



***Verrucospora vulgaris* (Agaricaceae, Agaricales), a Rare Tropical Species and a New Record for Thailand**

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ABSTRACT

During our study of basidiomycetes in northern Thailand, we collected *Verrucospora vulgaris*, a green agaric species with very irregular, angular and verrucose basidiospores. Since this is a rarely collected species, we provide a description and illustrations here. The species was originally described from tropical Africa and this is a first report for Thailand.

1. INTRODUCTION

The genus *Verrucospora* E. Horak belongs to family Agaricaceae, and two species have been presented [21]. *Verrucospora* was originally described in 1968 for *Lepiota verrocospora* Beeli from Congo [1]. Oberwinkler concluded that *Inocybe flavofusca* Henn. was identical to *L. verrucospora* [2], although Hennings described the spores as 'fusco-olivaceis' and the lamellae as 'flavidis, dein olivaceo-ferrugineis', which clearly are the colours of *Inocybe* spores [3]. There are two nomenclatural problems with the name *Verrucospora* [4]. Firstly, Horak placed *L. verrucospora* in *Verrucospora*, introducing the invalid combination *V. verrucospora* (Beeli) E. Horak [1]; Pegler introduced the new name *V. vulgaris* Pegler for this species [5]. Secondly, Shaw and Alcorn [6]

published the genus *Verrucispora* Shaw & Alcorn for a genus of anamorph ascomycetes. However, Kuyper [4] showed that the genus *Verrucospora* E. Horak was published in August 1967, and *Verrucispora* D.E. Shaw & Alcorn was published in December 1967, making *Verrucospora* E. Horak the legitimate name for the agaric genus [4]. Subsequently, Shaw and Alcorn proposed the name *Verrucisporota* D.E. Shaw & Alcorn for the genus [7].

Oberwinkler hypothesized that there might be a relationship between Agaricales and Thelephorales based on the spore shape and ornamentation which are similarly shaped, but dark-walled spores [2]. Pegler placed the genus in tribe Cystodermateae of the Agaricaceae [5], whereas Julich

erected a separate family, Verrucosporaceae for the genus [8]. A Chinese collection named *Verrucospora flavofusca* was also included in the AFTOL project [9]; based on the LSU, ITS, and SSU sequences this collection clearly belongs to the Agaricaceae s. str. and is not closely related to *Cystoderma* [9].

Thailand is located between the tropic of Cancer and the Equator, from 5° 37' to 20° 30' latitude north and from 97° 20' to 105° 39' longitude east. Northern Thailand is best known for hill tribe cultures and highest regional forest cover (43.5 %) [22]. We have been collecting *Agaricaceae* in northern Thailand since 2003, and have discovered numerous new and poorly known species [10-17, 20]. In this paper, we report on our collections of *V. vulgaris* which is a new record for Thailand. The taxon is fully described and illustrated.

2. MATERIALS AND METHODS

Fresh samples of mushrooms were collected in Huai Kok Ma (at Chiang Mai Province) and a forest Doi Tung (at Chiang Rai Province) in 2010. Macromorphological characters were noted as described by Vellinga *et al.* [14] and colour annotations followed Kornerup and Wanscher [18]. Microscopic characters were studied and illustrated from dry specimens, using a drawing tube attached to an Olympus CX-41 research compound microscope. Colour characters were observed in water, 2.5-10% of KOH, Melzer's reagent, cotton blue, Cresyl Blue, and Congo Red. Twenty-five spores per collection were observed and measured in side view. The notation (100,5,5) indicated that measurements were made on 100 spores in five samples in five collections. The following abbreviations were used: avl for average length, avw for average width, Q for quotient of length and width and avQ for average quotient. Specimens were deposited in the herbarium of Mae Fah Lung University (MFU).

3. RESULTS AND DISCUSSION

3.1 Taxonomy

Verrucospora vulgaris Pegler, *Kew Bull.*, Addit. Ser. 6: 384 (1977) (Figure 1 and 2)
 ≡ *Lepiota verrucispora* Beeli, *Bull. Soc. R. Bot. Belg.* 64: 218 (1932).

≡ *Verrucospora verrucispora* (Beeli) E. Horak, *Ber. schweiz. bot. Ges.* 77: 363 (1967), nom. inadmiss.

