NEW OR LITTLE KNOWN EPIPHYLLOUS LIVERWORTS, XXI. *RADULA CAMERUNENSIS* SP. NOV. (RADULACEAE, JUNGERMANNIOPSIDA)

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Abstract: The new epiphyllous species *Radula camerunensis* was discovered in the high rainfall area of the lowlands of SW Cameroon, as host of the new parasitic ascomycete *Epibryon platycarpum* Döbbeler et T. Franke. As all the investigated species of the former section *Epiphyllae* proved to be monophyletic, belonging to the same subclade of subgenus *Metaradula* in a recent molecular investigation, the status of *Radula* sect. *Epiphyllae* is tenable. The new species differs from all other members of sect. *Epiphyllae* by its long stalked discoid gemmae.

Key words: Bryoparasitic fungi, Cameroon, discoid gemmae, Radula, sect. Epiphyllae

INTRODUCTION

Peter Döbbeler, renowned specialist of bryoparasitic fungi, called my attention to an epiphyllous *Radula* species hosting a new species of ascomycetes, *Epibryum platycarpum* Döbbeler et T. Franke (DöBBELER 2016). The new ascomycete occurred exclusively on this host and was absent on *Radula flaccida* Lindenb. et Gottsche, which occurred on the same phanerogamic leaves. The specimens were collected from the high rainfall area in the lowlands of SW Cameroon by Thassilo Franke, biologist and producer of natural history films. The *Radula* host species had peculiar discoid gemmae with several rows of smaller cells along their margin. Investigating the specimen, I found other unique features as well and concluded that it belonged to a hitherto unknown epiphyllous *Radula* species.

The leaf-inhabiting *Radula* species were classified by SPRUCE (1884–1885) in the subgenus *Acroradula* (now subgenus *Radula*), and by CASTLE (1939, as nomen nudum) in the separate sect. *Epiphyllae* Castle ex Grolle (GROLLE 1970). YAMADA (1979) followed the classification of Castle. SCHUSTER (1980*a*) in a phylogenetic study considered Castle's classification artificial, claiming that the sect. *Epiphyllae* was polyphyletic and included advanced representatives of different groups. He described within the group of epiphyllous *Radula* species three

new sections (*Evansiae*, *Mammosae*, *Acuminatae*) and within sect. *Acuminatae* three new subsections (*Stenocalyces*, *Yanoellae*, *Acuminatae*). Sect. *Epiphyllae* Castle ex Grolle emend. R. M. Schust., thus, became restricted to only two species, *R. flaccida* and *R. pseudoflaccida* E. W. Jones. In Schuster's classifications of the former *Epiphyllae*, sect. *Evansiae* and sect. *Mammosae* are never gemmiparous while the subsections of sect. *Acuminatae* and the two species of sect. *Epiphyllae* are all gemmiparous. The species of the subsect. *Stenocalices*, subsect. *Yanoellae*, and sect. *Epiphyllae* have marginal gemmae, while those of subsect. *Acuminatae* have superficial gemmae.

Molecular genetic investigations by DEVOS *et al.* (2011) did not support Schuster's 1980 classification. They established that all investigated species of sect. *Epiphyllae* are a monophylum and constitute a subclade of subgenus *Metaradula* R. M. Schust. (SCHUSTER 1984). They also supplied a new diagnosis for subg. *Metaradula*, which had several defects, stating among others that lobule insertion is parallel to stem, directed to stem apex (valid only for part of the species) and that caducous leaves and gemmae are absent (which is not true for the majority of the members of sect. *Epiphyllae*). Schuster's original diagnosis of subg. *Metaradula* seems to be more appropriate.

As the new species from Cameroon has marginal gemmae, I compared it with the gemmiferous species from the sect. *Epiphyllae*. One of them, the Amero-African *Radula flaccida* has very elaborate, funnel shaped, 'flying saucer' like gemmae, while all others have simple, flat or slightly convex discoid gemmae either in the plane of leaf lobe or perpendicular to it (Fig. 1B-F). In the new species the gemmae are slightly convex and in the plane of leaf lobe. The striking feature of the new species is the elongated stalk cell of the gemmae, developing mostly on the antical margin (Fig. 1A).

