



Percentage distribution of foliicolous fungi of Maharashtra, India with respect to their disease symptoms: a novel study

R. Dubey ✉

A. D. Pandey

Botanical Survey of India, Western Regional Centre, Pune, India

Abstract: Fungal spores are continuously deposited on the leaf surfaces by wind impaction, sedimentation and rain wash-out from the atmosphere and splash-dispersal. They reside and act as asymptomatic mutualists, benign commensals or latent pathogens. A slight imbalance in this relation can lead to a pathogenic phase of the fungi, resulting in a variety of symptoms on leaves viz. discoloration, blight, sooty-spots, shot-hole, tar spot, powdery mildew, black mildews, downy mildews, rust, smuts, galls, sooty moulds and so on. With a view of studying the foliicolous fungi, a project entitled “Foliicolous fungi of Maharashtra” was undertaken and various areas were visited in different forest ecosystems of Maharashtra. A total of 429 isolates belonging 336 fungal species and intra specific taxa recorded during this study were categorized on the basis of foliage symptoms caused by them. Out of the entire recorded fungal taxa, 19.81% fungal species cause black mildews; 5.83% fungal species belong to only cercosporoid fungi causing leaf spots; 2.10% fungal species incite powdery mildews; 10.96% cause sooty molds; 1.63% incite anthracnose; 0.23% incite leaf smut; 3.50% incite leaf rust infection; 19.35 % incite leaf spots; 11.42% cause leaf blight/canker; 0.47% fungal species incite tar spots; 1.86% incite wilt diseases and the remaining 22.84% were found to be associated with leaves as facultative parasites/saprophytes/Hyperparasites. Therefore, above studies were designed to characterize foliicolous fungi that would further help in the conservation and management of biological resources and also increase the agriculture wealth of the nation.

Key words: Foliicolous fungi, symptoms, percentage occurrence

INTRODUCTION

India is endowed with diverse physiography, vegetation, ecosystem and habitats having huge potential for further explorations and utilization of its untapped biological diversity. Fungal spores are continuously deposited on the leaf surfaces by wind impaction, sedimentation and rain wash-out from the atmosphere and splash-dispersal. Though the leaf surface has long been considered a hostile environment for fungal colonists, it is a complex biological system, where interesting interactions occur between leaf and mycoflora intimately associated with its leaf systems (Andrews & Harris 2000). Kinkel (1997) reported that a thin nutrient film deposited from the atmosphere on the leaf surface further facilitates the microbial colonization for growing plants make new surfaces available for fungi. Fungal colonists reside and act as asymptomatic mutualists, benign commensals or latent pathogens. Subsequent studies (Ruinen 1961, Dickinson 1965, Dix & Webster 1995) suggested that an active population of fungi exist on the surface of physiologically active green leaves. A slight imbalance in this relation can lead to pathogenic phase of the fungi, resulting in a variety of symptoms viz., leaf discoloration, blight, sooty spots, leafy shot-hole, tar spot, powdery mildew, black mildews, downy mildews, rust, smuts, galls, sooty moulds and so on. Environmental changes in climate, CO₂ levels, UV radiation and air pollutants all affect the leaf surface fungal populations which in turn can exhibit altered growth and activity (Pugh & Buckley 1971). These fungal diseases have become important as they are dwindling country's economy and contributing to bio-deterioration or destruction of important plants resulting in economic losses. The potential of foliicolous fungi is not known much, from an application point of view; however, they constitute a fairly big group of micro-fungi.

Several excellent reviews emphasizing microbial colonization of leaf surface or related topics have appeared in recent years (Beattie & Lindow 1995, Jacques & Morris 1995, Andrews & Harris 2000,

Hirano & Upper 2000, Lindow & Leveau 2002, Lindow & Brandl 2003). A wide variety of fungi colonize the leaf even before senescence. Hogg & Hudson (1966) described the succession of fungi on leaves of *Fagus sylvatica*. Hogg (1966) elucidated the factors determining the natural succession of fungi on Beech leaves. Studies on the phylloplane mycoflora was conducted by Dickinson 1965 & 1967, Mishra & Tewari 1969, Mishra & Srivastava 1971, Ruscoe 1971, Pugh & Mulder 1971, Mishra & Kanaujia 1974, Fokkema et al. 1975, Dickinson & Skidmore 1976, Mishra & Tewari 1976, Warren 1976, Eicker 1976, Collins & Hayes 1976, Kumar & Gupta 1976.

