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Fungal Flora in Chiba Pref., Central Japan (V). Gasteromycetes 2. Additions to the Family Lycoperdaceae

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Abstract Eight species of Lycoperdaceae (Gasteromycetes) collected in Chiba Prefecture, a central area of Japan are reported: *Calvatia boninensis, C. craniiformis, C. rugosa, C. utriformis, Disciseda candida, Lycoperdon nigrescens, L. shimousanum* and *L. umbrinum*. Among them, *L. shimousanum* is a new species and *D. candida* is a new record for Japan. *C. boninensis, C. craniiformis, C. rugosa, C. utriformis* and *L. umbrinum* are newly recorded for Chiba Prefecture. Autofluorescense of basidiospores and capillitia of 4 *Calvatia, Disciseda* and *Lycoperdon* species are described by the fluorescence microscopic observation. Observations of their autofluorescense suggest their significance for morphological classification of Lycoperdaceae.

Key words: autofluorescence, Chiba Prefecture, Lycoperdaceae, new records, new species, taxonomy.

This paper is the second in a series of taxonomic studies on the family Lycoperdaceae in the herbarium of Natural History Museum and Institute, Chiba (CBM), Chiba Prefecture, Japan. The first paper (Kasuya, 2004a) discussed the 12 species of this family. In this paper, 8 species of the genera *Calvatia, Disciseda* and *Lycoperdon* were recognized. Among them, 1 species of *Lycoperdon* is a new species and 1 species of *Disciseda* are newly recorded from Japan. Furthermore, 5 species of *Calvatia* and *Lycoperdon* are newly recorded from Chiba Prefecture.

In lycoperdaceous fungi, autofluorescence specific to their basidiospores and capillitia is frequently observed (Kasuya, 2004b). Fluorescence can be observed easily by means of excitation with blue-violet radiation. In many cases, autofluorescence of organisms shows a characteristic pattern which is different from the image observed under visible light (Ando *et al.*, 1991). Therefore, the detailed structure of basidiospores and capillitia of lycoperdaceous fungi, which are difficult to recognize under visible light, may be described much more clearly using the fluorescence microscope. This paper describes autofluorescense of basidiospores and capillitia of 4 *Calvatia, Disciseda* and *Lycoperdon* species by the fluorescence microscopy.

Materials and Methods

All the materials examined were deposited in the herbarium of the Natural History Museum and Institute, Chiba (CBM). Macroscopic characters were described by the observations on dried materials. For visible light microscopic observations, free-hand sections of gleba and peridium were mounted in water, 5% KOH (w/v), and 1% cotton-blue lactophenol on glass slides. Forty or 50 randomly selected basidiospores were measured for each specimen under a light microscope. For scanning electron microscope (SEM) observations of the basidiospores of *L. shimousanum* Kasuya & Katum., a small portion of gleba was coated platinum-palladium in an ion sputter (Hitachi E-1030; Hitachi, Tokyo, Japan), and observed under a SEM (Hitachi S-800) operating at 15.0 kV. For the observations of the autofluorescence of basidiospores and capillitia, small portions of gleba of *Calvatia craniiformis* (Schwein.) Fr., *Disciseda candida* (Schwein.) Lloyd, *Lycoperdon nigrescens* Wahlenb. : Pers. and *L. shimousanum* were mounted in lactophenol, and were observed by a Zeiss Axiophot microscope equipped with an Epi-fluorescence system. Zeiss blue-violet filter (05 filter) was selected as an excitation filter; this filter has transmission at 395-440 nm.

Taxonomy

Calvatia boninensis S. Ito & Imai, Trans. Sapporo Nat. Hist. Soc. 16: 9, 1939.

Fig. 1.

Basidiomata large, subglobose to turbinate, 50-160 mm high, 30-170 mm broad, yellowish brown to dark brown, with thick white rhizomorphs up to 40 mm. Exoperidium made of very thin, papery, almost smooth scales, yellowish brown when young, later blackish brown, sloughing off at maturity. Endoperidium thin, shiny, yellowish brown to dark brown, later breaking away by degrees in fragments. Gleba pulverulent, white when young, later ochraceous. Subgleba well developed, spongy, ochraceous to dark brown.

Basidiospores ellipsoid to cylindric, rarely ovoid, minutely verrucose, (3.0-) 3.5-5.0 (-5.5) μm in diameter, with a hyaline pedicel up to 1.5 μm long. Basidia not observed. Capillitium *Lycoperdon*-type, 2.0-4.0 (-5.0) μm broad, rarely branched, brown to dark brown, septate, pores small, irregular, walls 0.5-1.0 μm thick, not pitted. Paracapillitium rarely present.

Habitat: On rich soil or humus in woodland.

Distribution: Japan (Imazeki *et al.*, 1988; Ito, 1959; Ito and Imai, 1939), Indonesia (Kasuya and Retnowati, 2005) and Thailand (Ruksawong and Flegel, 2001).

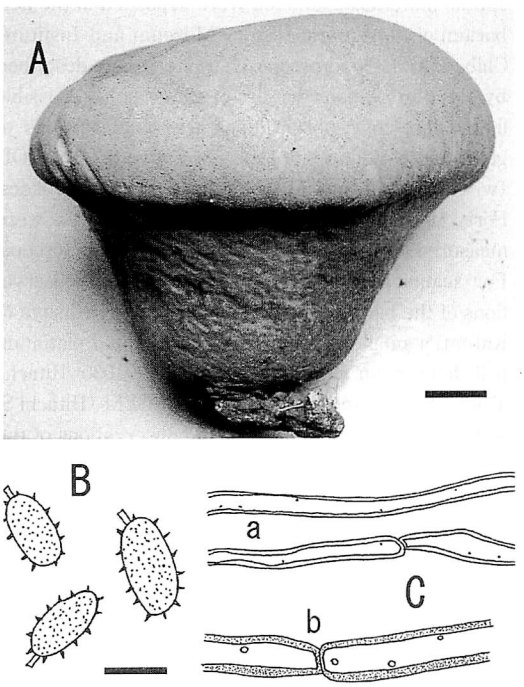

Fig. 1. *Calvatia boninensis* S. Ito & Imai (CBM-FB-15233). A, Basidioma; B, Basidiospores; C, Capillitia. a, capillitial thread; b, detail of capillitium. Scale bars A: 10 mm, B: 3.5 μm, Ca: 20 μm, Cb: 10 μm.