The asexual reproduction of bryophytes by means of gemmae has been studied in details by many authors since GOEBEL (1898, 1930), CAVERS (1910), and BUCH (1911). SCHUSTER (1980b) described in detail the development of complex, funnel form gemmae of *Radula flaccida*, observed earlier by GOEBEL (1930). RENNER and BRAGGINS (2004), discussing the gametophytic characters of the genus, showed that discoidal gemmae can be one or two cells thick, and that granular and roughly spherical gemmae occur in *Radula javanica* Gottsche. Only few authors, e.g. STEVENS (1910), paid attention to the development of simple discoid gemmae in the genus. He established that in *Radula complanata* gemma development starts with a protruding marginal leaf cell. Subsequently, the projecting portion is cut off by a wall. The inner cell is regarded as a stalk, the outer becomes the mother cell of the gemma. In the case of *R. complanata* the shape of the discoid gemma is quite irregular. After many cell divisions, when the gemma is ready to be separated, schizolytic splitting takes place in the wall



Fig. 1. Comparison of the gemma stalk of different *Radula* species with marginal gemmae in sect. *Epiphyllae.* – A = *Radula camerunenis* Pócs et Döbbeler, from the type. B = *Radula assamica* Steph.
C = *Radula nymanii* Steph. D = *Radula stenocalyx* Mont. E = *Radula tjibodensis* K. I. Goebel. F = *Radula yanoella* R. M. Schust.

between the stalk and the cells of the leaf. DEGENKOLBE (1938) showed the occurrence of a great variety of discoid gemmae in *Radula* and described the type of gemma occurring in the majority of sect. *Epiphyllae*, being regularly orbicular with smaller marginal cells in some cases.

The stalk cell in the gemmiferous species of sect. *Epiphyllae* is rounded, square or even compressed parallel to the leaf margin. In the new species, however, the stalk cell is elongate, already in juvenile stage, elevating the developing gemma above the leaf margin.

RESULTS

Radula camerunensis Pócs et Döbbeler, sp. nov. (Figs 2–5)

Diagnosis: *Radula camerunensis* differs from the other gemmiferous species of sect. *Epiphyllae* by its discoid gemmae having elongate stalk cells, which elevate the gemmae above the leaf margin. In addition, the new species has ligulate, forward directed lobules, without a pronounced mammilliform ventral sac.

Type: Cameroon, Southwest Province: Bimbia-Bonadikombo Forest Reserve (Mabeta-Moliwe Forest Reserve), lowland rainforest at sea level, with 5000 mm annual precipitation, epiphyllous, hosting the bryoparasitic ascomycete *Epibryum platycarpum* Döbbeler et T. Franke, which, being small, does not affect the growth of its host. *Radula camerunensis* thrives together with *Radula flaccida*, *Cololejeunea pusilla*, and *Cololejeunea microscopica* var. *africana*, none of them infected by the fungus. Coll.: Thassilo Franke, 27.11.2000, Döbbeler No. 8784 (holotype: EGR, isotype: NY, under the fungal name of *Epibryum platycarpum*, paratypes from the same collector, locality, and date: Döbbeler No. 8779, M; 8780 (holotype of *E. platycarpum*, M); No. 8789, TUR; No. 8778, B).

Description: In herbarium olive green, forming several cm² large colonies on living leaves of phanerogams, sometimes covering their whole surface. Many cases mixed with other epiphyllous liverworts, but easy to distinguish by the often developing, stalked discoid gemmae. Shoots irregularly branching, up to 10–20 mm length and 1.4–1.8 mm width. Stem width 50–64 μ m, in section 4–5 cells wide with 4 thin-walled medulla cells surrounded by 8–12 cortical cells with moderately thickened outer walls (Fig. 1*B*). This stem structure is typical for sect. *Epiphyllae* according to JONES (1977), belonging to his type '5'. Cell walls pale yellowish brown pigmented. Leaves moderately imbricate, lobe broadly ovate with widely rounded apex, 1–2.4 × 0.8–1.8 mm, nearly plane, dorsal base arching and with about ¼ of lobe length crossing the stem, dorsal base decurrent. Median cells 17–25, basal 25–40, marginal 8–10 × 6–10 µm, their wall thin with small trigones and intermediate thickenings. Oil bodies already disintegrated in the examined samples. Cuticle smooth. Keel straight or slightly curved, about ¼ of lobe length. Lobule broad ligulate or triangular, 320–480 μ m long with obtuse apex, at about half of its length fused and directed parallel to the stem, without large mamilliform ventral sac. It has only a dot-like (30–40 μ m diameter) rhizoid initial area with small (4–6 μ m) cells giving origin to the short (35–70 μ m), radially arranged, colourless rhizoids often dichotomic at their end. Other median cells 14–20 × 12–16 μ m, marginal cells 10–16 × 4–6 μ m.

