Studies on the species of *Lachnellula* in Hokkaido : their morphology, physiology, and pathogenicity*

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北海道における *Lachnellula* 属菌の種とその形態，生理，病原性に関する研究*

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Contents

* This paper is largely grounded on the thesis presented to the Hokkaido University
本論文は「北海道大学審査学位論文」である。

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〔北海道林業試験場報告　第 19 号　昭和 56 年 10 月　Bulletin of the Hokkaido Forest Experiment Station, No. 19, October, 1981

和 文 摘 要

第1章 緒 論

戦後，森林の復興のために北海道では，人工林の造成にあたり，トドマツ，カラマツの植栽のほかに多くの外国樹種も導入植栽され，人工林面積，植栽樹種とも急増した。このため各種の病害もふえ，*Lachnellula* 属菌によるがんしゅ病の発生もまた増加した。本論文でとりあつかった *Lachnellula* 属菌はヨーロッパでは19世紀初頭から，とくにカラマツがんしゅ病菌として *Lachnellula willkommii* が注目されてきた。我が国では 1957 年に長野県八ケ岳山麓のカラマツ林でこの菌が発見された。その後の調査で富士山，浅間山，八ケ岳山麓のカラマツ人工林，天然林でこの病害による激害林分が発見された。北海道では *Trichoscyphella calycina*（*Lachnellula calyciformis*）によるがんしゅ病が各地で恒常的にトドマソ幼齢林を加害している。さらに近年になって，ストローブマツ，ハイマツにマツ類がんしゅ病が発見された。*Lachnellula* によるがんしゅ病は高緯度地方，高海抜地に発生が多いため，我が国の高緯度地域を占める北海道では，今後，人工林め奥地化にともない *Lachnellula* 属菌によるがんしゅ病の多発が憂慮される。

本論文は道内各地からえた *Lachnellula* 属菌の形態，生理的性質，病原性を明らかにしようとするものであり，あわせて現在多発しているマツ類がんしゅ病被害地の分布，発生について考察を加えたものである。

第2章 *Lachnellula* 属の変遷

FUCKEL（1869）はコップ状の子のう盤，小さな子のう，単細胞で無色の子のう胞子をもつ有毛の無弁盤子菌に *Dasyscypha* 属を創設したが，その属の特徴の記載は簡単でまた，基準種のタイプ標本の指定もなく，基準となる側糸の特徴も明らかにしなかった。このため，この属は種々の変遷をへた後，NANNFELDT（1932）は繊維状の側糸をもつものに対して *Trichoscyphella* 属を創設した。さらに，DENNIS（1963）は子のう胞子が円形だという特徴だけで *Trichoscyphella* 属と区別されていた *Lachnellula* 属とこの *Trichoscyphella* 属を併合して *Lachnellula* 属とした。DHARNE（1965）は DENNIS の *Lachnellula* 属をうけいれるとともに，針葉樹に寄生し，子のう盤は柄をもち，外側は微細な毛でおおわれ，側糸は繊維状で，ときに連珠状の側糸をもつなどの特徴をもった無弁盤子菌類に制限した。

第3章 *Lachnellula* 属菌の一般的形態

子のう盤は寄主の樹皮上に単生あるいは群生し，ろう質あるいは肉質である。子のう盤は柄をもち，はじめ球状でのちに開き円盤形になり，高湿度の状態ではコップ形あるいは皿形になる。子のう盤の外側は毛でおおわれ，白，橙あるいは緑色である。毛の外側は微粒子でおおわれ，無色あるいは黄褐色，暗褐色でときに青緑色である。子実層は子のうと側糸からなり，黄色，橙色ときに赤色あるいは褐色である。子のうは円筒形で，そのなかに斜に1列あるいは2列，ときに不規則に8個の胞子をもつ。子のう胞子は平滑で無色，普通は単胞であるが発芽のとき2胞になる種もある。形は球形，楕円形，紡錘形などである。側糸は一般に子のうより長く，繊維形あるいは円筒形であり，ある種では連珠形の側糸がまじる。側糸は隔壁，微小な油滴をもつ。不完全世代の子実体は病患部にまれにできる。分生子殻子座は樹皮上に散生ないし群生し，はじめ埋生のちに表皮を破って表生する。分生子殻は迷路状になり，黄色，橙色あるいは褐色になる。分生子柄は殻窩の内面に並列し，単条あるいは分岐する。分生子は円筒形，楕円形，紡錘形等多形で，無色ときに1

～2 個の油滴をもち，分生子塊となって外界に噴出する。

第 4 章　我が国でのがんしゅ病研究史

白井がモミ上で発見し，HENNINGS（1900）によって *Dasyscypha abieticola* と命名されたものが，我が国でのがんしゅ病研究の最初である。その後，1957 年になって，長野県八ケ岳山麓でカラマツがんしゅ病菌である *Lachnellula willkommii* が発見され，精査の結果，富士山，浅間山，八ヶ岳山麓のカラマツ人工林，天然林でこの病害による激害林分が発見され，その病原菌，被害解析などについて詳細な報告が行われている。北海道では *Trichoscyphella calycina* によって，トドマツがんしゅ病が発生することを亀井（1962）が確かめた。その後，この病気の発生，病原菌についての多くの報告が行われた。マツ類のがんしゅ病については，佐保・高橋（1972），小口（1979）がその病原菌や被害についての報告を行った。

第 5 章　がんしゅ病の病徴と標徴

病斑は早春に罹病木の幹あるいは枝上に円形あるいは縦の楕円形にでき，患部は赤褐色になる。春から夏のはじめにかけて，患部の樹皮は陥没し，枯死した組織と生きている組織との間に明瞭な境界ができる。やがて，病患部の樹皮はざらざらになり，ひびがはいり，そこから樹脂が流出する。子のう盤は早春から晩秋にかけて病患部にできる。最後に罹病木は病患部から上部の幹あるいは枝は枯死する。5 年生以上の比較的太い幹をもつ樹木での幹の病斑は毎年進展し，がんしゅの形成が長年にわたって継続する。

第 6 章　材料と方法

道内各地で採集した標本を検鏡し，本属に属するかどうかを確認し，本属の 12 種をえ，それらの菌の特徴を測定するとともに，それらを描画した。この 12 種の生理実験を麦芽寒天，馬鈴薯寒天，ワックスマン寒天，ツァペック寒天の 4 種の培地を用いて行い，菌そうの発育調査のため，その直径を測定するとともに，菌そうの特徴を観察した。また寄主植物の殺菌枝を用い，それに接種し，分生子，子のう盤の形成を観察した。さらに培地上，殺菌枝上に生じた分生子の発芽の有無を確かめた。12 種の病原性を確かめるため，鉢植えの寄主植物に接種を行い，接種木の病斑の進展，分生子殻，子のう盤の形成，接種木の生否を観察した。

第 7 章　実験結果

1.　北海道産 *Lachnellula* 属菌 12 種をえた。このうち *L. aeruginosa* は新種であり，*L. abietis*, *L. ciliata*, *L. fuckelii*, *L. hahniana*, *L. occidentalis* の 5 種は日本新産種である。

2.　トドマツがんしゅ病菌とされていた *Trichoscyphella calycina* を *Lachnellula calyciformis* の異名とした。

3.　*L. aeruginosa*, *L. ciliata*, *L. pini* を除いた 9 種は麦芽，馬鈴薯寒天培地上で，また上記 3 種はワックスマン寒天培地上でよく発育した。一方全ての種はツァペック寒天培地上では発育がわるいか，まったく発育しなかった。

4.　*L. arida*, *L. calyciformis*, *L. hahniana*, *L. subtilissima* の 4 種は子のう盤を，また L. -*abiestis*, *L. aeruginosa*, *L. calyciformis*, *L. ciliata*, *L. microspora*, *L. subtilissima* の 6 種は分生子塊を麦芽寒天培地上に形成した。

5. *L. aeruginosa*, *L. fuckelii*, *L. hahniana*, *L. microspora*, *L. occidentalis*, *L. suecica* は小分生子を麦芽寒天培地上に，*L. pini* 馬鈴薯寒天培地上に形成した。

6. *L. abitis*, *L. calyciformis*, *L. ciliata*, *L. subtilissima* の 4 種は寄主植物の殺菌枝上に子のう盤を形成した。

7. *L. aeruginosa*, *L. calyciformis*, *L. ciliata*, *L. microspora*, *L. subtilissima* の 5 種は分生子塊を寄主植物の殺菌枝上に形成した。

8. 分生子は培養温度 15～25℃の範囲で 72 時間以内に麦芽寒天培地上でよく発芽した。

9. 分生子からの分離菌株は子のう胞子からの分離株と生理的性質がよく一致した。

10. *L. abietis*, *L. calyciformis*, *L. pini* はストローブマツの幹，枝上で病斑を進展させ，また *L. calyciformis* はトドマツ上で病斑を進展させた。これら 3 種は病斑上に子のう盤を形成し，*L. abietis*, *L. calyciformis* は分生子殻を病斑上に形成した。

11. 接種試験の結果 *L. calyciformis* はトドマツがんしゅ病菌であること，また *L. abietis*, *L. calyciformis*, *L. pini* の 3 種は，いずれもストローブ，マハイマツのがんしゅ病菌であることを確認した。

12. 現在多発しているマツ類がんしゅ病被害地は北海道中央部以北の最深積雪量が 2m以上，積雪日数が 150 日以上の地域のストローブマツ人工林と高海抜地のハイマツ天然林に分布している。

第 8 章～10 章　研究結果

研究結果を総括して，北海道産 *Lachnellula* 属の種の検索表，寄主目録，寄主範囲を記した。

I Introduction

Many species of the genus *Lachnellula* (*Dasyscypha, Trichoscyphella*), which have been reported in the literature are associated with canker diseases on numerous conifers. Especially, *Dasyscypha willkommii* has been well known for many years in Europe, Britai and North America as European larch canker fungus. And some other species, such as *Dasyscypha pini*, *D. calyciformis*, *D. pseudotsugae*, and *Trichoscyphella calycina* are reported as the fungi cause of coniferous canker diseases. The disease caused by these induced a great deal of losses to tho conifers.

In Japan, European larch canker fungus was first collected on the branches of Japanese larch (*Larix leptolepis* GORDON) at the foot of Mt. Yatsugatake in Nagano Prefecture in 1957 (ITO and ZINNO 1957), Thereafter, the serious outbreaks of the canker disease caused by this fungus have been found in the natural forests and the plantations of Japanese larch at altitude of 1,400～1,800 m around Mt. Yatsugatake, Mt. Fuji , and Mt. Asama in the central parts of the Main Island of Japan (ITO 1961 ; KOBAYASHI and UOZUMI 1962 ; ITO, ZINNO and KOBAYASHI 1963) .

In Hokkaido, *Trichoscyphella calycina* which causes the destractive damage to relatively young todo fir (*Abies sachalinensis* MAST.) plantations has been found by KAMEI (KAMEI 1962) . More recently, pine canker disease caused by *Lachnellula* spp. was found in eastern white pine (*Pinus strobus L.*) plantations and in Japanese stone pine (*P. pumila* REGEL) areas (TAKAHASHI and SAHO 1973 ; OGUCHI 1979) .

The purpose of the paresent paper is to identify the species of *Lachnellula* spp. collected in Hokkaido by through the morphological, physiological studies and to clarify their pathogenicity.

The author wishes to express his special appreciation to Professor Dr. Kazuyoshi MUTO, Hokkaido University, for his instructive criticism and sustained encouragement throughout the study. He also expresses his heartist thanks to Professor Dr. Tadao UI, and Assistant Professor Dr. Tsuneo IGARASHI, Hokkaido University, for helpful advice and kindness in reading the manuscript.

Greatful acknowledgement is made to Dr. Takao KOBAYASHI, Forestry and Forest Products Research Institute, and Mr. Ikuo TAKAHASHI, Tokyo University Forest in Hokkaido, for their useful suggestions and encouragment throughout the stuby.

II Historical review on the concept of *Lachnellula* KARST.

FRIES (1822) described one of the species under the name of *Peziza calycina* SCHUM. ex FR. and classified it in his tribe Dasyccyphae. His tride Dasyscyphae is hairy inoperculate Discomycetes with cup shaped apothecia, small ascus, and unicellular and hyaline ascospores. In 1869, FUCKEL raised the Friesian tribal name, Dasyscyphae to the status of the genus as *Dasyscypha*. He briefly described his new genus, but he defined neither morphological characters of the paraphyses nor name of the type specus. FUCKEL (1869) at the same time listed seven species under his new genus, of which only *Dasyscypha calycina* has the filamentous paraphyses, while the other six species have lanceolate paraphyses. KARSTEN (1871), in order to avoid confusion made by FUCKEL, created new genus *Lachnum* KARST. This genus was based on characters similar to those of *Dasyscypha* FUCK. but was limited to only species with lanceolate paraphyses. The species with filamentous paraphyses including *Dasyscypha calycina* SCHUM ex FR. were removed to *Helotium* FR. BOUDIER (1885) placed the hairy, stalked species with lanceolate paraphyses in *Dasyscypha* FUCK., ignoring *Lachnellula* KARST. BOUDIER (1885) included. *D. bicolor*, *D. cerina*, *D. virgenea*, and *D. brunneola* in this *Dasyscypha.* He also erected the genus *Trichoscypha* BOUD. for the species with filamentous paraphyses and included only species, *Trichoscypha calycina* (SCHUM. ex FR.) BOUD. REHM (1887-1896) accepted *Lachnum* KARST. for the species with lanceolate paraphyses and the species with filiform paraphyses were placed in *Dasyscypha* FUCK. NANNFELDT (1932) pointed out that the generic name, *Trichoscypha* BOUD. was a homonym of the older *Trichoscypha* HOOKER, and he revised BOUDIER's genus, *Trichoscypha* as *Trichoscypha* NANNF. He revived *Lachnumnum* KARST. for the species with an exciple of 'textura prismatica' and usually lanceolate paraphyses. HAHN and AYRRS (1934) followed REHN and named *Dasyscypha calcina* sensu FUCKEL as lectotipe for the *Dasyscypha*. DENNIS (1949), however, followed NANNFELDT (1932) in accdpting *Trichoscyphella* NANNF. but used *Dasyscypha* FUCK, in place of NANNFELDT's *Lachnum* KARST., since *Dasyscypha* antedates *Lachnum* KARST. and lanceolate paraphyses are not strictly a diagnostic character of *Lachnum* KARST. SEAVER (1951) followed PHILLIPS (1887) and placed the hairy inoperculate Discomycetes with lanceolate and filamentous paraphyses on conifers in *Lachnella* FR., DENNIS (1960) accepted NANNFELDT's genus, *Trichoscyphella* NANNF. for the species with cylindrical or moniliform paraphyses and for the species always occurring on the bark of conifers. But DENNIS (1949) amended *Dasyscypha* FUCK. as *Daysyscyphus* S. F. GRAY for the species with lanceolate paraphyses. Most species of *Dasyscypha* have distinctly lanceolate paraphyses, In 1962, DENNIS followed BOUDIER (1907) and reunited *Lachnellula* KARST. and *Trichoscyphella*, NANNF. and accepted the generic name *Lachnellula* KARST. DENNIS (1962), in his report : 'A reassessment of *Belonidium* MONT. & DUR., stated the reason why he united two genera as the followings : "It is difficult to distinguish these two genera because there are a number of species with minute subglobose or ovoid ascospores and the most logical course seems to be to unite them." And he extended the genus to the species with septate ascospores and also to the species occurring on wooby plants other than the c-

onifers. DHARNE (1965) followed DENNIS (1962) and accepted the generic name, *Lachnellula* KASST. but, to avoid confusion, he limited the genus to those hairy, inoperculate Discomycetes occurring on the conifers bark. Thus the seventeen species were included in this genus.

Ⅲ Morphology of *Lachnellula* KARST.

Apothecia occur solitary or aggregate on the outer bark of the host plants, and are waxy or fleshy tissue. Apothecia have a stipe or short-stipe, at first they are a globose form, their edges close tightly, then apothecia open up to be a rounded form, and expand as a cup-like or saucer-like form under moist condition. The externals of apothecia are hairy, white, buff, yellow, or bluish green. Hairs are minutely roughened on the outside, hyaline or yellowish brown to dark brown, occasionally bluish green. They are thin-walled, cylindrical with sub-acute, gently rounded extremites, and septate. Hymenium is aggregation of asci and paraphyses and it is yellow, orange, or orange-yellow to salmon-yellow, but a few species are red or brown. The outmost layer of the tissue, including the margin of the apothecium (ectal ex-cipulum), is 'texture glabulose' to 'texture oblita' type. The zone of the tissue enclosed between the hypothecium and the ectal excipulum (medullary exciplum) is 'texture intricate', which is composed of loosely interwoven, thin walled hyphae.

Asci are cylindrical, clavate, or cylindrically clavate with rounded or subacute apices, and usually contain eight ascospores, which are arranged obliquely uniseriately or biseriately, rarely partially biseriately or irregularly. Ascus pore is not always stained by iodine.

Ascospores are smooth, hyaline, and usually continuous inside the ascus, but some species become uniseptate upon germination. Ascospores are globose, elliptical, spindle-shaped, or elongate-elliptical to oblong-elliptical with obtuse or acute apices at one or both ends.

Paraphyses are generally longer than the asci and filiform, filamentous or cylindrical form with obtuse or subacute apices. Some species have the moniliform or submoniliform paraphyses which are interspersed with the paraphyses. They have septa and minute oil globules.

Pustules of imperfect stage of this genus infrequently occur in a group on the bark of cankered branch or stem. Conidial stroma immerse in the peridermal layer of the bark, and then erumpent through epiderm. Conidial cavities become labyrinthform with age and are yellow, orange yellow or light brown to dark brown.

Conidiophores arise from the innermost layer of cavity and are simple or verticillately branched. Conidiophores are noticeably septate.

Conidia are one cell and have various shape (namely cylindrical, elliptical, oblong-elliptical, spindle-shaped, ovoid, oblong, or spherical), and are hyaline and occasionally contain one or two globules. Conidia are exuded out in masses.

Ⅳ History of the canker disease of *Lachnellula* in Japan

A species of *Dasyscypha* was found by SHIRAI on Nikkô fir (*Abies brachyphylla* MAXIM. =A. *homolepis* S. et Z.) at Nikkô, Tochigi Prefecture and HENNINGS has described it as *Dasyscypha abieticola* P. HENN. et SHIR. (HENNINGS 1900). This fungus was the first reported as a species of *Dasyscypha* in Japan. Recently, UOZUMI (1965) reported the instance of this fungus on Japanese fir (*Abies firma* S. et Z.), Nikkô fir, and veitch fir (*A. veitchii* LINDL.), and he (1967) also reported that the Nikkô fir plantation in Mt. Fuji National Forest was suffered from this fungus. More recently, ITO (1973) revised the generic name of this fungus as *Trichoscyphella abieticola* (HENN.) K. ITO.

The larch canker caused by *Dasyscypha willkommii* in Europe was introduced by IDETA (1909) in his handbook KITAJIMA (1933) presented a brief account of a cankerous disease of Japanese larch (*Larix leptolepis* GORDON) occurring in the Tôhoku district, but he did not investigate its causal agent. KAMEI (1956) reported that he observed the larch canker fungus on Korean larch (*L. olgensis* var. *koreana* NAKAI) in Saghalien. In 1957, ZINNO collected a *Dasyscypha* sp. on branches of Japanese larch at the foot of Mt. Yatsugatake in Nagano Prefecture (ITO and ZINNO 1957). It was identified by ITO and ZINNO as *Dasyscypha willkommii* (HART.) REHM, and they reported the disease and the morphology of the causal fungus (ITO and ZINNO 1957). This was the first detailed report of Japanese larch canker disease in Japan. Recently, serious outbreaks of the canker have been found in the Japanese larch plantations at the east foot of Mt. Yatsugatake in Nobeyama National Forest (Nagano Prefecture), and the analyses of the damage were reported by ITO (1961) and KOBAYASHI and UOZUMI (1962). Since that time, further surveys have been carried out at the regions around Mt. Yatsugatake, Mt. Fuji, and Mt. Asama. The results of these surveys showed a more extensive distribution of the disease in Nagano, Yamanashi, and Shizuoka Prefectures, the central parts of the Main Island of Japan, and the disease affects severely larch trees not only in the plantations but also in the native forests (ITO, ZINNO, and KOBAYASHI 1963).

KAMEI (1961) collected a *Trichoscyphella* sp. on Kurile larch (*Larix gmelinii* GORD) in Wakkanai, the northern part of Hokkaido, and he reported it as *Trichoscyphella willkommii* NANNF., KOBAYASHI (1970) stated that Japanese larch canker fungus was native in Japan. SAHO and TAKAHASHI (1973) listed this species on *Larix* spp.

IDETA (1909) and HARA (1936) reported that silver fir (*Abies* spp.) was affected by *Dasyscypha calyciformis* (WILLD.) REHM. KOBAYASI, IMAZEKI, and ASUYAMA (1939) described this fungus in their book. KAMEI (1959) reported that todo fir (*Abies sachalinensis* MAST.) canker was caused by *Dasyscypha calyciformis.* This was the first report on todo fir canker disease in Japan. Thereafter, he (1961) revised this fungous name as *Trichoscyphella calycina* (SCHUM. ex FR.) NANNF. YOGO and ONO (1961) reported the correlation between the outbreak of this canker disease and the several agents (aphid, bark beetle, vole, and *Phomopsis* canker). YOKOTA and MATSUZAKI reported several studies of this disease and causal fungus (YOKOTA and MATSUZAKI 1966, 1968, 1971a, 1971b ; MATSUZAKI and YOKOTA 1965,

1970, 1972). UOZUMI (1965) reported the instance of this fungus on Nikkô fir and Kurile larch. OGUCHI (1972) found a *Trichoscyphella* sp. having large asci and ascospores on todo fir.

