Causonis sessilifolia (Vitaceae), a new species from Thailand

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Abstract
A new species, Causonis sessilifolia, from Thailand is described, based on morphological and phylogenetical methods. A full description, conservation assessment, a key, images and phylogenetic tree are provided. Diagnostic characters for this species are sessile leaves that are sometimes opposite and inflorescence insertion interfoliar.

Keywords
Causonis, Thailand, new taxon, taxonomy

Introduction
The genus Cayratia Juss. in the broad sense (s.l.) has consistently been found to be paraphyletic (Wen et al. 2007; Trias-Blasi et al. 2012; Lu et al. 2013; Parmar et al. 2021). To maintain the monophyly within Vitaceae, the species in Cayratia sect. Discypharia Suess. (Suessenguth, 1953; Latiff, 1981) which was later treated as Cayratia subg. Discypharia (Suess.) C.L.Li (Li 1996, 1998; Chen et al. 2007), were placed in the newly-re-instated genus Causonis Raf. (Wen et al. 2013; Parmar et al. 2021). This
A genus can be separated from the *Cayratia* s.l. as it lacks a distinct membrane enclosing the ventral infolds in seeds (Wen et al. 2013; Parmar et al. 2021). *Causonis* can be distinguished from other Vitaceae genera by plants being hermaphrodite, flowers 4-merous, inflorescences mostly axillary, but sometimes pseudo-axillary.

*Causonis* comprises about 16 species and four varieties and is found in tropical, subtropical and temperate Asia to Australia including the Pacific Islands (Parmar et al. 2021). Trias-Blasi and Parnell (2020) reported two *Causonis* species in Thailand, but the specimen from Nakhon Sawan in the northern floristic region of Thailand, was found to be morphologically distinct from other species reported from the region.

In autumn 2019, an expedition was conducted in a 5-hectare area adjacent to Kriangkrai River in the Nakhon Sawan Province (Fig. 1), because of concerns relating to high levels of deforestation in the lowland floodplain forests in Thailand. Additionally, very few specimens had been collected from this habitat. Generally, these areas are flooded every year during September-November when the water increases by 1–3 metres. This species was found in an open area along the riverbank.

**Methods**

This study is based on the material collected in October 2019 in Nakhon Sawan. Morphological characters were studied using a hand lens (30–60× magnification) and stereomicroscope and documented by photography. Collected specimens were thoroughly compared with protologues and types of all *Causonis* species occurring in Thailand and neighbouring regions. Additionally, herbarium material of *Causonis*, deposited in K, was studied (herbarium codes according to Thiers 2021). Herbarium vouchers for this study are deposited in BKF. The description follows the style and level of details outlined in Trias-Blasi and Parnell (2020), while the general terminology is based on Beentje (2016). The conservation assessment is based on the most recent version of the guidelines of IUCN Standards and Petitions Subcommittee (IUCN 2012).

Genomic DNA for the new taxon was extracted from silica-gel-dried leaf material using DNeasy Plant Mini Kit Qiagen (Qiagen, Hilden, Germany) following manufacturers’ protocols. All other sequences for different taxa were downloaded from GenBank. The DNAs were amplified for four chloroplast loci (*atpB-rbcL*, *trnC-petN*, *trnH-psbA* and *trnL-F*) following the protocols in Lu et al. (2018). The PCR products were purified and examined on a 1% agarose gel before being sent to Majorbio Company in Beijing, China, for sequencing on a Roche 454 sequencer with the same PCR amplification primers using a standard GS FLX Titanium sequencing kit XL+ (454 Life Sciences, Branford, CT, USA). The voucher specimens and the sequences’ GenBank accession numbers are provided in Table 1.

