Additional morphological notes and molecular-phylogenetic support for the distinct status of *Deinostigma cicatricosa* and *D. minutihamata* (Gesneriaceae)

Möller M.*1*, Bui H.Q.2*, Linh L.T.M.2*, Atkins H.J.1 & D.J. Middleton3

1Royal Botanic Garden Edinburgh, 20A Inverleith Row, Edinburgh EH3 5LR, Scotland, U.K.
2Institute of Ecology and Biological Resources, Vietnam Academy of Science and Technology, 18 Hoang Quoc Viet Road, Cau Giay Dist., Ha Noi, Vietnam
3Herbarium, Singapore Botanic Gardens, National Parks Board, 1 Cluny Road, 259569 Singapore
* E-mail: m.moeller@rbge.ac.uk

**Abstract:** During a recent molecular-phylogenetic revision of *Deinostigma*, material previously included in *Chirita minutihamata* D.Wood was assumed to belong to two different entities, *Deinostigma minutihamata* (D.Wood) D.J.Middleton & H.J.Atkins for material collected in Vietnam and *D. cicatricosa* (W.T.Wang) D.J.Middleton & Mich.Möller for material from China, although without supporting molecular evidence for the Vietnamese taxon. Here, we provide results in support of this decision in the form of a molecular phylogenetic analysis that includes material of *D. minutihamata* recently collected in Vietnam. This analysis shows that *D. cicatricosa* is more closely related to the other Chinese species, *D. cyrtocarpa* (D.Fang & L.Zeng) Mich.Möller & H.J.Atkins, than to the Vietnamese *D. minutihamata*. We also provide amended detailed descriptions of *D. minutihamata* and *D. cicatricosa*.

**Keywords:** China, *Chirita cicatricosa*, *Chirita minutihamata*, *D. cicatricosa*, Phylogeny, Taxonomy, Vietnam.

**Introduction**

Based on both molecular and morphological data, the genus *Deinostigma* W.T.Wang & Z.Y.Li (Gesneriaceae) was recently expanded from being a monotypic genus, with *D. poilanei* (Pellegr.) W.T.Wang & Z.Y.Li the only representative, to one that currently includes seven species (Möller *et al.*, 2016).

During the course of that study, the disjunct distribution of *Chirita minutihamata* D.Wood, as circumscribed by Wood (1974) and Wang *et al.* (1998), between central Vietnam and Guangxi in China was noted. While the plants were very similar in appearance, significant differences were also apparent, particularly in the longer, slender, more falcate fruits and the generally slightly larger flowers of the Chinese material. Together with the widely disjunct distribution, this led Möller *et al.* (2016) to treat them as separate species and the relevant new combinations were made: *D. minutihamata* (D.Wood) D.J.Middleton & H.J.Atkins for the Vietnam material, and *Deinostigma cicatricosa* (W.T.Wang) D.J.Middleton & Mich.Möller for the Chinese material. These combinations were made in anticipation of further studies, including through the use of molecular data when material of *D. minutihamata* became available, confirming that they were two distinct species.

During recent fieldwork by one of the authors, BHQ in Vietnam in June 2018, a collection was made in Quang Nam Province, Vietnam, conforming morphologically to *D. minutihamata*. Herbarium specimens and silica dried leaf material were collected. With this new material available for detailed comparative morphological and molecular phylogenetic studies, we are now able to investigate whether the recognition of *D. cicatricosa* as distinct from *D. minutihamata* is justified.
Materials and Methods

Plant materials

Materials of *D. minutihamata* were collected from Quang Nam Province, Vietnam, in June 2018 (B.H.Quang Coll no. 218, N 15°02'37.5'', E 108°02'19.9'', 692 m) for morphological and phylogenetic analyses (Fig. 1). Voucher specimens were deposited in the herbarium of the Institute of Ecology and Biological Resources, IEBR, Vietnam (HN, following the herbarium abbreviations of Thiers [continuously updated]).