Misapplied names -Verrucospora flavofusca (Henn.) Jülich, *Bibliotheca mycol.* 85: 401. 1982; *Horakia flavofusca* (Henn.) Oberw., *Sydowia* 28: 359 (1975)

Pileus 15-30 mm, first parabolic, expanding to subumbonate, often convex to plano-convex, with straight margin, with completely yellowish brown (5F6-7) surface, often with darker colour, greenish-brown (5D3), then surface breaking, yellowish brown (5F6-7) at centre or umbo, with fine squamules around center, with fine squamules more distant from each other around umbo toward margin, when dry squamules turning dark brown to black on greyish yellow (2B4) background, at margin with velar squamules or fine squamules; margin fringed when mature and exceeding lamellae. Lamellae not really free, but sinuate, slightly crowded, with 3 series of lamellulae, ventricose to broadly ventricose 4-6 mm wide, pastel yellow to light yellow (2A4, 2A5), with eroded and concolorous edge, sometimes turning slightly bluish white (20A2) at edge. Stipe 40-66 × 2.8-5 mm, cylindrical, some time wider at base or with bulb-like, 5-6 mm wide base, yellowish with (2A4) background at apex to base zone, slightly fibrillose and concolorous with background at apex, sometimes grayish-yellow (4B3, 4B4) from middle zone to base, glabrous or squamulose at annular zone downward base, concolorous with background, turned light brown (6D5-6) to dark brown (7F4, 7F5) and black when dried, with white to yellowish-white (4A2) rhizomorphic mycelium at base and connected to substrate. Annulus with annular zone of yellowish-with (2A4) fibrillose, squamulose, grayish-yellow to

light brown (4B3, 6D5-6). Context light yellow to grayish-yellow (2A5, 2B4) and 4-4.5 mm wide in pileus, sometimes turned bluish-white (20A2) but not much; in stipe, concolorous with surface, hollow, with white fibrils in hollow. Odour similar to odour of *Leucoagaricus naucinus*. Taste unknown. Spore print white in all collections.

Basidiospores [100, 5, 5] $6.4-8.6 \times 3.6-5.8 \mu\text{m}$, $\text{avl} \times \text{avw} = 7.3 \times 5.0 \mu\text{m}$, $Q = 1.25-1.63$, $\text{av}Q = 1.48$, in side view slightly angular or verrucose in young stage, developing numerous papillae, becoming irregularly angular, pentagonal or star-like, with irregular angles in frontal view, dextrinoid, congophilous, cyanophilous, not observed in Cresyl Blue. Basidia $26-28 \times 7.8-8.4 \mu\text{m}$, clavate, slightly curved at base, thick-walled, hyaline, 4-spored, rarely 1- or 2-spored. Lamella edge sterile, with crowded cheilocystidia. Cheilocystidia $23-45 \times 4.9-6.5 \mu\text{m}$, cylindrical, or slightly irregularly cylindrical, sometime cylindrical and swollen at apex, rarely short narrowly clavate to slightly utriform, slightly thick-walled, colourless. Pleurocystidia absent. Pileus covering a trichoderm made up of erect, irregular, narrowly clavate, narrowly cylindrical elements, $60-88 \times 13.6-15.2 \mu\text{m}$, brown-walled, with brown parietal and intracellular pigment, often with short clavate elements under long elements, $25-35 \times 9-12 \mu\text{m}$, with pale brown parietal and intracellular pigment. Squamules on stipe covering with same structure as those of pileus covering. Clamp connections present but not abundant.

Habitat and distributions: Growing solitary or in small groups, on humus soil with high nutrient source. Found in 2 kinds of deciduous forest; Huai Kok Ma (at Chiang Mai Province) is a forest with dominance with *Castanopsis* spp. and *Lithocarpus echinops* with abundant leaf compost in forest floor; and a forest Doi Tung (at Chiang Rai Province) is forest dominant with *Pinus kesiya* and coffee garden.

Material examined: THAILAND - Chiang Mai Prov., Muang Dist., Suthep Sub-Dist., Huai Kok Ma Village, $N18^{\circ}48.62' E098^{\circ}54.60'$, alt. 1145 m., 28 June 2010, J.M. Birkebak P81(MFLU 100599); ibidem, 29 June 2010, P. Sysouphanthong P83(MFLU10 0601); ibidem, 29 June 2010, P. Sysouphanthong P86 (MFLU10 0604); ibidem, 10 July 2010, J.M. Birkebak P109 (MFLU10 0627); Chiang Rai Prov., Mae Fah Luang Dist., Doi Tung Sub-Dist., $N20^{\circ} 17.37'$, $E99^{\circ} 49.08'$, Alt. 860 m., 17 July 2010, P. Sysouphanthong P122 (MFLU10 0640).