Foliicolous mycoflora of a variety of plants have attracted the attention of plant pathologists with a view to explore the ecological interactions between the pathogenic and the saprophytic fungi with regard to disease interactions (Newhook 1951, 1957, Last 1955, Wood & Tveit 1955, Last & Deighton 1965, Leben 1965). Rare studies have also been carried out on other aspects of foliicolous fungi, such as Barlocher (1992) studied colonization of aquatic mycoflora on the leaf surface. Abundance and diversity of microfungi in the leaf litter of a lowland rain forest in Costa Rica were conducted by Bills & Polishook (1994). Foliicolous ascomycetes were well documented by Hansford (1946) and Reynolds (1978a, 1978b, 1979, 1982). Strangely, efforts to culture and preserve the foliicolous fungi have so far been very rare (Ellis 1971, 1976, Matsushima 1971a, 1971b, 1975, Hawksworth 2001). Hughes (1976) collected and monographed many sooty moulds from New Zealand. Reynolds (1971, 1978a, 1978b, 1979, 1982, 1983, 1985) reported on collections of sooty moulds from the Neotropics. A detailed study of Foliicolous leaf spot fungi was also conducted by Braun (1999), Deighton (1959), Ellis (1971, 1976). Ou (1985) gave a detail account of common foliar parasites of grasses in tropical regions.

Foliicolous fungi have been fairly well documented from some parts of India, especially north-eastern regions. Except for a few stray reports, there has been no detailed document on these fungi, from southern India. Significant hitherto contributors for the study of foliicolous fungi of India are Prof. Kamal and his students in north-eastern India including Nepal (Kamal & Singh 1980, Kumar & Kamal 1979, Rai & Kamal 1982, Kamal et al. 1985, Verma & Kamal 1987a, 1987b, 1991). Diversity, distribution and taxonomy of foliicolous fungi from Terai forests of Uttar Pradesh, was recently accomplished by Shambhu Kumar (2015) and Mall et al. (2013). A respectable account of work on foliicolous fungi has also been conducted in Central India by Rajak & Pandey (1984) and Sahni (1964).

A highly regarded work has been conducted by many workers in Western Ghats of India on biodiversity, biological distribution and taxonomy of micro-fungi (Bhat 1993, 2010, Bhat & Raghukumar 2000, D'Souza & Bhat 2002, Prabhugaonkar 2011, Pratibha et al. 2010). Foliicolous fungi of Goa and its

adjoining areas were studied by Jalmi (2006). Thimmaiah et al. (2013) conducted a systematic survey of the foliicolous fungi of Kodagu, Karnataka. Foliicolous fungi of Kerala is well documented by Hosagoudar and his students (Hosagoudar & Robin 2011). Hosagoudar & Biju (2013) studied "foliicolous fungi of Silent Valley National Park", Kerala, India. In India, detailed morphological and cultural studies of rusts were undertaken by Thirumalachar (1949), Hosagoudar (1988) and Bagyanarayana and Braun (1999). Maharashtra state of India is known to possess many of the most diverse and unusual habitats of the globe and thereby constitute a major reservoir for the living organisms, be it plants, animals or microorganisms. Sporadic studies on foliicolous fungi, especially rust fungi, of Maharashtra was presented by Chavan (1968, 1975), Chavan & Bakare (1973), Chavan & Bhambure (1975), Chavan & Kulkarni (1974), Kundalkar & Patil (1980, 1981), Patil et al. (2011), Thite & Patil (1975), Patil & Thirumalachar (1970), Patwardhan (1964), Sathe (1965), Mundkar & Thirumalachar (1945, 1952), Ajrekar & Parandekar (1931) and Patil (1991). Studies on ascomycetes flora of Maharashtra was conducted by Borse & Pawara (2007), Chiplonkar (1970), Jadhav & Pawar (2009), Kalani (1961), Kale & Kale (1970), Pande (1973, 1979 & 1981a), Rao (1970), Patil & Thite (1974) and Ujjainkar (2003). Some reports on aquatic fungi of Maharashtra are also available (Borse & Pawara 2007, Borse & Patil 2007, Borse et al. 2008, Ghanwant & Reddy 2011, Nambiar & Raveendran 2011). Patil & Thite (1980, 1981) studied the fungi of Mahabaleshwar. Studies on Myxomycetes of South-West Maharashtra were conducted by Tembhure & Nanir (2011). In Maharashtra, although serious efforts were not made to document the foliicolous fungi exclusively, but studies carried out earlier revealed the distinct presence of fungi on leaves (Pande 1981b, Parandekar 1964, Pande & Bansude 1980, Patil & Magdum 1979, Patwardhan, 1969, Sawant & Papdiwal 2007, Patil & Pawar 1989, Singh et al. 2011).

