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## **Rediscovery of the Genus *Tsingya* Capuron (Sapindaceae) and Its Phylogenetic Position**

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# Rediscovery of the genus *Tsingya* Capuron (Sapindaceae) and its phylogenetic position

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## Abstract

BUERKI, S., D. R. DOHERTY, L. GAUTIER & M. W. CALLMANDER (2014). Rediscovery of the genus *Tsingya* Capuron (Sapindaceae) and its phylogenetic position. *Candollea* 69: 195-200. In English, English and French abstracts.

A recent intensive inventory in the Beanka region in western Madagascar has led to the rediscovery of a poorly known endemic and monotypic Malagasy genus: *Tsingya* Capuron (Sapindaceae). *Tsingya bemarana* Capuron was only known from the type collected in 1952 in the Bemaraha eroded limestone massif. This rediscovery allows to confirm its generic validity by providing a full taxonomical description of its only species, including its fruit so far unknown, and inferring its phylogenetic position, and to assess its conservation status.

## Key-words

SAPINDACEAE – *Tsingya* – Macphersonia group – Molecular Phylogeny

## Résumé

BUERKI, S., D. R. DOHERTY, L. GAUTIER & M. W. CALLMANDER (2014). La redécouverte du genre *Tsingya* Capuron (Sapindaceae) et sa position phylogénétique. *Candollea* 69: 195-200. En anglais, résumés anglais et français.

Un récent inventaire intensif dans la région de Beanka à l'ouest de Madagascar a conduit à la redécouverte d'un genre endémique malgache mal connu: *Tsingya* Capuron (Sapindaceae). *Tsingya bemarana* Capuron n'était connu que par le type, récolté en 1952 dans le massif calcaire érodé du Bemaraha. Cette redécouverte permet de confirmer sa validité générique en fournissant une description taxonomique complète de son unique espèce incluant son fruit jusqu'à lors inconnu, et en inférant sa position phylogénétique, ainsi que d'évaluer son statut de conservation.

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## Introduction

In his treatment of the Malagasy *Sapindaceae*, CAPURON (1969) described the monotypic genus *Tsingya* Capuron, endemic to the forests of the ruiniform karstic limestone (“tsingy” in Malagasy) of the Bemaraha Massif. The description was based on one fragmentary collection (*Service Forestier 6762*), which included inflorescences with flowers partly damaged by galls. However, CAPURON (1969) made some valuable observations in support of his new genus, which is nevertheless morphologically closely related to *Plagioscyphus* Radlk., which appears to possess similar fruits. CAPURON (1969) distinguished *Tsingya bemarana* Capuron by its i) absence of petals, ii) flattened, irregularly lobed and stellate pubescent disc and iii) stamens inserted into deep holes in the disc. Based on this evidence, Capuron tentatively classified *Tsingya* in the tribe *Schleichereae*, but urged for additional collections to be made (especially of mature fruiting material).

It took more than 60 years for this species to be rediscovered in the Beanka forests by a Malagasy botanist: R. Mitia Hanitrarivo, and for mature fruits to be collected. This region is in effect a northern extension of the Bemaraha massif occurring on the same basement rock. It has been the subject of intensive biodiversity inventories during the past years in conjunction with a collaborative effort to protect its mosaic of tsingy and white sands landforms and the plants and animals they support. A monograph has ensued on the biodiversity of this interesting area (GOODMAN & al., 2013), including a checklist of vascular plant species (GAUTIER & al., 2013), a mapping of forest cover (CHATELAIN & al., 2013) and a description of forest types (RAKOTOZAFY & al., 2013).

In this note, we reassess the generic validity of the genus *Tsingya*, and provide a full morphological description of *Tsingya bemarana*, inferring its phylogenetic position and assessing its conservation status. Plastid and nuclear DNA regions have been sequenced for the newly rediscovered collection of *Tsingya bemarana* and these data have been included into the dataset of BUERKI & al. (2010) spanning all the lineages of *Sapindaceae*.

## Phylogenetic position of *Tsingya* within Malagasy *Sapindaceae*

Sequences for four plastid markers (*matK*, *trnL* intron, *trnK-matK* and *trnL-trnF*) and the nuclear ITS region were generated for *Tsingya bemarana*. The DNA extraction, amplification and sequencing protocols for the nuclear and plastid markers are provided in BUERKI & al. (2009). The sequences will be submitted to GenBank as part of a forthcoming study on the “Macphersonia and Cupania groups”, but are available upon request. The phylogenetic position of *Tsingya bemarana* within *Sapindaceae* was inferred using maximum likelihood (ML) implemented in RAxML 7.2.8 (STAMATAKIS, 2006) with

1000 bootstraps followed by best-scoring ML tree. Although the analysis was partitioned, we followed the recommendation of STAMATAKIS (2006) and used the GTR+G model for all the DNA regions. The analysis was done using the facilities made available by the CIPRES portal in San Diego.

