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A COMPARATIVE STUDY OF THE BLACK STEM FUNGI ON LUCERNE AND RED CLOVER AND THE FOOTROT FUNGUS ON PEA[1]

Een vergelijkend onderzoek van de in associatie met voetrot optredende Phoma*-achtige schimmels bij luzerne, rode klaver en erwt*

G. H. BOEREMA, MARIA M. J. DORENBOSCH and LIDY LEFFRING[2]
Plant Protection Service (P.D.), Wageningen

North American (U.S.A., Canada) and Dutch strains of the black stem and footrot organisms were compared *in vitro* and some amplifying inoculation experiments were made. It appears that black stem of lucerne is associated in nearly all cases with one relatively uniform pycnidial fungus. From comparison with *Phoma herbarum*, the type species of the form-genus *Phoma*, it is concluded that the fungus, although quite different from *P. herbarum*, belongs to *Phoma* and not to *Ascochyta*. The correct name is therefore considered to be *Phoma medicaginis* Malbr. & Roum. and not *Ascochyta imperfecta* or *P. herbarum* f. *medicaginum*. Black stem of red clover is in general caused by a similar pycnidial fungus which, however, *in vitro* can always be distinguished from the lucerne fungus by its greater variability. This organism, the current name of which is *Phoma trifolii*, appears to be identical with the well-known footrot fungus of pea: *Ascochyta pinodella*. It is proposed to designate it as *Phoma medicaginis* var. *pinodella* (Jones) Boerema comb. nov. Incidentally *P. medicaginis* can be associated with black stem of red clover and *P. medicaginis* var. *pinodella* with black stem of lucerne. Inoculation experiments confirmed the pathogenicity of both organisms to lucerne and red clover. On pea only the variety *pinodella* has been found, although inoculation experiments proved that *P. medicaginis* can also attack pea.

INTRODUCTION

Each year many samples of lucerne, red clover and pea with footrot and leafspots are received by the diagnostic department of the Plant Protection Service at Wageningen. With both types of symptom there are always associated pycnidial fungi. Although these fungi are described in the literature as three different species, they do not show obvious differences *in vivo* and also show much resemblance *in vitro*. Our problem was to determine whether these fungi were identical or not.

REVIEW OF THE LITERATURE

The fungus on lucerne is usually described as *Ascochyta imperfecta* Peck (1912), that on red clover as *Phoma trifolii* E. M. Johnson & Valleau (1933) and that on pea as *Ascochyta pinodella* L. K. Jones (1927). In phytopathological literature all these fungi are characterized as weak parasites, needing certain unfavourable conditions to harm the plants.

In the United States comparisons have already been made between the fungi from lucerne and red clover, on which hosts the diseases are called "black stem" or "spring black stem". The results were rather conflicting.

[1] Accepted for publication 2 December, 1964.
[2] Present address: Floriculture Experiment Station, Aalsmeer.

JOHNSON & VALLEAU (1933) at first considered the causal organisms of both red clover black stem and lucerne black stem to be the same. On closer examination they observed differences between the two fungi and placed them in separate species. SCHENCK (1955) and SCHENCK & GERDEMANN (1956) also concluded that the lucerne fungus and the red clover fungus were distinct, though apparently closely related. Their lucerne isolates were always uniform in cultural appearance, whereas the red clover isolates were highly variable. Further they found small differences in pycnidial production, spore germination, growth rate and

TABLE 1. Origin, current names and type-indications of the isolates tested.
Herkomst, namen en type-aanduidingen van de onderzochte isolaties.

Isolate indication – origin		
American Type Culture Collection, Washington, D.C., U.S.A.		
Nr. 12087	*Medicago sativa*; cult. by HELEN S. Yu, U.S.D.A. 1955	
Centraalbureau voor Schimmelcultures, Baarn, the Netherlands		
	Medicago sativa	
	Trifolium pratense (stem)	
	Trifolium sp.	
Dr. M. F. KERNKAMP (via Dr. D. ROY WILCOXSON) Univ. Minnesota, U.S.A.		
Mi-A-3	*Medicago sativa*;	Michigan
M-A-40	id. ;	Minnesota
Io-A-8	id. ;	Iowa
SD-A-5	id. ;	South Dakota
M-A-43	id. ;	Minnesota
W-A-3	id. ;	Wisconsin
M-RC-11	*Trifolium pratense*;	Minnesota
Io-RC-1	id. ;	Iowa
Ill-RC-6	id. ;	Illinois
M-RC-10	id. ;	Minnesota
O-RC-1	id. ;	Ohio
Dr. H. W. MEAD, Res. Station, Saskatoon, Saskatchewan, Canada		
Nr. 48	*Medicago sativa* (seed);	Saskatchewan-1958
Nr. 58	id. (seed);	Saskatchewan-1958
Nr. 68	*Medicago sativa* subsp. *falcata* (stem);	Alberta-1959
Nr. 72	*Medicago sativa* (stem);	Brit. Columbia-1959
Own isolates from various locations in the Netherlands		
$L_1 - L_{12}$	*Medicago sativa* (stem, leaf and seed)	
$K_1 - K_7$	*Trifolium pratense* (stem, leaf and seed)	
$E_1 - E_{38}$	*Pisum sativum* (stem, leaf and seed)	