For phylogenetic studies, six sequences of four species were downloaded from GenBank, *D. cicatricosa* and *D. cyrtocarpa* (D.Fang & L.Zeng) Mich.Möller & H.J.Atkins from China, and *D. tamiana* (B.L.Burtt) D.J.Middleton & H.J.Atkins and *D. poilanei* from Vietnam. The outgroup consisted of 17 samples of 15 species in 9 genera implicated in the relationship spanning *Deinostigma* and *Primulina* Hance (Ranasinghe, 2017) (Table 1). The trees were rooted on *Didymocarpus antirrhinoides* A.Weber based on a comprehensive four plastid gene phylogeny (Ranasinghe, 2017).

Total DNA extraction and PCR amplification

Genomic DNA was extracted from leaf tissue ground in liquid nitrogen according to the CTAB protocol of Doyle and Doyle (1990). The quality of DNA was determined using 1.0% agarose gels. The purified genomic DNA was quantified using a BioRad Smartspec 3000 UV-Vis spectrophotometer (California, USA).

Sequences of the nuclear ribosomal internal transcribed spacers (ITS) and the plastid *trnL*-F intron spacer (*trnL*-F) were PCR-amplified using primers: ‘5P’ (5’-GGA AGG AGA AGT CGT AAC AAG G-3’) and ‘8P’ (5’-CAC GCT TCT CCA GAC TAC A-3’) (Möller & Cronk, 1997) and ‘c’ (5’-CGA AAT CGG TAG ACG CTA-3’) and ‘f’ (5’ATT TGA ACT GGT GAC ACG AG-3’) (Taberlet et al., 1991). The PCR reaction mixture contained 15µl Hotstart PCR Mastermix (Promega, USA), 1µl forward and reverse primers (10 µM), 2 µl DNA template and dH$_2$O up to a volume of 30µl. The PCR programme settings for ITS were: 95°C for 5 min, followed by 35 cycles of 95°C for 35 s, 55°C for 45 s and 72°C for 60 s, and finished with 72°C for 3 min; for *trnL*-F it was: 95°C for 5 min, followed by 35 cycles of 95°C for 35 s, 50°C for 30 s and 72°C for 45 s, and finished with 72°C for 3 min. Electrophoresis of 5µl PCR product on a 1.5% agarose gel in TEA buffer (1h, 100V) was carried out to check for amplification success and quality. PCR products were purified using ExoSAP-IT (Thermo Fisher Scientific, Buckinghamshire, UK) following the manufacturer’s protocol. Sequencing was performed at the Apical Scientific Sdn Bhd sequencing service (Selangor, Malaysia). Sequence clean-up and sequence assembly, was carried out by the Department of Molecular Systematics and Conservation Genetics (IBER).

Phylogenetic analysis

The sequences downloaded from GenBank and the newly acquired data were assembled in two matrices, one for ITS and one for *trnL*-F and included 24 samples each. The sequences were aligned online in MAFFT (v.7 online (https://mafft.cbrc.jp/alignment/server/) (Kuraku et al., 2013; Katoh et al., 2017) and adjusted manually. The two matrices were tested for phylogenetic incongruences with the incongruence length difference test (ILD) implemented in PAUP* v.4.0a163 (Swofford, 2002) as the partition homogeneity test, and was run for 100 replicates.

