum* to *D. kontumense*): the smaller **cf2.3a** with *Dendrobium* species without a divided lip, and the larger **cf2.3b** including species with 3-lobed lips (Ch11). Species belonging to this subgroup are mainly assigned to the sections *Crumenata* and *Aporum*. The last subcluster **cf2.4** (from *D. terminale* to *D. fulgidum*) is the most diverse compared to the others in terms of flower appearance. Starting with the **cf2.4a** group, which is characterized by unlobed lip, a different lip surface (Ch12) than **cf2.4b** and **cf2.4c**, lack of callus and a short mentum. In contrast, the **cf2.4b** and **cf2.4c** groups differ in the division of the lip into lobes, as well as glabrous, papillate or wrinkled lip surface compared to **cf2.4a**. Specimens from the following sections belong here: *Pedilonum* and *Calyptrochilus*.

In general, small sections are consistent in the two analyses with respect to morphology and flower patterns. On the other hand, large sections such as *Dendrobium*, *Stachyobium* or *Formosae* are already more morphologically variable and here the range of variation is already wider and varies depending on the set of characters analyzed.

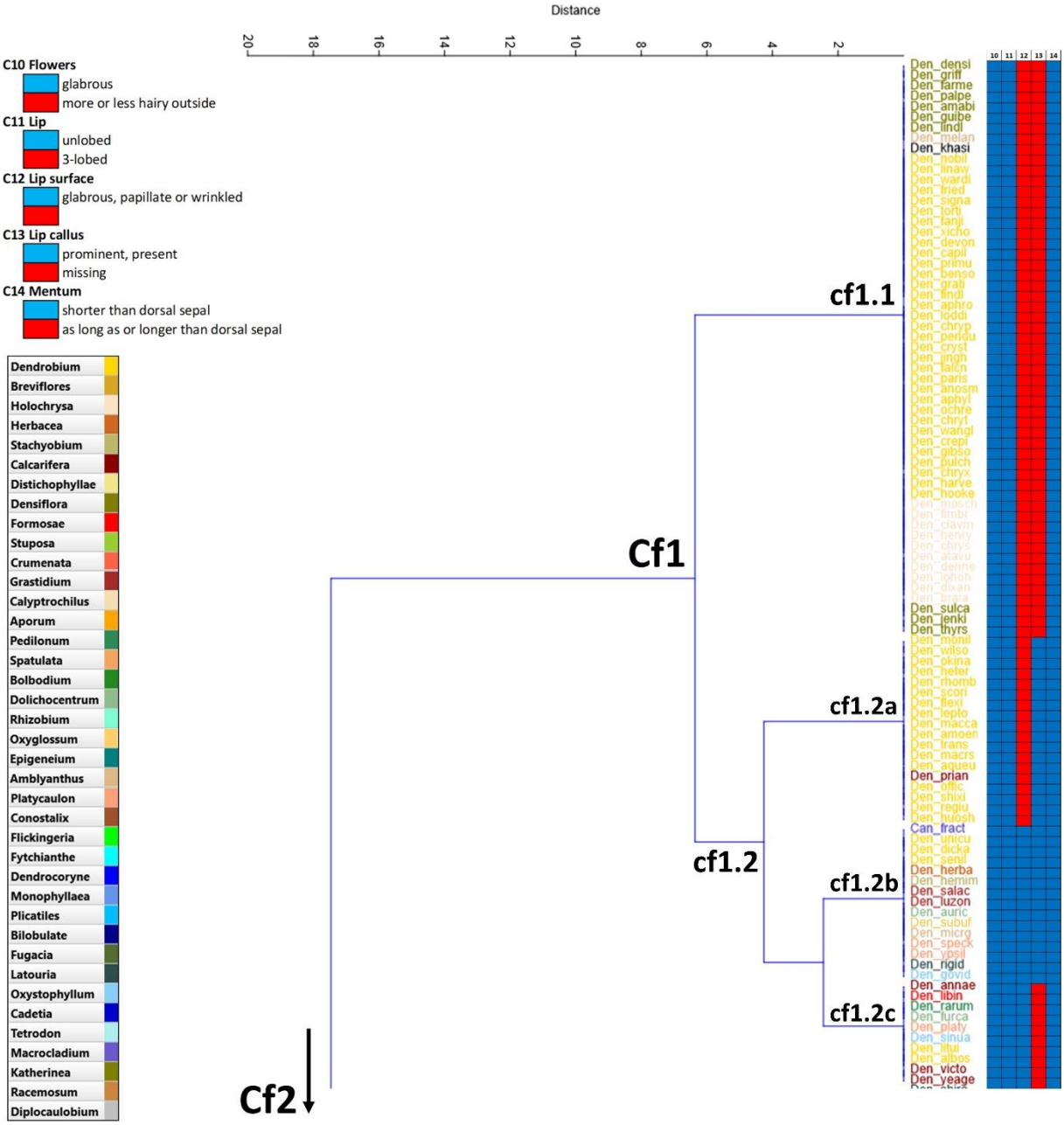

Fig. 8 The first part of the cluster analysis conducted using Ward's method, showing similarity in relation to floral characters for *Dendrobium sensu lato* species within cluster **Cf1**. The discussed subclusters are denoted by lowercase letters and numbers. A detailed description of floral characters can be found in Table S3 in the Additional file 3.

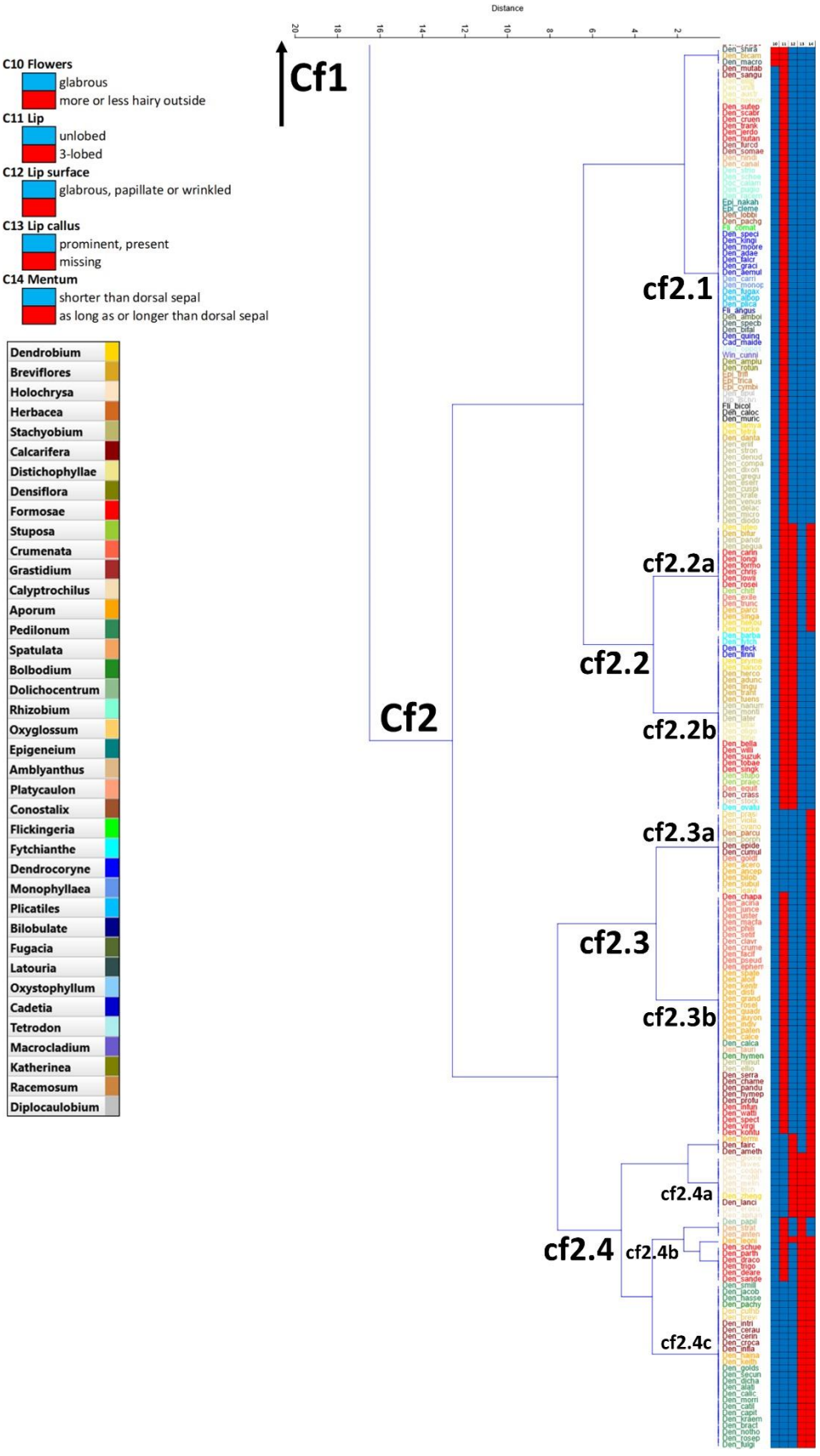

Fig. 9 The second part of the cluster analysis conducted using Ward's method, showing similarity in relation to floral characters for *Dendrobium sensu lato* species within cluster **Cf2**. The discussed subclusters are denoted by lowercase letters and numbers. A detailed description of floral characters can be found in Table S3 in the Additional file 3.

SIMPER analysis identified morphological characters as the most differentiating ones, and these were the same traits described above that divided the *Dendrobium sensu lato* into two major, large clusters - C1 and C2. These were characters related to the position of inflorescences on pseudobulbs from the current or previous year (Ch8 and Ch9), and the presence or absence of a three-lobed lip (Ch11), as well as the type of lip surface (Ch12). The overall average dissimilarity was 69.18% for the morphological characters used comprehensive in our study (Table 1A). In contrast, in the analysis based on floral traits, the presence or absence of a three-lobed lip (Ch11) was identified as the trait that most differentiated *Dendrobium sensu lato*, and it was this trait that divided *Dendrobium* into two main groups (clusters Cf1 and Cf2) described above. The next three traits related to the lip surface, mentum length and callus occurrence (Ch12, Ch14 and Ch13, respectively) had similar average similarity values, while the trait related to flower hairiness (Ch10) was of negligible importance in the analysis. In this case, the overall average dissimilarity equalled 63.53% (Table 1B).

NMDS and PCoA analyses showed a wide range of variability in *Dendrobium sensu lato*, but the sections overlapped considerably (Fig. 10). Despite the apparent and continuous morphological variation, the resulting image is the same as that visualized in the UPGMA cluster analysis. However, the cumulative percentage of explained variance by the first two axes in the PCoA analysis was only 39%, while the stress value was S=0.474, indicating the low quality of the fit in NMDS. This is largely due to the number of minor variables used in the ordering, as well as the close morphological similarity of the taxa, where their ranges of morphological variation overlapped. In both cases, the division of sections among *Dendrobium sensu lato* was along the second axis. In the case of the NMDS, the left side of the scatter plot grouped representatives for sections associated with cluster C2 in the UPGMA (e.g., sections *Dendrobium*, *Calcarifera*, *Calyptrochilus*, *Grastidium* and *Pedilonum*) (Fig. 10A), while on the opposite side were species belonging to cluster C1. An almost identical picture (although it appears slightly more ordered) was obtained in the PCoA analysis, with the difference that on the left side of the plot were concentrated representatives of sections related to cluster C1, while on the other side of the axis were characteristic sections for cluster C2 (e.g., sections *Formosae*, *Aporum*, *Crumenata*, *Dendrocoryne*, *Oxyglossum*, *Spatulata*, *Rhizobium*, *Racemosum*) (Fig. 10B).

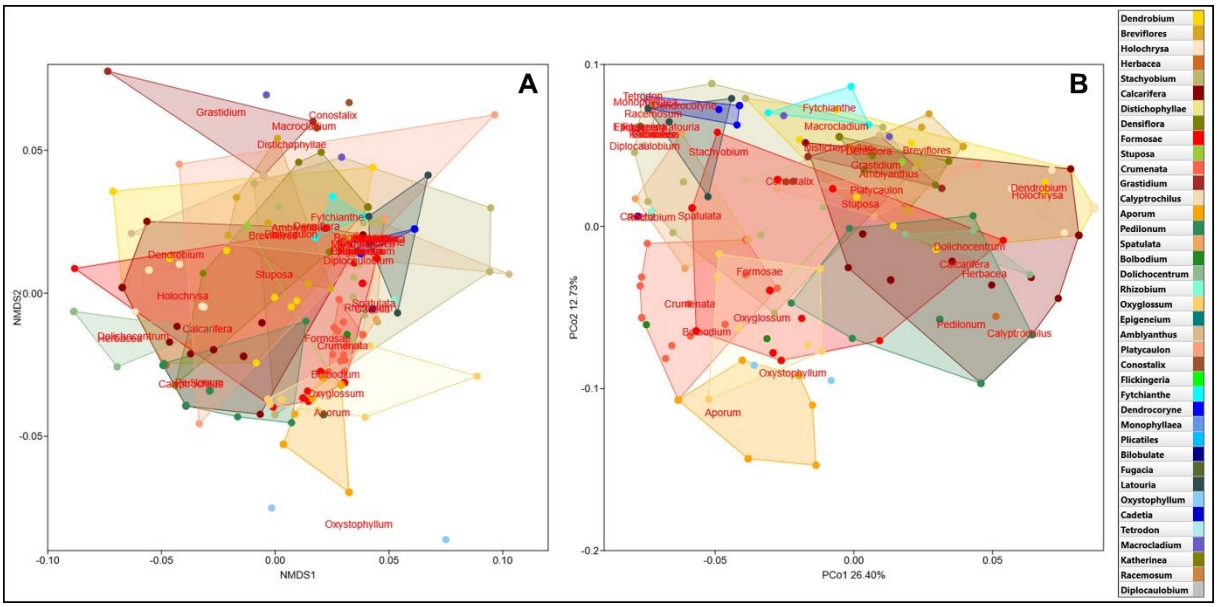

Fig. 10 Non-metric multidimensional scaling analysis, NMDS (**A**) and principal coordinate analysis, PCoA (**B**) showing the two-dimensional ordering of *Dendrobium sensu lato* specimens, based on 14 morphological characters. Convex hulls for each *Dendrobium* section were added to the plots.

In subsequent NMDS and PCoA analyses conducted only for floral traits, there was again a large overlap in the ranges of morphological variation, and thus a high similarity between the observed floral patterns for among individual *Dendrobium* sections (Fig. 11). The cumulative percentage of explained variance by the first two axes in the PCoA analysis was almost 70%, while the stress value was S=0.378, indicating the low quality of the matching in NMDS. As with the overall analysis, the picture obtained here is very similar to that visualized in the UPGMA cluster analysis. Again, the distribution of sections among *Dendrobium sensu lato* followed the second axis in the PCoA analysis (Fig. 11B). Representatives of sections associated with the **Cf2** flower cluster were concentrated on the left side of the plot, while sections specific to the **Cf1** flower cluster were located on the other side of the axis.

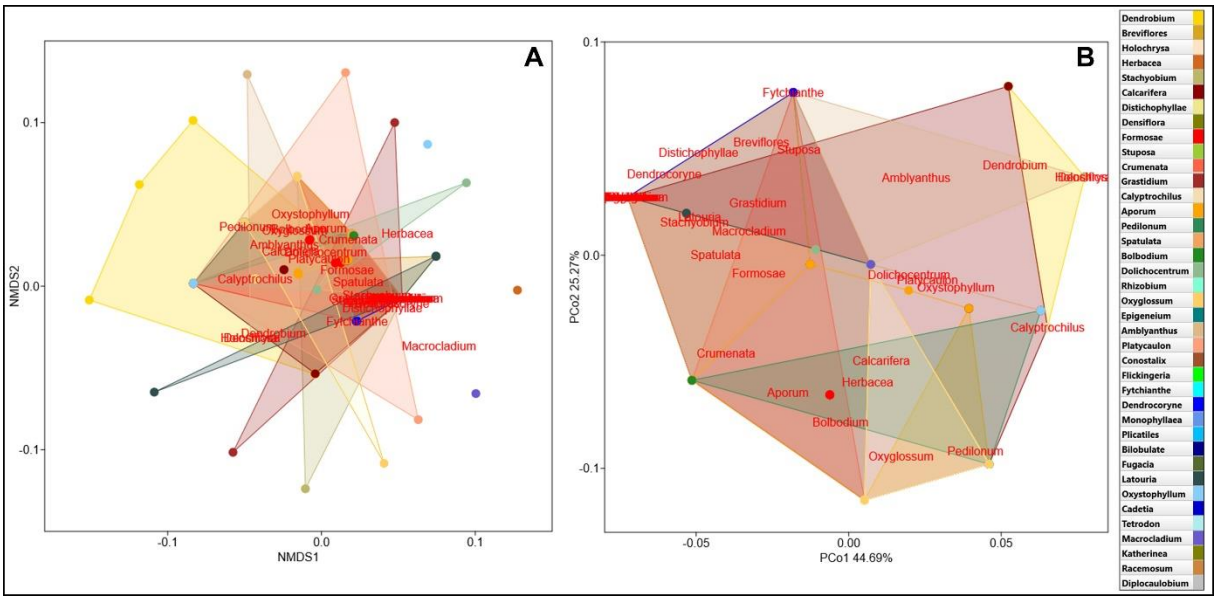

Fig. 11 Non-metric multidimensional scaling analysis, NMDS (**A**) and principal coordinate analysis, PCoA (**B**) showing the two-dimensional ordering of *Dendrobium sensu lato* specimens, based on only 5 floral characters. Convex hulls for each *Dendrobium* section were added to the plots.

Table 1 SIMilarity PERcentage (SIMPER) analysis for 14 morphological characters (**A**) and only for 5 floral traits (**B**) showing the contribution of each trait in distinguishing between taxa within *Dendrobium sensu lato*. Overall average divergence was 69.18% for A and 63.53% for B, respectively. For a detailed description of the morphological characters, see Table S3 in Additional file 3 (Av. dissim. - average dissimilarity; Contrib. % - percentage of similarity explained by individual traits; Cumulat. % - cumulative percentage of Bray-Curtis similarity).