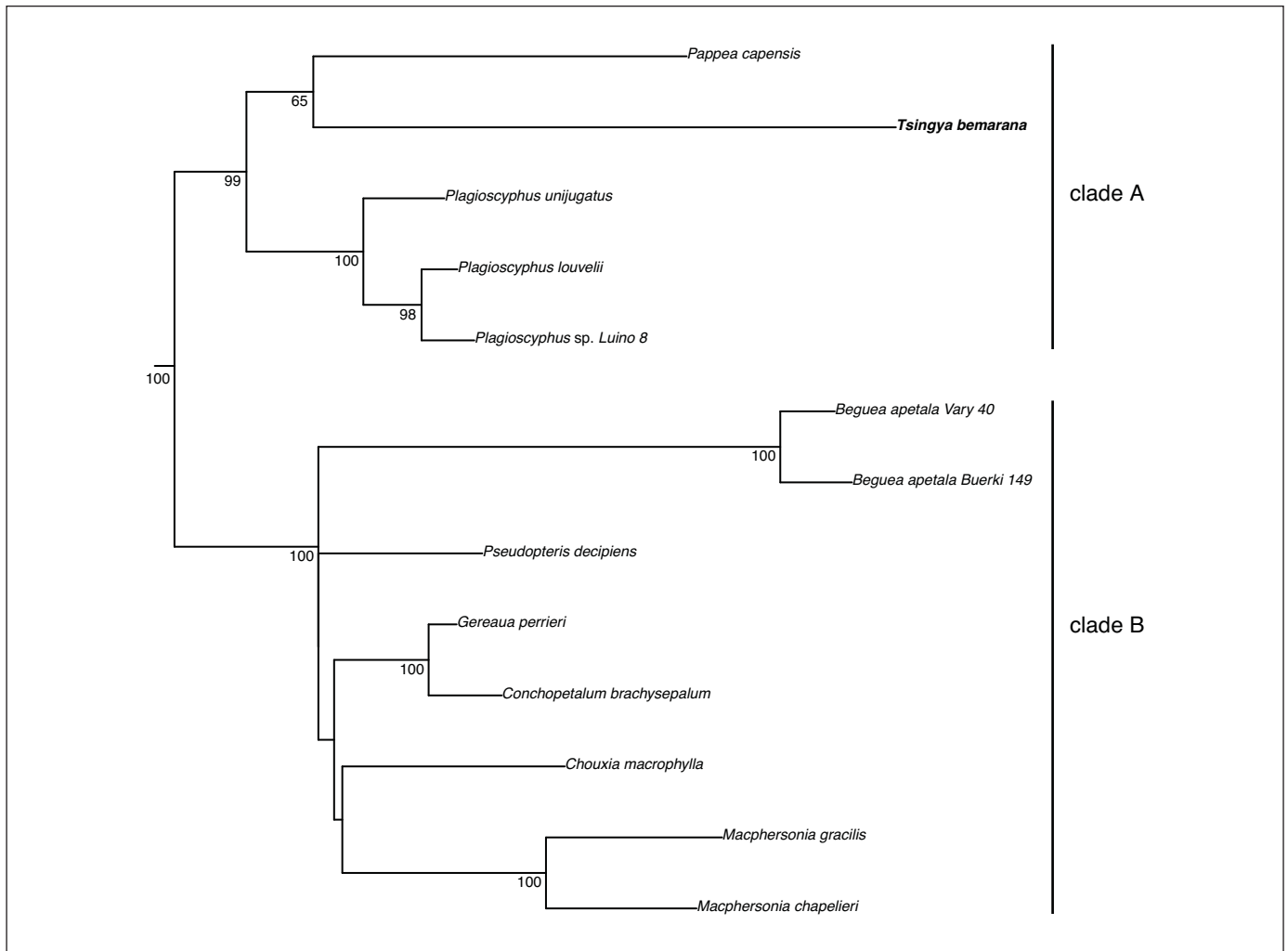
The ML inference strongly supported *Tsingya bemarana* as part of the “Macphersonia group”, which includes most of the Malagasy endemic genera of *Sapindaceae* (Fig. 1; see BUERKI & al., 2010 for an in depth discussion on these genera). This group is subdivided into two clades and *Tsingya* belongs to clade A (bootstrap support, hereafter BS: 99%) together with the South African *Pappea capensis* Eckl. & Zeyh. and the Malagasy *Plagioscyphus* (Fig. 1). Although the ML analysis suggests that *Tsingya* is closely related to *Pappea* Eckl. & Zeyh., this relationship is weakly supported (BS: 65%) and additional DNA regions will have to be sequenced to confirm this result. However, there are several key morphological traits allowing us to distinguish *Tsingya* from *Pappea*: e.g. paripinnate leaves in *Tsingya* (vs. simple in *Pappea*), monoecious breeding system (vs. dioecious) and absence of petals (vs. 4-6 petals). Overall, the phylogenetic inference is in agreement with the hypothesis made by CAPURON (1969) based on morphological data alone and supports *Plagioscyphus* (the monophyly of this genus is strongly supported with a BS of 100%) as the closest taxon to *Tsingya*. This result provides ample justification for the recognition of *Tsingya* (see below for a full description of the species) and may provide new insights towards the understanding of the diversification and biogeography of the endemic Malagasy angiosperm genera (see BUERKI & al., 2013).

