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Fungal Disease Complex on a Nutritional, Medicinal and Economically Important Forage Crop - Maize from Bahraich (U.P.) India

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Abstract

During survey for the foliicolous fungi from diversified habitats of Bahraich (U.P.) India the authors came across an important crop for the locality *Zea mays* Linn. which belongs to Poaceae- the grass family is commonly called as zea, corn, silk corn, makka, barajowar. It is most widly distributed world's crop with short lifecycle which requires warm weather, appropriate apprension and management It is a valuable livestock feed, as a human food and raw material for several industries. On critical study of maize plants, we found the living leaves to be infected with *Alternaria, Curvularia* and *Drechslera* sp. On critical morphotaxonomic study it was found to be new species and the same taxon has been described and illustrated as plurivorus hyphomycetus fungi *Alternaria ziamaydis* sp. nov.; dematiaceous hyphomycetous fungi *Culvularia zeae* sp nov. and *Drechslera indiana* sp nov. The review of available literatures reveals that there has been no record of these fungus from India on this host so far. Therefore, this host for the three new species is a new record to Indian mycoflora from Bahraich (U.P) India.

Keywords: Foliicolous Fungi; *Zea mays*; North Central; Bahraich; Uttar Pradesh; India.

Introduction

Fungi are ubiquitous in nature and have occupied almost all places in the biosphere. These are the most important organisms in the universe.bacause of their vital functions in the maintenance of ecosystem, on human beings and their related activities. Fungi are often directly involved in our day to day life and play a crucial role in the nutrient cycling, decomposition and nutrient transport from soil to plants. These are extremely adaptable and can break down many substances including some toxic substances. This adaptability accounts for the presence of fungi in different environment around the world.

Some are plant and human pathogens, where as others are mutualistic symbionts. They are of great economic importance in brewing, baking, pharmaceutical industries, biometrical tools and several have been cultivated as substitute for food. Fungi also cause huge economic loss each year by causing spoilage of food, destruction of materials used by man, disease of plants, etc. Hence, fungi have both positive and negative effect of the mankind. These roles are played by fungi because of their diversity and abundance, roles in natural and altered ecosystem, land use, planning and management, etc. Fungi differ in their morphology, ecology, life history strategies, etc. The world of fungi provides an endless source of biodiversity. Only a very small part of the total fungal wealth has been subjected for the scientific studies. Thus we have very little information available with us. Further study has to be made to explore the hidden wealth of fungi.

Owing to the diverse climatic and altitudinal conditions, India is rich in the phanerogamic flora which is the chief hosts for the parasitic fungi. In comparison with phanerogams, the number of fungi known for this country is too less obviously because of the partial exploration.

The fungi, in general, which attack the cultivated

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plants, have got much importance because of their direct effect on mankind. Several other groups of fungi whose occurrence are mostly restricted to wild plants are less destructive to cultivated plants and hence receive less attention.

The leaves provide a very suitable habitat for the growth and development of fungal pathogen by providing ample surface area and nutrient supply. Such leaf inhabiting fungi are known as Foliicolous Fungi and the invaded area of the leaf appears as leaf spot or leaf lesion. Taxonomic studies of such fungal forms have been generally considered as only of academic interest but the taxonomic treatment of a fungal organism is the first requirement for any study concerning its biology. Correct identification of a fungus absolutely free from ambiguities is vital for its employment in applied disciplines. In fact without being equipped for ascertaining the correct identity of a fungal pathogen all studies concerning its phytopathological aspects would be misleading. The weed and forest plants serve as reservoirs of leaf spot pathogens which may spread to agricultural and horticultural plants.