Specimens examined: Chiba Pref., Imba-gun, Sakae-machi, Ryukakuji, July 28, 1991, coll. Chiba Mycol. Club s.n., CBM-FB-3199; Chiba Pref., Chiba-shi, Chuo-ku, Aoba-cho, September 18, 1991, coll. T. Fukiharu s.n., CBM-FB-3308; same place, August 17, 1993, coll. T. Fukiharu s.n., CBM-FB-9021; same place, June 29, 1994, coll. R. Onuma s.n., CBM-FB-11194; same place, September 24, 1994, coll. T. Fukiharu s.n., CBM-FB-11553; same place, June 25, 1998, coll. Y. Abe s.n., CBM-FB-16529; same place, July 7, 1998, coll. T. Fukiharu s.n., CBM-FB-16660; same place, June 30, 2000, coll. Y. Koyama s.n., CBM-FB-24678; Chiba Pref., Chiba-shi, Inage-ku, Inage-higashi, July 7, 1993, coll. F. Koshino s.n., CBM-FB-5608; Chiba Pref., Chiba-shi, Midori-ku, Noro-cho, July 21, 1986, coll. Y. Horie s.n., CBM-FB-1902; same place, September 17, 1994, coll. Chiba Mycol. Club s.n., CBM-FB-11461; Chiba Pref., Chiba-shi, Mihama-ku, Makuhari-nishi, October, 2, 1996, coll. T. Wada s.n., CBM-FB-15233; Chiba Pref., Chosei-gun, Ichinomiya-machi, Torami, October 27, 1987, coll. F. Koshino s.n., CBM-FB-24605; Chiba Pref., Awa-gun, Tomiura-machi, Tarada, September 20, 2000, coll. T. Kawana s.n., CBM-FB-24838.

Japanese name: Ou-noutake.

Remarks: This species is newly reported here from Chiba Prefecture. This species is morphologically similar to *C. craniiformis* (Schwein.) Fr., but the basidiospores of these two species are quite different. *Calvatia boninensis* has ellipsoid to cylindric basidiodpores, but those of *C. craniiformis* are globose to subglobose. *Calvatia boninensis* was originally described from Bonin Islands, Southeastern Japan (Ito and Imai, 1939), later also collected from tropical, subtropical to warm-temperate areas of Eastern Asia (Imazeki *et al.*, 1988; Ito, 1959; Kasuya and Retnowati, 2005; Ruksawong and Flegel, 2001).

Calvatia craniiformis (Schwein.) Fr., Summa Veg. Scand.: 442, 1849.

Fig. 2.

Basidiomata large, subglobose to turbinate, 40-120 mm high, 40-200 mm broad, yellowish brown to reddish brown, with thick white rhizomorphs up to 30 mm. Exoperidium made of very thin, papery, smooth to minutely furfuraceous scales, pale ochraceous when young, later dark brown, sloughing off at maturity. Endoperidium thin, shiny, yellowish brown to reddish brown, later breaking away by degrees in fragments. Gleba pulverulent, white when young, later ochraceous to brown. Subgleba well developed, spongy, ochraceous to brown.

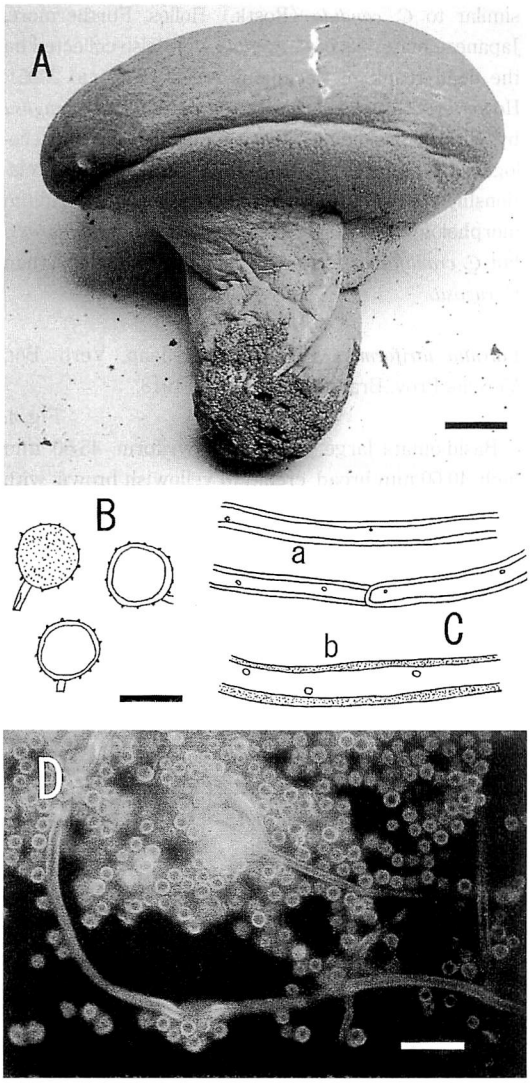

Fig. 2. *Calvatia craniiformis* (Schwein.) Fr (CBM-FB-706). A, Basidioma; B, Basidiospores; C, Capillitia. a, capillitial thread; b, detail of capillitium; D, Autofluorescence of basidiospores and capillitia. Scale bars A: 7 mm, B: 4.0 μm, Ca: 20 μm, Cb: 10 μm, D: 13 μm.

Basidiospores globose to subglobose, minutely verrucose, (3.0-) 3.5-4.5 (-5.0) μm in diameter excluding ornaments or (3.5-) 4.0-5.0 (-5.5) μm in diameter including ornaments, with a hyaline pedicel up to 3.0 μm long; walls, ornaments and pedicels of basidiospores fluoresce. Basidia not observed. Capillitium *Lycoperdon*-type, 2.0-6.0 (-7.0) μm broad, occasionally dichotomously branched, yellowish brown to dark brown, septate, pores small, irregular, abundant, walls 0.5-1.5 μm thick, not pitted, walls and septa fluoresce. Paracapillitium absent.