Figure 1. Fresh basidiomata of *Verrucospora vulgaris*. a-c (MFLU10 0640), d-e (MFLU10 0627), f (MFLU100601), g (MFLU100599), h (MFLU100604).

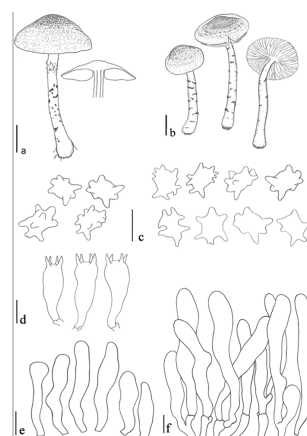


Figure 2. *Verrucospora vulgaris* (a, c-f = MFLU10 0640) (b = MFLU100599). A, b. basidiomata and a section, c. basidiospores, d. basidia, e. cheilocystidia, f. pileus covering. Scale bar (a-b= 10 mm, a-e = 10 μm).

4. DISCUSSION

Verrucospora vulgaris is recognized by greyish yellow to greenish yellow basidiomata, yellowish brown fine squamules on pileus, sinuate and pastel yellow to light yellow lamellae, cylindrical stipe covered with greenish yellow fibrils and yellowish brown squamules, irregularly angular, papillate to verrucose white spores, clavate to cylindrical cheilocystidia, trichodermal pileus- and stipe covering, and clamp connections.

Verrucospora vulgaris varies in colours from collection to collections; two collections from forest of Huai Kok Ma, MFLU10 0627 and MFLU100601 show yellowish brown colours. While two other collections, viz. MFLU100599 and MFLU 100604 from this area are same with a collection, MFLU100640 from Forest of Doi Tung show greenish-brown. However, all collections share the microcharacters (see Figure 1 & 2).

This species is specially found in tropical region, and this is second report of distribution of the species outside tropical Africa [1, 5]. The Thai collections are very similar to the material of *V. vulgaris* described from tropical Africa (Congo and Tanzania) [1,2,5,19]. Pegler noted that the pileal squamules were made up of loosely arranged, branching hyphae with strongly developed clavate-cylindrical apices, 20-90 × 4-11 μm, with a slightly thickened brown wall," and that the pileal surface was an "irregular epithelium of clavate elements, 33-55 × 7-20 μm, sometimes containing a brown vacuolar sap" [5]. The Thai collections have a trichodermal pileus covering, with cylindrical-clavate elements, and shorter clavate cells. We assume that these structures are similar to those as previously described [5].

Lepiota citrophylla, a species with free lamellae, spurred basidiospores from Thailand macroscopically resembles *V. vulgaris* because of the greenish yellow to yellowish brown basidiomata with brown

to dark brown squamules on pileus and stipe. Both two species share similar soil type, forest types of *Castanopsis* spp., *Lithocarpus polystachyus* in forest of Huai Kok Ma and *Pinus kesiya* in forest of Doi Tung [15]. Microscopically, they differ, especially in spore shape.

However, *Verrucospora flavofusca* (Henn.) Jülich was certified to belong to the *Agaricaceae* by phylogenetic tree based on the LSU, ITS, and SSU sequences, and it related to *Lepiota cristata* and *Coprinus comatus* [9]. The 2 species present in this genus [21]. *Verrucospora flavofusca* [8] and *V. vulgaris* Pegler [5] are very similar in their morphology. Unfortunately, we do not have molecular data to confirm that they are different. So we propose to name *Verrucospora vulgaris* in materials of this study.

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REFERENCES

- [1] Horak E., Remarques critiques sur quelques champignons du Congo (Afrique), *Ber. Schw. Bot. Ges.*, 1968 ["1967"]; 77: 362-375.
- [2] Oberwinkler F., Eine agaricoide Gattung der Thelephorales, *Sydowia*, 1976; 78: 359-361.
- [3] Hennings P., Fungi camerunenses novi III. *Engl. Bot. Jahrb.*, 1902; 30: 52.
- [4] Kuyper T.W., *Verrucospora* Horak or *Horakia* Oberwinkler (Fungi)?, *Taxon*, 1990; 39: 498-499.
- [5] Pegler D.N., A preliminary Agaric flora of East Africa, *Kew Bull. Add. Ser.*, 1977; 6: 1-615.