India is located to the north of the Equator, lies between 8^{o} 4' and 37^{o} 6' north latitude and 68^{o} 7' and 97^{o} 25' east latitude, The country measures 3214 kilometres from south to north and 2933 km from east to west with a total land area being 32,87,263 square kilometres. India is the one of the twelve mega biodiversity countries of the world lying in between Tropic eighteen biodiversity hot spots located in the Western Ghats and in the Eastern Himalayas. The Himalayas rise as a of Capricorn and Tropic of Cancer, and has two of the world's virtual wall beyond the snow line. Above the alluvial plain lies the Tarai strip, a seasonally marshy zone of sand and clay soil. The Tarai region has higher rain fall than does the plains, and downward rushing rivers originating from the Himalayas slow down and spread out in the flatter tarai zone depositing fertile silt and reproductive spores during the monsoon season and receding in the dry season. The Tarai, as a result has high water level and moist subtropical conditions a luxuriant turnover of green vegetation year around. The climatological and topographical conditions also favour the luxuriant growth and development of foliicolous fungi. The North Tarai Region of Uttar Pradesh is next only to Eastern and Western Ghats, as having the highest biodiversity in general and the highest diversity of plant inhabiting fungi in particular. The region's high biodiversity offers an ideal opportunity for the morpho taxonomic exploration of the fungal organisms in general and foliicolous fungi in particular. We surveyed the North Tarai forests for Foliicolous Fungi during kharif season of 2014 & 2015 near surrounding agricultural fields of Bahraich Forest Range representing Bahraich Forest Division, Bahraich (U.P.).

Scope of Study

The Foliicolous Fungi causes huge losses every year in different parts of the world. The fungal pathogens producing leaf spots infect a large variety of hosts including most of the crops, forests and other plants. The destruction caused by these pathogens is a serious problem before us. The focus of this research is identification & documentation of foliicolous fungi which may assist in the development of new fungicides and ideas how to reduce the severity of infection as well as in in the protection of floral diversity from the infection of these pathogens as well as in the conservation of valuable flora of the area.

Materials and Methods

During survey and collection, infected plant parts where noticed were collected carefully in the field and notes were made regarding their pathogen city, nature of colonies infection, locality, altitude. For each collection a separate field number was given. Each infected plant parts was collected separately in polythene bags along with host twig preferably with the reproductive parts to facilitate the identity of corresponding hosts. These collections were pressed neatly and dried between sheets of blotting papers. The host plants were identified by matching them with authentic herbarium material and also consulting the experts.

In the laboratory, Hosagoudar, Biju & Appaiah (2006) nail polish technique was used to study the structural and morphological characters of fungi. Since the desired quality and quantity of nail polish is difficult to procure from the market, the problem is resolved by preparing a xylene - thermocol solution. Five millilitersi or desired quantity of xylene is poured in a container. Very bright and clean thermocol cut into minute pieces is added to xylene. It is mixed thoroughly till getting it to a particular consistency and poured it into air tight bottle for the use. A drop of xylene – thermocol solution applied on the selected colonies, carefully thinned with the help of a fine brush without disrupting the colonies.

The treated colonies along with their host plants kept in dust free chamber for half an hour. When the applied solution dried, a thin colorless "film" or flip is formed with the colonies firmly embedded in it.. A

drop of DPX was added on a clean slide and flip was spread properly on it. Care was taken to avoid air bubbles while mounting. One or two more drops of DPX were again added on the flip and clean cover glass brings out the excess DPX and it was removed after drying. These slides were labelled and placed in dust free chamber for one or two days for drying. Slides were prepared in cotton-blue lacto phenol mixture and were examined. Camera Lucida drawings were made and the morpho taxonomic determination of the taxa was done using available literature. The fungal taxa were identified using microscopic preparation. The fungal holotype specimen has been deposited for allotment of accession number from HCIO. The Mycobank No. from the Fungal Database Nomenclature and Species Banks was procured.

Result and Discussion

During our survey of the surrounding fields of crop plants for foliicolous micro fungi during kharif season of 2014 & 2015 we came across fields of *Zea mays* Linn. which belongs Poaceae- the grass family commonly called as zea, corn, silk corn, makka, barajowar. It is most widly distributed world's crop with short lifecycle which requires warm weather, appropriate apprension and management It is a valuable livestock feed, a valuable human food and raw material for several industries. etc severely infected with leaf spot pathogen. It is cultivated for grains as well as fodder. Multitudes of maize subspecies are identified and classified depending upon the extent of starch each have. Maize is a crop having short life cycle and requires warm weather, appropriate apprehension and management. It is valuable livestock feed, as human food and raw material for several industries.