The data matrices were analysed by maximum parsimony (MP) and Bayesian Inference (BI) in PAUP and MrBayes v.3.2.6 (Ronquist & Huelsenbeck, 2003; Ronquist et al., 2012). Parsimony trees were obtained from 10,000 random addition sequence trees that were optimized using MulTrees, SteepestDescent, and Tree-Bisection-Reconnection (TBR). Node support was estimated with 10,000 bootstrap replicates, each comprising
Fig. 1. Known distribution points of *Deinostigma* species. Those included in the phylogenetic analyses are shown as circles and those not included as triangles: *D. cicatrica* (W.T.Wang) D.J.Middleton & Mich.Möller (light blue circles) and *D. cyrtocarpa* (D.Fang & L.Zeng) Mich.Möller & H.J.Atkins (yellow circle) in Guangxi, China, and *D. tamiana* (B.L.Burtt) D.J.Middleton & H.J.Atkins (light green circle), *D. minutihamata* (D.Wood) D.J.Middleton & H.J.Atkins (red circles), *D. poilanei* (Pellegr.) W.T.Wang & Z.Y.Li (dark blue circles), and the unsampled *D. cycnostyla* (B.L.Burtt) D.J.Middleton & H.J.Atkins (pink triangle), and *D. eberhardtii* (Pellegr.) D.J.Middleton & H.J.Atkins (dark green triangles).
Table 1. List of samples included in the molecular phylogenetic analyses including country of origin, collection information, voucher deposition and GenBank accession numbers for ITS and \textit{trn}LF

| Species                   | Country        | Collection                                      | Deposited               | ITS          | \textit{trn}LF   |
|---------------------------|----------------|------------------------------------------------|-------------------------|--------------|-----------------|
| \textit{Agalmyla clarkei} (Elmer) B.L.Burtt | Indonesia     | RBGE-Philippine National Herbarium Expedition 1999 (99) 13 | E                       | FJ501360     | FJ501540        |
| \textit{Agalmyla paucipilosa} Hilliard & B.L.Burtt | Indonesia     | P. Smith & L. Galloway 261                      | E                       | HQ632990     | HQ632893        |
| \textit{Deinostigma cicatricosa} (W.T.Wang) D.J.Middleton & Mich.Möller | China          | M. Moeller & Y.G. Wei MMO07-1148                | IBK/E                   | KU990890     | KU990886        |
| \textit{Deinostigma cicatricosa} (W.T.Wang) D.J.Middleton & Mich.Möller | China          | W.B. Xu s.n.                                   | IBK                      | JX506925     | JX506817        |
| \textit{Deinostigma cyrtocarpa} (D.Fang & L.Zeng) Mich.Möller & H.J.Atkins | China          | M. Moeller & Y.G. Wei MMO 06-908                | IBK/E                   | KU990889     | KU990885        |
| \textit{Deinostigma cyrtocarpa} (D.Fang & L.Zeng) Mich.Möller & H.J.Atkins | China          | W.B. Xu s.n.                                   | IBK                      | JX506885     | JX506777        |
| \textit{Deinostigma minitihamata} (D.Wood) D.J.Middleton & H.J.Atkins | Vietnam        | B.H. Quang 218                                 | HN                      | MT066216     | MT075730        |
| \textit{Deinostigma poilanei} (Pellegr.) W.T.Wang & Z.Y.Li | Vietnam        | R. Rybkova HB222                                | -                       | KU990892     | KU990888        |
| \textit{Deinostigma tamiana} (B.L.Burtt) D.J.Middleton & H.J.Atkins | Vietnam        | Soviet-Vietnam Expedition (Liberec BG, Czech Republic & St. Petersburg BG) 01/114 | E                       | KU990891     | KU990887        |
| \textit{Didymocarpus antirrhinoide} A.Weber | Malaysia       | K. Jong 9009 (RBGE cult. 19650167)              | E                       | DQ912671     | FJ501513        |
| \textit{Didymostigma obtusum} (Clarke) W.T.Wang | China          | M. Moeller et al. 08-1310                       | E                       | HQ632971     | HQ632875        |
| \textit{Didymostigma trichanthera} C.X.Ye & X.G.Shi | China          | M. Moeller et al. 08-1335                       | E                       | HQ632972     | HQ632876        |
| \textit{Hemiboea fangii} Chun ex Z.Y.Li | China          | M. Moeller et al. 08-1284                       | E                       | HQ632979     | HQ632882        |
| \textit{Hemiboea longgangensis} Z.Y.Li | China          | Y.G. Wei 07550                                 | IBK                     | HQ632986     | HQ632889        |
| \textit{Metapetrocosmea peltata} (Merr. & Chun) W.T.Wang | China          | Y.G. Wei 07-702                                 | IBK                     | HQ632968     | HQ632872        |
| \textit{Oreocharis acaulis} (Merr.) Mich.Möller & A.Weber | China          | M. Moeller et al. 08-1328                       | E                       | HQ633012     | HQ632916        |
| \textit{Oreocharis henryanra} Oliv. | China          | M. Moeller et al. 10-1691                       | E                       | JF697574     | JF697586        |
| \textit{Petrocodon dealbatus} Hance | China          | Xie Qingjian J-042 (US 422841)                  | US                      | FJ501358     | FJ501537        |
| \textit{Petrocodon dealbatus} var. denticulatus (W.T.Wang) W.T.Wang | China          | Y.G. Wei 2010-03                                | IBK                     | JF697578     | JF697590        |
Bayesian inference analyses were implemented using substitution models selected separately for the ITS spacers, the 5.8S gene and \textit{trn}\textsubscript{L-F} in MrModelTest2_64bit (Nylander, 2004), under the Akaike Information Criterion (Akaike, 1974), and were GTR+G for the ITS spacers, SYM+I+G for the ITS-5.8S gene, and GTR+I for \textit{trn}\textsubscript{L-F}. Two independent runs of four Markov Chain Monte Carlo (MCMC) chains were run for one million generations, sampling every 1,000 generations. A stop-rule was implemented when the average standard deviation of split frequencies reached 0.01, and after removing the burn-in set to 10% of the sampled trees, a majority rule consensus tree was built from the remaining sampled trees, providing also the posteriori probabilities for each node.

