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THE TYPE AMYLOSTEREUM (BASIDIOMYCETES)
PARTIAL INTERCOMPATIBILITY BETWEEN SIMILAR SPECIES

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Le genre *Amylostereum* (Basidiomycetes) intercompatibilités partielles entre espèces allopatriques
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Summary

Studies of 4 species of *Amylostereum* of which *A. Ferreum* (Berk. An Curt.) nov. comb., distinguishing characters (Key), mycelial data. Two of the three-holarctic species (*A. Laevigatum* and *A. Chailletii*) give numerous hybrid mycelia with the neotropical *A. Ferreum*. Hypothesis on speciation process. The essential parts of the discussion are translated into English at the end of the paper.

The type *Amylostereum* Boid. (1958 a) was created for three holarctics, formerly described as: *A. chailletii* (Pers.: fr.) Boid. the species type, *A. areolatum* (Fr.) Boid. and *A. laevigatum* (Fr.) Boid. All three are hosts to conifers. If the last mentioned has been placed in the type *Peniophora* for a long time, the other two were *Stereum*, which many considered difficult to distinguish from each other. No other species has been reported to fall under the type *Amylostereum* since its creation. However, *Stereum ferreum* Berk. And Curt. 1868 originated from Cuba, *Stereum xylostroma* Lloyd 1922 described the Equator, both in unspecified forests, and *Lopharia ochracea* Cunn. 1963, which was harvested in New Zealand on *Dacrydium cupressinum* and *Pinus radiata* have characteristics of the type *Amylostereum*. The problem is then to find out whether these have the right to be known as autonomous species or should be synonym with one of the three *Amylostereum* given above.

The harvests made at Borneo, by E.J.H. Corner and others, between the altitudes 1 700 and 3 300 m, coming from Guadeloupe of *Podocarpus coriaceus* leads to the belief that the type *Amylostereum* exists in all areas where the Bymnosperms are distributed and particularly in *Podocarpaceae* (Plate 1).

It seemed desirable to study the basidioms of the floral or the most diverse geographic zones at the hand of live crops. If possible these should come from multiple types of Gymnosperms, of which the actual distribution and geological history are much better known than those of the mushroom. In effect, thanks to live crops received at Lyon, which have been rehumidified it was possible to obtain sporulation and to isolate the necessary monocaryotic cultures necessary for tests of intercompatibility-internoncompatibility. Thus one could try to know the number of 'biological' species of *Amylostereum*, their geographical distribution, their actual hosts and at the hand of these particulars, it could be possible to reconstitute the history of a type of Basidiomycete.

An call has been lauched to Botanists of the Southern hemisphere and the Far East particularly. In spite of the welcome received from many places at our demand, the results were extremely meagre, which we can say is due to the non-existence or the rarity of *Amylostereum* in certain regions of the globe. In contrast, a call to the temperate North region and particularly in Europe has enabled us to receive a number of copies from divers conifers - spontaneous or introduced. Further on we give the list of crops, which have permitted the obtaining of the cultures.

Since the start of classic studies by Buchner (1928), it was known that certain Hymenoptera *Siricidae*, which lay their eggs in weakened conifers, which deposits stored mildew in the 'intersegmental glands' besides. According to Francke-Grossmann (1967) 'strains of fungi, isolated from intersegmental pouches of *Sirex juvencus*, *S. Noctilio*, *S. Cyaneus* and *U. Gigas* were kindly determined by Nobles... as probably *Amylostereum chailletii* or a related species...'. Gaut (1969) has, through interfertilities and electrophoretic comparisons, identified the mushroom which destroys the *Pinus radiata*, which was introduced in Australia, as *A. areolatum*. It has been innoculated by *Sirex noctilio*, but has never been profitably observed in nature. This same *A. areolatum* was discovered in Japan in the glands of the *Sirex nitobei* (Kobayashi et al. 1978), and attacks the *Pinus desiflora* and *thunbergii*.

A. MATERIAL USED AND INTRASPECIFIC CONFRONTATIONS

Underneath we provide the list of the crops at the origin of the cultures and a list of the cultures received. They will be placed under the name that the intercompatibility tests affirm. For each of the four species, in alphabetical order, the geographical distribution is briefly presented after the particulars of literature.

1. *Amylostereum areolatum* (Fr.) Boid.

This species which, as demonstrated by Jahn (1971), essentially grows on *Picea abies*, is known in central and eastern Europe; it has only been shown in Finland once (Eriksson et al. 1978). As we shall see, it is identified by cultures from samples taken from the glands of the *Sirex noctilio* in Australia and Tasmania, and of the *Sirex nitobei* in Japan (loc.cit). We have, as herbarium, an English crop of *Pinus sp.* And crops from the soviet Far East of *Abies holophylla*.

