

# THE ECOLOGICAL AND THE TAXONOMIC PROBLEMS IN FUSARIA\*

BY C. V. SUBRAMANIAN

(University Botany Laboratory, Madras 5)

THE small contribution which I wish to make to this Symposium will deal with two aspects of the study of Fusaria in which I have been interested for some years past, viz., the ecology and the taxonomy of Fusaria, especially those which occur in soils.

## THE ECOLOGICAL PROBLEM IN FUSARIA

Much of the work on the ecological problem in the Fusaria has been aimed at elucidating the possible method of perennation of several soil-borne Fusaria. The work of Sadasivan (1939) on *Fusarium culmorum* gave an impetus to investigations along the same lines on vascular wilt Fusaria and it was shown that *F. vasinfectum*, the cotton wilt pathogen, like *F. culmorum*, possessed competitive saprophytic ability and could be classified as a soil-inhabitant (Subramanian, 1946). Further work on the occurrence of Fusaria in black cotton and other soils from southern India indicates that several other species of *Fusarium* can also be placed in the soil-inhabitant group. This list would include *F. chlamyosporum* Wr. and Rg., *F. avenaceum* (Fr.) Sacc., *F. equiseti* (Corda) Sacc., *F. scirpi* Lamb. and Fautr., *F. scirpi* Lamb. and Fautr. v. *acuminatum* (Ell. and Ev.) Wr., *F. scirpi* Lamb. and Fautr. v. *caudatum* Wr., *F. oxysporum* Schlecht., *F. javanicum* Koord., *F. javanicum* Koord. v. *radicicola* Wr., *F. solani* (Mart.) App. and Wr., *F. solani* (Mart.) App. and Wr. v. *martii* (App. and Wr.) Wr., *F. solani* (Mart.) App. and Wr. v. *minus* Wr., *F. solani* (Mart.) App. and Wr. v. *striatum* (Sherb.) Wr. and *F. caeruleum* (Lib.) Sacc. Stover's (1953) work indicates that *F. oxysporum* Schlecht. v. *cubense* (Smith) Wr. and Rg., which causes the panama disease of bananas exhibits a similar behaviour.

Whilst the soil-inhabitant type of behaviour appears to be the most common in this genus, a contrasting type of behaviour is shown by *F. udum* Butler which causes a vascular wilt of *Cajanus cajan* in India. That this species is distinct from *F. oxysporum* Schlecht. emend. Snyder and Hansen has been stressed by Subramanian (1955). *F. udum* has been shown

\* Paper read to the Indian Academy of Sciences at its Annual Meeting at Belgaum in December, 1954, at the Symposium entitled "Soil Micro-organisms and Plant Well-being".

(Agnihotrudu, 1954) to be incapable of colonising dead plant debris in soils and its saprophytic survival is limited to tissues originally invaded during its pathogenic phase—a behaviour similar to that of *Ophiobolus graminis*.

Work on saprophytic behaviour of soil *Fusaria* is obviously important from the point of view of control of the diseases caused by them and future work should be aimed at screening as many *Fusaria* as possible for their saprophytic behaviour in soils.

#### THE TAXONOMIC PROBLEM

One of the major difficulties encountered in all work with *Fusaria* is, indeed, their taxonomy. Notwithstanding the earlier work of Appel, Wollenweber, Reinking, Sherbakoff and others (see Wollenweber and Reinking, 1935) and the more recent studies of Snyder and Hansen (1940, 1941, 1945, 1954), the taxonomic problem of the *Fusaria* still remains. Wollenweber and Reinking's (1935) is, indeed, the most outstanding and authoritative treatise on this difficult subject and Wollenweber's monographic treatment (Wollenweber, 1931, 1943) brings together in an admirable manner exhaustive descriptions of the various taxa and also lists of synonymy, hosts, exsiccatae and distribution. Snyder and Hansen's revision of the genus is based essentially on the broader groupings of the genus proposed by Wollenweber and Reinking, and alterations are effected only with reference to the species concept. Both systems have their protagonists and it is not necessary here to dwell at length on the comparative merits and demerits of the two systems (for which please see Padwick, 1941). On the other hand, it would be worthwhile discussing possibilities of a newer approach to the solution of this old problem.