SAHO and TAKAHASHI (1972) reported that the canker disease of eastern white pine (*Pinus strobus* L.) was caused by *Lachnellula fuscosanguinea*. Next year, they revised the causal fungus as *Lachnellula pini* (BRUNCH.) DENNIS (TAKAHASHI and SAHO 1973). OGUCHI (1979) reported that canker disease of *Pinus strobus P. pumila* was caused by *Lachnellula* spp. in Hokkaido.

SAHO and TAKAHASHI (1973) listed *Lachnellula arida*, *L. microspora*, *L. subtilissima*, and *L. suecica*, *Abies*, *Larix*, and *Pinus* in their list : 'A check list and host index of fungi on forest trees collected at the Tokyo University Forest in Hokkaido, Japan (1960-1972) : More recently, these fungi were described by TAKAHASHI (TAKAHASHI 1979).

V Symptoms and signs of the disease caused by *Lachnellula* spp.

In spring, just after the snow disappears, the lesions develop circularly or longitudinally on stems and branches of young trees. The infected parts become reddish brown. From spring to early summer, the diseased bark is clearly depressed, and there appears a clear distinction between the dead and the healthy tissues. Then the bark of infected tissue become roughened and cracked, and white resin flows from the lesions. From early spring to late autumn, apothecia appear on the lesions. Eventually, cankers girdle the small branches or stems of the young trees resulting in the death of upper or outer parts. On the other hand, diseased trees older than five years survive, but the lesions on the stems develop yearly forming perennial cankers.

VI Materials and methods

A Materials

Many materials of *Lachnellula* spp. were collected from various localities in Hokkaido, and some herbarial materials were collected in the Main Island of Japan. The various tree species are tested in the present paper and they are showed summarily in Table 1,

Table 1. Host plant and number of materials tested in the present paper
表－1 寄主と標本数

Host 寄主		Locality 場所	Hokkaido	Honshû	Total
Abies	*sachalinensis*	(トドマツ)	44		44
	holophylla	(チョウセンモミ)	3		3
	homolepis	(ウラジロモミ)	3		3
	veitchii	(シラビソ)	2		2
	alba	(ヨーロッパモミ)	2		2
	mariesii	(オオシラビソ)		1	1
Larix	*gmelinii*	(グイマツ)	7		7
	olgensis var. koreana	(チョウセンカラマツ)	3		3
	gmelinii×*leptolepis*	(グイマツ×カラマツ)	3		3
	leptolepis	(カラマツ)	1	1	2
Pinus	*strobus*	(ストローブマツ)	12	1	13
	pumila	(ハイマツ)	10	3	13
	sylvestris	(ヨーロッパアカマツ)	3		3
	banksiana	(バンクスマツ)	2		2
Picea	*jezoensis*	(エゾマツ)	1		1
Pseudotsuga	*taxifolia*	(ダグラスモミ)	1		1
	Total		97	6	103

As shown in Table 1, the material collected from *Picea* was only one. In fact, it may be said that the present work is concerned with *Lachnellula* spp. on *Abies*, *Larix*, and *Pinus*.

B Methods

1 Morphological studies

Many of the materials were hand-sectioned to make certain whether materials belong to the member of *Lachnellula*. In order to examine fine morphology of the fungi, several abequate pieces made by hand-sectioning were mounted in either a drop of SHEAR's mounting fluid* (ITO 1959) or sterilized water, and covered with a cover-glass and sealed with nail enamel. Fruit body produced on cultural medium or sterilized twigs was usually squashed in the SHEAR's fluid or sterilized water, and in some cases it was sectioned and treated in the same manner as the above-mentioned. MELZER reagent** (KORF 1953) is applied to examine iodin reaction of ascus pore, and cotton blue*** (KOBAYASHI and INDO 1955) was used to stain the textures of the sectioned apothecium.

* Potassium acetate 1g, Alcohol 30 ml, Glycerol 20 ml, Distilled water 50 ml.

** Iodine crystals 0.5g, Potassium iodide 1.5 g, Chloral hydrate 20 g, Distilled water 20 ml.

*** Alcohol saturated with cotton blue 10 ml, Glycerol 10 ml, Distilled water 80 ml.

2 Physiological studies

a Growth of colony on various agar media

Isolates of *Lachnellula* spp. used in this studies were isolated by the modified KEIT's method (KEIT 1915 ; KAWAMURA 1934) from the sectioned or squashed apothecium of several fungi, and the single spore cultures were obtained. Stock cultures were maintained on slants of malt extract agar at 15℃ under dark condition. In this cultural experiments, the following four agar media were used, and they were sterilized by autoclaving for 10 minutes at 120℃, and 20 ml of several agar media were poured into Petri dishes (9 cm diam.). These Petri dishes were inoculated with respective fungi by placing one inoculum disc in the center of the agar medium in each dish, The inoculum was prepared by using a sterilized cork borer (6−7mm diam,) to cut disc of agar containing mycelia from the edge of 4～6 - week - old colonies. The inoculated Petri dishes were incubated at 15℃ under dark condition. The amount of growth was determined by those two diameters measured at right angles to each other to give the average diameter of each colony with a millimeter ruler. The mycelial growth was observed and measured at intervals of 10 days for 2 months.

Media used are as follows :

1) Malt extract agar	
Malt extract (Difco Ladoratories)	20 g
Agar	20 g
Distilled water	1,000 ml
2) Potato dextrose agr	
Potato dextrose agar (Difco Laboratories)	39 g
Distilled water	1,000 ml
3) Waksman's solution agar	
KH_2PO_4	1 g
$MgSO_4 \cdot 7H_2O$	0.5 g
Glucose	10 g
Pepton	5 g
Agar	20 g
Distilled water	1,000 ml
4) Czapek's solution agar	
$NaNO_3$	2 g

K_2HPO_4	1 g
$MgSO_4 \cdot 7H_2O$	0.5 g
KCI	0.5 g
$FeSO_4 \cdot 7H_2O$	0.01 g
Sucrose	30 g
Agar	20 g
Distilled water	1,000 ml

b Growth on stersterilized twigs

Test tubes (20cm×2.5cm) containing 5 ml malt extract agar and a twig of todo fir (*Abies sachalinensis* MAST.), Japanese larch (*Larix leptolepis* GORD), or eastern white pine (*Pinus strobus* L.), 5 - 10 mm in diameter and 10 - 15 cm in length, were autoclaved at 120℃ for 20 minutes. After sterilization, the agar medium in test tubes was inoculated with mycelia of respective isolates. The test tubes were incubated at 15℃ under dark condition for about 3 months, and then they were kept under laboratory conditions.

3 Pathogenic studies

Inoculation experiments were carried out to determine the pathogenicity of collected *Lachnellula* spp. The inocula used were obtained from pure cultures prepared from single asco-spore isolates of each species. The potted trees were prepared for this experiments. These potted trees were divided into the two groups ; the first one was composed of untreated healthy trees and designated as the healthy group, the second one was composed of trees pretreated with knifed girdles of the stems, and designated as the unhealthy group. Inoculation were made on the portions of the ste stems or branches of the several trees. First, a part of the bark was burned with the head of a red-hot nail. Then a cross slit was made on the burned part with a sterilized scalpel. In the inoculation pots, inoculum from agar media was inserted into each cross slit on the burned bark. The same procedure, with the exception of inoculum, was conducted on the trees in the check pots. Here, sterilized agar block was used as inocula instead of mycelial pieces. Inoculated parts were covered with sterilized gauze, soaked with sterilized water, and again covered with polyethylene sheet for 14 days. The inoculated trees were kept under snow for one or more winter seasons.

4 General references and abbreviations

Scientific name of the trees generally followed UEHARAs 'Illustrated woody plants (in Japanese)' (1959), and REHDER's 'Manual of cultivated trees and shrubs' (1965).

In this paper, collector's name was abbreviated as follows : T. O. : Takeo OGUCHI.

Ⅶ Experimental results

A *Lachnellula* spp. on *Abies*

1 Morphological studies

a Lachnellula aeruginosa OGUCHI sp. nov. (Fig. 1).

Apotheciis sparsis, solitariis, erumpentibus, stipitatis, primo globosis, dein cyathiformi-bus, extus atroviridibus vel obscuro-virentibus, dein isabellinis vel melanoviridibus, 1 - 3 mm diam ; pilis elongatis, cylindraceis, septatis, minutissime verrucosis, primo viridi-flavis, dein subgriseo-olivaceis vel luteolis ; disco primo aurantiaco vel isabellino dein succineo vel ochraceo ; ascis octosporis, clavatis vel subcylindraceis, 43.5 - 65×4.5 - 6.5 μm, plerumque 50 - 60×5 - 6.5 μm ; ascosporis monostichis, continuis, hyalinis, subglobosis vel ovoideis, 3.5 - 6×2 - 5 μm, plerumque 4 - 5×2.5 - 4 μm ; paraphysibus filiformibus, flexuosis 65 - 82 μm longis, 2.5 - 3.5 μm crassis.

Conidiis in cultura hyalinis, continuis, globosis vel ellipticis, 2 - 4.8×1.5 - 3 μm, plerum-que 2.5 - 4×2 - 3 μm ; microconidiis hyalinis, continuis, oblongo-ellipsoideis vel allantoideis, 3 - 6×0.8 - 1.5 μm, plerumque 4 - 5×1.2 μm.

Apothecia scattered, solitary, erumpent, stalked, at first globose, then cup-shaped, ex-ternally dark herbage green to dull green when fresh, isabelline to greenish black when aged, externally green to dark green, covered with excipular hairs. Hairs cylindrical, septate, minutely roughened. Discs orange to isabell-ine when fresh, amber to ochreous when aged.

Asci 8 - spored, clavate to subcylindrical, 43.5 - 65×4.5 - 6.5 μm, mostly 50 - 60×5 - 6.5 μm. Ascospores uniseriate, continuous, hya･line, subglobose to ovoid, 3.5 - 6×2 - 5 μm, mostly 4 - 5×2.5 - 4 μm. Paraphyses filiform, flexuous, 65 - 82×2.5 - 3.5 μm.

Conidial stage was not observed in nature but conidial masses are produced on maltextract agar. Conidia continuous, spherical to elliptical, hyaline, 2 - 4.8×1.5 - 3 μm, mostly 2.5 - 4×2 - 3 μm, Conidia germinate on malt extract agar. Microconidia were produced on malt extract agar and sterilized twigs of todo fir. Microconidia continuous, hyaline, elliptical-oblong or allantoid, 3 - 6×0.8 - 1.5 μm, mostly 4 - 5 × 1 - 1.2 μm. Micro conidia did not germinate.

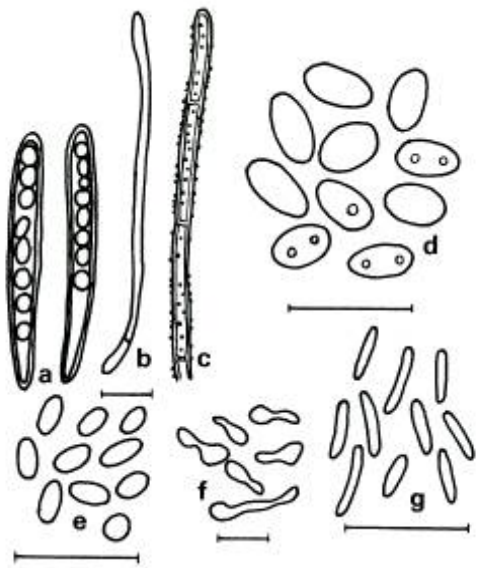


Fig.1. (図－1) *Lachnellula aeruginose* OGUCHI sp. nov

a : Asci and ascospores 子のうと子のう胞子

b : Paraphysis 側糸 c : Hair 毛

d : Ascospores 子のう胞子 e : Conidia 分生子

f : Germinating conidia 分生子の発芽

g : Microcondia 小分生子 (├──┤ =10 μm)

Host and Material : *Abies sachalinensis* MAST. (Todo - matsu) ——— Yamabe, Furano, VI - 9, 1973 by I. TAKAHASHI ; Yamabe, Furano, VII - 24, 1973 by T. O. ; Yamabe, Furano, VII - 30, 1974 by T. O. The type speciman has been deposited in Hokkaido Forest Experiment Station, Bibai, Hokkaido (Holotype HFPL - 1).

Distribution : Asia (Japan)

Note : The species which have externally greenish apothecium have hitherto been recognized in genera related to *Lachnellula*. PHILLIPS (1887) described *Lachnella tricolor* (SOW.) PHILL. This specific epithet, *tricolor*, refers to the blue-grey exterior, yellowish hymenium, and nearly white stem. MORGAN (1902) described *Lachnum viridulum* MASSE and MORGAN, and this species has dark green disc, and the externalis pale green. KANOUSE (1935) described *Lachnella tricolor* var. *microspora* KANOUSE. SEAVER (1951) reported *Lachnlla viridicome*, *L. pulveracea*, and *L. microspora* as having greenish hairs. But *L. pulverancea* and *L. microspora* are identical with the species of MORGAN (1902) and KANOUSE (1935). These species occurred on rotten, decaying wood and *Quercus* sp.

This new species has similar appearance to *Lachnella viridicoma*, but the species is evidently distinguished from *L. viridicoma* by the shape and size of ascospores and the habitat. This species is a rare species and easily recognized by its blue exterior.

b *Lachnellula calyciformis* (WILLD. ex FR.) DHARNE

Phytop. Zeits., 53 : 124, 1965. (Fig. 2)

Synonym : *Dasyscypha calyciformis* (WILLD.) REHM Rabenh. Krypt-Fl., III : 834, 1896 ; MAUBLANC, Bull. Soc. Mycol. France, 20 : 232, 1904 ; SCHELLENBERG, Mitt. Schweiz. Zentralanst. Forstl. Versuchswesen, 8 : 269, 1905 ; KOBAYASHI, IMAZEKI and ASUYAMA, Nippon Inkwasyokubutu Dukan : 299, 1939 ; BINGHAM and EHRLICH, Mycol., 35 : 98, 1943 ; KUJALA, Commun. Inst. For. Fenn., 38 : 22, 1950 ; KAMEI, Hoppô Ringyô Sôsho, 12 : 83, 1959.

Trichoscyphella calycina (SCHUM. ex FR.) NANNF, Nova Acta Reg. Soc, Sci. Upsal. ser, VI, 8 : 300, 1932 ; KAMEI, Res. Bull. Coll. Exp. Forests, Coll. Agr. Hokkaido Univ., 21 : 235, 1962.

Apothecia scattered or grouped, erumpent, stalked, at first globular and closed, then opening in a cup-shaped and expanding to saucer-shaped under moist conditions. Externally white, covered with excipular hairs. Hairs septate, minutely roughened, hyaline, gently rounded extremities. Discs orange to orange-yellow, 0.2 - 0.5 mm rarely reaching to 3 mm in diameter.

Asci cylindrical to cylindrical-clavate, apex obtusely rounded, 40 - 57.5×3.7 - 6.2 μm, mostly 40 - 50×4.5 - 5 μm, 8 - spored. Ascospores obliquely uniseriate, elliptic, continuous, hyaline, 4.5 - 7×2 - 3.5 μm, mostly 5 - 7×2 - 3 μm. Paraphyses filiform, with rounded or subacute extremities, minute oil globules, 52 - 72×2 - 3 μm.

Conidial stage consisting waxy, fleshy, erumpent, stromata with irregular, labyrinthiform cavities in which the conidia are abstricted from the tips of slender, acutely pointed, simple or verticillately branched conidipholes. Conidia continuous, spherical to elliptical, hyaline, 2.5 - 5×1.5 - 2.2 μm,

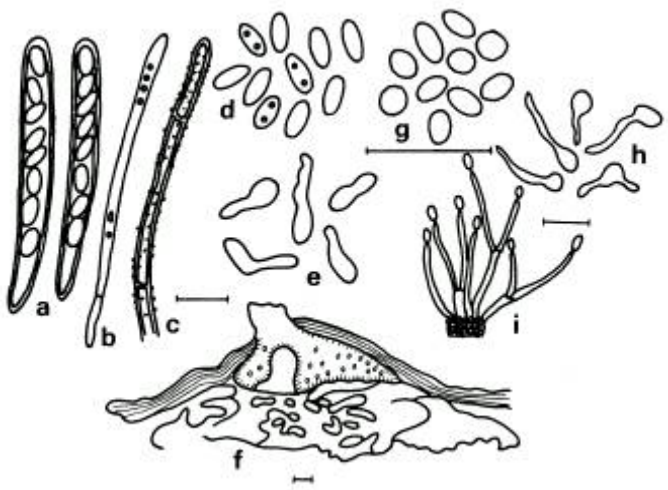


Fig.2.（図－2）　*Lachnellula calyciformis*（WILLD. ex FR.）DHARNE

a : Asci and ascospores　子のうと子のう胞子　b : Paraphysis　側糸
c : Hair　毛　d : Ascospores　子のう胞子
e : Germinating ascospores　子のう胞子の発芽　f : Pycnidium　分生子殻
g : Conidia　分生子　h : Germinating conidia　分生子の発芽
i : Conidiophores　分生子柄（├──┤ : a～e, g～i ＝10μm ; f＝μm）

mostly 3 - 4×1.5 - 2μm. Conidia germinate on malt extract agar.

Host and Material : *Abies mariesii* MAST.（Ôsirabiso）——— Kiso, Nagano, Ⅶ - 26, 1967 by T. HAMA. *Abies sachalinensis* MAST.（Todo-matsu）——— Tôhoro, Nemuro, Ⅶ - 5, 1969 by T. O. ; Yamabe, Furano, V - 13, 1971 by T. O. ; Kimobetsu, Shiribeshi Ⅵ - 2, 1971 by T. O. ; Yoshida, Takikawa, XI - 16 1971 by T. O. ; Kôshunai, Bibai Ⅶ - 20, 1972 by T. O. ; Toyotomi, Sôya, Ⅶ - 25, 1972 by T. O. ; Kôshunai, Bibai, Ⅷ - 11, 1972 by T. O. ; Kôshunai, Bibai, Ⅳ - 28, 1973 by T. O. ; Kôshunai, Bibai V - 10, 1973 by T. O. ; Kôshunai, Bibai, V - 24, 1973 by T. O. ; Kôshunai, Bibai, Ⅵ - 9, 1974 by T. O. ; Nopporo, Ebetsu, Ⅷ - 2, 1974 T. O. ; Pifuka, Kamikama, X - 8, 1974 by T. O. ; Fûren, Kamikawa X - 17 by T. O. ; Nishishibetsu, Shibetsu, X - 17, 1974 by T. O. ; Kôshunai, Bibai, XI - 7, 1974 by T. O. ; Rumoi, Ⅶ - 10, 1975 by T. O. ; Ikomanbetsu, Kamikawa, Ⅶ - 25, 1975 by T. O. ; Honbetsu, Ikeda, Ⅸ - 9, 1975by M. AKIMOTO ; Yamabe, Furano, Ⅸ - 11, 1975 by T. O. ; Shintoku, Tokachi, X - 15, 1975 by T. O. ; Nayoro, X - 23, 1975 by T. O. ; Pifuka, Kamikawa, Ⅷ - 11, 1977 by T. O. ; Horokanai, Sorachi, Ⅵ - 20, 1977 by S. YOKOTA ; Ashoro, Tokachi, Ⅵ - 26, 1977 by Y. MURATA ; Kôshunai. Bibai, Ⅷ - 6, 1977 by T. O. ; Shintotsukawa, Sorachi, Ⅷ - 12, 1977 by M. AKIMOTO ; Kôshunai. Bibai, Ⅸ - 28, 1977 by T. O. ; Kôshunai, Bibai, X - 27, 1977 by T. O. *Abies veitchii* LINDL.（Shirabiso）——— Kôshunai, Bibai, XI - 7, 1974 by T. O. ; Kôshunai, Bibai, Ⅳ - 20, 1978 by T. O. *Abies homolepis* S. et Z.（Urajiro-momi）———Kôshunai, Bibai, Ⅸ - 7, 1974 byT. O. *Abies holophylla* MAX.（Chôsen-momi）———Kôshunai, Bibai, V - 27, 1975 by T. O. ; Shintoku, Tokachi, X - 15, 1976 by T. O. *Abies alba* MILL.（Yôroppa-momi）— Kôhunai. Bibai, Ⅳ - 20, 1978 by T. O. ; Yamabe, Furano, X - 5, 1978 by T. O.

Distribution : Asia（Japan）, North America, Europe, and Australia（New Zealand）

Note : KAMEI（1959）found a canker disease of todo fir（*Abies sachalinensis* MAST. ）and identifi-

ed the causal fungus as *Dasyscypha calyciformis* (WILLD.) REHM. In 1962, he accepted KORF's opinion and revised the scientific name of the causal fungus to *Trichoscyphella calycina* (SCHUM. ex FR.). NANNF. KORF wrote in his personal communication with KAMEI: "Perhaps the next available epithet for your fungus would then be *subtilissima* COOKE, as *Lachnellula subtilissima* (COOKE) comb. nov." But the fungus causing the canker disease of todo fir is apparently different from *L. subtilissima* in the size and the shape of the ascospores.

MAUBLANC (1904), DENNIS (1962), and ROBAC (HEPTING 1971) stated that the name *D. calycina* should not be used because of the confusion existing among *D. calycina* SCHUM., *D. calycina* FRIES, and *D. calycina* FUCHEL, and *D. calycina* SCHUM. ex FR. *Trichoscyphella calycina*, identified by KAMEI (1962) as the causal fungus of todo fir canker disease, is the same species that has hitherto been known as *D. calyciformis* (Table 2). DHARNE (1965) proposed a new combination *Lachnellula calyciformis* (WILLD ex FR.) DHARNE. The author accepts DHARNE's scientific name for the fungus causing the canker disease of todo fir.