Cultivated crops:

LY 3978, of *Picea abies*, St-Bon, Savoie (France), 4 September, 1961

LY 4328, of *Abies holophylla*, regio Primorsk, reservatum Kedrovaja Pdj., Oriens extremis (U.R.S.S.), 18 September, 1961, leg. E. Parmasto, P. 15298

LY 1922, dead trunk of *Picea* or *Abies*(?), the Boreon, Alpes-Maritimes (France), altitude 1500 m, 18 Sept. 1964.

LY 9026 and 9027, on *Picea abies*, Zwieseler Waldhaus, 750 m altitude, Baviere (Germany), 6 Dec, 1987, leg. I. Nuss.no 1596 and 1595.

Cultures received:

- 1) LY 4466, isolated from *Pinus radiata* after laying of the *Sirex noctilio* close to Hobart (Tasmania), determined *A. areolatum* by Gaut (1969)
- 2) Cultures received from T. Terashita and isolated in Japan in intersegmental sacs of *Sirex nitobei* in September 1971. Only the culture 'Sirex 7' from the origin Seto (Aichi Prefecture) will be used. This culture does not show arthrospores in March 1982.

On the contrary, all the other cultures of *A. areolatum* form arthrospores on malt-agar.

Intercompatibility between origins cited above:

The French culture 4922 is used by Gaut (1969) to identify the destructive mushroom of *Pinus Radiata* in Tasmania and Australia (no LY 4466). However, he has obtained 16 crosses between the 4 poles of 4922 and 4 Tasmanian neohaplont of which for results were positive, 8 imperfect and 4 negative. This French crop 4922 has proved compatible with 4328 (4 times on 4) and least completely with 9026 (5 times on 16) and 9027 (9 times on 12). 9026 and 9027 which between them gave 10 positive results on 12, cross well with Gaut's mushroom 4466: 6 positive results of 7 tried. Finally, the Japanese culture no 7 *Sirex nitobaei* dicaryotise the monocaryons of 4328.

The incomplete positive results are not due to alleles in common of polarity, which we could have discovered as having been used, in general, with the monosperms of poles preliminarily determined.

The crop 3878 is very meagre and not typical, but because of the presence of arthrospores in the culture it could be *A. areolatum*. Not having obtained isolated monosperms, we have deducted 92 cultures of mono-oidiens; monocaryotic at a young stage, in 1963. These cultures form rings regularly in three to five weeks. The dicaryon 3978 has dicaryotised the monocaryon of 4328 according to the phenomenon of Buller. Would it be about a *slow homothal natural hybrid* such as those obtained in the laboratory between *Dichostereum durum* and *sordulentum*, of which the monoconidien cultures, which are monocaryotic in the beginning and then form rings in the same way (Lanquetin and Boidin 1983)

2. *Amuylostereum chailletii* (Pers.:Fr.)Boid.

This species is much more widely distributed than the preceding one. It inhabits the whole of Europe where it grows mostly on *Abies Alba* and also on *Picea abies*, sometimes on *Larix europaea* and *Cedrus atlantica*. It is found in North America on diverse *abies* (*concolor*, *lasiocarpa*, *grandis*...), on several species of *Picea*, on *Pseudotsuga menziesii*, etc. (Burt, 1920, Lentz 1955, Gilbertson and Budington 1970, Gilbertson, Marint and Lindsey 1974, Martin and Gilbertson 1980). Rhill and Bakshi (1966) found it for the first time in the Indies on *Picea smithiana* and Thind (1973) and Rattan (1977) found it in the Himalayas on *Abies pindro* and *Cedrus deodara*. We have the crops of Georgie (U.R.S.S.) on *Abies nordmanniana* and *Picea orientalis*, of the Indies (U.P.) on *Picea morinda* as herbarium.

Cultivated crops

LY 4573, on conifers, Pyrenees-Atlantiques (France), 10 November 1963, leg. J. Beller.

LY 4622, on *Abies alba*, forest of Bizanos, Pyrenees-Atlantiques, 15 January 1964, leg. J. Beller.

LY 4855, on *Abies alba*, col du Cucheron, Savoie (France), 18 June 1964, leg. J.C. Leger.

LY 4893, on *Picea abies*, Peira Cava, Alpes-Maritimes (France), 15 September, 1974.

LY 8908, on *Abies alba*, les Bois noirs, St-Proest, Loire (France) 29 September 1978

LY 8943-8945-8949 A-8950 and 8951, on *Abies* sp., Ostergotland, Hjassatroget (Sweden), 31 October 1978, leg. T. Hallinback.

LY 8952, on *Abies* sp., Upland, Hacksta, Sundet (Sweden), 28 October 1978, leg. G. Eriksson.

LY 9525-9526 and 9528, on *Abies alba*, la Dagoniere, Mont Pilat (France), 27 September 1979.

LY 9536, on *Cedrus atlantica*, Luberon, Vaucluse (France), 12 December 1979, leg. A. David.

