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BRIEF ARTICLES

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NOMURAEA VIRIDULUS, A NEW ENTOMOGENOUS FUNGUS FROM TAIWAN

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The genus *Nomuraea* erected by Kish, Samson and Allen (1974) was characterized by the formation of mono- or synnematus conidiophores bearing verticils of metulae and phialides in compact clusters encircling the stalk mostly below the septa; phialides usually in whorls, broadly cylindrical or sometimes with a swollen base, neck very short or absent; and conidia green to slightly purple in mass (Samson, 1974). Samson (1974) recognized two species, *N. rileyi* (Farlow) Samson and *N. atypicola* (Yasuda) Samson; in both species the teleomorphic state remains unknown. *N. rileyi* was entomopathogenic with a potential for biocontrol of insect pests, particularly lepidopterans (Ignoffo et al., 1976; Ignoffo and Garcia, 1985, 1989), whereas *N. atypicola* was a spider parasite, common in gardens and roadsides in Japan (Samson, 1974). Hocking (1977) described a new species, *N. anemonoides* Hocking from Australian soil, which contrasts with the previously described *Nomuraea* species by its synnematus rugulose conidophores bearing irregular verticils of phialides alone or metulae and phialides at most septa along the whole length of the conidiophore, and also by globose, ellipsoidal, or ellipso-pedunculate conidia. *N. anemonoides*, although probably a saprophyte, can be lethal to lepidopteran larvae when exposed at high dosage (Ignoffo and Garcia, 1989).

In the course of a study of entomogenous fungi from Taiwan, on many occasions a distinctive fungus was found frequently on cicada (*Cryptotympana facialis* Walker) cadavers. The specific fungus turned out to be a *Nomuraea* sp. but produced huge greenish, cylindrical, slightly curved or allantoid conidia, which can be readily

distinguished from known *Nomuraea* spp. and thus it is described as new. Illustrations and diagnosis were based on cultures on malt-extract agar (MEA) and on cicadas as well. Color nomenclature for colonies, mycelia and conidia was adapted from the color standard of Kornerup and Wanscher (1978). Preparation of microscopic structures for scanning electron microscopy followed previously described methods (Tzean and Estey, 1978).

Nomuraea viridulus Tzean, Hsieh, Chen et Wu,
sp. nov. FIGS. 1–10

Coloniae in maltis extractis agaribus ad 25 C in 14 diebus, crescentes, lente attingentes 1.5–2.3 cm diametro, centrales velutinae, zonatae, fairnosae, praesertim distinctae ubi sporulae graves, cinerascens virides, ad obscure virides, margine floccosae albae. Reversum centrum luteolum ad luteolum brunneum, subcentrum ad marginem obscure viride ad incolorem. Odor diffusis. Pigmentum demptum exsudatum liquidum. Synnemata absens. Mycelium ramosum septatum, laevis paries, album 2.5–3.8 μm latum. Conidiophorum ascendens mazime saepe ab aereo, subinde ab superficie laevis paries, hyalinum, longitudine usque ad 210 μm , verticillatum, ramosum vel phialis vel saepe cum irregulari subterminali, laterali vel infero plano ramoso, 1–2 vel in verticillato orienti prope septum; rami late clavati, clavati vel cylindrici 11.7–31.6 \times 2.6–3.8 μm ; phiales adpressae 3–8 per ramum vel subinde solitariae portatae directae in conidophoris, ovalis-cylindrica, ellipsoideae. Collum absens vel vix manifestum 5.3–9.2 \times 3.3–4.6 μm . Conidia catenulata non septata, laevis-paries, cylindrica, ellipsoidea, plerumque leviter curva vel allantoida, separata extrema aliquando apiculata, solitaria, cinerascens, lutea, in massis obscure cinerascens viridia ad perviridia 14.4–19.4 \times 3.8–4.4 μm . Chlamydospora absens. In insecti hospite, mycelium primum album, floccosum, tegens sutura, invicem ad viride caesiam ad pallidum viride, velutinum, in aetate, cinerascens viride ad hebetem viridem cum gravi sporulato, ramosum. Phiales et conidia plus minusve pingua, sed breve qum illa in cultura; constitantia 4–12, phiales per ramum, ramus oriens non limitatus ab septo, subinde proliferatus novus