Code van de onderzochte isolaties – herkomst

De onderste kolom heeft betrekking op Nederlandse isolaties; stem = *stengel,*

spore septation. There was, however, no significant difference in spore size between the two isolates. All isolates were more pathogenic on the host from which they were originally isolated than on others.

ELLINGBOE & KERNKAMP (1957) and ELLINGBOE (1958, 1959a), on the other hand, considered the fungi causing black stem of lucerne and red clover to be identical. In their comparative study of many monoconidial isolates from both hosts, collected from eight states in North-America, they found the same variability in cultural type, radial growth, conidial production, spore septation

rent name	Cultural type (see text)
Ascochyta imperfecta	I
Ascochyta imperfecta	I
Phoma trifolii	II
Phoma trifolii	I
oma herbarum var. *medicaginis*	I
id.	I
id.	I
id.	I
id.	I
id.	I
id.	I
id.	I
id.	II
id.	II
id.	II
Ascochyta imperfecta	I
id.	I
id.	I
id.	I
Ascochyta imperfecta	Mostly I Once II
Phoma trifolii	Mostly I Once I
Ascochyta pinodella	Always II
identificeerd als	*Cultuurtype (zie tekst)*

1 = *zaad*, leaf = *blad*, mostly = *meestal*, once = *eenmaal*, always = *altijd*.

and spore size. EDMUNDS (1958) and EDMUNDS & HANSON (1957, 1960) also considered that only one pathogen is involved in black stem of lucerne and red clover. But they found, as did SCHENCK & GERDEMANN (l.c.), that the red clover isolates studied were more variable in cultural characteristics and in ability to produce pycnidia than the lucerne isolates. Their lucerne isolates attacked lucerne more severely than red clover, whereas most isolates from red clover were less pathogenic on both lucerne and red clover than were lucerne isolates. By inoculation experiments the fungus was proved to be pathogenic also to other Leguminosae.

These different opinions between American authors have also led to differences in nomenclature. This question will be discussed at the end of this paper. In these American studies, no European isolates were available for comparison. In Europe the fungi on lucerne and red clover are usually referred to two different species, but no comparisons are known in the literature. In contrast to the situation in the United States the clover disease is not regarded as of economic importance in Europe. For this reason only few data are available on European isolates from red clover.

Comparison of *Ascochyta pinodella* from pea with the black stem fungi of lucerne and clover is not known in the literature. Only SPRAGUE (1929) with his comparative study of leguminous *Ascochyta*'s observed that the growth rate of lucerne isolates is rapid, like that of *A. pinodella*.

MATERIALS AND METHODS

Comparison of the organisms

Our own isolates from the three hosts were compared (Table 1) with isolates obtained from culture collections at Washington and Baarn and from Dr. M. F. KERNKAMP (U.S.A.) and Dr. H. W. MEAD (Canada). The isolates were studied in plate cultures at room temperature on five different media, cherry agar (300 ml juice of 500 g cherries + 1300 ml H_2O + 27,5 g agar), potato glucose agar, oat agar, malt agar and Conn's agar (for recipes see AINSWORTH, 1961).

Pathogenicity tests

Three of our own isolates from lucerne, red clover and pea were used for comparative inoculations on the three plant species. In the first test the roots of the plants were inoculated with these isolates to obtain footrot symptoms, while in the second test the leaves were inoculated. The re-isolates from the leafspots obtained in the second test were used for root inoculation in a third test.

The material for the root inoculations was prepared by fragmenting sporulating plate cultures on cherry agar with tap water in a food blendor. The root systems of four-week old seedlings were washed and then dipped in this suspension. The seedlings were then potted in sterile soil and kept at room temperature in a glasshouse. Each of the two root inoculation tests included 20 lucerne, 20 red clover and 20 pea plants for each of the isolates and a control treated with a suspension of agar in water. In transplanting and washing the root systems, the plants were slightly damaged but this was not considered to be any disadvantage in obtaining comparative results with these weak parasites.