Cultures:

Sirex gigas no 1: isolated from a female *Sirex* during laying on *Picea abies* (?), col de Curvillat close to Hauteville, Ain (France), 15 September 1978, leg. J. Bussy.

21501, isolated from *Tsuga heterophylla*, Cowichan lake, B.C. (Canada), coll. R.E. Foster, det. M.K. Nobles 1948.

The cultures grow more slowly than those of the other *Amylostereum* and certian stocks grow particularly badly, such as 8908, of which the table of polarity could only be obtained; 5 growths on 8 were positive with 8950, and 3 on 9 with 459.

Die former cultures (549 to 4893) proved to be intercompatible at the time of their harvest. When confronted with 8943, 8945, 8949 A, 8950, 8951 and 8952, 459 gave 80 positive results on 108 pairs even though the cultures 459 has aged by 32 years. Several monocaryons which most often represent the four poles of each of the other Swedish and French crops have given 127 positive results on 136 growths with LY 8950 of Sweden, with the exception of 9536, harvested on *Cedrus* which was only compatible for 5 times on 14.

Of the neohaplonts obtained as from the culture *Sirex gigas* no 1 as those obtained from the Canadian dicaryon 2105, have given positive results with the 8950.

3. *Amylostereum ferreum* (Berk. And Curt.) comb.nov.

Found in Cuba, Found in Costa-Rica by Ptuillard (in Lloyd 1901), then by Burt (1920) in Jamaica, each time in indeterminate woods. Bagchee and Bakshi (loc.cit.) found thim in the Himalayas on *Taxus baccata*; it could therefore be a case of *A. laevigatum*.

Taken crops and cultures:

LY 8146 and 8149, on *Podocarpus criaceus*, Morne Leger, alt. 1 000 m, Parc National de Guadeloupe, 6 October 1976.

LY 8931, on *Podocarpus lamtertii*, Itaimbezinho, alt. 1 000 m, Rio Grande do Sul (Brazil), 4 October 1978, leg. R.T. Guerrero I.C.N. 6615.

These three crops are totally intercompatible (24 times on 24). It is worth mentioning that all three are found on the *Podocarpus* which could be the habitual hosts of this species.

4. *Amylostereum laevigatum* (Fr.) Boid.

Known in all of Eurasia on diverse *Juniperus*, specifically on *J. Communis* where it is standard, sometimes on *Taxus baccata*. Dennis et al. (1977) found it on *Curessus lusitanica* in Acores. Burt (1925) sites only two American crops: one in the state of Ne york, and the other in Canada. They do not occur in juniper mushrooms in Arizona, for example, according to Gilbertson and lindsey (1975). Its distribution in America is still to be specified.

Cultivated crops:

LY 4950, on *Juniperus communis*, col de Braus, Alpes-Maritimes (France), 18 September 1964.

LY 1827, on *J. Communis ssp. Nana*, Combe de la Loubatiere, Mont Ventoux, Vaucluse (France), 7 October 1978, Leg. P. Lanquetin.

LY 8934, on *J. Communis*, Courmangoux, Jura (France), 24 October 1978, leg. P. Lanquetin

LY 8944, on *Thuja sp.* Ostergotland, Hjassatorget (Sweden), 31 October 1978, leg. T. Hallingback

LY 8946, on *Juniperus communis*, Halland, Saro Vasterskog (Sweden), 31 October 1978, leg. N. Hallenberg

LY 8941, 8947, 8948 A and B, one the bark of a live *Taxus baccata*, same place, date and collector.

LY 8949 B and 8956, on *Abies sp.* Ostergotland, Hjassatorget (Sweden) 31 October 1978, leg. T. Hallingback.

LY 9201, on *Taxus baccata*, Egg-Erzberg, alt. 700 m (Switzerland), March 1979, leg. B. Erb.

LY 9474, on *Juniperus sabina*, route du col de Vars, Guillestre, Haute-Alpes (France), 22 July 1979.

The monosperms of the crops reported at *A. laevigatum*, were all confronted with the four poles of LY 8927. 183 positive results were obtained on 208 pairs. It is with LY 8946 that 24 negative results on 80 were noted. Below can be seen that the 8946 harvested on *J. Communis* in Sweden have spores which are longer ($x=7.81$) than those of the other crops on junipers (for example for 8927 $x=5.94$).

It can be observed that *A. laevigatum* is not only present on *Juniperus communis*, but *J. Nana* and *J. Sabina* as well as *Taxus baccata* are normal hosts of this *Amylostereum*. N. Hallenberg and T. Hallingback (1974) have already demonstrated the intercompatibility between crops on *Juniperus* and *Taxus* in Sweden, but also showed that the form occurring on *Taxus* has bigger spores than the one on *Juniperus*.