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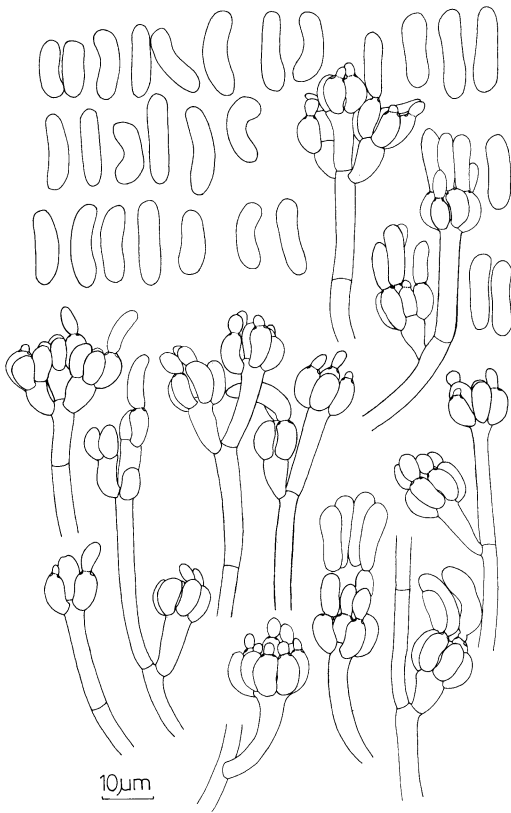


FIG. 1. *Nomuraea viridulus*. Characteristics of conidiophores, metulae, phialides and conidia in culture media (MEA).

ramus. Conidiophora subliliter aspra ad aspros paries. Synnemata et chlamydospora absens.

HOLOTYPE: in *Cryptotympana facialis* Walker, Homoptera, Hsintien, Taipei, Taiwan, R.O.C., 22 June 1989, PPH14; ISOTYPUS IMI.

Colonies on MEA growing slowly, at 25 C in 14 days, 1.5–2.3 cm diam, zonate, velutinous to farinose, greyish green to dull green (26D-E4-5, 27D-E4-6), margin floccose, white. Reverse center yellowish to yellowish brown (5C-F4-6), subcentral to margin dull green (26-27D-E3-4) to colorless. Odor and diffuse pigment lacking. Exudate clear colorless. Synnemata absent. Mycelium branched, septate, smooth-walled, hyaline, 2.5–3.8 μm wide. Conidiophores ascending most commonly from aerial, occasionally from surface or submerged hyphae, septate, straight or sinuous, smooth-walled, hyaline, up to 210 μm in length, 3.1–5 μm in length, bearing divergent, terminal, verticillate metulae or phialides, or with