A) Morphological traits	**Av. dissim.**	**Contrib. %**	**Cumulat. %**
8	7.43	10.75	10.75
11	7.41	10.70	21.45
12	6.96	10.06	31.51
9	6.77	9.78	41.29
14	6.55	9.47	50.76
13	6.36	9.19	59.95
7	6.17	8.92	68.87
3	5.52	7.98	76.85
4	4.44	6.41	83.26
5	4.01	5.80	89.06
6	3.54	5.12	94.18
2	2.56	3.70	97.88
1	1.23	1.78	99.66
10	0.23	0.34	100
B) Floral traits	**Av. dissim.**	**Contrib. %**	**Cumulat. %**
11	18.17	28.60	28.60
12	15.90	25.02	53.62
14	14.72	23.16	76.78
13	14.18	22.32	99.10
10	0.569	0.896	100

Discussion

Dendrobium is a large genus with a wide geographical distribution. Despite numerous studies, a consistent classification of the group as a whole and for individual sections has yet to be developed. There is still a lack of comprehensive data to at least determine the sectional affiliation of individual species. The identification of morphological traits allows us to understand the genetic relationships between particular species [49]. Unfortunately, in the case of *Dendrobium*, such studies are clearly not sufficient to formulate even preliminary conclusions about the relationship. It is a complex and problematic group. A situation of this genus now resembles the problems that researchers dealing with the genus *Pleurothallis sensu lato* encountered before Luer's fundamental works. *Dendrobium* has a high morphological diversity and overlapping differences. This is confirmed by the results of Adams, Zhitao et al. and our own (Figs. 10 and 11) [11, 29]. As can easily be seen, the phylogeny of *Dendrobium* also needs to be clarified (Figs 1-5), and what is more, it is not entirely consistent with morphology (Figs 6-9). The result obtained for the ITS marker (Figs 1-5) shows that *Dendrobium sensu latissimo* is a monophyletic group, contrary to the work of Clements [34]. However, our result also proves that the nominal section, as traditionally understood, is not monophyletic (Figs 2-4). To clarify this situation, we focused on and combined two main aspects - morphological variability and phylogenetic relationships. This allowed us to more thoroughly analyze species from the nominal section and from the remaining sections where we had doubts about taxonomic affiliation, based on comprehensive information. The species belonging to the section *Dendrobium*, according to different authors and our results, are listed in Table S6 (Additional file 4).

We also plotted the distribution of the analyzed *Dendrobium* species on the cladogram with the potential evolution of floral characters (Fig. S4 in Additional file 2). This allowed to check whether morphologically similar taxa occur in the same geographic regions. Based on the information obtained, we conclude that the center of diversity of *Dendrobium* species can probably be located in the island part of Asia. From there, *Dendrobium sensu lato* could potentially migrate in three directions: Australia and New Zealand, secondly India and Sri Lanka, and finally China, Korea and Japan (Fig. S4 in Additional file 2). This is a small step towards analyzing the migration of species that may have spread from tropical to subtropical regions [4].

A comparison of morphological similarity analyses, ancestral state reconstructions for morphological traits, and phylogenetic analysis showed that vegetative traits are much less variable

than the floral ones. We suggest that floral traits in *Dendrobium* representatives evolved several times independently (Figs. S3 and S4, Additional file 2). Therefore, taxa placed in the same section did not always form a consistent clade on the phylogenetic tree, such as species in the nominal section. Morphological similarity analyses also did not fully reflect previously proposed sectional divisions. Section *Oxystophyllum* is presented by only two species in our study but they are included in one clade (A1, Fig. 1). However, in the morphological analysis, they were placed in two separate clusters, cf1.2c and cf1.2b (Fig. 8), simply because *D. sinuatum* does not have a callus on the lip. In contrast, clade B1 (Fig. 1) is consistent. It includes species of *Racemosum*, *Epigeneium* and *Katherinea*. This time, the proposed sectional division was reflected in both the phylogenetic results and the majority of morphological features (Figs. 1, 6 and 9). Considering our results and the works of Clements or Pridgeon et al., one has to think about the position of these taxa (especially *Oxystophyllum*) [10, 34, 50]. With their inclusion in the genus as a whole, *Dendrobium* will still be a monophyletic taxon. Treating these taxa as separate genera, they are sisters to *Dendrobium sensu stricto*.

Worth considering is the section *Holochrysa*. This is a paraphyletic group, and some species seem to fit phylogenetically and morphologically into the nominal section (clade d3, Figs. 2 and 3). Of note is *D. braianense*, which is grouped with *D. capillipes* and *D. albosanguineum* (section *Dendrobium*) on the ITS tree (clade d6, Fig. 4). But these taxa do not join other species of the nominal section. Also, analysis of morphological similarity showed that these species grouped together and with other nominal section taxa (Figs. 7-8). Most species in the typical section have inflorescences produced on last year's pseudobulbs and placed laterally along the pseudobulbs, *D. braianense* also has these, but additionally has an elongated inflorescence. We propose to include *D. brainense* in the section *Dendrobium* based on the results obtained. The question that comes to mind is whether the remaining species of the *Holochrysa* section should be included in the nominal section, as the results of our analyses and Xiang et al. suggest [4]. It is worth considering. On the ITS tree, representatives of this section were grouped as an independent polytomous lineage (subclade d3.1, Fig. 2) that evolved from the common ancestor, the same as taxa of the nominal section (node d3, Fig. 2). However, in d3.1 were also placed *D. chrysotoxum*, *D. pulchellum* and *D. brymerianum* (sect. *Dendrobium*), *D. stuposum* and *D. praecinctum* (sect. *Stuposa*), *D. dantaniense* and *D. bicameratum* (sect. *Breviflores*), *D. somae* (sect. *Grastidium*) and *D. khasianum*. The analysis of floral characters also aggregates taxa of *Holochrysa* together with species of the section *Dendrobium*, but the differences,

e.g. in the structure of the inflorescences, make treating all species as one section still a matter of debate (Figs. 2 and 8, Fig. S3). Also noteworthy is *D*. *henryi* (subclade d3.3, Fig. 3), which is representative of *Holochrysa*. The results, both phylogenetic and morphological analyses, prove that this taxon should be included in the nominal section (Figs. 3, 7 and 8). In addition, similar conclusions obtained by Yuan et al. and Xu et al. [51, 52]. The last species from the section *Holochrysa* that we would like to mention, *D*. *lohohense*, was placed as an independent polytomous branch from node d3 (Fig. 2), from a potential common ancestor, as well as other species of the nominal section. The morphological similarity analysis (Figs 7-8) shows that it clusters together with representatives of the section *Dendrobium*, but the only feature that distinguishes this species is the inflorescence placed apically on pseudobulbs (Figs 7-8). Reconstruction of the ancestral state indicates the plesiomorphic nature of this character (Fig. S3 in Additional file 2). Interestingly, the absence of an elongate inflorescence placed laterally along the pseudobulbs and having only the above variant is not characteristic of either *Dendrobium* or *Holochrysa* (Figs 7-8).

The representatives of the section *Breviflores* were also grouped together with the species of the typical section (clade d3, Figs 2-3). This needs to be discussed like the taxa in the section *Holochrysa*. Surprisingly, *D*. *hercoglossum* (sect. *Breviflores*) formed a strongly supported clade (PP=1, BS=96, Fig. 3) together with *D*. *nobile*, *D*. *linavianum* and *D*. *regium* (sect. *Dendrobium*). This is puzzling, because analyses of morphological characters clearly group this species with the other taxa belonging to the *Breviflores* (Figs. 7 and 9). It has also been considered *Breviflores* instead [7, 9, 51, 53, 54, 55]. In addition, the presence of elongated inflorescences and the other results obtained also led us to leave this species in its current group (Fig. S3 in Additional file 2). A similar situation occurs with the remaining species in this section.

We also noted several species that have never been placed in nominal sections but are grouped in clade d3 (Figs 2-3), e.g. *F. bicolor*, *D*. *khasianum*, or *D*. *calocephalum*. However, based on our results, we believe that although they are closely related to species in the section *Dendrobium*, their morphology is so different that they should be excluded from the typical section.

D. *capillipes* and *D*. *albosanguineum* (clade d6, Fig. 4) were placed beyond the clade with the section *Dendrobium* (clade d3, Figs 2-3). UPGMA analysis for 14 morphological characters divides that both species are clustered with representatives of the nominal section (cluster C2, Figs 6-7), but in different subgroups, e1.3b and e1.1. There are no differences in pseudobulbs or leaves, probably

these characters are relatively consistent in all taxa of the typical section (Fig. 7). In the analysis of the morphology of the floral pattern, these two species are also divided into two subgroups (cf1.2c and cf1.1, Fig. 8). In this case the differences are more pronounced. Despite the presence of some deviations, it seems to us that both species should be included in the nominal section, as recognized by Peyachoknagul et al. [55]. However, we have more doubts in the case of *D*. *capillipes*. The number of leaves in this species varies from three to five. Takamiya et al. show that this is unique for both sections *Holochrysa* and *Dendrobium* [30]. Nevertheless, they treated this species as *Holochrysa*, while Wang et al. classified it in the section *Dendrobium*, and Schuiteman and Xiang et al. treat this taxon as unplaced [7, 2, 4].

D. *gibsonii* and *D*. *senile* were also not connected to the nominal section. They form independent branches that evolved from the ancestor at node D2 (Fig. 3). Takamiya et al. treated *D*. *gibsonii* as part of the section *Holochrysa* [30]. However, the results of our analyses do not confirm the close relationship with taxa of this section. Moreover, the correct assignment of taxa to these two sections is still controversial and the results so far are inconclusive. There are many morphological characters that suggest a combination of these sections. This is also supported by our molecular analysis. Although *D*. *gibsonii* is phylogenetically outside the d3 clade (Figs. 2 and 3), in our opinion, its set of characters allows this species to belong to the section *Dendrobium* (Fig. 8). This proposal has been confirmed by other authors [7, 53, 54]. The situation is different in the case of *D*. *senile*. Not only the phylogeny, but also the vegetative and floral characters do not correspond to the remaining species of the nominal section (Figs. 2 and 8). Therefore, we propose that this species be separated from the section *Dendrobium*. Interestingly, Wood included this species in the section *Formosae* [9], and Xiang et al. treated it as an unplaced species [4].

We were also interested in two other species, *D*. *brymerianum* and *D*. *hancocki*, which Wood placed in the sections *Densiflora* and *Holochrysa*, respectively [9]. The morphological analyses carried out also do not indicate that they belong to the typical section (Figs. 6 and 9). It should be noted that on the phylogenetic tree obtained, they are included in the nominal section (clade d3, Figs 2-3). Despite their phylogenetic affiliation, we prefer to separate these species from the nominal group. Although some authors put them here [54, 35]. In the case of *D. brymerianum*, apart from morphological differences, we noticed its relatively narrow and unusual range of occurrence compared to other species of the type of section.

D. *zhenghuoense*, the species that was grouped with most taxa of the nominal section in the strongly supported clade d3 (PP=1, BS=98, Figs 2-3), is morphologically distinct. The lack of callus on the lip, the mentum as long as or longer than the dorsal sepal, the elongate inflorescence, and the presence of swollen and leafy pseudobulbs at the apex are convergent characters that appear several times, probably in response to specific habitat requirements and pollinator pressure (Figs S1, S3 and S4 in Additional file 2). It is worth noting that Chen et al. obtained similar phylogenetic results [56]. The authors point out the similarity to *D*. *hekounse*, but we think that *D*. *hekounse* is also morphologically different from other members of the nominal section. Despite the phylogenetic affiliation, *D*. *zhenghuoense* has a clearly different set of morphological characters and in our opinion does not belong to the section *Dendrobium*.

Working on a genus as complex as *Dendrobium* requires comparing data from as many sources as possible. When we analyze the results, it is easy to see that some differences or similarities often overlap and do not provide clear answers. Our studies show that the morphological similarity analysis of both vegetative and floral characteristics is relatively consistent in the case of less representative sections. On the other hand, numerous representative sections, such as the nominal one and closely related, e.g. *Holochrysa* or *Breviflores,* are already more morphologically variable, especially in the floral characters. This may be related to the potential pollinator. Phylogenetically, however, they form a fairly coherent and homogeneous group, with a few exceptions such as *Flickingeria bicolor*, *D*. *khasianum*, or *D*. *calocephalum*. This makes it more difficult to delineate between them, or, as Schuiteman have suggested, to tend to group them together [2]. There is such a remarkable similarity in the entire nominal section, especially between the floral patterns that are observed. But we found some arguments that led us to include or exclude some species from this section.

Currently, two extreme approaches to distinguishing taxonomic units of generic rank can be observed. The first, is based on combining into a single taxon, let's call it a supergenus, all taxa having a monophyletic origin, even if they show discontinuities in structure e.g. morphological. Examples from recent years can be multiplied, *Maxillaria* [57], *Bifrenaria* [58], *Herminium* [59], *Oncidium* [60]. Their species range corresponds to the previously distinguished subtribes. Although we obtain monophyletic groups, the problem of their internal classification is relegated to the intrageneric level. The second approach is to distinguish groups that are monophyletic but show distinct, unique combinations of

morphological traits. This approach is difficult, as it requires learning and analyzing not only the sequence of selected markers, but also the exact morphological or anatomical structure of the species which are analyzing.

Dendrobium is an excellent illustration of both approaches to genus definition and taxonomy. Schuitemann proposes a broad view of the genus, which includes all genera of the subtribe Dendrobiinae [2]. On the other hand, Clements [34] and Clements & Jones [33] divided *Dendrobium* into as many as 3 subtribes and a total of 50 genera. In our opinion, *Dendrobium* should be broken down into several genera, but the proposals of the Australian researchers are premature. As our study shows, most of the genera they distinguish are well nested in other *Dendrobium* clades or show polyphyletism.

Our results clearly show that *Dendrobium sensu lato* is a genus with an intricate history in which it is possible to observe different rates of evolution, the loss of certain characteristics and the appearance of others in the process of migration to new areas and under the influence of adaptation to local pollinators, in which episodes of hybridization have also occurred. The taxonomy of such a large, diverse, wide-ranging group is and must be complex. Therefore, many years of intensive research are still ahead of us to find the key to total evidence taxonomy.

Conclusions

Results of our analyses show that *Dendrobium sensu lato* is likely to have undergone convergent evolution. Many floral features have evolved multiple times independently. This leads to numerous classification problems within the genus. We did not get a clear answer on how to classify species within the entire genus. For this reason, in this study we focused mainly on the nominal section. Thus, we treat the determination of species within the nominal section as a priority. It is worth keeping in it only those species that, based on their set of characters, leave no doubt about their membership. Because of such a large group, it is best to take small steps, analyze individual sections, and then put all the data together. Otherwise, studying the entire genus may cause many problems and ambiguities.

Convergent evolution can be the result of adaptation to pollinators or, more commonly, the result of hybridization. This is a common phenomenon in plants, making it difficult to establish a consistent classification system, especially for a genus as numerous and diverse as *Dendrobium*. In this article,

we have not performed analyses that could clearly confirm or refute this theory. However, it is worth paying special attention to this issue in the future.

Supplementary Information

Additional file 1. A list of GenBank ID numbers. Table S1. List of species of *Dendrobium* used in the molecular study for ITS marker including GenBank Accession Number.
Additional file 2. Ancestral state reconstruction of morphological features. Table S2. Data matrix used for ancestral state reconstruction of morphological features, where taxa characters were coded for the presence (1, 2 - yes) or absence (0 - no) of a feature. Fig. S1. Ancestral state reconstruction of morphological characters (pseudobulbs) of *Dendrobium* taxa (according to Table S2). Fig. S2. Ancestral state reconstruction of morphological characters (leaves) of *Dendrobium* taxa (according to Table S2). Fig. S3. Ancestral state reconstruction of morphological characters (inflorescence) of *Dendrobium* taxa (according to Table S2). Fig. S4. Ancestral state reconstruction of morphological characters (lip and mentum) of *Dendrobium* taxa (according to Table S2).Circles next to taxon names indicate geographic distribution.
Additional file 3. Morphological similarities. Table S3. List of morphological characters used to describe *Dendrobium* sensu lato species included in the cluster and multivariate analyses. Table S4. Data matrix of 14 morphological traits used in the analysis of morphological variation in *Dendrobium* sensu lato. The data were transformed according to Table S3 in this supplementary file. Table S5. SIMPER analysis identifying the percentage of similarity and dissimilarity of all morphological traits studied (A) and only floral traits (B) for each section of *Dendrobium* sensu lato. For a detailed description of morphological characters is provided in Table S3 in this supplementary file. Fig. S5. Two-way UPGMA cluster analysis based on Jaccard similarity coefficients, showing morphological relationships within *Dendrobium sensu lato*, with regard to the division into sections. A detailed description of the morphological characters can be found in Table S3 in the additional file. Fig. S6. Two-way cluster analysis conducted using Ward's method, showing relationships within floral traits only for *Dendrobium sensu lato*, with regard to the division into sections. A detailed description of floral characters can be found in Table S3 in the additional file.
Additional file 4. Summary of the taxonomy of the *Dendrobium* section. Table S6. Nominal section species according to various authors and the results obtained.

Declarations

Ethics approval and consent to participate

Not applicable

Consent for publication

Not applicable

Availability of data and materials

All data generated or analysed during this study are included in this published article [and its supplementary information files].

Competing interests

The authors declare that they have no competing interests.

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Author's contributions

A.B.-H., M.D., D.L.S conceived the study. A.B-H. analysed data, presented data on species occurrence, prepared the figures/tables, conducted molecular analyses and wrote the manuscript M.D. performed phylogenetic analyses, analysed data, wrote the manuscript and agreed to serve as author responsible for contact and ensure communication. N.O. performed ASR analyses, prepared figures, wrote the manuscript. A.M.N. performed morphological similarities analyses, prepared figures/tables and wrote the manuscript D.L.S analysed data, prepared a list of morphological features of species and wrote the manuscript. All the authors reviewed the manuscript.

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Supplementary Materials

Table S1. List of species of *Dendrobium* used in the molecular study for ITS marker including GenBank Accession Number.