The material for the leaf inoculations was prepared by shaking dislodged

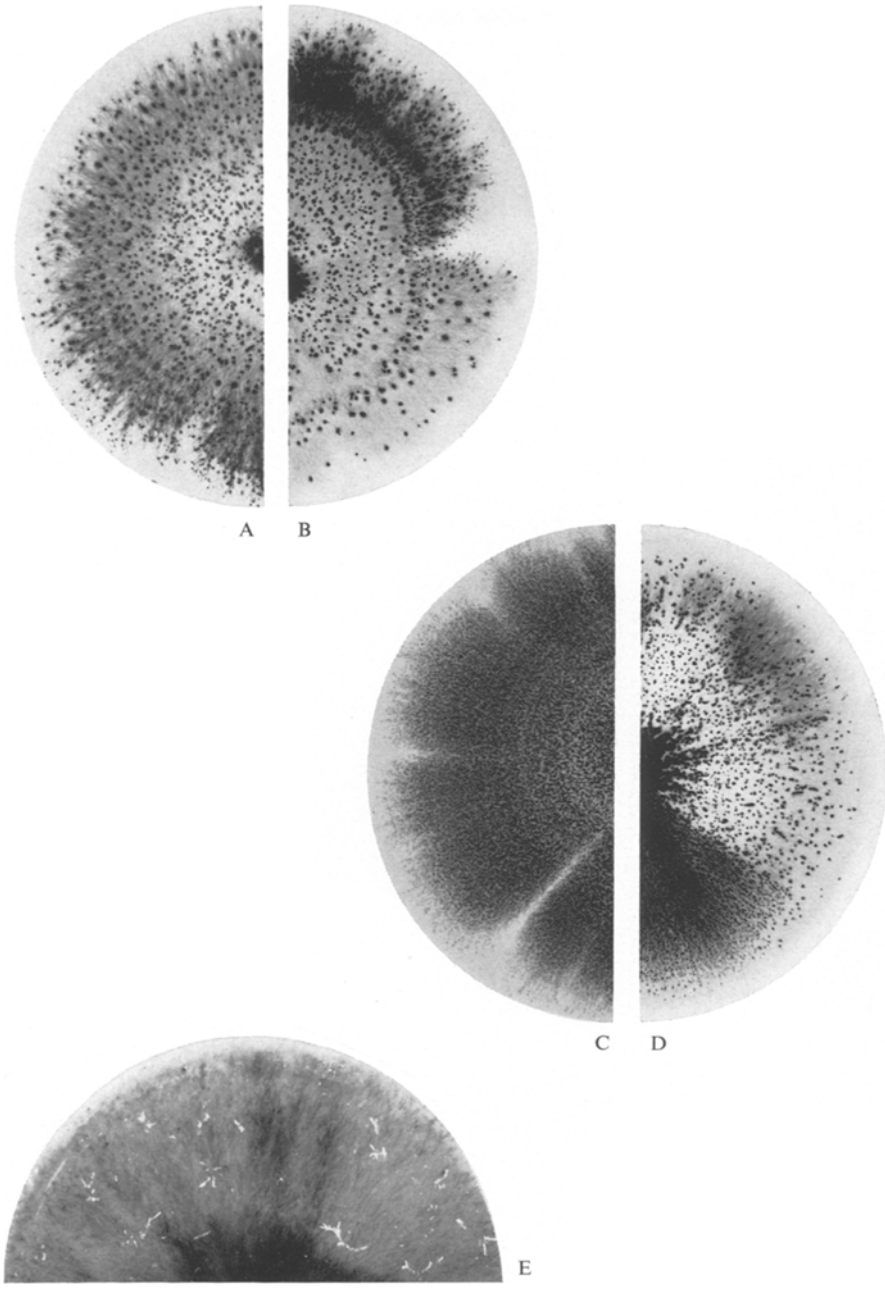

FIG. 1. Fungus-type I *(Phoma medicaginis)*.
A, B, C, D. Four different strains after three weeks' growth on cherry agar. / *Vier verschillende stammen gefotografeerd na een groei van drie weken op kers-agar.*
E. Production of crystals on malt agar. / *Vorming van kristallen op mout-agar.*