A common feature of the Wollenweber-Reinking and the Snyder-Hansen classifications and also the more recent revision by Gordon (1952, 1954) is the implicit faith laid in the fundamentals of the classification of the genus as enunciated nearly thirty years ago by Wollenweber, Sherbakoff, Reinking, Johann and Bailey (1925), a pre-requisite for taxonomic study of any given taxon being a pure culture derived from a single spore. It is necessary to grow such a single spore culture on several culture media and record their cultural characters at different stages of growth and also study the morphology before it could be placed in any known species. The procedure involved in specific identification is thus lengthy and complicated. Such a procedure would certainly be worthwhile if there is some hope of arriving at a correct specific identification. Unfortunately, this is seldom the case and the species concept of Wollenweber and Reinking is based on a mixture of morphological and cultural characters, of which the latter are highly variable and hence

are of little taxonomic value. The Snyder-Hansen system and also Gordon's revision both suffer from the same defects since the main modification effected is the reduction in the number of species on the basis of observations on the variability in cultures of the various taxa.

A different approach to *Fusarium* taxonomy was offered by the recent work of Miller (1945, 1946 *a*, 1946 *b*) in Canada who indicated the need to base the classification of the genus on a study of the 'wild type' in the various species. Miller's 'wild type' refers to a pure culture obtained immediately after isolation from the substratum. On the basis of a study of first cultures of a few isolates of *Elegans Fusaria* obtained from diseased muskmelon plants in Canada, Miller concluded that the 'wild type' in the genus *Fusarium* is the type that usually "occurs in nature as a form (or forms) that, when cultured on most artificial media, produces abundant aerial mycelium on which conidia, mostly non-septate, are borne rather sparsely" (Miller, 1945, p. 41). However, continued culturing, according to Miller, results in the loss of the 'wild type', being crowded out by mutants which are characterised by prolific sporulation (0-3- or more-septate conidia) and an adpressed type of growth with little or no aerial mycelium. Following up this hypothesis, Miller (1946 *a*, 1946 *b*) has further suggested that the traditional sporodochium in the genus *Fusarium* is synonymous with patch mutants encountered during his studies and, hence, has suggested that these should not be the basis for a sound classification of the genus. In my studies of first cultures of various parasitic *Fusaria* obtained from different genera of host plants and of saprophytic forms obtained from soils, no confirmation could be obtained of Miller's concept of a stereotyped and uniform 'wild type' (Subramanian, 1951). First cultures of both pathogenic and saprophytic *Fusaria* were not characterised by any uniform 'wild type', but fell into several intergrading types from the typically sporodochial or pionnotal to the typically mycelial. The *Fusaria* studied included *F. caeruleum* (Lib.) Sacc., *F. conglutinans* Wr. v. *citrinum* Wr., *F. culmorum* (W. G. Sm.) Sacc., *F. equiseti* (Corda) Sacc., *F. javanicum* Koord. and its variety *radicicola* Wr., *F. oxysporum* Schlecht., *F. scirpi* Lamb. and Fautr. and its varieties *acuminatum* (Ell. and Ev.) Wr., and *longipes* (Wr. and Rg.) Wr., *F. semitectum* Berk. and Rav., *F. solani* (Mart.) App. and Wr., and its varieties, *martii* (App. and Wr.) Wr., *minus* Wr., and *striatum* (Sherb.) Wr., and *F. vasinfectum* Atk. In a later investigation on several isolates of *Fusaria* obtained from corms of gladiolus plants showing yellows at Cambridge (England) during 1950, I found that these *Fusaria* exhibited a wide range of variation in cultural characters like amount of aerial mycelium colouration on different media, presence or absence of sporodochia and,

pionnotes, etc., but the morphological features of the conidia were remarkably constant. More recently, a similar wide range of variation in cultural characters has been reported in the case of first cultures of *F. udum* also (Subramanian, 1955). Here again the morphology of the micro- and macroconidia in the first cultures showed little variation compared to the cultural characters. Studies of this kind on 'wild types' or first cultures thus point to the fact that a given species may exhibit a wide range of variation in cultural characters but not in morphological characters relating to the conidia. Nevertheless, the 'wild types', as defined by Miller, are not of universal application in the genus *Fusarium* since they do not conform to any single definite pattern and since in the definition suggested by Miller the emphasis has been on cultural characters.