Species	GenBank Accession Number
D. acerosum	AB847639.1
D. acinaciforme	KJ210408.1
D. adae	EU430371.1
D. aduncum	JN388580.1
D. aemulum	EU430372.1
D. alaticaulinum	AB593493.1
D. albopurpureum	MK483264.1
D. albosanguineum	AB593494.1
D. aloifolium	AY239951.1
D. amabile	MK522209.1
D. amboinense	AB894133.1
D. amethystoglossum	MK522216.1
D. amoenum	AB593497.1
D. amplum	KX600512.1
D. anceps	HM054548.1
D. annae	AB593498.1
D. anosmum	KP743544.1
D. antennatum	ON694122.1
D. aphanochilum	AB593500.1
D. aphrodite	AB593501.1
D. aphyllum	KJ210415.1
D. aqueum	JF713085.1
D. atavus	AB593502.1
D. aurantiacum	MK522265.1
D. auriculatum	AB593503.1
D. austrocaledonicum	AB593504.1
D. auyongii	AB847643.1
D. barbatulum	HM054578.1
D. bellatulum	KJ210421.1
D. bensoniae	AB593505.1
D. bicameratum	KX792018.1
D. bifalce	EU430373.1
D. bifarium	AB593506.1
D. bifurcatum	AB593507.1
D. bilobulatum	ON694111.1
D. bracteosum	AY239954.1
D. braianense	AB593510.1
D. brevicaule	ON694117.1
D. brymerianum	KJ210423.1
D. calcaratum	AB593512.1
D. calceolum	AB593513.1
D. caliculi-mentum	AB593514.1
D. calocephalum	KY745822.1
D. camptocentrum	AY239955.1
D. canaliculatum	EU430375.1
D. candidum	HM590391.1
D. capillipes	JN388582.1
D.capituliflorum	AY239956.1
D. cariniferum	AB847645.1
D. carrii	EU430376.1
D. catenatum	KJ210487.1
D. catillare	AB847646.1
D. ceraula	AY239958.1
D. cerinum	AY239959.1
D. chameleon	AB593527.1
D. chapaense	EF641112.1
D. chittimae	KY966520.1

D. christyanum	KJ210426.1
D. chrysanthum	JN388584.1
D. chryseum	KY966523.1
D. chrysocrepis	AB593531.1
D. chrysotoxum	MK522232.1
D. clavator	KC507762.1
D. clavatum	HM590387.1
D. codonosepalum	AB847649.1
D. compactum	AB847650.1
D. confusum	AY239961.1
D. crassifolium	ON694114.1
D. crepidatum	KJ210432.1
D. crocatum	KJ944631.1
D. cruentum	AB593536.1
D. crumenatum	JN388587.1
D. crystallinum	HQ114243.1
D. cumulatum	AB593541.1
D. cuspidatum	KY966531.1
D. cuthbertsonii	AB593542.1
D. cyanocentrum	AY239964.1
D. dantaniense	MW807360.1
D. dearei	AB847651.1
D. delacourii	AB593545.1
D. denneanum	KY499220.1
D. densiflorum	HQ114254.1
D. denudans	AB593547.1
D. devonianum	HQ114244.1
D. dichaeoides	ON694118.1
D. dickasonii	KY966534.1
D. diodon	AB593550.1
D. distichum	AB593551.1
D. dixanthum	MK522194.1
D. dixonianum	KY966536.1
D. draconis	HM054628.1
D. elliottianum	KY966537.1
D. ellipsophyllum	KF143455.1
D. ephemerum	AB593555.1
D. epidendropsis	AY485696.1
D. equitans	HM590388.1
D. eriiflorum	AB593556.1
D. erosum	AB593557.1
D. eserre	AB593558.1
D. exile	KJ210444.1
D. faciferum	AB847652.1
D. fairchildiae	AB593559.1
D. falconeri	AB593560.1
D. falcorostrum	EU430377.1
D. fanjingshanense	KJ210449.1
D. farmeri	AB593561.1
D. fimbriatum	AB593562.1
D. findlayanum	JN388589.1
D. finniganense	EU430378.1
D. fleckeri	EU430380.1
D. flexicaule	MG779630.1
D. formosum	AY239967.1
D. friedericksianum	AB593565.1
D. fugax	MW775086.1
D. fulgidum	AY239968.1
D. furcatopedicellatum	AF521611.1
D. furcatum	AB593566.1
D. fytchianum	AB593567.1
D. gibsonii	AB593568.1

D. glomeratum	AB593535.1
D. goldfinchii	AY239969.1
D. goldschmidtianum	KF143463.1
D. gracilicaule	EU430382.1
D. grande	KC507779.1
D. gratiosissimum	JN388590.1
D. govidjoae	AY239971.1
D. gregulus	AB593572.1
D. griffithianum	AB593573.1
D. guibertii	AB593574.1
D. haemoglossum	HM054639.1
D. hainanense	JN388575.1
D. hancockii	JN388591.1
D. harveyanum	JN388594.1
D. hasseltii	AB593577.1
D. hekouense	ON694112.1
D. hemimelanoglossum	AB593578.1
D. henryi	KJ210455.1
D. herbaceum	HM054663.1
D. hercoglossum	HM590381.1
D. heterocarpum	JN388593.1
D. hookerianum	AB593584.1
D. huoshanense	MK522199.1
D. hymenanthum	KC507768.1
D. hymenophyllum	AB847655.1
D. indivisum	AY239972.1
D. inflatum	AY239973.1
D. infundibulum	KJ210459.1
D. intricatum	AB593586.1
D. jacobsonii	AB593587.1
D. jenkinsii	JN388595.1
D. jerdonianum	MK169314.1
D. jinghuanum	LC716011.1
D. junceum	AY239975.1
D. keithii	KY966552.1
D. kentrophyllum	AB593591.1
D. khasianum	MN213674.1
D. kingianum	MW521091.1
D. kontumense	AB593592.1
D. kraemeri	AB847657.1
D. kratense	AB847658.1
D. laevifolium	AB593593.1
D. lamyaiae	AB593595.1
D. lancifolium	AY239976.1
D. laterale	AB847660.1
D. lawesii	AY239977.1
D. leonis	AY239978.1
D. leptocladum	AB593598.1
D. libingtaoi	KY062492.1
D. linawianum	AB593599.1
D. lindleyi	JN388568.1
D. linguella	AB593601.1
D. lituiflorum	AB593602.1
D. lobbii	AB593603.1
D. loddigesii	HQ114220.1
D. lohohense	JN388574.1
D. longicornu	KF143483.1
D. lowii	AB593606.1
D. luteolum	AB593607.1
D. luzonense	MK522200.1
D. maccarthiae	AB593608.1
D. macfarlanei	AB847662.1

D. macrophyllum	ON694116.1
D. macrostachyum	HM054696.1
D. maguanense	KY062482.1
D. melanostictum	AB593610.1
D. melinanthum	AB593611.1
D. microbulbon	HM054707.1
D. microglaphys	AB593612.1
D. minutiflorum	KF143488.1
D. miyakei	KJ210473.1
D. mohlianum	AY239980.1
D. moniliforme	AB873180.1
D. monophyllum	EU430387.1
D. monticola	DQ058799.1
D. moorei	EU430388.1
D. morrisonii	AY239982.1
D. moschatum	AB593616.1
D. muricatum	LC227999.1
D. mutabile	AY239984.1
D. nanum	MK169315.1
D. nemorale	AB593618.1
D. nindii	AY239985.1
D. nobile	AB593619.1
D. nothofagicola	AY239986.1
D. nutantiflorum	MH763847.1
D. ochreatum	HM054720.1
D. officinale	KC205176.1
D. okinawense	KF143495.1
D. oligophyllum	AB847665.1
D. oppositifolium	LC227998.1
D. ovatum	HM054734.1
D. pachyglossum	AB593623.1
D. pachyphyllum	AB593624.1
D. palpebrae	AB593625.1
D. panduratum	MK169322.1
D. panduriferum	AB847666.1
D. papilio	AY239987.1
D. parciflorum	AB593628.1
D. parcum	AB593629.1
D. parishii	AB593630.1
D. parthenium	AB847668.1
D. patentilobum	AB847669.1
D. peguanum	HM054746.1
D. pendulum	KF143498.1
D. philippinense	AY239988.1
D. platygastrium	AB593635.1
D. plicatile	KY966573.1
D. porphyrochilum	AB593637.1
D. praecinctum	MN213685.1
D. prasinum	AB593639.1
D. prianganense	AB593640.1
D. primulinum	JN388598.1
D. profusum	AB593642.1
D. pseudotenellum	KF143502.1
D. pugioniforme	ON694120.1
D. pulchellum	KJ210492.1
D. quadrilobatum	KC507778.1
D. quinquelobum	ON694123.1
D. racemosum	EU430389.1
D. rarum	AB593644.1
D. regium	AB593645.1
D. rhombeum	AB593646.1
D. rigidifolium	ON694115.1

D. roseiodorum	AB593648.1
D. roseipes	AB593649.1
D. rosellum	AB593650.1
D. rotundatum	KX522631.1
D. ruckeri	KF143504.1
D. salaccense	JN388577.1
D. sanderae	AB593654.1
D. sanguinolentum	AB593655.1
D. scabrilingue	AB593656.1
D. schoeninum	EU430390.1
D. schuetzei	AB593658.1
D. scoriarum	MG779633.1
D. secundum	AY239993.1
D. senile	AB593661.1
D. serratilabium	AY239994.1
D. setifolium	KC507763.1
D. shiraishii	AB894132.1
D. shixingense	MG779628.1
D. signatum	AB593662.1
D. sinense	KF143510.1
D. singaporense	KC507765.1
D. singkawangense	AB593663.1
D. sinuatum	AY239995.1
D. smillieae	AB593664.1
D. somae	MK522202.1
D. spatella	KF143513.1
D. speciosum	AY239998.1
D. speckmaieri	AB593665.1
D. spectabile	AB894141.1
D. spectatissimum	AB593666.1
D. stockeri	AB593667.1
D. stratiotes	ON694121.1
D. striolatum	ON694119.1
D. strongylanthum	KJ210501.1
D. stuposum	JN388599.1
D. subulatum	KC507766.1
D. subuliferum	AY240000.1
D. sulcatum	KF143517.1
D. sutepense	AB593671.1
D. suzukii	AB593672.1
D. taurinum	AB894142.1
D. terminale	MK522256.1
D. tetrachromum	AB847672.1
D. thyrsiflorum	KT778758.1
D. tipuliferum	MH766907.1
D. tobaense	AB593677.1
D. tortile	AB593678.1
D. trankimianum	AB847673.1
D. transparens	KF143520.1
D. trantuanii	AB847674.1
D. trichostomum	AB593680.1
D. trigonopus	KF143522.1
D. trinervium	KY966588.1
D. truncatum	AY240002.1
D. tuensangense	MW807365.1
D. unicum	MK522213.1
D. uniflorum	AB593683.1
D. usterioides	AY240003.1
D. venustum	AB847676.1
D. victoriae-reginae	AB593684.1
D. violaceum	AB593685.1
D. virgineum	AB972347.1

D. wangliangii	KF143524.1
D. wardianum	KT778762.1
D. wattii	KF143525.1
D. williamsonii	ON694113.1
D. wilsonii	MK522247.1
D. xichouense	KJ210514.1
D. yeageri	AY240006.1
D. ypsilon	AB593688.1
D. zhenghuoense	KX530078.1
Cadetia maideniana	AY239948.1
Cannaeorchis fractiflexa	AY239949.1
Diplocaulobium ischnopet	AY240007.1
Dockrillia calamiformis	AY240008.1
Epigeneium clemensiae	KF143530.1
Epigeneium cymbidioides	AY240011.1
Epigeneium nakaharaei	AY240012.1
Epigeneium tricallosum	JF706721.1
Epigeneium triflorum	AY240013.1
Flickingeria angustifolia	KF143532.1
Flickingeria bicolor	AY485720.1
Flickingeria comata	HM590389.1
Winika cunninghamii	AY240019.1
Bulbophyllum inunctum	JF428110.1
Bulbophyllum macranthu	EF195933.1

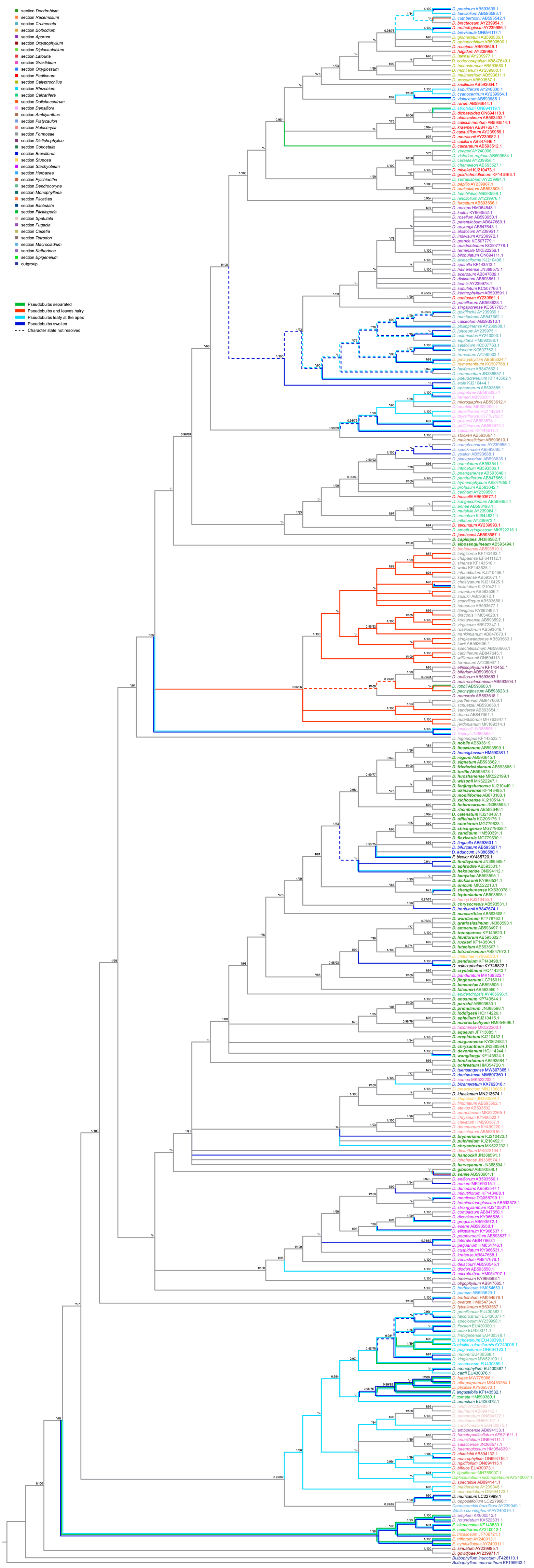


Fig. S1. Ancestral state reconstruction of morphological characters (pseudobulbs) of *Dendrobium* taxa (according to Table S2).

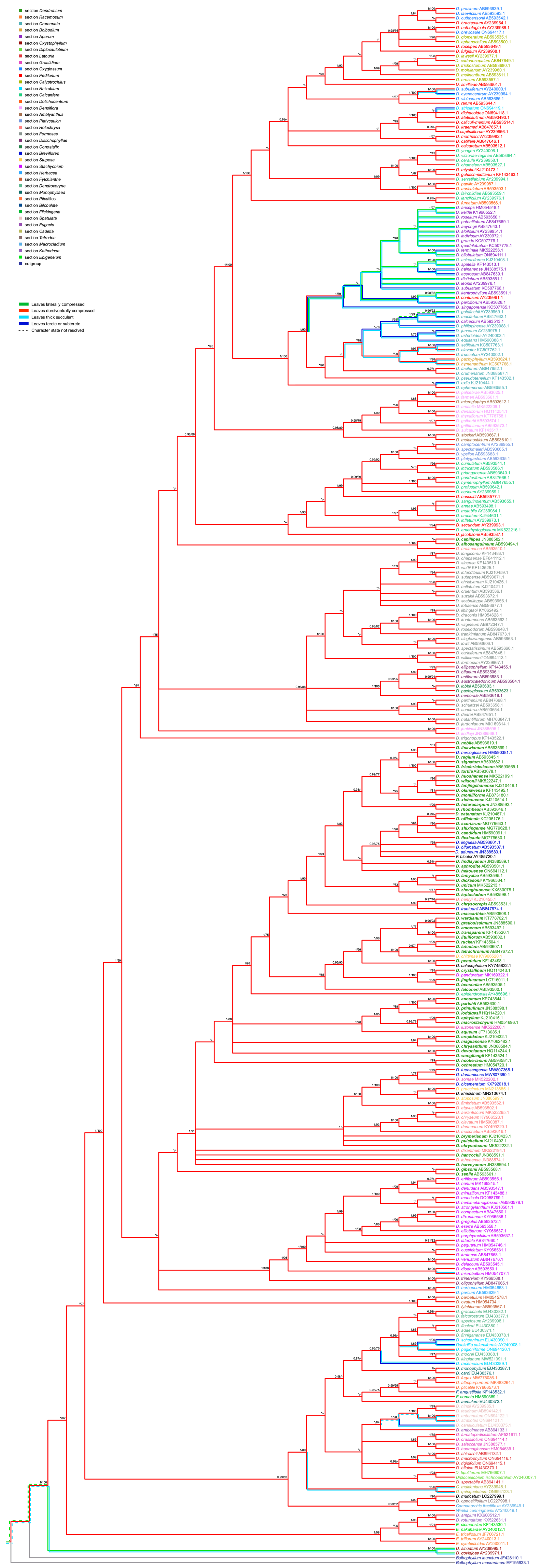


Fig. S2. Ancestral state reconstruction of morphological characters (leaves) of *Dendrobium* taxa (according to Table S2).

section *Dendrobium*
section *Racemosum*
section *Crumenata*
section *Bolbodium*
section *Aporum*
section *Oxystophyllum*
section *Diplocaulobium*
section *Latouria*
section *Grastidium*
section *Oxyglossum*
section *Pedilonum*
section *Calyptrochilus*
section *Rhizobium*
section *Calcarifera*
section *Dolichocentrum*
section *Densiflora*
section *Amblyanthus*
section *Platycaulon*
section *Holochrysa*
section *Formosae*
section *Distichophyllae*
section *Conostalix*
section *Breviflores*
section *Stuposa*
section *Stachyobium*
section *Herbacea*
section *Fytchianthe*
section *Dendrocoryne*
section *Monophyllaea*
section *Plicatiles*
section *Bilobulate*
section *Flickingeria*
section *Spatulata*
section *Fugacia*
section *Cadetia*
section *Tetrodon*
section *Macrocladium*
section *Katherinea*
section *Epigeneium*
outgroup

Inflorescence elongate
Inflorescence produced on last years pseudobulbs
Inflorescence placed laterally along pseudobulbs
Character state not resolved

Fig. S3. Ancestral state reconstruction of morphological characters (inflorescence) of *Dendrobium* taxa (according to Table S2).

section *Dendrobium*
section *Racemosum*
section *Crumenata*
section *Bolbodium*
section *Aporum*
section *Oxystophyllum*
section *Diplocaulobium*
section *Latouria*
section *Grastidium*
section *Oxyglossum*
section *Pedilonum*
section *Calyptrochilus*
section *Rhizobium*
section *Calcarifera*
section *Dolichocentrum*
section *Densiflora*
section *Amblyanthus*
section *Platycaulon*
section *Holochrysa*
section *Formosae*
section *Distichophyllae*
section *Conostalix*
section *Breviflores*
section *Stuposa*
section *Stachyobium*
section *Herbacea*
section *Fytchianthe*
section *Dendrocoryne*
section *Monophyllaea*
section *Plicatiles*
section *Bilobulate*
section *Flickingeria*
section *Spatulata*
section *Fugacia*
section *Cadetia*
section *Tetrodon*
section *Macrocladium*
section *Katherinea*
section *Epigeneium*
outgroup

Lip surface tomentose papillose, glandular to hairy
Lip 3-lobed
Mentum as long as or longer than dorsal sepal
Lip callus absent
Character state not resolved

1 - Himalaya
2 - India / Sri Lanka
3 - Burma, Indochina, Sumatra, Java, Borneo & Philippines
4 - China, Korea & Japan
5 - Australia, New Zeland, New Guinea & Celebes

Fig. S4. Ancestral state reconstruction of morphological characters (lip and mentum) of *Dendrobium* taxa (according to Table S2).Circles next to taxon names indicate geographic distribution.

Table S2. Data matrix used for ancestral state reconstruction of morphological features, where taxa characters were coded for the presence (1, 2 - yes) or absence (0 - no) of a feature.