Results

The ILD test returned a maximum value ($P=1.0$) which indicated that no incongruence between the data sets existed. As such the phylogenetic analyses were performed on combined data. The combined matrix had 1,583 characters of which 1,098 were constant (69.4%), 157 variable (9.9%) and 328 parsimony-informative (20.7%). The MP analysis retained one most parsimonious tree of 1,068 steps length, a consistency index (CI) of 0.6442 and retention index (RI) of 0.6933 that was fully resolved. Convergence of the BI runs was satisfactory (Appendix 1). The topology of the MP and BI trees were identical except for two branches that collapsed in the latter that also had no support in the MP analysis (Figs. 2 & 3).

In both analyses, \textit{Deinostigma} was monophyletic with high branch support (MPBS=85%; BIPP=0.97). It is sister to \textit{Metapetrocosmea} W.T.Wang (MPBS=100%; BIPP=1) and distant from the \textit{Primulina} samples that were sister to the \textit{Petrocodon} Hance samples with very high branch support (MPBS=99%; BIPP=1). The samples of the Chinese species of \textit{Deinostigma}, \textit{D. cyrtocarpa} and \textit{D. cicatricosa}, are sister and monophyletic (MPBS=62%; BIPP=0.76). The species pair \textit{D. tamiana} and \textit{D. poilanei} from Vietnam are also sister (MPBS=99%; BIPP=1). The sample of \textit{D. minutihamata} from Vietnam is sister to the two Chinese species in the MP analysis (MPBS=<50%), and on a polytomy with these and the clade of the sister pair \textit{D. tamiana} and \textit{D. poilanei} in the BI analysis.