In my opinion, the solution of the problem warrants an entirely new approach. It would appear that too much emphasis has been laid in the past on artificial cultures of organisms in taxonomic studies and firm adherence to the idea that, for taxonomic purposes, organisms should be studied under strictly controlled nutritional levels and other conditions in the laboratory is not always essential. In the eagerness to obtain cultures, the natural material is seldom examined and more often than not a non-sporulating culture has been obtained by continued subculturing of the fungus in synthetic media. The fungi have then to be fed on special concocted foods to obtain a culture in *normkultur* or *hochkultur*. In the case of several vascular wilt Fusaria, the pathogen is confined essentially to the vascular tissues of the host during the earlier stages of pathogenesis, but gradually invades the outer tissues progressively with the death of the infected plants and, indeed, sporodochia and numerous conidia are produced only during the final phase of the disease, following death of plant parts. Thus, the *hochkultur* stage in most of these specialised vascular wilt Fusaria appears on host tissues killed by the fungus but not earlier, and attempts to obtain, on artificial media, the *hochkultur* of the fungus from invaded tissues during earlier phases of the disease have very often failed. The case of Fusaria which cause foot rots and root rots appears to be different in that the invasion of the host tissues is more general during early phases of the disease and the conidia are produced earlier too. In any case, the conidia, in all species of Fusaria, are produced in the infected tissues or substrata sooner or later and such conidia as occur on tissues invaded in nature should be considered normal for all taxonomic purposes. There is also the question of variation among pure cultures of Fusaria and the limitations of working with such pure cultures would be realised by all those who have keenly followed the work of Hansen (1938) on the dual phenomenon in the Fungi Imperfecti.

All this circumlocutory procedure can be avoided by studying *Fusaria* as they occur in nature. Systematic work and taxonomic studies on various other genera of the Fungi Imperfecti are being carried out with precision without the aid of artificial cultures and solely on specimens collected directly from the field and occurring on the natural substratum. Such a study of *Fusaria* as they occur in nature would obviate all limitations resulting from the use of artificial cultures; and, as for the possible criticism that, in nature, spores of more than one species may occur side by side, thereby vitiating specific determination and taxonomic groupings, it may be pointed out that no two spores which are morphologically indistinguishable, having, of course, regard to their origin and dispersal, need be considered as taxonomically different. Another possible objection to the procedure recommended here would be the view that the taxonomic picture obtained by examining material procured directly from nature would be vitiated by the fact that conditions prevailing in nature may not be uniform and that a specific diagnosis drawn up out of such material may not be comparable with that of a specimen of the same species collected under different conditions of habitat, etc. This difficulty can be overcome by including in studies of this type a representative collection of specimens from different localities, etc. No taxonomic study would be perfect without a comprehensive investigation of a number of individuals of a given taxon obtained from different areas, different substrata, etc. What is important is the range of variation in key characteristics of a taxon as it occurs in nature rather than under laboratory-made artificial cultures. Cultures certainly have a major place in the study of the biology, physiology and various other aspects of molds, but their importance in taxonomy has been overemphasised. *Hochkultur* or *normkultur* cultures, if readily available, could certainly be used for taxonomic purposes; but the point I wish to emphasise here is that such cultures are not usually available, especially if the cultures had been maintained on rich synthetic media over a long period and in such cases the material obtained directly from nature provides the complete taxonomic picture. That appears to me the easiest and the most straightforward and simple approach to the solution of the problem and hence well worthy of serious consideration. Under the taxonomic procedure suggested here, the various species of *Fusaria* would be what they are in nature and not what they appear to be in culture.