Species	pseudobulbs (0-approximate vs. 1-separated)	pseudobulbs & leaves (0-glabrous vs. 1-hairy)	pseudobulbs (0-leafy throughout vs. 1-leafy at the apex)	pseudobulbs (0-reedlike vs. 1-swollen)	leaves (0-dorsiventrally vs. 1-laterally compressed)	leaves thick succulent (0-no vs. 1-yes)	inflorescence (0-short vs. 1-elongate)	inflorescence produced on (0-current vs. 1-last year pseudobulbs)	inflorescence places (0-apically on vs. 1-laterally along) pseudobulbs	flowers (0-glabrous vs. 1-more or less hairy outside)	lip (0-unlobed vs. 1-3-lobed)	lip surface (0-glabrous, papillate or wrinkled vs. 1-tomentose, papillose, glandular to hairy)	lip callus (0-prominent, present vs. 1-missing)	mentum (0-shorter vs. 1-as long as or longer) than dorsal sepal
Dendrobium nobile	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium linawianum	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium officinale	0	0	0	0	0	0	0	1	1	0	0	1	0	0
Dendrobium shixingense	0	0	0	0	0	0	0	1	1	0	0	1	0	0
Dendrobium regium	0	0	0	0	0	0	0	1	1	0	0	1	0	0
Dendrobium wardianum	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium friedericksianum	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium signatum	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium tortile	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium huoshanense	0	0	0	0	0	0	0	1	1	0	0	1	0	0
Dendrobium fanjingshanense	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium moniliforme	0	0	0	0	0	0	0	1	1	0	0	1	0	0
Dendrobium wilsonii	0	0	0	0	0	0	0	1	1	0	0	1	0	0
Dendrobium okinawense	0	0	0	0	0	0	0	1	1	0	0	1	0	0
Dendrobium xichouense	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium heterocarpum	0	0	0	0	0	0	0	1	1	0	0	1	0	0
Dendrobium devonianum	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium capillipes	0	0	0	0	0	0	1	1	0	0	0	1	1	0
Dendrobium rhombeum	0	0	0	0	0	0	0	1	1	0	0	1	0	0
Dendrobium scoriarum	0	0	0	0	0	0	0	1	1	0	0	1	0	0
Dendrobium flexicaule	0	0	0	0	0	0	0	1	1	0	0	1	0	0
Dendrobium primulinum	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium bensoniae	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium brymerianum	0	0	0	1	0	0	1	1	0	0	1	1	0	0
Dendrobium gratiosissimum	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium hekouense	0	0	1	1	0	0	0	1	1	0	1	1	0	1
Dendrobium findlayanum	0	0	0	1	0	0	0	1	1	0	0	1	1	0
Dendrobium aphrodite	0	0	0	1	0	0	0	1	1	0	0	1	1	0
Dendrobium leptocladum	0	0	0	0	0	0	0	1	1	0	0	1	0	0
Dendrobium zhenghuoense	0	0	1	1	0	0	1	1	0	0	0	1	1	1
Dendrobium lamyaiae	0	0	0	0	0	0	0	1	0	0	1	0	0	0
Dendrobium unicum	0	0	0	0	0	0	0	1	1	0	0	0	0	0
Dendrobium dickasonii	0	0	0	0	0	0	0	1	1	0	0	0	0	0
Dendrobium loddigesii	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium chrysocrepis	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium maccarthiae	0	0	0	0	0	0	0	1	1	0	0	1	0	0
Dendrobium lituiflorum	0	0	0	0	0	0	0	1	1	0	0	0	1	0
Dendrobium amoenum	0	0	0	0	0	0	0	1	1	0	0	1	0	0
Dendrobium transparens	0	0	0	0	0	0	0	1	1	0	0	1	0	0
Dendrobium ruckeri	0	0	0	0	0	0	0	1	1	0	1	1	0	1
Dendrobium luteolum	0	0	0	0	0	0	0	1	1	0	1	1	0	1
Dendrobium tetrachromum	0	0	0	0	0	0	0	1	1	0	1	0	0	0
Dendrobium pendulum	0	0	0	1	0	0	0	1	1	0	0	1	1	0
Dendrobium crystallinum	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium jinghuanum	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium falconeri	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium parishii	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium anosmum	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium aphyllum	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium ochreatum	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium chrysanthum	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium macrostachyum	0	0	0	0	0	0	0	1	1	0	0	1	0	0
Dendrobium aqueum	0	0	0	0	0	0	0	1	1	0	0	1	0	0
Dendrobium wangliangii	0	0	1	1	0	0	1	1	0	0	0	1	1	0
Dendrobium crepidatum	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium gibsonii	0	0	0	0	0	0	1	1	1	0	0	1	1	0
Dendrobium pulchellum	0	0	0	0	0	0	1	1	1	0	0	1	1	0
Dendrobium chrysotoxum	0	0	1	0	0	0	1	1	0	0	0	1	1	0
Dendrobium harveyanum	0	0	0	0	0	0	1	1	1	0	0	1	1	0
Dendrobium hookerianum	0	0	0	0	0	0	1	1	0	0	0	1	1	0
Dendrobium hancockii	0	0	0	1	0	0	1	0	0	0	1	1	0	0
Dendrobium senile	0	1	1	1	0	0	1	1	0	0	0	0	0	0
Dendrobium albosanguineum	0	0	0	0	0	0	1	1	0	0	0	0	1	0
Dendrobium hercoglossum	0	0	0	0	0	0	1	1	1	0	1	1	0	0
Dendrobium aduncum	0	0	0	0	0	0	0	1	1	0	1	1	0	0
Dendrobium linguella	0	0	0	0	0	0	0	1	1	0	1	1	0	0
Dendrobium bifurcatum	0	0	0	0	0	0	1	1	1	0	1	1	0	1
Dendrobium trantuanii	0	0	0	1	0	0	0	1	1	0	1	1	0	0
Dendrobium bicameratum	0	0	1	0	0	0	0	1	1	1	1	0	0	0
Dendrobium tuensangense	0	0	0	0	0	0	0	1	1	0	1	1	0	0
Dendrobium dantaniense	0	0	0	0	0	0	0	1	1	0	1	0	0	0
Dendrobium moschatum	0	0	0	0	0	0	1	1	0	0	0	1	1	0
Dendrobium fimbriatum	0	0	0	0	0	0	1	1	0	0	0	1	1	0
Dendrobium clavatum	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium henryi	0	0	0	0	0	0	0	1	1	0	0	1	1	0
Dendrobium chryseum	0	0	0	0	0	0	1	1	0	0	0	1	1	0
Dendrobium atavus	0	0	0	0	0	0	1	1	0	0	0	1	1	0
Dendrobium denneanum	0	0	0	0	0	0	1	1	0	0	0	1	1	0
Dendrobium lohohense	0	0	0	0	0	0	0	1	0	0	0	1	1	0
Dendrobium dixanthum	0	0	0	0	0	0	1	1	0	0	0	1	1	0
Dendrobium braianense	0	0	0	0	0	0	1	1	1	0	0	1	1	0
Dendrobium parcum	0	0	0	0	0	0	0	1	1	0	0	0	0	1
Dendrobium herbaceum	0	0	0	0	0	0	0	1	0	0	0	0	0	0
Dendrobium minutiflorum	0	0	0	1	0	0	1	0	0	0	1	0	0	1

Dendrobium panduratum	0	0	0	0	0	0	1	0	0	0	1	1	0	1
Dendrobium porphyrochilum	0	0	0	0	0	0	1	0	0	0	0	0	0	1
Dendrobium nanum	0	0	0	1	0	0	1	0	0	0	1	1	0	0
Dendrobium eriiflorum	0	0	0	0	0	0	1	0	0	0	1	0	0	0
Dendrobium strongylanthum	0	0	0	0	0	0	1	0	0	0	1	0	0	0
Dendrobium denudans	0	0	0	0	0	0	1	0	0	0	1	0	0	0
Dendrobium hemimelanoglossum	0	0	0	0	0	0	1	1	0	0	0	0	0	0
Dendrobium monticola	0	0	1	1	0	0	1	0	0	0	1	1	0	0
Dendrobium compactum	0	0	0	0	0	0	1	0	1	0	1	0	0	0
Dendrobium dixonianum	0	0	0	0	0	0	1	0	0	0	1	0	0	0
Dendrobium gregulus	0	0	1	1	0	0	1	0	0	0	1	0	0	0
Dendrobium eserre	0	0	0	0	0	0	1	0	0	0	1	0	0	0
Dendrobium elliottianum	0	0	1	1	0	0	1	0	0	0	1	0	0	1
Dendrobium peguanum	0	0	0	1	0	0	1	1	0	0	1	1	0	1
Dendrobium laterale	0	0	1	1	0	0	1	0	0	0	1	1	0	0
Dendrobium cuspidatum	0	0	0	0	0	0	0	0	0	0	1	0	0	0
Dendrobium kratense	0	0	0	0	0	0	1	1	1	0	1	0	0	0
Dendrobium venustum	0	0	0	0	0	0	1	0	0	0	1	0	0	0
Dendrobium delacourii	0	0	0	0	0	0	1	0	0	0	1	0	0	0
Dendrobium microbulbon	0	0	1	1	0	1	1	0	0	0	1	0	0	0
Dendrobium diodon	0	0	1	0	0	0	1	0	0	0	1	0	0	0
Dendrobium intricatum	0	0	0	0	0	0	0	1	1	0	0	0	1	1
Dendrobium lancifolium	0	0	0	0	0	0	0	1	1	0	0	1	1	1
Dendrobium fairchildiae	0	0	0	0	0	0	0	1	1	0	0	1	0	1
Dendrobium epidendropsis	0	0	0	0	0	0	1	1	1	0	0	0	0	1
Dendrobium cumulatum	0	0	0	0	0	0	1	1	1	0	0	0	0	1
Dendrobium victoriae-reginae	0	0	0	0	0	0	0	1	1	0	0	0	1	0
Dendrobium serratilabium	0	0	0	0	0	0	0	1	1	0	1	0	0	1
Dendrobium chameleon	0	0	0	0	0	0	0	1	1	0	1	0	0	1
Dendrobium yeageri	0	0	0	0	0	0	0	1	1	0	0	0	1	0
Dendrobium ceraula	0	0	0	0	0	0	0	1	1	0	0	0	1	1
Dendrobium prianganense	0	0	0	0	0	0	0	1	1	0	0	1	0	0
Dendrobium panduriferum	0	0	0	0	0	0	0	1	1	0	1	0	0	1
Dendrobium hymenophyllum	0	0	0	0	0	0	1	1	1	0	1	0	0	1
Dendrobium profusum	0	0	0	0	0	0	1	1	1	0	1	0	0	1
Dendrobium cerinum	0	0	0	0	0	0	1	1	1	0	0	0	1	1
Dendrobium mutabile	0	0	0	0	0	0	1	1	0	0	1	0	0	0
Dendrobium sanguinolentum	0	0	0	0	0	0	0	1	0	0	1	0	0	0
Dendrobium annae	0	0	0	0	0	0	0	1	1	0	0	0	1	0
Dendrobium crocatum	0	0	0	0	0	0	0	1	0	0	0	0	1	1
Dendrobium inflatum	0	0	0	0	0	0	0	1	1	0	0	0	1	1
Dendrobium amethystoglossum	0	0	0	0	0	0	1	0	0	0	0	1	0	1
Dendrobium ellipsophyllum	0	0	0	0	0	0	0	0	1	0	1	0	0	0
Dendrobium bifarium	0	0	0	0	0	0	0	0	1	0	1	1	0	0
Dendrobium uniflorum	0	0	0	0	0	0	0	0	1	0	1	0	0	0
Dendrobium austrocaledonicum	0	0	0	0	0	0	0	0	1	0	1	0	0	0
Dendrobium nemorale	0	0	0	0	0	0	0	0	1	0	1	0	0	0
Dendrobium oligophyllum	0	0	0	0	0	0	0	0	1	0	1	1	0	0
Dendrobium trinervium	0	0	0	0	0	0	0	0	1	0	1	1	0	0
Dendrobium sulcatum	0	0	1	1	0	0	1	0	0	0	0	1	1	0
Dendrobium jenkinsii	0	0	1	1	0	0	0	0	0	0	0	1	1	0
Dendrobium thyrsiflorum	0	0	1	0	0	0	1	0	0	0	0	1	1	0
Dendrobium densiflorum	0	0	1	0	0	0	1	1	0	0	0	1	1	0
Dendrobium griffithianum	0	0	1	1	0	0	1	0	0	0	0	1	1	0
Dendrobium farmeri	0	0	1	1	0	0	1	0	0	0	0	1	1	0
Dendrobium palpebrae	0	0	1	0	0	0	1	0	0	0	0	1	1	0
Dendrobium amabile	0	0	0	0	0	0	1	0	0	0	0	1	1	0
Dendrobium guibertii	0	0	1	1	0	0	1	0	0	0	0	1	1	0
Dendrobium lindleyi	0	0	1	1	0	0	1	0	0	0	0	1	1	0
Dendrobium draconis	0	1	0	0	0	0	0	0	0	0	1	0	1	1
Dendrobium trigonopus	0	0	0	0	0	0	0	1	0	0	1	0	1	1
Dendrobium cariniferum	0	1	0	0	0	0	0	0	0	0	1	1	0	1
Dendrobium bellatulum	0	1	1	1	0	0	0	0	0	0	1	1	0	0
Dendrobium williamsonii	0	1	0	0	0	0	0	0	0	0	1	1	0	0
Dendrobium longicornu	0	1	0	0	0	0	0	0	0	0	1	1	0	1
Dendrobium formosum	0	1	0	0	0	0	0	0	0	0	1	1	0	1
Dendrobium christyanum	0	1	0	0	0	0	0	0	0	0	1	1	0	1
Dendrobium infundibulum	0	1	0	0	0	0	0	0	0	0	1	0	0	1
Dendrobium wattii	0	1	0	0	0	0	0	0	0	0	1	0	0	1
Dendrobium sutepense	0	0	0	0	0	0	0	1	0	0	1	0	0	0
Dendrobium scabrilingue	0	1	0	0	0	0	0	0	0	0	1	0	0	0
Dendrobium cruentum	0	1	0	0	0	0	0	0	0	0	1	0	0	0
Dendrobium suzukii	0	1	0	0	0	0	0	0	0	0	1	1	0	0
Dendrobium tobaense	0	1	0	0	0	0	0	0	0	0	1	1	0	0
Dendrobium singkawangense	0	1	0	0	0	0	0	0	0	0	1	1	0	0
Dendrobium lowii	0	1	0	0	0	0	0	0	0	0	1	1	0	1
Dendrobium spectatissimum	0	1	0	0	0	0	0	0	0	0	1	0	0	1
Dendrobium virgineum	0	1	0	0	0	0	0	0	0	0	1	0	0	1
Dendrobium kontumense	0	1	0	0	0	0	0	0	0	0	1	0	0	1
Dendrobium roseiodorum	0	1	0	0	0	0	0	0	0	0	1	1	0	1
Dendrobium trankimianum	0	1	0	0	0	0	0	0	0	0	1	0	0	0
Dendrobium dearei	0	1	0	0	0	0	0	0	0	0	1	0	1	1
Dendrobium jerdonianum	0	1	0	0	0	0	0	0	0	0	1	0	0	0
Dendrobium nutantiflorum	0	1	0	0	0	0	0	0	0	0	1	0	0	0
Dendrobium sanderae	0	1	0	0	0	0	0	0	0	0	1	0	1	1
Dendrobium schuetzei	0	0	0	0	0	0	0	0	0	0	1	0	1	1
Dendrobium libingtaoi	0	0	0	0	0	0	0	0	1	0	0	0	1	0
Dendrobium chapaense	0	1	0	0	0	0	0	1	0	0	1	0	0	1
Dendrobium parthenium	0	0	0	0	0	0	0	0	0	0	1	0	1	1
Dendrobium stuposum	0	0	0	0	0	0	0	1	0	0	1	1	0	0
Dendrobium chittimae	0	0	0	0	0	0	0	0	0	0	1	1	0	1
Dendrobium praecinctum	0	0	0	0	0	0	0	1	0	0	1	1	0	0
Dendrobium acinaciforme	0	0	0	0	1	1	0	0	0	0	1	0	0	1
Dendrobium exile	0	0	0	1	2	1	0	0	0	0	1	1	0	1
Dendrobium equitans	0	0	0	0	2	1	0	0	0	0	1	1	0	0