Thus, it is evident from the above-cited literature that no systematic and serious studies of foliicolous fungi have been undertaken in Maharashtra and only sporadic reports are available. All these observations motivated to look at the phylloplane as a distinct micro-habitat for the study of leaf surface.

In the light of above, the present study was designed to characterize foliicolous fungi of Maharashtra that would further help in the conservation and management of biological resources and also increase the agricultural wealth of the nation. The study involved a reconnaissance survey (2010–2015) in the natural stands and forest plantations throughout the Maharashtra State, encompassing different types of forest ecosystems, viz., Southern Indian moist deciduous forests, Western (montane) subtropical hill forests, Montane subtropical forest, Southern tropical dry deciduous forests, Open scrub

forests, West coast semi-evergreen forests, Southern tropical thorn forests, Garden plantation & City area. Depending upon the appearance, the symptoms are termed as discoloration, blight, sooty-spot, shot-hole, tar spot, powdery mildew, black mildews, downy mildews, rust, smuts and so on (Rangaswami, 1998). Recent studies conducted on foliicolous fungi in Maharashtra state of India have shown that high diversity of fungi has been observed in plant-associated microhabitats such as leaf surface.

MATERIALS AND METHODS

In the present study, dicot, monocot, bryophytes and pteridophytes plants were considered equally for collecting foliicolous fungi. The samples included live, senescing and moribund leaves with some kind of infections. The leaves with infection were plucked from herbaceous plants, bushes, climbers and tree plants and brought to the laboratory in separate collection bags. Each sample with a manifestation of foliicolous fungi was sorted out. Infected samples were brought to the laboratory in the aluminium foil bags so that saprophytes may not attack them. Further processing of infected samples was carried out by microscopic studies by preparing the slides in a drop of routine mounts (Lacto phenol, Cotton blue or Lactofuschins) and glycerine separately following different methods as hand sections, microtome techniques. Fungi with sporulating structures were observed under a compound microscope for detailed diagnostic features which aided in their identification.

Fungal species belonging to Ascomycota were identified from Dennis (1978). Hosagoudar (1996, 2008, 2013), as well as Hosagoudar (2012) were consulted for the identification of Black mildew fungi. Digital images were taken using Digital colour CCD Camera (Nikon DS Fi1) attached to a Nikon eclipse 50i microscope with interference optics. Scanning Electron Microscope (SEM) images of some fungal specimens were also captured. All the holotypes are maintained systematically in Botanical Survey of India, Western Regional Centre Herbarium, Pune and few new findings have been deposited in Ajrekar Mycological Herbarium, Pune. Descriptions of all new findings have been submitted to Mycobank.

RESULTS

The results presented in this report are based on an investigation carried out over a period of five and a half years, from November, 2010 to March, 2016, on the floristic foliicolous fungi associated with plants of Maharashtra. All the major geographical regions and forest types of Maharashtra were thoroughly explored during this period. In the present study, fungi occurring on leaves of angiosperm (both dicot and monocot), gymnosperm and pteridophytic plant species have been collected and studied in detail from