Habitat: On rich soil or humus in woodland.

Distribution: Common in temperate to warm-temperate areas in Japan. Also known from China (Liu, 1984; Fan *et al.*, 1994), Indonesia (Kasuya and Retnowati, 2005), Canada (Coker and Couch, 1928), United States (Coker and Couch, 1928; Zeller and Smith, 1964) and Mexico (Urista *et al.*, 1985).

Specimens examined: Chiba Pref., Abiko-shi, Nakabyo, November, 1, 1992, coll. Y. Kudo s.n., CBM-FB-4847; Chiba Pref., Imba-gun, Sakae-machi, Ryukakuji, October 6, 1991, coll. Y. Horie s.n., CBM-FB-6058; Chiba Pref., Sakura-shi, Sakura, October 4, 1997, coll. Chiba Mycol. Club s.n., CBM-FB-16073; Chiba Pref., Chiba-shi, Chuo-ku, Aoba-cho, June 11, 1998, coll. Y. Abe s.n., CBM-FB-16468; Chiba Pref., Chiba-shi, Chuo-ku, Inohana, July 26, 1986, coll. Y. Horie s.n., CBM-FB-1907; Chiba Pref., Chiba-shi, Midori-ku, Noro-cho, September 26, 1994, coll. R. Onuma s.n., CBM-FB-11630; Chiba Pref., Chiba-shi, Wakaba-ku, Tabeta-cho, June 16, 1998, coll. K. Otsuta s.n., CBM-FB-16513; Chiba Pref., Kimitsu-shi, Ozaki, October 2, 1992, coll. Chiba Mycol. Club s.n., CBM-FB-4509; Chiba Pref., Kamogawa-shi, Izumi, October 22, 1987, coll. F. Koshino s.n., CBM-FB-706.

Japanese name: Noutake.

Remarks: This species is newly reported here from Chiba Prefecture. *Calvatia craniiformis* is the most common Japanese species of *Calvatia*. Surface ornamentation and pedicels of basidiospores of *C. craniiformis* can be observed more clearly under fluorescense microscopy than visible light.

Calvatia rugosa (Berk. & M. A. Curtis) Reid, Kew Bull. 31: 671, 1976.

Fig. 3.

Basidiomata subglobose to depressed globose, 20-45 mm high, 30-100 mm broad, pale ochraceous to reddish brown, but turning yellow when bruised before maturity, with thick, white rhizomorphs up to 50 mm long. Exoperidium made of very thin, furfuraceous spines, pale yellowish brown to ochraceous when young, later dark brown, sloughing off at maturity. Endoperidium thin, shiny, yellowish brown to reddish brown at upper part, white to cream at base, later breaking away by degrees in fragments. Gleba pulverulent, white when young, later ochraceous to brown. Subgleba minutely developed or sometimes absent, compact, pale yellow to ochraceous.

Basidiospores globose, minutely verrucose, warts up to 0.5 μm high, (3.0-) 3.5-4.5 μm in diameter excluding ornaments or (3.5-) 4.0-5.0 μm in diameter including ornaments, pale yellowish brown to ochraceous, with a hyaline pedicel, 1.0-1.5 μm long. Basidia not observed.

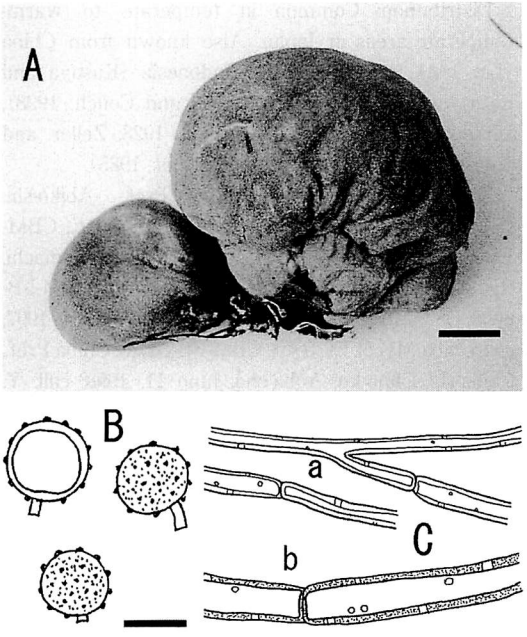

Fig. 3. *Calvatia rugosa* (Berk. & M. A. Curtis) Reid (CBM-FB-14411). A, Basidiomata; B, Basidiospores; C, Capillitia. a, capillitial thread; b, detail of capillitium. Scale bars A: 10 mm, B: 4.0 μm, Ca: 20 μm, Cb: 10 μm.

Capillitium *Lycoperdon*-type, 2.5-4.5 μm broad, occasionally branched, yellowish brown, septate, often breaking into short particles at septations, pores abundant, small, walls very thin, 0.5-1.0 μm thick, pitted. Paracapillitium absent.

Habitat: On the dead trunk of deciduous trees, especially Fagaceae.

Distribution: Japan, Europe (Sarasini, 2005), United States (Coker and Couch, 1928), Central America (Reid, 1976), South America (Rick, 1930) and Oceania (Cunningham, 1944).

Specimen examined: Chiba Pref., Ichihara-shi, Tsukizaki, October 21, 1995, coll. Chiba. Mycol. Club s.n., CBM-FB-14411.

Japanese name: Irogawari-hokoritake.