Dendrobium junceum	0	0	0	1	2	1	0	0	0	0	1	0	0	1
Dendrobium usterioides	0	0	1	1	2	1	1	0	0	0	1	0	0	1
Dendrobium macfarlanei	0	0	0	0	2	1	1	0	0	0	1	0	0	1
Dendrobium goldfinchii	0	0	1	1	1	1	1	0	0	0	0	0	0	1
Dendrobium philippinense	0	0	1	1	2	1	0	0	0	0	1	0	0	1
Dendrobium truncatum	0	0	0	0	0	0	0	0	0	0	1	1	0	1
Dendrobium setifolium	0	0	1	1	2	1	0	0	0	0	1	0	0	1
Dendrobium clavator	0	0	1	1	0	1	0	0	0	0	1	0	0	1
Dendrobium crumenatum	0	0	0	0	0	0	0	0	0	0	1	0	0	1
Dendrobium faciferum	0	0	1	1	0	0	0	1	0	0	1	0	0	1
Dendrobium pseudotenellum	0	0	1	1	0	0	0	0	0	0	1	0	0	1
Dendrobium ephemerum	0	0	1	1	0	0	0	1	0	0	1	0	0	1
Dendrobium salaccense	0	0	0	0	0	0	0	0	1	0	0	0	0	0
Dendrobium luzonense	0	0	0	0	0	0	0	0	1	0	0	0	0	0
Dendrobium furcatopedicellatum	0	0	0	0	0	0	0	0	1	0	1	0	0	0
Dendrobium somae	0	0	0	0	0	0	0	0	1	0	1	0	0	0
Dendrobium crassifolium	0	0	0	0	0	0	0	0	1	0	1	1	0	0
Dendrobium erosum	0	0	0	0	0	0	0	1	0	0	0	1	1	1
Dendrobium aphanochilum	0	0	0	0	0	0	0	1	0	0	0	1	1	1
Dendrobium glomeratum	0	0	0	0	0	0	0	1	0	0	0	1	1	1
Dendrobium lawesii	0	0	0	0	0	0	0	1	0	0	0	1	1	1
Dendrobium codonosepalum	0	0	0	0	0	0	0	1	0	0	0	1	1	1
Dendrobium mohlianum	0	0	0	0	0	0	0	1	0	0	0	1	1	1
Dendrobium melinanthum	0	0	0	0	0	0	0	1	0	0	0	1	1	1
Dendrobium trichostomum	0	0	0	0	0	0	0	1	0	0	0	1	1	1
Dendrobium hainanense	0	0	0	0	2	1	0	0	0	0	0	0	1	1
Dendrobium parciflorum	0	0	0	0	2	1	0	0	0	0	1	1	0	1
Dendrobium terminale	0	0	0	0	2	1	0	0	0	0	0	1	0	1
Dendrobium spatella	0	0	0	0	1	1	0	0	0	0	1	0	0	1
Dendrobium aloifolium	0	0	0	0	1	1	0	0	0	0	1	0	0	1
Dendrobium kentrophyllum	0	0	0	0	1	1	0	0	0	0	1	0	0	1
Dendrobium acerosum	0	0	0	0	2	1	0	0	0	0	0	0	0	1
Dendrobium distichum	0	0	0	0	1	1	0	0	0	0	1	0	0	1
Dendrobium leonis	0	0	0	0	1	1	0	0	0	0	1	1	1	1
Dendrobium grande	0	0	0	0	1	1	0	0	0	0	1	0	0	1
Dendrobium rosellum	0	0	0	0	1	1	0	0	0	0	1	0	0	1
Dendrobium quadrilobatum	0	0	0	0	1	1	0	0	0	0	1	0	0	1
Dendrobium auyongii	0	0	0	0	1	1	0	0	0	0	1	0	0	1
Dendrobium anceps	0	0	0	0	1	1	0	0	0	0	0	0	0	1
Dendrobium keithii	0	0	0	0	1	1	0	0	0	0	0	0	1	1
Dendrobium indivisum	0	0	0	0	1	1	0	0	0	0	1	0	0	1
Dendrobium patentilobum	0	0	0	0	1	1	0	0	0	0	1	0	0	1
Dendrobium singaporense	0	0	0	0	2	1	0	0	0	0	1	1	0	1
Dendrobium bilobulatum	0	0	0	0	1	1	0	0	0	0	0	0	0	1
Dendrobium subulatum	0	0	0	0	2	1	0	0	0	0	0	0	0	1
Dendrobium calceolum	0	0	0	0	1	1	0	0	0	0	1	0	0	1
Dendrobium goldschmidtianum	0	0	0	0	0	0	0	1	1	0	0	0	1	1
Dendrobium secundum	0	0	0	0	0	0	1	0	0	0	0	0	1	1
Dendrobium calcaratum	1	0	0	0	0	0	1	1	1	0	1	0	0	1
Dendrobium dichaeoides	0	0	0	0	0	0	1	1	0	0	0	0	1	1
Dendrobium alaticaulinum	0	0	0	0	0	0	1	1	0	0	0	0	1	1
Dendrobium caliculi-mentum	0	0	0	0	0	0	1	1	0	0	0	0	1	1
Dendrobium morrisonii	0	0	0	0	0	0	0	1	1	0	0	0	1	1
Dendrobium catillare	0	0	0	0	0	0	0	1	1	0	0	0	1	1
Dendrobium capituliflorum	0	0	0	0	0	0	0	1	0	0	0	0	1	1
Dendrobium kraemeri	0	0	0	0	0	0	0	1	1	0	0	0	1	1
Dendrobium bracteosum	0	0	0	0	0	0	0	1	1	0	0	0	1	1
Dendrobium nothofagicola	0	0	1	1	0	0	0	0	0	0	0	0	1	1
Dendrobium roseipes	0	0	0	0	0	0	0	1	0	0	0	0	1	1
Dendrobium fulgidum	0	0	0	0	0	0	0	1	0	0	0	0	1	1
Dendrobium smillieae	0	0	0	0	0	0	1	1	0	0	0	0	1	1
Dendrobium rarum	0	0	0	0	0	0	1	1	0	0	0	0	1	0
Dendrobium jacobsonii	0	0	0	0	0	0	0	1	1	0	0	0	1	1
Dendrobium hasseltii	0	0	0	0	0	0	1	1	0	0	0	0	1	1
Dendrobium stratiotes	0	0	0	0	0	1	1	0	0	0	1	0	1	0
Dendrobium antennatum	0	0	0	0	0	1	1	0	0	0	1	0	1	0
Dendrobium nindii	0	0	0	0	0	0	1	0	0	0	1	0	0	0
Dendrobium taurinum	0	0	0	0	0	0	1	0	0	0	1	0	0	1
Dendrobium canaliculatum	0	0	1	1	2	1	1	0	0	0	1	0	0	0
Dendrobium pachyphyllum	0	0	1	1	0	1	0	0	0	0	0	0	1	1
Dendrobium hymenanthum	0	0	1	0	0	1	0	0	0	0	1	0	0	1
Dendrobium papilio	0	0	0	0	0	0	0	1	0	0	1	0	1	0
Dendrobium auriculatum	0	0	0	0	0	0	0	1	0	0	0	0	0	0
Dendrobium furcatum	0	0	0	0	0	0	0	1	0	0	0	0	1	0
Dendrobium striolatum	1	0	1	0	2	1	1	0	0	0	1	0	0	0
Dendrobium schoeninum	1	0	1	0	2	1	1	0	0	0	1	0	0	0
Dockrillia calamiformis	1	0	1	0	2	1	1	0	0	0	1	0	0	0
Dendrobium pugioniforme	1	0	1	0	0	1	0	0	0	0	1	0	0	0
Dendrobium racemosum	1	0	1	0	2	1	1	0	0	0	1	0	0	0
Dendrobium cuthbertsonii	0	1	1	0	0	0	0	0	0	0	0	0	1	1
Dendrobium laevifolium	0	0	1	0	0	0	0	0	0	0	0	0	0	1
Dendrobium prasinum	0	0	1	1	0	0	0	0	0	0	0	0	0	1
Dendrobium brevicaule	0	0	1	0	0	0	0	0	0	0	0	0	1	1
Dendrobium violaceum	0	0	1	1	0	0	0	1	0	0	0	0	0	1
Dendrobium cyanocentrum	0	0	1	0	2	1	0	0	0	0	0	0	0	1
Dendrobium subuliferum	0	0	1	0	0	1	0	0	0	0	0	0	0	0
Epigeneium nakaharaei	0	0	1	1	0	0	0	0	0	0	1	0	0	0
Epigeneium clemensiae	0	0	1	1	0	0	0	0	0	0	1	0	0	0
Dendrobium microglaphys	0	0	1	0	0	0	0	0	0	0	0	0	0	0
Dendrobium melanostictum	0	0	0	0	0	0	0	0	1	0	0	1	1	0
Dendrobium stockeri	0	0	0	0	0	0	0	0	1	0	1	1	0	0
Dendrobium speckmaieri	0	0	0	1	0	0	0	1	1	0	0	0	0	0
Dendrobium ypsilon	0	0	0	1	0	0	0	0	0	0	0	0	0	0
Dendrobium platygastrium	0	0	0	0	0	0	1	0	0	0	0	0	1	0
Dendrobium lobbii	1	1	0	0	0	0	0	0	1	0	1	0	0	0

Dendrobium pachyglossum	0	1	0	0	0	0	0	0	1	0	1	0	0	0
Flickingeria comata	0	0	1	1	0	0	0	0	0	0	1	0	0	0
Dendrobium ovatum	0	0	0	0	0	0	1	0	0	0	1	1	0	0
Dendrobium barbatulum	0	0	1	1	0	0	1	1	1	0	1	1	0	0
Dendrobium fytchianum	0	0	0	0	0	0	1	1	0	0	1	1	0	0
Dendrobium speciosum	0	0	1	1	0	0	1	1	0	0	1	0	0	0
Dendrobium kingianum	0	0	1	1	0	0	1	0	0	0	1	0	0	0
Dendrobium moorei	0	0	1	1	0	0	1	0	0	0	1	0	0	0
Dendrobium fleckeri	0	0	1	1	0	0	0	0	0	0	1	1	0	0
Dendrobium adae	0	0	1	1	0	0	1	1	0	0	1	0	0	0
Dendrobium finniganense	0	0	1	0	0	0	0	0	0	0	1	1	0	0
Dendrobium falcorostrum	0	0	1	1	0	0	1	0	0	0	1	0	0	0
Dendrobium gracilicaule	0	0	1	0	0	0	1	0	0	0	1	0	0	0
Dendrobium aemulum	0	0	1	0	0	0	1	0	0	0	1	0	0	0
Dendrobium carrii	0	0	1	0	0	0	1	0	0	0	1	0	0	0
Dendrobium monophyllum	0	0	1	1	0	0	1	0	0	0	1	0	0	0
Dendrobium fugax	1	0	1	1	0	0	0	0	0	0	1	0	0	0
Dendrobium albopurpureum	1	0	1	1	0	0	0	0	0	0	1	0	0	0
Dendrobium plicatile	1	0	1	1	0	0	0	0	0	0	1	0	0	0
Flickingeria angustifolia	1	0	1	1	0	0	0	0	0	0	1	0	0	0
Dendrobium amboinense	0	0	1	1	0	0	0	0	0	0	1	0	0	0
Dendrobium macrophyllum	0	0	1	1	0	0	1	0	1	1	1	0	0	0
Dendrobium rigidifolium	0	0	1	0	0	1	1	0	0	0	0	0	0	0
Dendrobium shiraishii	0	0	1	0	0	0	1	0	0	1	1	0	0	0
Dendrobium spectabile	0	0	1	0	0	0	1	0	0	0	1	0	0	0
Dendrobium bifalce	0	0	1	0	0	0	1	0	0	0	1	0	0	0
Dendrobium govidjoae	0	0	0	0	1	1	0	0	0	0	0	0	0	0
Dendrobium sinuatum	0	0	0	0	1	1	0	0	0	0	0	0	1	0
Dendrobium quinquelobum	0	0	1	0	0	1	0	0	0	0	1	0	0	0
Cadetia maideniana	0	0	1	0	0	1	0	0	0	0	1	0	0	0
Dendrobium oppositifolium	0	0	1	1	0	0	1	0	0	0	1	0	0	0
Cannaeorchis fractiflexa	0	0	0	0	0	0	1	0	1	0	0	0	0	0
Winika cunninghamii	0	0	0	0	0	0	1	0	1	0	1	0	0	0
Dendrobium amplum	1	0	1	1	0	0	0	0	0	0	1	0	0	0
Dendrobium rotundatum	1	0	1	1	0	0	0	0	0	0	1	0	0	0
Epigeneium triflorum	1	0	1	1	0	0	1	0	0	0	1	0	0	0
Epigeneium tricallosum	1	0	1	1	0	0	0	0	0	0	1	0	0	0
Epigeneium cymbidioides	1	0	1	1	0	0	1	0	0	0	1	0	0	0
Dendrobium tipuliferum	0	0	1	0	0	0	0	0	0	0	1	0	0	0
Diplocaulobium ischnopetalum	0	0	1	0	0	0	0	0	0	0	1	0	0	0
Flickingeria bicolor	0	0	1	1	0	0	0	0	0	0	1	0	0	0
Dendrobium calocephalum	0	0	1	1	0	0	0	0	0	0	1	0	0	0
Dendrobium khasianum	0	0	0	0	0	0	0	0	0	0	0	1	1	0
Dendrobium muricatum	0	0	1	1	0	0	1	0	0	0	1	0	0	0

C1 Pseudobulbs
- approximate
- separated

C2 Pseudobulbs and leaves
- glabrous
- hairy

C3 Pseudobulbs
- leafy throughout
- leafy at apex

C4 Pseudobulbs
- reed-like
- swollen

C5 Leaves
- dorsiventrally compressed
- laterally compressed
- terete or subterete

C6 Leaves thick, succulent
- no
- yes

C7 Inflorescence
- short
- elongate

C8 Inflorescence produced
- on current pseudobulbs
- on last year's pseudobulbs

C9 Inflorescence placed
- apically on pseudobulbs
- laterally along psedobulbs

C10 Flowers
- glabrous
- more or less hairy outside

C11 Lip
- unlobed
- 3-lobed

C12 Lip surface
- glabrous, papillate or wrinkled

C13 Lip callus
- prominent, present
- missing

C14 Mentum
- shorter than dorsal sepal
- as long as or longer than dorsal sepal

- Dendrobium
- Breviflores
- Holochrysa
- Herbacea
- Stachyobium
- Calcarifera
- Distichophyllae
- Densiflora
- Formosae
- Stuposa
- Crumenata
- Grastidium
- Calyptrochilus
- Aporum
- Pedilonum
- Spatulata
- Bolbodium
- Dolichocentrum
- Rhizobium
- Oxyglossum
- Epigeneium
- Amblyanthus
- Platycaulon
- Conostalix
- Flickingeria
- Fytchianthe
- Dendrocoryne
- Monophyllaea
- Plicatiles
- Bilobulate
- Fugacia
- Latouria
- Oxystophyllum
- Cadetia
- Tetrodon
- Macrocladium
- Katherinea
- Racemosum
- Diplocaulobium

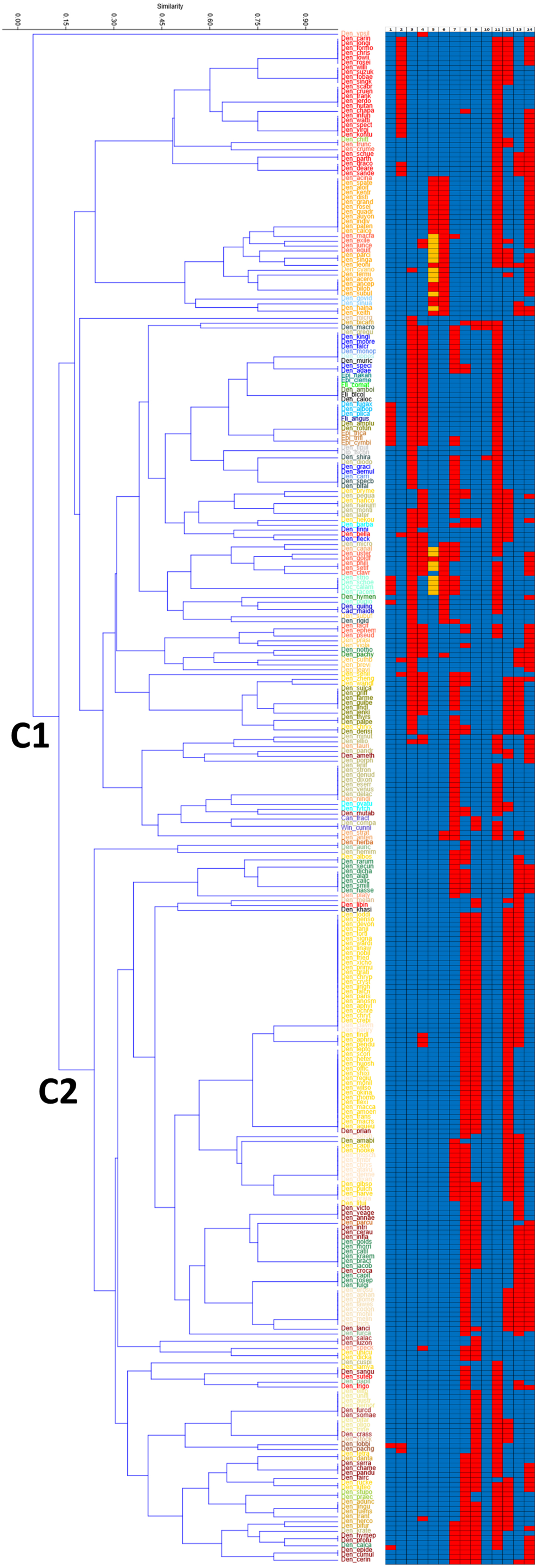


Fig. S5. Two-way UPGMA cluster analysis based on Jaccard similarity coefficients, showing morphological relationships within *Dendrobium sensu lato*, with regard to the division into sections. A detailed description of the morphological characters can be found in Table S3 in the additional file.

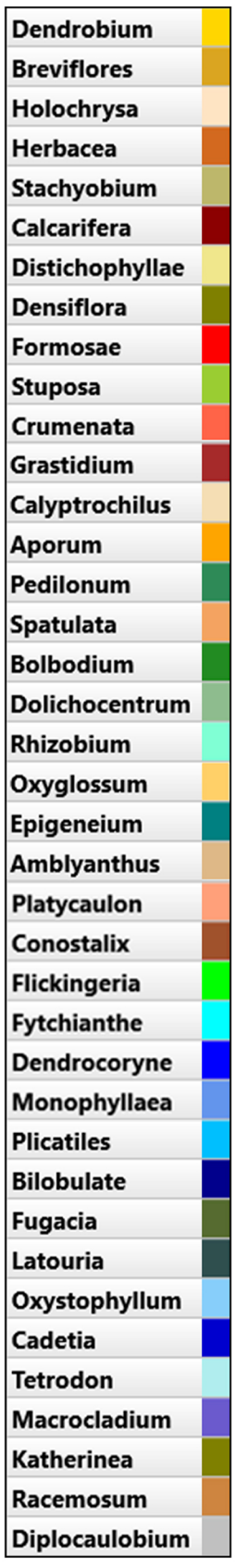
C10 Flowers
- glabrous
- more or less hairy outside

C11 Lip
- unlobed
- 3-lobed

C12 Lip surface
- glabrous, papillate or wrinkled

C13 Lip callus
- prominent, present
- missing

C14 Mentum
- shorter than dorsal sepal
- as long as or longer than dorsal sepal



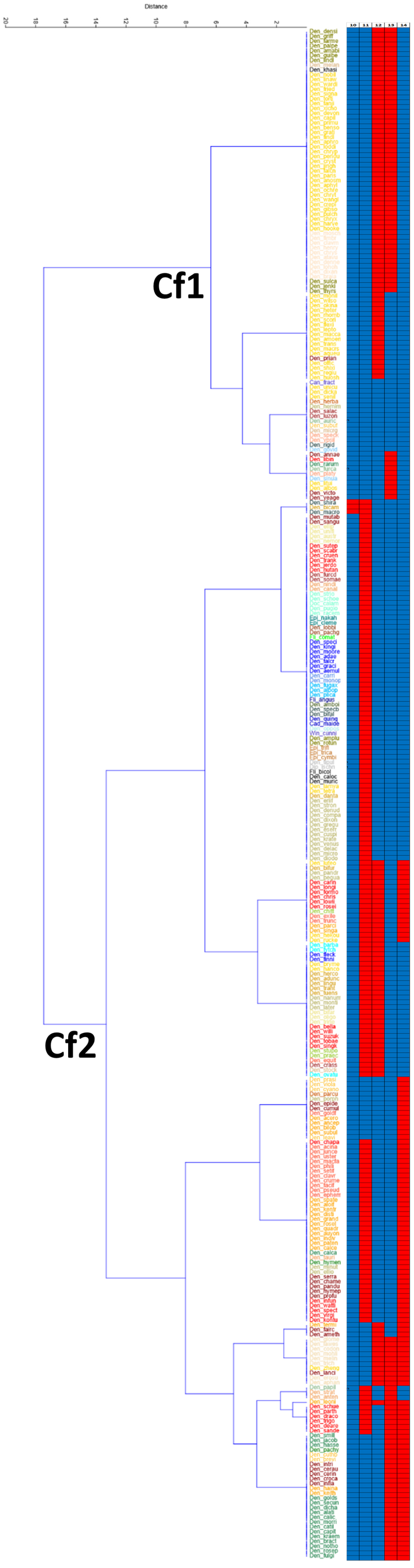


Fig. S6. Two-way cluster analysis conducted using Ward's method, showing relationships within floral traits only for *Dendrobium sensu lato*, with regard to the division into sections. A detailed description of floral characters can be found in Table S3 in the additional file.