74 major localities of the State of Maharashtra. A total of 188 genera of fungi are documented in this study, of these 139 fungal genera are being represented by single species. Another 49 genera are represented by more than one species. Some fungi were encountered after a period of 35 years or more from India, viz. *Conidiocarpus betle* (Syd. P. Syd & E.J. Butler) T. Bose, *Asterina woodfordiae* V. P. Sahni, *Cercospora blumeicola* S. Das, *Cercospora careyae* T. S. Ramakr & K. Ramakr., *Meliola diospyri* Syd. & P. Syd. *Helicomina costii* M.A. Salam & P.N. Rao was recorded after a period of 65 years from India. During the study, it was examined that different fungal species inflict various kinds of symptoms over the leaf surface and cause the deterioration of the whole plant, which include leaf spots, leaf blotches, leaf blight/rot/canker, powdery mildews, black mildews, leaf rust, leaf smut, sooty moulds, tar spots, wilt etc. (Fig 1. and Fig 2.).

A total of 336 fungal species and intraspecific taxa recorded were categorized on the basis of foliage symptoms caused by them (Fig. 3). Out of the entire recorded 429 isolates belonging to 336 fungal species and intra specific taxa, 85 (19.81%) fungal species cause black mildews; 25 (5.83%) cercosporoid fungi cause leaf spots; 9 (2.10%) species cause powdery mildews; 47 (10.96%) species cause sooty moulds; 7 (1.63 %) species cause anthracnose; 1 (0.23 %) species causes leaf smut; 15 (3.50 %) species cause leaf rust infection; 83 (19.35%) species cause leaf spots; 49 (11.42%) species cause leaf blight/canker; 2 (0.47%) species cause tar spots; 8 (1.86%) species cause wilt diseases and the remaining 98 (22.84%) species are associated with leaves as facultative parasites/saprophytes/Hyperparasites. It was also noted that a single fungal species may cause more than one symptom or it may be associated with other fungal species over the leaf surface. The results show that among obligate fungi, the black mildews which constitute 85 species (19.81% of total) were found extensively distributed in the different forest ecosystems. Further, 98 fungal species inhabit the leaf substratum in form of facultative parasite/saprophytes/hyperparasite for their growth and survival.

DISCUSSION

Floristic details and biological associations when investigated systematically, as done in this study, lead to a treasure of information getting unfolded before us. It is something like nature revealing secrets before us. It was examined that very low number of foliicolous species have been reported earlier and not the true number of foliicolous fungal species occurring in Maharashtra, which reflects the small study efforts made to map them. Although it was assumed that the pleomorphic nature of fungi has been found adding to the complexity of classification, leaf diseases such as blights, wilt, rust, smuts, cankers, leaf spots, etc on a number of economically

and medicinally important plants often result in a huge economic loss. It was also observed that these fungal diseases interfere in plant's natural metabolism such as photosynthesis, respiration, uptake of water

and minerals. Due to fungal diseases, many of these unique gene sources of plants and the potential of producing new drugs of great benefit to mankind may be lost forever.



Fig. 1. Symptoms/diseases of foliicolous fungi. a. Leaf Spots; b. Leaf blights; c. Leaf discoloration; d. Black mildews; e. Sooty molds; f. Gall formation; g. Gall formation by Exobasidiales; h. Gall formation by Graphiolales



Fig. 2. Symptoms/diseases caused by foliicolous fung. i. Hypertrophy; j. Powdery mildews; k. Downy mildews; l. Rust disease; m. Smut on grasses; n. Flyspeck fungi on leaf surface; o. Tar spots; p. Sooty spots formed by insects and fungal association