Remarks: This species is newly reported here from Chiba Prefecture. Previously, this species was reported as *C. rubroflava* (Cragin) Lloyd in Japan (Yoshimi and Hongo, 1989). However, *C. rubroflava* is considered a synonym of *C. rugosa* because both have yellowish to reddish endoperidium, small subgleba and globose basidiospores (Reid, 1976; Sarasini, 2005). In Chiba Prefecture, this species sometimes occurs on the dead trunk of deciduous trees, especially on Fagaceae, although there was no previous report of this substrate as a habitat of this species. Basidioma morphology and microscopic characterers of this species are similar to *C. candida* (Rostk.) Hollós. Furthermore, Japanese materials of *C. candida* were also collected on the dead trunk of deciduous trees (Kasuya, 2005). However, *C. candida* is easily separated from *C. rugosa* by whitish basidioma. These morphological and ecological characteres of *C. candida* suggest the close relationships between *C. rugosa* and *C. candida*. Peridium morphology of this species is similar to *C. craniiformis*, but *C. craniiformis* has more developed subgleba than *C. rugosa*.

Calvatia utriformis (Bull. : Pers.) Jaap, Verh. Bot. Vereins Prov. Brandenburg 59: 37, 1918.

Fig. 4.

Basidiomata large, globose to pyriform, 45-60 mm high, 40-60 mm broad, cream to yellowish brown, with white rhizomorphs up to 20 mm. Exoperidium made of thick polygonal plates, furfuraceous spines present among the plates, white when young, and then cream to yellowish brown, later sloughing off. Endoperidium thin, papery, shiny, white to greyish brown, fragile on the upper part when maturity. Gleba pulverulent, white when young, later yellowish brown. Subgleba well developed, spongy, white to ochaceous.

Basidiospores globose, almost smooth, 3.5-4.5 μm in diameter, with or without a short hyaline pedicel up to 0.5 μm long. Basidia not observed. Capillitium

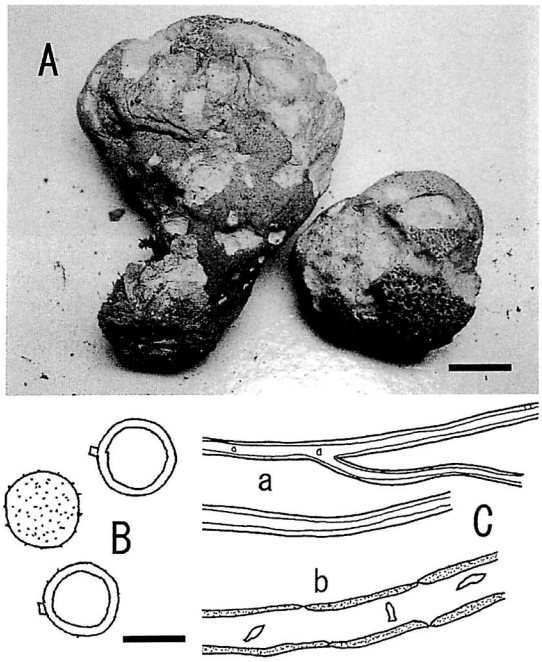

Fig. 4. *Calvatia utriformis* (Bull. : Pers.) Jaap (CBM-FB-5595). A, Basidiomata; B, Basidiospores; C, Capillitia. a, capillitial thread; b, detail of capillitium. Scale bars A: 15 mm, B: 4.0 μm, Ca: 20 μm, Cb: 10 μm.

Lycoperdon-type, 2.5-11.0 μm broad, dichotomously branched, yellowish brown, aseptate, pores slit-like or irregular, abundant, walls 0.5-2.0 μm thick, pitted. Paracapillitium absent. Exoperidial plates made of hyaline, subglobose sphaerocysts up to 30-50 μm in diameter.

Habitat: On sandy soil among grass.

Distribution: Japan, Europe (Kreisel, 1989; Lange, 1990), South Africa (Bottomley, 1948), Kazakhstan (Shvarcman and Filimonova, 1970), China (Liu, 1984), Himalaya (Ahmad, 1941; Kreisel, 1989), Mongolia (Dörfelt and Bumzaa, 1986), Canada (Coker and Couch, 1928), United States (Coker and Couch, 1928; Zeller and Smith, 1964) and Mexico (Urista *et al.*, 1985).

Specimen examined: Chiba Pref., Chiba-shi, Mihama-ku, Takahama, coll. F. Koshino s.n., CBM-FB-5595.

Japanese name: Kikumetake.

Remarks: This species is newly reported here from Chiba Prefecture. Previously, this species was reported as *C. caelata* (Bull.) Morgan in Japan (Yoshimi and Hongo, 1989). However, *C. caelata* is considered a synonym of *C. utriformis* because both have the same structures of peridium and similar morphology of basidiospores and capillitial pores (Kreisel, 1989). This species is distinguished from other Japanese species of *Calvatia* by the exoperidial polygonal plates, and the presence of slit-like pores in capillitium. *Calvatia excipuliformis* (Pers.: Pers.) Perdeck also has slit-like capillitial pores, but this species is clearly separated from *C. utriformis* by the high stipitum basidiomata, and prominently warted basidiospores. *Calvatia utriformis* has one variety, *i.e.*, *C. utriformis* var. *hungarica* (Hollós) F. Smarda. It is morphologically similar to the variety *utriformis* except its remarkably depressed basidiomata and not developed subgleba (Calonge, 1998; Martin, 1997).