Dioicous. Male gametoecia on apex of the mean stem and side branches, often forming a raceme at shoot ends. Male spikes up to 2 mm length and 0.4 mm width, consisting of 10–20 pairs of slightly falcate, 280 μ m long and 240 μ m wide bracts with hypostatic lobules almost equalling the lobe length. The male spike sometimes ends in a normal leafy shoot. Female gametoecia on shoot apex or branches, usually with one innovation. Perichaetial leaves not specially differentiated. Perianth very narrowly conical, trumpet shaped, 1–2 mm long and 0.3–0.6 mm wide at its undulate mouth. The lower 3/5 of perianth built up of several layers, subtended by a stem-perigynium with cells of the outer wall oblong rectangle or sigmoid, 30–50 × 10–20 μ m, while the upper 2/5 of perianth is unilayered with irregular, near isodiametric cells of 16–30 × 10–20 μ m size. Sporophyte consists of a 2–2.5 mm long seta and a narrow cylindrical capsule, with 600–750 μ m long and 40 μ m wide valves with 4–8 rows of 80–160 μ m long and 12 μ m wide, elongate, brown pigmented cells in their outer walls.

Vegetative reproduction by discoid gemmae, developing at the antical margin of leaf or rarely on the male bract lobe. Their stalk cell from the beginning of its growth already elongate, slightly widens upwards, 25-36 µm long, keeping away the gemma from the lobe margin. The gemmae reach on the mother plant 250–400 μ m diameter but detached, mature gemmae 560–640 \times 600–640 μ m, reaching the size of smaller leaves, somewhat reniform, having shallowly notched base at the insertion point of stalk, which finally detaches from the body of gemma. Supposedly the gemmae continue their growth after detachment, as this large size never occurs until they are attached to the leaf. Mature gemmae slightly convex (under pressure of coverslip they crack) and in the plane of the leaf lobe; in cross section they are generally unilayered but here and there can be two cells thick. The median cells are isodiametric, polygonal, 13–25 µm diameter while 5–8 rows of marginal cells are smaller, quadrate, only 6–12 μ m, forming a rim around the large celled, 12-18 cells wide interior part. In cross section the median cells are much higher than the marginal ones, hence the gemma is thinning towards its margin. Unfortunately, I was not able to observe germinating gemmae except one, which gave birth to another, sprouting gemma.



Fig. 2. Radula camerunensis Pócs et Döbbeler. – A = Habit, ventral view, and two detached, enlarged gemma. B = Stem, transversal section. C = Lobe base, with the rhizoid initial area comparing its small cells with those of the lobe and lobule. D = Median transversal section of a disciform gemma not yet released from the lobe margin. E = Outer layer of the lower part of perianthium. F = Single layer of the upper part of perianthium. All drawn from the type.



Fig. 3. *Radula camerunensis* Pócs et Döbbeler. – A = Habit, ventral view. B = Leaf, ventral view. C = Median lobe cells. D and E = Stalked gemmae on the antical lobe margin. All drawn from the type.



Fig. 4. Radula camerunensis Pócs et Döbbeler. – A = Margin of a detached gemma. B = Young gemma and gemma initials on the antical lobe margin. C = Mature, already detached gemma. Being concave, cracked when flattened under the pressure of coverslip. D = Stalked base of a maturing gemma. E = Raceme on a shoot end formed by male branches. F = Part of a male branch. All drawn from the type.



Fig. 5. Radula camerunensis Pócs et Döbbeler. – A = Male branch with a gemma bearing enlarged lobus. B = Capsule valves. C = Perianth mouth. D = Relatively small, young perianth. All drawn from the type.