Table S3. List of morphological characters used to describe *Dendrobium sensu lato* species included in the cluster and multivariate analyses.

Ch1 Pseudobulbs
- approximate
- separated

Ch2 Pseudobulbs and leaves
- glabrous
- hairy

Ch3 Pseudobulbs
- leafy throughout
- leafy at apex

Ch4 Pseudobulbs
- reed-like
- swollen

Ch5 Leaves
- dorsiventrally compressed
- laterally compressed
- terete or subterete

Ch6 Leaves thick, succulent
- no
- yes

Ch7 Inflorescence
- short
- elongate

Ch8 Inflorescence produced
- on current pseudobulbs
- on last year‘s pseudobulbs

Ch9 Inflorescence placed
- apically on pseudobulbs
- laterally along psedobulbs

Ch10 Flowers
- glabrous
- more or less hairy outside

Ch11 Lip
- unlobed
- 3-lobed

Ch12 Lip surface
- glabrous, papillate or wrinkled
- tomentose, papillose, glandular to hairy

Ch13 Lip callus
- prominent, present
- missing

Ch14 Mentum
- shorter than dorsal sepal
- as long as or longer than dorsal sepal

Table S4. Data matrix of 14 morphological traits used in the analysis of morphological variation in *Dendrobium sensu lato* . The data were transformed according to Table S3 in this supplementary file.

ID	Taxon	Code	Section	1	2	3	4	5	6	7	8	9	10	11	12	13	14
1	*Dendrobium nobile*	Den_nobil	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	1	0
2	*Dendrobium linawianum*	Den_linaw	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	1	0
3	*Dendrobium officinale*	Den_offic	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	0	0
4	*Dendrobium shixingense*	Den_shixi	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	0	0
5	*Dendrobium regium*	Den_regiu	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	0	0
6	*Dendrobium wardianum*	Den_wardi	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	1	0
7	*Dendrobium friedericksianum*	Den_fried	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	1	0
8	*Dendrobium signatum*	Den_signa	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	1	0
9	*Dendrobium tortile*	Den_torti	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	1	0
10	*Dendrobium huoshanense*	Den_huosh	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	0	0
11	*Dendrobium fanjingshanense*	Den_fanji	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	1	0
12	*Dendrobium moniliforme*	Den_monil	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	0	0
13	*Dendrobium wilsonii*	Den_wilso	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	0	0
14	*Dendrobium okinawense*	Den_okina	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	0	0
15	*Dendrobium xichouense*	Den_xicho	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	1	0
16	*Dendrobium heterocarpum*	Den_heter	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	0	0
17	*Dendrobium devonianum*	Den_devon	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	1	0
18	*Dendrobium capillipes*	Den_capil	Dendrobium	0	0	0	0	0	0	1	1	0	0	0	1	1	0
19	*Dendrobium rhombeum*	Den_rhomb	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	0	0
20	*Dendrobium scoriarum*	Den_scori	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	0	0
21	*Dendrobium flexicaule*	Den_flexi	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	0	0
22	*Dendrobium primulinum*	Den_primu	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	1	0
23	*Dendrobium bensoniae*	Den_benso	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	1	0
24	*Dendrobium brymerianum*	Den_bryme	Dendrobium	0	0	0	1	0	0	1	1	0	0	1	1	0	0
25	*Dendrobium gratiosisssimum*	Den_grati	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	1	0
26	*Dendrobium hekouense*	Den_hekou	Dendrobium	0	0	1	1	0	0	0	1	1	0	1	1	0	1
27	*Dendrobium findlayanum*	Den_findl	Dendrobium	0	0	0	1	0	0	0	1	1	0	0	1	1	0
28	*Dendrobium aphrodite*	Den_aphro	Dendrobium	0	0	0	1	0	0	0	1	1	0	0	1	1	0
29	*Dendrobium leptocladum*	Den_lepto	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	0	0
30	*Dendrobium zhenghuoense*	Den_zheng	Dendrobium	0	0	1	1	0	0	1	1	0	0	0	1	1	1
31	*Dendrobium lamyaiae*	Den_lamya	Dendrobium	0	0	0	0	0	0	0	1	0	0	1	0	0	0
32	*Dendrobium unicum*	Den_unicu	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	0	0	0
33	*Dendrobium dickasonii*	Den_dicka	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	0	0	0
34	*Dendrobium loddigesii*	Den_loddi	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	1	0
35	*Dendrobium chrysocrepis*	Den_chryp	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	1	0
36	*Dendrobium maccarthiae*	Den_macca	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	0	0
37	*Dendrobium lituiflorum*	Den_litui	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	0	1	0
38	*Dendrobium amoenum*	Den_amoen	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	0	0
39	*Dendrobium transparens*	Den_trans	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	0	0
40	*Dendrobium ruckeri*	Den_rucke	Dendrobium	0	0	0	0	0	0	0	1	1	0	1	1	0	1
41	*Dendrobium luteolum*	Den_luteo	Dendrobium	0	0	0	0	0	0	0	1	1	0	1	1	0	1
42	*Dendrobium tetrachromum*	Den_tetra	Dendrobium	0	0	0	0	0	0	0	1	1	0	1	0	0	0
43	*Dendrobium pendulum*	Den_pendu	Dendrobium	0	0	0	1	0	0	0	1	1	0	0	1	1	0
44	*Dendrobium crystallinum*	Den_cryst	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	1	0
45	*Dendrobium jinghuanum*	Den_jingh	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	1	0
46	*Dendrobium falconeri*	Den_falcn	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	1	0
47	*Dendrobium parishii*	Den_paris	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	1	0
48	*Dendrobium anosmum*	Den_anosm	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	1	0
49	*Dendrobium aphyllum*	Den_aphyl	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	1	0
50	*Dendrobium ochreatum*	Den_ochre	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	1	0
51	*Dendrobium chrysanthum*	Den_chryt	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	1	0
52	*Dendrobium macrostachyum*	Den_macrs	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	0	0
53	*Dendrobium aqueum*	Den_aqueu	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	0	0
54	*Dendrobium wangliangii*	Den_wangl	Dendrobium	0	0	1	1	0	0	1	1	0	0	0	1	1	0
55	*Dendrobium crepidatum*	Den_crepi	Dendrobium	0	0	0	0	0	0	0	1	1	0	0	1	1	0
56	*Dendrobium gibsonii*	Den_gibso	Dendrobium	0	0	0	0	0	0	1	1	1	0	0	1	1	0
57	*Dendrobium pulchellum*	Den_pulch	Dendrobium	0	0	0	0	0	0	1	1	1	0	0	1	1	0
58	*Dendrobium chrysotoxum*	Den_chryx	Dendrobium	0	0	1	0	0	0	1	1	0	0	0	1	1	0
59	*Dendrobium harveyanum*	Den_harve	Dendrobium	0	0	0	0	0	0	1	1	1	0	0	1	1	0
60	*Dendrobium hookerianum*	Den_hooke	Dendrobium	0	0	0	0	0	0	1	1	0	0	0	1	1	0
61	*Dendrobium hancockii*	Den_hanco	Dendrobium	0	0	0	1	0	0	1	0	0	0	1	1	0	0
62	*Dendrobium senile*	Den_senil	Dendrobium	0	1	1	1	0	0	1	1	0	0	0	0	0	0
63	*Dendrobium albosanguineum*	Den_albos	Dendrobium	0	0	0	0	0	0	1	1	0	0	0	0	1	0
64	*Dendrobium hercoglossum*	Den_herco	Breviflores	0	0	0	0	0	0	1	1	1	0	1	1	0	0
65	*Dendrobium aduncum*	Den_adunc	Breviflores	0	0	0	0	0	0	0	1	1	0	1	1	0	0
66	*Dendrobium linguella*	Den_lingu	Breviflores	0	0	0	0	0	0	0	1	1	0	1	1	0	0
67	*Dendrobium bifurcatum*	Den_bifur	Breviflores	0	0	0	0	0	0	1	1	1	0	1	1	0	1
68	*Dendrobium trantuanii*	Den_trant	Breviflores	0	0	0	1	0	0	0	1	1	0	1	1	0	0
69	*Dendrobium bicameratum*	Den_bicam	Breviflores	0	0	1	0	0	0	0	1	1	1	1	0	0	0
70	*Dendrobium tuensangense*	Den_tuens	Breviflores	0	0	0	0	0	0	0	1	1	0	1	1	0	0
71	*Dendrobium dantaniense*	Den_danta	Breviflores	0	0	0	0	0	0	0	1	1	0	1	0	0	0
72	*Dendrobium moschatum*	Den_mosch	Holochrysa	0	0	0	0	0	0	1	1	0	0	0	1	1	0
73	*Dendrobium fimbriatum*	Den_fimbr	Holochrysa	0	0	0	0	0	0	1	1	0	0	0	1	1	0
74	*Dendrobium clavatum*	Den_clavm	Holochrysa	0	0	0	0	0	0	0	1	1	0	0	1	1	0
75	*Dendrobium henryi*	Den_henry	Holochrysa	0	0	0	0	0	0	0	1	1	0	0	1	1	0
76	*Dendrobium chryseum*	Den_chrys	Holochrysa	0	0	0	0	0	0	1	1	0	0	0	1	1	0
77	*Dendrobium atavus*	Den_atavu	Holochrysa	0	0	0	0	0	0	1	1	0	0	0	1	1	0
78	*Dendrobium denneanum*	Den_denne	Holochrysa	0	0	0	0	0	0	1	1	0	0	0	1	1	0
79	*Dendrobium lohohense*	Den_lohoh	Holochrysa	0	0	0	0	0	0	0	1	0	0	0	1	1	0
80	*Dendrobium dixanthum*	Den_dixan	Holochrysa	0	0	0	0	0	0	1	1	0	0	0	1	1	0
81	*Dendrobium braianense*	Den_braia	Holochrysa	0	0	0	0	0	0	1	1	1	0	0	1	1	0
82	*Dendrobium parcum*	Den_parcu	Herbacea	0	0	0	0	0	0	0	1	1	0	0	0	0	1
83	*Dendrobium herbaceum*	Den_herba	Herbacea	0	0	0	0	0	0	0	1	0	0	0	0	0	0
84	*Dendrobium minutiflorum*	Den_minut	Stachyobium	0	0	0	1	0	0	1	0	0	0	1	0	0	1
85	*Dendrobium panduratum*	Den_pandr	Stachyobium	0	0	0	0	0	0	1	0	0	0	1	1	0	1
86	*Dendrobium porphyrochilum*	Den_porph	Stachyobium	0	0	0	0	0	0	1	0	0	0	0	0	0	1
87	*Dendrobium nanum*	Den_nanum	Stachyobium	0	0	0	1	0	0	1	0	0	0	1	1	0	0
88	*Dendrobium eriiflorum*	Den_eriif	Stachyobium	0	0	0	0	0	0	1	0	0	0	1	0	0	0
89	*Dendrobium strongylanthum*	Den_stron	Stachyobium	0	0	0	0	0	0	1	0	0	0	1	0	0	0
90	*Dendrobium denudans*	Den_denud	Stachyobium	0	0	0	0	0	0	1	0	0	0	1	0	0	0
91	*Dendrobium hemimelanoglossum*	Den_hemim	Stachyobium	0	0	0	0	0	0	1	1	0	0	0	0	0	0
92	*Dendrobium monticola*	Den_monti	Stachyobium	0	0	1	1	0	0	1	0	0	0	1	1	0	0
93	*Dendrobium compactum*	Den_compa	Stachyobium	0	0	0	0	0	0	1	0	1	0	1	0	0	0
94	*Dendrobium dixonianum*	Den_dixon	Stachyobium	0	0	0	0	0	0	1	0	0	0	1	0	0	0
95	*Dendrobium gregulus*	Den_gregu	Stachyobium	0	0	1	1	0	0	1	0	0	0	1	0	0	0
96	*Dendrobium eserre*	Den_eserr	Stachyobium	0	0	0	0	0	0	1	0	0	0	1	0	0	0
97	*Dendrobium elliottianum*	Den_ellio	Stachyobium	0	0	1	1	0	0	1	0	0	0	1	0	0	1
98	*Dendrobium peguanum*	Den_pegua	Stachyobium	0	0	0	1	0	0	1	1	0	0	1	1	0	1
99	*Dendrobium laterale*	Den_later	Stachyobium	0	0	1	1	0	0	1	0	0	0	1	1	0	0
100	*Dendrobium cuspidatum*	Den_cuspi	Stachyobium	0	0	0	0	0	0	0	0	0	0	1	0	0	0
101	*Dendrobium kratense*	Den_krate	Stachyobium	0	0	0	0	0	0	1	1	1	0	1	0	0	0
102	*Dendrobium venustum*	Den_venus	Stachyobium	0	0	0	0	0	0	1	0	0	0	1	0	0	0
103	*Dendrobium delacourii*	Den_delac	Stachyobium	0	0	0	0	0	0	1	0	0	0	1	0	0	0
104	*Dendrobium microbulbon*	Den_micro	Stachyobium	0	0	1	1	0	1	1	0	0	0	1	0	0	0
105	*Dendrobium diodon*	Den_diodo	Stachyobium	0	0	1	0	0	0	1	0	0	0	1	0	0	0
106	*Dendrobium intricatum*	Den_intri	Calcarifera	0	0	0	0	0	0	0	1	1	0	0	0	1	1
107	*Dendrobium lancifolium*	Den_lanci	Calcarifera	0	0	0	0	0	0	0	1	1	0	0	1	1	1
108	*Dendrobium fairchildiae*	Den_fairc	Calcarifera	0	0	0	0	0	0	0	1	1	0	0	1	0	1
109	*Dendrobium epidendropsis*	Den_epide	Calcarifera	0	0	0	0	0	0	1	1	1	0	0	0	0	1
110	*Dendrobium cumulatum*	Den_cumul	Calcarifera	0	0	0	0	0	0	1	1	1	0	0	0	0	1