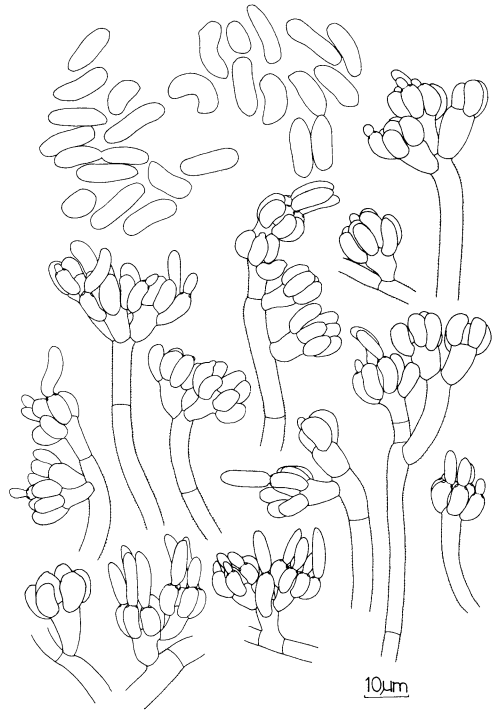
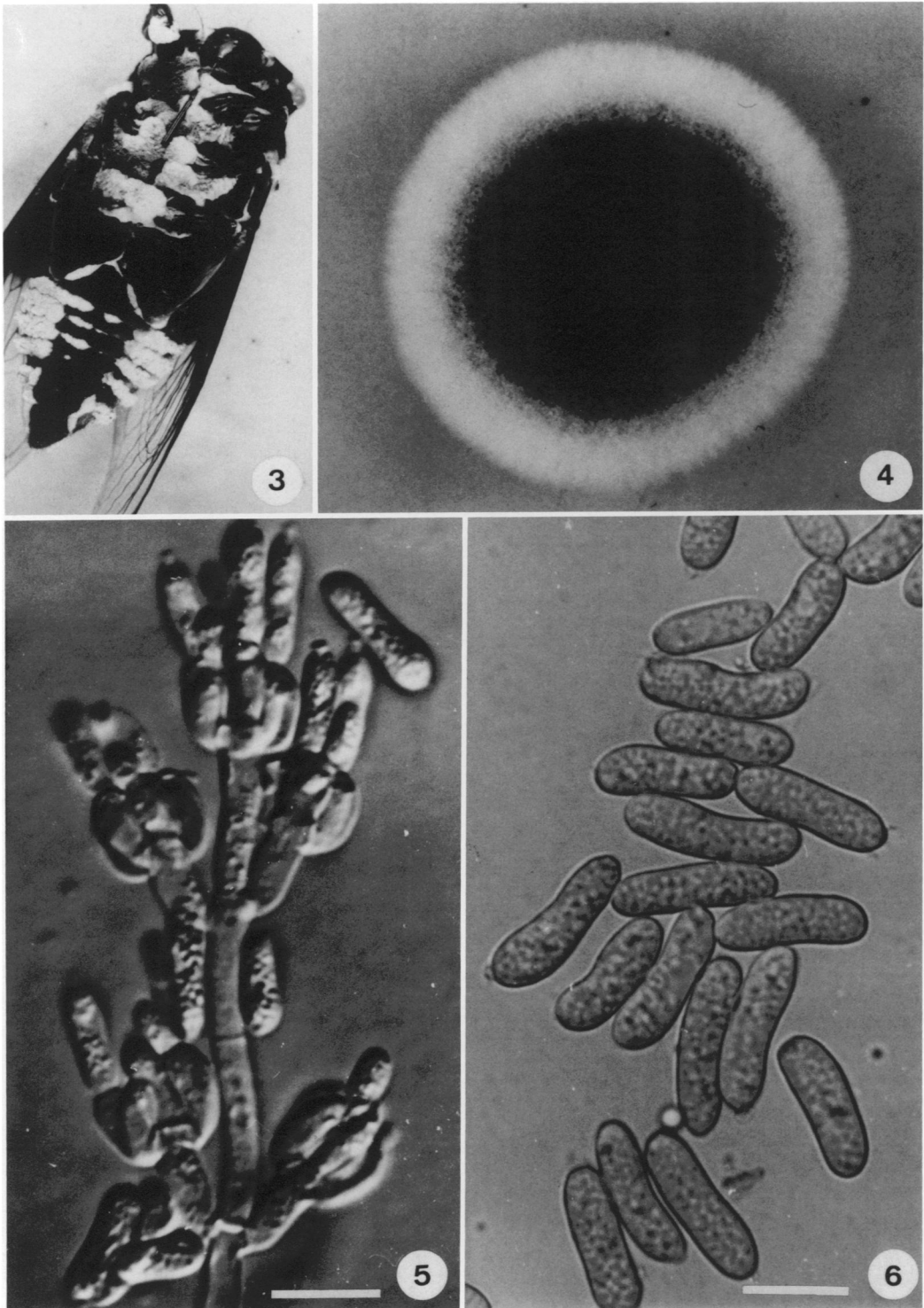