## Taxonomy

*Tsingya bemarana* Capuron in Mém. Mus. Natl. Hist. Nat., sér. B, Bot. 19: 104. 1969. (Fig. 2).

**Typus:** MADAGASCAR. Prov. Mahajunga: Plateau calcaire du Bemahara, aux env. de Tsiandro, [18°39'55”S 44°43'42”E], XI.1952, galled fl. & imm. fr., *Service Forestier 6762* (holo-: P [P00076184]!; iso-: P [P00076183, P00214682] images seen, P [P00363062]!, TEF [TEF000597]!).

Monoecious trees with platanoid bark exfoliating in plates, 13-25 m tall, 0.2-0.5 m in diam., young branches covered with dense fascicled-stellate pubescence. Leaves alternate, 15-25 × 20-23 cm, paripinnately compound, (4-)6-8-foliolate; leaflets ovate to elliptic, margin entire, petiolulate (2-7 mm), sub-membranous, (5-)9-12(-16) × (2.5-)3-4(-5) cm, discolor, dark green and shiny above and pale green below; base attenuate; apex acuminate; margin entire. Inflorescences axillary, racemose, 4-7 cm in length. Flowers small, 3-4 mm in diam., regular; sepals 5, valvate; petals absent; disc flattened, irregularly lobed, 2-2.5 mm in diameter, stellate pubescent; stamens 8-10, c. 4 mm in length, inserted



**Fig. 1.** – Maximum likelihood phylogenetic tree showing the position of *Tsingya bemarana* Capuron in the “Macphersonia group”. Bootstrap support values are displayed below the branches.

into deep holes in the disc, filaments distinct; staminodia resembling short stamens; ovary 3-locular; style terminal, 3 mm in length, with stigmatic lines; ovule 1 per locule. *Fruit* indehiscent, pear-shaped berry, shortly apiculate, somehow asymmetrical, 3-3.5 × 2-2.5 cm, covered by stellate trichomes, 1-seeded by abortion, the seed surrounded by a fleshy, translucent arillode, and bearing a hilum scar the entire ventral length [adapted in part from CAPURON (1969) and SCHATZ (2001)].

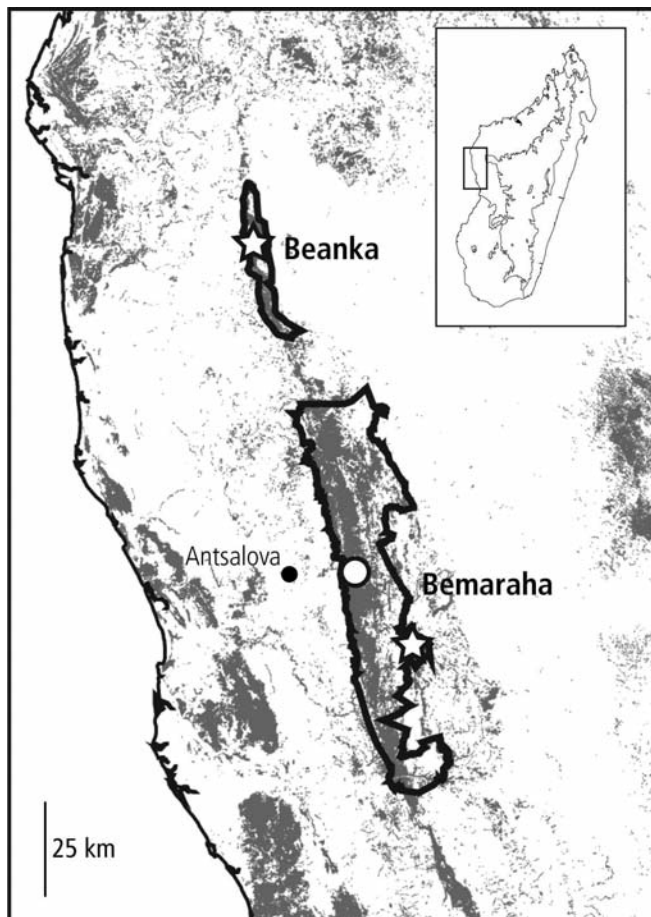
*Habitat and Ecology.* – The recent collection of *Tsingya bemarana* came from sample site B020 of the forest study at Beanka, which was characterized by forest cover of 100% on exposed eroded limestone rock, and by the presence of: *Pandanus flagellibracteatus* Huynh (*Pandanaceae*) and *Omphalea occidentalis* Leandri (*Euphorbiaceae*) forest, with closed