DISCUSSION

According to our present knowledge sect. Epiphyllae Castle ex Grolle contained 13 species, if we do not mention other facultative epiphyllous species as Radula lindenbergiana Gottsche et C. Hartm. (Pócs 1982) in the Caucasus Mts or Radula cavifolia Hampe ex Gottsche et al. From the 13 epiphyllous Radula 3 lack gemmae, while 10 are gemmiferous, including the recently described neotenic, semi-thalloid Radula yanoella R. M. Schust. and R. aguirrei R. M. Schust. (CASTLE 1939, GROLLE 1970, SCHUSTER 1980a, 1991) from the Neotropics and the Asian Radula grandilobula Promma et Chantanaorrapint (PROMMA and CHANTANAORRAPINT 2015). With the description of the new species R. camerunensis Pócs et Döbbeler, the number of species in sect. Epiphyllae is raised to 14. According to the traditional view sect. Epiphyllae belonged to subgenus Acroradula Spruce (present subg. Radula), according to recent molecular investigations (DEVOS et al. 2011) to subg. Metaradula R. M. Schust. DEVOS et al. also showed that the gemmiferous members of sect. Epiphyllae are a monophylum, forming a separate subclade of subg. Metaradula characterized also by the presence of gemmae and their highly reduced stem anatomy (JONES 1977). As the investigated species of the former section *Epiphyllae* proved to be monophyletic, within the same subclade of subgenus Metaradula according to recent molecular investigation, the status of Radula sect. Epiphyllae is tenable. Among the gemmiferous members of the *Epiphyllae* the new species is unique by its elongated stalk cell, which elevates the discoid gemmae above the lobe margin (in the plane of lobe). The large median cells of the gemma are surrounded by several rows of smaller and thinner marginal cells. This characteristic is shared with Radula tjibodensis and with R. nymanii, but its ligulate, forward directed lobule differentiates the new species from them. In addition, the gemmae of *R. tjibodensis* are perpendicular to the lobe surface and those of R. nymanii are developing on the ventral (postical) lobe margin (SINGH et al. 2016, Fig. 2). Therefore, the taxonomic position of the new species seems to be quite separate from the other members of *Epiphyllae*. The host specificity of the parasitic ascomycete supports this. It is interesting that the new African species is morphologically closest to two Asiatic species. This may reflect the importance of stochastic long range dispersal in the Epiphyllae (e.g. PATIÑO et al. 2016) and suggest that greatly restricted distribution does not necessarily mean lower diversification rates.

Conservation aspects: As the bryoflora of Cameroon is poorly known, mostly from old German collections, the discovery of the new species does not come as a surprise. However, as the lowland forests in West Africa are very much decimated, the coincidence of a new ascomycete exclusively parasitising a new species of liverworts is noteworthy and may indicate the richness and uniqueness of the habitat. Bimbia-Bonadikombo Community Forest Reserve in the high rainfall area of the Southwest Province of Cameroon near Limbe town has been known for its high biodiversity with eight threatened vertebrate species including chimpanzees, and merits special attention and a high level of protection. Due to the surrounding population pressure, it will not be an easy task, but promising measures have already been taken (NGALIM and TERENCE 2016).

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Összefoglaló: Peter Döbbeler mikológus (München), a mohalakó mikrogombák ismert kutatója hívta fel a figyelmemet egy érdekes epiphyll (levéllakó) *Radula* fajra a kameruni sík vidéki esőerdőkből, Thassilo Franke gyűjtéséből. Ez a *Radula* faj a kizárólagos gazdanövénye az általa leírt, tudományra új *Epibryon platycarpum* Döbbeler et T. Franke mohaparazita gombának (DöBBELER 2016), bár vele együtt egy másik *Radula* faj is tömegesen előfordul ugyanazokon a leveleken. Anyagát tanulmányozva ez a faj *Radula camerunensis* Pócs et Döbbeler néven, mint a tudományra új faj került leírásra. Az új faj egyedülálló tulajdonsága a *Radula* fajok között, hogy discoid gemmáinak (vegetatív szaporító testek) nyélsejtje megnyúlt és a gemmát eltartja a levélke szélétől. Ezáltal a 14., trópusi esőerdőkben élő epiphyll *Radula* faj vált ismertté. A közelmúltban végzett molekuláris vizsgálatok (DEVOS *et al.* 2011) igazolták CASTLE (1939) nyomán GROLLE (1970) felfogását a monofiletikus *Epiphyllae* szekció önállóságáról és létjogosultságáról, szemben Schuster későbbi (SCHUSTER 1980*a*) felfogásával, aki ezt az egységet polifiletikusnak tartotta és számos szekcióra bontotta. A mai felfogás szerint az *Epiphyllae* szekció a *Metaradula* R. M. Schust. alnemzetségbe tartozik (DEVOS *et al.* 2011), bár e szerzők *Epiphyllae* diagnózisának megfogalmazása hibás.