- [6] Shaw D.E. and Alcorn J.L., The genus *Verrucispora* gen. nov. (Fungi imperfecti) on Proteaceae in New Guinea and Queensland, *Proc. Linnean Soc. New South Wales*, 1967; **92**: 171-173.
- [7] Shaw D.E. and Alcorn J.L., New names for *Verrucispora* and its species. *Aust. Syst. Bot.*, 1993; **6**: 273-276.
- [8] Julich W., Higher taxa of Basidiomycetes, *Bibliotheca Mycol.*, 1982; **85**: 1-485.
- [9] Matheny P.B., Curtis J.M., Hofstetter V., Aime M.C., Moncalvo J.-M., Ge Z.-W., Yang Z.-L., Slot J.C., Ammirati J.F., Baroni T.J., Bougher N.L., Hughes K.W., Lodge D.J., Kerrigan R.W., Seidl M.T., Aanen D.K., DeNitis M., Daniele G.M., Desjardin D.E., Kropp B.R., Norvell L.L., Parker A., Vellinga E.C., Vilgalys R. and Hibbett D.S., Major clades of *Agaricales*: a multi-locus phylogenetic overview, *Mycologia*, 2007; **98**: 984-997.
- [10] Le T.H., Nuytinck J., Verbeken A., Lumyong S. and Desjardin D.E., *Lactarius* in Northern Thailand: 1. *Lactarius* subgenus Piperites, *Fungal Divers.*, 2007a; **24**: 173-224.
- [11] Le H.T., Stubbe D., Verbeken A., Nuytinck J., Lumyong, S. and Desjardin D.E., *Lactarius* in Northern Thailand: 2. *Lactarius* subgenus Plinthogali, *Fungal Divers.*, 2007; **27**: 61-94.
- [12] Zhao R.-L., Desjardin D.E., Soyong K., and Hyde K.D., A new species of bird's nest fungi: characterisation of *Cyathus subglobisporus* sp. nov. based on morphological and molecular data, *Persoonia*, 2008; **21**: 71-76.
- [13] Zhao R.-L., Desjardin D.E., Soyong K., Perry P.A. and Hyde K.D., A monograph of *Micropsalliota* in northern Thailand based on morphological and molecular data, *Fungal Divers.*, 2010; **45**: 33-79.
- [14] Vellinga E.C., Sysouphanthong P. and Kevin D.H., The family *Agaricaceae*: phylogenies and two new white-spored genera, *Mycologia*, 2011; **103**: 494-509.
- [15] Sysouphanthong P., Hyde K.D., Chukeatirote E., Bahkali A.H. and Vellinga E.C., *Lepiota* (Agaricales) in northern Thailand - 1. *L.* section *Stenosporae*, *Mycotaxon* 2011a; **117**: 53-85.
- [16] Sysouphanthong P., Hyde K.D., Chukeatirote E., Bahkali A.H. and Vellinga E.C., *Lepiota* (Agaricales) in northern Thailand - 2 *Lepiota* section *Lepiota*, *Crypt. Mycol.*, 2012; **33**(1): 25-42.
- [17] Zhao R.L., Karunarathna S.C., Raspé O., Parra L.A., Guinberteau J., Moinard M., De Kesel A., Barroso G., Desjardin D., Courtecuisse R., Hyde K.D., Guelly A.K. and Callac P., Major clades in tropical *Agaricus*, *Fungal Divers.*, 2011; **51**(1): 279-296.
- [18] Kornerup A. and Wanscher J.H., *Methuen Handbook of Colour*, London, Eyre Methuen, UK, 1978.
- [19] Beeli M., Fungi goossensiani IX. Genre *Lepiota*, *Bull. Soc. Royal Bot. Belgique*, 1932; **64**: 206-219.
- [20] Wisitrassameewong K., Karunarathna C.K., Thongklang N., Zhao R.L., Callac P., Chukeatirote E., Bahkali A.H. and Hyde K.D. *Agaricus subrufescens*: new to Thailand, *Chiang Mai J. Sci.*, 2012; **39**(2): 281-291.
- [21] Kirk P.M., Cannon P.F., Minter D.W. and Stalpers J.A., *Ainsworth & Bisby's Dictionary of the Fungi* (10th ed), Wallingford, UK, CAB International, 2008.
- [22] Gardner S., Sidisunthorn P. and Anusarnsunthorn V., *A Field Guide to Forest Trees of Northern Thailand*. Bangkok: Kobfai publishing project, 2000.