Here are our measures on spores in KOH-floxin, classified according to the function of their supports. (Average of 30 spores of profile)

Table translated on text.

It is found that if the spores of the crops on *Taxus* (Sweden and Switzerland) are in general of a larger size than those of the crops on *Juniperus*, the gathering 8946 (Sweden, on *Juniperus*) have even larger spores (plate III, 1).

With the *Vararia* of Gabon (1980) we have found that the yields L/1 of the order of 1,40 between the average height of the longest spores (L) and the average height of the shortest spores (l) does not prevent their intercompatibility. This is confirmed when L is taken on LY

9201 (say 7,46) and l on 8949 B (say 5, 70) the yield $L/l = 1,31$. However the crop with the longest spores is 8946 (on *Juniperus*, Sweden) and L/l then equals 1,37; in this case we have demonstrated a lesser intercompatibility of the 8946 with the 8927 (24 negative results on 80)

1. *Alaevigatum*: a, LY 8946, crop with big spores on *Juniperus communis*, Sweden; b, LY 8934, crop with small spores, on *J. Communis*, Jura; c, LY 9201, crop with big spores, on *Taxus baccata*, Switzerland.
2. *A. areolatum*, LY 4922, Alpes-Maritime
3. *A. chailletii*: d, LY 8950, with small spores, on *Abies*, Sweden; e, LY 4622, on *Abies*, Pyrenees-Atlantiques.

The intercompatibilities tell us that the Gymnosperms introduced in Sweden can be hosts to *A. laevigatum*, that is the *Thuja sp.* (for LY 8944) and *Abies sp.* (for 8949 B and 8956). Particularly to be noted is the case of the crop 8949: *A. laevigatum* (8949 B) and *A. chailletii* (8949 A), that is the fir, of which the natural area of distribution does not include Scandinavia, is able, in Sweden, to both shelter and make the two *Amylostereum* premises fruitful.

5. *Amylostereum sp.*

A) Crops which only gave negative results:

LY 8939, on *Pseudotsuga*, Univesrity Endowment Lands, Vancouver B.C. (Canada), 25 October 1978.

LY 9479, on *Abies alba*, forest of Boscodon, Alpes de Haute-Provence (France), 25 July 1979.

Cultures received

- Culture of H. Francke-Grossmann, isolated by *Sirex (Paururus) juvenicus*, Reinbeck (Germany), determined *St. Chailletii*.

- Culture F 10302, from dead *Abies balsanea*, St-Anne, Power Co, Lac Vert, Prov. Quebec (Canada), 23 October 1941, coll. A.W. Mc Callum, cult. M.K. Nobles.

The crop 8939 has been given two tests to create a table of polarity, each with 10 different monocaryons (2 times 45 confrontations); two confrontations were positive, and in 4 other cases several hooks were observed. These cultures grow extremely badly. The intercompatibility tests with the three European *Amylostereum* were all negative. Through its morphology the crop reports to *A. chailletii*. We have only one culture of polysperms of the crop 9479, and they are deprived of mildew. According to the phenomenon of Buller as in the confrontations done with 12 neohaplonts, no positive results were obtained with the three species of *Amylostereum*.

The cultures F10302 and that of Francke-Grossmann on *Sirex juvencus* have given no positive results in confrontations with *A. chailletii* 8950, *A. laevigatum* 8927 and *A. areolatum* 9027. In all these cases it depends on whether the cultures are very old or whether they grow with difficulty.

- B) Canadian cultures received under the name of *St. Chailletii*, which have given, according to the phenomenon of Buller, several positive results which are difficult to interpret:

B. DESCRIPTIVE PART

- 1) The type *Amylostereum* Boid. 1958

Sticky to reflexive Basidioms of a brown colour; context of diminished brown formed from genetically ringed hyphae and brown, skeletal hyphae, or not the horizontal context. The thick hymenium formed from numerous pseudocystids then cystids with a thick, brownish edge, carrying hyaline crystals; small, unremarkable gloecocystids, continually not very dense, positive syloaldehyde, often carrying a terminal schizopapilla; homobasids straightly claviformed with 4 sterigmata. Spores with a thin wall, smooth amyloids, white in great numbers. Tetrapolar heterothal. Normal nuclear behavior. Crossed with conifers, type *Stereum chailletii* (Pers:Fr.)

0 – Not the horizontal context nor cortex; mushroom displayed at an adherent margin, thickness of 300-1 800 μ m; large cystide of 7-12 μ m with roots. Spores 5,8-7,8x3,5-4-(4,8) μ m(*); common in Eurasia on *Juniperus spp.*; on *Taxus baccata* it achieves great thickness; sometimes on *Thuja sp.*

A. laevigatum (Fr.) Boid.

0 – a context of horizontal hyphes more or less thick; the mushroom can also be reflective, which shows it to be sterile; smaller cystids than at *A. laevigatum*

1 – Context generally underdeveloped and much thinner than the very crassulent hymenium; basiodoms with spots or blotches which thicken rapidly (0,5- 2,5 mm), with a rapidly thinning margin, only detached on 1 to 3 mm, showing a sombre, sterile front; non-sticky and breakable. Oblong spores of the front ellipses of the profile, 5-6 x 3-3,5 μ m. On *Podocarpus spp.* Central and South America.