Geneious 8.1.9 was used to assemble forward and reverse sequences (Kearse et al. 2012). Geneious was also used to edit contiguous sequences and check chromatograms for base validation. Following that, sequences were aligned using MAFFT 1.3.1 (Katoh et al. 2002) and then manually adjusted in Geneious. On the CIPRES Science Gateway (Miller et al. 2010), phylogenetic analyses were first performed for individ-
Causonis sessilifolia (Vitaceae), a new species from Thailand

Causonis sessilifolia (Vitaceae), a new species from Thailand

ual DNA loci using the Maximum Likelihood (ML) approach in RAxML-HPC2 on XSEDE (8.2.12) using the GTR + G model with 1000 bootstrap replicates (Stamatakis 2014). Single tree analyses did not detect well-supported topological conflicts amongst individual DNA loci (i.e. ML BS < 70%; Hillis and Bull 1993). As a result, for further phylogenetic analyses, the four chloroplast DNA loci were concatenated. For the four chloroplast (4cp) dataset, partitioned ML and Bayesian Inference (BI) analyses were performed and the best fitting models for individual data partitions were selected using PhyML 3.0 (Guindon et al. 2010) with the Akaike Information Criterion (AIC). The nucleotide substitution model GTR + G was found to be the most suitable for trnC-petN and trnL-F and HKY85 + G for atpB-rbcL and trnH-psbA. MrBayes 3.2.6 was used to conduct Bayesian analysis on the CIPRES Science Gateway (Ronquist et al. 2012). For a total of 10,000,000 generations, four Markov Chain Monte Carlo analyses were conducted, with one tree sampled every 1,000 generations. The standard deviation between the split frequencies was found to be less than 0.01, indicating that enough generations had been completed. Following the burn-in of the first 25% of trees, the remaining trees were used to determine a 50% majority-rule consensus tree and posterior probabilities (PP). The trees obtained from ML and BI were analysed for topological conflicts through FigTree v.1.4.4 (Rambaut 2018).

**Taxonomic treatment**

*Causonis sessilifolia* Trias-Blasi & G.Parmar, sp. nov.
urn:lsid:ipni.org:names:77222603-1

**Diagnosis.** Morphologically, *Causonis sessilifolia* and *Causonis japonica* (Thunb. ex Murray) Raf. share similarity in possessing 5-foliolate leaves, but the former taxon has sessile leaves and 2–5-furcate tendrils (vs. petiolate leaves and 2–3-furcate tendrils in *C. japonica*)
Table 1. Voucher information and GenBank accession numbers for the sequences used in this study.

| Taxon                                      | Voucher No. | Locality                  | atpB-rbcL | trnC-petN | trnH-psbA | trnL-F   |
|--------------------------------------------|-------------|---------------------------|-----------|-----------|-----------|----------|
| Causonis australica L.M.Lu & Jackes        | AU015 (PE)  | Australia, Queensland     | MW408585  | MW408375  | MW408696  | MW408491 |
| Causonis clematidea (F.Muell.) Jackes      | Wen 12184 (US) | Thailand, Nakhon Sawan   | KC166297  | KC166475  | KC166552  | KC166625 |
| Causonis cornicalata (Benth.) J.Wen & L.M.Lu | YSL4758 (PE) | China, Taiwan             | MW408551  | MW408342  | MW408665  | MW408460 |
| Causonis daliensis (C.LLi) G.Parmar & L.M.Lu | VN2014116 (PE) | Vietnam, Lam Dong         | MW408540  | MW408333  | MW408654  | MW408450 |
| Causonis fugongensis (C.LLi) G.Parmar & L.M.Lu | CPG36648 (PE) | Myanmar, Kachin           | MW408564  | MW408355  | MW408676  | MW408473 |
| Causonis japonica (Thunb.) Raf. var. japonica | Wen 8537 (US) | Japan, Chiba              | KC166313  | KC166488  | KC166564  | KC166637 |
| Causonis japonica var. pseudotrifolia (W.T.Wang) G.Parmar & J.Wen | Wen 8085 (US) | China, Chongqing          | AB234920  | KC166498  | KC166573  | AB235006 |
| Causonis maritima (Jacks) Jackes           | AU020 (PE)  | Australia, Queensland     | MW408567  | MW408358  | MW408679  | MW408476 |
| Causonis mollis (Wall. ex M.A.Lawson) G.Parmar & J.Wen | Wen 9403 (US) | China, Taiwan             | KC166314  | JF437193  | JF437079  | JF437299 |
| Causonis sessilifolia Trias-Blasi & G.Parmar | CPG23617 (PE) | Vietnam, Vinh Phuc       | MW408535  | MW408328  | MW408650  | MW408445 |
| Causonis timorienensis var. mekongensis (C.Y.Wu ex W.T.Wang) G.Parmar & L.M.Lu | OK338627 (PE) | Thailand, Nakhon Sawan    | OK338627  | OK338628  | OK338629  | OK338630 |
| Causonis trifolia (L.) Mabbb. & J.Wen        | CPG27533 (PE) | China, Yunnan             | MW408575  | MW408365  | MW408678  | --       |
| Causonis trifolia (L.) Mabbb. & J.Wen        | CPG388701 (PE) | India, Kerala             | MW408576  | MW408366  | MW408688  | --       |
| Causonis trifolia (L.) Mabbb. & J.Wen        | LA17 (PE)   | Laos, Luang Namtha        | MW408577  | MW408367  | MW408689  | MW408484 |
| Pseudocayratia pengiana Hsu & J.Wen          | CPG191178 (PE) | Thailand, Chiang Mai     | KC166323  | KC166500  | KC166574  | AB235007 |
| Pseudocayratia speciosa J.Wen & L.M.Lu       | YSL4764 (PE) | Indonesia, Bali          | KC428757  | KC428783  | KC428800  | KC428819 |