canopy and low emerging tree cover (RAKOTOZAFY & al., 2013). This forest type has an average canopy of 12.5 m, and the tree collected formed part of the canopy. The species was listed by BOLLIGER (2014) as one of the 58 species of Beanka that are apparently exclusive to limestone. It would seem to be restricted to the dry deciduous forests on eroded limestone (“tsingy”) of western Madagascar in the Bemaraha and Beanka regions (Fig. 3).

*Conservation status.* – Despite an intensive botanical inventory in the Beanka region, only one tree was found, and the species has not been collected from Bemaraha since 1952. Nevertheless, the forest of Behandrao, South of Tsiandro on the eastern slopes of Bemaraha (Fig. 3), where the type has been collected, is under protection and very few collections have been made in this region since the 1950’s. This is also



Fig. 2. – Herbarium scan of *Tsingya bemarana* Capuron collected by Hanitrarivo, Bolliger & Rakotozafy 167 kept at G showing the pyriform fruit.



**Fig. 3.** – Detailed map of western Madagascar showing the distribution of *Tsingya bemarana* Capuron (herbarium collections: stars; observation: circle) in Bemaraha and Beanka, plotted on a map of forest cover in 2000 (grey) following HARPER & al. (2007).

true for the Ambodiriana valley within the protected area to the west of Antsalova (Fig. 3). With only two collections known and one reliable field observation (LEANDRI, 1954), an “Extent of Occurrence” of 437 km<sup>2</sup>, an “Area of Occupancy” (AOO) of 27 km<sup>2</sup> and three subpopulations (calculation following CALLMANDER & al., 2007), within a fully Protected Area (Bemaraha) and a Protected Area that holds temporary protection (Beanka), *Tsingya bemarana* is assigned a preliminary status of “Vulnerable” (VU D2) following IUCN Red List Categories and Criteria (2012). Despite the fact that all known populations are currently under protection, *Tsingya bemarana* is rare and known only from three locations.

**Observations.** – In CAPURON (1969: 85)’s flower identification key to the *Schleicheraceae*, *Tsingya* keys out next to *Beguea* Capuron based on the absence of corolla. While working on a revision of *Beguea* for Madagascar, SCHATZ & LOWRY (pers. comm.) have therefore hypothesized that *Tsingya* may be conspecific with *Beguea*. In CAPURON (1969: 86)’s fruit identification key, *Tsingya* keys out next to *Plagioscyphus*

based on the hilum scar that runs on the entire ventral length of its seed. The phylogenetic inference is in agreement with the hypothesis that *Tsingya* is in fact closely related to *Plagioscyphus* in Madagascar (see above). The characters of the mature fruit of *Tsingya* recently discovered is in congruence with the phylogenetic evidence; *Tsingya* can easily be distinguished from *Beguea* by its pubescent pyriform fruits (vs. glabrous globular to ovoid) (Fig. 2). *Tsingya* differs from *Plagioscyphus* by the type of indument on its fruit (stellate trichomes in *Tsingya* vs. simple in *Plagioscyphus*) and the corolla (absent vs. present). The monoecious breeding system of *Tsingya* is also unique in the “Macphersonia group” (all the other genera are dioecious or polygamous; BUERKI & al., 2010) and represents a good morphological synapomorphy for this genus.

**Specimens examined.** – MADAGASCAR. Prov. Mahajanga: Bemaraha, forêt de Behandrao, [18°49’37”S 44°52’25”E], 29.XI.1952, LEANDRI & al. 1969 (= Service Forestier 6762) (P [P00076181, P00076182]); Beanka, Partie N, bord de la rivière Bokarano, 17°54’36”S 44°28’46”E, 222 m, 10.II.2012, fr., HANITRARIVO, BOLLIGER & RAKOTZAFY 167 (G [G00376555]), K, L, MO, P, TEF).

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