Table 2. Dimension of the hitherto known of *Lachnellula calyciformis*
表−2 今までにしられている *Lachnellula calyciformis* の測定値

Scientific epithet 学名	Size of ascus 子のうの大きさ (μm)	Size of ascospore 子のう胞子の大きさ (μm)	Size of paraphysis 側糸の大きさ (μm)	Worker 著者
Helotium calyciforme (WILLD.) WETTSTEIN	50 - 68×4.5 - 5.5	5 - 11×1.5 - 3	1 - 1.5 wide	WETTSTEIN (1887)
Dasyscypha calyciformis (WILLD.) REHM	50 - 60×4.5 - 5	5 - 7×2.5 - 3	1 - 2 breit	REHM (1896)
Dasyscypha calyciformis (WILLD.)	40 - 50×4 - 4.5	6 - 8×2.5 - 3		MAUBLANC (1904)
Dasyscpha calyciformis WILLD.	50 - 60×4 - 7	5 - 8×2 - 3	50 - 60	SCHELLENBERG (1939)
Dasyscypha calyciformis (WILLD.) REHM	50 - 60×4.5 - 5	5 - 7.5×2.5 - 3		KOBAYASHI et al (1939)
Dasyscypha calyciformis (WILLD. ex FR.) REHM	36 - 58.5×4 - 6 (43 - 54×4.5 - 5.5)	4 - 7.5×1.5 - 3.5 (4.5 - 7×2 - 3)	45 - 77×1 - 3 (55 - 65×1.5 - 2.5)	BINGHAM & EHRLICH (1943)
Dasyscypha calyciformis (WILLD.) REHM	50 - 56×4 - 4.8	4.8 - 7.2×2.5 - 3.5		KUJALA (1950) —
Trichoscyphella calycina (SCHUM. ex FR.) NANNF.	40 - 50×3 - 7	5 - 7.5×2 - 3	50 - 60	KAMEI (1962)
Lachnellula calyciformis (WILLD. ex FR.) DHARNE	50 - 63×4 - 5.5	4 - 7.5×2.5 - 3.5	60 - 70×1.5	DHARNE (1965)
Lachnellula calyciformis (WILLD. ex FR.) DHARNE	40 - 57.5×3.7 - 6.2 (40 - 50×4.5 - 5)	4.5 - 7×2 - 3 (5 - 7×2 - 3)	52 - 72×2 - 3	Writer

c *Lachnellula ciliata* (HAHN) DENNIS Persoonia 2 : 183, 1962 ;
DHARNE, Phytop. Zeits. 53 : 120, 1965. (Fig. 3)

Synonym : *Dasyscypha ciliata* HAHN, Mycol. 32 : 141, 1940
Lachnella ciliata (HAHN) SEAVER, North Amer. Cup-fungi (Inoperc.) : 247, 1951.

Apothecia waxy, fleshy, short stalked, usually scattered, occasionally grouped, at first globular and closed, then opening as a flat discunder moist conditions, laterally compressed and closed when dry. Externally white, covered with excipular hairs. Hairs cylindrical, septate, minutely roughened, hyaline. Discs orange to peach, commonly 0.5 - 1 mm in diameter.

Asci clavate, with obtusely rounded apices, 60 - 82.5×7.5 - 12 μm, mostly 65 - 80×7.5 - 10 μm, 8 - spored. Ascospores obliquely uniseriate, ovoid to ellipsoid, continuous, hyaline, 5 - 13×4 - 6.5 μm, mostly 7 - 11 × 4 - 6 μm. Paraphyses out-ranking asci, filiform, septate, of equal diameter or slightly swollen at the tips, with minute oil globules, 85 - 145×2 - 3 μm.

Conidial stage was not observed innature but conidial masses were produced on malt extract agar. Conidia continuous, spherical to elliptical, hyaline, 2 - 4.5×2 - 3.5 μm, mostly 2.5 - 3.5×2.2 - 3 μm. Conidia germinated on malt extract agar.

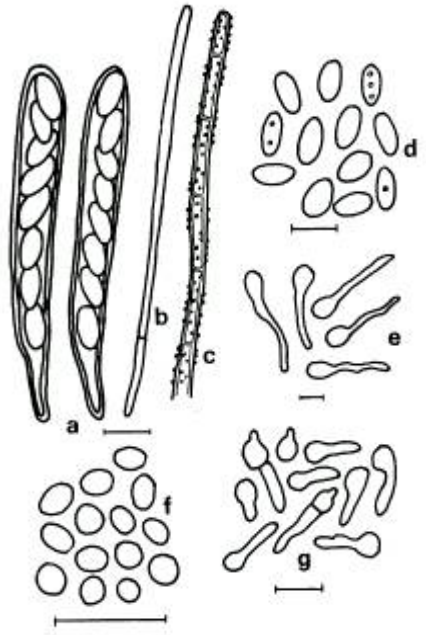


Fig.3. (図－3) *Lachnellula ciliata* (HAHN) DENNIS
a : Asci and ascospores 子のうと子のう胞子
b : Paraphysis 側糸 c : Hair 毛
d : Ascospores 子のう胞子 e : Germinating ascospores 子のう胞子の発芽 f : Conidia 分生子
g : Germinating conidia 分生子の発芽
(⊢——⊣ =10μm)

Host and Material : *Abies sachalinansis* MAST. (Todo-matsu) ——— Mikasa, Sorachi, VI - 11, 1974 by M. AKIMOTO ; Nayoro, X - 18, 1974 by T. O.

Distribution : Asia (Japan) and North America.

Note : This species, on Douglas fir (*Pseudotsuga taxifolia*) was first described by HAHN (1940) and he proposed it as *Dasyscypha ciliata* HAHN. SEAVER (1951) reported this species as *Lachnella ciliata* (HAHN) SEAVER in his book : 'The North America Cup-fungi (Inoperculates)' and his discription of the species is the same with HAHN's. HAHE (1940) stated that the species was restricted in the habitat to Douglas fir and its conidial stage was observed neither in nature nor on culture. The present fungus occurred on todo fir, and conidia were produced on malt extract agar. For these reasons, the species which has the same morphological characteristics with the present fungus was investigated in the literature, and the present fungus was compared with *Lachnellula agassizii* and *L. gallica* which were mentioned by HAHN (1940) and DHARNE (1965) as similar species to *L. ciliata.* The present fungus is distinguished from *L. agassizii* by the sizes of asci and ascospores. *ciliata* has larger sizes and it differs from *L. gallica* in having shorter asci and smaller ascospores (Table 3). It seems that the present fungus

is identical with *L. ciliata*. This is new to Japanese fungus flora.

Table 3. Comparision of morphology of *Lachnellula ciliata*
表－3 *Lachnellula ciliata* 近縁種の形態比較

	Size of ascus 子のうの大きさ（μm）	Size of ascospore 子のう胞子の大きさ（μm）	Paraphysis 側糸 Form 形	Paraphysis 側糸 Size 大きさ（μm）	Worker 著者
L. gallica	95 - 110×9 - 10	10 - 12×6 - 7	filiform		KARSTEN & HARIOT (1880)
	92 - 105×6.5 - 8.5	7 - 10×4.5 - 7	filiform	100 - 105×1.5 - 2.5	DHARNE（1965）
L. agassizii		6.5 - 7.5×4			SACCARDO（1889）
	43.5 - 67.5×4 - 6	5 - 9.5×2 - 4	spatulate	47.5 - 94.5×1 - 3.5	BINGHAM & EHRLICH（1934）
	(47 - 60×4.5 - 5.5)	(5.5 - 8.5×2.5 - 3.5)		(65 - 85×2 - 3)	
	60 - 95×3 - 4	6 - 10×3 - 4	clavate		SEAVER（1951）
	55 - 60×3 - 4.5	6 - 8×3 - 4.5	spatulate	60 - 70×1.5 - 3.5	DHARNE（1965）
L. ciliata	63 - 92.8×6 - 12	8 - 12.4×4 - 6.6	filiform		HAHN（1940）
	(70 - 80×7 - 10)				
	70 - 80×7 - 10	8 - 12×4 - 6	filiform		SEAVER（1951）
	60 - 82.5×7.5 - 12	5 - 13×4 - 6.5	filiform	85 - 145×2 - 3	Writer

d *Lachnellula fuckelii*（BRES. ap. REHM）DHARNE phytop. Zeits., 53 : 131, 1965.（Fig. 4）

Synonym : *Dasyscypha willkommii* HARTIG var. Fuckelii BRESAD., REHM, Rabenh. Krypt-Fl. Bd. I, Abt. Ⅱ, 833, 1896.

Apothecia scattered, often overlap one an - other, erumpent, short stalked, at first globu-lar and closed, then opening in a rounded form, sometimes irregular, external white to buff, covered with excipular hairs. Hairs cylindrial, septate, minutely roughened, hyaline. Discs orange to brownish orange, 0.8 - 1.2 mm in diameter.

Asci cylindrical, apex obtusely rounded, 87.5 - 125×8.5 - 12.5 μm, mostly 95 - 115×8.5 - 12 μm, 8-spored. Ascospores obliquely uniseriate, ellipsoidal, continuous, hyaline, 10 - 16.5×4.2 - 7 μm, mostly 10 - 15×5 - 7 μm. Paraphyses filiform, with round apex, m-

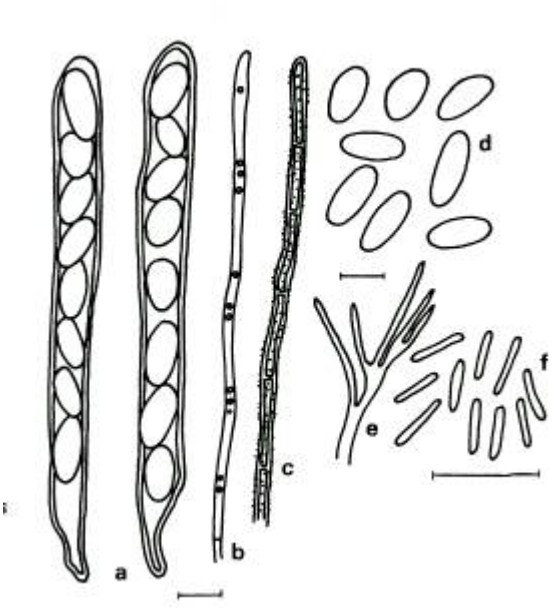


Fig.4.（図－4） *Lachnellula fuckelii*（BRES. ap. REHM）DHARNE

a : Asci and ascospores 子のうと子のう胞子
b : Paraphysis 側糸 c : Hair 毛
d : Ascospores 子のう胞子
e : Microconidiophore 小分生子柄
f : Microconidia 小分生子（├──┤＝10 μm）

inute oil globules, 100 - 137×2 - 3 μ m.

Conidial stage was not observed in nature or on culture. Microconidia were produced on malt extract agar and sterilized twigs of todo fir. Microconidia continuous, hyaline, elliptical-oblong or allantoid, 5 - 9×0.8 - 1.2 μ m, mostly 5 - 7×1 μ m. Microconidia did not germinate.

Host and Material ; *Abies sachalinensis* MAST. (Todo - matsu) Horonobe, Sôya, Ⅶ - 26, 1972 by T. O.

Distribution : Asia (Japan) and Europe.

Note : According to DHARNE (1965), at first, REHM described *Dasyscypha calycina* var. *minor*, which was collected by BRESADOLA on *Pinus* and the variety was named on the bases of the size of its asci and ascospores. Later REHM (1896) revised the variety to *D. willkommii* var. *fuckelii*, which was descrided by BRESADOLA as the synonym of *D. calycina* FUCKEL not SCHUM, and he pointed out the shorter size of asci and smaller size of asccspores thn those of *D. willkommii*. DHARNE (1965) proposed a new combination, *Lachnellula fuckelii*, and he erected the variety to species.

This species is exeeedingly rare in Hokkaido and the author found a scanty collection of this species occurring saprophytically on a dead stem of todo fir. This is new to Japanese fungus flora.

e *Lachnellula microspora* ELLIS & EVERHALT Proc. Sci. Phila. : 451, 1893 ; SEAVER, North Amer, Cup-fungi (Inoperc.) : 283, 1951 ; SAHO and TAKAHASHI, Rept. Tottori Mycol. Inst. (Japan) : 706, 1973. (Fig. 5)

Apothecia scattered, erumpent, very short stalked, at first closed, then opening in a cup-shaped, reaching a diameter of 1 - 2.5 mm, ex-ternally brownish red, covered with excipular hairs. Hairs cylindrical, septate, minutely roughened, rufous. Discs pale orange when fresh, pele brownish red when dry.

Asci clavate-cylindrical, 45 - 57.5× 4.5 - 6 μ m, mostly 50 - 55×5 - 5.5 μ m, 8 - spored. Asco-spores obliquely uniseriate, globose to subglobose, continuous, hyaline, 3 - 5×2.5 - 4 μ m, mostly 3 - 4×3 - 3.5 μ m. Paraphyses filiform , with rounded extremities, 60 - 80×1.2 - 2 μ m.

Conidial stage was not observed in nature but conidial masses are produced on malt ex-tract agar. Conidia continuous, globose to sub-globose, hyaline, 4 - 4.5× 1.2 - 4 μ m, mostly 4 - 4.5×1.5 - 2.5 μ m. Conidia germinate on malt ex-tract agar. Microconidia were produecd on malt extract agar and sterilized twigs of todo fir.

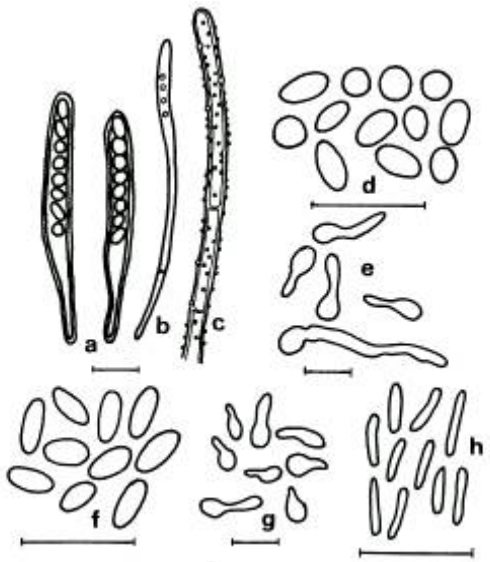


Fig.5. (図－5) *Lachnellula microspora* ELLIS and EVERHALT

a : Asci and ascospores 子のうと子のう胞子

b : Paraphysis 側糸 c : Hair 毛

d : Ascospores 子の胞子 e : Germinating ascospores 子のう胞子の発芽 f : Conidia 分生子 f : Germinating conidia 分生子の発芽

h : Microconidia 小分生子 (⊢⊣ =10 μ m)

Microconidia continuous, hyaline, elliptical oblong or allantoid, 4 - 7×0.8 - 1.5μm, mostly 4.5 - 6×1μm. Microconidia did not germinate.

Host and Material : *Abies sachalinensis* MAST. (Todo-matsu) ———— Yamabe, Furano, Ⅷ - 26, 1971 by I. TAKAHASHI ; Yamabe, Furano, Ⅶ - 24, 1973 by T. O. ; Yamabe, Furano, Ⅶ－30, 1974 by T. O.

Distribution : Asia (Japan) and North America

Note : This species was first described as inhabitant of spruce by ELLIS and EVERHALT (1893). SEAVER (1951) described this species as one of two species of *Lachnellula* KARST. and his description is identical with ELLIS and EVERHALT. SAHO and TAKAHASHI (1973) listed this species on todo fir and Japanese stone pine (*Pinus pumila* REGEL) as new to Japanese fungus flora in their list, but they did not describe the morphological characteristics. More recently, TAKAHASHI (1979) described this species. *L. microspora* is similar in brown appearance to *Lachnellula arida*, but the species has somewhat smaller ascospores than those of *L. arida*.

f *Lachnellula subtilissima* (COOKE) DENNIS

Persoonia 2 : 184, 1962 ; DHARNE, Phytop. Zeits. 53 : 121, 1965 ; DENNIS Brit. Ascom. : 155, 1968 ; SAHO and TAKAHASHI, Rept. Tottori Mycol. Inst. (Japan) 10 : 706, 1973. (Fig. 6)

Synonym : *Peziza subtilissima* COOKE, Grevillea 3 : 121, 1871.
Dasyscypha subtilissima (COOKE) SACC., Syll. Fung. 8 : 438, 1889 ; REHM, Rabenh. Krypt-Fl. Ⅲ : 833, 1896 ; HILEY, The fungal disease of the common larch 76, 1919 ; KUJALA, Commun. Inst. Fenn. 38 : 30, 1950. *Trichoscypha subtilissima* COOKE, BOUDIER, Discom. D' Europe : 125, 1907.

Apothecia solitary or grouped, erumpent, short stalked, at first closed, opening in a urn-shaped. Externally white or buff, covered with excipular hairs. Hairs cylindrical, septate, minutely roughened, hyaline, gently rounded extremities. Discs vivid yellow to orange, 0.8 - 2.5 mm in diameter.

Asci narrow cylindrical to cylindrical, apex rounded, 47.5 - 62.5×5 - 7μm. mostly 50 - 57.5×5 - 6.5μm, 8 - spored. Ascospores irregularly biseriate, fusiform clavate, continuous, hyaline, 5 - 9×1.5 - 3μm, mostly 6 - 8×2 - 2.5μm. Paraphyses filiform, overtopping the asci, with yellow oil globules, 52.5 - 75×2 - 2.5μm.

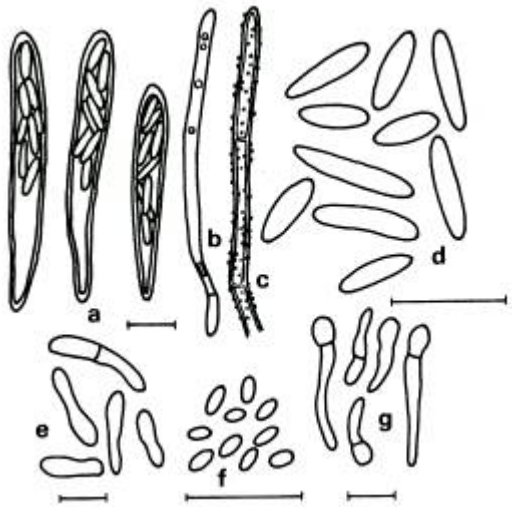


Fig.6. (図－6) *Lachnellula subtilissima* COOKE

a : Asci and ascospores 子のうと子のう胞子
b : Paraphysis 側糸 c : Hair 毛
d : Ascospores 子のう胞子
e : Germinating ascospores 子のう胞子の発芽
f : Conidia 分生子 g : Germinating conidia 分生子の発芽 (├──┤ =10μm)

Table 4. Host and dimension of the hitherto known of *Lachnellula subtilissima*
表－4 今までにしられている *Lachnellula subtilissima* の寄主と測定値

Scientific epiyhet 学名	Host 寄主	Size of ascus 子のうの大きさ （μm）	Size of ascospore 子のう胞子の大きさ （μm）	Worker 著者	
Peziza（*Dasyscypha*）*subtilissima* COOKE	Bark of firs		9μ long	COOKE	(1871)
Lachnella subtilissima（COOKE）PHILL.			10 - 12×2	PHILLIPS	(1887)
Dasyscypha subtilissima（COOKE）SACC.	Abietis et Pini silvestris		10 - 12×2	SACCARDO	(1889)
Dasyscypha subtilissima（COOKE）REHM	An Rinde von Nadelbaumen	50 - 65×5 - 7	10 - 12×2	REHM	(1896)
Dasyscypha subtilissima SACC.	Larch, silver fir and spruce？		8 - 10×2	HILEY	(1919)
Dasyscypha subtilissima（CKE）SACC.			6.5 - 10.5×2 - 3	JØRSTAD	(1925)
Dasyscypha subtilissima（CKE）SACC			5.2 - 9×2.5 - 4	KUJALA	(1950)
Lachnella subtilissima（COOKE）DENNIS	*Larix, Picea, Pinus, Abies*	45 - 60×4 - 5.5	7 - 12×1.5 - 2 - 5	DHARNE	(1965)
Lachnellula subtilissima（COOKE）DENNIS	*Pinus*		6 - 12×2 - 2.5	DENNIS	(1968)
Lachnellula subtilissima（COOKE）DENNIS	*Abies*	47.5 - 62.5×5 - 7	5 - 9×1.5 - 3	Writer	

Conidial stage was not observed in nature but conidial masses are produced on malt extract and potato dextrose agar. Conidia continuous, spherical to elliptical, hyaline, 2 - 3×1 - 1.5μm, mostly 2 - 2.5×1 - 1.5μm. Conidia germinate on malt extract agar.

Host and Material : *Pinus banksiana* LAM.（Bankus - matsu）——— Yamabe, Furano, IX - 27, 1976 by I. TAKAHASHI ; *Abies holophylla* MAX.（Chôsen - moni）——— Kôshunai, Bibai, V - 10, 1974 by T. O. ; Kôshunai, Bibai, XI - 7, 1974 by T. O.

Distribution : Asia (Japan), North America, and Europe.

Note : This species was first described as *Peziza subtilissima* by COOKE (1871). COOKE's morphological dimension was only within the length of ascospores, and thereafter many mycologists have reported the sizes of ascus and ascospore of this species (Table 4) SAHO and TAKAHASHI (1973) first listed this species on *Pinus banksiana*, *P. pumila*, and *P. strobus* in Japan, but they did not report the morphological characteristics. More recently TAKAHASHI (1979) described this species.

The ascospores, the author's material, are smaller than the others as in Table 4, but no species in the genus *Lachnellula* has the characteristic ascospores as *L. subtilissima*. This species was placed in *Lachnellula subtilissima* for this reason.