A. ferreum (Berk. And Curt.) Boid. and Lanq.

1 – Developed context, generally much thicker than the hymenium, also the specimens are solid and can be reflective.

2 – Cylindrical spores 6.2-8 x 3-4 μ m (report length on breadth > 2).

The cortex only limits the reflective parts and is absent from the parts which stick well to trunks. The basiodoms is spread out to reflective on 0,5 to 1 cm of the beam, and therefore of a thickness less than 1mm; the superior front becomes smooth fast. Frequently on *Abies*, sometimes on *Picea*, *Larix*, *Cedrus*. Europe, Asia, North America.

2- Shorter spores, oblong, 4,8-6,5 x 2,8-3,8 μ m (R L/1 < 2).

Cortex present even in the spread out, sticky parts at the support where it can carry a dark brown tomentum. Under the magnifying glass, the cortex seems like a black edge, underlined by a clear line, formed by the basis of the context. Basiodom fast reflective, forming covers of 1 to 2 cm beams, 1 to 2 mm thick, often interwoven, between which the mushroom can take on a much thicker aspect. On *Picea* in Europe, on *Abies holphylla* in Siberia.

A. areolatum (Chaill. Ap. Fr.) Boid.

2) Description of *Amylostereum ferreum* (Berk. And Curt.) Boid. and Lanq (pl. II,1)

Small stains of a thick, tough series, spread out except at the margin, often closely raised, able to melt together to become larger marks. Closely detached part (maximum 3-4 mm) showing a sterile surface which is concentrically arranged, black brown, smooth. Hymenium of a more or less pinkish brown (SYR 6,2 to 6,5/3). Pale brownish flesh, clearer on the surface. From the cut rapidly achieves 1600 to 2000 μ m at a little distance from the margin. It forms with age a sort of darker cortex, blackish under the magnifying glass, dark brown in KOH floxin which is only a superficial part of the base subhorizontal hyphes. The layer of the base hyphes is irregularly developed sometimes of a thickness around 200 μ m, formed for a large part of skeletal hyphes scarcely passing 110 μ m length x 4 μ m, thickened at the front, brown (it is the type of horizontal cystids which do not have very deep roots.) and distinct, generating hyphes, frequently thickened and curled at the front. The hyphes do not differ in the cortex

but are more coloured there, and not melted together. At the summit of this zone the Hyphes stand up rapidly and there is a hymenial zone which is very thick (e.g. 1400 to 1750 μ m), without clear stratification, formed by cystids at the brown front, often very thick, more or less rooted on their superior part, 16-55x3,2-6 μ m, and on generating, curled hyphes. On the surface these cystids often have a thinner front and a paler tint with a summit carrying a heavier hyalin root; they measure in the region of 30-40x4-6 μ m with crystals; all the passages exist in these young cystides, and the deeper less rooted but thicker and more colored at the edge. Among the innumerable cystids, the gleocystids of 4-5 μ m with a thin edge, irregularly sub cylindrical or soften in their superior part, with an unremarkable content (several oily droplets) but ended by one or two schizopapilla. Several thin Hyphes, 1,5 μ m, attain or surpass the hymenial surface. Generally non emergent basids, size 4-4,8 μ m at the summit, at 4 sterigmata, the length of approximately 4 μ m ($x = 5,38 \pm 0,34 \times 3,22 \pm 0,20$ for 8146, $4,96 \pm 0,89 \times 3,38 \pm 0,37$ for 8149), at the edge smooth and thin, therefore clearly amyloid.

The description was given according to the Guadeloupean crops cited above.

Other crops examined

Cuba: *Stereum ferreum*, 199, Wright (Curtis) HOLOTYPE (in K).

Jamica: LY 3494, St-Andrew Parish, ca 4000-4900 feet, 5 September 1975, leg. A.L.Welden 1005

The Brazilian harvest (LY 8931) shows a cortex which is more developed and carrying a tomentum at the height of 300-350 μ m; it seems to have vegetated several times. Its spores are a little more elongated, 5-7x2,7-3,5 μ m ($x=5,98 \pm 0,56 \times 2,97 \pm 0,17$). If one takes into account the different crops examined, it is rather difficult to oppose *A. ferreum* with the *Amylostereum* of Europe. Through its structure it comes close to *A. areolatum*, which it does not carry, not having the same tendency to reflect, its context is always thinner than the hymenium; one is reminded by this fact of the thick species *A. laevigatum* on *Taxus*. Burt (1920, p.203) noticed this resemblance and writes: '*S. Ferreum* is at least closely related to *S. Areolatum*, a european species occurring on *Taxus*'. These last words lead one to believe that the author in fact thought of *A. laevigatum*, of which the thick specimens are similar, but which are totally deprived of a horizontal context and have longer spores.