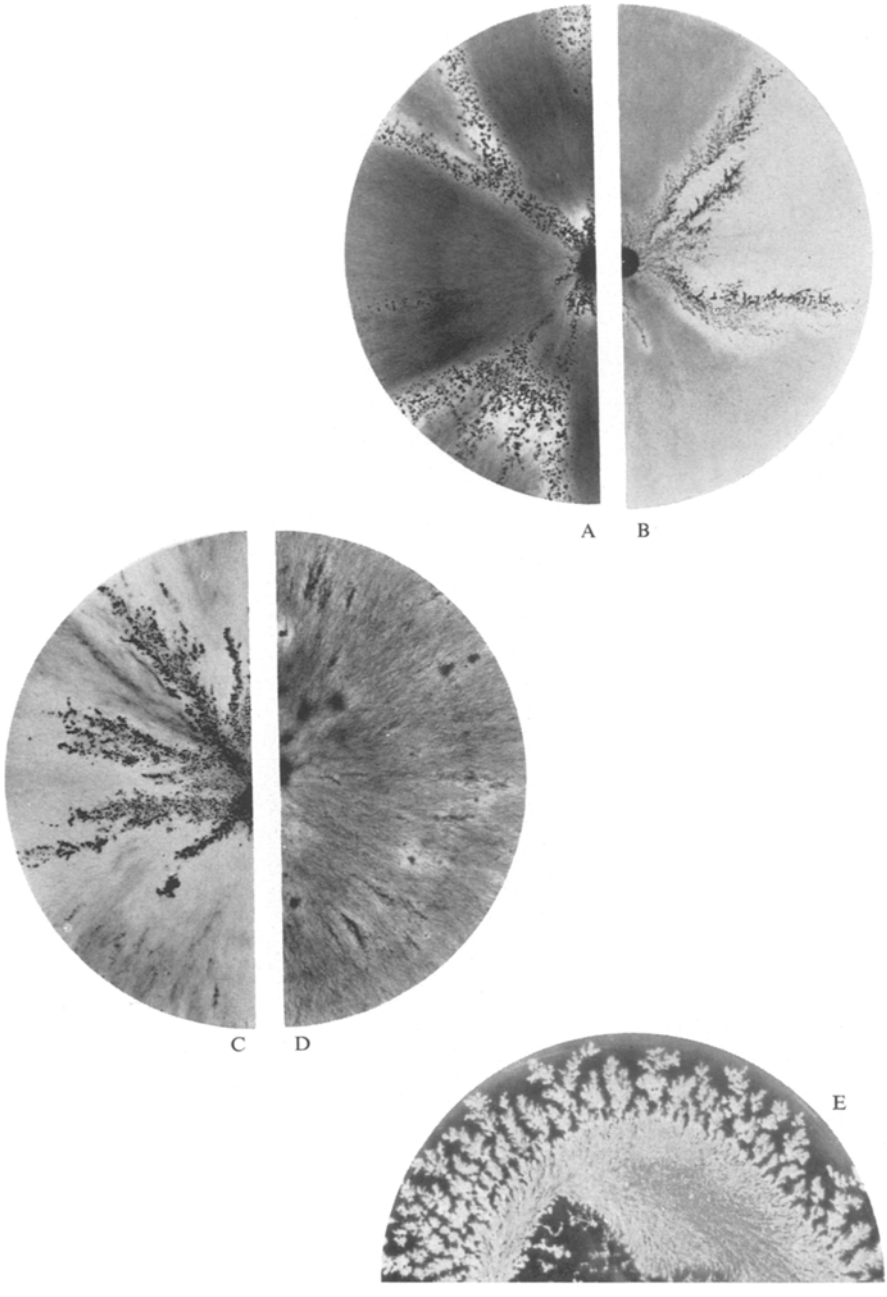

FIG. 2. Fungus-type II *(Phoma medicaginis* var. *pinodella)*.
A, B, C, D. Four different strains after three weeks' growth on cherry agar. / *Vier verschillende stammen, gefotografeerd na een groei van drie weken op kers-agar.*
E. Production of crystals on malt agar. / *Vorming van kristallen op mout-agar.*

ripe pycnidia in tap water. The suspension was sprayed with an atomizer on to 30 fresh detached washed leaves of the plant species tested. The controls were treated with pure tap water. The inoculated leaves and the controls were placed on moistened filter paper in sterile Petri plates kept in the dark at 10°C.

RESULTS

Comparison of the organisms

It was observed with the naked eye, that the various isolates could be grouped into two rather distinct cultural types:

I. Isolates very uniform in cultural appearance on all the media used[3] (Fig. 1)

In this group a flat grey felty mat is produced. It darkens gradually, sectorially or in concentric zones, until carbonaceous. Numerous relatively large pycnidia are regularly scattered over the mat. On malt agar after about a month some small crystals occasionally appear in the medium (Fig. 1e).