2　Physiological　studies

a　Growth of colony on various agar media

The mycelial growth on the above-mentioned four agar media is show in Fig. 7, and their cultural characteristics are given in Table 5. *L. calyciformis*, *L. fuckelii*, *L. microspora*, and *L. subtilissima* grew well on malt extract and potato dextrose agar, whereas *L. aeruginosa* and *L. ciliata* grew aest on Waksman's solution agar. Among six species, *L. calyciformis* grew the best on malt extract agar, and the diameter of its colony grew about twice as long as the diameter of *L. ciliata* or *L. microspora* on malt extract agar at the and of two months' incubation. Growth of mycelial colonies on Czapek's solution agar was generally poor or kone.

In this cultural experiment, buff, pinkish or milky white masses like a brop were observed on inocula of *L. ciliata*, *L. fuckelii*, and *L. subtilissima* and these masses were conidia or microconidia. *L. subtilissima* produced one apothecium on malt extract agar.

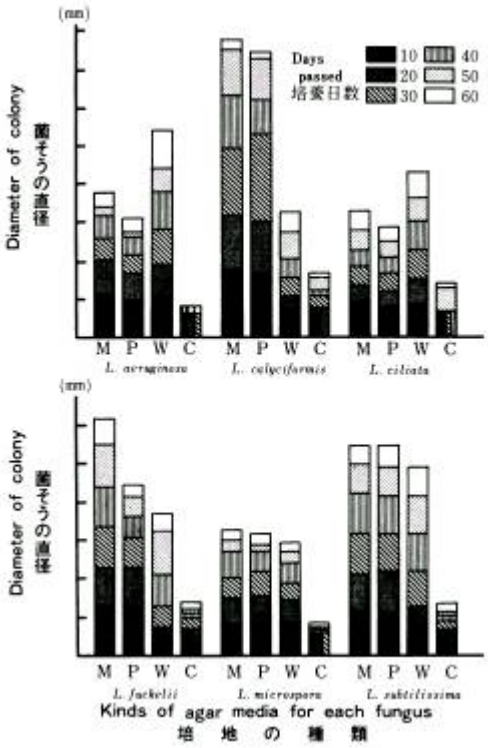


Fig.7.　Mycelial growth on various agar media

図－7　培地上における菌そうの発育

M：Malt extract agar　麦芽寒天

P：Potato dextrose agar　馬鈴薯寒天

W：Waksman's solution agar ワックスマン寒天

C：Czapek's solution agar ツァペック寒天

Table 5.　Cultural characteristics of *Lachnellula* spp.　on various agar media

表－5　*Lachnellula* 属菌の各種培地上の培養特徴

Medid 培地 / Species 種名	Malt extract 麦芽	Potato dextrose 馬鈴薯	Waksman ワックスマン	Czapek ツァペック
L. aeruginosa	c. dense, white, circular at margin ; a. m. long, entangied	c. dense, mat-like, rosy buff at center, white at margin ; a. m. partly long, entangled.	c. fan-sgape, white to pale ochreous, irregular at margin ; a. m. quite poor.	Growth of c. none.
L. calyciformis	c. vigorous, thin, furrow, white to buff, circular at margin ; a. m. partly dense, long, entangled ; white m. m. scattered on c.	c. vigorous, thin, concentric white, circular at margin ; a. m. partly dense, short ; pale saffron-yellow ; m. m. scattered on c.	c. fan-shape, dense, white to buff, irregular at margin ; a. m. quite sparse.	c. fan-shape, thin, white, irregular at margin ; a. m. none.
L. ciliata	c. dense, pale buff, irregular at margin ; a. m. very short, partly dense ; buff, dropy masses oozed out on inocula.	c. dense. pale buff, irregular at margin ; a. m. short, almost none.	c. vigorous, thin, pale ochreous, almost circular at margin ; a. m. none.	Growth of c. poor, rougth, circular or irregular at margin ; a. m. none.
L. fuckelii	c. thin, white, circular at margin ; a. m. long, entangled around inocula ; ochreous m. m. scattered on inocula.	c. decse, white, irregular at margin ; a. m. long, entangled ; milky white, dropy masses oozed out on inocula.	c. dense, pale ochreous, circular at margin ; a. m. quiet sparse.	c. rough, circular at margin ; a. m. none.
L. microspora	c. dense, white to buff, irregular at margin ; a. m. very rough, long, entangled.	c. dense, mat-like, white to pale buff, circular at margin ; a. m. short, entangled.	c. dense, mat-like, white to grey, greyish red at center, irregular at margin ; a. m. dense, short.	c. quite poor ; a. m. long, white only on inocula.
L. subtilissima	c. vigorous, dense, fan shape, white, irregular at margin ; a. m. short, dense ; buff, dropy masses oozed out on inocula ; a apothecium was produced on innocula.	c. vigorous, dense, mat-like, white, irregular at margin ; a. m. none ; pink, dropy masses oozed out on inocula.	c. vigorus, dense, buff, partly ochreous, almost circular at margin ; a. m. quite sparse.	c. dense, white, circular at margin ; a. m. none.

c. : colony　菌そう，a. m. : aerial hyphae　気中菌糸，m. m. : mycelial masses　菌糸塊

b Growth on sterilized twigs

It was observed that mycelia grew and fruit bodies produced on sterilized twigs of some tree species. The results are given in Table 6. In about 2 to 3 months after inoculation, five species except *L. fuckelii* produced pycnidial bodies on the sterilized twigs of *Abies sachalinensis*. At first, white mycelial masses formed on the bark. Then buff, pinkish or milky drops of conidia oozed from these pycnidial mnasses. *L. calyciformis* produced pycnidia on the sterilized *Larix leptolepis* and *Pinus strobus*. Apothecia were produced within 4 to 6 months of incubation. *L. fuckelii* produced neiter pycnidium nor apothecium.

Table 6. Production of fruit body of *Lachnellula* spp. on various sterilized twigs
表－6 各種殺菌枝上における *Lachnellula* 属菌の子実体形成

Species 種名	Tree species 樹種	No. of twigs tested 供試枝数	Production of Pycnidia 分生子殻形成	Production of Apothecia 子のう盤形成
L. aeruginosa	*Abies sachalinensis*	3	－ ～ ＋	－
L. calyciformis	*Abies sachalinensis*	3	－ ～ ＋＋	－ ～ ＋
	Larix leptolepis	3	－ ～ ＋	－ ～＋＋
	Pinus strobus	3	＋＋～＋＋＋	－ ～ ＋
L. ciliata	*Abies sachalinensis*	7	－ ～＋＋＋	－
	A. homolepis	3	－ ～ ＋＋	－ ～ ＋
	A. veitchii	2	－ ～＋＋＋	－ ～＋＋
L. fuckelii	*Abies sachalinensis*	6	－	－
	Larix leptolepis	3	－	－
L. microspora	*Abies sachalinensis*	3	－ ～ ＋	－
L. subtilissima	*Abies sachalinensis*	8	－ ～ ＋＋	－
	A. homolepis	4	－	－ ～ ＋
	A. veitchii	5	－	－ ～＋＋

－：none ない， ＋：sparse わずか， ＋＋：many 多い， ＋＋＋：abundant 豊富

Table 7. Production of conidia and microconidia of *Lachnellula* spp. on various substrata
表－7 *Lachnellula* 属菌の各種基質上における分生子と小分生子の形成

Species 種名	Conidia 分生子			Microconidia 小分生子	
	Malt extract agar 麦芽寒天	Potato dextrose agar 馬鈴薯寒天	Sterilized twig 殺菌枝	Malt extract gar 麦芽寒天	Sterilized twig 殺菌枝
L. aerugiosa	●		As ●	●	As ●
L. calyciformis	●		As ● Ll ●		
L. ciliata	●		As ● Av ● Ah ●		
L. fuckelii				●	As ●
L. microspora	●		As ●	●	As ●
L. subtilissima	●	●	As ●		

●：Produced 形成， As：*Abies sachalinensis* （トドマツ）， Av：*A. veitchii* （シラビソ），
Ah：*A. homolepis* （ウラジロモミ）， Ll：*Larix leptolepis* （カラマツ）

c　Production of conidia and microconidia on various substrata

Production of conidia and microconidia was observed on various agar media and sterilized twigs of some tree species. As shown in Table 7, five species, except *L. fuckelii*, produced conidia on malt extract agar and sterilized twigs of *Abies sachalinensis*. *L. calyciformis*, *L. ciliata*, and *L. subtilissima* did not produce microconidia on any agar medium and sterilized twig. On the other hand, *L. aeruginosa*, *L. fuckelii*, and *L. microspora* produced microconidia on malt extract agar and sterilized twigs of *Abies sachalinensis*.

d　Germination of conidia

The results of germination tests are given in Table 8, 9. Conidia of five isoletes hardly germinated within 24 hours after incubation, but they almost germinated within 48 to 72 hours, and their germ-tube grew to 50 - 80 μm. In the test, there is remarkable differences in germination percentage of conidia. It is considered to be influenced by the age of conidia.

e　Relation between temperatur and germination of conidia

Conidia of *L. calyciformis* and *L. ciliata* germinated at from 10 to 25℃ with optimum 15 - 20℃,

Table 8. Germination of conidia of *Lachnellula* spp. inoculated after 48 hours on malt extract agar.

表－8　接種 48 時間における *Lachnellula* 属菌の分生子の発芽（麦芽培地）

Species 種名	Germination percentage 発芽率 (%)	Maximum length of germ-tube 発芽管の長さ (μm)
L. aeruginosa	77.7	33.8
L. calyciformis	80.7	25.0
L. ciliata	84.3	20.2
L. microspora	26.3	20.0
L. subtilissima	0.3	12.5

Average of three slides ; 100 conidia were counted from each slide ; incudated at 15℃ under dark condition

スライド 3 枚の平均 ; 各スライドの分生子 100 個を測定 ; 培養温度 15℃暗黒

Table 9. Germination of conidia of *L. microspora* and *L. subtilissima* after 72 hours on malt extract agar.

表－9　接種 72 時間における *L. microspora* と *L. subtilissima* の分生子の発芽（麦芽培地）

Species 種名	Germination percentage 発芽率 (%)	Maximum length of germ-tube 発芽管の長さ (μm)
L. microspora	81.3	62.5
L. subtilissima	61.0	50.0

Average of three slides ; 100 conidia were counted from each slide ; incubated at 15℃ under dark condition

スライド 3 枚の平均 ; 各スライドの分生子 100 個を測定 ; 培養温度 15℃暗黒

Table 10. Relation between temperature and germination of conidia on malt extract agar

表－10　麦芽培地上における分生子の発芽と温度との関係

Temp. 温度 (℃)	*L. calyciformis*		*L. ciliata*	
	Germination percentage 発芽率 (%)	Maximum length of germ-tube 発芽管の長さ (μm)	Germination percentage 発芽率 (%)	Maximum length of germ-tube 発芽管の長さ (μm)
0	0	－	0	－
5	0	－	0	－
10	2.6	8.0	6.4	5.0
15	60.4	40.8	77.6	30.0
20	43.4	70.0	88.2	75.0
25	24.6	60.0	83.6	62.5
30	0	－	0	－

Average of five slides ; 100 conidia were counted from each slide ; inoculated after 72 hours.

スライド 5 枚の平均 ; 各スライドの分生子 100 個を測定 ; 接種 72 時間後

but could not germinate at 30℃ or below 10℃ (Table 10).

3 Pathogenic studies

Inoculation experiment was carried out to determine the pathogenicity of these fungi. The potted 5-year-old teees of *Abies sachalinensis* were prepared, and inoculations were made on 3～4-year-old or 2～3-year-old portions of stems or branches on December 22, 1972. The results were observed in July and November 1973. They are summarized in Table 11.

1) Inoculation with *L. calyciformis*

Three trees (C, D, and F) developed lesions, and five trees (A, B, D, E, and F) produced pycnidia, and seven trees with the exception of tree D were dead on July 4, 1973. On November 9, 1973, three trees (C, D, and F) stopped the development of lesion, and four trees (A, B, E, and F) produced apothecia, and tree D was died.

2) Inoculation with *L. fuckelii*

Tree B developed lesion in July, 1973, but its development had stopped until November 9, 1973. The stem of tree E was broken out by the pressure of snow during winter. None of trees produced pycnium and spothecium.

3) Inoculation with the other species

None of trees developed lesions and produced either pycnidium or apothecium.

4 Discussion

The sizes of ascus and ascospore of *L. fuckelii* are the largest among six species, and the sizes of ascus of three species, *L. aeruginosa*, *L. microspora*, and *L. subtilissima*, are quite similar (Fig. 8). The size of ascospore of *L. calyciformis* is also similar to the one of the first and the second species (Fig. 9). These species, however, can be distinguished easily in the colour of apothecium. *L. calyciformis* and *L. subtilissima* have similar colour apothecium but they are distinguished in the shape of ascospores.

The description of conidia and their germination on the genus *Lachnellula* are scarce in the literature. MAUBLANC (1904), SCHELLENBERG (1905), KAMEI (1962), and DHARNE (1965) described the imperfect stage of *L. calyciformis*. MAUBLANC (1904) and

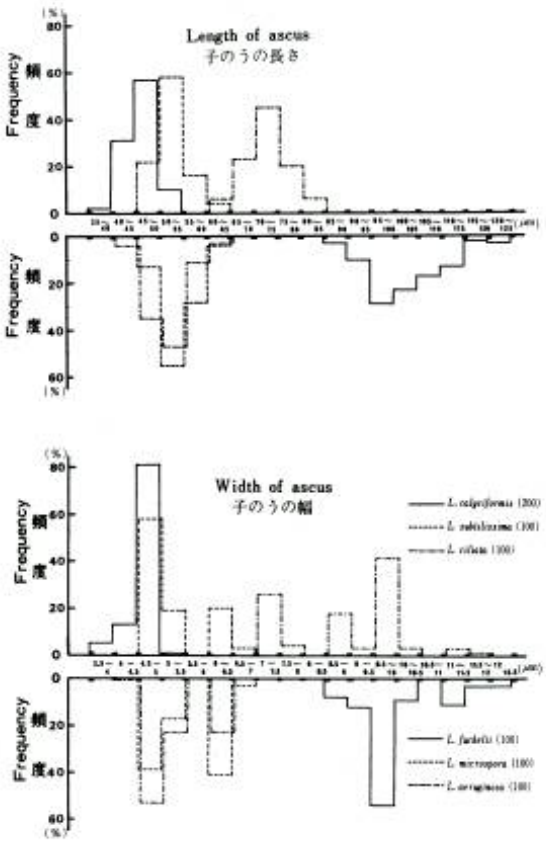


Fig.8. Histogram of length and width of ascus in 6 species of *Lachnellula* on *Abies* (Total account of measured asci)

図－8 モミ属上の *Lachnellula* 属菌 6 種の子のうの長さと幅の度数分布図（計測個数）

Table 11. Inoculation experiment with *Lachnellula* spp. on *Abies sachalinensis*
表－11 *Lachnellula* 属菌のトドマツへの接種試験

Species 種名	Health group 健康状態	Tree designations 名称	Development of lesions 病斑の進展	Production of fruit body a)		Condition of trees at the end of experiment 実験後の接種木の状態
				Pycnidia 分生子殻の形成	Apothecia 子のう盤の形成	
L. aeruginosa	Healthy	A	−	−	−	healthy
		B	−	−	−	healthy
		C	−	−	−	healthy
	Unhealthy	D	−	−	−	dead
		E	−	−	−	dead
		F	−	−	−	dead
L. calyciformis	Healthy	A	−	+	+	dead
		B	−	+	+	dead
		C	+	−	−	dead
		D	+	+	−	dead
		E	−	+	+	dead
	Unhealthy	F	+	+	+	dead
		G	−	−	−	dead
		H	−	−	−	dead
L. ciliata	Healthy	A	−	−	−	healthy
		B	−	−	−	healthy
		C	−	−	−	healthy
	Unhealthy	D	−	−	−	dead
		E	−	−	−	dead
		F	−	−	−	dead
L. fuckelii	Healthy	A	−	−	−	healthy
		B	−	−	−	healthy
		C	−	−	−	healthy
		D	−	−	−	healthy
		E	−	−	−	dead
	Unhealthy	F	−	−	−	dead
		G	−	−	−	dead
		H	−	−	−	dead
L. microspora	Healthy	A	−	−	−	healthy
		B	−	−	−	healthy
		C	−	−	−	healthy
	Unhealthy	D	−	−	−	dead
		E	−	−	−	dead
		F	−	−	−	dead
L. subtilissima	Healthy	A	−	−	−	healthy
		B	−	−	−	healthy
		C	−	−	−	healthy
	Unhealthy	D	−	−	−	dead
		E	−	−	−	healthy
		F	−	−	−	dead
Check	Healthy	A	−	−	−	healthy
		B	−	−	−	healthy
		C	−	−	−	healthy
	Unhealthy	D	−	−	−	dead
		E	−	−	−	dead
		F	−	−	−	dead

a) －: none ない, ＋: sparse わずか

KAMEI（1962）reported spermatia or microconidia. DHARNE（1965）described conidia and microconidia, but he did not mention about germination of conidia. Therefore, it is not evident whether he distinguished conidia and microconidia. SCHELLENBERG（1905）stated that "Die kleinen Conidien keimen sehr schwer. Nur in einigen wenigen Fällen habe ich die Keimung beobachten." And he illustrated the germinating conidia. In the present experiment, five species except *L. fuckelii* produced abundant conidia on some substrata, and these fresh cohidia germinated well on lnalt extract agar (Fig. 1, 2, 3, 5, and 6).

The physiological characters of culture isolated from conidia have close agreement with those obtained from ascospores. There can be no doubt as to the genetic connection between the two forms. According to YOKOTA and MATSUZAKI（1971）, ascospores of *Trichoscyphella calycina*（*L. calyciformis*) could not germinate at below 10℃ and above 30℃, and as shown in Table 10, conidia of *L. calyciformis* also did notgerminate at these temperatures in incubation after 72 hours.

As the results of the present inoculation experiment, *L. calyciformis* is the only parasitic species among the six . of *Lachnellula* on *Abies sachalinensis*. The relatively-young plantations of todo fir, which are mostly less than 10 years after afforestation, are frequently attacked by this fungus, and destractive damage has been encountered in Hokkaido. According to BROWNE（1968）and HEPTING（1971）, *L. calyciformis* usually inhabit saprophytically on dead or dying twigs of various conifers, but it often becomes actively parasitic on young

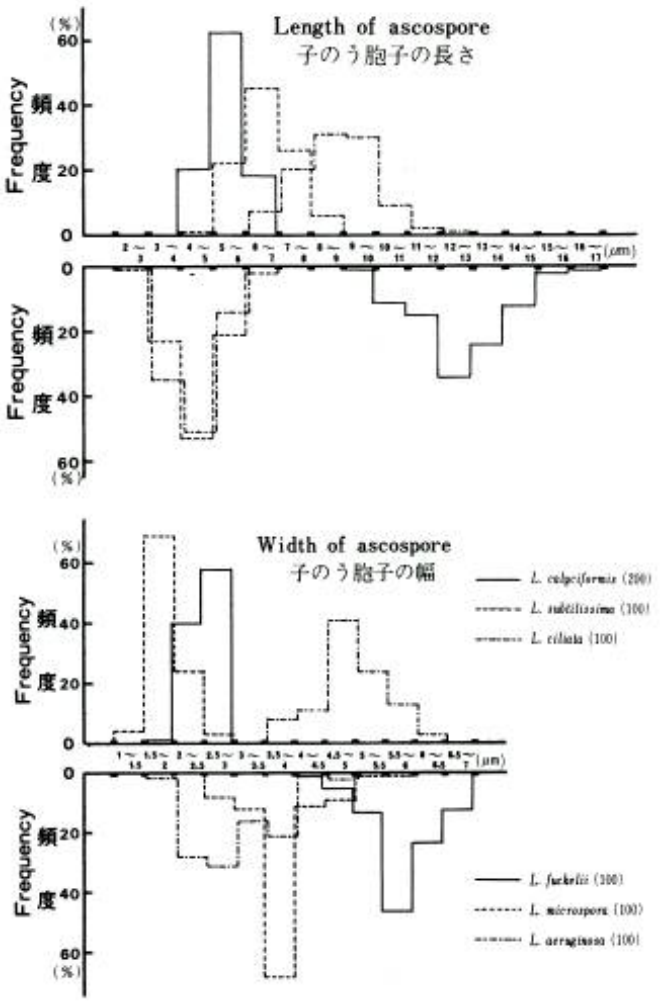


Fig.9. Histogram of length and width of ascospore in 6 species of *Lachnellula* on *Abies* (Total account of measured ascospores)

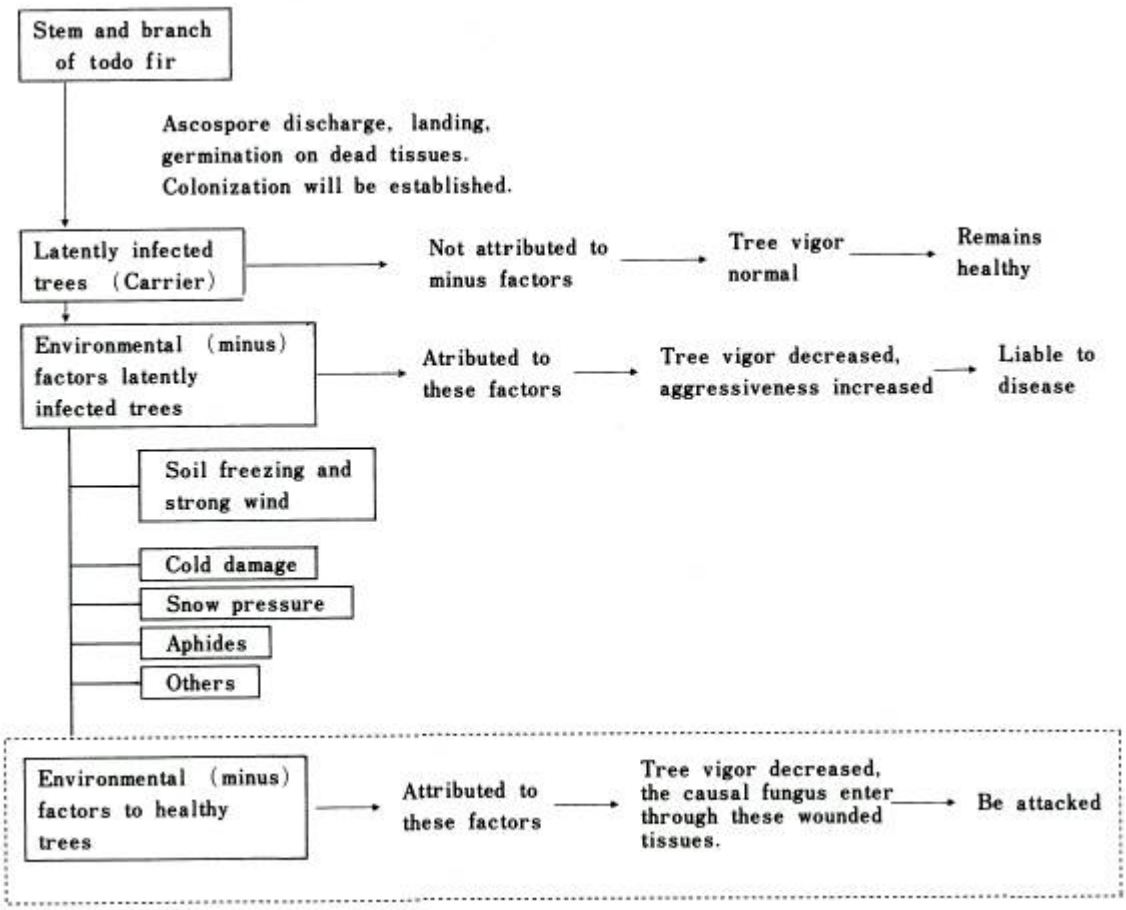
図－9 モミ属上の *Lachnellula* 属菌6種の子のう胞子の長さと幅の度数分布図（計測個数）

twigs and stems of some tree species under unfavorable conditions for the host plants. YOG and ONO (1961) reported the relation between the affections of aphides, bark beetles, or *Phomopsis* parasitisms and the outbreak of the canker disease of todo fir. They stated that 13 per cent of the infected trees were parasitized by aphides, and the aphid parasitism was more important than the other agents. KAMEI (1962) reported that cold injury among the influences of environmental factors acts as a predisposing factor of the disease.