Maize is native of South America but extensively cultivated in various other countries as well like India, Thailand, Pakistan and China, and in several parts of Philippines. It is considered as staple article of food in some islands and provinces. It is widely grown in temperate and tropic regions with well drained and fertile soil.

Pollens and seeds are the nutritious and edible parts of maize. Seeds are consumed in raw and cooked form that serves as good source of carbohydrates. Corn contain vitamin B-complex such as B1 (thiamine), B2 (niacin), B3 (riboflavin), B5 (panto thenic acid) and B6 that makes it commendable for hair, skin, digestion, heart and brain. It contains vitamin C, A and K together with large amount of beta carotene and fair amount of selenium that helps to improve thyroid gland and play important role in proper functioning of immune system. It has higher content of protein and fat as compared to other cereals. Corn silk contains maizeric acid, fixed oils, resin, sugar, mucilage, salt and fibres essential for our diet. Corn syrup is useful in manufacturing of jams, jellies, and other sweets and as a additive for cane sugar and maple syrup.

Edible oils obtained from seeds are useful in salad and for cooking. Roasted seeds are used as coffee substitute. Phyto chemical secondary metabolites such as saponin, allantoin, sterol, stigmasterol, alkaloids, hordenine and polyphenols are found in leaves, seeds and corn silk.

From the ancient time corn has been used to pacify kapha, pitta, anorexia, general debilities, emaciation and haemorrhoids. It is a potent antioxidant that guards body from harming by free radicals responsible for cellular damage and/or cancer. It has the potential to alleviate pain and possess analgesic activity as well. Helping production of sex related hormones assemble it good for sexual health especially for men with erectile dysfunctions. It is believed to improve symptoms of rheumatism as B-complex is able to improve joint motility.

Major nutrient of corn silk is potassium that is powerful diuretic. In Europe and some other countries such as French,Spain, Greece including India; corn silk is used to conquer urinary tract infections and kidney stones. While in China it has been widely taken in case of fluid retention and jaundice. Corn silk improves blood pressure and support liver functioning as well as producing bile. Roots, leaves and corn silk as decoction are used for bladder while the decoction of cob as tea is used for stomach complaints. It act as a good emollient for ulcer, wound and swelling. In some places decoction of corn silk and parched corn is extremely useful in nausea and vomiting. The oil present in corn (rich in embryo) is far and wide used for cooking and manufacture of soaps. Sticky gum contains dextrin used for sealing envelopes and labels. Corn starch is well recognized for its uses in cosmetics and pharmaceutical industries as diluents. Corn seeds are functional in making alcohol and stem fibres for manufacture of paper.

The crop is a significant dietary food for one third of the world population. It is a major source of dietary energy and nutritional security for poor farmers and consumers because of the essential minerals and nutrients.

India being a rich source of raw material may provide a health promoting food, promising for the economic growth of the country and nutritious food at low cost may be processed for global utilization (Mall and Tripathi, 2016).

On consultation of the literature available viz, Fungi of India (Bilgrami & Jamaluddin,1979,1981; Sarbhoy, Varshney and Agarwal, 1996), forty six fungal species representing twenty eight fungal genera had been recorded on this host plant. On primarily stage of study three different type of spores were noticed. On critical study the living leaves were found to be infected with a plurivorus hyphomycetus fungi *Alternaria*; dematiaceous hyphomycetous fungi *Culvularia* and *Drechslera*. On critical examinations (Ellis, 1971,1976; Ellis and Ellis, 1997) and comparison with other known species found on the sane host or a host of the same family these were found to be a new species. The same are named as *Alternaria zeamaydis* sp. nov., described as *Alternaria zeamaydis* Kumar, Mall and Tripathi sp.nov., *Curvularia zeae* sp. nov., descrived as *Curvularia zeae* Kumar, Mall and Tripathi sp.nov.& *Drechslera indica* sp. nov., described as *Drechslera indiana* Kumar, Mall and Tripathi sp. nov.

Alterneria zeamaydis Kumar, Mall and Tripathi sp.nov.

