Observations on Chytridiaceous Parasites of Phanerogams

XXVI. *Physoderma gerhardti* Schroeter on *Phalaris arundinacea* L.

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Abstract. Host symptoms and morphology of the endo- and epibiotic stages of a *Physoderma* on *Phalaris arundinacea* L. are described. It is considered identical with a *Physoderma* on *Agropyron repens* earlier described. Both are referred to *Physoderma gerhardti* Schroeter found in 1883 by Gerhardt from near Liegnitz, Schlesien.

Key words: *Physoderma* on *Phalaris* — Michigan (U.S.A.) — Morphology — *P. gerhardti* Schroeter.

In the course of studies of the chytrid genus *Physoderma*, a group of obligate parasites, primarily of vascular marsh plants, which abound in the vicinity of the Biological Station, Douglas Lake, Cheboygan Co., Mich., U.S.A., one has been found occasionally on the common Reed Canary Grass, *Phalaris arundinacea* L. The most productive of the sites has been a very wet swale in Section 23, Hebron Township, which in 1975 was covered with several feet of standing water.

HOST SYMPTOMS

Physoderma-infected plants were readily recognizable in late June and first 2 weeks of July by the presence on them of varicolored submerged leaves. This color pattern was caused by the fungus producing in the green host tissue extensive necrotic areas which frequently covered the entire half of the blade up to the midrib. Such inundated leaves had of course been readily available to infection by zoospores from recently germinated overwintered resting spores of the *Physoderma*. In dry springs such as in 1964 when standing water at the site existed for only a short time infection was scanty, and the lowermost leaves alone bore the fungus, which under these conditions were difficult to detect.

Within necrotic areas of the leaf blade are found clusters or "swarms" of dark reddish-brown to nearly black low pustules or spots 1–2 mm long by 0.25–0.5 mm wide (Fig. 1). These spots usually remain discrete, scattered objects, although along the margins of the blade they may occasionally fuse to form a streak. On the sheaths, spots are up to 2 mm long and are often a bright rust color which with age become nearly black. By the end of the 2 week in July in our area the whole infected leaf has become dark brown and the by now faint shadowy scattered evidences of the fungus are almost impossible to find. Infection at this time appears more generally distributed in the leaves. There is no evidence of systemic infection, such as stunting and only the leaves were attacked.

ENDOBIOTIC SYSTEM

The complex and highly specialized internal structure of an aquatic grass makes a study of the endobiotic system of the parasite somewhat difficult. All tissues of the leaf except those of the vascular system are invaded by the *Physoderma*. It will be recalled that the endobiotic vegetative system of *Physoderma* is strongly polycentric and originates by an infecting zoospore producing a primary "turbinate cell" in the host epidermis (Figs. 12 and 13) from which rhizoids emerge (Fig. 11). It consists of very delicate occasionally branched rhizoids on which are placed in intercalary fashion the secondarily produced "turbinate cells". The latter are nucleated, one or several-segmented swellings (Fig. 3a) which by the extension of rhizoids from them further replicate the thallus as it wanders from cell to cell within the host tissue. In *Physoderma* no lysis of infected tissue ordinarily occurs.