The crop type, such as the crops of Jamaica (Burt, Welden) or of Costa Rica (Pouillard in Lloyd 1901) are in indetermined woods. The three live crops which we have studied are all on *Podocarpus* (*P. Coriaceus* in Guadeloupe, *P. Lambertii* in South Brazil); it therefore seems that *A. ferreum* is more or less co-existent with the type *podocarpus* or with *Podocarpaceae*. The researcher of numerous species would then need *Podocarpus* from America, oriental and meridional Africa, South West Asia and Oceania, as well as for example on *Dacrydium* in Chili and insulinde, on *Acmopyle* and New Cledon ... or also on *Phyllocladus*, Philipines in New Zealand (cf. Pl. 1). One would like to know whether *A. Ferreum* has followed these trees since the disjunction of the continent of Gondwana; and if, in the course of this long deviation of continents, whether or not there was the appearance of new species of *Amylostereum*.

3) Cultural characteristics of *Amylostereum ferreum*

Spores: non-fertilised LY 8149).

Monosperms (LY 8146-8149-8931). The spores germinate at the end of 48 hours and the isolated monosperms are formed from hyphes.

Polarity: The 10 monosperms of the Guadeloupean origin LY 8194 is divided into 4 poles:

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The appearance of 28 isolated monosperms of the Brazilian origine LY 8931 confirms the tetrapolarity of this species.

Numbers not typed over – look on page 224

Polysperms

- growth: varies from one culture to another, most often in covered woods within for weeks.
- Aspect: the margin is more or less regular. At six weeks the aspect of the cultures are rather uniform: the overhead mycelium is cottony and low, becoming muffled on cutting and its edges. Naturally whitish (10 YR8/3), it sometimes attains (in the cultures LY 8149) alutace (10 YR 8/4 to 8/6) and is even cream-coloured (7,5 YR 7/4) only on the cutting. The overhead mycelium of LY 8931 is less abundant and does not completely hide the area. The reverse of the cultures are unchanged and has no smell. Therefore we can say that the effective confrontations to establish the polarity, released 'n smell close to that of humid conifer sawdust.

Microscopy

- overhead mycelium: it is formed

1 degree of straight overhead Hyphes 1,2-2-(3) μ m, regular with constant rings, homogenous content, thin or distinct edge. At the base of the overhead mycelium, against the agar-agar, several larger hyphes, x 4-6-(7) μ m, irregular, with constant rings.

2 of pseudocystids, (70)-160-230-(335)x3,5-5-(7) μ m, which are terminal articles of greater or lesser length, lightly enlarged and finely regulated sometimes with small brownish marks. Their thickened edges, x0,5-1-(1,5) μ m, become thinner towards the base.

3 of straight sylvocystids with guttulous content, 32-105x1,5-2-(3) μ m, (more easily observable in young cultures) showing 5 to 7 small schizopapilla only visible in the red Congo. (Plate II, 1d).

4 sorts of very thin fibres, x1-1,2 μ m at the distinct edge, which is not dextrinoid.

- submerged mycelium: hyphes, x 2-4-(6) μ m, rather irregular, thin at the edge, curled.

- cytology: hyphes with regularly binucleated articles.

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We have found that all the *Amylostereum* have a normal nuclear behaviour and are tetrapolar. We resume their code here:

Not typed, look at page 225

A. areolatum is easily distinguishable in culture through the forming of arthrospores on the dicaryotic mycelium (Siepmann and Zycha 1968, Boidin in Gaut 1969) where they are on one or two stones; they also exist on monocaryotic mycelium. It is necessary then to remark that the mildew content in the intersegmental glands of a *Sirex gigas* from Col de Curvillat (Ain, France) have permitted the easy obtaining of one culture which is identified as *A. chailletii*, a culture which does not show arthrospores on malt-agar. *A. chailletii* would only be capable of dislocating its mycelium in symbiotic conditions.

B. TESTS OF INTERCOMPATIBILITY BETWEEN THE FOUR SPECIES OF AMYLOSTEREUM

Having found, according to preliminary tests, that the monocaryons of *A. ferreum* was susceptible to give several ringed mycelium as well as with *A. chailletii* with *A. laevigatum* it seemed necessary to confront, in every sense, the four known species of *Amylostereum*; except for the few crops of monospores that we do not have, the confrontations were effected between the poles of the different origins.