Infection spots amphigenous, irregularly spreding leaf margins, grayish black to dark black. Colnies amphiphyllous, effuse, spred over the infection spot, necrotic spot along the margin of the leaf lamina. Mycelium of hyphae immersed, stromatic. Stromata 2-4 celled, immersed, spherical, pseudoparenchymatous measuring up to 16 micron in diameter. Conidiophores, macromematous, mononematous, straight or flexous, simple, cylindrical branch to unbranched, thick walled, 3-4 septate, brown 40-60 micron long and 5-7 micron wide. Conidiogenous cells integrated terminal to intercalary sympodial, polyteretic, bearing thickened conidial scars. Conidia muriform, solitary or in chain, acropleurogenous, obclavate or ellipsoidal or ovoid, straight to slightly curved, some times beak rostrate with scar at tip, olivaceous brown to dark brown, thick walled in the broadest parts rarely at the septa bearing 5-6 transverse septa with longitudinal or oblique septa, 40-55 micron long and 10-15 micron wide., germinating conidia present.

On living leaves of *Zea mays* Linn. (Poaceae), Barapatthar, Bahraich; Bahraich Forest Division, Bahraich (UP) India, August 22, 2015; GPS 27° 31′ 23.055" Latitude; 81° 36′ 38.270" Longitude; 276 ft. Altitude Leg; Ajay Kumar, T.P. Mall and S.C. Tripathi BRH-01379, AK- 0379 (Icotype), HCIO- Holotype, Mycobank MB: 818881/22.10.2016.

Curvularia zeae Kumar, Mall and Tripathi sp.nov.

Colonice amphigenous, effues, hairy, at first grey later black. Conidiophores upto 55-60 ¼ long and 4-6 ¼ wide broun. Conidia 2 septate , with middle cells brown or dark brown and end cells much paler 28-32x10-14 ¼ .

On critical examinations (Ellis, 1971, 1976; Ellis and Ellis, 1997) and comparison with other known species *Curvularia lunata* (Wakker) Boedija (Subramanium, 1953) reported on the same host plant also it was found to be a new species. The same is named as Curvularia zeae sp. nov. and described as ***Curvularia zeae*** Kumar, Mall and Tripathi sp.nov.

Table 1: Comparison of morphotaxonomic features of *Alternaria tenuis* Nees with *Alternaria zeamaydis* sp. nov.

Morphotaxonomic features	***Alternaria tenuis* Nees (Bhaskaran,1972)**	***Alternaria zeamaydis* sp. nov.**
Colonoies	Colonies usually balck or olivaceous black	Colonies amphiphyllous, effuse, brown
Mycelium	--------	Mycelium internal, Stroma absent
Conidiophore	Conidiophore arising singly or in small groups, simple or branched, pale to golden brown 50x3-6 micron with one several scars	Conidiophore in fascicle, macromematous, mononematous, straight or flexous, simple, cylindrical branch to unbranched, thick walled, 3-4 septate, brown 45-60 micron long and 4-6 micron wide.
Conidia	Conidia in straight or branched chains, obclavate upto 8 transverse and serval longitudinal septa Conidia 20-63x9-18 micron	Conidia acropleurogenous, solitary to catenate, dry obclavate to ellipsoidal to ovoid, rostrum present, 4-5 transvercely sepate and two obliquely septate, Conidia 46-56x10-14 micron

The above comparative account shows that the morphotaxonomic features of the present collection is different from those of *Curvularia lunata,* therefore, proposal of a new taxon of separate species rank, for the present collection, is deemed justified.

***Drechslera indiana** Kumar, Mall and Tripathi sp.nov.*

Infection spot epiphyllous thin dark brown to black, confluent coalescing to cover almost entire leaf surface with original shape intact; conidiophores simple, emerging solitary, dark brown to olivaceous, geniculate, bearing 3-6 conidia occasionally more, producing the first conidium at a distance of about 25 µm from the base 240--260 x 8-11µm, 5-8 septate, Conidia straight or more often slightly curved to one side, tapering slightly towards both the abruptly rounded end, cylindrical, slightly broader near the