In the *Physoderma* on *Phalaris* these rhizoids are richly branched distally (Fig. 3), a feature not usually
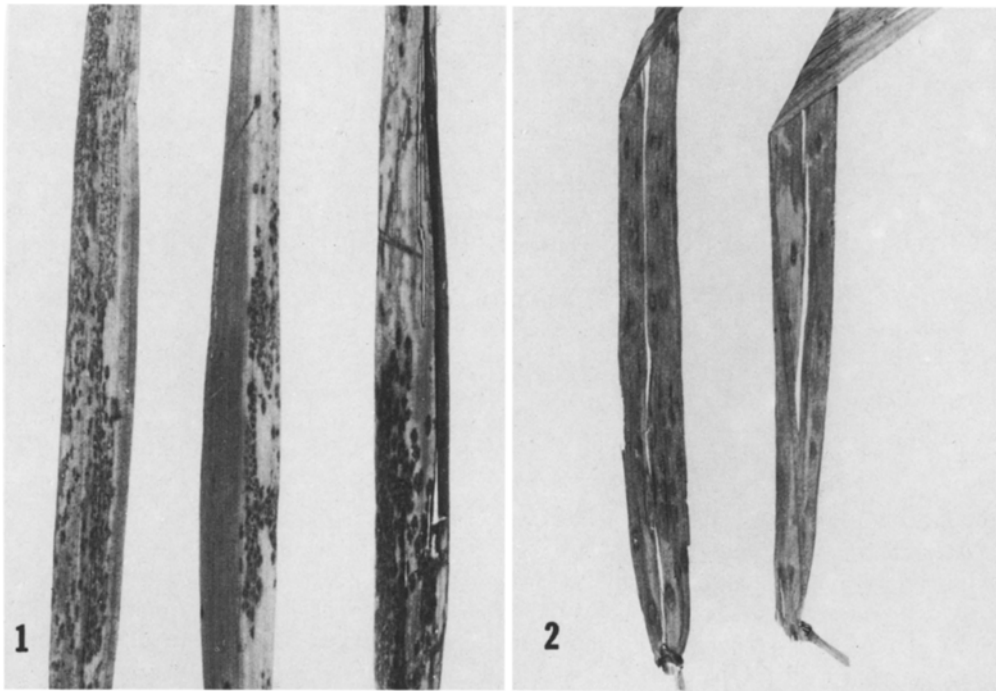
Figs. 1 and 2
Leaves of *Phalaris arundinacea* infected by *Physoderma gerhardti*

Fig. 1
Portions of leaves of Michigan material showing small pustules produced by fungus, some distinctly clumped. Light portions of leaf yellowed, necrotic; dark portions green

Fig. 2
Portions of leaves of *Phalaris* collected at Liegnitz by Gerhardt in 1883, and considered lectotype of *P. gerhardti* Schroeter

observed in other *Physodermas*. The turbinate cells, for example, those in the host leaf sheath where they are best observed, are ovate, often medianly constricted intercalary structures (Figs. 3–5), are occasionally 1–3-partitioned, and $10-20 \times 6-10$ µm. In tissues of the leaf blade, except for the epidermis, both turbinate cells and rhizoids are difficult to observe due to crowding of parts. The disproportionately large ovoid amber to brown thick-walled resting spores are the most conspicuous objects borne on the rhizoidal system. Each originates as a small thin-walled, globose, colorless object formed at the tip of a short lateral tube projected from one cell of the turbinate cell (Fig. 4a). Their origin and subsequent development can best be followed in the sheath and in epidermal cells of the blade. In other tissues these processes cannot be observed because of the crowding of parts and presence of dark disorganized host contents. Here, nothing can be seen but the mass of developing or already mature resting spores (Figs. 6–8). Maturing resting spores, sometimes ornamented with delicate antler-like processes (Fig. 5b), bear in their contents

numerous scattered refractive globules. Further maturation involves gradual centripetal thickening and coloration of the wall. The contents now bear globose and more like-sized refractive structures disposed around a large central vacuole (Fig. 5a). At full maturity the resting spore is slightly ellipsoidal with one face somewhat flattened, and with a 1.51 µm thick, clear, amber- to brown-colored wall (Figs. 3 and 5). Shape of the resting spore as well as its size, varies in the different tissues (Figs. 6–8). A medianly constricted type of resting spore is often found lying singly and loosely in a large cell (Figs. 9 and 10). Clearly, this shape is not due to restrictions imposed by that of the host cell. In the leaf blade compact parenchyma they are primarily ellipsoidal, as stated, and varied from $17.5-31.5 \times 14-21$ µm, averaging 23.3×17.6 µm. In the aerenchyma (Fig. 8) spores tended generally to be larger (up to 35×25 µm).