111	*Dendrobium victoriae-reginae*	Den_victo	Calcarifera	0	0	0	0	0	0	0	1	1	0	0	0	1	0
112	*Dendrobium serratilabium*	Den_serra	Calcarifera	0	0	0	0	0	0	0	1	1	0	1	0	0	1
113	*Dendrobium chameleon*	Den_chame	Calcarifera	0	0	0	0	0	0	0	1	1	0	1	0	0	1
114	*Dendrobium yeageri*	Den_yeage	Calcarifera	0	0	0	0	0	0	0	1	1	0	0	0	1	0
115	*Dendrobium ceraula*	Den_cerau	Calcarifera	0	0	0	0	0	0	0	1	1	0	0	0	1	1
116	*Dendrobium prianganense*	Den_prian	Calcarifera	0	0	0	0	0	0	0	1	1	0	0	1	0	0
117	*Dendrobium pandurifferum*	Den_pandu	Calcarifera	0	0	0	0	0	0	0	1	1	0	1	0	0	1
118	*Dendrobium hymenophyllum*	Den_hymep	Calcarifera	0	0	0	0	0	0	1	1	1	0	1	0	0	1
119	*Dendrobium profusum*	Den_profu	Calcarifera	0	0	0	0	0	0	1	1	1	0	1	0	0	1
120	*Dendrobium cerinum*	Den_cerin	Calcarifera	0	0	0	0	0	0	1	1	1	0	0	0	1	1
121	*Dendrobium mutabile*	Den_mutab	Calcarifera	0	0	0	0	0	0	1	1	0	0	1	0	0	0
122	*Dendrobium sanguinolentum*	Den_sangu	Calcarifera	0	0	0	0	0	0	0	1	0	0	1	0	0	0
123	*Dendrobium annae*	Den_annae	Calcarifera	0	0	0	0	0	0	0	1	1	0	0	0	1	0
124	*Dendrobium crocatum*	Den_croca	Calcarifera	0	0	0	0	0	0	0	1	0	0	0	0	1	1
125	*Dendrobium inflatum*	Den_infla	Calcarifera	0	0	0	0	0	0	0	1	1	0	0	0	1	1
126	*Dendrobium amethystoglossum*	Den_ameth	Calcarifera	0	0	0	0	0	0	1	0	0	0	0	1	0	1
127	*Dendrobium ellipsophyllum*	Den_ellip	Distichophyllae	0	0	0	0	0	0	0	0	1	0	1	0	0	0
128	*Dendrobium bifarium*	Den_bifar	Distichophyllae	0	0	0	0	0	0	0	0	1	0	1	1	0	0
129	*Dendrobium uniflorum*	Den_unifl	Distichophyllae	0	0	0	0	0	0	0	0	1	0	1	0	0	0
130	*Dendrobium austrocaledonicum*	Den_austr	Distichophyllae	0	0	0	0	0	0	0	0	1	0	1	0	0	0
131	*Dendrobium nemorale*	Den_nemor	Distichophyllae	0	0	0	0	0	0	0	0	1	0	1	0	0	0
132	*Dendrobium oligophyllum*	Den_oligo	Distichophyllae	0	0	0	0	0	0	0	0	1	0	1	1	0	0
133	*Dendrobium trinervium*	Den_trine	Distichophyllae	0	0	0	0	0	0	0	0	1	0	1	1	0	0
134	*Dendrobium sulcatum*	Den_sulca	Densiflora	0	0	1	1	0	0	1	0	0	0	0	1	1	0
135	*Dendrobium jenkinsii*	Den_jenki	Densiflora	0	0	1	1	0	0	0	0	0	0	0	1	1	0
136	*Dendrobium thyrsiflorum*	Den_thyrs	Densiflora	0	0	1	0	0	0	1	0	0	0	0	1	1	0
137	*Dendrobium densiflorum*	Den_densi	Densiflora	0	0	1	0	0	0	1	1	0	0	0	1	1	0
138	*Dendrobium griffithianum*	Den_griff	Densiflora	0	0	1	1	0	0	1	0	0	0	0	1	1	0
139	*Dendrobium farmeri*	Den_farme	Densiflora	0	0	1	1	0	0	1	0	0	0	0	1	1	0
140	*Dendrobium palpebrae*	Den_palpe	Densiflora	0	0	1	0	0	0	1	0	0	0	0	1	1	0
141	*Dendrobium amabile*	Den_amabi	Densiflora	0	0	0	0	0	0	1	0	0	0	0	1	1	0
142	*Dendrobium guibertii*	Den_guibe	Densiflora	0	0	1	1	0	0	1	0	0	0	0	1	1	0
143	*Dendrobium lindleyi*	Den_lindl	Densiflora	0	0	1	1	0	0	1	0	0	0	0	1	1	0
144	*Dendrobium draconis*	Den_draco	Formosae	0	1	0	0	0	0	0	0	0	0	1	0	1	1
145	*Dendrobium trigonopus*	Den_trigo	Formosae	0	0	0	0	0	0	0	1	0	0	1	0	1	1
146	*Dendrobium cariniferum*	Den_carin	Formosae	0	1	0	0	0	0	0	0	0	0	1	1	0	1
147	*Dendrobium bellatulum*	Den_bella	Formosae	0	1	1	1	0	0	0	0	0	0	1	1	0	0
148	*Dendrobium williamsonii*	Den_willi	Formosae	0	1	0	0	0	0	0	0	0	0	1	1	0	0
149	*Dendrobium longicornu*	Den_longi	Formosae	0	1	0	0	0	0	0	0	0	0	1	1	0	1
150	*Dendrobium formosum*	Den_formo	Formosae	0	1	0	0	0	0	0	0	0	0	1	1	0	1
151	*Dendrobium christyanum*	Den_chris	Formosae	0	1	0	0	0	0	0	0	0	0	1	1	0	1
152	*Dendrobium infundibulum*	Den_infun	Formosae	0	1	0	0	0	0	0	0	0	0	1	0	0	1
153	*Dendrobium wattii*	Den_watti	Formosae	0	1	0	0	0	0	0	0	0	0	1	0	0	1
154	*Dendrobium sutepense*	Den_sutep	Formosae	0	0	0	0	0	0	0	1	0	0	1	0	0	0
155	*Dendrobium scabrilingue*	Den_scabr	Formosae	0	1	0	0	0	0	0	0	0	0	1	0	0	0
156	*Dendrobium cruentum*	Den_cruen	Formosae	0	1	0	0	0	0	0	0	0	0	1	0	0	0
157	*Dendrobium suzukii*	Den_suzuk	Formosae	0	1	0	0	0	0	0	0	0	0	1	1	0	0
158	*Dendrobium tobaense*	Den_tobae	Formosae	0	1	0	0	0	0	0	0	0	0	1	1	0	0
159	*Dendrobium singkawangense*	Den_singk	Formosae	0	1	0	0	0	0	0	0	0	0	1	1	0	0
160	*Dendrobium lowii*	Den_lowii	Formosae	0	1	0	0	0	0	0	0	0	0	1	1	0	1
161	*Dendrobium spectatissimum*	Den_spect	Formosae	0	1	0	0	0	0	0	0	0	0	1	0	0	1
162	*Dendrobium virgineum*	Den_virgi	Formosae	0	1	0	0	0	0	0	0	0	0	1	0	0	1
163	*Dendrobium kontumense*	Den_kontu	Formosae	0	1	0	0	0	0	0	0	0	0	1	0	0	1
164	*Dendrobium roseiodorum*	Den_rosei	Formosae	0	1	0	0	0	0	0	0	0	0	1	1	0	1
165	*Dendrobium trankimianum*	Den_trank	Formosae	0	1	0	0	0	0	0	0	0	0	1	0	0	0
166	*Dendrobium dearei*	Den_deare	Formosae	0	1	0	0	0	0	0	0	0	0	1	0	1	1
167	*Dendrobium jerdonianum*	Den_jerdo	Formosae	0	1	0	0	0	0	0	0	0	0	1	0	0	0
168	*Dendrobium nutantiflorum*	Den_nutan	Formosae	0	1	0	0	0	0	0	0	0	0	1	0	0	0
169	*Dendrobium sanderae*	Den_sande	Formosae	0	1	0	0	0	0	0	0	0	0	1	0	1	1
170	*Dendrobium schuetzei*	Den_schue	Formosae	0	0	0	0	0	0	0	0	0	0	1	0	1	1
171	*Dendrobium libingtaoi*	Den_libin	Formosae	0	0	0	0	0	0	0	0	1	0	0	0	1	0
172	*Dendrobium chapaense*	Den_chapa	Formosae	0	1	0	0	0	0	0	1	0	0	1	0	0	1
173	*Dendrobium parthenium*	Den_parth	Formosae	0	0	0	0	0	0	0	0	0	0	1	0	1	1
174	*Dendrobium stuposum*	Den_stupo	Stuposa	0	0	0	0	0	0	0	1	0	0	1	1	0	0
175	*Dendrobium chittimae*	Den_chitt	Stuposa	0	0	0	0	0	0	0	0	0	0	1	1	0	1
176	*Dendrobium praecinctum*	Den_praec	Stuposa	0	0	0	0	0	0	0	1	0	0	1	1	0	0
177	*Dendrobium acinaciforme*	Den_acina	Crumenata	0	0	0	0	1	1	0	0	0	0	1	0	0	1
178	*Dendrobium exile*	Den_exile	Crumenata	0	0	0	1	2	1	0	0	0	0	1	1	0	1
179	*Dendrobium equitans*	Den_equit	Crumenata	0	0	0	0	2	1	0	0	0	0	1	1	0	0
180	*Dendrobium junceum*	Den_junce	Crumenata	0	0	0	1	2	1	0	0	0	0	1	0	0	1
181	*Dendrobium usterioides*	Den_uster	Crumenata	0	0	1	1	2	1	1	0	0	0	1	0	0	1
182	*Dendrobium macfarlanei*	Den_macfa	Crumenata	0	0	0	0	2	1	1	0	0	0	1	0	0	1
183	*Dendrobium goldfinchii*	Den_goldf	Crumenata	0	0	1	1	1	1	1	0	0	0	0	0	0	1
184	*Dendrobium philippinense*	Den_phili	Crumenata	0	0	1	1	2	1	0	0	0	0	1	0	0	1
185	*Dendrobium truncatum*	Den_trunc	Crumenata	0	0	0	0	0	0	0	0	0	0	1	1	0	1
186	*Dendrobium setifolium*	Den_setif	Crumenata	0	0	1	1	2	1	0	0	0	0	1	0	0	1
187	*Dendrobium clavator*	Den_clavr	Crumenata	0	0	1	1	0	1	0	0	0	0	1	0	0	1
188	*Dendrobium crumenatum*	Den_crume	Crumenata	0	0	0	0	0	0	0	0	0	0	1	0	0	1
189	*Dendrobium faciferum*	Den_facif	Crumenata	0	0	1	1	0	0	0	1	0	0	1	0	0	1
190	*Dendrobium pseudotenellum*	Den_pseud	Crumenata	0	0	1	1	0	0	0	0	0	0	1	0	0	1
191	*Dendrobium ephemerum*	Den_ephem	Crumenata	0	0	1	1	0	0	0	1	0	0	1	0	0	1
192	*Dendrobium salaccense*	Den_salac	Grastidium	0	0	0	0	0	0	0	0	1	0	0	0	0	0
193	*Dendrobium luzonense*	Den_luzon	Grastidium	0	0	0	0	0	0	0	0	1	0	0	0	0	0
194	*Dendrobium furcatopedicellatum*	Den_furcd	Grastidium	0	0	0	0	0	0	0	0	1	0	1	0	0	0
195	*Dendrobium somae*	Den_somae	Grastidium	0	0	0	0	0	0	0	0	1	0	1	0	0	0
196	*Dendrobium crassifolium*	Den_crass	Grastidium	0	0	0	0	0	0	0	0	1	0	1	1	0	0
197	*Dendrobium erosum*	Den_erosu	Calyptrochilus	0	0	0	0	0	0	0	1	0	0	0	1	1	1
198	*Dendrobium aphanochilum*	Den_aphan	Calyptrochilus	0	0	0	0	0	0	0	1	0	0	0	1	1	1
199	*Dendrobium glomeratum*	Den_glome	Calyptrochilus	0	0	0	0	0	0	0	1	0	0	0	1	1	1
200	*Dendrobium lawesii*	Den_lawes	Calyptrochilus	0	0	0	0	0	0	0	1	0	0	0	1	1	1
201	*Dendrobium codonosepalum*	Den_codon	Calyptrochilus	0	0	0	0	0	0	0	1	0	0	0	1	1	1
202	*Dendrobium mohlianum*	Den_mohli	Calyptrochilus	0	0	0	0	0	0	0	1	0	0	0	1	1	1
203	*Dendrobium melinanthum*	Den_melin	Calyptrochilus	0	0	0	0	0	0	0	1	0	0	0	1	1	1
204	*Dendrobium trichostomum*	Den_trich	Calyptrochilus	0	0	0	0	0	0	0	1	0	0	0	1	1	1
205	*Dendrobium hainanense*	Den_haina	Aporum	0	0	0	0	2	1	0	0	0	0	0	0	1	1
206	*Dendrobium parciflorum*	Den_parci	Aporum	0	0	0	0	2	1	0	0	0	0	1	1	0	1
207	*Dendrobium terminale*	Den_termi	Aporum	0	0	0	0	2	1	0	0	0	0	0	1	0	1
208	*Dendrobium spatella*	Den_spate	Aporum	0	0	0	0	1	1	0	0	0	0	1	0	0	1
209	*Dendrobium aloifolium*	Den_aloif	Aporum	0	0	0	0	1	1	0	0	0	0	1	0	0	1
210	*Dendrobium kentrophyllum*	Den_kentr	Aporum	0	0	0	0	1	1	0	0	0	0	1	0	0	1
211	*Dendrobium acerosum*	Den_acero	Aporum	0	0	0	0	2	1	0	0	0	0	0	0	0	1
212	*Dendrobium distichum*	Den_disti	Aporum	0	0	0	0	1	1	0	0	0	0	1	0	0	1
213	*Dendrobium leonis*	Den_leoni	Aporum	0	0	0	0	1	1	0	0	0	0	1	1	1	1
214	*Dendrobium grande*	Den_grand	Aporum	0	0	0	0	1	1	0	0	0	0	1	0	0	1
215	*Dendrobium rosellum*	Den_rosel	Aporum	0	0	0	0	1	1	0	0	0	0	1	0	0	1
216	*Dendrobium quadrilobatum*	Den_quadr	Aporum	0	0	0	0	1	1	0	0	0	0	1	0	0	1
217	*Dendrobium auyongii*	Den_auyon	Aporum	0	0	0	0	1	1	0	0	0	0	1	0	0	1
218	*Dendrobium anceps*	Den_ancep	Aporum	0	0	0	0	1	1	0	0	0	0	0	0	0	1
219	*Dendrobium keithii*	Den_keith	Aporum	0	0	0	0	1	1	0	0	0	0	0	0	1	1
220	*Dendrobium indivisum*	Den_indiv	Aporum	0	0	0	0	1	1	0	0	0	0	1	0	0	1
221	*Dendrobium patentilobum*	Den_paten	Aporum	0	0	0	0	1	1	0	0	0	0	1	0	0	1
222	*Dendrobium singaporense*	Den_singa	Aporum	0	0	0	0	2	1	0	0	0	0	1	1	0	1
223	*Dendrobium bilobulatum*	Den_bilob	Aporum	0	0	0	0	1	1	0	0	0	0	0	0	0	1

224	*Dendrobium subulatum*	Den_subul	Aporum	0	0	0	0	2	1	0	0	0	0	0	0	0	1
225	*Dendrobium calceolum*	Den_calce	Aporum	0	0	0	0	1	1	0	0	0	0	1	0	0	1
226	*Dendrobium goldschmidtianum*	Den_golds	Pedilonum	0	0	0	0	0	0	0	1	1	0	0	0	1	1
227	*Dendrobium secundum*	Den_secun	Pedilonum	0	0	0	0	0	0	1	0	0	0	0	0	1	1
228	*Dendrobium calcaratum*	Den_calca	Pedilonum	1	0	0	0	0	0	1	1	1	0	1	0	0	1
229	*Dendrobium dichaeoides*	Den_dicha	Pedilonum	0	0	0	0	0	0	1	1	0	0	0	0	1	1
230	*Dendrobium alaticaulinum*	Den_alati	Pedilonum	0	0	0	0	0	0	1	1	0	0	0	0	1	1
231	*Dendrobium caliculi-mentum*	Den_calic	Pedilonum	0	0	0	0	0	0	1	1	0	0	0	0	1	1
232	*Dendrobium morrisonii*	Den_morri	Pedilonum	0	0	0	0	0	0	0	1	1	0	0	0	1	1
233	*Dendrobium catillare*	Den_catil	Pedilonum	0	0	0	0	0	0	0	1	1	0	0	0	1	1
234	*Dendrobium capituliflorum*	Den_capit	Pedilonum	0	0	0	0	0	0	0	1	0	0	0	0	1	1
235	*Dendrobium kraemeri*	Den_kraem	Pedilonum	0	0	0	0	0	0	0	1	1	0	0	0	1	1
236	*Dendrobium bracteosum*	Den_bract	Pedilonum	0	0	0	0	0	0	0	1	1	0	0	0	1	1
237	*Dendrobium nothofagicola*	Den_notho	Pedilonum	0	0	1	1	0	0	0	0	0	0	0	0	1	1
238	*Dendrobium roseipes*	Den_rosep	Pedilonum	0	0	0	0	0	0	0	1	0	0	0	0	1	1
239	*Dendrobium fulgidum*	Den_fulgi	Pedilonum	0	0	0	0	0	0	0	1	0	0	0	0	1	1
240	*Dendrobium smillieae*	Den_smill	Pedilonum	0	0	0	0	0	0	1	1	0	0	0	0	1	1
241	*Dendrobium rarum*	Den_rarum	Pedilonum	0	0	0	0	0	0	1	1	0	0	0	0	1	0
242	*Dendrobium jacobsonii*	Den_jacob	Pedilonum	0	0	0	0	0	0	0	1	1	0	0	0	1	1
243	*Dendrobium hasseltii*	Den_hasse	Pedilonum	0	0	0	0	0	0	1	1	0	0	0	0	1	1
244	*Dendrobium stratiotes*	Den_strat	Spatulata	0	0	0	0	0	1	1	0	0	0	1	0	1	0
245	*Dendrobium antennatum*	Den_anten	Spatulata	0	0	0	0	0	1	1	0	0	0	1	0	1	0
246	*Dendrobium nindii*	Den_nindi	Spatulata	0	0	0	0	0	0	1	0	0	0	1	0	0	0
247	*Dendrobium taurinum*	Den_tauri	Spatulata	0	0	0	0	0	0	1	0	0	0	1	0	0	1
248	*Dendrobium canaliculatum*	Den_canal	Spatulata	0	0	1	1	2	1	1	0	0	0	1	0	0	0
249	*Dendrobium pachyphyllum*	Den_pachy	Bolbodium	0	0	1	1	0	1	0	0	0	0	0	0	1	1
250	*Dendrobium hymenanthum*	Den_hymen	Bolbodium	0	0	1	0	0	1	0	0	0	0	1	0	0	1
251	*Dendrobium papilio*	Den_papil	Dolichocentrum	0	0	0	0	0	0	0	1	0	0	1	0	1	0
252	*Dendrobium auriculatum*	Den_auric	Dolichocentrum	0	0	0	0	0	0	0	1	0	0	0	0	0	0
253	*Dendrobium furcatum*	Den_furca	Dolichocentrum	0	0	0	0	0	0	0	1	0	0	0	0	1	0
254	*Dendrobium striolatum*	Den_strio	Rhizobium	1	0	1	0	2	1	1	0	0	0	1	0	0	0
255	*Dendrobium schoeninum*	Den_schoe	Rhizobium	1	0	1	0	2	1	1	0	0	0	1	0	0	0
256	*Dockrillia calamiformis*	Doc_calam	Rhizobium	1	0	1	0	2	1	1	0	0	0	1	0	0	0
257	*Dendrobium pugioniforme*	Den_pugio	Rhizobium	1	0	1	0	0	1	0	0	0	0	1	0	0	0
258	*Dendrobium racemosum*	Den_racem	Rhizobium	1	0	1	0	2	1	1	0	0	0	1	0	0	0
259	*Dendrobium cuthbertsonii*	Den_cuthb	Oxyglossum	0	1	1	0	0	0	0	0	0	0	0	0	1	1
260	*Dendrobium laevifolium*	Den_laevi	Oxyglossum	0	0	1	0	0	0	0	0	0	0	0	0	0	1
261	*Dendrobium prasinum*	Den_prasi	Oxyglossum	0	0	1	1	0	0	0	0	0	0	0	0	0	1
262	*Dendrobium brevicaule*	Den_brevi	Oxyglossum	0	0	1	0	0	0	0	0	0	0	0	0	1	1
263	*Dendrobium violaceum*	Den_viola	Oxyglossum	0	0	1	1	0	0	0	1	0	0	0	0	0	1
264	*Dendrobium cyanocentrum*	Den_cyano	Oxyglossum	0	0	1	0	2	1	0	0	0	0	0	0	0	1
265	*Dendrobium subuliferum*	Den_subuf	Oxyglossum	0	0	1	0	0	1	0	0	0	0	0	0	0	0
266	*Epigeneium nakaharaei*	Epi_nakah	Epigeneium	0	0	1	1	0	0	0	0	0	0	1	0	0	0
267	*Epigeneium clemensiae*	Epi_cleme	Epigeneium	0	0	1	1	0	0	0	0	0	0	1	0	0	0
268	*Dendrobium microglaphys*	Den_micrg	Amblyanthus	0	0	1	0	0	0	0	0	0	0	0	0	0	0
269	*Dendrobium melanostictum*	Den_melan	Amblyanthus	0	0	0	0	0	0	0	0	1	0	0	1	1	0
270	*Dendrobium stockeri*	Den_stock	Amblyanthus	0	0	0	0	0	0	0	0	1	0	1	1	0	0
271	*Dendrobium speckmaieri*	Den_speck	Platycaulon	0	0	0	1	0	0	0	1	1	0	0	0	0	0
272	*Dendrobium ypsilon*	Den_ypsil	Platycaulon	0	0	0	1	0	0	0	0	0	0	0	0	0	0
273	*Dendrobium platygastrium*	Den_platy	Platycaulon	0	0	0	0	0	0	1	0	0	0	0	0	1	0
274	*Dendrobium lobbii*	Den_lobbi	Conostalix	1	1	0	0	0	0	0	0	1	0	1	0	0	0
275	*Dendrobium pachyglossum*	Den_pachg	Conostalix	0	1	0	0	0	0	0	0	1	0	1	0	0	0
276	*Flickingeria comata*	Fli_comat	Flickingeria	0	0	1	1	0	0	0	0	0	0	1	0	0	0
277	*Dendrobium ovatum*	Den_ovatu	Fytchianthe	0	0	0	0	0	0	1	0	0	0	1	1	0	0
278	*Dendrobium barbatulum*	Den_barba	Fytchianthe	0	0	1	1	0	0	1	1	1	0	1	1	0	0
279	*Dendrobium fytchianum*	Den_fytch	Fytchianthe	0	0	0	0	0	0	1	1	0	0	1	1	0	0
280	*Dendrobium speciosum*	Den_speci	Dendrocoryne	0	0	1	1	0	0	1	1	0	0	1	0	0	0
281	*Dendrobium kingianum*	Den_kingi	Dendrocoryne	0	0	1	1	0	0	1	0	0	0	1	0	0	0
282	*Dendrobium moorei*	Den_moore	Dendrocoryne	0	0	1	1	0	0	1	0	0	0	1	0	0	0
283	*Dendrobium fleckeri*	Den_fleck	Dendrocoryne	0	0	1	1	0	0	0	0	0	0	1	1	0	0
284	*Dendrobium adae*	Den_adae	Dendrocoryne	0	0	1	1	0	0	1	1	0	0	1	0	0	0
285	*Dendrobium finniganense*	Den_finni	Dendrocoryne	0	0	1	0	0	0	0	0	0	0	1	1	0	0
286	*Dendrobium falcorostrum*	Den_falcr	Dendrocoryne	0	0	1	1	0	0	1	0	0	0	1	0	0	0
287	*Dendrobium gracilicaule*	Den_graci	Dendrocoryne	0	0	1	0	0	0	1	0	0	0	1	0	0	0
288	*Dendrobium aemulum*	Den_aemul	Dendrocoryne	0	0	1	0	0	0	1	0	0	0	1	0	0	0
289	*Dendrobium carrii*	Den_carri	Monophyllaea	0	0	1	0	0	0	1	0	0	0	1	0	0	0
290	*Dendrobium monophyllum*	Den_monop	Monophyllaea	0	0	1	1	0	0	1	0	0	0	1	0	0	0
291	*Dendrobium fugax*	Den_fugax	Plicatiles	1	0	1	1	0	0	0	0	0	0	1	0	0	0
292	*Dendrobium albopurpureum*	Den_albop	Plicatiles	1	0	1	1	0	0	0	0	0	0	1	0	0	0
293	*Dendrobium plicatile*	Den_plica	Plicatiles	1	0	1	1	0	0	0	0	0	0	1	0	0	0
294	*Flickingeria angustifolia*	Fli_angus	Bilobulate	1	0	1	1	0	0	0	0	0	0	1	0	0	0
295	*Dendrobium amboinense*	Den_amboi	Fugacia	0	0	1	1	0	0	0	0	0	0	1	0	0	0
296	*Dendrobium macrophyllum*	Den_macro	Latouria	0	0	1	1	0	0	1	0	1	1	1	0	0	0
297	*Dendrobium rigidifolium*	Den_rigid	Latouria	0	0	1	0	0	1	1	0	0	0	0	0	0	0
298	*Dendrobium shiraishii*	Den_shira	Latouria	0	0	1	0	0	0	1	0	0	1	1	0	0	0
299	*Dendrobium spectabile*	Den_specb	Latouria	0	0	1	0	0	0	1	0	0	0	1	0	0	0
300	*Dendrobium bifalce*	Den_bifal	Latouria	0	0	1	0	0	0	1	0	0	0	1	0	0	0
301	*Dendrobium govidjoae*	Den_govid	Oxystophyllum	0	0	0	0	1	1	0	0	0	0	0	0	0	0
302	*Dendrobium sinuatum*	Den_sinua	Oxystophyllum	0	0	0	0	1	1	0	0	0	0	0	0	1	0
303	*Dendrobium quinquelobum*	Den_quinq	Cadetia	0	0	1	0	0	1	0	0	0	0	1	0	0	0
304	*Cadetia maideniana*	Cad_maide	Cadetia	0	0	1	0	0	1	0	0	0	0	1	0	0	0
305	*Dendrobium oppositifolium*	Den_oppos	Tetrodon	0	0	1	1	0	0	1	0	0	0	1	0	0	0
306	*Cannaeorchis fractiflexa*	Can_fract	Macrocladium	0	0	0	0	0	0	1	0	1	0	0	0	0	0
307	*Winika cunninghamii*	Win_cunni	Macrocladium	0	0	0	0	0	0	1	0	1	0	1	0	0	0
308	*Dendrobium amplum*	Den_amplu	Katherinea	1	0	1	1	0	0	0	0	0	0	1	0	0	0
309	*Dendrobium rotundatum*	Den_rotun	Katherinea	1	0	1	1	0	0	0	0	0	0	1	0	0	0
310	*Epigeneium triflorum*	Epi_trifl	Racemosum	1	0	1	1	0	0	1	0	0	0	1	0	0	0
311	*Epigeneium tricallosum*	Epi_trica	Racemosum	1	0	1	1	0	0	0	0	0	0	1	0	0	0
312	*Epigeneium cymbidioides*	Epi_cymbi	Racemosum	1	0	1	1	0	0	1	0	0	0	1	0	0	0
313	*Dendrobium tipuliferum*	Den_tipul	Diplocaulobium	0	0	1	0	0	0	0	0	0	0	1	0	0	0
314	*Diplocaulobium ischnopetalum*	Dip_ischn	Diplocaulobium	0	0	1	0	0	0	0	0	0	0	1	0	0	0
315	*Flickingeria bicolor*	Fli_bicol	unspecified	0	0	1	1	0	0	0	0	0	0	1	0	0	0
316	*Dendrobium calocephalum*	Den_caloc	unspecified	0	0	1	1	0	0	0	0	0	0	1	0	0	0
317	*Dendrobium khasianum*	Den_khasi	unspecified	0	0	0	0	0	0	0	0	0	0	0	1	1	0
318	*Dendrobium muricatum*	Den_muric	unspecified	0	0	1	1	0	0	1	0	0	0	1	0	0	0