“–” represents missing sequences.

Type material. **Thailand. Northern floristic region:** Nakhon Sawan, Muang, Kriangkrai subdistrict, abandoned area at the bridge of Kriangkrai Canal, 15°44’40”N, 100°11’9”E, 23 October 2019, M. Poopath & S. Duangjai 2511 (holotype, BKF! (SN229663 (Fig. 5)), isotype BKF! (SN229662)).

Description. Slender herbaceous climber. Stem cylindrical, 2–5 mm diameter, branched, glabrous, young stems purplish-green, hairy with some bent hairs to glabrous; tendril 2–5-furcate, slender, wiry, leaf-opposed, cylindrical, with a straight section, then bifurcating and coiling, 5–10 cm long, glabrous. Leaves compound, pedately 5-foliolate, alternate or opposite; petiole absent, central petiolule 0.5–1.5(–3) cm long, middle petiolules sessile, lateral petiolules 0.5–2 mm long, mostly glabrous, sometimes with bent hairs; central leaflet blade lanceolate to slightly rhombic, 2–5 by 1–2.5 cm, base cuneate; middle leaflet blade lanceolate, 1–3 by 0.5–1.5 cm; lateral leaflet blade lanceolate often with a single asymmetric acute lobe, 0.5–1.75 by 0.2–1.25 cm, base cuneate; margin broadly denticulate, apex acuminate to mucronate; adaxial and abaxi-
Causonis sessilifolia (Vitaceae), a new species from Thailand

Figure 2. *Causonis sessilifolia* A habit B pedately 5-foliolate leaves C flowers and opposite leaves D fruits. Images: Sutee Duangjai and Manop Poopath.

...al sides glabrous, mid-rib raised on upper surface, each leaflet with 2–5 pairs of lateral veins, if leaflet lobed then lower lateral vein may be more distinct. *Inflorescence* rami-fied, interfoliar or pseudo-terminal, mostly dividing dichotomously, with numerous ramifications, 0.7–2 by 1–2.5 cm, lax, erect; peduncle 2–8 cm long, glabrous, pedicel
Figure 3. *Causonis sessilifolia*. Habitat. Images: Manop Poopath.

Figure 4. Phylogeny of selected species of *Causonis* including *C. sessilifolia*, based on the combined chloroplast dataset (*atpB-rbcL, trnC-petN, trnH-psbA* and *trnL-F*). Maximum Likelihood bootstrap values and Bayesian posterior probability values are indicated above branches respectively.

0.75–2 mm long, glabrous. Buds ovoid, 1–2 by 1–2 mm. *Calyx* cupuliform, entire, margin sinuate, 0.5–1 by 1–1.25 mm, glabrous, dark red. *Corolla* petals 4, ovate, 1.5–2 by 1.25–2 mm, apex cucullate, glabrous, dark red. *Stamens* 4; filaments flattened, broadening at the base, 1 mm long, cream; anthers elliptic, medifixed, 0.5–1 mm long, cream. *Ovary* adnate to the disc; disc with 4 distinct lobes, cupular, 0.5 by 1.5 mm,
Causonis sessilifolia (Vitaceae), a new species from Thailand

Figure 5. Holotype of Causonis sessilifolia.

glabrous, dark red outside and whitish inside. Style conical, 0.5 mm long; stigma inconspicuous, dark red. Fruit berry, globose, 4–7 mm in diameter, glabrous, smooth, green. Seeds 2, 4–5 by 3 mm, adaxial side with two faces, abaxial side convex and ovate with a linear chalazal knot (Fig. 2).

