MYCENA LACRIMANS, A RARE SPECIES FROM AMAZONIA, IS BIOLUMINESCENT

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Mycena lacrimans (Agaricales, Mycenaceae) is redescribed from material collected recently near the type locality and is for the first time reported as bioluminescent. A comprehensive description, illustrations, photographs, comparison with phenetically similar species, and discussion of phylogenetic relationships are provided.

Keywords. Basidiomycete, conservation, mycenoid fungi, taxonomy.

INTRODUCTION

Mycena lacrimans Singer was originally described from a single basidiome collected on an undetermined dicotyledonous fallen leaf at Reserva Ducke (a reserve of the Instituto Nacional de Pesquisas da Amazônia, INPA) near Manaus in Amazonas State, Brazil. A comprehensive description in Latin was published (Singer, 1989), but no illustrations of salient micromorphological features, photographs, comparison with similar species or indication of infrageneric taxon for the species were provided. More significantly, the species was not reported as luminescent. Until recently, the species had been known only from this single basidiome. Basidiomes of this poorly known species have now been collected along highway BR-319 approximately 120 km south of Reserva Ducke. This new material allows us to supplement the original description with additional taxonomically significant data, illustrations and photographs, and provides quality DNA for sequencing and phylogenetic analyses. We report for the first time that *Mycena lacrimans* is strongly luminescent on the stipe apex.

MATERIALS AND METHODS

Fresh material was collected, described when fresh, photographed in light and dark with a Nikon D70 digital camera with a 92 mm F/2.8D macro lens, and dried for preservation. Micromorphological features were described from dried material rehydrated in 100% ethanol followed by either distilled water, 3% KOH, Congo red plus KOH, or Melzer's reagent. Spore statistics include: x_m , the arithmetic mean of the spore length by spore width for *n* spores measured in a single specimen,

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reported as $x_{\rm mr}$, the range of spore means, and $x_{\rm mm}$, the mean of spore means (± standard deviation) for two specimens examined; Q, the quotient of spore length and spore width in any one spore, indicated as a range of variation in *n* spores measured; $Q_{\rm m}$, the mean of Q values in a single specimen, reported as $Q_{\rm mr}$, the range of $Q_{\rm m}$ values, and $Q_{\rm mm}$, the mean of $Q_{\rm m}$ values for two specimens examined. Specimens are deposited in INPA and SFSU.

Results

Mycena lacrimans Singer, Fieldiana: Bot., n.s. 21: 78 (1989). – Type: Brazil, Amazonas State, Manaus, Reserva Forestal Ducke, 4 xii 1978, *Singer* B 11429 (holo INPA [82.356]). Figs 1, 2.

Basidiomes (Fig. 1) mycenoid, fragile. *Pileus* 1–6 mm in diameter, convex to convex with applanate or shallowly depressed disc, sulcate with scalloped margin, glabrous when fresh, minutely resinous-granulose when dried, dull, dry (not viscid), slightly subhygrophanous, when young pale greyish white with a hint of greyish orange on the disc, in age disc and sulcae pale greyish brown or paler with a nearly white margin; drying grey to greyish yellow. *Context* very thin. *Lamellae* adnate to arcuate or shallowly subdecurrent, distant to subdistant (10–15) with 1 (rarely 2) series of lamellulae, moderately broad, white to creamy white; edge concolorous with face, resinous-granulose when dried. *Stipe* 8–30 × 0.2–0.7 mm, central, terete, cylindrical above with a slightly enlarged base, subinsititious, pliant, dull to shiny, dry, subhygrophanous, glabrous to very minutely pruinose, white overall when young, base darkening to pale greyish orange; pale yellow when dried.

Basidiospores (Fig. 2A) (6–)6.5–8.5 × 3.2–4.5 μ m [x_{mr} = 7.3–7.9 × 3.8–3.9 μ m, $x_{\rm mm} = 7.6 \pm 0.44 \times 3.8 \pm 0.04 \,\mu{\rm m}, Q = 1.8-2.4, Q_{\rm mr} = 1.9-2.1, Q_{\rm mm} = 2.0 \pm 0.1,$ n = 10-25 spores per two collections], ellipsoid to pip-shaped, smooth, hyaline, amyloid, thin-walled. Basidia (Fig. 2B) 19–24 \times 5.4–6.5 µm, clavate, hyaline, 4-spored, with sterigmata up to 6.5 µm long. Basidioles clavate. Pleurocystidia absent. Lamellar edge heteromorphous. Cheilocystidia (Fig. 2C) abundant, subcylindrical to clavate or broadly clavate; main body $27-40 \times 6.5-12 \mu m$, apex covered with relatively large and knob-like spinulae $1.2-5 \times 1.2-2.5 \mu m$, upper 70% below apex with smaller, relatively dense spinulae $0.5-1 \times 0.5 \ \mu m$, base of cells smooth, thinwalled, hyaline to pale yellow in 3% KOH, non-amyloid, non-gelatinous but exudative, with amorphous to small-plaque-like, pale yellowish brown exudates adhered to apices in H₂O, exudates soluble in KOH and Melzer's. Pileipellis a very thin cutis of several cell layers thick overlaying a thick hypodermium. Pileipellis hyphae repent, interwoven, 2–5 µm in diameter, smooth, highly branched, thin-walled, non-gelatinous to subgelatinous, giving rise to terminal cells (Fig. 2D) $6.5-12 \mu m$ in diameter that are densely spinulose over terminal 50–75% of the cell and smooth at the base (similar to the cheilocystidia), hyaline, non-amyloid, thinwalled or with walls up to 1 mm thick, non-gelatinous but exudative. Hypodermium of