II. Isolates very variable in cultural characteristics and in ability to produce pycnidia (Fig. 2)

These isolates usually produce many different sectors, *e.g.* sectors with predominantly dark or white mycelial growth with only occasional small pycnidia and sectors with less copious aerial mycelium and abundant small pycnidia. In general these isolates are faster growing than those of type I. On malt agar these isolates always produce abundant crystals arranged in a fan shaped pattern (Fig. 2e).

Microscopic examination showed that the isolates of both cultural types produce dark coloured thick-walled chlamydospores, either singly or in chains. With the isolates of type I, however, these chlamydospores are found only in old cultures, whereas in cultures of type II they are already present after a few days. The mycelium of both types is microscopically very similar. In both types the pycnidial primordium arises meristogenously. Usually a single hypha gives rise to the primordium, but occasionally adjacent hyphae take part in the formation (simple and compound meristogenous origin, cf. KEMPTON, 1919). The structure of the pycnidial wall in type I differs somewhat from that of type II. In type I one can speak of "*textura globulosa*"; in type II one finds wall structures intermediate between the globulosa texture and the "*textura prismatica*" as defined by STARBÄCK (1895). The pycnidiospores are in both cases produced by a budding of the meristogenous cells of the inner pycnidial wall. The mean spore size of both cultural types does not differ significantly on the various media. Most spores are non-septate. The percentage of 1-septate spores, always low, varies considerably, however. It was independent of the cultural type and apparently influenced by the medium.

The relationship between the two cultural types and the plants from which the isolates originated is shown in Table 1. It appears that nearly all isolates of type I originated from lucerne. Three exceptions concerned red clover. The isolates of type II were all from red clover or pea, with only one exception from lucerne. It was impossible to see any difference between isolates of type II from red clover or pea.

[3] In spite of this narrow variability, the isolates, of course, show small differences (Fig. 1a-d).

Pathogenicity tests

In the comparative experiments with inoculations of the root systems, peas always showed symptoms (Table 2). These consisted of a yellow discoloration of the lower leaves and a brownish-black discoloration of the root collar and stem base. Also a large part of the root systems showed a dark discoloration.

The results of the first experiment with root inoculations were rather disappointing for lucerne and red clover. Hardly any symptoms were observed (Table 2). In the second root inoculation experiment the same fungus strains were used but this time after one passage through the leaves of the plant species tested. In this case also lucerne and red clover showed clear symptoms (Table 2), which were very similar to the symptoms on pea, *viz.* yellow lower leaves and brownish-black stems and roots. In both experiments re-isolation was always possible when symptoms appeared. No differences were observed in symptoms caused by the different strains. The untreated plants remained healthy in both experiments.

TABLE 2. Results of the two root inoculation experiments a and b. In the second experiment (b) the fungus strains were used after one passage through the leaves of the plant species tested.
— = not attacked, + = attacked, ++ = very severely attacked
Resultaten van twee wortel-inoculatieproeven a *en* b *op luzerne, rode klaver* (red clover) *en erwt* (pea). Untreated = *onbehandeld. Bij de tweede proef* (b) *werden de stammen (strains) gebruikt na „passage" door bladeren van de toetsplanten.*
— = *niet aangetast*, + = *aangetast*, ++ = *zwaar aangetast*

	Strain L[2] Type I (from lucerne)		Strain K[5] Type II (from red clover)		Strain E[7] Type II (from pea)		Untreated	
	a	b	a	b	a	b	a	b
Lucerne	—	+	—	+	—	+	—	—
Red clover	+	+	—	+	—	+	—	—
Pea	+	++	+	++	+	++	—	—

The comparative experiments with inoculation on leaves always gave clear symptoms; these showed small differences on pea, clover and lucerne. However, on the same plant species no differences were observed between the three strains of the inoculum. In all cases re-isolations were possible. Some of these were used for the second root inoculation experiment.

DISCUSSION AND CONCLUSIONS

It appears that in general only one fungus type (cultural type I) is responsible for black stem of lucerne in North America (U.S.A. and Canada) and in the Netherlands. This type is characterized by its uniform appearance in culture (Fig. 1) and a delayed production of chlamydospores and crystals. With black stem of red clover, on the other hand, another fungus type (cultural type II) is usually present both in the United States and in the Netherlands. In contrast to type I on lucerne this type is characterized by a very variable appearance in culture (Fig. 2) and a quick, abundant production of chlamydospores and crystals.

In rare instances type I was isolated from red clover and type II from lucerne (Table 1). This corresponds with the fact that in experiments on lucerne and red clover black stem symptoms could be obtained with both types.