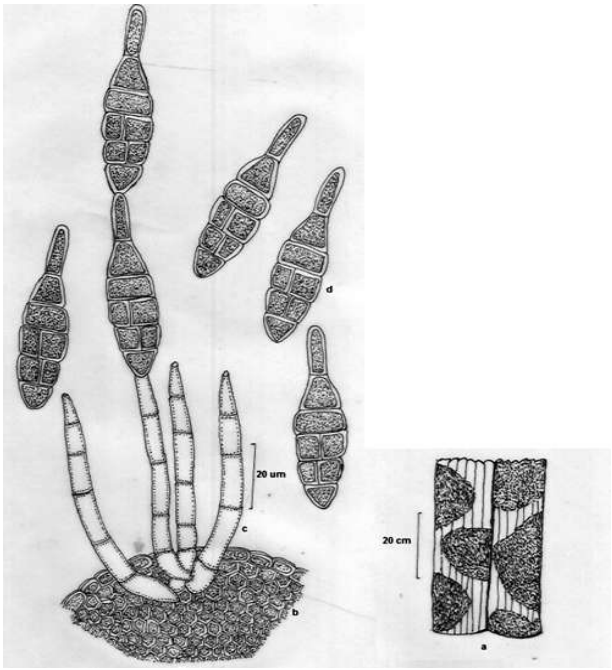

Fig. 1: *Alternaria zeamaydis* Kumar, Mall and Tripathi sp. Nov. **A:** Infected leaf, **B:** Stroma, **C:** Conidiophore, **D:** Conidia

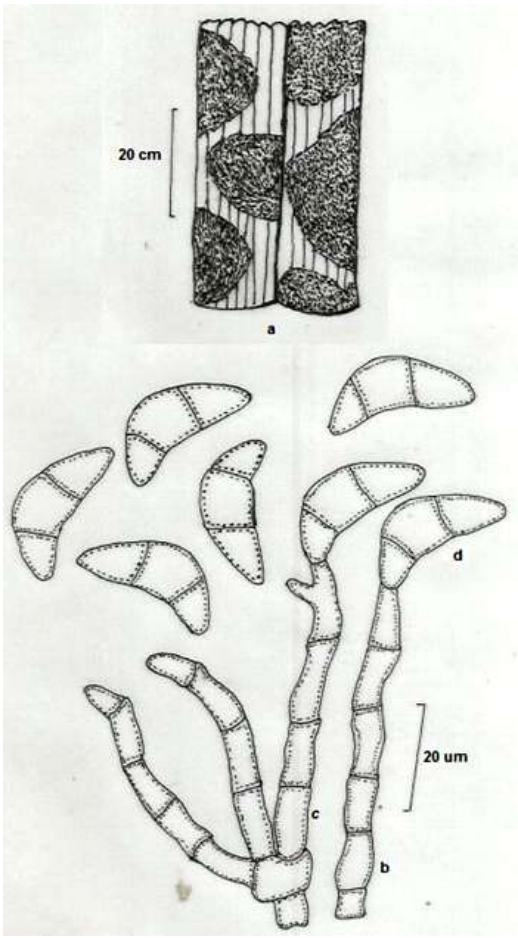

Fig. 2: *Curvularia zeae* Kumar, Mall and Tripathi sp. Nov. A: Infected leaf, B: Stroma, C: Conidiophore, D: Conidia

middle, light fuligeno0us when young later turning in into light to dark brown in color at maturity; 35-45X9-11 µm, 5-6 septate, non constricted at the septum..

On living leaves of *Zea mays* Linn. (Poaceae), Barapatthar, Bahraich; Bahraich Forest Division, Bahraich (UP) India, August 22, 2015; GPS 27° 31' 23.055" Latitude; 81° 36' 38.270" Longitude; 276 ft. Altitude

Leg; Ajay Kumar, T.P. Mall and S.C. Tripathi BRH-01379, AK- 0379 (Icotype), HCIO- Holotype, Mycobank MB: 818881/22.10.2016. .

The above compairative account shows that the marphotaxonomic features of the present collection is different from those of *Drechslera* state of *Setosphaeria taurica* Ellis, 1971 therefore, proposal of a new taxon of separate species rank, for the present collection, is deemed justified.