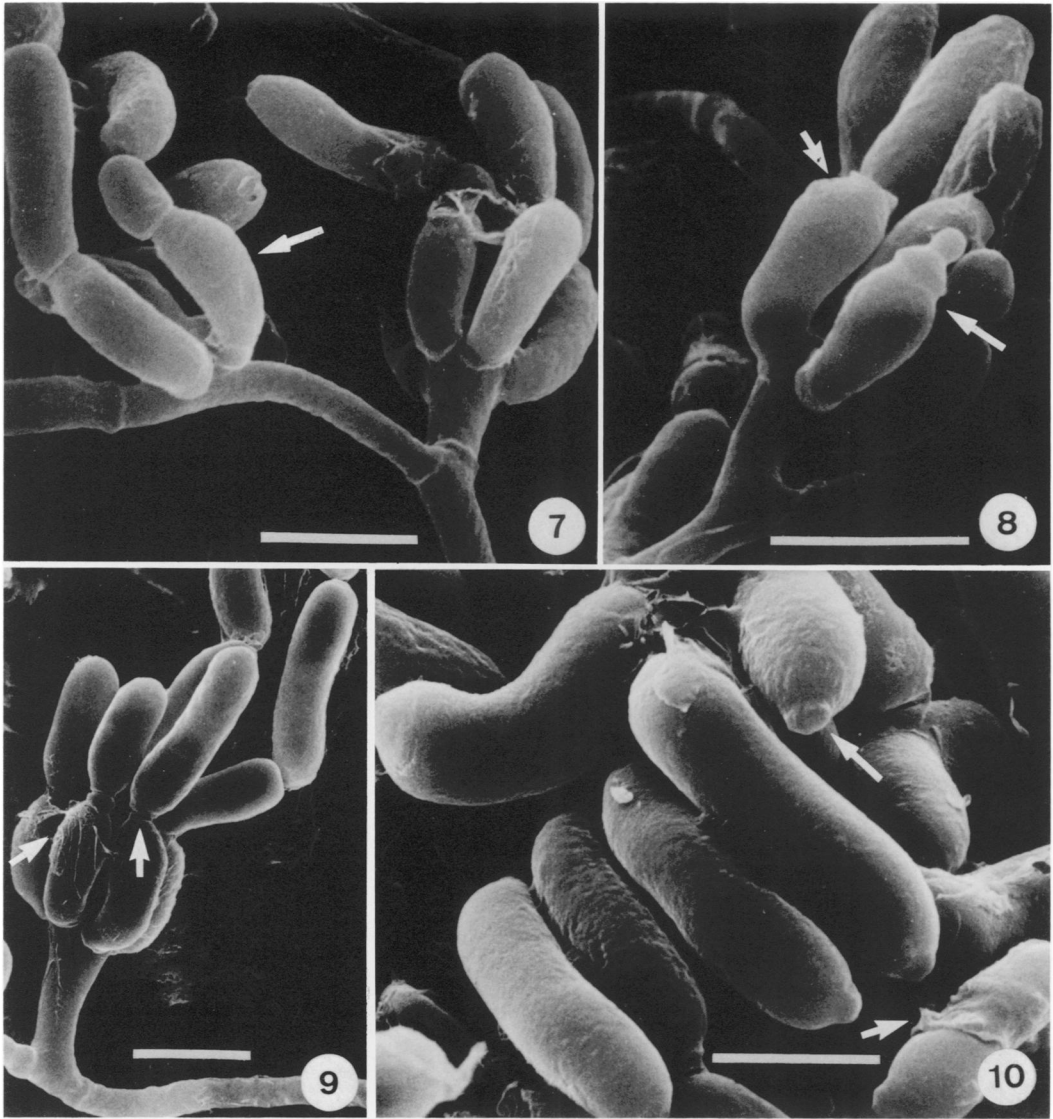