Ascochyta pinodella, the well-known footrot fungus of pea, was further found to be identical with the above mentioned type II from red clover. Type I was never isolated by us from pea; nevertheless by inoculation this type also induced the characteristic footrot and leafspot symptoms on pea. Apart from the different cultural appearance and the somewhat larger pycnidia of type I, both types are morphologically very similar *e.g.* with regard to characteristics of the mycelium, chlamydospores, pycnidia and pycnidiospores.

In their researches on black stem of lucerne and red clover most American authors, also, have distinguished between the two above mentioned cultural types. JOHNSON & VALLEAU (1933), SCHENCK (1955) and SCHENK & GERDEMANN (1956) apparently always isolated type I from lucerne and type II from red clover. They therefore concluded that the lucerne fungus and the red clover fungus are two different species. EDMUNDS & HANSON (1960) also separated the two cultural types in correlation with the two hosts (see also EDMUNDS, 1958), but apparently from pathogenicity tests they concluded that only one pathogen was involved. Only ELLINGBOE & KERNKAMP (1957) and ELLINGBOE (1958, 1959a,b) found no correlation between cultural types and the host from which they were isolated. In our opinion this may have been due partly to the variability of type II, which in some cases can be isolated from lucerne, just as type I sometimes occurs on red clover. Possibly, also, in concentrating on the small differences within type I these authors did not notice the characteristic differences between types I and II[4]. In any case it is a fact that of the strains received from Dr. KERNKAMP (Table 1) the lucerne isolates were all of type I and the red clover isolates, with only two exceptions, of type II.

All the American authors mentioned observed, as we did, that the isolates from lucerne and red clover could attack both plant species. Most of them, however, observed a great variability in pathogenicity between their isolates and in general the isolates were more pathogenic on the host from which they were isolated. It therefore seems probable that examination of a greater number of Dutch or other European strains might reveal more variation in pathogenicity than our inoculation experiments have done (see also the English work on the black stem of lucerne by TOOVEY, WATERSTON & BROOKS, 1936). The method of inoculation, of course, also plays an important role with these weak parasites. Further, MEAD & CORMACK (1961) observed that isolates from lucerne (cf. Table 1) could attack pea and this accords with our own results.

Summarizing we may draw the following conclusions:

1. In general the black stem diseases of lucerne and red clover are caused, both in the Netherlands (and perhaps in the whole of Europe) and in America (U.S.A. and Canada) by two different, though very similar, organisms.
2. The common black stem organism of red clover is identical with *Ascochyta pinodella* from pea.
3. Both organisms are variable in pathogenicity, but in general they can attack lucerne and red clover as well as pea.

[4] Compare the variants mentioned and illustrated by CORMACK (1945), which in our opinion relate to type I. The variability of type II is of an altogether different order.

Taxonomic considerations

The first question is whether the organisms are two different species or two forms or varieties of one and the same species. It is known that imperfect stages of non-related organisms may show considerable morphological similarity. It was because of this that SCHENCK & GERDEMANN (1956) did not want to decide whether they dealt with one variable species or with two different species. We agree that the system of classification of the Deuteromycetes is wholly artificial but it is a necessary system for practical morphological identification and we must use it in accordance with its rules. In our opinion therefore we have to consider the two cultural types as varieties of one and the same fungus imperfectus because no substantial morphological differences were observed.

A second question is the proper generic position of the fungus. Looking at the existing generic definitions of the Sphaeropsidales with hyaline spores it is impossible to decide to what form-genus this organism properly belongs. The original diagnoses of the relevant genera are often based on unstable characteristics such as the number of septa in the spores (*e.g.* non-septate as against one-septate) and the substrate on which the fungus occurs *in vivo* (*e.g.* leaf or stem). This explains why this fungus through the years has been assigned to several different genera. To make a justifiable choice we have compared the fungus with the type species of the form-genera concerned, viz. *Phoma* Sacc., *Phyllosticta* Pers. ex Desm., *Ascochyta* Lib. and *Diplodina* West. As criteria we used the structure of the pycnidia, the mode of origin of the pycnidiospores and the nature of the septa.

Phyllosticta convallariae Pers., the type species of *Phyllosticta*, produces relatively large conidia (ca. 8×10–$12\ \mu$) on small, stick-like sporophores which soon disappear[5]. In our fungus, however, the small conidia are produced by budding of the meristematic cells of the inner pycnidial wall. The type species of *Diplodina*, *D. salicis* West., produces pycnidia which open by splitting so that the pycnidia become cupulate. Our fungus, on the contrary, has pycnidia with a predetermined opening or ostiole. Moreover, in *D. salicis* the spores originate on true sporophores[6].