Germination of the resting spores was secured after they had been held for about 6 months at $1-2°$ C and then transferred to shallow dishes of charcoal water in a light chamber at $20°$ C. This process did

Figs. 3–15.   Endobiotic system, resting spores and their germination of *Physoderma gerhardti*

Figs. 3–5.   Portions of host cells (*Phalaris*) showing rhizoids bearing turbinate cells (Fig. 3a) many of which have produced resting spores at tip of a short lateral outgrowth (Fig. 4a, for example). Unusual, much-branched, distal elements of rhizoids shown in Figure 3. In Figure 5a and b antler-like outgrowths on immature and mature resting spores are seen

Fig. 6.   Masses of resting spores crowded into aerenchyma showing variation in size

Fig. 7.   Same in cells along either side of midrib showing great variation in size and shape of resting spores
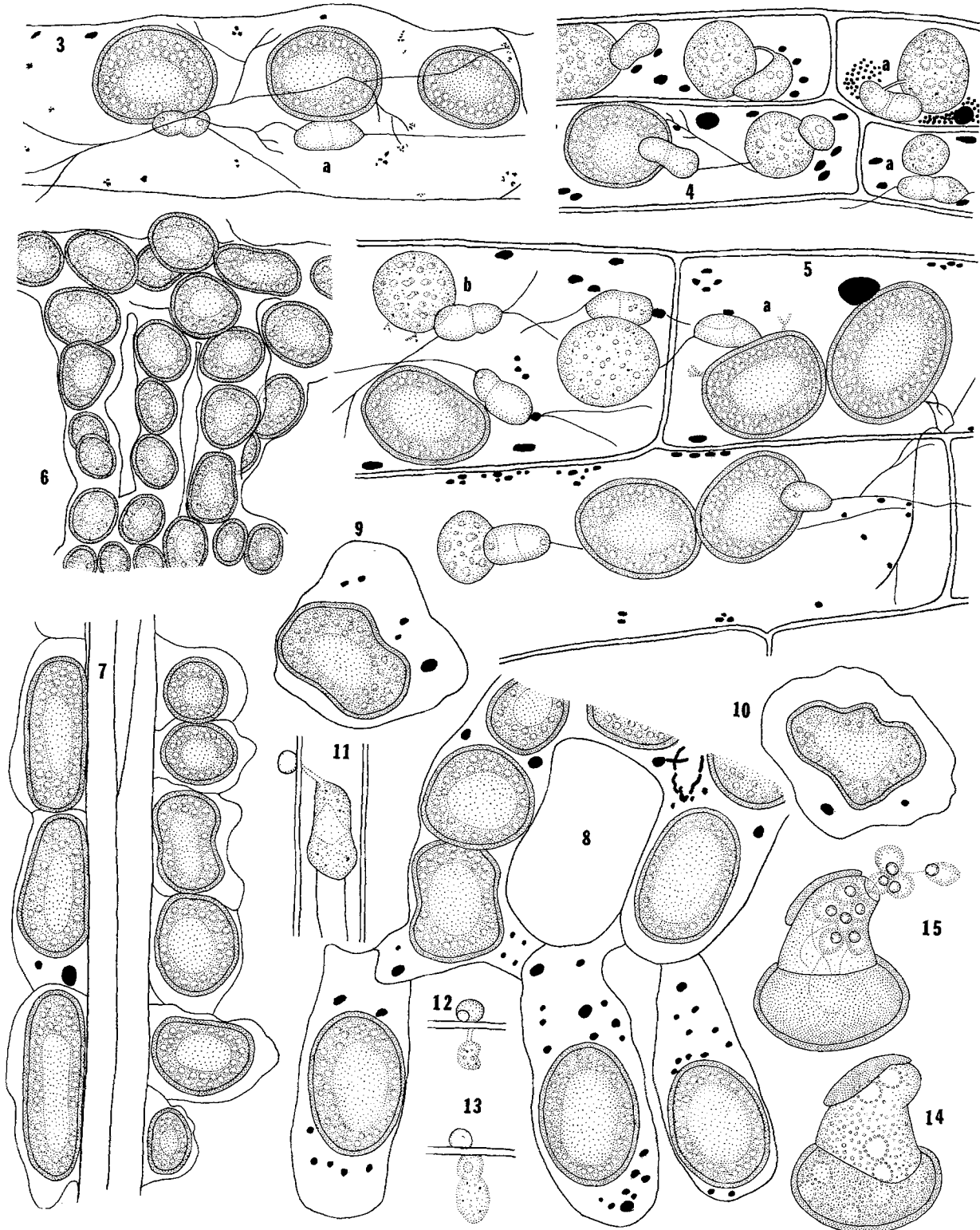
Fig. 8.   Aerenchyma with host contents masking all but resting spores of parasite