YOKOTA and MATSUZAKI (1971) dealt with the mechanism of the disease development on todo fir canker disease and demonstrated that the pathogen is always present on macroscopically healthy parts of todo fir trees, and when necrotic tissue is produced by predisposing factors, for instance, frost, snow pressure, and aphid affection, the pathogen grows on the necrotic tissue and forms a canker. And they showed the diagrammatic scheme of the mechanism of the disease development (Table 12). But from the result of the present inoculation experiment, it is considered that a direct mechanism, as shown in the part surrounded with a dotted line in Table 12, should be included in their scheme.

Table 12. Diagrammatic scheme of the mechanism of development in todo fir canker

表－12 トドマツががんしゅ病発病機構図



B *Lachnellula* spp. on *Larix*

1. Morphological studies

a *Lachnellula arida* (PHILL.) DENNIS

Persoonia 2 : 183, 1962 ; DHARNE, Phytop. Zeits. 53 : 136, 1965 ; SAHO and TAKAHASHI, Rept. Tottori Mycol. Inst. (Japan) 10 : 706, 1973. (Fig. 10)

Synonym : *Peziza arida* PHILL., Grevilles 5 : 117, 1877.
Dasyscypha arida (PHILL.) SACC., Syll. Fung. 8 : 455, 1889.
Lachnella arida (PHILL.) SEAVER, North Amer. Cup-fungi (Inoperc.) : 268, 1951.

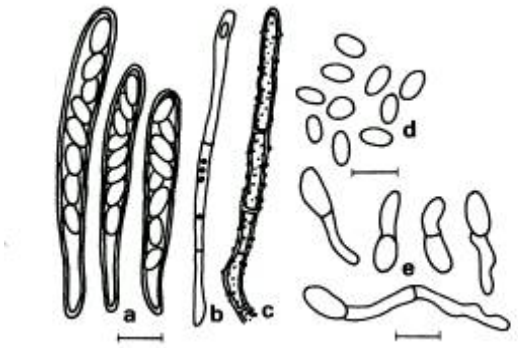


Fig.10. (図－10) *Lachnellula arida* (PHILL.) DENNIS
a : Asci and ascospores 子のうと子のう胞子
b : Paraphysis 側糸 c : Hair 毛 d : Ascospores 子のう胞子 e : Germinating ascospores 子のう胞子の発芽 (├──┤ =10μm)

Apothecia scattered or grouped, erumpent, stalked, at first globular, closed, opening as a flat saucer-like under moist conditions, closed with enrolled margins when dry. Externally brown, covered with excipular hairs. Hairs cylindrical, septate, minutely roughened, brown, gently rounded extremities. Discs yellow to orange yellow, 0.6 - 1.8 mm in diameter.

Asci clavate, apex obtusely rounded, 45 - 61×5 - 8.5μm, mostly 50 - 57.5×5 - 7.5μm, 8-spored. Ascospores obliquely uniseriate or partly biseriate, ellipsoid, continuous, hyaline, 5 - 10×2.5 - 6μm, mostly 5.5 - 7×3 - 5μm. Paraphyses outranking asci, filiorm, septate, slightly swollen at the tips, with minutely oil globules 70 - 85×2.5μm.

Conidial stage was not observed in nature and on culture.

Host and Material : *Larix gmelinii* GORDON (Gui-matsu) ──── Yamabe, Furano, VI－19, 1971 by I. TAKAHASI.

Distribution : Asia (Japan), North America, and Europe.

Note : SAHO nd TAKAHASHI (1973) first listed this species in Japan, but they did not report the morphological characteristics. More recently, TAKAHASHI (1979) descrided this species.

b *Lachnellula hahniana* (SEAVER) DENNIS

Persoonia 2 : 184, 1962. (Fig. 11)

Synonym : *Dasyscypha calycina* FUCKEL, Symb. Mycol. : 305, 1869 ; HAHN and AYERS, Mycol. 26 : 77, 1934 KUJALA, Commun. Inst. For. Fenn. 38 : 24, 1950.
Lachnella Hahniana SEAVER, Horth Amer. Cup-fungi (Inoperc.) : 245, 1951.
Trichoscyphella hahniana (SEAVER) MANNERS, Trans. Brit. Mycol. Soc. 36 : 364,

1953 ; DENNIS, Brit. Cup Fungi : 97, 1960.

Apothecia scattered or grouped, erumpent, short stalked, at first globular, closed, opening in a round form, expanding under moist conditions to more or less flat disc. Externally white, covered with excipular hairs. Hairs hyaline, cylindrical, septate, thinwalled, minutely roughened, with rounded or slightly swollen extremities Discs ochraceous to salmon-orange, 1 - 3 mm in diameter.

Asci clavate, frequently swollen toward the apex with obtusely rounded apices, 125 - 150× 10 - 15 μ m, mostly 130 - 145×11 - 14 μ m, 8-spored. Ascospores obliquely uniseriate, elongate-elliptic or elliptic-oblong , occasion-nally fusiform or pointed at one end, continu-uous, hyaline, 17.5 - 27×6 - 8.5 μ m, mostly17.5 - 23.5 × 6 - 7.5 μ m. Paraphyses exceeding the asci, filiform, septate, commonly intermixed with irregularly submonihform paraphyses , with swelling near their extremities, obtusely rouded or subacute apices, occasionally spathulate , with minute oil globules.

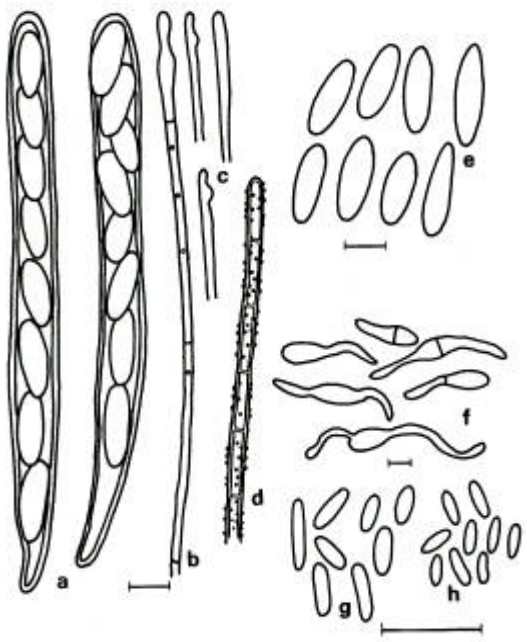


Fig.11. (図－11) *Lachnellula hahniana* (SEAVER) DENNIS

a : Asci and ascospores 子のうと子のう胞子
b : Paraphysis 側糸 c : Shape of paraphyses 側糸の形 d : Hair 毛 e : Ascospores 子のう胞子
f : Germinating ascospores 子のう胞子の発芽
g : Microconidia on malt extract agar 麦芽寒天培地上の小分生子 h : Microconidia on sterilized twigs of *Larix leptolepis* カラマツ殺菌枝上の小分生子
(⊢⊣ =10 μ m)

Conidial stage was not observed in nature and on culture but microconidial masses were produced on malt extract agar and sterilized twigs of *Larix leptolepis*. Microconidia continuous, hyaline, elliptic-oblong or allantoid, 3 - 6.5×1 - 2.2 μ m, mostly 4 - 5×1－2 μ m. Microconidia did not germinate.

Host and Material ; *Larix olgensis* var. *koreana* NAKAI (Chôsen-karamatsu) ――― Yamabe, Furano, Ⅵ - 9, 1971 by I. TAKAHASHI. *L. gmelinii* GORDON (Gui-matsu) ――― Yamabe, Furano, Ⅶ - 24, 1973 by T. O. ; Yamabe, Furano, X - 6, 1978 by T. O.

Distribution : Asia (Japan), North America, and Europe.

Note : HAHN and AYERS (1934) described four large-spored and white-excipled species of *Dasyscypha*, and they stated that *Dasyscypha calycina* FUCKEL has submoniliform paraphyses and that it apparently differs from *D. willkommii* in morphology and pathogenicity. At the same time they described the two species, *D. occidentalis* and *L. oblongospora*. SEAVER (1951) referred *D. calycina* FUCKEL to *Lachnella hahniana* SEAVER MANNERS (1953) rejected SEAVER's generic name and revised it to *Trichoscyphella hahniana* (SEAVER) MANNERS. In 1962, DENNIS proposed a new combination as *Lachnellula hahniana* (SEAVER) DENNIS.

c *Lachnellula occidentalis* (HAHN et AYERS) OGUCHI comb. nov. (Fig. 12).

Synonym : *Dasyscypha occidentalis* HAHN and AYERS, Mycol. 26 : 90, 1934.

Lachnella occidentalis (HAHN & AYERS) SEAVER, North Amer. Cup-fungi (Inoperc.) : 244, 1951.

Apothecia scattered or grouped, erumpent, stalked, at first globular, closed, then opening in a roundish form, margin inclosed, urn-like, becoming widly expanded, saucerlike under moist conditions, laterally compressed. Externally white, covered with excipular hairs. Hairs flexuous, cylindrical, septate, minutely roughened, hyaline, thin-walled, gently rounded extremities. Discs orange-buff to salmon-orange, 1 - 3 mm in diameter.

Asci clavate, apex obtusely rounded, 85 - 120×7.5 - 12.5 μm, mostly 87.5 - 105×10 - 12.5 μm, 8-spored. Ascospores obliquely uniseriate, ellipsoid, continuous, hyaline, 12 - 20×4 - 7.5 μm, mostly 13 - 16×5 - 7 μm. Paraphyses exceeding asci, flexuous, filiform or spathulate, septate, with minute oil globules, 100 - 140×1 - 2.5 μm.

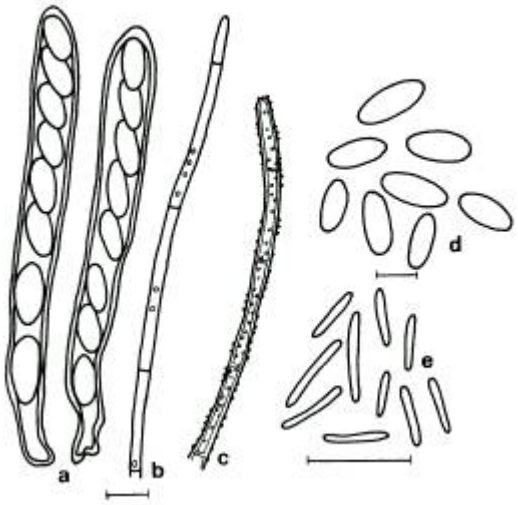


Fig.12. (図－12) *Lachnellula occidentalis* (HAHN et AYERS) OGUCHI comb. nov.
a : Asci and ascospores 子のうと子のう胞子
b : Paraphysis 側糸 c : Hair 毛 d : Asco-spres 子のう胞子 e : Microconidia 小分生子
(├──┤ =10 μm)

Conidial stage was not observed in nature and oh culture but microconidial masses were produced on malt extract agar and sterilized twigs of *Larix leptolepis*. Microconidia continuous, hyaline, cylindrical or allantoid, 5 - 10×0.8 - 1.2 μm, mostly 6 - 8×1 μm. Microconidia did not germinate.

Host and Material : *Larix leptolepis* GORDON (Kara-matsu) ——— Shintoku, Tokachi, Ⅶ - 29, 1972 by T O.

Distribution : Asia (Japan) and North America.

Note : *Daysyscypha occidentalis* was first described by HAHN and AYERS (1934). Then DHARNE (1965) combined *D. occidentalis* and *Lachnellula hahniana*, because *D. occidentalis* has submoniliform paraphyses and all other characters of *D. occidentalis* closely rssemble to *L. hahniana*. He proposed these combined one as *Lachnellula occidentalis* (HAHN and AYERS) DHARNE. But the present materials had on submoniliform paraphyses and so *D. ocicdentaliss* and *L. hahniansa* were treated as different species in this paper. New combination of the present fungus is proposed as *Lachnellula occidentalis* (HAHN et AYERS) OGUCHI.

This species is new to Japanese fungus flora and is rare species in Hokkaido. The present materials were collected from fallen branches of *Larix leptolepis* at Shintoku near obihiro City.

d *Lachnellula suecina* (de BY. ex FUCKEL) NANNF.

DHARNE, Phytop. Zeits. 53 : 118, 1965 ; SAHO and TAKAHASHI, Rept. Tottori Mycol Inst. (Japan) 10 : 706, 1973. (Fig. 13)

Synonym : *Helotium chrysophthalmum* (PESS.) KARST., Mycol. Fennica 1 : 155, 1871. *Lachnellula chrysophthalma* (PERS.) KARST., NANNFELDT. Nova Acta Reg. Soc. Sci. Upsal. Ser. IV. 8 : 300, 1932 ; SEAVER. North Amer. Cupfungi (Inoperc.) : 283, 1951. *Trichoscypha chrsophthalma* (KARST.) BOUD ., Discom D' Europe : 125, 1907.

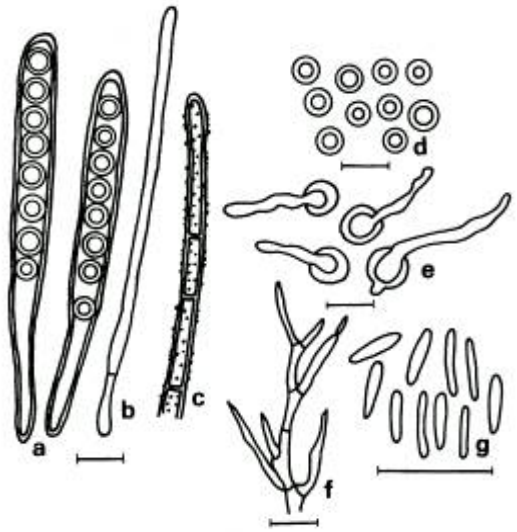


Fig.13. (図－13) *Lachnellula suecica* (de BY. ex FUCKEL) NANNFELDT

a : Asci and ascospores 子のうと子のう胞子
b : Paraphysis 側糸 c : Hair 毛 d : Ascospores 子のう胞子 e : Germinating ascospores 子のう胞子の発芽 f : Microconidiophore 小分生子柄
g : Microconidia 小分生子 (├──┤ =10μm)

Apothecia scattered or grouped, erumpent, short-stalked, at first globular, expanding under moist conditions and becoming flat saucer-like. Externally white, covered with excipular hairs. Hairs septate, hyaline, cylindrical, minutely roughened. Discs orange-red, 1 - 3 mm, under moist comditions up to 5 - 6 mm in diameter.

Asci cylindrical, with gently rounded apices, 58 - 90×5 - 7.5μm, mostly 70 - 80×7.5μm, 8-spored. Ascospores regularly uniseriate, globose, continuous, hyaline, 5 - 7.5μm in diameter. Paraphyses filiform, septate, 65 - 96×2.5 - 3.6μm.

Conidial stage was not observed in nature and on culture but microconidial masses were produced on malt extract agar and sterilized twigs of *Larix leptolepis*. Microconidia continuous, hyaline, cylindrical to oblong-elliptjcal, 4 - 7×0.8 - 1.5μm, mostly 4.5 - 6×0.8 - 1.2μm. Microconidia did not germinate.

Host and Material : *Larix olgensis* var. *koreana* NAKAI (Chôsen-karamatsu) ──── Yamabe, Furano, Ⅶ - 27, 1971 by I. TAKAHASHI. *L. gmelinii* GORDON (Gui-matau) ──── Yamabe, Furano, Ⅶ - 24, 1973 by T. O. ; Yamabe, Furano, Ⅶ－30, 1974 by T. O. *L. gmelinii* ×*L. leptolepis* ──── Yamabe, Furano, Ⅶ - 30, 1974 by T. O.

Distribution : Asia (Japan), North America, and Europe.

Note : This species is the type species of *Lachnellula* KARST. and the genus *Lachnellula* was distinguished from the genus *Trichoscyphella* NANNF. by its globose ascospores until DENNIS (1962) united these two genera.

e *Lachnellula calyciformis* (WILLD. ex FR.) DHARNE (Fig. 2)

Morphology of this species habiting *Larix* is the same as the characters that it was described on *Abies*.

Host and Material : *Larix gmelinii* GORDON (Gui-matsu) ──── Yamabe, Furano, Ⅶ - 24, 1973 by T. O.

Distribution : Asia (Japan), North America, and Europe.

2 Physiological studies

a Growth of colony on various agar media

The mycelial growth on the above-mentioned four agar media is shown in Fig. 7 and 14, and their cultural characteristics are given in Table 5 and 13, The present five species grew best on malt extract agar. *L. occidentalis* grew slowly on any agar media. *L. hahniana* did not grow on Czapek's solution agar. Three species except *L. arida* and *L. calyciformis* produced microconidia on malt extract agar. *L. arida*, *L. calyciformis*, and *L. hahniana* produced apothecio on malt extract agar in several stock cultures.

b Growth on sterilized twigs

It was observed that mycelia grew and fruit bodies produced on sterilized twigs of *Larix leptolepis*. The results are given in Table 14. About 2 to 3 months after inoculation, the bark of aterilized twigs was covered with white, dense or thin mycelia, and 3 to 4 months after inoculation, white mycelial masses were produced on the mycelial mats. Then pale yellow or orange-

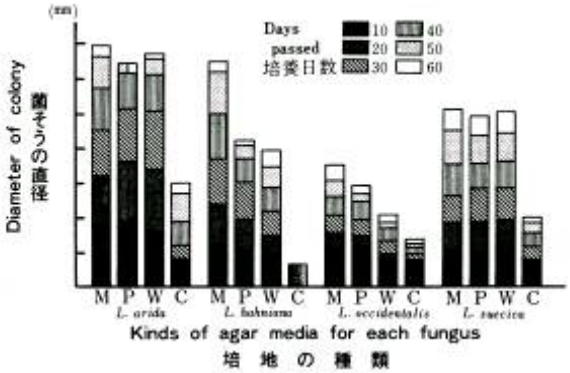


Fig.14. Mycelial growth on various agr media

図－14 培地上における菌そうの発育

M : Malt extract agar 麦芽寒天 P : Potato dextrose agar 馬鈴薯寒天 W : Waksman's solution agar ワックスマン寒天 C : Czapek's solution agar ツァペック寒天

Table 13. Cultural characteristics of *Lachnellula* spp. on various agar media

表－13 *Lachnellula* 属菌の各種倍地上の培養特徴

Media 培地 / Species 種名	Malt extract 麦芽	Potato dextrose 馬鈴薯	Waksman ワックスマン	Czapek ツァペック
L. arida	c. dense, mat-like, white, circular at margin ; a. m. long, entangled at center ; white m. m. scattered on c.	c. dense, mat-like, white, circular at margin ; a. m. long, entangled, short at center,	c. dense, concentric, white, wavy at margin ; a. m. long.	c. white at margin, orange at center, irregular at margin ; a. m. long, dense, entangled; black sclerotium-like body being around inoculum.
L. hahniana	c. concentric, white, dense, at center, rough at margin ; a. m. short, entangled.	c. concentric, mat-like, white to reddish orange, circular at margin ; a. m. short	c. dense, mat-like, rugous, white, at center, buff at margin ; a. m. very short.	c. quite poor.
L. occidentalis	c. dense, white, circular at margin ; a. m.short, rough.	c. unven, white to buff, irregular at margin ; a. m. short, dense.	c. uneven, white, irregular at margin ; a. m. very short, dense.	c. very thin, irregular, white, pinkish at center ; a. m. none.
L. suecica	c. thin, concentric, greyish red, white at nargin ; a. m. short ; buff m. m. scattered on c.	c. dense, mat-like, white, partly pale greyish red, irregular at margin ; a. m. almost none.	c. dense, mat-like, white, pale greenish yellow at center, circular at margin ; a. m. almost none.	c. poor, thin, white, pale greenish yellow at center ; a. m. none.

c. : colony 菌そう, a. m. : aerial hyphae 気中菌糸, m. m. : mycelial masses 菌糸塊

yellow to brownish orange microconidial masses oozed from mycelial masses with exception of *L. arida* and *L. calyciformis*. *L. calyiformis* produced pycnidia and apothecia , but the others produced neither pycnidium nor apothecium within 6 to months of incubation.