Phenology. Flowering and fruiting in October.
Etymology. The specific epithet “sessilifolia” refers to the sessile leaves of the taxon.

Distribution and habitat. Thailand (Northern floristic region, Nakhon Sawan). Lowland floodplain forest, along the bank of canal in open areas; 30 m alt. (Figs 1, 3).

Conservation status. This species is only known from the type locality and, therefore, has an Area of Occupancy (AOO) and Extent of Occupancy (EOO) of 4 km². This suggests that it might be Critically Endangered as the AOO is less than 10 km² and is only found in one location. The species has been found to grow outside any protected areas and in an abandoned area next to a canal. This could mean the species is more vulnerable than others as it is unprotected. Additionally, all the surrounding areas are used for agriculture and, therefore, it is likely this forested area might also be transformed for this use. Due to this threat, the restricted AOO and number of locations, we think that this taxon could be driven to being Critically Endangered or Extinct in a very short time; therefore, we assess the taxon as VUD2 (IUCN 2012).

Taxonomic remarks. Phylogenetically, this pedately 5-foliolate leaved species lies in a clade previously known with species of exclusively trifoliolate leaves such as Causonis trifolia (L.) Mabb. & J.Wen and Causonis maritima (Jackes) Jackes (Parmar et al. 2021), but C. sessilifolia lacks the petiolate leaves found in C. trifolia and C. maritima. In particular, this species is phylogenetically most closely related to C. maritima (ML BS = 74%; Bayesian PP = 0.99; Fig. 4), but morphologically differs from it in having pedately 5-foliolate leaves (vs. trifoliolate), leaves sometimes opposite (vs. leaves always alternate), leaves sessile (vs. leaves petiolate), inflorescence insertion interfoliar (vs. axillary), calyx and corolla glabrous (vs. hairy).

Key to Causonis in Thailand (including Causonis maritima)

1 Leaves 3-foliolate ................................................................. 2  
  – Leaves 5-foliolate ................................................................. 3 

2 Tendrils 2–3-furcate, tips without adhesive disc; hooked hairs confined to mid-vein on adaxial surface ............................................................... 1. C. maritima  
  – Tendrils 3–5-furcate, tips with adhesive disc; hairs all over abaxial and adaxial surfaces ........................................................................................................ 2. C. trifolia

3 Petiole present; inflorescence leaf-opposed, pseudoaxillary or axillary ........... 3. C. japonica  
  – Petiole absent; inflorescence interfoliar ........................................ 4. C. sessilifolia

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Causonis sessilifolia (Vitaceae), a new species from Thailand

References

Beentje H (2016) The Kew Plant Glossary, an Illustrated Dictionary of Plant Terms (2nd edn). Kew Publishing, Royal Botanic Gardens Kew, 184 pp.

Chen Z, Ren H, Wen J (2007) Vitaceae. In: Wu Z, Hong D, Raven P (Eds) Flora of China. Science Press and Missouri Botanical Garden Press, Beijing, St. Louis, Vol. 12, 173–222.

Guindon S, Dufayard J, Lefort V, Anisimova M, Hordijk W, Gascuel O (2010) New algorithms and methods to estimate Maximum-Likelihood phylogenies: Assessing the performance of PhyML 3.0. Systematic Biology 59(3): e30721. https://10.1093/sysbio/syq010

Hillis D, Bull J (1993) An empirical test of bootstrapping as a method for assessing confidence in phylogenetic analysis. Systematic Biology 42(2): 182–192. https://doi.org/10.1093/sysbio/42.2.182

IUCN (2012) Threats classification scheme (Version 3.2.). http://www.iucnredlist.org/documents/Dec_2012_Guidance_Threats_Classification_Scheme.pdf [Accessed 13.08.2021]

Katoh K, Misawa K, Kuma K, Miyata T (2002) MAFFT: A novel method for rapid multiple sequence alignment based on fast Fourier transform. Nucleic Acids Research 30(14): 3059–3066. https://doi.org/10.1093/nar/gkf436

Kearse M, Moir R, Wilson A, Stones-Havas S, Cheung M, Sturrock S, Buxton S, Cooper A, Markowitz S, Duran C, Thierer T, Ashton B, Meintjes P, Drummond A (2012) Geneious basic: An integrated and extendable desktop software platform for the organization and analysis of sequence data. Bioinformatics (Oxford, England) 28(12): 1647–1649. https://doi.org/10.1093/bioinformatics/bts199

Latiff A (1981) Studies in Malesian Vitaceae V. The genus Cayratia in the Malay Peninsula. Sains Malaysiana 10(2): 129–139.

