Studies On Fungi Associated With Sorghum (Sorghum bicolor) (L.) Moench

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Abstract: Sorghum is a cereal plant drown the grass family. Sorghum is third most produced grain in the country. Farmer mainly used the crop as livestock food in production of ethnol. This has made India the second largest producer of sorghum in the world. The sorghum bran is low in protein and ash and rich in fiber components. Sorghum is widely cultivated as a food and oil seed crop. Cultivation of sorghum offers health and economic benefits but one of the most important challenge faced by sorghum growers is the management of destructive disease.

Crop is susceptible to a variety of plant pathogen like bacteria, fungi, viruses and nematodes, resulting in poor. The fungus in sorghum like *Fusarium*, *penicillum* and *Cladomonas* as the major species causes substantial loss to plant sorghum. Sorghum is susceptible to fungal and bacterial microflora under certain environmental condition Sorghum is subjected to infectious disease which can sometime limit production. Every year, seed and seedling diseases of grain sorghum common in India. Medicinal plant extract used to control of the phytopathogen

INTRODUCTION

Sorghum is a cereal plant drown from the grass family and has been ranked among the top leading cereals grown around the world. early species of sorghum were discovered in Australia ,Africa, India, as well as parts of the pacific ocean. Sorghum grown well in areawith warm climates.and the production of alcoholic beverages and biofuels. Nutritionist categorize sorghum as very healthy, as it is rich with essential nutrients that are significant in the body. In united state of America as a cereal, sorghum is the third most produced grain in the country. USA as the world leading producer of sorghum, with a production total of 11.5 million metric tons. (UK Deb, MCS Bantian, .Roy and P Parthasaray Rao.Global sorghum production Scenario (2004) Pages 21-38in sorghum genetic enhancement: research process,dissemination and imapct. Patancheu 502 324, Andhra pradesh, India: International crops research institute for the Semi-Arid Tropics.)

Farmers mainly used the crop as livestock food and in the production of ethnol. Region with semi-arid climate in India choose to cultivate sorghum. Due to ideal climate, local do not require minimal fertilizers, which makes its cultivation economical. This has made India the second largest producer of sorghum in the world. India produces an average of 7.5 million metric tons of sorghum. The areas in India where sorghum is largely grown include the Maharashtra and Karnataka region Nigeria ranks as the top sorghum producing country in Africa, and the third largest in the world. The average production of sorghum in Nigeria to be 7.4 million metric tons, almost equalling India which rank second with 7.5 million metric tons. Sorghum in Nigeria is mostly grown for food, while other minor uses include livestock food and industrial raw material for production of malt and beer. Corn in Mexico is the leading crop, as it provides local with their significant food. In fact Mexico has become a major supplied of corn to other countries across the world. The average production, as 6.1 million metric tons. Jawar is one of the important food and fodder cereal crops cultivated across India. Sorghum popularly known as Jawar in India. It is being cultivated in Maharashtra and its contribution to total cereal production in state is 51 % Maharashtra has a total area of 66.77 lakh hectares under sorghum. Out of this total area 30.48 lakh hectares are grown in kharif and rabbi. D.R.Bapat, Senior Sorghum Breeder. Mahatma Phule Agricultural University, Rahuri. (Rabbi sorghum Production strategy for Maharashtra).Inhouse Review of cereals ,Sorghum and millet.A-ogram 7-11April 1987,ICRISAT Center, Patancheru,A.P.)

Oil Content- the seeds oil was later fractionated in to eight fatty acids. The total oil contents in the seed of orghum ranged from 5.0 to 8.2 %. The results revealed that oleic acid (0.43-0.56%) linoleic acids (27.59-50.73%) , stearic acids (1.9-2.59%) & palmitic acids (11.73-20.18%) was present in the seeds oil of different sorghum varieties when analysed by GC-MS. The large scale production of seed oil after refining process can contribute towards alleviation of edible oil. Sorghum grain was found to contain appropriately 50 times more wax and two-third as much oil as can. The sorghum bran is low in protein and ash and rich in fiber components. The germ fraction in sorghum is rich in ash protein and oil but very poor in starch. Over 68% of total mineral matter and 75% of the oil of whole kernel Is located in the germ fraction. (Fred A.Kummerow, The composition of sorghum grain oilandropogon sorghum var. Vulgaris. May 1946)