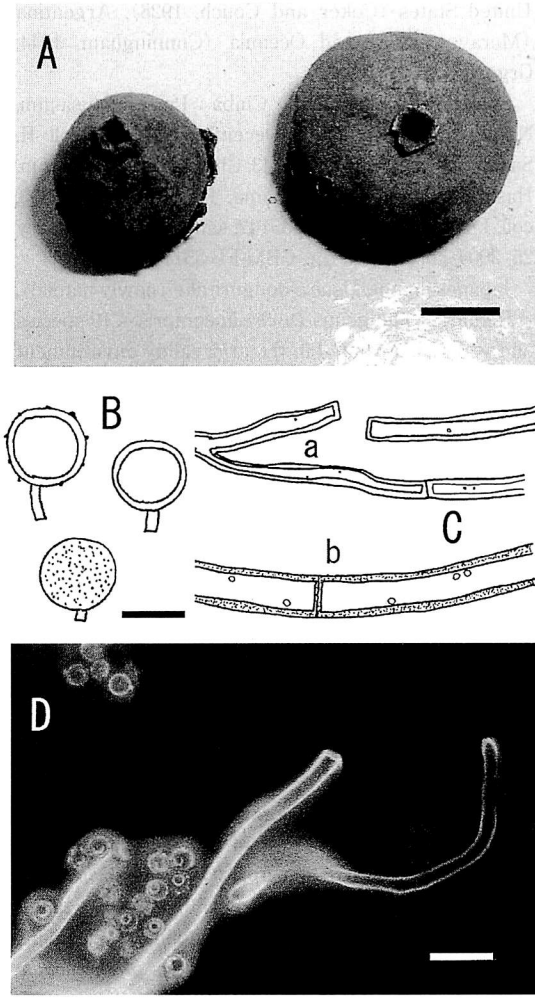

Fig. 5. *Disciseda candida* (Schwein.) Lloyd (CBM-FB-35143). A, Basidiomata; B, Basidiospores; C, Capillitia. a, capillitial thread; b, detail of capillitium; D, Autofluorescence of basidiospores and capillitia. Scale bars A: 7 mm, B: 5.0 μm, Ca: 20 μm, Cb: 10 μm, D: 15 μm.

Disciseda candida (Schwein.) Lloyd, Mycol. Notes 1(10): 100, 1902.

Fig. 5.

Basidiomata in small groups or solitary, hypogaeous when young, later subhypogaeous or epigaeous, globose to depressed globose, 10-20 mm broad, white to whitish grey. Exoperidium thin, brittle, brown, fallen away except for the basal area leaving a cup-shaped remaining at maturity, encrusted with sand. Endoperidium white to whitish grey, papery, pulverulent, tough, opening with a small apical pore up to 4 mm wide. Gleba white when young, later olivaceous brown, powdery. Subgleba absent.

Basidiospores globose, almost smooth or minutely verrucose, warts up to 0.5 μm high, 4.0-5.0 μm broad excluding ornaments or 5.0-6.0 μm broad including ornaments, with a short, hyaline pedicel up to 0.5 μm long, yellowish to olivaceous brown; walls, ornaments and pedicels of basidiospores fluoresce. Basidia not observed. Capillitium *Lycoperdon*-type, 3.0-6.0 μm broad, filamentous, rarely branched, septate, breaking into short particles at septations, pores abundant, yellowish brown, walls 1.0 μm thick, not pitted, strongly fluoresce. Paracapillitium absent.

Habitat: On dry, sandy soil among grass near the sea shore.

Distribution: Japan, Europe (Calonge, 1998; Kers, 1975; Moravec, 1954), South Africa (Bottomley, 1948),

United States (Coker and Couch, 1928), Argentina (Moravec, 1954) and Oceania (Cunningham, 1944; Grgurinovic, 1997).

Specimens examined: Chiba Pref., Sousa-gun, Nosaka-machi, Notehama, December 21, 2003, coll. H. Sakamoto s.n., CBM-FB-35143; Chiba Pref., Sambu-gun, Hasunuma-mura, Minamihama, December 21, 2003, coll. I. Asai s.n., CBM-FB-35142; same place, February 25, 2004, coll. I. Asai s.n., CBM-FB-35144.

Japanese name: Usuiro-donguritake (newly named).

Remarks: The genus *Disciseda* comprises 19 species and widely distributed in the arid, sandy environment of the world (Grgurinovic, 1997; Kreisel, 1976; Moravec, 1954). There are 2 species of *Disciseda* known to have almost smooth to minutely verrucose basidiospores: *D. australis* G. H. Cunn. and *D. cervina* (Berk.) Hollós (Cunningham, 1944; Grgurinovic, 1997). However, *D. candida* is clearly distinguished from both *D. australis* and *D. cervina* by its more whitish endoperidium and not conspicuous, small apical pore. Another 2 species of *Disciseda* are previously described to have whitish endoperidium and small apical pore like *D. candida*: *D. bovista* (Klotsch) Henn. and *D. subterranea* (Peck) Coker & Couch (Kers, 1975; Mitchel *et al.*, 1975). However, *D. bovista* is easily separated from *D. candida* by its large, verrucose basidiospores (6.0-9.0 μm broad, Kers, 1975). *Disciseda subterranea* is also distinguished from *D. candida* by its more smaller basidiomata up to 7-15 mm broad and more spinulose basidiospores (Mitchel *et al.*, 1975). Morphological characteres of the Japanese specimens are well identical with the descriptions of *D. candida* (Cunningham, 1944; Grgurinovic, 1997; Kers, 1975; Moravec, 1954), and those Japanese materials are identified as *D. candida*. This species is newly reported here from Japan.

Surface ornamentation and pedicels of basidiospores of *D. candida* can be observed more clearly under fluorescense microscopy than visible light. As a result, fluorescence microscopy is possibly useful to observations on basidiospores of *Disciseda*. Therefore, fluorescence microscopy should be applied to morphological classification of this genus. In Japan, only 1 species of this genus, *D. subterranea* was hitherto known (Kasuya, 2004a; Yoshimi and Hongo, 1989). Both *D. candida* and *D. subterranea* are frequently collected from coastal sand dunes in Japan.

Lycoperdon nigrescens Wahlenb. : Pers., Synops. Meth. Fung.: 146, 1801.

Fig. 6.

Specimens examined: Chiba Pref., Funabashi-shi, Oujinbo-cho, October 14, 2000, coll. Chiba Mycol. Club s.n., CBM-FB-30038; Chiba Pref., Chiba-shi, Chuo-ku, Aoba-cho, December 18, 1997, coll. Y. Abe s.n., CBM-FB-16308; Chiba Pref., Ichihara-shi, Shimono, October 27, 1993, coll. Chiba Mycol. Club s.n., CBM-FB-9779.