FIG. 2. *Nomuraea viridulus*. Characteristics of conidiophores, metulae, phialides, and conidia on cicada, *Cryptotympana facialis* Walker. Note that the conidiogenous structures and conidia are usually shorter and broader than those on culture media.

irregularly subterminal lateral or lower levels metulae, one to two or in whorl arising near septa; metulae broadly clavate, or cylindrical, 11.7–31.6 \times 2.6–3.8 μm ; phialides appressed three to eight per metulae, or occasionally sessile and solitary borne directly on the conidiophores, oval-cylindrical, ellipsoidal, neck absent or barely perceptible, 5.3–9.2 \times 3.3–4.6 μm . Conidia catenulate, nonseptate, smooth-walled, cylindrical, ellipsoidal, usually slightly curved or allantoid, sometimes apiculate, greyish yellow, in mass dull greyish green to dark green, 14.4–19.4 \times 3.8–4.4 μm . Chlamydospores absent. Under scanning electron microscopy, phialides and conidia usually covered with a layer of mucilage. On insect hosts, mycelium initially white, floccose, covering the sutures, turning vivid blue (22A8) to light green (27D-E4-5), velutinous, in age greyish green to dull green, with heavy sporulation, metulae, phialides and conidia usually broader and shorter than those in culture media; 4–12 phialides per metulae, growing along the



FIGS. 3-6. The characteristics of habitat, colony, conidiophores, conidiogenous cells and conidia of *Nomuraea viridulus*. 3. Naturally infested cicada, *Cryptotympana facialis*, body juncture overlaid with velutinous mycelium and dull-green conidia. 4. Colony on MEA at 25 C in 14 days without illumination. 5. Conidiophore bearing terminal verticillate phialides and subterminal or lateral metulae and verticillate phialides, metulae sometimes arising near septa. 6. Conidia cylindrical, ellipsoidal, some slightly curved or allantoid. Bars = 10 μ m.



FIGS. 7–10. Scanning electron micrographs of the sporulating structures and conidia of *Nomuraea viridulus*. 7. Phialides verticillate or solitary (arrow) borne on metulae or ascending directly from the fertile hyphae. 8. Phialides neck absent or barely perceptible, apex truncate (arrow), or somewhat thickened in conidiogenesis (arrow). 9. Conidia catenulate, schizolytic (right arrow), phialides oval-cylindrical, covered with distinct mucilage (left arrow). 10. Conidia cylindrical, some curved, apiculate (upper arrow), some with mucilage (lower arrow). Bars = 5 μ m.

whole length of the conidiophores at random not restricted at septa, occasionally metulae proliferating; conidiophores finely roughened to roughened. Synnemata and chlamydospores absent.

SPECIMEN EXAMINED. On *Cryptotympana facialis*, Ho 44, Hsintien, Taipei, Taiwan, R.O.C. 22 June, 1989, HOLOTYPE PPH 14 (dried culture) and exotype PPH14E (living culture), deposited in the Department

of Plant Pathology and Entomology, National Taiwan University, Taipei, Taiwan. PPH14E was also deposited in the Culture Collection and Research Center (CCRC 32589), Hsinchu, Taiwan. ISOTYPES NY, IMI. Other specimen examined, exclusively on cicada, Ho 1, Furong, Taipei, 13 July 1988; Ho 5, Chingmeei, Taipei, 17 Aug. 1988; Ho 7, Sirting Park, Pingtung, 10 Sept. 1988; Homoptera, Ho 49, Full-Moon Mt., Taoyuen, 16 July 1989; Ho 57, Fushan, Taipei, 9 Aug. 1989; Ho 51, Botanical Garden, Taipei, 29 July 1989;

Ho 68, Paoming Temple, Keelung, 25 Aug. 1989; Ho 72, Nanjen Mt., Pingtung, 24 Sept. 1989.