Table 1. Continued

<i>Asterina jasmini</i> Hansf.	+	-	-	-	-	-	-	-	-	-	-	-
<i>Asterina jasminicola</i> W. Yamam	+	-	-	-	-	-	-	-	-	-	-	-
<i>Asterina morellae</i> Hosag., C.K. Biju & T.K. Abraham	+	-	-	-	-	-	-	-	-	-	-	-
<i>Asterina woodfordiae</i> V.P. Sahni	+	-	-	-	-	-	-	-	-	-	-	-
<i>Asterina wrightiae</i> Syd.	+	-	-	-	-	-	-	-	-	-	-	-
<i>Asterina wrightii</i> Berk. & M.A. Curtis	+	-	-	-	-	-	-	-	-	-	-	-
Asterostomella state of <i>Asterina jasmini</i> Hansf	+	-	-	-	-	-	-	-	-	-	-	-
Asterostomella state of <i>Asterina jasminicola</i> W. Yamam.	+	-	-	-	-	-	-	-	-	-	-	-
<i>Asterostomula pavettae</i> Hosag. & Sabeena	+	-	-	-	-	-	-	-	-	-	-	-
<i>Bahusandhika indica</i> (Subram.) Subram.	-	-	-	-	-	-	-	-	-	-	-	+
<i>Balladyna pavettae</i> Boedijn	+	-	-	-	-	-	-	-	-	-	-	-
<i>Balladyna ugandensis</i> Syd.	+	-	-	-	-	-	-	-	-	-	-	-
<i>Balladyna vanderystii</i> (Hansf.) Arx	+	-	-	-	-	-	-	-	-	-	-	-
<i>Balladyna velutina</i> (Berk. & M.A. Curtis) Höhn	+	-	-	-	-	-	-	-	-	-	-	-
<i>Beltrania mangiferae</i> Munjal & J.N. Kapoor	-	-	-	-	-	-	-	+	-	-	-	+
<i>Beltrania querna</i> Harkn	-	-	-	-	-	-	-	-	-	-	-	+
<i>Beltrania rhombica</i> Penz.	-	-	-	-	-	-	-	+	-	-	-	+
<i>Beltraniella spiralis</i> Piroz. & S.D. Patil	-	-	-	-	-	-	-	+	-	-	-	+
<i>Botryosporium longibrachiatum</i> (Oudem.) Maire	-	-	-	-	-	-	-	-	-	-	-	+
<i>Botryosporium madrasense</i> Raghuk.	-	-	-	-	-	-	-	-	-	-	-	+
<i>Calonectria morgani</i> Crous, Alfenas & M.J. Wingf.	-	-	-	-	-	-	-	-	+	-	+	-
<i>Camarosporium rubicola</i> (Sacc.) Sacc.	-	-	-	-	-	-	-	-	+	-	-	-
<i>Camposporium</i> sp.	-	-	-	+	-	-	-	-	-	-	-	-
<i>Capnodium coartatum</i> Chomnunti & K.D. Hyde	-	-	-	+	-	-	-	-	-	-	-	-
<i>Capnodium</i> sp. 1	-	-	-	+	-	-	-	-	-	-	-	-
<i>Capnodium</i> sp. 2	-	-	-	+	-	-	-	-	-	-	-	-
<i>Capnodium</i> sp. 3	-	-	-	+	-	-	-	-	-	-	-	-
<i>Capnodium</i> sp. 4	-	-	-	+	-	-	-	-	-	-	-	-
<i>Catenularia cubensis</i> Hol.-Jech.	-	-	-	-	-	-	-	-	-	-	-	+
<i>Cercospora apii</i> Fresen	-	+	-	-	-	-	-	-	-	-	-	-
<i>Cercospora blumeicola</i> S. Das	-	+	-	-	-	-	-	-	-	-	-	-
<i>Cercospora careyae</i> T.S. Ramakr. & K. Ramakr.	-	+	-	-	-	-	-	-	-	-	-	-
<i>Cercospora ricinella</i> Sacc. & Berl.	-	+	-	-	-	-	-	-	-	-	-	-
<i>Cercospora thumbergiae</i> Boedijn	-	+	-	-	-	-	-	-	-	-	-	-
<i>Chaetomella acutiseta</i> B. Sutton & A.K. Sarbhoy	-	-	-	+	-	-	-	-	-	-	-	-
<i>Chaetospermum camelliae</i> Agnihotr.	-	-	-	-	-	-	-	-	-	-	-	+
<i>Chalara siamense</i> Pinnoi	-	-	-	-	-	-	-	-	+	-	-	-
<i>Chalara</i> sp.	-	-	-	-	-	-	-	-	-	-	-	+
<i>Chloridium indicum</i> Subram.	-	-	-	-	-	-	-	-	+	-	-	-
<i>Ciliochorella mangiferae</i> Syd.	-	-	-	-	-	-	-	+	-	-	-	-
<i>Circinotrichum olivaceum</i> (Speg.) Piroz	-	-	-	-	-	-	-	+	-	-	-	-
<i>Cirsosia vateriae</i> Hosag.	+	-	-	-	-	-	-	-	-	-	-	-
<i>Cladosporium aecidiicola</i> Thum.	-	-	-	-	-	-	-	+	-	-	-	-
<i>Cladosporium cladosporioides</i> (Fresen.) G.A. de Vries	-	-	-	+	-	-	-	+	-	-	-	-
<i>Cladosporium colocasiae</i> Sawada	-	-	-	+	-	-	-	+	+	-	-	-