Table 14. Production of fruit body of *Lachnellula* spp. on sterilized twigs of *Larix leptolepis*

表－14 カラマツ殺菌枝上における *Lachnellula* 属菌の子実体形成

Species 種名	No. of twigs tested 供試枝数	Production of	
		Pycnidia 分生子殻形成	Apothecia 子のう盤形成
L. arida	3	－	－
L. calyciformis	3	－～＋	－～＋＋
L. hahniana	5	－	－
L. occidentalis	10	－	－
L. suecica	3	－	－

－：none ない，＋：sparse わずか，＋＋：many 多い

3 Pathogenic studies

Inoculation experiment was carried out to determine the pathogenicity of these fnngi. The potted 3-year-old trees of *Larix leptolepis* were prepared, and inoculations were made on 2～3-year-old portions of branches on November 6, 1973. The results were observed on July 1, 1974. The results are summarized in Table 15.

1) Inoculation with *L. arida*

None of trees developed lesions, and three trees (A, B, and D) exuded resin from inoculated portions. None of trees produced pycnidia, but three trees (A, C, and E) produced apothecia on the dead bark around inoculated portions.

2) Inoculation with *L. hahniana*

None of trees developed lesions, and produced pycnidium. All trees produeed apothecia on the dead bark around inoculated portions.

3) Inoculation with the other species

None of trees developed lesions and produced either pycnidium or apothecium.

4 Discussion

Lachnellula arida and *L. suecica* have small ascospores, but these two species are easily distinguished each other, because *L. arida* has brown excipled apothecia and *L. suecica* has white excipled and globose ascospores. *L. hahniana* and *L. occidentalis* have large ascospores, but L. hahniana has larger asci and ascospores than those of *L. occidentalis* (Fig. 15, 16).

According to BOYCE (1961) and HEPTING (1971), *L. arida* is a saprophytic fungus on conifers in North America. SAHO and TAKAHASHI (1972) reported *L. suecica* as the causal fungus of larch canker disease. But from the results of the present inoculation experiment, it is considered that *L. suecica* usually lives saprophytically on dead twigs of various conifers. HAHN and AYERS (1934) carried out the inoculation experiment with *Dasyscypha calycina* FUCKEL (*Lachnellula hahniana*) on living larch and Douglas fir, and they stated as the followings : "Artificial inoculation with *D. calycina* on living larch and Douglas fir (blue from) did not succeed. The fungus was able, however, to produce fruiting bodies

on the same form of Douglas fir which was dying at the time of inoculation, thereby showing its saprophytic nature." MANNERS (1953) carried out the inoculation experiment with *Trichoscyphella hahniana* (*Lachnellula hahniana*) on European larch, and this experiment did not succeed, too, and he observed young apothecia occurring on the dead bark around lesions.

In the present inoculation experiment, none of the trees drees developed lesions, and artificial inoculation with five species of *Lachnellula* failed on living larches. However the trees inoculated with *L. arida* and *L. hahniana* produced a few apothecia on the dead bark around the inoculated parts.

Table 15. Inoculation experiment with *Lachnellula* spp. on *Lrix leptolepis*
表－15 *Lachnellula* 属菌のカラマツへの接種試験

Species 種名	Health group 健康状態	Tree designations 名称	Development of lesious 病斑の進展	Production of fruit body a)		Condition of trees at the end of experiment 実験後の接種木の状態
				Pycnidia 分生子殻の形成	Apothecia 子のう盤の形成	
L. arida	Healthy	A	−	−	+	healthy
		B	−	−	−	healthy
		C	−	−	+	healthy
	Unhealthy	D	−	−	−	healthy
		E	−	−	+	healthy
		F	−	−	−	dead
L. calyciformis	Healthy	A	−	−	−	healthy
		B	−	−	−	healthy
		C	−	−	−	healthy
	Unhealthy	D	−	−	−	healthy
		E	−	−	−	dead
		F	−	−	−	healthy
L. hahniana	Healthy	A	−	−	+	healthy
		B	−	−	+	healthy
		C	−	−	+	healthy
	Unhealthy	D	−	−	+	healthy
		E	−	−	+	healthy
		F	−	−	+	healthy
L. occidentalis	Healthy	A	−	−	−	healthy
		B	−	−	−	dead
		C	−	−	−	healthy
	Unhealthy	D	−	−	−	healthy
		E	−	−	−	dead
		F	−	−	−	healthy
L. suecica	Healthy	A	−	−	−	healthy
		B	−	−	−	healthy
		C	−	−	−	healthy
	Unhealthy	D	−	−	−	dead
		E	−	−	−	healthy
		F	−	−	−	healthy
Check	Healthy	A	−	−	−	healthy
		B	−	−	−	healthy
		C	−	−	−	healthy
	Unhealthy	D	−	−	−	healthy
		E	−	−	−	healthy
		F	−	−	−	healthy

a) －：none ない, ＋：sparse わずか

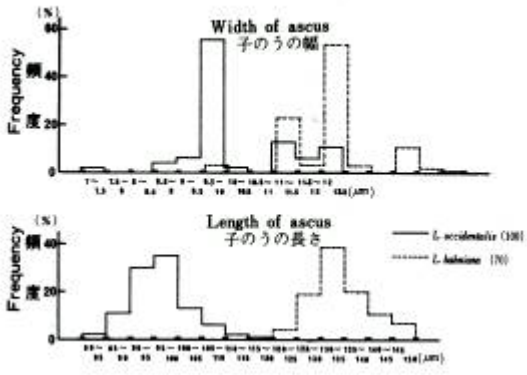


Fig.15. Histogram of length and width of ascus in 2 species of *Lachnellula* on *Larix* (Total account of measured asci)

図－15 カラマツ属上の *Lachnellula* 属菌 2 種の子のうの長さと幅の度数分布図（計測個数）

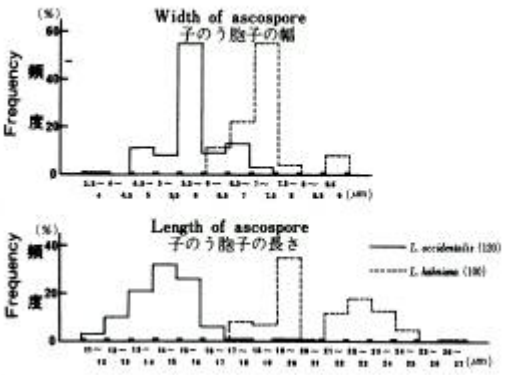


Fig.16. Histogram of length and width of ascospore in 2 species of *Lachnellula* on *Larix* (Total account of measured ascospores)

図－16 カラマツ属上の *Lachnellula* 属菌 2 種の子のう胞子の長さと幅の度数分布図（計測個数）

C *Lachnellula* spp. on *Pinus*

1 Morphological studies

a *Lachnellula abietis* (KARST.) DENNIS Persoonia 2 : 183, 1962. (Fig. 17).

Synonym : *Helotium abietis* KARST., Mycol. Fennica 1 : 154, 1871.

Dasyscypha abietis (KARST.) SACC., Syll. Fung. Ⅷ : 438, 1889 ; KUJALA. Commun. Inst. For. Fenn, 38 : 22, 1950 ; DHARNE, Phytop. Zeits. 53 : 139, 1965.

Trichoscpha abietis BOUD., Discom. D'Europe : 125, 1907.

Trichoscyphella abietis NANNF., Nova Acta Reg. Soc. Sci. Upsal・Ser. Ⅳ, 8 : 298, 1932.

Apothecia scattered or grouped, erumpent, stalked, at first globular, closed, opening in a round form, externally white, covered with excipular hairs. Hairs cylindrical, septate, minutely roughened, hyaline. Discs yellow to orange, 0.5 - 2 mm in diameter.

Asci cylindrical, with rounded apices, 60 - 87.5×5 - 8.5 μ m, mostly 60 - 80×5 - 7.5 μ m, 8 - spored. Ascospores obliquely uniseriate, elliptical to nallow kidney-shaped, occasionally with two oil globules, continuous, rarely bicellular, hyaline, 7 - 16×1.5 - 5 μ m, mostly 8 - 14×2 - 5 μ m. Paraphyses filiform overtopping the asci, septate, with many oil globules, 70 - 100×2 - 2.5 μ m.

The conidial stage consisting waxy, fleshy, erumpent, stroma with irregular, labyrinthiform cavities in which the conidia are abstricted from the tips of slender, acutely pointed, simple or verticillately branched conidiophores. Conidia continuous, spherical to elliptical, hyaline, 2 - 4 μ m in diameter. Conidia germinate on malt extract agar.

Host and Material : *Pinus strobus* L. (Sutorôbu-matsu) ——— Osamunai, Sorachi, XII - 25, 1974 by T. UOZUMI ; Fukagawa XI - , 1975 by Fukagawa District Forest office ; Kamioka, Gifu, IX - , 1975 by H. HOSOE.

Distribution : Asia （Japan）, North America, and Europe.

Note : DHARNE（1965）placed this species in the genus *Dasyscyphus* S, F. GRAY for the reason that this species differs from the other species of *Lachnellula* in the shapes of the ascospores and asci and in the smooth walled hairs. But the author's material has granulated hairs, which are identical with those of other species of *Lachnellula*. This species is easily distinguished from *L. calyciformis* by the size and the shape of the ascospores. This is new to Japanese fungus flora.

b *Lachnellula pini*（BRUNCH.）DENNIS

Persoonia 2 : 184, 1962 ; TAKAHASHI and SAHO, Trans, Hokkaido Branch, Jap. For. Soc. 22 : 103, 1973 ; SAHO and TAKAHASHI, Rept. Tottori Mycol. Inst. （Japan）10 : 706, 1973.（Fig. 18）

Synonym : *Dasyscypha pini*（BRUNCH.）HAHN and AYERS, Mycol. 26 : 487, 1934.
Lachnellula fuscosanguinea（REHM）DENNIS, SAHO and TAKAHASHI, For. Protection（Tokyo）21 : 210, 1972.

Apothecia scattered or grouped, occurring abundantly on resinous cankers, distinctly stalked, at first globular, urn-shaped, expanding as a flat disc, externally brown, covered with excipular hairs. Hairs cylindrical, septate, minutely roughened, brown.

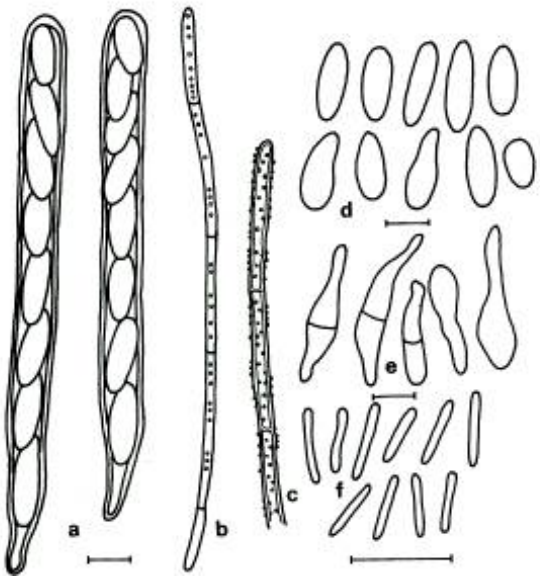


Fig.18.（図－18） *Lachnellula pini*（BRUNCH.）DENNIS

a : Asci and ascospores 子のうと子のう胞子
b : Paraphysis 側糸 c : Hair 毛
d : Ascospores 子のう胞子 e : Germinating ascospores 子のう胞子の発芽 f : Micro-conidia 小分生子 （├──┤ ＝10μm）

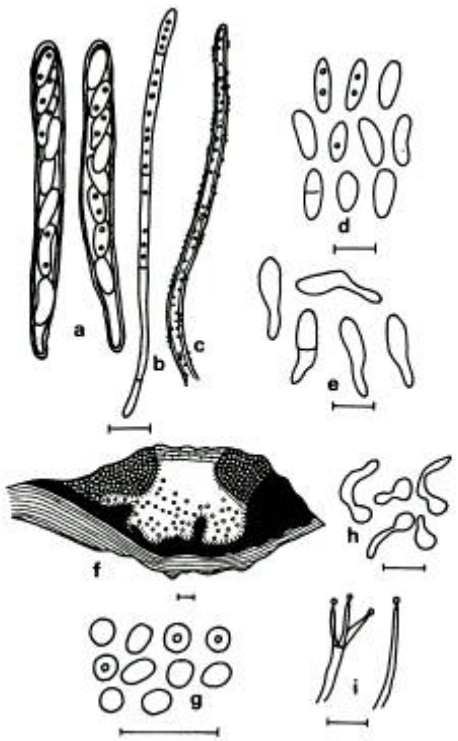


Fig.17.（図－17） *Lachnellula abietis*（KARST.）DENNIS

a : Asci and ascospores 子のうと子のう胞子
b : Paraphysis 側糸 c : Hair 毛
d : Ascospores 子のう胞子
e : Germinating ascospores 子のう胞子の発芽
f : Pycnidium 分生子殻
g : Conidia 分生子
h : Germinating conidia 分生子の発芽
i : Conidiophores 分生子柄
（├──┤ : a ～ e, g ～ i＝10μm ; f ＝100μm）

Discs bright orange to yellow ochre, 1 - 2 mm, occasionally 4 mm in diameter.

Asci cylindrical to clavate, 110 - 165×10 - 12.5 μm, mostly 110 - 155×10 - 11.5 μm, 8 - spored. Ascospores uniseriate, elongate elliptical, occasionally pyriform, with one apex obtuse, the other tapering to a subacute or acute extremity, hyaline, continuous, occasionally becoming bicellular on germination, 12.5 - 25×3.5 - 8.5 μm, mostly 15 - 22.5×6 - 7.5 μm, Paraphyses filamentous, outranking asci, septate, with yellow oil globules, 110 - 150×2 - 3 μm.

Conidial stage was not observed in nature and on culture. A few microconidia were produced on potato dextrose agar. Microconidia cylindrical to sausage-form, hyaline, 2.5 - 5.2×0.4 - 0.5 μm, mostly 3×0.5 μm. Microconidia did not germinate.

Host and Material : *Pinus pumila* REGEL (Hai-matsu) ——— Yamabe, Furano, VI - 16, 1972 by I. TAKAHASHI ; Higashikawa, Kamikawa, VII - 21, 1973 by T, O. ; Higashikawa Kamikawa, VIII - 20, 1973 by T. O. ; Higashikawa, Kamikawa, VII - 18, 1974 by T. O. ; Shintoku, Tokachi, VII - 19, 1974 by M. AKIMOTO ; Biei, Kamikawa, VII - 20, 1974 by M. AKIMOTO ; Ashoro, Tokachi, VI - 26, 1977 by Y. MURATA. *Pinus strobus* L. (Sutorôbu-matsu) ——— Yamabe, Furano, VII - 10, 1972 by I. TAKAHASHI ; Yamabe, Furano, VII - 24, 1973 by T. O. ; Yamabe, Furano, VII - 30, 1974 by T. O.

Distribution : Asia (Japan), North America, and Europe.

Note : This species was confused with *L. fuscosanguinea* for a long time, but HAHN and AYERS (1934) noted that there are some differences between these two species. SAHO and TAKAHASHI (1972) found a brown excipled *Lachnellula* on *Pinus strobus* and *P. pumila* at high altitudes on the Tokyo University Forest near Furano City, in the central Hokkaido, and identified it as *L. fuscosanguinea*. Then TAKAHASHI and SAHO (1973) revised its specific name to *L. pini*.

c *Lachnellula calyciformis* (WILLD, ex FR.) DHARNE (Fig. 2)

Morphology of this species habiting *Pinus* is the same as the characters that it was described on *Abies*.

Host and Material : *Pinus banksiana* LAMB. (Bankusu-matsu) —— Sarakitomanai, Sôya, VII - 11, 1979 by T. O. *P. strobus* L. (Sutorôbu-matsu) —— Yamabe, Furano, VIII - 1, 1979 by T. O. *P. sylvestris* L. (Yôroppa-akamatsu) —— Sarakitomanai, Sôya, VII - 11, 1979 by T. O. ; Yamabe, Furano, VIII - 1, 1979 by T. O.

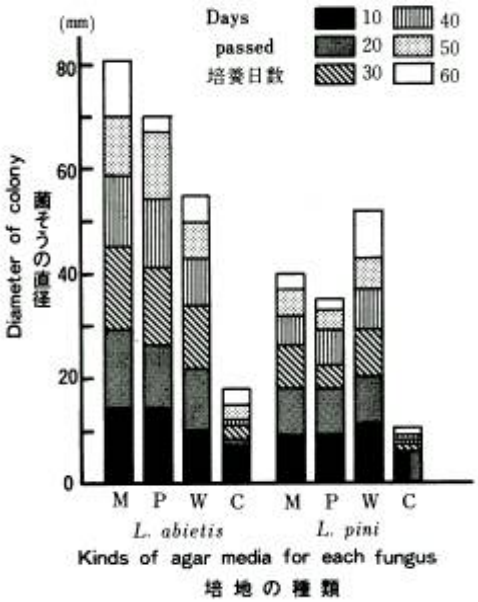


Fig.19. Mycelial growth on various agar media

図－19 培地上における菌そうの発育

M : Malt extract agar 麦芽寒天

P : Potato dextrose agar 馬鈴薯寒天

W : Waksman's solution agar ワックスマン寒天

C : Czapek's solution agar ツァペック寒天

2 Physiological studies

a Growth of colony on various agar media

The mycelial growth on the above-mentioned four agar media is shown in Fig. 7 and 19, and their cultural characteristics are given in Table 5 and 16. *L. abietis* and *L. calyciformis* grew well on malt extract agar and potato dextrose agar. *L. pini* grew best oh Waksman' s solution agar. In these cultural experiments the fungi did not produce apothecium and pycnidium on agar media, but *L. abietis* and *L. calyciformis* produced conidia on malt extract agar. Microconidia of *L. pini* were produced on potato dextrose agar in several stock cultures.

Table 16. Cultural characteristics of *Lachnellula* spp. on various agar media
表－16 *Lachnellula* 属菌の各種培地上の培養特徴

Media 培地 / Species 種名	Malt extract 麦芽	Potato dextrose 馬鈴薯	Waksman ワックスマン	Czapek ツァペック
L. abietis	c. vigorous, concentric, wavy, white, circular at margin ; a. m. rough, short, entangled, white, pale yellow at center.	c. vigorous, grey, circular at margin ; a. m. dense, short, white, pale yellow at center.	c. vigorous, pale brown, irregular at margin ; a. m. short, dense, white at center.	c. white almost circular at margin ; a. m. sparse, short, entangled, white at center.
L. pini	c. white, circular at margin ; a. m. short, mat-like, buff at center, white at margin.	c. white, circular at margin ; a. m. short, mat-like, buff at center, white at margin.	c. vigorous, pale vinaceous at center, white, circular at margin ; a. m. long, dense, white.	c. quiet poor ; a. m. only around inoculum, white, sparse.

c.： colony 菌そう, a. m. ：aerial hyphae 気中菌糸

b Growth on sterilized twigs

It was observed that mycelia grew and fruit bodies produced on sterilized twigs of *Pinus strobus* L. The results are given in Table 17. About 2 to 3 months after inoculatis *L. abietis* and *L. calyciformis* produced pycnidial bodies on the sterilized twigs. At first, white mycelial masses formed on the bark. Then white or pale pinkish-white drops of conidia oozed from the pycnidial masses. Apothecia of *L. abietis* and *L. calyciformis* were produced within 4 to 6 months of incubation. *L. pini* produced neither pycnidia nor apothecia.

3 Pathogenic studies

Inoculation experiment were carried out to determine the pathogenicity of these fungi.

The potted 5～7-year-old trees of *Pinus strobus* L. were prepared, and inoculation were made on 2～4-year-old portions of stems or branches. The results are summarized in Table 18.

1）Inoculation with *L. abietis*

able 17. Production of fruit body of *Lachnellula* spp. on sterilized twigs of *Pinus strobus*

表－17 ストローブマツ殺菌枝上における *Lachnellula* 属菌の子実体形成

Species 種名	No. of twigs tested 供試枝数	Production of Pycnidia 分生子殻形成	Production of Apothecia 子のう盤形成
L. abietis	3	＋ ～ ＋＋	－ ～ ＋
L. calyciformis	3	＋＋～＋＋＋	－ ～ ＋
L. pini	3	－	－

－：none ない, ＋：sparse わずか, ＋＋：meny 多い, ＋＋＋：abundant 豊富

Two to three years old branches of 6-year-old *Pinus strobus* were inoculated on November 17, 1976. The results were observed on September 19, 1977. The branches of all the trees developed lesions, and produced many pycnidia and apothecia elcept tree A' s.

2) Inoculation with *L. calyciformis*

Inoculations were made on 2～3-year-old branches of 7-year-old *P. strobus* on December 12, 1977. Observations were made July 25, 1978. Inoculated branches of all trees except tree D developed lesions, The two trees (D and E) died, and the remainders (A～C and F) developed cankers, from which resin exuded. No branches had produced either pycnidia or apothecia until November 30, 1978.