Figs. 9 and 10.   Two parenchyma cells showing irregular shape of resting spores seems not to be influenced by shape of host cell

Fig. 11.   Establishment of first turbinate cell and rhizoids in host by germinated R.S. zoospore

Figs. 12 and 13.   Earlier stages in infection by R.S. zoospore

Figs. 14 and 15.   Two stages in germination of resting spores of *Phalaris* parasite

Figs. 11—13 of *Agropyron* parasite; all others of *Phalaris* parasite. All Figures (except Fig. 6) ×825; Fig. 6 ×400
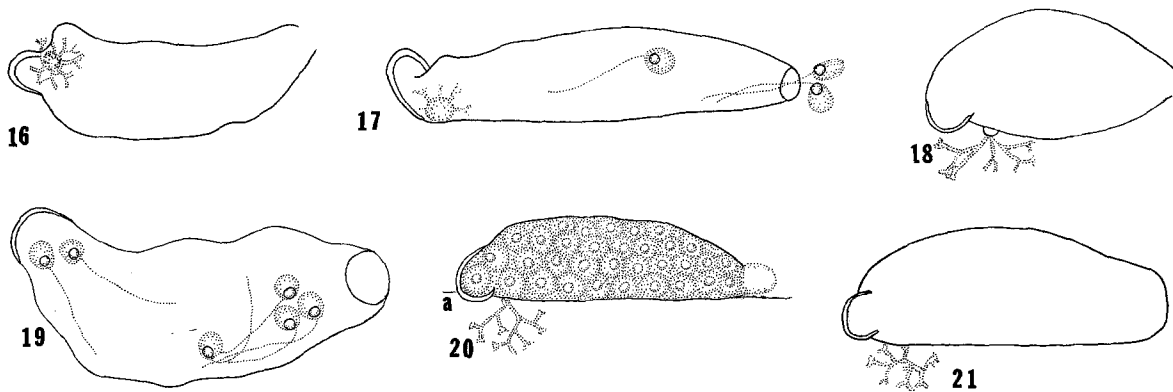
Figs. 16—21. Unilaterally developed epibiotic sporangia of *P. gerhardti*

Figs. 16, 17, and 19. Sporangia (*top view*), some with zoospores, formed by R.S. zoospores of *Phalaris* parasite on *Elymus canadensis*. At the left end of the procumbent, sessile, epibiotic sporangium is the relatively unexpanded part of the original cyst of the R.S. zoospore. In Figure 16 the rhizoidal system in the epidermal cell is also visible at this end. The opposite end bears the discharge orifice for the zoospores

Figs. 18, 20, and 21. Sporangia (*side view*) produced by parasite on *Agropyron* on *Phalaris*. In Figure 18, a small apophysis is formed from which stubby rhizoids emerge. The others lack this structure. In Figure 20 the zoospores are mature and about to be discharged through an orifice produced upon the deliquescence of the blunt protruding papilla. All Figures ×825

not differ in any essentials from that described in detail for a *Physoderma* on *Agropyron repens* (Sparrow et al., 1961) and will not be repeated here. In Figures 14 and 15 are shown the circumcissiley dehisced cap of wall material, finger-like zoosporangium and "R.S." zoospores[1] (Fig. 15) emerging through an apical pore from the sporangium. These zoospores are slightly ellipsoidal, 6—7 µm in diameter with a single colorless globule and 30 µm long posterior flagellum. In motion they become distinctly elongate and 10—12 × 5—6 µm. Such zoospores give rise either to a new endobiotic, polycentric stage or to a second type of thallus, the so-called epibiotic one.