Yield loss- sorghum is widely is widely cultivated as a food and oil seed crop. Cultivation of sorghum offers health and economic benefits but one of the most important challenge faced by sorghum growers is the management of distructive disease. The crop is susceptible to a variety of plant pathogen like bacteria, fungi, viruses and nematodes, resulting in poor. Yields and quality of the produce. Among the most devastating fungal disease of sorghum are the Rust (*Puccinia* purpurea) leaf blight (Exserohilum turcicum),grass mosaic virus, sorghum ergot,Fusarium stolk rot, Head smut (sporisorium reilanum), Penicillum species charcoal rot causes substantial loss to plant stand and yield combined yield losses due to incidence of the diseases in sorghum.sorghum ergot (clavliceps africona) Sorghum ergot is a fungus whose spores compete with pollen at flowering. Sorghum

ergot has been found in late flowering commercial grain crops in northern. The fungus infect sorghum heads at flowering and is flavoured by mild temperature (15-30c). high humidity and overcast condition. The disease reduce yield through poor seed set and cause harvesting difficulties difficulties from the sticky honesydew on the heads. Fungal bodies, called sclerates, which replace the seed are toxic to livestock, particularly cows,sows and cattle. Fusarium stalk rot (fusarium species).fusarium stalk rot in sorghum is caused by a range of fusarium species cladomonas as the major species causing disease In India. Rust (Puccinia purpurea) -early symptoms on leaves are small purple -red or tan spot. Head smut (sporisorium relianum).

Sorghum vulgare- sorghum crop production has considerably increased in severable countries during the past few years. Sorghum is 5th important cereal after wheat, rice, and maize and signicicantly dietary food for one third of the world population. These crops are the principle sources of energy, protein, vitamins and minerals for million of the poorest people in these region and sustain the lives of the poorest rural people and will continue to do so in these foreseeable future. Sorghum is subjected to infectious disease which can sometimes limit production. Sorghum is susceptible to fungal and bacterial micro flora under certain environmental condition. The mycoflora not only threatent plant growth but also affect food,quality, causing huge economic losses. Every year, seed and seedling diseases of grain sorghum are common in India. (Varaprasad Bobbarala,.etal 2012)

Medicinal plant extract- plant extract used to control of the phytopathogen have been obtained mainly from tree plant such as *Eukalyptus* and neem (24%) of the studies with extract and herbaceous species like garlic, gingiber,turmeric and ocimum (54%). (J.R. Stangarlin, O.J. Kunh.,etal 2011)

We are used to the plant extract such *as Tinospora, ocimum, cyzygium, zingiber*,lime etc. Are used to control the growth of fungi like as *Alternaria alternata, Alternaria tenuis, Aspergillus flavus, penicllium chrysoginum, penicillium citrinum, Rhizopus stolonifer,curvularia lunata, cladosporium* etc.

From these presented numbers we can have an overview of the research on alternative control of disease in plants. Beside that other researches have been made to identify plants whose extracts have compouds with biological action against phythopathogens or biological action against phytopathogens or that induce plant resistance.

MATERIAL AND METHOD

Isolation of fungal species.

The externally and internally seed borne fungi are isolated by two important methods which are commonly used in laboratory and research instituents. The methods are known as followed:

1)Blotter paper method- This is very simple, most convevient and efficient of all the incubation methods pair of sterile while blotter papers of 9.5 cm. diameter was shaked in sterile distilled water and were place in presterilized petri plates 5 seeds of test sample per petriplates. Were then placed at equal distance on moist blotter. The plates were incubated at 27° c under diurnal conditions, on day of incubation, seeds were first examined under storoscopic microscope for determine the various fungal growths. The identification and further confirmation of seed born fungi was made by preparing slides of the fungi.

Fifty sorghum seeds were surface sterilized with 0.3% silver nitrate solution for 1 min. followed by immersion in sterile distilled water for 1 min.

Composition of media and media used for isolation.

1)PDA (potato Dextrose Agar)-

Peeled potato-200 gm.

Dextrose – 20 gm.

Agar –agar- 20 gm.