My own findings are that it would be possible to identify *Fusaria* as they occur in their natural habitats without the aid of artificial cultures on synthetic or natural media; but here again a pre-requisite would be the reclassification of the genus on the basis of investigation of natural material and type and other exsiccatae of the various taxa. This raises the funda-

mental question of the characteristics of the genus which are of value taxonomically. The genus was established by Link in 1809. It is usually placed in the Moniliales-Tuberculariaceæ-Mucedineæ-Phragmosporæ and includes by description hyphomycetes with septate, branched, hyaline vegetative hyphæ and producing hyaline conidia typically in sporodochia, the sporodochia being considered the distinguishing characteristic of the Tuberculariaceæ. Three asexual spore forms are recognised, *viz.*, the micro-conidia, the macro-conidia and the chlamydospores. The conidia, in general, are slime spores and are borne typically on phialides. That the sporodochium is a hallmark of the genus is shown by the occurrence of sporodochia in a number of species of this genus as they occur in nature, but less frequently on culture media. However, in artificial cultures many forms have been reported to occur which lack sporodochia, but otherwise agreeing with the genus in other characteristics. Such non-sporodochial forms (*e.g.*, *Fusarium poæ* (Pk.) Wr., *F. chlamydosporum* Wr. and Rg., *F. semitectum* Berk. and Rav., *F. camptoceras* Wr. and Rg., *F. bostrycoides* Wr. and Rg., *F. conglutinans* Wr., *F. orthoceras* App. and Wr., and *F. angustum* Shérb.) are also included within the genus by Wollenweber and Reinking (1935) and by Snyder and Hansen (1940, 1941, 1945). Indeed, the *raison d'être* of this classification appears to be the same as the one recently stressed by Hughes (1953, p. 578), *viz.*, "that the species whose conidiophores are solitary, in tufts on a small stroma or in synnemata, are all congeneric." Apart from the application of this conclusion to the Fungi Imperfecti as a whole, its application to the genus *Fusarium* appears logical and should have approval, unless it be that future studies on natural material reveal the consistent occurrence of sporodochia in all species of the genus.

What then is the hallmark of the genus *Fusarium*? To my mind, it would be the typical hyaline, falcate or sickle-shaped phragmo-phialospores produced by the fungus. The classification of the genus should be based primarily on the nature of these phialospores, *i.e.*, their shape, septation and size. Some other characteristics, also recorded from specimens obtained directly from nature, like the nature of the conidiophores, the manner of conidial formation, the occurrence of smaller or micro-conidia and their morphology are also important. Most other characteristics like the presence or absence of chlamydospores and all cultural characters would appear to be of no value in taxonomy. Pathogenicity has been used as a criterion to classify species, varieties and forms by Wollenweber and Reinking and to classify forms by Snyder and Hansen. There is considerable evidence pointing to the fact that pathogenicity is a highly variable factor and it has been shown that several *Fusaria* have a wider host range than suspected. For instance,

*F. oxysporum* Schlecht. emend. Snyder and Hansen f. *vasinfecta* (Atk.) Snyder and Hansen is now known to be pathogenic to plants other than cotton, and the classification of the *Elegans* and other sections by Snyder and Hansen on the basis of selective pathogenicity has little experimental evidence to support it when applied to the genus as a whole. In such cases it would be superfluous and fantastic to accord any taxonomic status to the forms on the basis of their pathogenicity to one or more hosts. On the other hand, these forms could be given a biologic status within the genus since what they exhibit is biologic specialisation. The main argument in favour of this point of view is that biologic characters are highly variable especially in the case of facultative parasites and hence their use in taxonomy should be discouraged. A further criticism which would apply here is that the conditions optimum for pathogenicity are themselves ill-defined and variable so that it would be unwise to found a classification on such a criterion.

A thorough study, on these lines, of *Fusaria* as they occur in nature, collected from geographically different areas and from diverse substrata coupled with an enquiry towards an understanding of type specimens and other *exsiccatæ*, wherever possible, would pave the way for a taxonomic revision of the genus which would at once be easier to use and more in keeping with the characteristics of its individuals as they occur in nature.