F1G. 1. Photographs of *Mycena lacrimans* (*Braga-Neto* 25) taken in natural light and in the dark under its own illumination. Natural light photographs 200 ASA, F/8–F/13, 1/15 second exposure; luminescence photographs 400 or 800 ASA, F/9–F/16, 30 second exposure. Scale bar: 5 mm for upper row left and right; 3 mm for all other photographs.



F1G. 2. Micromorphological features of *Mycena lacrimans*: A, basidiospores (a, *Braga-Neto* 25; b, holotype); B, basidia and basidiole (*Braga-Neto* 25); C, cheilocystidia (a, *Braga-Neto* 25; b, holotype); D, terminal cells of pileipellis hyphae (a, *Braga-Neto* 25; b, holotype); E, caulocystidia (*Braga-Neto* 25). Scale bar: 10 μm.

fusoid to vesiculose cells $12-30 \ \mu\text{m}$ in diameter, weakly dextrinoid, thin-walled, nongelatinous to subgelatinous, with pale brown to fuscous plasmatic pigments. *Pileus trama* hyphae interwoven, cylindrical, dextrinoid. *Lamellar trama* regular, dextrinoid. *Stipe tissue* monomitic; cortical hyphae $2-4 \ \mu\text{m}$ in diameter, parallel to subinterwoven, cylindrical, branched, smooth, hyaline, dextrinoid, thin-walled, subgelatinous, giving rise to caulocystidia; medullary hyphae similar but parallel and up to 20 $\ \mu\text{m}$ in diameter. *Stipitipellis* of erect to repent caulocystidia; *caulocystidia* (Fig. 2E) similar to the cheilocystidia, $32-58 \times 5-12 \mu m$, cylindrical to subclavate, spinulose with apical spinulae slightly larger than lateral spinulae (but not usually as large as those on the cheilocystidia), smooth over lower portion of cell, hyaline, non-amyloid, non-gelatinous, exudative. *Clamp connections* observed on basidia, cystidia, pileipellis and stipitipellis hyphae, absent elsewhere.

Bioluminescence. Intense, bright greenish yellow at stipe apex, weakening downwards towards the stipe base, weak on pileus disc from transmitted light generated at stipe apex, and non-luminescent elsewhere (Fig. 1).

Habit, habitat and distribution. Scattered on fallen dicotyledonous leaves and small twigs in terra-firme evergreen forests, more common near water bodies (*igapós* – flooded forests, and *igarapés* – small creeks). Brazil, Amazonas State.

Specimens examined. BRAZIL. Amazonas State, Manaus, Reserva Forestal Ducke, 4 xii 1978, Singer B 11429 (holo INPA [82.356]); along federal highway BR-319 near km 83, 3°37'12.7"S, 60°12'44.8"W, 15 v 2005, *R. Braga-Neto* 25 (INPA, SFSU).

Notes. Mycena lacrimans is a unique species that is not easily placed in any currently recognized infrageneric group. It is characterized by the presence of densely spinulose, cylindrical to clavate, exudative cells as cheilocystidia and as terminal cells of the pileipellis and stipitipellis. We presume that Singer (1989) chose the epithet (*lacrima* = tear) to reflect the exudative property of the cystidia. The apical spinulae of these cells are knob-like and much larger than the lateral rod-like spinulae, and the cells arise from smooth, non-spinulose hyphae. In addition, the pileipellis and stipitipellis hyphae that generate these exudative cells are subgelatinous, very narrow and interwoven, the species forms a well-developed dextrinoid hypodermium of vesiculose to inflated cells, the lamellar trama hyphae are dextrinoid, and the spores are amyloid. In combination, these features suggest affinity to Mycena sect. Aspratiles Maas Geest. & de Meijer (1997). Mycena aspratilis Maas Geest. & de Meijer, the only member of the section, differs from *M. lacrimans* in having thick-walled $(1-2.5 \,\mu\text{m})$ densely spinulose cystidia that lack larger apical spinulae, and in forming conspicuously diverticulate pileipellis and stipitipellis hyphae. Moreover, pleurocystidia are present, none of the cystidia was reported as exudative, and the species was not reported as luminescent. Maas Geesteranus & de Meijer (1997) suggested that Mycena sect. Aspratiles was allied with Mycena sect. Polyadelphia but differed mainly in the presence of thick-walled cystidia in the former. Mycena lacrimans differs significantly from members of Mycena sect. Polyadelphia in that all members of the latter section have pileipellis and stipitipellis hyphae that are densely spinulose and non-gelatinous, and that the cystidia are non-exudative. The phylogenetic placement of *Mycena lacrimans* in parsimony and Bayesian analyses of a preliminary molecular dataset generated from nuclear large ribosomal subunit (nLSU) rDNA sequences of over 100 species of mycenoid fungi (including 60 Mycena sensu lato species representing 26 sections of the genus) was equivocal (data not shown; Desjardin, Moncalvo and Perry, unpublished). We hope to resolve the phylogenetic relationships of this interesting species during the course of our on-going molecular studies utilizing a multi-locus approach (Desjardin, Moncalvo and Perry – NSF Grant No. DEB-0542445).