Discussion

Our molecular phylogenetic analyses confirm the suggestion by Möller et al. (2016) that the collections subsumed under \textit{Chirita minutihamata} by Wood (1974) and Wang et al. (1998) belong indeed to two different species, since the material from China, now regarded as \textit{D. cicatricosa}, is sister to \textit{D. cyrtocarpa} with reasonable support, rather than to the Vietnamese specimen of \textit{D. minutihamata} (Figs. 2 & 3). This makes geographic sense since the two Chinese species are more closely related to each other than to the Vietnamese species (Figs. 1–3). The morphological case to support the separate status of \textit{D. minutihamata} and \textit{D. cicatricosa} has been made above and previously concerning the corolla size and fruit shape (Möller et al., 2016). Amended descriptions are provided below. From these, corolla colour, filament indumentum and fruit size can be added to the list of characters differentiating the two species. A summary of the main morphological characters differentiating all three species is provided in Table 2 and photographic images are provided in Fig. 4.

The phylogenies we reconstruct here do not completely reflect the distribution of \textit{Deinostigma} species across the range of the genus, since the strongly supported sister pair in Vietnam, \textit{D. tamiana} and \textit{D. poilanei}, are rather disjunct in the north on the one hand and the centre and south of Vietnam on the other. However, these disjunctions may represent an artefact of undersampling, and
Fig. 2. Single most parsimonious tree of 1068 steps length (CI=0.6442; RI=0.6933), based on combined ITS and trnL-F data for the relationships among Deinostigma species. Bootstrap values along the branches. * denotes values <50%.
additional fieldwork may yet uncover more
distribution points for some of the species, or as yet
undiscovered species. Vietnam is known to be
undercollected (Middleton et al., 2019) and the
consequences of low collection density have been
demonstrated in other Gesneriaceae genera, such as
Oreocharis Benth., which until recently was only
known from one species in Vietnam, but which in
the last two years has increased to eight species due
to new collections from recent expeditions (Möller
et al., 2018). Most of these Oreocharis species are
local endemics and only one, O. aurea Dunn, is
widespread in southern Yunnan and northern
Vietnam (Chen et al., 2018). Such patterns of high
levels of narrow endemism and rarer widespread
distributions within a genus are common in
Gesneriaceae (Middleton et al., 2019) and may also
be present in Deinostigma, where most species have a
narrow distribution and only D. minutihamata and D.
poilanei appear to be more widespread (Fig. 1).
However, more fieldwork is needed, particularly in
Vietnam and perhaps also in Laos, to obtain a complete
picture of the distribution of Deinostigma species.

Deinostigma cicatricosa (W.T.Wang) D.J.Middleton & Mich.Möller, Gard. Bull.
Singapore 68(1): 155 (2016). Chirita cicatricosa
W.T.Wang, Bull. Bot. Res., Harbin 1(4): 69 (1981).

**Type:** CHINA, Guanxi, Dongxing, Banba Commune, Renbei, 03.10.1976, D. Fang et al. 1525
(holo GXMI [GXMI050619]).

Vernacular name: 多痕奇柱苣苔, Duó hén qí zhù jù tái (Chinese)

Perennial herbs. Stems decumbent or erect, densely
pubescent with hairs of varying lengths, the longer
hairs mostly glandular but with scattered eglandular
hairs and small hooked hairs, glabrescent with age,
peg-like bases of fallen leaves persistent. Leaves
alternate, crowded towards branch apices,
internodes 3–8 mm; petioles 1.3–6 cm long, densely
pubescent with longer glandular hairs and short
hooked hairs; blade ovate to elliptic, 1.8–7.5 × 1.4–
4.3 cm, 0.9–2.2 times as long as wide, base cuneate
to subcordate, apex short acuminate, margins
crenate, secondary veins 4–5 on each side of midrib,
sparingly to densely pubescent above and beneath.
Inflorescences axillary, few-flowered, sometimes
only 1-flowered, to 10 cm long, all axes with longer
 glandular hairs and short hooked hairs; peduncle
4.5–5 cm long; bracts ovate, c. 7.5 × 3.2 mm, apex
acuminate; pedicels 6–9 mm long. Calyx 5-lobed;
lobes divided to base, narrowly elliptic, 10.5–13 ×
2.1–2.4 mm, apex acuminate, densely pubescent as
on inflorescence axes. Corolla infundibuliform, 45–