#### REFERENCES

- Agnihotrudu, V. .. "Soil conditions and wilt diseases in plants. Rhizosphere microflora in relation to fungal wilts," *Thesis* approved for the Ph.D. degree of the Madras University, 1954 (Unpublished).
- Gordon, W. L. .. "The occurrence of *Fusarium* species in Canada, II. Taxonomy and prevalence of *Fusarium* species in cereal seed," *Canad. J. Bot.*, 1952, **30**, 209-51.
- \_\_\_\_\_ .. "The occurrence of *Fusarium* species in Canada, III. Taxonomy of *Fusarium* species in the seed of vegetables, forage and miscellaneous crops," *ibid.*, 1954, **32**, 576-90.
- Hansen, H. N. .. "The dual phenomenon in imperfect fungi," *Mycologia*, 1938, **30**, 442-55.
- Hughes, S. J. .. "Conidiophores, conidia, and classification," *Canad. J. Bot.*, 1953, **31**, 577-659.
- Miller, J. J. .. "Studies on the *Fusarium* of muskmelon wilt, I. Pathogenic and cultural studies with particular reference to the cause and nature of variation in the causal organism," *Canad. J. Res., C*, 1945, **23**, 16-43.
- \_\_\_\_\_ .. "Cultural and taxonomic studies on certain *Fusaria*, I. Mutation in culture," *ibid.*, *C*, 1946 *a*, **24**, 188-212.

- Miller, J. J. .. "Cultural and taxonomic studies on certain *Fusaria*, II. The taxonomic problem in *Fusarium* with particular reference to section *Elegans*," *Canad. J. Res., C*, 1946 *b*, 24, 213-23.
- Padwick, G. W. .. "The genus *Fusarium*, VI. A recent attempt at mass revision," *Indian J. agric. Sci.*, 1941, 11, 663-74.
- Sadasivan, T. S. .. "Succession of fungi decomposing wheat straw in different soils, with special reference to *Fusarium culmorum*," *Ann. appl. Biol.*, 1939, 26, 497-508.
- Snyder, W. C. and Hansen, H. N. .. "The species concept in *Fusarium*," *Amer. J. Bot.*, 1940, 27, 64-7.
- \_\_\_\_\_ .. "The species concept in *Fusarium* with reference to section *Martiella*," *ibid.*, 1941, 28, 738-42.
- \_\_\_\_\_ .. "The species concept in *Fusarium* with reference to *Discolor* and other sections," *ibid.*, 1945, 32, 657-66.
- \_\_\_\_\_ .. "Variation and speciation in the genus *Fusarium*," *Ann. N.Y. Acad. Sci.*, 1954, 60, 16-23.
- Stover, R. H. .. "The effect of soil moisture on the growth and survival of *Fusarium oxysporum* f. *cubense* in the laboratory," *Phytopathology*, 1953, 43, 499-504.
- Subramanian, C. V. .. "The saprophytic activity of *Fusarium vasinfectum* the cotton wilt pathogen in the soil, I. Colonisation of cotton root bits buried in the soil," *J. Indian bot. Soc.*, M. O. P. Iyengar Commn. Vol., 1946, 209-13.
- \_\_\_\_\_ .. "Is there a 'wild type' in the genus *Fusarium*?" *Proc. nat. Inst. Sci., India*, 1951, 17, 403-11.
- \_\_\_\_\_ .. "Studies on South Indian *Fusaria*, IV. The 'wild type' in *Fusarium udum* Butler," *J. Indian bot. Soc.*, 1955, 34 (1), in press.
- Wollenweber, H. W. .. "*Fusarium*-Monographie: Fungi parasitici et saprophytici," *Z. Parasitenk.*, 1931, 3, 269-516.
- \_\_\_\_\_ .. "*Fusarium*-Monographie, II. Fungi parasitici et saprophytici," *Zbl. Bakt.*, 1943, II Abt., 106, 106-202.
- \_\_\_\_\_, and Reinking, O. A. *Die Fusarien, ihre Beschreibung, Schadwirkung und Bekämpfung*, 1935, 355 pp, Paul Parey, Berlin.
- \_\_\_\_\_, Sherbakoff, C. D., Reinking, O. A., Johann, H. and Bailey, A. A. "Fundamentals for taxonomic studies of *Fusarium*," *J. agric. Res.*, 1925, 30, 833-43.