1) Confrontations between the three European species

- 236 confrontations *A. laevigatum* x *A. chailletii*: 236 negative results. They were done between recent crops LY 8927, 8949 B, 8956 for *A. laevigatum* and LY 8943, 8945, 8949 A, 8950, 8951, 8952, 9525, 9526, 9528, 9536 and DAOM 21501 (neohaplonts) for *A. chailletii*.
- 64 confrontations *A. laevigatum* x *A. areolatum*: 64 negative results. They were done between LY 8927 9049, 9201 for *A. laevigatum* and the LY 4922, 9026 and 9027 for *A. areolatum*.
- 100 confrontations *A. chailletii* x *A. areolatum*: 100 negative results. They were effected between LY 459, 8950, 9525, 9526, 9528 for *A. chailletii* and LY 9026 and 9027 for *A. areolatum*. It can be added that the 12 negative confrontations between the four poles of 9027 and the neohaplonts of *Sirex gigas* no 1.

2) Confrontations between *A. ferreum* and the three European *Amylostereum*.

A. ferreum x *A. areolatum*: 88 negative results on 88 confrontations.

A. ferreum x *A. Chailletii*: on 120 confrontations, 53 have at least given several localised rings on contact, but sometimes one of the two partners is completely dicaryotised. Table 1 details one part of these confrontations. The obtained results with neohaplonts of a North American crop of *A. chailletii* (21501) does not differ from those given by European monocaryons.

A. ferreum x *A. laevigatum*: 260 confrontations are presented in tables II and III.

Diverse results were found: totally negative, appearance of several distorted rings with hooks, rings close to the contact line, practically complete invasion of the territory of a haplont which was practically never that of *A. ferreum*. Several hybrid myceliums were addressed to R. Siepmann, who has succeeded in fructifying *Amylostereum areolatum* (1968).

TABLE TRANSLATED ON PAGE – SEE P 227

The American *A. ferreum* is then able to dicaryotise irregularly at the same time as the European monocaryons *A. chailletii* and *A. laevigatum*. One therefore has a sort of triangular intercompatibility.

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In Europe, nobody discusses the fact that *A. laevigatum* and *A. chailletii* would be two good species, which are morphologically and ecologically well distinguished, and the negative results (236 on 236) confirms this.

Several triangular results of this type were shown but never up until now, between morphologically distinct species.

Macrae (1967) finds that 11 North American origins of *the Hirschioporus abietinus* poroide divide themselves into two sub-divisions which are totally interincompatible; but these two sub-divisions are completely compatible with a German crop and partially compatible with three Norwegian origins.

Mounce and Macrae (1938) even finds that 45 North American origins of *Fomes pinicola* are divided into two groups A and B which are practically interincompatible (7 partially positive results on 103 crosses of origin). Seven European and Japanese origins are compatible with group A and partially (104 positive results for 306 negative results) with group B.

The same results are reported (Matsumoto et al. 1982) between a *Typhula ishikariensis* of the U.S.A. and the 'biotypes A and B' of Japan from the island Hokkaido, interincompatible with each other.

In the particular case of the *Armillariella mellea* are visually distinguished through myceliums, five American and European biological species reveals a case of triangular compatibility

between the B species of Europe and the IV and X species of America (Anderson et al. 1980). Two hypotheses can be formulated:

A0 The two American *H. Abietinus* or the *Fomes pinicola* A and B or the biotypes A and B of Matsumoto are the races of the same species between which there exists a heterogenic incompatibility (see Esser and Blaich 1973). The different genes of the polarity genes for example prohibits interfertility between the species A1 and C1 of the Ascomycete *Sordaria fimicola* (cf. Olive 1956) both compatible with the race C4. That is the explanation which we have suggested (Boidin in Clemenceon 1977, p292 and 298)

This interpretation is only applicable to *Amylostereum* during formation (thanks to an ecological isolation and especially a temporary geographical isolation) then becomes similar could have disappeared through repeated fusions or introgressions, could be lead to erect impassible barriers among themselves, which they have done. The raising of incomplete barriers give sterile hybrids, where there is a handicap in reproduction, a spread of genetic capability; the origins which are the most rapidly and effectively isolated therefore have an advantage and can dominate and then supplant the others. These insurpassible barriers have not been drawn towards *A ferreum* of which the area of distribution would have been distinct from those of the other three *Amylostereum* for a long time.

One could think to use this second hypothesis for *H. Abietinus* and *F. Pinicola*. *Hirschioporus abietinus* would, in North America, have been an aggregate of the two intercompatible twin species, having then put up effective sterility barriers. The European origins of *H. Abietinus*, - of the same title as the South and Central American *Amylostereum ferreum* - have on the contrary not placed up such barriers, and are close to the ancestral species, have retained the possibility to dicaryotise the American twins.

One could repeat this for the *Fomes pinicola* A and B, twin species here incompletely 'finished', because they are still very partially intercompatible.