3) Inoculation with *L. pini*

Inoculations were made on 3～4-year-old portions of the stems of 5-year-old *P. strobus* on November 15, 1973. The results were observed in July 1974, April 1975, May 1976, and September 1976. The tree (E) produced apothecia and was dead on April 25, 1975. The two trees (C and D) produced many apothecia and were dead on May 24, 1976. The tree (B) produced apothecia and was dead on September 25, 1976. The tree (A) developed a canker and produced abundant apothecia until November 30, 1978.

Table 18. Inoculation experiment with *Lachnellula* spp. on *Pinus strobusw*
表－18 *Lachnellula* 属菌のストローブマツへの接種試験

Species 種名	Health group 健康状態	Tree designations 名称	Development of lesions 病斑の進展	Production of fruit body [a]		Condition of trees at the end of experiment 実験後の接種木の状態
				Pycnidia 分生子殻の形成	Apothecia 子のう盤の形成	
L. abietis	Healthy	A	+	−	−	healthy
		B	+	++	+	dead
		C	+	++	+	helthy
	Unhealthy	D	+	+++	+	healthy
		E	+	+++	+	dead
		F	+	++	+	dead
L. calyciformis	Healthy	A	+	−	−	healthy
		B	+	−	−	healthy
		C	+	−	−	healthy
	Unhealthy	D	−	−	−	dead
		E	+	−	−	dead
		F	+	−	−	healthy
L. pini	Healthy	A	+	−	++	healthy
		B	+	−	+	dead
		C	+	−	++	dead
	Unhealthy	D	+	−	++	dead
		E	+	−	+	dead
Check	Healthy	A	−	−	−	healthy
		B	−	−	−	healthy
		C	−	−	−	healthy
	Unhealthy	D	−	−	−	healthy
		E	−	−	−	healthy
		F	−	−	−	healthy

－：none ない, ＋：sparse わずか, ＋＋：many 多い, ＋＋＋：abundant 豊富

4 Discussion

L. pini has larger asci and ascospores than those of *L. abietis* (Fig. 20 and 21). The two species are easily distinguished each other with the naked eye, decause *L. pini* has brown excipled apothecia and *L. abietis* has white excipled apothecia.

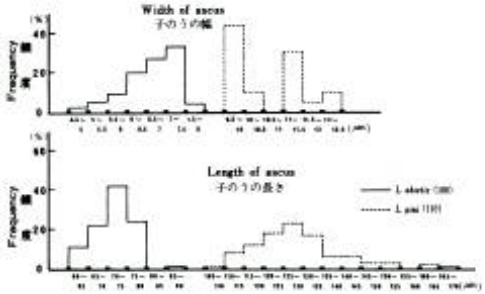


Fig.20. Histogram of length and width of ascus in 2 species of *Lachnenllula* on *Pinus* (Total account of measured asci)

図－20 マツ属上の *Lachnellula* 属菌 2 種の子のうの長さと幅の度数分布図（計測個数）

In the present experiments, *L. abietis* and *L. calyciformis* produced abundant conidia on malt extract agar and sterilized tiwgs as well as on inoculated twigs and stems. These fresh conidia germinated well on malt extract agar. *L. pini* did not produce conidia on any substrata, but a few microconidia were produced on potato dextrose agar. However, they did not germinate. According to HAHN and AYERS (1934), this fungus did not produce any conidial atage, including microconidia, on the various media.

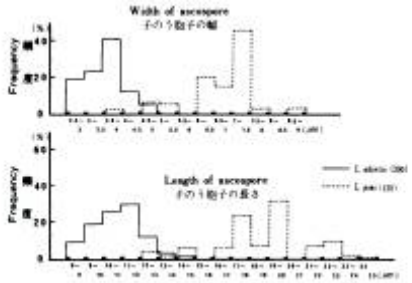


Fig.21. Histogram of length and width of ascospore in 2 species of *Lachnenllula* on *Pinus* (Total account of measured ascospores)

図－21 マツ属上の *Lachnellula* 属菌 2 種の子のう胞子の長さと幅の度数分布図（計測個数）

L. calyciformis has been a well-known causal agent of todo fir canker, but with respect to pine it has been unknown. *Pinus strobus* and *P. pumila* are new hosts of the fungus in Japan. According to KUJALA (1950) and HEPTING (1971), *L. abietis* is a saprophytic fungus on *Picea* in southern Finland and North America. Considering the results of the present inoculation experiment, *L. abietis* and *L. calyciformis* usually live saprophytically on dead or dying twigs of various conifers, but they often become actively parasitic on the yonung twigs and stems of pines under unfavorable conditions for the host plants, for instance, young forests in high altitudes or on unfavorable sites.

Fig.22. Distribution of the canker disease of *Pinus strobus* and *P. pumila*

図－22 ストローブマツとハイマツのがんしゅ病の分布

L. pini was reported from North America, Canada, northern Europe, and Cyprus, where it occurred at high latitudes and high altitudes (BROWNE 1968). STILLINGER (1929) stated that the heavily infected areas in North America occ-

ur at high elevations of 4,500 to 6,000 feet where white pine is near its altitudinal limits or rather unfavorable for white pine growth. *L. pini* has been reported from British Washigton, Idaho, Montana, and Michigan at elevations of 1,200 to 7,500 feet on *Pinus albicaulis*, *P. monticola*, and *P. strobus* (HAHN and AYERS 1934). According to KURKELA and NOROKORPI (1979), mortality of pines due to canker disease by *L. pini* at different altitudesin a locality (Laanila) of northern Finland was the followings: below 200 m, 6.4%; 200～300 m, 20.1%; above 300 m, 32.0%. At the same time they stated that the percentage of infected pine increases with the altitude and that it increases most rapidly at height of 150 to 200 m in northern Sweden (the basins of Kalix and Lule rivers) and *L. pini* occurs most severely at altitudes of 400 to 700 m in central Sweden (Norrland). According to TAKAHASHI and SAHO (1973), the areas damaged by *L. pini* range at elevation from 700 to 1,100 m on the Tokyo University Forest near Furano City in Hokkaido. The other infected areas in Hokkaido occur in *Pinus pumila* forests at mountains over 1,500 m.

All the diseases caused by *Lachnellula* on *Pinus strobus* and *P. pumila* have been recorded as *L. pini* since TAKAHASHI and SAHO (1973) reported from Hokkaido the *Lachnellula* canker disease of pine caused by *L. pini*. However, after critical examination of the canker disease on *Pinus strobus* and *P. pumila*, it was indicated that *L. abietis* and *L. calyciformis* also might cause this disease. The possibility that all three *Lachnellula* species may cause the death of pines was realized through inoculation experiments.

The distribution of the canker disease of pines caused by *Lachnellula* spp. is shown in Fig. 22, and the localities of the damaged areas are given in Table 19. The data in this table were based upon the 'Annual report of damage to the forest by diseases and pests and its protection (in Japanese)' which was published by Hokkaido Forest Protection Association.

Table 19. Distribution of damaged *Pinus strobus* plantations and *P. pumila* forests in Hokkaido

表－19　北海道におけるストローブマツ造林地とハイマツ林の被害地

Year 年	Locality 場所	Host 寄主	Damaged area 被害面積 (ha)
1973	Furano	*P. strobus*	0
	Furano	*P. pumila*	1
		Subtotal	1
'74	Nayoro	*P. strobus*	59
	Nakagawa	ditto	20
	Kamikawa	ditto	5
	Asahikawa	ditto	12
		Subtotal	96
'75	Fûren	*P. strobus*	11
	Nayoro	ditto	94
	Nakagawa	ditto	44
	Shimokawa	ditto	146
	Asahi	ditto	9
	Kamikawa	ditto	5
	Numata	ditto	28
	Fukagawa	ditto	29
	Asahikawa	ditto	5
	Enbetsu	ditto	16
	Furano	*P. pumila*	3
		Subtotal	390
'76	Sarufutsu	*P. strobus*	98
	Hamatonbetsu	ditto	85
	Fukagawa	ditto	18
	Asahikawa	ditto	5
	Rumoi	ditto	80
	Nakagawa	ditto	62
	Nayoro	ditto	68
	Fûren	ditto	11
	Furano	ditto	0
		Subtotal	427
'77	Sarufutsu	*P. strobus*	66
	Hamatonbetsu	ditto	60
	Wakkanai	ditto	46
	Utanobori	ditto	17
	Nakagawa	ditto	14
	Shimokawa	ditto	138
	Asahi	ditto	10
	Numata	ditto	14
	Fukagawa	ditto	19
	Hokuryû	ditto	32
	Rumoi	ditto	60
		Subtotal	476
'77	Shimokawa	*P. strobus*	313
	Nakagawa	ditto	37
	Hokuryû	ditto	13
	Fukagawa	ditto	38
	Numata	ditto	7
	Rumoi	ditto	5
	Obira	ditto	10
		Subtotal	432
		Total	1,813

The damage was hardly noticed until 1973, but the infected areas increased thereafter. The damage of *Pinus strobus* occurred mainly in young plantations less than ten years old, and that of *P. pumila* was found in the natural forests at the higher elevations around Mt. Ashibetsu, Mt. Tokachi, Mt. Asahi, and Mt. Meakan. The reports of the disease on both pines were from the areas with more than 2-meter snowfall (Fig. 23) and with more than 150-day snowfall a year (Fig. 24). HAHN and AYERS (1934) noted that the isothermal line of − 3.9 to − 1.1 ℃ (the monthly mean temperature in January) passed through the Pacific Northwest, the Upper Peninsula of Michigan, and Scandinavia, where *Dasyscyphapini* has been reported.

Fig.23. Maximum snow depth line (after WADACHI 1960)

図−23 最高積雪深（和達 1960）

Fig.24. Days with snow cover (after WADACHI 1960)

図−24 積雪日数（和達 1960）

In Hokkaido, the monthly mean temperature in January in the areas where *Lachnellula* canker occurs, is −6 to −12℃. And these areas are covered with more than 2-meter snowfall from October to April or May. However, low winter temperatures do not act as a predisposing factor in the development of the disease decause all of the young trees are protected by the thick snow cover. Among the environmental factors associated with the development of *Lachnellula* canker disease, the amount of snowfall and the duration of the snow mantle seem to be most important. The close relationship between a large amount of snowfall and the occurrence of the canker disease was already noted by YOKOTA et al. (1974, 1975) in the case of *Scleroderris* canker of todo fir. At the same time, they pointed out that the canker fungus was able to colonize on the bark which suffered from early frost damage, prior to the first snowfall. In the case of *Lachnellula* canker, the same phenomena may occur in the infected areas, and long dark humid conditions under the snow may act as a predisposing factor in the development of the canker disease. In addition to these unfavorable conditions, trees are wounded by the settling force and the creeping pressure of the snow. The causal fungi easily enter through these wounds, and gradually develop lesions under the snow.

D *Lachnellula calyciformis* on *Pinus* and *Pseudotsuga*

Morphology of this species habiting *Picea* and *Pseudotsuga* is the same as the characters that it was described on *Abies*.

Host and Material : *Pseudotsuga taxifolia* (POIR.) BRITT. (Dagurasu-momi) ——— Yamabe, Furano, Ⅷ - 1, 1979 by T. O.

Ⅷ Key to the species of *Lachnellula* KARST. in Hokkaido

Hairs of apothecium hyaline, appearing white
- Spores elongate-ellipsoid, over 20 μm long
  - Spores 17 - 27 μm long ····· *L. hahniana*
- Spores ellipsoid to ovoid, not over 20 μm long
  - Spores 12 - 20 μm long ····· *L. occidentalis*
  - Spores 10 - 17 μm long ····· *L. fuckelii*
  - Spores 7 - 16 μm long ····· *L. abietis*
  - Spores 5 - 13 μm long ····· *L. ciliata*
  - Spores 4.5 - 7 μm long ····· *L. calyciformis*
- Spores fusiform clavate, not over 12 μm long
  - Spores 5 - 9 μm long ····· *L. subtilissima*
- Spores globose, not over 10 μm in diameter·····
  - Spores 5 - 7.5 μm in diameter ····· *L. suecica*

Hairs of apothecium brown, appearing brown
- Spores elongate-ellipsoid, not over 20 μm long
  - Spores 12 - 15 μm long····· *L. pini*
- Spores globose to ellipsoid, not over 10 μm long
  - Spores 5 - 10 μm long····· *L. arida*
  - Spores 3 - 5 μm long····· *L. microspora*

Hairs of apothecium greenish, appearing green
- Spores subglobose to ovoid, not over 6 μm long
  - Spores 3.5 - 6 μm long····· *L. aeruginosa*

IX Host index

Abies alba MILL.
Lachnellula abietis, *L. calyciformis*, *L. subtilissima*
A. balsamea (L.) MILL.
Lachnellula arida, *L. calyciformis*, *L. hahniana*
A. cephalonica LOUD.
Lachnellula calyciformis
A. concolor LINDL. et GORD.
Lachnellula arida, *L. calyciformis*
A. firma S. et Z.
Lachnellula calyciformis
A. fraseri (PURSH) POIR.
Lachnellula calyciformis
A. grandis LINDL.
Lachnellula arida, *L. calyciformis*
A. holophylla MAX.
Lachnellula calyciformis, *L. subtilissima*
A. homolepis S. et Z.
Lachnellula calyciformis
A. lasiocarpa (HOOK.) NUTT.
Lachnellula arida
A. magnifica A. MURR.
Lachnellula arida
A. mariesii MAST.
Lachnellula calyciformis
A. nordmanniana SPACH.
Lachnellula calyciformis
A. pindrow ROYEL.
Lachnellula arida, *L. subtilissima*
A. procera REHD.
Lachnellula arida
A. sachalinensis MAST.
Lachnellula aeruginosa, *L. calyciformis*, *L. ciliata*, *L. fuckelii*, *L. microspora*
A. sibirica LEDEB.
Lachnellula calyciformis
A. veitchii LINDL.
Lachnellula calyciformis

Larix decidua MILL.
Lachnellula arida, *L. calyciformis*, *L. hahniana*, *L. occidentalis*, *L. subtilissima*, *L. suecica*
L. gmelinii GORDON
Lachnellula arida, *L. calyciformis*, *L. hahniana*, *L. suecica*
L. gmelinii × *L. leptolepis*
Lachnellula arida, *L. calyciformis*, *L. suecica*
L. laricina (DUROI) K. KOCH.
Lachnellula calyciformis, *L. hahniana*, *L. occidentalis*, *L. suecica*
L. leptolepis GORDON
Lachnellula calyciformis, *L. hahniana*, *L. occidentalis*, *L. suecica*
L. lyallii PARL.
Lachnellula hahniana
L. occidentalis NUTT.
Lachnellula arida, *L. calyciformis*, *L. occidentalis*, *L. suecica*
L. olgensis var. *Koreana* NAKAI
Lachnellula arida, *L. hahniana*, *L. suecica*
L. olgensis var. *Koreana* × *L. leptolepis*
Lachnellula calyciformis
Picea abies (L.) KARST.
Lachnellula abietis, *L. calyciformis*, *L. subtilissima*
P. engelmanni (PARRY) ENGELM.
Lachnellula arida, *L. suecica*
P. excelsa LINK.
Lachnellula arida, *L. calyciformis*, *L. fuckelii*, *L. subtilissima*, *L. suecica*
P. glehnii MAST.
Lachnellula calyciformis
P. jezoensis CARR.
Lachnellula calyciformis
P. mariana B. S. P.
Lachnellula hahniana
P. pungens ENGELM.
Lachnellula arida
P. rubens SARG.
Lachnellula abietis
P. sitchensis CARR.
Lachnellula calyciformis, *L. suecica*
Pinus albicaulis ENGELM.
Lachnellula arida, *L. pini*

P. attenuata LEMM.
Lachnellula calyciformis
P. banksiana LAMM.
Lachnellula calyciformis, *L. pini*, *L. subtilissima*, *L. suecica*
P. cembra L.
Lachnellula arida, *L. suecica*
P. contorta DOUGL.
Lachnellula arida, *L. calyciformis*, *L. suecica*
P. densiflora S. et Z.
Lachnellula subtilissima
P. flexilis JAMES
Lachnellula arida
P. halepensis var. *brutia* (TEN.) HENRY
Lachnelluly pini
P. koraiensis S. et Z.
Lachnellula suecica
P. montana MILL.
Lachnellula calyciformis, *L. fuckelii*, *L. subtilissima*, *L. suecica*
P. monticola DOUGL.
Lachnellula calyciformis, *L. pini*, *L. suecica*
P. mugo TURRA.
Lachnellula calyciformis, *L. subtilissima*
P. mugo var. *pumilo* (HAENKE) ZENARI
Lachnellula calyciformis
P. mugo var. *rostrata* (ANT.) HOOPES.
Lachnellula subtilissima
P. nigra ARNOLD
Lachnellula calyciformis, *L. pini*, *L. subtilissima*
P. nigra var. *caramanica* (LOUD.) REHD.
Lachnellula pini
P. nigra var. *poiretiana* (ANT.) ASCHERS & GRAEBN
Lachnellula calyciformis
P. ponderosa LAWS.
Lachnellula calyciformis
P. pumila REGEL
Lachnellula calyciformis, *L. microspora*, *L. pini*, *L. subtilissima*, *L. suecica*
P. radiata D. DON.
Lachnellula calyciformis
P. rigida MILL.
Lachnellula calyciformis

P. strobus L.

Lachnellula abietis, *L. calyciformis*, *L. pini*, *L. subtilissima*

P. sylvestris L.

Lachnellula calyciformis, *L. hahniana*, *L. pini*, *L. subtilissima*, *L. suecica*

P. thunbergii PARLATORE

Lachnellula subtilissima

Pseudotsuga taxifolia (POIR.) BRITT.

Lachnellula arida, *L. calyciformis*, *L. ciliata*, *L. hahniana*

Tsuga mertensiana SARG.

Lachnellula arida

X Host range

Lachnellula abietis (KARST.) DENNIS

Abies alba, *Picea abies*, *P. rubens*, *Pinus strobus*

Lachnellula aeruginosa OGUCHI

Abies sachalinensis

Lachnellula arida (PHILL.) DENNIS

Abies balsamea, *A. concolor*, *A. grandis*, *A. lasiocarpa*, *A. magnifica*, *A. pindrow*, *A. procera*, *Larix decidua*, *L. gmelinii*, *L. gmelinii*, *L. gmelinii* × *L. leptolepis*, *L. occidentalis*, *L. olgensis* var. *koreana*, *Picea engelmanni*, *P. excelsa*, *P. pnugens*, *Pinus albicaulis*, *P. cembra*, *P. contorta*, *P. flexilis*, *Pseudotsuga taxifolia*, *Tsuga mertensiana*

Lachnellula calyciformis (WILLD. ex FR.) DHARNE

Abies alba, *A. balsamea*, *A. cephalonica*, *A. concolor*, *A. firma*, *A. fraseri*, *A. grandis*, *A. holophylla*, *A. homolepis*, *A. mariesii*, *A. nordmanniana*, *A. sachalinensis*, *A. sibirica*, *A. veichii*, *Larix decidua*, *L. gmelinii* × *L. leptolepis*, *L. occidentalis*, *Picea abies*, *P. excelsa*, *P. glehnii*, *P. jezoensis*, *P. sitchensis*, *Pinus attenuata*, *P. banksiana*, *P. contorta*, *P. montana*, *P. monticola*, *P. mugo*, *P. mugo* var. *pumilo*, *P. nigra*, *P. nigra* var. *poiretiana*, *P. ponderosa*, *P. pumila*, *P. radiata*, *P. rigida*, *P. strobus*, *P. sylvestris*, *Pseudotsuga taxifolia*

Lachnellula ciliata (HAHN) DENNIS

Abies sachalinensis, *Pseudotsuga taxifolia*

Lachnellula fuckelii (BRES. ap. REHM) DHARNE

Abies sachalinensis, *Picea excelsa*, *Pinus montana*

Lachnellula hahniana (SEAVER) DENNIS

Abies balsamea, *Larix decidua*, *L. gmelinii*, *L. laricina*, *L. leptolepis*, *L. lyallii*, *L. olgensis* var. *koreana*, *Picea mariana*, *Pinus sylvestris*, *Pseudostuga taxifolia*

Lachnellula microspora ELLIS & EVERHALT

Abies sachalinensis, *Pinus pumila*

Lachnellula occidentalis (HAHN et AYERS) OGUCHI

Larix decidua, *L. laricina*, *L. leptolepis*, *L. occidentalis*

Lachnellula pini (BRUNCH.) DENNIS

Pinus albicaulis, *P. banksiana*, *P. halepensis* var. *brutia*, *P. monticola*, *P. nigra*, *P. nigra* var. *caramanica*, *P. pumila*, *P. strobus*, *P. sylvestris*.

Lachnellula subtilissima (COOKE) DENNIS

Abies alba, *A. holophylla*, *A. pindrow*, *Larix decidua*, *Picea abies*, *P. excelsa*, *Pinus banksiana*, *P. densiflora*, *P. montana*, *P. mugo*, *P. mugo* var. *rostrata*, *P. nigra*, *P. pumila*, *P. strobus*, *P. sylivestris*, *P. thunbergii*

Lachnellula suecica (de BY. ex FUCKEL) NANNF.

Larix decidua, *L. gmelinii*, *L. gmelinii* × *L. leptolepis*, *L. laricina*, *L. leptolepis*, *L. occidentalis*, *L. olgensis* var. *koreana*, *Pices engelmanni*, *P. excelsa*, *P. sitchensis*, *Pinus banksiana*, *P. cembra*, *P. contorta*, *P. koraiensis*, *P. montana*, *P. monticola*, *P. pumila*, *P. sylvestris*.

Summary

1. Morphological studies

1) Twelve species of *Lachnellula* have been collected from Hokkaido and they were identified.

2) *Lachnellula aeruginosa* OGUCHI was descrided and illustrated as a new species and L. occidentalis was proposed as a new combination, *Lachnellula occidentalis* (HAHN et AYERS) OGUCHI.