Epibiotic Stage

The epibiotic stage consists of a sporangium resting on an epidermal cell of the host, attached to a short, endobiotic bushy complex of rhizoids (Figs. 16—21). Development of this stage in *Physoderma* on *Phalaris* was followed most successfully on seedlings of *Elymus canadensis*, which were readily infected by the fungus[2]. Once the R.S. zoospore became encysted on the host surface, a penetration tube was produced at the tip of which, endobiotically, a series of short bushy rhizoids formed which remained within the single epidermal cell (Figs. 18, 20, and 21). The spore body then gradually enlarged and elongated in one direction only, i.e., at right angles to the axis of the penetration

[1] Zoospores from germinated resting spores
[2] *Phalaris* seedlings could rarely be obtained when needed and mature plant parts were too massive and obscure for this purpose. The epibiotic stage has been shown, however (Sparrow, 1975), to maintain its essential features on different hosts

tube and hence parallel with the surface of the epidermis. The rest of the spore (approximately one-half) remained a nearly unchanged hemisphere but did undergo with time enlargement and thickening of its wall. Such a "unilateral type" of development (Sparrow, 1975) results in a finger-like or slipper-shaped, occasionally gibbose, sporangium body lying along the plane of the host cell surface (Fig. 20), with one end at maturity producing a discharge papilla and the other near the penetration tube bearing the thick-walled hemispherical remains of the zoospore cyst (Fig. 20a). These epibiotic zoosporangia are 50—100 µm long by 14—27 µm in diameter. The zoospores formed in them and discharged upon the deliquescence of the papilla (Fig. 17), are 4—5 µm in diameter, with a colorless globule and single posterior flagellum. They are distinctly elongate during motility and capable of intervals of amoebod movement.

A summary of the morphological features of the *Phalaris* parasite is given in Table 1, where data are also given for the apparently identical parasite studied on *Agropyron repens* (Sparrow et al., 1961) collected at the same site and for Gerhardt's fungus on *Phalaris* collected at Liegnitz in Schlesien in 1883, considered here to be the lectotype of *Physoderma gerhardti* Schroeter.

Host Range

Only a few data were collected on the host range of the *Phalaris* parasite. These were obtained by methods outlined in an earlier paper (Sparrow and Griffin, 1964). The results are given in Table 2 which includes

Table 1. Principle morphological features of Physodermas on *Phalaris arundinacea* and *Agyropyron repens* from Michigan, and from Liegnitz (coll. Gerhardt, 1883)

Host	Host symptoms	Endobiotic vege-tative stage	Resting spores	R.S. zoospores	Epibiotic stage	Epibiotic zoospores
Phalaris arundinacea, Michigan material	Clusters on blade and sheath of separate reddish brown to blackish fusiform-ovate low pustules, 1—2 mm long by 0.25—0.5 mm wide. More scattered on old leaves	Rhizoids richly branched distally t.c. ovate, 1—3 celled, 10—20 × 6—10 µm	Bright amber to brown wall 1.51 µm thick. Variable in size sometimes according to tissue, 17.5—31.5 ×14—21 µm (av. 23.3 × 17.6 µm). Up to 35 × 25 µm in aerenchyma	6—7 µm dia. with colorless globule, 30 µm long flagellum; 10—12 × 5—6 µm in motion	Unilateral develop-ment ± elongate, slipper-shaped occasionally gibbose 50—100 × 14—27 µm. Rhizoids sparse, bushy, somewhat coarse	4—5 µm dia. with colorless globule; flagellum 20—25 µm long
Agropyron repens Michigan, (Sparrow et al., 1961)	Dark brown streaks, less than 1.0—20 mm long × 0.5 mm wide mostly 4—6 mm long	Rhizoids not notably branched distally; t.c. sub-cylindrical to ovate, 2—3 celled 12—14 µm long × 5—7.5 µm wide	Pale amber wall 1.58 µm thick; 15—34 × 12—22 µm (av. 21.5 × 16.6 µm)	10—12 × 5—6 µm; globule 3—4 µm dia.	Unilateral develop-ment; elongate, saccate, sometimes gibbose 17—65 × 5—20 µm; rhizoids bushy, coarse	5 µm dia. with colorless or orange-colored globule
Phalaris arundinaceae, Liegnitz, 1883 Gerhardt (lectotype of *Physoderma gerhardti* Schroeter)	Scattered fusiform or ovate brown spots, dispersed along whole leaf blade and sheath occasionally fusing to form dark dark patches; 3—5 mm long by up to 1.0 mm wide	—	Pale amber wall, 1.44 mm thick variable in size and shape, 14.0—31.5 × 10.5—31.5 µm (av. 22.4 × 16.5 µm)	—	—	—