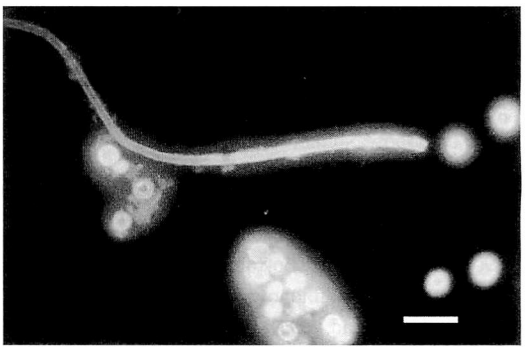

Fig. 6. Autofluorescence of basidiospores and capillitia of *Lycoperdon nigrescens* Wahlenb.: Pers (CBM-FB-16308). Scale bar: 15 μm.

Japanese name: Kuro-hokoritake.

Remarks: Surface ornamentation and short pedicels of basidiospores of this species cannot be recognized under fluorescence microscopy, but under visible light, these are clearly observed.

Lycoperdon shimousanum Kasuya & Katum., sp. nov.

Figs. 7-8.

Basidiomata in caterva parva formantia vel gregaria, 20-65 mm lata, 35-75 mm alta, turbinato-piriformia, cremea vel ochracea. Exoperidium primo albidum vel creameum, deinde pallide brunnescens vel fusco-brunnescens, spinis fragilibus conicis vel subconicis 1-2 mm longis denique secedentibus interdum granulis cremeis pulverulentibus commixtis obtectum, ex cellulis globosis vel subglobosis 5.0-7.0 μm diam hyalinis compositum. Endoperidium papyraceum, cremeum vel ochraceum, lucidum, ordinatione reticuli instructum. Gleba juvenilis alba, dein ochracea vel brunnea, pulverulenta, sine pseudocolumella. Subgleba bene crescens, insigniter magna, usque ad 60 μm crassa et 40 μm alta, superficie gyrosa vel spongiosa vel alveolata, cremea vel ochracea. Basidiosporae globosae vel subglobosae, episporio 4.0-5.0 μm diam (verruculo 0.5 μm alto incluso 4.5-5.5 μm diam), olivaceo-brunneae, sine pedicello; paries cum verruculis fluorescens. Basidia non observata. Capillitium flavo-brunneum vel olivaceo-brunneum, 3.0-8.5 μm crassum, elasticum, aseptatum, interdum dichotomice ramosum, poris non abundantibus praeditum; pariete 1.0-1.5 μm crasso non foveolato 3-stratoso. Paracapillitium absens.

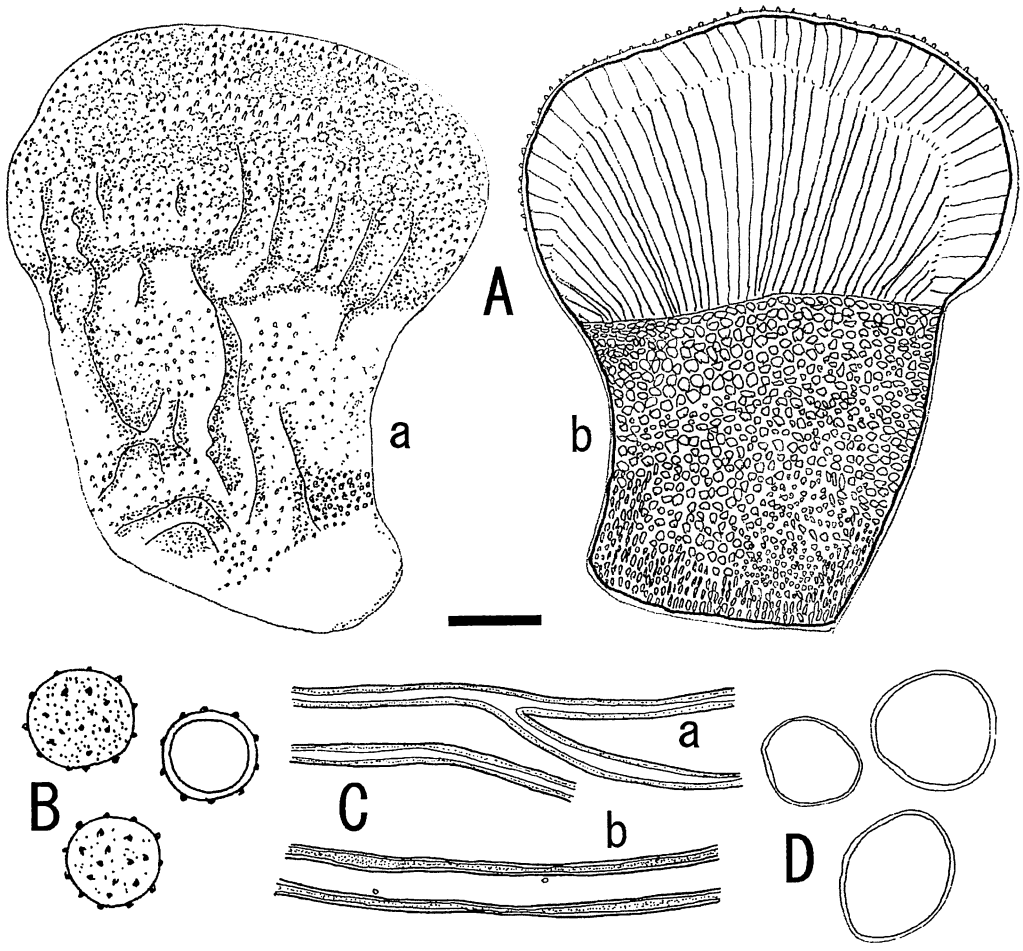

Fig. 7. *Lycoperdon shimousanum* Kasuya & Katum. (CBM-FB-3250, holotype). A, Basidiomata. a, surface of basidioma; b, section of basidioma; B, Basidiospores; C, Capillitia. a, capillitial thread; b, detail of capillitium; D, Sphaerocysts of exoperidium. Scale bars A: 13 mm, B: 4.5 μm, Ca: 20 μm, Cb: 10 μm, D: 5.0 μm.

Holotypus: CBM-FB-3250, ad terram in sylva, Ryukakuji, Sakae-machi, Imba-gun, Chiba Pref. in Japonia, 6 October 1991, a "Chiba Mycological Club" leg., in Herbario fungorum Musei et Instituti Historiae Naturalis Chibaensis (CBM) conservatus.