Li C (1996) New taxa in Vitaceae from China. Chinese Journal of Applied and Environmental Biology 2(1): 43–53.

Li C (1998) Vitaceae: Flora Reipublicae Popularis Sinicae, no. 2, vol. 48 Science Press, Beijing, China, 1–3, 12–208.

Lu LM, Wang W, Chen ZD, Wen J (2013) Phylogeny of the non-monophyletic Cayratia Juss. (Vitaceae) and implications for character evolution and biogeography. Molecular Phylogenetics and Evolution 68(3): 502–515. https://doi.org/10.1016/j.ympev.2013.04.023

Lu LM, Cox C, Mathews S, Wang W, Wen J, Chen ZD (2018) Optimal data partitioning, multispecies coalescent and Bayesian concordance analyses resolve early divergences of the grape family (Vitaceae). Cladistics 34(1): 57–77. https://doi.org/10.1111/cla.12191

Miller M, Pfeiffer W, Schwartz T (2010) Creating the CIPRES science gateway for inference of large phylogenetic trees. Proceedings of the Gateway Computing Environments Workshop (GCE), New Orleans, Louisiana, 45–52. https://doi.org/10.1109/GCE.2010.5676129

Parmar G, Dang VC, Rabarijona RN, Chen ZD, Jackes BR, Barrett RL, Zhang ZZ, Niu YT, Trias-Blasi A, Wen J, Lu LM (2021) Phylogeny, character evolution and taxonomic revision of Causonis Raf., a segregate genus from Cayratia Juss. (Vitaceae). Taxon: 1–31. https://doi.org/10.1002/tax.12562

Rambaut A (2018) FigTree, version 1.4.4. Program distributed by the author. Institute of Evolutionary Biology, University of Edinburgh. http://tree.bio.ed.ac.uk/software/figtree
Ronquist F, Teslenko M, van der Mark P, Ayres D, Darling A, Höhna S, Larget B, Liu L, Suchard M, Huelsenbeck J (2012) MrBayes 3.2: Efficient Bayesian phylogenetic inference and model choice across a large model space. Systematic Biology 61(3): 539–542. https://doi.org/10.1093/sysbio/sys029

Stamatakis A (2014) RAxML version 8: A tool for phylogenetic analysis and post-analysis of large phylogenies. Bioinformatics (Oxford, England) 30(9): 1312–1313. https://doi.org/10.1093/bioinformatics/btu033

Suessenguth K (1953) Vitaceae. In: Engler A, Prantl K (Eds) Die Natürlichen Pflanzenfamilien. 20d. Duncker & Humbolt, Berlin, 174–333.

Thiers B (2021) Index Herbariorum: a global directory of public herbaria and associated staff. New York Botanical Garden’s Virtual Herbarium. https://sweetgum.nybg.org/ih/ continuously updated [accessed on 15 September 2021]

Trias-Blasi A, Parnell JAN (2020). Causonis. In: Chayamarit K, Balslev H (Eds) Flora of Thailand, Bangkok, 14(4): 600–603.

Trias-Blasi A, Parnell JAN, Hodkinson TR (2012) Multi-gene region phylogenetic analysis of the grape family (Vitaceae). Systematic Botany 37(4): 941–950. https://doi.org/10.1600/036364412X656437

Wen J, Nie Z-L, Soejima A, Meng Y (2007) Phylogeny of Vitaceae based on the nuclear GAI1 gene sequences. Canadian Journal of Botany 85(8): 731–745. https://doi.org/10.1139/B07-071

Wen J, Lu L, Boggan JK (2013) Diversity and Evolution of Vitaceae in the Philippines. Philippine Journal of Science 142: 223–244.