Conclusion

The above comparative accounts shows that the morphotaxonomic features of the present collection are quite different from there comparable fungal strains. Therefore, proposal of the new taxon of both species rank, f or the present collection is justified.

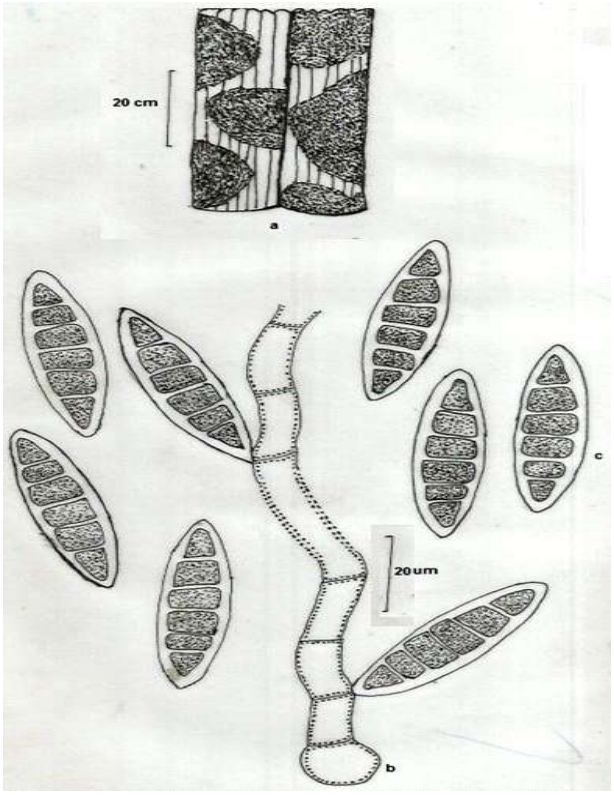

Fig. 3: *Drechslera indiana* Kumar, Mall and Tripathi sp. Nov. **A:** Infected leaf, **B:** Conidiophore, **C:** Conidia.

Table 2: Comparison of morphotaxonomic features of *Curvularia lunata* (Wakker) Boedija with *Curvularia zeae* sp. nov.

Morphotaxonomic features	*Curvularia lunata* (Wakker) Boedija	*Curvularia zeae* sp. nov.
Symptom	Colonies effuse	Colonice amphigenous, effues, hairy , at first grey later black
Conidiophore	----------	Conidiophores upto 55-60 µ long and 4-6 µ wide broun.
Conidia	3 septate, 20-32X12-15	Conidia 2 septate , with middle cells brown or dark brown and end cells much paler 28-32x10-14 µ .

Table 3: Comparison of morphotaxonomic features of *Drechslera* state of *Cochliobolus carbonus* Nelson, 1959 with *Drechslera indiana* sp. nov.

Morphotaxonomic Features	*Drechslera* state of *Cochliobolus carbonus* Nelson,1959	*Drechslera indiana* sp. nov.
Symptoms	On Zea mays attacking aerial part,dark brown or black mycelium giving charred appearence .	Infection spot epiphyllous thin dark brown to black, confluent coalescing to cover almost entire leaf surface with original shape intact;
Conidiophore	Conidiophore arising singly or in small groups, straight or flexuous, mid to dark brown or olivaceous brown, up to 250µ long and5-8µthick.	.conidiophores simple, emerging solitary, dark brown to olivaceous, geniculate, bearing 3-6 conidia occasionally more, producing the first conidium at a distance of about 25 µm from the base 240--260 x 8-11µm, 5-8 septate
Conidia	Conidia curved, some times straight, occasionally almost cylindrical but usually broader in middle and tapering towards the rounded end end, 30-100X12-18µ, finally becoming dark orvery dark brown or olivaceous brown, hilum not very conspicuous.	Conidia straight or more often slightly curved to one side, tapering slightly towards both the abruptly rounded end, cylindrical, slightly broader near the middle, light fuligeno0us when young later turning in into light to dark brown in color at maturity; 35-45 x 9-11 µm, 5-6 septate, non constricted at the septum..

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