IMPLICATIONS FOR CONSERVATION

Highways have been a major determinant of deforestation in the Amazon (Laurance et al., 2001, 2002; Nepstad et al., 2001), causing loss of biodiversity and disturbance of environmental processes such as water cycling and carbon storage (Fearnside, 2005). The intention of Brazil's Federal Government to reconstruct BR-319, a degraded highway that links Manaus to Porto Velho, is a major conservation concern in the state of Amazonas (Fearnside & Graça, 2006). Efforts are being made to pave the route, although economic benefits of improved transportation seem to be illusory; for example, shipment of such freight by trucks is much less efficient in terms of energy use and labour costs than transportation by ship (Fearnside & Graça, 2006). It is well known that the existence of protected areas can significantly slow the advance of deforestation and biodiversity losses in the Amazon (Ferreira et al., 2005). However, there are currently only a few protected reserves along highway BR-319 (Fearnside & Graça, 2006). Most of the unknown global fungal diversity is expected to be in the tropics (Lodge et al., 1995; Hawksworth & Rossman, 1997; Hawksworth, 2001), and fungi are poorly known in these regions. We found the rare and exciting Mycena lacrimans with its luminescent basidiomes only from areas along BR-319 that are currently not under protection. We emphasize the necessity of reserves within areas of great scientific interest along the BR-319 route, and strongly encourage the creation and implementation of such reserves.

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REFERENCES

FEARNSIDE, P. M. (2005). Deforestation in Brazilian Amazonia: History, rates and consequences. *Conservation Biol.* 19: 680–688.

- FEARNSIDE, P. M. & GRAÇA, P. M. L. A. (2006). BR-319: Brazil's Manaus–Porto Velho highway and the potential impact of linking the arc of deforestation to Central Amazonia. *Environm. Managem.* 38: 705–716.
- FERREIRA, L. V., VENTICINQUE, E. M. & ALMEIDA, S. S. (2005). O desmatamento na Amazônia e a importância das áreas protegidas. *Estudos Avançados* 19: 1–10.
- HAWKSWORTH, D. L. (2001). The magnitude of fungal diversity: the 1.5 million species estimate revisited. *Mycol. Res.* 105: 1422–1432.
- HAWKSWORTH, D. L. & ROSSMAN, A. Y. (1997). Where are all the undescribed fungi? *Phytopathology* 87: 888–891.
- LAURANCE, W. F., COCHRANE, M. A., BERGEN, S., FEARNSIDE, P. M., DELAMÔNICA, P., BARBER, C. *et al.* (2001). The future of the Brazilian Amazon. *Science* 291: 438–439.
- LAURANCE, W. F., ALBERNAZ, A. K. M., SCHROTH, G., FEARNSIDE, P. M., BERGEN, S., VENTINCINQUE, E. M. & COSTA, C. (2002). Predictors of deforestation in the Brazilian Amazon. J. Biogeogr. 29: 737–748.
- LODGE, D. J., CHAPELA, I., SAMULES, G. et al. (1995). A survey of patterns of diversity in non-lichenized fungi. *Mitt. Eidgenössisch. Forschungsanstalt für Wald, Schnee und Landschaft* 70: 157–173.
- MAAS GEESTERANUS, R. A. & DE MEIJER, A. A. R. (1997). Mycenae paranaenses. Kon. Ned. Akad. Wetensch. Verh., Afd. Natuurk., Tweede Reeks, deel 97: 1–164.
- NEPSTAD, D. C., CARVALHO, G., BARROS, A. C., ALENCAR, A. A., CAPOBIANCO, J. P., BISHOP, J. *et al.* (2001). Road paving, fire regime feedbacks, and the future of Amazon forests. *Forest Ecol. Managem.* 154: 395–407.
- SINGER, R. (1989). New taxa and new combinations of Agaricales (Diagnoses Fungorum Novorum Agaricalium IV). *Fieldiana: Bot.*, n.s., 21: 1–133.

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