### Table 2. Comparison of characteristics of three species in Deinostigma, D. cyrtocarpa, D. cicatricosa and D. minutihamata.

| Characters                          | D. cyrtocarpa     | D. cicatricosa    | D. minutihamata   |
|------------------------------------|-------------------|-------------------|-------------------|
|                                    | (D.Fang & L.Zeng) | W.T.Wang)         | D.J.Middleton &   |
|                                    | Mich.Möller & H.J.Atkins | Mich.Möller     | Mich.Möller &     |
|                                    |                   |                   | H.J.Atkins        |
| Leaf                               | 3–15 × 1.5–6.5 cm | 1.8–7.5 × 1.4–4.3 cm | 2.6–10.2 × 1.8–4 cm |
| Corolla length                     | 3.3–5.5 cm        | 4.5–5.8 cm        | 3.7–5.2 cm        |
| Corolla colour                     | Dark purple       | Dark purple       | Pale purple to violet, with a few darker lines ventrally |
| Filament indumentum                | Glandular puberulent, densely bearded apically | Densely long pubescent | Glabrous except sparsely pubescent at apex |
| Ovary length                       | 7–9 mm            | c. 13 mm          | c. 13 mm          |
| Fruit position                     | Plagiocarpic, at ±90° angle to pedicel | Orthocarpic, ± straight in relation to pedicel | Orthocarpic, ± straight in relation to pedicel |
| Fruit shape and size               | Straight, 1.5–2 cm long | Curved, 4.5–5 cm long | Straight, 2–2.3 cm long |
| Fruit dehiscence                   | Predominantly loculicidally along the upper suture | Loculicidally dehiscing along both sutures into 2 valves | Loculicidally dehiscing along both sutures into 2 valves |
Fig. 3. Bayesian inference majority rule consensus tree with average branch lengths based on combined ITS and trnL-F data for the relationships among Deinostigma species. Posterior probabilities along the branches.
Notes on Deinostigma minutihamata

58 mm long; dark purple, tube 33–37.5 mm long, lobes orbicular, apices rounded; upper lobes 7–9 × 7–9 mm, lateral lobes 8–10 × 11 mm, lower lobe 8–9.5 × 9.2–11.5 mm, pubescent outside with glandular hairs, glabrous inside. Fertile stamens 2; filaments slightly curved, 10–12 mm long, densely long pubescent; anthers coherent, 1.5–2 × 3.5–5.5 mm, densely pubescent; staminodes 3, 8.5–10 mm long, densely pubescent. Disc 5-lobed, c. 1 mm high. Ovary c. 13 mm long, densely glandular pubescent; style c. 18 mm long, densely glandular pubescent; stigma chiritoid, lower lip 2-lobed, c. 5.5 mm long. Mature capsule curved, 4.5–5 cm long. Seeds not seen.

Flowering & fruiting: Flowering from October–December and fruiting from November.

Habitat: Growing in montane forests.

Distribution: Endemic to China (Guangxi).

Specimen examined: CHINA, Guangxi (as Kwangtung on label, Kwangtung–Tonkin border), Fang Cheng district, Kung Ping Shan and vicinity, semi-woody, growing in thicket, 25–30.08.1936, W.T.Tsang 26711 (E [E00627703]).

Deinostigma minutihamata (D.Wood) D.J.Middleton & H.J.Atkins, Gard. Bull. Singapore 68(1): 158. 2016. Chirita minutihamata D.Wood, Notes Roy. Bot. Gard. Edinburgh 31: 370. 1972. Primulina minutihamata (D.Wood) Mich.Möller & A.Weber, Taxon 60: 783. 2011. Type: VIETNAM, Kon Tum, Dak Glei district, Ngok Pa Not, 2300 m, 12.12.1946, E. Poilane 35803 (holo P [P00602518] isotype P [P00602519]). Fig. 4e–j

Vernacular name: Bảo xuân móc nhỏ (Vietnamese).