3) *L. abietis*, *L. ciliata*, *L. fuckelii*, *L. hahniana*, and *L. occidentalis* were new to Japanese fungus flora.

4) *Trichoscyphella calycina* (SCHUM. ex FR.) NANNF., which was considered to be causal fungus of the canker disease of todo fir, was treated as a synonym of *Lachnellula calyciformis* (WILLD. ex FR.) DHAENE.

2. Physiological studies

a Growth of colony on various agar media

Malt extract, potato dextrose, Waksman's solution, and Czapek's solution agar were used in this experiment.

1) Good growth of nine species except *L. aeruginosa*, *L. ciliata*, *L. pini* were recognized on malt extract &nd potato dextrose agar at 15℃ under dark condition and the above-mentioned three species grew well on Waksman's solution agar. On the other hand, growth of all mycelial colonies on Czapek's solution agar was generally poor or none.

2) *L. aeruginosa*, *L. calyciformis*, *L. hahniana*, and *L. subtilissima* produced apothecia on malt extract agar and *L. abietis*, *L. aeruginosa*, *L. calyciformis*, *L. ciliata*, *L. microspora*, and *L. subtilissima* probuced conidial masses on malt extract agar.

3) *L. aerginosa*, *L. fuckelii*, *L. hahniana*, *L. microspora*, *L. occidentalis*, and *L. suecica* produced microconidia on malt extract agar and *L. pini produced* those on potato dextrose agar.

b Growth of sterilized twigs

Twigs of *Abies sachalinensis*, *Larix leptolepis*, and *Pinus strobus* were used in this experiment.

1) *L. abietis*, *L. calyciformis*, *L. ciliata*, and *L. subtilissima* produced apothecia on sterilized twigs of *A. sachalinensis*, *L. leptolepis*, or *P. strobus*.

2) *L. aeruginosa*, *L. calyciformis*, *L. ciliata*, *L. microspora*, and *L. subtilissima* produced conidial masses on sterilized twigs of *A. sachalinensis*, *L. leptolepis*, or *P. strobus*.

3) *L. aeruginosa*, *L. fuckelii*, and *L. microspora* produced microconidia on sterilized twigs of *A. sachalinensis*.

c Germination of conidia

1) Conidia which were produced on agar media and sterilized twigs, germinated well on malt

extract agar at 15～25℃ within 72 hours after incubation.

2) The physiological characters of the culture isolated from conidia have close agreement with those obtained from ascospores.

3. Pathogenic studies

Inoculation experiment were carried out on potted *A. sachalinensis*, *L. leptolepis*, or *P. strobus*.

1) *L. abietis*, *L. calyciformis*, and *L. pini* developed lesions on stems or branches of *P. strobus* and *L. calyciformis* developed lesions on *A. sachalinensis*, too. These three species produced apothecia and *L. abietis* and *L. calyciformis* produced pycnidia on lesions.

2) It was confirmed that *L. calyciformis* was causal fungus of the canker disease of todo fir, and the possibility that *L. abietis*, *L. calyciformis*, and *L. pini* caused the death of pines was realized through inoculation experiment.

4. Distribution of the canker disease of pine

1) The canker disease of pine spreads widely in the plantation of *Pinus strobus* having heavy snowfall (more than 2 - m) and long duration with snow cover (more than 150 - day), and in the natural forests of *P. pumila* at high elevations.

2) The damaged areas caused by this disease were about 1,800 ha from 1973 to 1978.

5. Reference

1) Key to the species of *Lachnellula* in Hokkaido, host index, and host range were presented.

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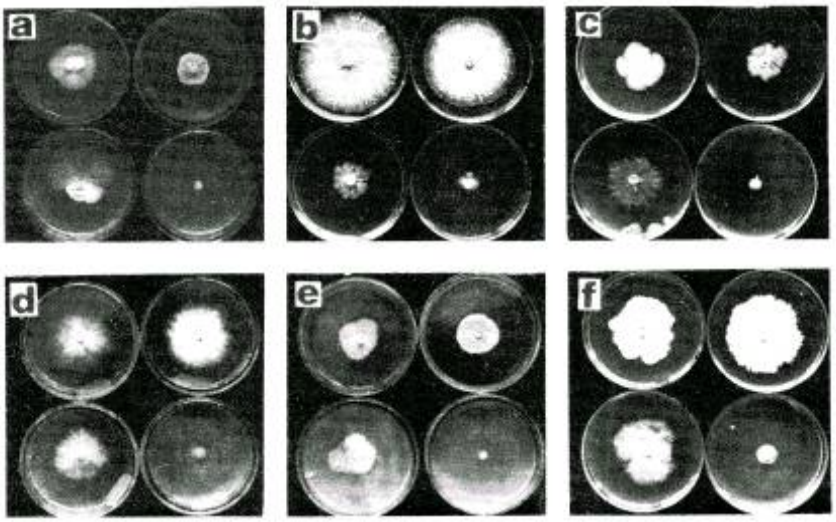

Photo.1.　Growth of colony on various agar media

写真－1　各種培地上の菌そうの発育

a : *L. aeruginosa*　b : *L. calyciformis*　c : *L. ciliata*　d *: L. fuckelii*

e : *L. microspora*　f : *L. subtilissima*

Upper left : Malt extract agar　Right : Potato dextrose agar

Lower left : Waksman' s solution agar　Right : Czapek' s solution agar

上左：麦芽寒天培地　右：馬鈴薯寒天培地

下左：ワックスマン寒天培地　右：ツァペック寒天培地

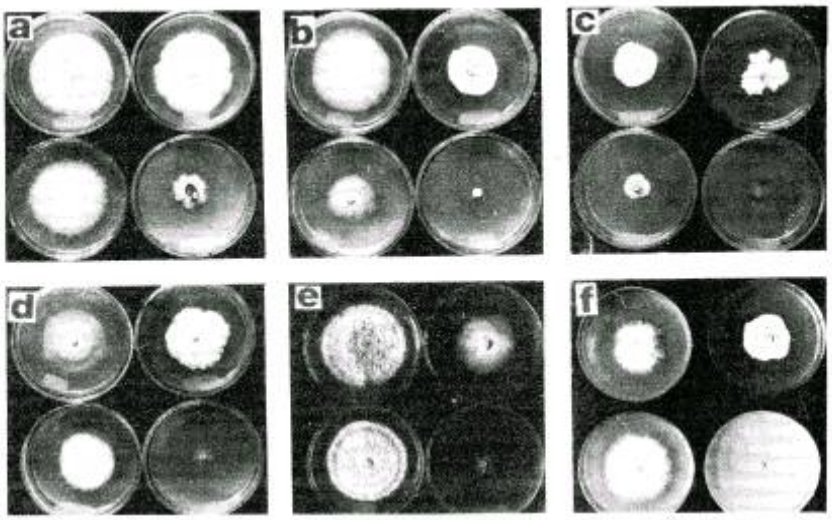

Photo.2.　Growth of colony on various agar media

写真－2　各種培地上の菌そうの発育

a : *L. arida*　b : *L. hahniana*　c : *L. occidentalis*　d : *L. suecica*

e : *L. abietis*　f : *L. pini*

Upper left : Malt extract agar　Right : Potato dextrose agar

Lower left : Waksman' s solution agar　Right : Czapek' s solution agar

but e is upper right : Waksman' s solution agar, lower left : Potato dextrose agar

上左：麦芽寒天培地　右：馬鈴薯寒天培地

下左：ワックスマン寒天培地　右：ツァペック寒天培地

但し e は上右：ワックスマン寒天培地　下左：馬鈴薯寒天培地

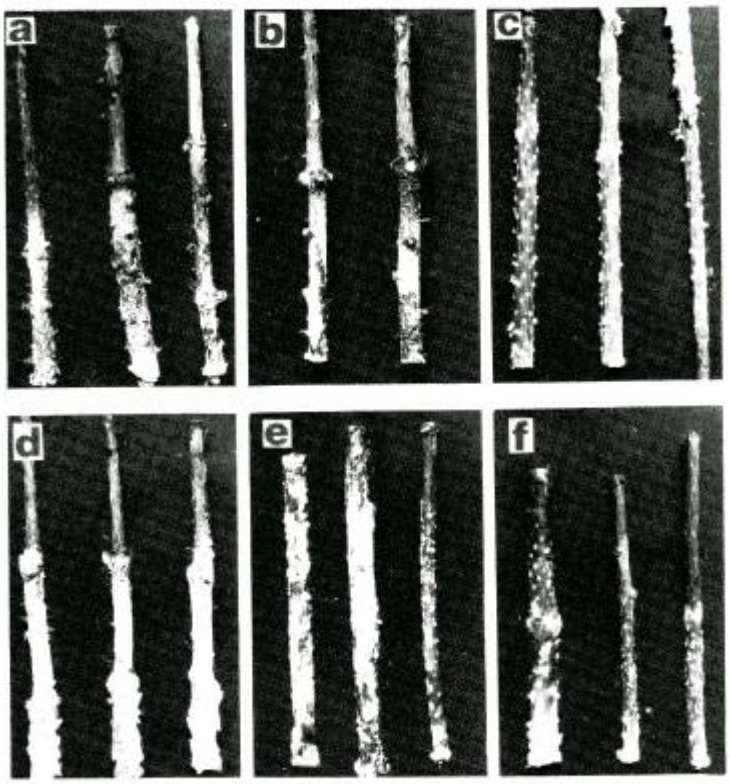

Photo.3. Mycelial growth on sterilized twigs of *Abies sachalinensis*

写真－3　トドマツ殺菌枝上の菌糸の発育

a : *L. aeruginosa*
b : *L. calyciformis*
c : *L. ciliata*
d : *L. fuckelii*
e : *L. microspora*
f : *L. subtilissima*

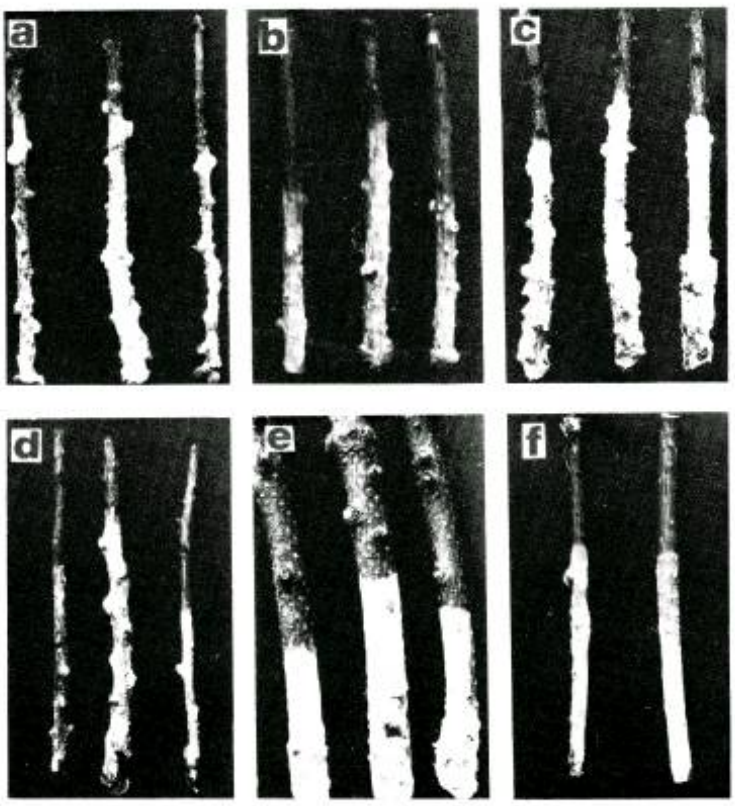

Photo.4. Mycelial growth on sterilized twigs

写真－4　殺菌枝上の菌糸の発育

a : *L. arida* on *Larix leptolepis*
b : *L. hahniana* on ditto
c : *L. occidentalis* on ditto
d : *L. suecica* on ditto
e : *L. abietis* on *Pinus strobus*
f : *L. pini* on ditto

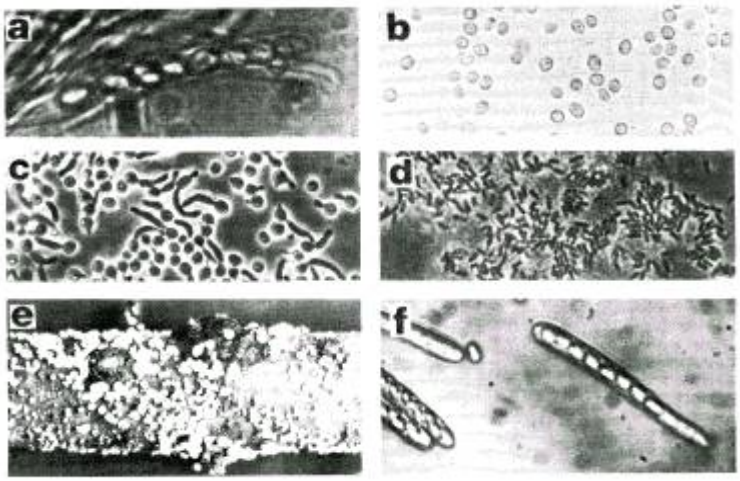

Photo.5. Apothecia on host plant and microscopical photographs of *Lachnellula* spp.

写真－5　寄主上の子のう盤と顕微鏡写真

a～b：*Lachnellula aeruginosa*

a：Ascus and ascspores　子のうと子のう胞子

b：Conidia　分生子　　c：Germinating conidia　分生子の発芽

d：Microconidia　小分生子

e～f：*Lachnellula calyciformis*

e：Apothecia on *Abies sachalinensis*　トドマツ上の子のう盤

f：Asci and ascospores　子のうと子のう胞子

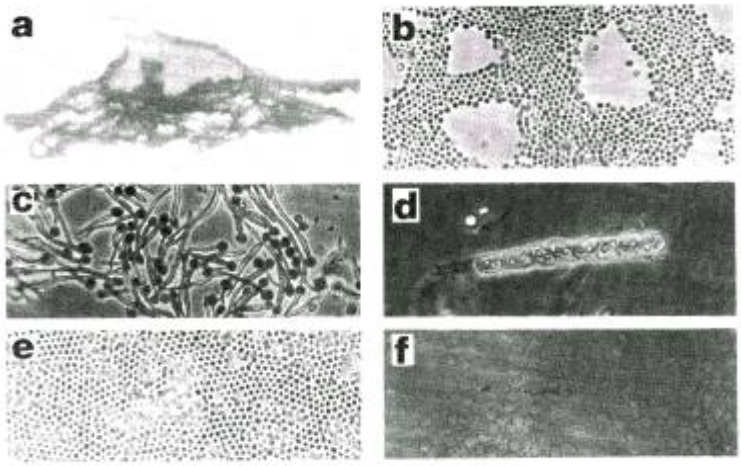

Photo.6. Microscopical photographs of *Lachnellula* spp.

写真－6　顕微鏡写真

a～c：*Lachnellula calyciformis*

a：Pycnidium on *Abies sachalinensis* トドマツ上の分生子殻

b：Conidia　分生子　c：Germinating conidia　分生子の発芽

d～f：*Lachnellula ciliata*

d：Ascus and ascospores　子のうと子のう胞子

e：Conidia　分生子

f：Germinating conidia　　分生子の発芽

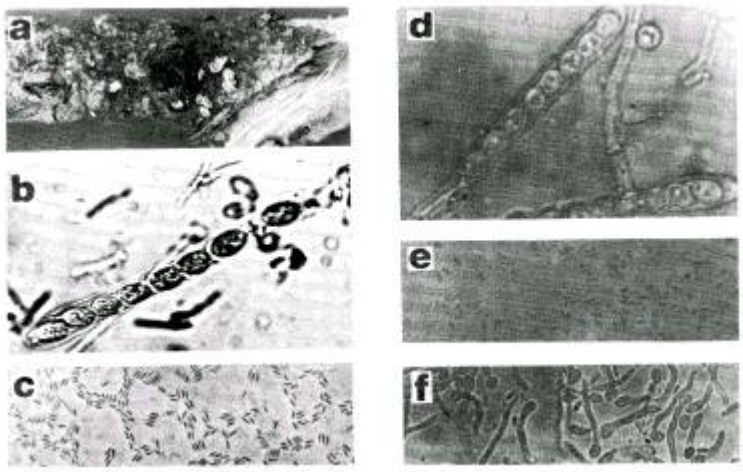

Photo.7. Apothecia on host plant and microscopical photographs of *Lachnellula* spp.

写真－7　寄主上の子のう盤と顕微鏡写真

a～b：*Lachnellula fuckelii*

a：Apothecia on *Abies sachalinensis* トドマツ上の子のう盤

b：Ascus and ascospores 子のうと子のう胞子

c：Microconidia　小分生子

d～f：*Lachnellula microspora*

d：Asci and ascospores 子のうと子のう胞子

e：Conidia　分生子

f：Germinating conidia 分生子の発芽

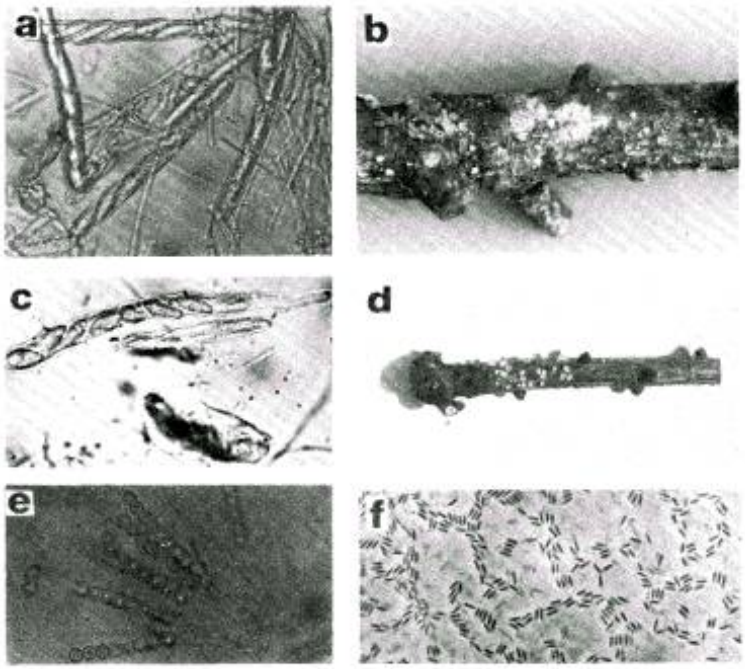

Photo.8. Apothecia on host plants and microscopical photographs of *Lachnellula* spp.

写真－8　寄主上の子のう盤と顕微鏡写真

a : Asci and ascospores of *Lachnellula hahniana*

L. hahniana　子のうと子のう胞子

b～c : *Lachmellula occidentalis*

b : Apothecia on *Larix leptolepis*　カラマツ上の子のう盤

c : Ascou and ascospores　子のうと子のう胞子

d～f : *Lachnellula suecica*

d : Apothecia on *Larix gmelinii*　グイマツ上の子のう盤

e : Asci and ascospores　子のうと子のう胞子

f : Microconidia　小分生子

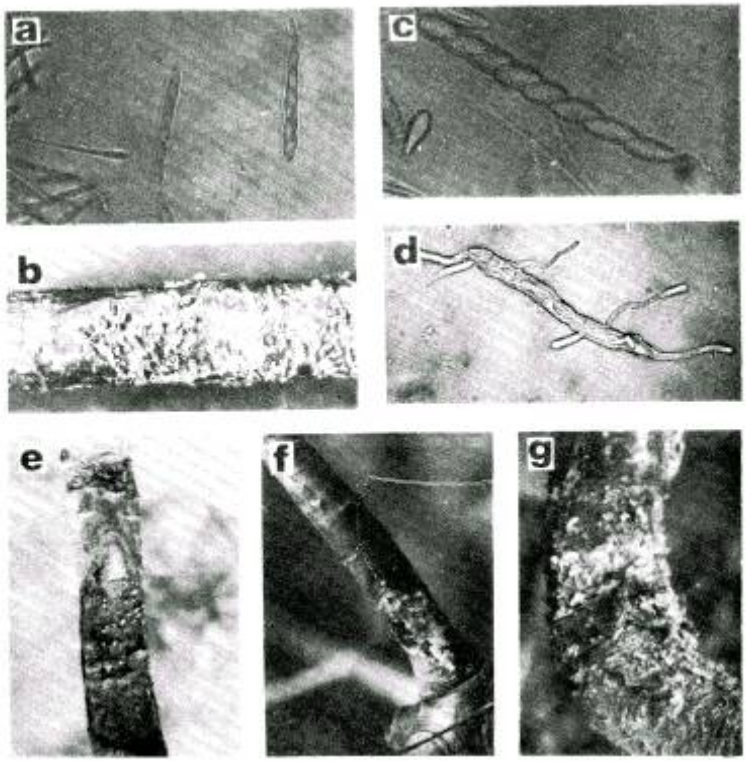

Photo.9. Apothecia on host plant and microscopical photographs of *Lachnellula* spp.　and results of inoculation experiment on *Pinus strobus*

写真－9　寄主上の子のう盤，顕微鏡写真とストローブマツへの接種試験結果

a : Asci and ascospores of *Lachnellula abietis*

L. abietis　の子のうと子のう胞子

b～d : *Lachnellula pini*

b : pothecia on *Pinus strobus*　ストローブマツ上の子のう盤

c : Ascus and ascospores　子のうと子のう胞子

d : Germinating ascospores in ascus　子のう内の子のう胞子の発芽

e : Lesion and pycnidia on branch inoculated with *L. abietis*

L. abietis　を接種した枝の病斑と分生子殻

f : Lesion on branch inoculated with *L. calyciformis*

L. calyciformis　を接種した枝の病斑

g : Canker and apothecia on stem inoculated with *L. pini*

L. pini を接種した枝のがんしゅと子のう盤