Distilled water- 1000 ml.

pH- 5.6

1) CZA (Czapek Dox Agar)
2) Agar- 15 gm.
3) Sodium nitrate- 3 gm.
4) Potassium chloride- 0.5 gm
5) Magnesium sulphate- 0.5 gm.
6) Diapotassium hydrogen phosphate- 1 gm
7) Ferrous sulphate- 0.1 gm.
8) Sucrose- 30 m.

Distilled water- 1000 ml
pH.-5.6.

2) **Agar plate method**- surface sterilized seeds were then placed on freshly prepared potato dextrose agar (PDA) plates (five seeds per plate). And incubated for three days t 25°c. pure culture of different out growing fungi were obtained by transferring fungal colonies to new PDA plates using sterile wireloop and incubating the plates for 5-7 days aat 25°c. pure cultures of each isolated were then stored at 4°c. in vials. Containing 2.5 ml of sterile distilled water for further use.

Identification of fungi:

The Identification of the fungal species is based on morphological characteristics of the colony and microscopic examinations.

Morphology of colony: waxy, velvetty, fluffy, leathery and cottany etc.

Vegetative Structure: Unicellular hyphae, coenocytic or septate.

Asexual Spores: zoospores, sporangiospores, conida or blastospores.

Sexual Spores: oospores, zygospores, ascospores, basdiospores.

5. Reprodutive bodies: ascocarpous, Basidicarpous, simple conidiophores, synnamata, sporodochia, pycnidia.

The fungi were identified with the help of standard procedure and relevant literature.

Biocontrol activites of botanicals:

Five plants were collected from local areas of marketyard of Ahmednagar and area of khadki, khandala, Arangav in Ahmednagar. All plants were identified with the help of Flora of Ahmednagar author: S.G Pradhan; N.P.Singh.

The list of common Medicinal Plant used in Study

Sr no	Common Name	Botanical Name	Family	Plant Part Used
1	Tulasi	Ocimum tenuiflorum	Lamiaceae	Leaves
2.	Jhambhul	Syzygium cumini	Myrtaceae	Leaves
3.	Gulvel	Tinospora cordifolia	Menispermaceae	Leaves

Preparation of plant extract:

Fresh sample were washed in tap water and finally washed thrice using sterilized distilled water. They were crushed in a sterilized distilled water. They were crushed in a sterilized pestle and morter by adding a little quantity of sterile distilled water just enough to crush the sample easily. The extracts were collected by filtering through the whatman filter paper. Finally, filtrates thus obtained from the leaves were used as stock solution

Result and Discussion

Table-1 Incidence of different specis of fungi on sorgum. (Agar plate method).

Sr. No	Name of fungi	Isolated from marketyard	Sorghum warehouse
1.	*Alternaria alternate*	25%	30%
2.	*Aspergillus Flavus*	40%	50%
3.	*Penicillium citrinum*	30%	35%
4.	*Penicillium chrysoginum*	25%	30%
5.	*Rhizopus stolonifer*	20%	30%
6.	*Alternaria tenuis.*	30 %	25 %
7.	*Fussarium oxysporum*	40 %	55 %
8.	*Cladosporium herbarum*	25 %	30 %
9.	*Curvularia lunata*	25 %	20 %

Fungi which associated with sorghum was isolated with the help of Agar plate method and returns were summerized in table number 1. From the observation it was recorded that nine different fungi were associated with sorghum grains like(*Alternaria alternata, Aspergillus flavus, Penicillium citrinum, penicillium chrysoginum,Rhizopus stolonifer, Alternata tenuis, Fussarium oxysporium, Cladosporium herbarum, Curvularia lunata).*

Aspergillus flavus are predominent over sorghum (50%) as compare to others. Then the most dominant fungal genera were Fusarium oxysporum(55%) and the Aspergillus species with a high frequency of 50% and with a relative percentage of 40%and50% respectively. The other fugal genera such as Alternata spp(30%). The genus Fusarium was the most dominent fungi among 9 fungi reported in this study. Fusarium is one most economically important genera of fungi causing diseases on a wide variety of plants and derived product.Maximum inciedence was recorded on sorghum grains collected from farmers warehouse. Penicllium citrinum

was shown their dominance over sorghum grains collected over sorghum grains collected from warehouse (35%) followed by market yard (30%) and penicillium chrysoginum (30%) followed by market yard (25%).