Table 2. Results of cross-inoculations of certain grasses using *Physoderma* from *Phalaris* and *Agropyron* as inocula

Physoderma inoculum source	Hosts	Epibotic stage	Endobiotic stage	Resting spores	T/P[a]
Phalaris arundinacea Swale, Cheboygen Co., Mich.	*Agropyron repens*	+	+	+	3/16
	Glyceria striata	−	−	−	1/4
	Elymus canadensis	+	+	+	4/15
	E. caput-medusae	+	+	+	1/3
Agropyron repens. Same site (Sparrow and Griffin, 1964)	*Phalaris arundinacea*	+	+	+	11/45
	Glyceria striata	+	−	−	10/37
	Elymus canadensis	+	+	+	5/22
	E. caput-medusae	+	ptc[b]	−	2/8

[a] T = number of trials; P = number of host plants
[b] Primary turbinate cell

data previously obtained on the *Physoderma* parasite of *Agropyron repens* found in the same locality (Spar-row et al., 1961). It should be noted that contrary to a previous report (Sparrow and Griffin, 1964), resting spores of the *Agropyron* parasite were formed in *Phalaris arundinacea* but only when young vigorous adventitious host sprouts were substituted for seed-lings. The latter in our hands consistently died during the time necessary for complete resting spore ma-turity.

The susceptability of *Elymus* spp. to infection is interesting since these hosts were also infected by the *Agropyron* parasite. On the other hand, too much significance should not be attached to the lack of in-

fection in *Glyceria*, since seedlings were not available. The most significant fact is that the two paratites (one on *Phalaris arundinacea* and the other on *Agropyron repens*) living in the same swale infect each other.

DISCUSSION

An examination of Table 1 will reveal that from its morphology the *Physoderma* on *Phalaris arundinacea* is very similar, indeed, to that found on *Agropyron repens* collected at the same site (Sparrow et al., loc. cit.). Variations in host symptoms here are not regarded as unduly important (Sparrow and Griffin, 1964). No significant differences in shape or size are to be found in the epibiotic stage, both having similarly shaped sporangia which are developed in unilateral fashion. The *Agropyron* parasite, however, produces some sporangia with orange pigmented globules in their zoospores, a feature of as yet unknown significance and found in several other taxa of the genus where zoospores with colorless globules are also found (*P. macular*, Clinton, 1902; *P. lycopi*, Sparrow, 1957; *Physoderma* on *Asclepias*, Sparrow and Johns, 1970). In their endobiotic system it may be seen that there is also great similarity. It was found, however, that the distal parts of the rhizoidal system of the *Phalaris* parasite became more subdivided into delicate branches of limited extent than was usually observed in the *Agropyron* fungus. No significant differences are to be found in the resting spores, especially as to size, and in particular, wall thickness. Indeed, it will be noted that the same may be said for resting spores of what has been here selected as the lectotype of Schroeter's (1885, 1897) *Physoderma gerhardti* on *Phalaris*, namely Gerhardt's collection of 1883 (Fig. 2).

It will be recalled that Schroeter based his species primarily on material of *Phalaris arundinacea* collected by Gerhardt in the "Schwarzwasserbruch bei Liegnitz, 12.10.83". He also declared that *Physodermas* found on *Glyceria aquatica (maxima)* at Liegnitz and near Breslau, and on *Alopecurus pratensis*, belonged to his new species. No cross-inoculation studies were evidently made to support the identity of the fungi on *Phalaris* and *Glyceria* nor were any made by subsequent collectors of "*Physoderma gerhardti*", save Koch (1966). This investigator found a *Physoderma* on *Glyceria maxima* in Denmark which he identified with Schroeter's species. He secured resting spore germination but was unable to transmit his fungus from *Glyceria* to *Phalaris arundinacea, Alopecurus pratensis, Dactylis glomerata* and *Glyceria fluitans*. If Schroeter's taxon is to include species on *Glyceria* and other grasses, proof from host range studies should be forthcoming. Our own failure to infect *Glyceria* to resting spore