Etymology: "shimousa" means the old name of collected place of the type specimen.

Basidiomata in small groups or gregarious, 20-65 mm broad, 35-75 mm high, turbinate to pyriform, cream to ochraceus. Exoperidium white to cream when young, later becoming pale brown to blakish brown, made up of fragile, subconical to conical spines 1-2 mm long, later sloughing off, often mixed with cream pulverulent granules. Endoperidium papery, cream to ochraceous, shiny, showing reticulate pattern after exdoperidium sloughing off. Gleba white when young, later ochraceous to olivaceous brown, pulverulent, without pseudocolumella. Subgleba very well developed, remarkably large, up to 60 mm broad, 40 mm high, surface gyrose, spongy to alveolate, cream to ochraceous.

Basidiospores globose to subglobose, verrucose, warts up to 0.5 μm high, 4.0-5.0 μm broad excluding ornaments or 4.5-5.5 μm broad including ornaments, olivaceous brown, pedicel lacking; walls and ornaments of basidiospores fluoresce. Basidia not observed. Capillitium *Lycoperdon*-type, yellowish brown to olivaceous brown, 3.0-8.5 μm broad, elastic, pores not abundant, aseptate, sometimes dichotomously branched. Walls of capillitium 1.0-1.5 μm thick, not pitted, consist of 3-layer; outer layer thin, 0.1-0.3 μm thick, middle layer 0.5-1.0 μm thick, and inner layer 0.2-0.5 μm thick, only outer and inner layers fluoresce. Paracapillitium absent. Exoperidium composed of

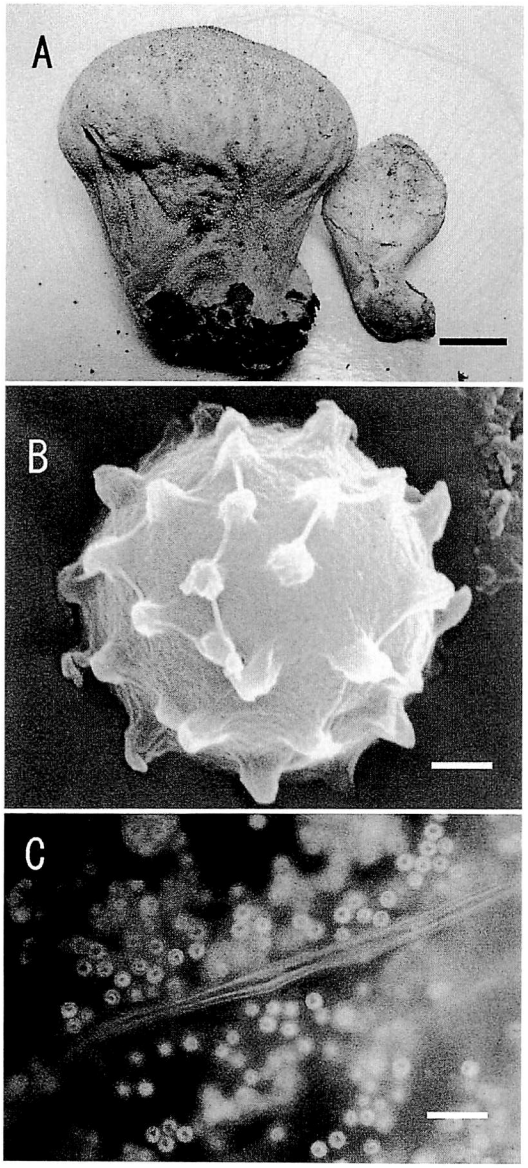

Fig. 8. *Lycoperdon shimousanum* (CBM-FB-3250, holotype). A, Basidiomata; B, SEM micrograph of basidiospore; C, Autofluorescence of basidiospores and capillitia. Scale bars A: 20 mm, B: 1.0 μm, C: 15 μm.

globose to subglobose sphaerocysts, 5.0-7.0 μm broad, hyaline.

Habitat: On rich soil or humus in woodland.

Distribution: Japan (Chiba). Known only from type locality.

Specimen examined: Chiba Pref., Imba-gun, Sakae-machi, Ryukakuji, October 6, 1991, coll. Chiba Mycol. Club s.n., CBM-FB-3250 (holotype).

Japanese name: Shimousa-ashibuto-hokoritake (newly named).

Remarks: This species is characterized by the presence of spongy to alveolate, well developed, large subgleba, non-pedicellate, verrucose basidiospores and thick (1.0-1.5 μm), 3-layered walls of capillitium. Exoperidium mixed with conical spines and pulverulent granules, and reticulate endoperidium of gleba place *L. shimousanum* at Stirps *'L. Perlatum'* in Section *Lycoperdon* (Demoulin, 1971). Hitherto, 3 species of Stirps *'L. Perlatum'* are known in the world (Demoulin, 1971). There are 2 species belonging to this stirps known to have spongy subgleba and verrucose badisiospores: *L. nigrescens* and *L. perlatum* Pers. (Demoulin, 1971). However, *L. shimousanum* is distinguished from both *L. nigrescens* and *L. perlatum* by the well developed, large subgleba, non-pedicellate basidiospores and thick capillitial walls, moreover from *L. nigrescens* by the pale brownish exoperidial spines. In the genus *Lycoperdon*, another 2 species are known to have spongy, well developed, large subgleba like *L. shimousanum*: *L. lividum* Pers. and *L. mammiforme* Pers. (Calonge, 1998; Kreisel, 1962). However, *L. shimousanum* is easily distinguished from those 2 species by its exoperidium forming subconical to conical spines or pulverulent granules, reticulate endoperidium of gleba and non-pedicellate basidiospores. Only 1 species, *L. umbrinoides* Dissing & M. Lange was previously described to have conspicuously thick-walled capillitium (1.0-2.5 μm broad, Dissing and Lange, 1962; Kasuya, 2004c; Sarasini, 2005) in this genus. However, this species is clearly separated from *L. shimousanum* by its blackish, smooth endoperidium and warty basidiospores with long pedicel. As above, no species of *Lycoperdon* is known to have spongy to alveolate, well developed, large subgleba, non-pedicellate, verrucose basidiospores and thick capillitial walls. Therefore, *L. shimousanum* is described here as a new species.