Table S5. SIMPER analysis identifying the percentage of similarity and dissimilarity of all morphological traits studied (A) and only floral traits (B) for each section of *Dendrobium sensu lato* . For a detailed description of morphological characters is provided in Table S3 in this supplementary file.
The average changes in the percentage of each section of *Dendrobium sensu lato* in differences in the Bray-Curtis dissimilarity matrix are also reported.

A)

Characters	Av. dissim.	Contrib. %	Cumulat. %	Mean Dendrobium	Mean Breviflores	Mean Holochrysa	Mean Herbacea	Mean Stachyobium	Mean Calcarifera	Mean Distichophyllae	Mean Densiflora	Mean Formosae	Mean Stuposa	Mean Crumenata	Mean Grastidium	Mean Calyptrochilus	Mean Aporum	Mean Pedilonum	Mean Spatulata	Mean Bolbodium	Mean Dolichocentrum	Mean Rhizobium
8	7.433	10.75	10.75	0.984	1	1	1	0.136	0.952	0	0.1	0.1	0.667	0.133	0	1	0	0.889	0	0	1	0
11	7.405	10.70	21.45	0.111	1	0	0	0.909	0.333	1	0	0.967	1	0.933	0.6	0	0.667	0.0556	1	0.5	0.333	1
12	6.961	10.06	31.51	0.889	0.75	1	0	0.227	0.19	0.429	1	0.367	1	0.2	0.2	1	0.19	0	0	0	0	0
9	6.767	9.781	41.29	0.841	1	0.3	0.5	0.0909	0.81	1	0	0.0333	0	0	1	0	0	0.389	0	0	0	0
14	6.554	9.474	50.77	0.0635	0.125	0	0.5	0.227	0.714	0	0	0.6	0.333	0.933	0	1	1	0.944	0.2	1	0	0
13	6.357	9.189	59.96	0.571	0	1	0	0	0.429	0	1	0.233	0	0	0	1	0.143	0.944	0.4	0.5	0.667	0
7	6.172	8.922	68.88	0.190	0.25	0.7	0	0.955	0.333	0	0.9	0	0	0.2	0	0	0	0.444	1	0	0	0.8
3	5.519	7.977	76.86	0.0794	0.125	0	0	0.273	0	0	0.9	0.0333	0	0.533	0	0	0	0.0556	0.2	1	0	1
4	4.437	6.414	83.27	0.143	0.125	0	0	0.364	0	0	0.6	0.0333	0	0.667	0	0	0	0.0556	0.2	0.5	0	0
5	4.009	5.795	89.07	0	0	0	0	0	0	0	0	0	0	1.07	0	0	1.29	0	0.4	0	0	1.6
6	3.538	5.115	94.18	0	0	0	0	0.0455	0	0	0	0	0	0.667	0	0	1	0	0.6	1	0	1
2	2.558	3.698	97.88	0.0159	0	0	0	0	0	0	0	0.833	0	0	0	0	0	0	0	0	0	0
1	1.233	1.783	99.66	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0.0556	0	0	0	1
10	0.2346	0.3391	100	0	0.125	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Characters	Mean Oxyglossum	Mean Epigeneium	Mean Amblyanthus	Mean Platycaulon	Mean Conostalix	Mean Flickingeria	Mean Fytchianthe	Mean Dendrocoryne	Mean Monophyllaea	Mean Plicatiles	Mean Bilobulate	Mean Fugacia	Mean Latouria	Mean Oxystophyllum	Mean Cadetia	Mean Tetrodon	Mean Macrocladium	Mean Katherinea	Mean Racemosum	Mean Diplocaulobium	Mean unspecified
8	0.143	0	0	0.333	0	0	0.667	0.222	0	0	0	0	0	0	0	0	0	0	0	0	0
11	0	1	0.333	0	1	1	1	1	1	1	1	1	0.8	0	1	1	0.5	1	1	1	0.75
12	0	0	0.667	0	0	0	1	0.222	0	0	0	0	0	0	0	0	0	0	0	0	0.25
9	0	0	0.667	0.333	1	0	0.333	0	0	0	0	0	0.2	0	0	0	1	0	0	0	0
14	0.857	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
13	0.286	0	0.333	0.333	0	0	0	0	0	0	0	0	0	0.5	0	0	0	0	0	0	0.25
7	0	0	0	0.333	0	0	1	0.778	1	0	0	0	1	0	0	1	1	0	0.667	0	0.25
3	1	1	0.333	0	0	1	0.333	1	1	1	1	1	1	0	1	1	0	1	1	1	0.75
4	0.286	1	0	0.667	0	1	0.333	0.667	0.5	1	1	1	0.2	0	0	1	0	1	1	0	0.75
5	0.286	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0
6	0.286	0	0	0	0	0	0	0	0	0	0	0	0.2	1	1	0	0	0	0	0	0
2	0.143	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
1	0	0	0	0	0.5	0	0	0	0	1	1	0	0	0	0	0	0	1	1	0	0
10	0	0	0	0	0	0	0	0	0	0	0	0	0.4	0	0	0	0	0	0	0	0

Av. dissim. - average dissimilarity; Contrib. % - percentage of dissimilarity explained by individual traits; Cumulat. % - cumulative percentage of Bray-Curtis similarity.

B)

Characters	Av. dissim.	Contrib. %	Cumulat. %	Mean Dendrobium	Mean Breviflores	Mean Holochrysa	Mean Herbacea	Mean Stachyobium	Mean Calcarifera	Mean Distichophyllae	Mean Densiflora	Mean Formosae	Mean Stuposa	Mean Crumenata	Mean Grastidium	Mean Calyptrochilus	Mean Aporum	Mean Pedilonum	Mean Spatulata	Mean Bolbodium	Mean Dolichocentrum	Mean Rhizobium
11	18.17	28.6	28.6	0.111	1	0	0	0.909	0.333	1	0	0.967	1	0.933	0.6	0	0.667	0.0556	1	0.5	0.333	1
12	15.9	25.02	53.62	0.889	0.75	1	0	0.227	0.19	0.429	1	0.367	1	0.2	0.2	1	0.19	0	0	0	0	0
14	14.72	23.16	76.78	0.0635	0.125	0	0.5	0.227	0.714	0	0	0.6	0.333	0.933	0	1	1	0.944	0.2	1	0	0
13	14.18	22.32	99.1	0.571	0	1	0	0	0.429	0	1	0.233	0	0	0	1	0.143	0.944	0.4	0.5	0.667	0
10	0.5692	0.896	100	0	0.125	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Characters	Mean Oxyglossum	Mean Epigeneium	Mean Amblyanthus	Mean Platycaulon	Mean Conostalix	Mean Flickingeria	Mean Fytchianthe	Mean Dendrocoryne	Mean Monophyllaea	Mean Plicatiles	Mean Bilobulate	Mean Fugacia	Mean Latouria	Mean Oxystophyllum	Mean Cadetia	Mean Tetrodon	Mean Macrocladium	Mean Katherinea	Mean Racemosum	Mean Diplocaulobium	Mean unspecified
11	0	1	0.333	0	1	1	1	1	1	1	1	1	0.8	0	1	1	0.5	1	1	1	0.75
12	0	0	0.667	0	0	0	1	0.22	0	0	0	0	0	0	0	0	0	0	0	0	0.25
14	0.857	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
13	0.286	0	0.333	0.333	0	0	0	0	0	0	0	0	0	0.5	0	0	0	0	0	0	0.25
10	0	0	0	0	0	0	0	0	0	0	0	0	0.4	0	0	0	0	0	0	0	0

Av. dissim. - average dissimilarity; Contrib. % - percentage of dissimilarity explained by individual traits; Cumulat. % - cumulative percentage of Bray-Curtis similarity.

Table S6. Nominal section species according to various authors and the results obtained.

	Section						
Species	Wood (2006)	Wang et al. (2009)	Xiang et al. (2013)	Takamiya et al. 2014	Zheng et al. (2020)	Liu & Chen (2011) Chen et al. (2010) Chen et al. (2016)	Our results
Dendrobium albosanguineum	*Dendrobium*			*Dendrobium*			*Dendrobium*
Dendrobium amoenum	*Dendrobium*		*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium anosmum	*Dendrobium*			*Dendrobium*			*Dendrobium*
Dendrobium aphrodite	*Dendrobium*			*Dendrobium*			*Dendrobium*
Dendrobium aphyllum	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium aqueum	*Dendrobium*		*Dendrobium*		*Dendrobium*		*Dendrobium*
Dendrobium bensoniae	*Dendrobium*		*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium brymerianum	*Densiflora*	*Dendrobium*	*Dendrobium*	*Dendrobium*			unresolved
Dendrobium capillipes	*Holochrysa*	*Dendrobium*	unresolved	*Holochrysa*			unresolved
Dendrobium chrysanthum	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium chrysocrepis	*Dendrobium*			*Dendrobium*			*Dendrobium*
Dendrobium chrysotoxum	*Densiflora*	*Chrysotoxae*	*Dendrobium*	*Densiflora*	*Dendrobium*		*unresolved*
Dendrobium crepidatum	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium crystallinum	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium devonianum	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium dickasonii	*Dendrobium*						*Dendrobium*
Dendrobium falconeri	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium fanjingshanense			*Dendrobium*		*Dendrobium*		*Dendrobium*
Dendrobium findlayanum	*Dendrobium*			*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium flexicaule	*Dendrobium*		*Dendrobium*		*Dendrobium*		*Dendrobium*
Dendrobium friedericksianum	*Dendrobium*			*Dendrobium*			*Dendrobium*
Dendrobium gibsonii	*Holochrysa*	*Dendrobium*	*Dendrobium*	*Holochrysa*	*Dendrobium*		*Dendrobium*
Dendrobium gratiosissimum	*Dendrobium*		*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium hancockii	*Holochrysa*	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*		unresolved
Dendrobium harveyanum	*Densiflora*		*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium hekouense						*Dendrobium*	*Dendrobium*
Dendrobium heterocarpum	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium hookerianum	*Dendrobium*		*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium huoshanense			*Dendrobium*		*Dendrobium*		*Dendrobium*
Dendrobium jinghuanum					*Dendrobium*		*Dendrobium*
Dendrobium lamyaiae	*Dendrobium*			*Dendrobium*			*Dendrobium*
Dendrobium leptocladum			*Dendrobium*	*Dendrobium*			*Dendrobium*
Dendrobium linawianum	*Dendrobium*		*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium lituiflorum	*Dendrobium*		*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium loddigesii	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium luteolum	*Formosae*			*Dendrobium*			*Dendrobium*
Dendrobium maccarthiae	*Dendrobium*			*Dendrobium*			*Dendrobium*
Dendrobium macrostachyum	*Dendrobium*		*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium moniliforme	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium nobile	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium ochreatum	*Dendrobium*		*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium officinale	*Dendrobium*	*Dendrobium*	*Dendrobium*				*Dendrobium*
Dendrobium okinawense			*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium parishii	*Dendrobium*		*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium pendulum	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium primulinum	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium pulchellum	*Holochrysa*		*Dendrobium*	*Holochrysa*	*Dendrobium*		unresolved
Dendrobium regium	*Dendrobium*			*Dendrobium*			*Dendrobium*
Dendrobium rhombeum	*Dendrobium*			*Dendrobium*			*Dendrobium*
Dendrobium ruckeri	*Dendrobium*		*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium scoriarum				*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium senile	*Formosae*		*Dendrobium*	*Dendrobium*	*Dendrobium*		unresolved
Dendrobium shixingense						*Dendrobium*	*Dendrobium*
Dendrobium signatum	*Dendrobium*			*Dendrobium*			*Dendrobium*
Dendrobium tetrachromum				*Dendrobium*			*Dendrobium*
Dendrobium tortile	*Dendrobium*			*Dendrobium*			*Dendrobium*
Dendrobium transparens	*Dendrobium*		*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium unicum	*Dendrobium*		*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium wangliangii			*Dendrobium*		*Dendrobium*		*Dendrobium*
Dendrobium wardianum	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*	*Dendrobium*		*Dendrobium*
Dendrobium wilsonii			*Dendrobium*		*Dendrobium*		*Dendrobium*
Dendrobium xichouense			*Dendrobium*		*Dendrobium*		*Dendrobium*
Dendrobium zhenghuoense						*Dendrobium*	unresolved

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obejmował:

- udział w zaprojektowaniu badania i ustaleniu wspólnej koncepcji pracy

- przeprowadzenie badań laboratoryjnych

- przygotowanie tabel i rycin

- sporządzenie schematów obrazujących występowanie i zagęszczenie analizowanych struktur na warżkach

- opracowanie oraz interpretację wyników

- przygotowanie treści artykułu oraz edycję ostatecznej wersji manuskryptu

- konsultacje ze współautorami

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obejmował:

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- przeprowadzenie badań laboratoryjnych

- przeprowadzenie obliczeń na podstawie których sporządzono wykresy

- opracowanie oraz interpretację wyników

- przygotowanie wykresów, figur i tabel

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obejmował:

- udział w zaprojektowaniu badania i ustaleniu wspólnej koncepcji pracy

- udział w przeprowadzeniu analiz molekularnych

- analizę danych i interpretację wyników

- opracowanie bazy danych dotyczących zasięgu występowania gatunków

- przygotowanie treści artykułu

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obejmował:

- opracowanie koncepcji artykułu wraz ze współautorami

-weryfikację oznaczeń materiału do badań

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-naniesienie poprawek w tekście

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-opracowanie koncepcji artykułu wraz ze współautorami

-przygotowanie zestawu cech morfologicznych dla poszczególnych gatunków

-udział w interpretacji wyników

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obejmował:

-przeprowadzenie analiz filogenetycznych

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-przeprowadzenie analiz filogenetycznych

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obejmował:

-przygotowanie preparatów do badań w mikroskopie skaningowym oraz wykonanie zdjęć struktur mikromorfologicznych przy użyciu SEM

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-przeprowadzenie analiz zmienności morfologicznej

-sporządzanie tabel

-udział w napisaniu manuskryptu

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