Subshrub or perennial herbs, to 40 cm tall. Stems decumbent, branched, densely pubescent with hairs of varying lengths, the longer hairs mostly glandular but with scattered glandular hairs and small hooked hairs, glabrescent with age, peg-like bases of fallen leaves persistent. Leaves alternate, crowded towards branch apices, internodes 3–6 mm; petioles 1.7–4 cm long, densely pubescent with glandular and eglandular hairs; blade ovate to elliptic, 2.6–10.2 × 1.8–4 cm, 1.2–3 times as long as wide, base cuneate, rarely to obtuse, apex short acuminate, margins weakly crenate, secondary veins 4–5 on each side of midrib, densely pubescent above and beneath. Inflorescences axillary, few-flowered, sometimes only 1-flowered, to 8.2 cm long, all axes with longer glandular and eglandular hairs and shorter hooked hairs; peduncle to 2.7 cm long; bracts ovate, c. 13 × 7 mm, apex acuminate; pedicels 5–28 mm long. Calyx 5-lobed; lobes divided to base, narrowly ovate, 16–17 × 2–2.2 mm, apex acuminate, densely pubescent as on inflorescence axes. Corolla infundibuliform, 37–52 mm long; pale blue to violet with few darker lines ventrally, lobes orbicular, apices rounded; tube 28–35 mm long; upper lobes c. 9 × 7 mm, lateral lobes c. 5 × 7 mm, lower lobe c. 7 × 7 mm, pubescent outside with glandular and eglandular hairs, glabrous inside. Fertile stamens 2; filaments slightly curved, c. 11.6 mm long, densely pubescent at apex; anthers coherent, 1.4–2 × 3–5 mm, densely pubescent; staminodes 3, c. 8 mm long, pubescent at apex. Disc not seen. Ovary c. 13 mm long, densely glandular pubescent; style c. 15 mm long, densely glandular pubescent; stigma chiritoid, lower lip 2-lobed, c. 3 mm long. Capsules narrowly fusiform, straight, 2–3.5 cm long, sparsely pubescent. Seeds 0.4–0.5 × 0.15–0.2 mm.

Flowering & fruiting: Flowering from April–June and fruiting from July–December.

Habitat: Growing on rocks in primary forest.

Distribution: Endemic to Vietnam.

Specimens examined: VIETNAM, Kon Tum, Dak Glei district, Massif du Ngok Pan, 2300 m, 12.12.1946, E. Poilane 35781 (P [P03884219], NW slope of Ngoc Linh mountain system above Long Nam village, 1700–1900 m, 04.04.1995, Averyanov, N.T. Hiep, P.K. Loc VH1165 (P [P03884218], E [E00267299], HN [HN0000031083]), W slope of Ngoc Linh mountain system on elevation to Ngoc Gua peak, 1900–2000 m, 10.04.1995, Averyanov, N.T. Hiep, P.K. Loc VH1316 (P [P03884217], HN [HN0000031082]). Lam Dong, Lac Duong district, Da Chay municipality, 29 km to NE from Dalat city, 2150 m, 01.05.1997,
Fig. 4. Photographic images of Deinostigma cicatricosa (W.T.Wang) D.J.Middleton & Mich.Möller (a, b), its phylogenetically closest relative D. cyrtocarpa (D.Fang & L.Zeng) Mich.Möller & H.J.Atkins (c, d), and D. minutihamata (D.Wood) D.J.Middleton & H.J.Atkins (e–j): a, c, e. Habit; b, d, f. Flowers; g. Adaxial and abaxial side of leaves; h. Cut open flower; i. Stamens (left) and staminodes (right); j. Calyx and pistil. a, c, e. scale bars = 2 cm, b, d, f, h scale bars = 1 cm, g–j scales in mm (photos a & c by M. Möller; b & d by Yi-Gang Wei; e–j by H.Q. Bui).
Averyanov, N.T. Hiep, P.K. Loc VH4492 (HN [HN0000031034]). Quang Nam, Nam Tra My district, Tra Linh commune, Tra Cang village, N 15°02'37.5", E 108°02'19.9", 692 m, 18.06.2018, Quang 218 (HN).