Here we doubtlessly have interesting examples illustrating the continual speciation with the lignivore Basidiomycetes. One must remember that *Podocarpus* existed in Europe at the end of the tertiary era. One could imagine that since their distribution, the adaptation to different supports has led to the disjunction of species which are morphologically distinct and sexually strangers, but was not sufficient to still make *A. chailletii* and *laevigatum* entirely incompatible with their ancestor. *A. areolatum*, although morphologically closer to *A. chailletii* than to *A. laevigatum* today, must have diverged earlier. (Table V)

3) Case of the two Canadian cultures DAOM 21326 and 21498

According to the phenomenon of Buller, the culture 21326 has been confronted:

- a- with *A. areolatum* LY 9026 and 9027: 9 negative results on 9.
- b- With *A. chailletii* LY 8950 and 9528: rings are observed locally in 5 branches on 12
- c- With *A ferreum* LY 8149 and 8931: 5 times on 6 rings were seen on certain hyphae.
- d- With *A. laevigatum* LY 8927 and 8948: 5 times out of eleven rare rings were observed.

In the same way the culture DAOM 21498 was confronted:

- a- with *A. areolatum* LY 4922, 9026 and 9536: 10 negative results on 10.
- b- With *A. chailletii* LY 8950, 8951, 9528 and 9536: 12 locally positive results with 24 tests.
- c- With *A. ferreum* LY 8949 and 8931: 4 negative results on 4
- d- With *A. lavaegatum* LY 8927, 8941 and 8944: 8 results more or less positive on 14 trials.

It seems that in these two cases the results are totally negative with *A. areolatum*. This confirms the preceding conclusions on the clearest (and oldest?) autonomy of *A. areolatum*.

It is more difficult to draw parties from the other results. The two Canadian origins showed themselves compatible with none of the three other known species, and can only be attributed to one given species of *Amylostereum*. If 21326 has formed several rings with *A. ferreum*, the results for 21498 are negative, but only 4 trunks were observed and one can only draw a conclusion on one difference of significant behaviour between the two Canadian dicaryons.

The studie of the exsiccatum corresponding to the culture 21326 (for the other, isolated from the forest, exsiccatum does not exist) reminds one of *A. laevigatum*, whose presence in North America is not established. Let us remember that the European *A. laevigatum*, contrary to the two Canadian crops, has never made rings with *A. chailletii*. If the Canadian mushroom had to be reported at *A. laevigatum* (one must wish for new crops and a deepened study with regard to haploid cultures) one could here have had a supplementary case of partial intercompatibility between refined, but geographically further species: here Canada Europe and Canada Guadeloupe and Brazil.

D. Study of certain exotic specimens of *Amylostereum* in herbarium

The type of *Stereum xylostroma* Lloyd 1922 (coll. Rev. L. Mille, Ecuador, cat. No 6422 in BPI), compared by Lloyd with *Stereum sulcatum* which he shows exteriorly and without doubt an *A. ferreum*. *Lopharia ochracea* G.H. Cunningham 1963 is a New Zealand speicies which we know through two specimens (fragments of two paratypes): PDD 7582, on *Dacrydium cupressinum*, Te Whaiti, distr. Auckland, New0Zealand, June 1950, leg. J.M. Dingley, det. G.H. Cunn, and PDD 17454, same host, Pukekure, distr. Westland, New-Zealand, Nov. 1954, leg. J.M. Dingley, det. G.H. Dunningham. The type which we have not seen, was harvested on *Pinus radiata*; it differs according to G.H. Cunningham by its 'pilei reflexed... to 5mm radius'... The two examined specimens are indisputably *Amylostereum* but they are practically deprived of context, and it is not possible to pronounce the validity of this species. It could only be harvested in New Zealand recently and it is necessary to obtain spores and cultures for confrontations.

Thanks to E.J.H. Corner we could study four original specimens of the mountain of Borneo:

RSNB 742, Kinabalu, alt. 3 300 m, on *Dacrydium* (?), 10 July 1961.

RSNB 1913, Kinabalu, Mesilau, alt. 1 800 m, 21 April 1961.

RSNB 5414, Kinabalu, Bembangan River, alt. 1 800 m, 22 February 1964 and 5623, same place, 3 March 1964, all three on fallen trunks, without doubt from *Phyllocladus* (which is also part of *Podocarpaceae*).

These harvest all have the genre characteristics: cystids with a thick, brown edge, gloeocystids with terminal schizopapilla, amyloids, smooth rings... The structure is not clearly different from that of *A. ferreum*, with an often underdeveloped context, a very thick hymenium, a fast thickening and non reflective margin. Cystids and gloeocystids are the most strongly calibrated and are distributed less closely, giving a view of generic hyphes with a thin edge, dense and fused together, simulating a pseudoparenchyme. Measures on spores and cultures were used to compare the Indonesian *Amylostereum* better with American crops. We are convinced that it would be very interesting to confront crops on *Podocarpaceae* from further points of the globe.