Nomuraea viridulus has been encountered on cicada cadavers on many occasions during a 2 year survey from Taiwan. This fact indicates its common occurrence and suggests that it has been overlooked in the past. The distinguishing characteristics of *N. viridulus* consist of erect or flexuous conidiophores bearing irregular verticils of phialides alone, or metulae and phialides, along their whole length randomly and not restricted at septa, appressed whorls of phialides, phialides without collula and huge cylindrical, ellipsoidal, usually slightly curved or allantoid conidia. The characteristics of metulae and phialides of *N. viridulus* show close resemblance to *N. rileyi* and *N. atypicola*; however, the former's phialides and metulae are apparently longer and broader than the latter's. Also there is a striking difference in size and shape of conidia between them (Samson, 1974) making the distinction easy. Though the irregular verticils of phialides alone, or metulae and phialides, along the whole length of conidiophores of *N. viridulus* are similar to *N. anemonoides*, between them a conspicuous distinction exists not only in size of metulae and phialides but also in size and shape of conidia, and they can be differentiated from each other (Hocking, 1977). For comparison all the currently known *Nomuraea* species with their key microscopic traits were tabulated (TABLE I). In other aspects, *N. viridulus* also shows some similarity to several species of *Paecilomyces* and *Metarhizium* (Samson, 1974; Tulloch, 1976). However, in most species of *Paecilomyces* colonies are never true green, have thinner hyphae and smaller conidiogenous structures, with flask-shaped phialides in divergent clusters and tapering abruptly into a long thin neck (Samson, 1974; Brown and Smith, 1957). While in *Metarhizium*, conidiophores are variable in length, usually much shorter than in *Nomuraea*, with apical loosely penicillate or verticillate branches, each branch bearing two to five metulae and phialides, metulae clavate, phialides elongate cylindrical, with central constriction, and in most species conidiophores usually are aggregated into sporodochia, with stromatic base, and conidia laterally adhering together to form a prismatic column (Samson, 1974). These traits aid in separation of *Paecilomyces* and *Metarhizium* from *N. viridulus* despite similarity in microscopic features in one way or another.

TABLE I
COMPARISON OF THE MORPHOLOGICAL CHARACTERISTICS OF FOUR CURRENTLY KNOWN *NOMURAEA* SPECIES

Species	Colonies	Metulae (μm)	Phialides (μm)	Conidia (μm)	Reference
<i>N. anemonoides</i>	greyish green dull green	4.5-6.5 \times 3-4.5	4-6.5 \times 3-4	globose, ellipsoid, ellipso-pedunculate, 5.5 \times 3-4	Hocking, 1977
<i>N. atypicola</i>	purple dark purple	6-7 \times 3-5	3.5-5 \times 2-2.5	cylindrical, 4-6 \times 1.2-1.5	Samson, 1974
<i>N. rileyi</i>	olive green Malachite green	4-6.5 \times 2.2-3.5	4.7-6.5 \times 2.3-3	broadly ellipsoidal, cylindrical, 3.5-4.5 \times 2-3.1	Samson, 1974
<i>N. viridulus</i>	greenish green dull green	11.7-31.6 \times 2.6-3.8	5.3-9.2 \times 3.3-4.6	cylindrical, ellipsoidal, allantoid, 14.4-19.4 \times 3.8-4.4	Tzean et al., 1992

Inasmuch as *N. viridulus* is widespread, adapts and survives well in nature as is evident from a 2 year survey, it might be capable of initiation of an epizootic in certain insect pests. Such potential for biocontrol merit further exploration, particularly in pathogenicity and host specificity in light of the past records (Samson et al., 1988; Ignoffo et al., 1976).

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Key Words: entomogenous fungi, Hyphomycetes, *Nomuraea*, taxonomy

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STEPHANOCYSTS AS NEMATODE-TRAPPING AND INFECTING PROPAGULES

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The term stephanocysts was coined by Boidin (1950) to describe a unique structure present in the context of the hymenium of a specific group of resupinate Basidiomycetes, especially in the genus *Hyphoderma*. Stephanocysts are bicelled, consisting of a cup-like basal cell and a terminal globose cell. At the juncture of the two cells, a row of spines surrounds the circumference. Boidin (1950) claimed that these spines arise from the upper cell, but this opinion has not been

confirmed by later workers (Burdshall, 1969; Hal-lenberg, 1990). The circumscription for the stephanocysts has been further broadened to include a reduced, one-celled type discovered in *H. puberum* (Fr.: Fr.) Wallr. (Boidin, 1958). The reduced stephanocyst consists of a swollen cell arising directly from the hyphal elements, which is surrounded by spines at the circumference and resembles the upper globose cell of the other form (Boidin, 1958; Burdshall, 1969). Additional atypical stephanocysts were found in *H. comptum* (Jack.) Jülich (Jülich, 1976), *H. echinocystis* Er-

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