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Appendix A:

Characteristics of the Bayesian Inference analysis for the combined ITS and trnL-F data for the relationships among Deinostigma species.

Average standard deviation of split frequencies: 0.009947

Analysis stopped because convergence diagnostic hit stop value.
Analysis completed in 6 mins 21 seconds
Analysis used 380.80 seconds of CPU time

Likelihood of best state for “cold” chain of run 1 was -7709.24
Likelihood of best state for “cold” chain of run 2 was -7709.59

Acceptance rates for the moves in the “cold” chain of run 1:
With prob. (last 100) chain accepted proposals by move
25.1 % (28 %) Dirichlet(Revmat[all])
45.0 % (34 %) Slider(Revmat[all])
24.8 % (24 %) Dirichlet(Pi[1])
29.9 % (31 %) Slider(Pi[1])
24.2 % (25 %) Dirichlet(Pi[2])
27.8 % (21 %) Slider(Pi[2])
31.7 % (27 %) Multiplier(Alpha[2,3])
31.9 % (19 %) Slider(Pinvar[1,3])
3.2 % (1 %) ExtSPR(Tau[all],V[all])
4.5 % (7 %) ExtTBR(Tau[all],V[all])
7.1 % (3 %) NNI(Tau[all],V[all])
7.6 % (3 %) ParsSPR(Tau[all],V[all])
26.6 % (31 %) Multiplier(V[all])
24.5 % (22 %) Nodeslider(V[all])
21.0 % (27 %) TLMultiplier(V[all])

Acceptance rates for the moves in the “cold” chain of run 2:
With prob. (last 100) chain accepted proposals by move
23.2 % (31 %) Dirichlet(Pi[2])
27.1 % (23 %) Slider(Pi[2])
32.1 % (27 %) Multiplier(Alpha[2,3])
31.4 % (32 %) Slider(Pinvar[1,3])
3.0 % (4 %) ExtSPR(Tau[all],V[all])
4.5 % (7 %) ExtTBR(Tau[all],V[all])
7.0 % (7 %) NNI(Tau[all],V[all])
7.7 % (6 %) ParsSPR(Tau[all],V[all])
26.6 % (28 %) Multiplier(V[all])
24.2 % (23 %) Nodeslider(V[all])
21.3 % (24 %) TLMultiplier(V[all])

Chain swap information for run 1:

|   | 1  | 2  | 3  | 4  |
|---|----|----|----|----|
| 1 | 0.68 | 0.43 | 0.25 |   |
| 2 | 73791 | 0.71 | 0.47 |   |
| 3 | 74312 | 73845 | 0.72 |   |
| 4 | 74369 | 74219 | 74464 |   |

Chain swap information for run 2:

|   | 1  | 2  | 3  | 4  |
|---|----|----|----|----|
| 1 | 0.67 | 0.43 | 0.25 |   |
| 2 | 73767 | 0.70 | 0.46 |   |
| 3 | 74334 | 74477 | 0.72 |   |
| 4 | 74180 | 74114 | 74464 |   |

Upper diagonal: Proportion of successful state exchanges between chains
Lower diagonal: Number of attempted state exchanges between chains

Chain information:

| ID | Heat |
|----|------|
| 1  | 1.00 (cold chain) |
| 2  | 0.91 |
| 3  | 0.83 |
| 4  | 0.77 |

Heat = 1 / (1 + T * (ID - 1))
(where T = 0.10 is the temperature and ID is the chain number)