Table 1. Continued

<i>Zygosporium majus</i> Piroz.	-	-	-	+	-	-	-	-	-	-	+
<i>Zygosporium masonii</i> S. Hughes	-	-	-	+	-	-	-	-	-	-	+
<i>Zygosporium minus</i> S. Hughes	-	-	-	+	-	-	-	-	-	-	+
<i>Zygosporium oscheoides</i> Mont.	-	-	-	+	-	-	-	-	-	-	+
Total	85	25	9	47	7	1	15	83	49	2	98

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پراکنش نسبی قارچ های برگ‌ی در مهراشترا هندوستان با توجه به نشانه های بیماری: یک بررسی جدید

راشمی دابی ✉ و آمیت دیواکار پاندی

تحقیقات گیاه شناسی هندوستان، مرکز منطقه ای غربی، پونا، هندوستان

چکیده: اسپوره‌های قارچی به طور مداوم با وزش باد، همراه غبار و بارش باران از اتمسفر و سطح زمین روی سطح گیاهان قرار می‌گیرند. آنها در آنجا مستقر شده و به صورت همزیست های فاقد نشانه عمل می‌کنند، شروع به همسفرگی کرده و یا به صورت بیمارگرهای نهان ظاهر می‌شوند. یک عدم توازن جزئی در این ارتباط می‌تواند منتهی به مرحله بیمارگری قارچ ها شده که نتیجه آن ایجاد نشانه های مختلف روی برگ ها نظیر تغییر رنگ، سوختگی، لکه های تیره، غربالی، لکه قیری، سفیدک پودری، دوده ای شدن، سفیدک کرکی، زنگ، سیاهک، گال، کپک دوده ای، و غیره می‌باشد. برای بررسی اجمالی قارچ های برگ‌ی، یک پروژه با عنوان قارچ های برگ‌ی مهراشترا هندوستان ارائه گردید و مناطق مختلفی از این ناحیه در اکوسیستم های مختلف جنگلی مورد بازدید قرار گرفت. در مجموع، ۴۲۹ جدایه متعلق به ۳۳۶ گونه و درون گونه قارچی در طول این تحقیق به دست آمد که بر پایه نشانه های برگ‌ی که ایجاد می‌کردند، دسته بندی شدند. از همه آرایه های قارچی به دست آمده، ۱۹/۸۱٪ از آرایه ها سبب دوده ای شدن (black mildew)، ۵/۸۳٪ از آرایه های که متعلق به قارچهای سرکوسپورئید بودند، سبب لکه برگ‌ی، ۲/۱۰٪ از آرایه ها سبب سفیدک پودری، ۱۰/۹۶٪ از آرایه ها سبب کپک دوده ای، ۱/۶۳٪ سبب آنتراکنوز، ۰/۲۳٪ سبب سیاهک برگ‌ی، ۳/۵۰٪ سبب زنگ، ۱۹/۳۵٪ سبب لکه برگ‌ی، ۱۱/۴۲٪ سبب سوختگی/شانکر، ۰/۴۷٪ سبب لکه قیری، ۱/۸۶٪ سبب پژمردگی می‌شوند و معلوم شد که ۲۲/۸۴٪ باقیمانده به صورت پارازیت/سaprofیت/هیپرپارازیت همراه برگ ها هستند. از اینرو، مطالعه بالا که برای تعیین ویژگی های قارچ های برگ‌ی انجام شده است، می‌تواند برای حفظ و مدیریت منابع بیولوژیکی و همچنین افزایش بهره وری کشاورزی مفید باشد.

کلمات کلیدی: قارچهای برگ‌ی، نشانه ها، وقوع نسبی