In *Ascochyta pisi* Lib., the type species of *Ascochyta*, only the fully mature fruit-bodies have an opening; this opening may be called a p o r u s in contrast to the predetermined opening or o s t i o l u m of our fungus. The spores in *A. pisi* are not produced by a budding process as in our fungus, but originate as protrusions which are detached by the development of a septum. This was established by means of electron microscope studies (BREWER & BOEREMA, 1965). Differences in the type of septation are also important. Electron microscope observation has shown (BREWER & BOEREMA, l.c.) that the usually two- or more-celled spores of *A. pisi* are "distoseptate" as defined by LUTTRELL (1963), which means that the cells in the conidia are surrounded by individual saclike walls as in the conidia of *Helminthosporium*. As against this the two-celled spores of our fungus appear "euseptate" *sensu* LUTTRELL (l.c.); i.e., the septum consists of a diaphragm

[5] It agrees with the characteristics of the form-genus *Phyllostictina* Syd., cf. PETRAK & SYDOW (1927:210). This should mean that *Phyllostictina* (1916) is a later synonym of *Phyllosticta* (1818).

[6] *D. salicis* is identical with the type species of *Discella* Berk. & Br.: *Discella carbonacea* (Fr.) Berk. & Br., cf. GROVE (1937: 148). This means that *Diplodina* (1857) is a later synonym of *Discella* (1850).

merging peripherally with the lateral walls. Taking into account all these fundamental differences we concluded that our fungus does not belong to the genus *Ascochyta*.

Phoma herbarum West. (1852), the type species of *Phoma* (BOEREMA, 1964), has many features in common with our fungus. The structure of the ostiolate pycnidia and the mode of origin of the pycnidiospores in *P. herbarum* agree well with those of our fungus. *P. herbarum* generally produces only one-celled spores, but two-celled spores can also be found and those have proved to be "euseptate" (BREWER & BOEREMA, 1965). Therefore it is concluded that our fungus belongs to the genus *Phoma*.

The next point to consider is that of the correct specific epithet. The oldest names of the fungus all concern the host lucerne. For the synonymy of the fungus on this host we can refer to SCHENCK & GERDEMANN (1956) and EDMUNDS & HANSON (1960). The oldest valid name appears to be *Phoma herbarum* West. f. *medicaginum* Rabenhorst (1862, see the list of synonyms). However, the present knowledge of *Phoma herbarum* (BOEREMA, 1964) indicates that this is quite a different species. Transfer of the forma "*medicaginum* Rab." to the species-rank is not possible, because that would be a later homonym of the next oldest name of the fungus: *Phoma medicaginis* Malbr. & Roum. (1886, see the list of synonyms). It is therefore concluded that the combination made by MALBRANCHE & ROUMEGUÈRE is the correct valid name of the fungus. In general on lucerne only type I of the fungus as described above occurs. We think the name *P. medicaginis* (and its synonyms on lucerne) should obviously be applied to this type (type I). Consequently type II can be considered as a variety of *P. medicaginis* and we must use for it the earliest available epithet. This proved to be "*pinodella*" of *Ascochyta pinodella* Jones (1927), described from pea. In accordance with the fact that only type II has been found in the field on pea, the description of the type culture of *A. pinodella* given by WEHLBURG (1932) makes it clear that this name concerns type II. The same holds true for *Phoma trifolii* E. M. Johnson & Valleau (1933).

The synonymy of the fungus is then as follows:

PHOMA MEDICAGINIS Malbr. & Roum. [var. *MEDICAGINIS*]
in Roum., Fungi gall. exs. (Cent. 37), Nr. 3675. 1886;
in Rev. mycol., Toulouse 8:91. 1886

Syn.[7]: *Phoma herbarum* West. f. *medicaginum* West. in Rab., Fungi europ. exs. ed. nov. ser. 2 (Cent. 5), Nr. 455b. 1862 [= Ausg. 3]; ref. SCHLECHTENDAL in Bot. Ztg. 20:199. 1862 [often cited as *P. herbarum* var. *medicaginis*].

:*Diplodina medicaginis* Oud. in Ned. Kruidk. Arch. ser. 3, 2: 884, 885. 1903.

:*Ascochyta imperfecta* Pk. in Bull. N.Y. St. Mus. 157:21. 1912.

Habitat:
Associated with footrot and leafspots on *Medicago sativa* L. ("black stem disease"). Incidentally also on other Leguminosae.