REFERENCES

- BUCH, H. (1911): Die Brutorgane der Lebermoose. PhD Thesis, Alexander University, Helsingfors, 70 pp.
- CASTLE, H. (1939): A revision of the genus Radula. Part II. Subgenus Acroradula. Section 1. Epiphyllae. Ann. Bryol. 12: 21–47.
- CAVERS, F. (1910): On asexual reproduction and regeneration in Hepaticae. *New Phytol.* 2: 121–133, 155–165.

DEGENKOLBE, W. (1938): Brutorgane bei beblätterten Lebermoosen. - Ann. Bryol. 10: 43-96.

DEVOS, N., RENNER, M. A. M., GRADSTEIN, S. R., SHAW, J. and VANDERPOORTEN, A. (2011): Molecular data challenge traditional subgeneric divisions in the leafy liverwort Radula. – *Taxon* **60**(6): 1623–1632.

DÖBBELER, P. (2016): Three new ascomycetes on epiphyllous liverworts. - Karstenia 56: 47-54.

GOEBEL, K. (1898): Organographie der Pflanzen, 2. Teil, 1st ed. – Gustav Fischer, Jena, 698 pp.

- GOEBEL, K. (1930): Organographie der Pflanzen, 2. Teil, 3. A. 3rd ed. Gustav Fischer, Jena, pp. 622–1471.
- GROLLE, R. (1970): Radula castlei sp. nov. und Anmerkungen zur Gattung Radula. *Bryologist* 73: 662–668. https://doi.org/10.2307/3241278
- JONES, E. W. (1977): African Hepatics XX. The genus Radula Dumortier. J. Bryol. 9: 461–504. https://doi.org/10.1179/jbr.1977.9.4.461
- NGALIM, R. N. M. and TERENCE, S. (2016): The Bimbia-Bonadikombo Community Forest, South West Region of Cameroon: biodiversity potentials, problems and prospects, 1. – Int. J. Forestry and Horticulture 2(3): 5–18. https://doi.org/10.20431/2454-9487.0203002
- PATIÑO, J., WANG, J., RENNER, M. A., GRADSTEIN, S. R., LAENEN, B., DEVOS, N., SHAW, A. J., and VANDERPOORTEN, A. (2016): Range size heritability and diversification patterns in the liverwort genus Radula. – *Mol. Phyl. Evol.* **106**: 73–85. http://dx.doi.org./10.10166j.ympev.2016.09.020
- Pócs, T. (1982): An epiphyllous liverwort community from the Caucasus Mountains. Bryol. Beiträge 1: 13-22.
- PROMMA CH. and CHANTANAORRAPINT, S. (2015): The epiphyllous Radula (Radulaceae, Marchantiophyta) in Thailand, with the description of Radula grandilobula sp. nov. – *Cryptogamie*, *Bryol.* 36(3): 217–234.
- RENNER, M. A. M. and BRAGGINS, J. E. (2004): The sterile gametophyte as a source of informative characters in the genus Radula (Radulaceae, Hepaticae). *Nova Hedwigia* 78(1–2): 243–268. https://doi.org/10.1127/0029-5035/2004/0078-0243
- SCHUSTER, R. M. (1980*a*): Phylogenetic studies on Jungermanniidae II. Radulinae (Part I). *Nova Hedwigia* **32**: 637–693.
- SCHUSTER, R. M. (1980b): The Hepaticae and Anthocerotae of North America. Vol. 4. Columbia University Press, New York, 1334 pp.
- SCHUSTER, R. M. (1984): Diagnoses of some new taxa of Hepaticae. Phytologia 56(2): 65-74.
- SCHUSTER, R. M. (1991): On neotenic species of Radula. J. Hattori Bot. Lab. 70: 51-62.
- SINGH, D. K., DEY, M. and SINGH, D. (2016): The genus Radula in the Andaman and Nicobar Islands, India. – J. Bryol. 38(3): 195–210. http://dx.doi.org./10.1179/1743282015Y.0000000032
- SPRUCE, R. (1884–1885): Hepaticae of the Amazon and the Andes of Peru and Ecuador. London. (reprinted in 1984 as *Contrib. New York Bot. Garden* 15: XII+588 pp.).
- STEVENS, N. E. (1910): Discoid gemmae in the leafy hepatics of New England. Bull. Torrey Bot. Club 37: 365–373.
- YAMADA, K. (1979): A revision of Asian taxa of Radula, Hepaticae. J. Hattori Bot. Lab. 45: 201–322.

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