Observations on the detailed structures of capillitial walls are difficult by the usual microscopical methods. However, 3-layered capillitial walls can be clearly observed under fluorescence microscopy. This result suggests that the structures of capillitial walls can be possibly applied to a new taxonomic characters. Therefore, intensive re-examinations on morphology of capillitial walls of *Lycoperdon* using fluorescence microscopy are needed.

Lycoperdon umbrinum Pers. : Pers., Synops. Meth. Fung.: 147, 1801.

Fig. 9.

Basidiomata in small groups, 20-40 mm broad, 25-45

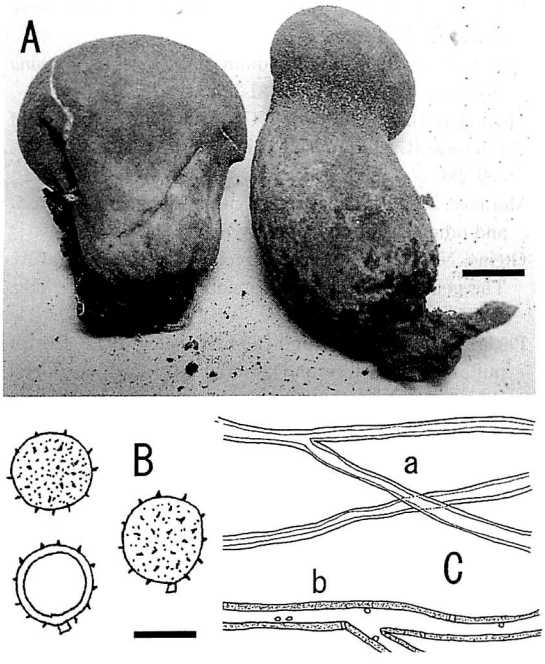

Fig. 9. *Lycoperdon umbrinum* Pers. : Pers. (CBM-FB-11572). A, Basidiomata; B, Basidiospores; C, Capillitia. a, capillitial thread; b, detail of capillitium. Scale bars A: 7 mm, B: 3.5 μm, Ca: 20 μm, Cb: 10 μm.

mm high, subpyriform to pyriform, pale brown to yellowish brown. Exoperidium made of densely, slender, convergent spines, 0.5-1 mm long, pale brown, brown to blackish brown, later sloughing off, not surrounded by granules or warts. Endoperidium papery, shiny, pale brown to grayish brown at the upper part than subgleba, lower ochraceous to yellowish brown. Gleba white when young, later olivaceous brown to sometimes purplish, with indistinct pseudocolumella. Subgleba well developed, spongy to alveolate, olivaceous brown to greyish brown.

Basidiospores globose to subglobose, minutely verrucose, warts up to 0.5-1.0 μm high, (3.5-) 4.0-5.5 μm broad excluding ornaments or (4.0-) 4.5-6.5 μm broad including ornaments, olivaceous brown, with a short, hyaline pedicel up to 1.0 μm long. Basidia not observed. Capillitium *Lycoperdon*-type, 2.5-8.0 μm broad, elastic, pores abundant, aseptate, dichotomously branched, walls 0.5-1.0 μm thick, pitted. Paracapillitium usually abundant.

Habitat: On rich soil or humus in woodland, mainly under conifers.

Distribution: Japan, Europe (Calonge and Demoulin, 1975; Kreisel, 1962; Lange, 1948; Pegler *et al.*, 1995), East Africa (Oteino, 1967), United States (Coker and Couch, 1928) and Mexico (Calderón-Villagómez and Pérez-Silva, 1989).

Specimen examined: Chiba Pref., Chiba-shi, Chuo-ku, Aoba-cho, June 25, 1998, coll. Y. Abe s.n., CBM-FB-16528; same place, July 1, 1998, coll. T. Fukiharu s.n., CBM-FB-16555 and 16556; Chiba Pref., Chosei-gun, Chonan-machi, Kasamoriji, September 24, 1994, coll. E. Sano s.n., CBM-FB-11572.

Japanese name: Tsubu-hokoritake.

Remarks: This species is newly reported here from Chiba Prefecture. This species is morphologically very similar to *L. estonicum* Demoulin, but its basidiospores have long pedicels up to 20 μm long (Demoulin, 1972). In Japanese members of *Lycoperdon*, basidioma structure of this species is similar to *L. umbrinoides*, but the latter has more warty basidiospores and the very thick-walled capillitium (Dissing and Lange, 1962; Kasuya, 2004c; Sarasini, 2005).

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千葉県菌類誌(V) 腹菌類. 2. ホコリタケ科の追加

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前報に引き続き，千葉県立中央博物館に収蔵されている千葉県産ホコリタケ科菌類標本について分類学的検討をおこない，以下の8種を報告した: *Calvatia boninensis* (オオノウタケ), *C. craniiformis* (ノウタケ),

C. rugosa（イロガワリホコリタケ），*C. utriformis*（キクメタケ），*Disciseda candida*（ウスイロドングリタケ，新称），*L. nigrescens*（クロホコリタケ），*L. shimousanum*（シモウサアシブトホコリタケ，新称），および *L. umbrinum*（ツブホコリタケ）．これらのうち，無性基部が発達し，胞子の小柄を欠き，さらに3層からなる厚い弾糸壁を持つ新種 *L. shimousanum* を記載した．また，*D. candida* は日本新産種である．さらに，*C. boninensis, C. craniiformis, C. rugosa, C. utriformis,* および *L. umbrinum* は千葉県新産種である．ノウタケ属，ドングリタケ属およびホコリタケ属に属する4種の菌について，蛍光顕微鏡観察に基づき，胞子と弾糸に認められた自家蛍光を記載した．これらの自家蛍光の観察は，ホコリタケ科の形態分類において重要である可能性が示唆された．