[7] EDMUNDS & HANSON (1960) also included in the synonymy "*Phoma medicaginis* Fckl. (1869)" and *Ascochyta pisi* Lib. var. *medicaginis* Sacc. (1920). The name ascribed to FUCKEL does not exist. FUCKEL "1869" (1870) only made the combination *Pleospora medicaginis*, a perfect stage, and remarks: "Fungus pycnidium: *Phoma herbarum* Westend. Form. *Medicaginis* in Rbh. F. eur. 455.b.". However, such a connection with a perfect stage has not been proved. Examination of original material of *A. pisi* var. *medicaginis* in the SACCARDO-herbarium at Padova (collected by VERNON & SIMMONS at Wendover, Wyoming, U.S.A.) proved this to be a different species.

PHOMA MEDICAGINIS var. *PINODELLA* (L.K. Jones) Boerema comb. nov.

Basionym: *Ascochyta pinodella* L.K. Jones in Bull. N.Y. St. agric. Exp. Sta. 547:10. 1927.
Syn.: *Phoma trifolii* E.M. Johnson & Valleau in Bull. Ky agric. Exp. Sta. 339:73, 74. 1933.

Habitat:
Associated with footrot and leafspots (pod spots) on *Pisum sativum* L. and *Trifolium pratense* L. ("black stem disease"). Occurs also on other Leguminosae.

SAMENVATTING

Bij het mycologisch, diagnostisch onderzoek op de Plantenziektenkundige Dienst wordt men regelmatig geconfronteerd met voetziekteverschijnselen, respectievelijk bladvleksymptomen bij luzerne, klaver en erwt, waarbij *Phoma*-achtige schimmels zijn betrokken, welke morfologisch vrijwel niet van elkaar te onderscheiden zijn. Om de identiteit van deze schimmel(s) op te helderen werd een vergelijkend onderzoek ingesteld, waarbij behalve de morfologische kenmerken ook de totale habitus in vitro en de pathogeniteit werd onderzocht. Daar men in de Verenigde Staten van Noord-Amerika bij het vergelijkend onderzoek van de desbetreffende *Phoma*-achtige schimmels van klaver en luzerne tot geheel tegenstrijdige conclusies was gekomen, werden ook vele Amerikaanse schimmelisolaties van luzerne en klaver bij dit onderzoek betrokken (tabel 1).

Het bleek, dat de schimmelisolaties van de drie planten wat betreft de kenmerken van de pycniden en de sporen zeer veel overeenkomst vertonen. Op één uitzondering na waren de schimmelisolaties van luzerne echter steeds te onderscheiden door hun relatief regelmatige groei en het feit dat alleen in oude cultures chlamydosporen en kristallen worden gevormd (type I; zie tabel 1 en fig. 1). De schimmelisolaties van klaver en erwt waren op enkele uitzonderingen na gekenmerkt door een grote variatie van de groei in vitro en een snelle, sterke ontwikkeling van chlamydosporen en kristallen (type II; tabel 1 en fig. 2). Het was niet mogelijk hierbij twee groepen te onderscheiden in relatie tot hun herkomst van klaver of erwt.

Bij de pathogeniteitsproeven bleek dat typische isolaties van respectievelijk luzerne, klaver en erwt bij alle drie planten dezelfde symptomen kunnen veroorzaken, waarbij echter de erwt zich steeds het meest gevoelig toonde (zie tabel 2). Dit maakt het begrijpelijk dat de luzerneschimmel (type I) ook enkele malen werd geïsoleerd van klaver en dat de klaver-erwt-schimmel (type II) ook eenmaal werd geïsoleerd van luzerne (de bovengenoemde uitzonderingen, zie tabel 1). De schimmelisolaties van erwt waren zonder uitzondering van het type II.

Uit een en ander werd geconcludeerd dat wij hier het beste kunnen spreken van twee variëteiten van één en dezelfde schimmel. Vergelijking met de typesoort van het geslacht *Phoma* Sacc. leidde verder tot de conclusie dat deze schimmel thuishoort in het geslacht *Phoma*. De oudst geldige naam van de schimmel is dan *Phoma medicaginis* Malbr. & Roum. (syn. *Phoma herbarum* f. *medicaginum; Ascochyta imperfecta*), welke naam dus slaat op type I. Voorgesteld wordt de algemeen op klaver en erwt voorkomende variëteit van deze schimmel (type II) aan te duiden als *Phoma medicaginis* var. *pinodella* (Jones) Boerema comb. nov. (syn. *Ascochyta pinodella, Phoma trifolii*).

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