stage is not, as indicated, considered of much significance since in the few trials the host species used was not the same as Schroeter's, and obscure mature host parts were employed, not seedlings. Further cross-inoculation studies using *Glyceria* and *Phalaris* should be attempted. This is important since a considerable amount of material on *Glyceria* spp. has been distributed as *Physoderma gerhardti* by European collectors (for example, Krieger, "Fungi saxonici" No. 592, etc.) and is found in various exsiccati.

It is believed that the two *Physodermas* found in the same swale, one on *Phalaris*, the other on *Agropyron* (Sparrow et al., 1961) relate to the same taxon. They are both referred to Schroeter's *P. gerhardti* on *Phalaris arundinacea*. We cannot, of course, ever be absolutely sure of this identity since there is no living material from Gerhardt's 1883 collection. A study of *Physodermas* from his collecting site — if it still exists — might prove informative. It is most fitting that a species of this genus should comemmorate one who was so keen a field collector of these fungi.

Physoderma gerhardti Schroeter Emend.
In: Cohn. Kryptogamen-Flora von Schlesien 3(1), 194 (1885)

Epibiotic sporangia unilateral in development, at maturity procumbent, elongate, saccate or slipper-shaped, occasionally gibbose; a thick-walled variously expanded hemispherical portion of the zoospore cyst at one end near the point of origin of the short stubby, delicate or more often coarse, sometimes apophysate rhizoidal system, the opposite end terminated by a somewhat blunt, broad discharge papilla; variable in size, $17-100 \times 5-27$ μm; zoospores globose at discharge $4-5$ μm in diameter with a single colorless or orange-colored globule and $20-25$ μm long posterior flagellum, forming a temporary motionless mass at the discharge orifice before individually dispersing, when swimming, body more fusiform and $6-7 \times 4-5$ μm; internally proliferous. Endobiotic system composed of delicate rhizoids which are moderately or richly branched distally and which extend through many cells of the host, bearing occasional subcylindrical to ovate $0-3$-partitioned turbinate cells $10-20$ μm long $\times 5-10$ μm wide. Resting spores somewhat ellipsoidal or occasionally irregular and medianly constricted in outline, usually slightly flattened on one surface with a pale to rich bright amber-colored to brown wall $1.44-1.58$ μm thick; contents bearing a large central vacuole surrounded by numerous small bright globules; variable in size sometimes in the same host cell, sometimes according to tissue of host, inclusively $14-35 \times 10.5-24.5$ μm, formed at the tip of a lateral outgrowth of a segment of the turbinate

cell; germination upon the circumscissile dehiscence of a broad cap of wall material which is displaced by the slow elongation and protrusion of a thin-walled endosporangium from the spore body; the endosporangium somewhat broad and finger-like, 13—23 μm long by 12—22 μm at base, bearing at maturity a prominent apical discharge papilla 6—10 μm broad and 16—48 uniguttulate zoospores; the latter emerging passively through an orifice formed upon deliquescence of the papilla, spherical at first and 6—7 μm in diameter with a single colorless globule and basal granule; when swimming, the body more elongate and 10—12×5—6 μm, the posterior flagellum 30 μm long.

Lectotype, des. mihi:

Parasitic in *Phalaris arundinacea* L. Coll. Gerhardt "Bruche bei Liegnitz, 12.10.83" (see also Schroeter, "Pilze Schlesiens" No.281), Schlesien, Poland, In: Herb. Inst. Bot. Univ. Wratislavensis.

Michigan material: On *P. arundinacea* L., July 13, 1962 (coll. F.K.S.) et seq.; on *Agropyron repens* (L.) Beauv., June 14, 1960 (coll. R. M. Johns), et seq.; swale, Sect. 23, Hebron Township, Cheboygan Co., Mich., U.S.A. In: Univ. Michigan Herbarium.

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