Table-2 Incidence of different species of fungi on sorghum.(Blotter method).

Sr no	Name of Fungi	Isolated from Marketyard	Sorghum warehouse
1.	*Alternaria alternate*	25%	20%
2.	*Aspergillus flavus*	30%	35%
3.	*Penicilliumcitrinum*	15%	20%
4.	*Penicilliumchrysoginum*	20%	22%
5.	*Rhizopus stolonifer*	45%	50%

Fungi which associated with sorghum was isolated with the help of Blotter plate method results were summerized in table number 2. From the observation it was recorded that five different fungi were associated with sorghum grains, like (*Alternaria alternata*, *Aspergillus flavus*, *Penicillium citrinum*, *Penicllium chrysoginum*, *Rhizopus stolonifer* etc.) percentage of isolated fungi species which occurred in sorghum grain samples collected from marketyard and warehouse Rhizopus stolonifer was the most prevalent fungi in all examined sample. The Rhizopus stolonifer is the predominent and comprised (50%) of the total fungal count of the isolates. The most dominant fungal genera were Rhizopus stolonifer (50%)and the Aspergillus group was the most prevalent fungi in samples, Aspergillus flavus may to a certain extent already be present on grain brought into the ware house.
Maximum incidence was recorded on sorghum grains collected from farmer warehouse. Penicillium citerinum was shown their dominance over sorghum grains collected from warehouse (20%) followed by market yard (15%) and the penicllium chrysoginum are collect from warehuse (22%) followed by market yard (20%).

Effect of leaf extract on fungal growth (in gram)

Name of Fungi	*Syzygium cumini*		*Tinospora cordifolia*		*Ocimum tenuifolia*		Control	
	Wet Wt	Dry Wt	Wet Wt	Dry Wt	Wet Wt	Dry Wt	Wet Wt	Dry Wt
Aspergillus flavus	6.68	0.50	5.24	0.14	5.08	0.15	8.20	0.60
Rhizopus stolonifer	4.45	0.34	4.34	0.34	3.40	0.38	6.45	0.50
Penicillium citrinum	3.99	0.46	4.25	0.45	3.29	0.59	5.78	0.80
Penicillium chrysogenum	3.68	0.39	2.88	0.42	2.83	0.34	4.33	0.47

Effect of different leaf extracts against the selected pathogen was tested pathogen was tested and result. From the result it was observed that crude leaf extract of *tinospora cordifolia* retard the growth of *Penicillium* species (0.42gm) as well as Rhizopus stolonifer(0.34) compare to control leaf extract. The maximum inhibition plant extract *Ocimum tenuiflorum* Aspergillus flavus 0.15 followed by leaf extract of osmium tenuiflorum against pencillium cetrinum (2.83)gm as compare to control. The maximum inhibition. Plant extract ocimum tenuiflorum (0.59)

All the leaf extract were useful for the control of fungal growth they show inhibitory activity against the selected pathogen.

Bioefficiancy- Bioefficiancy of plant extract against selected fungi. The five medicinal plant are used in the biocontrol agent. These five medicinal plant are collected from the

All plants were identified with the help of taxonomic literature such as flora of ahmedanagar.

The list of common medicinal plant used in the study,

1) Syzygium cumini (Jambhul)

2) Tinospora cordifolia (Gulvel)

3) *Ocimum tenuiflorum (Tulas)*

4) *Zingiber officinale (Ale)*

5) *Citrus limon (lemon)*

One gram of leaves of selected plant then crush into 10ml acetone, with the help of morter and pestal.

These five plant are used as a biocotrol agent. These plant are easily available in the surrounding hence biofertilizer or biopesticide are not use to control fungal growth.

Discussion-

Leaf extracts of Tinospora cordifolia retards the growth of Aspergills flavus as well as leaf extract of Ocimum tenuiflorum also retards the growth of Aspergillus flavus similar type of result was reported by (stangarlin, etal 2011) from the result it was observed that crude leaf extract of eucalyptus and neem (24%) retarted the growth of penicillium, aspergillus and Rhizopus (9%of works).

The leaf extract of Syzygium cumini retards the growth of Rhizopus stolonifer as well as leaf extract of Syzygium stolonifer. Similar type of results was reported by (Zakaria Awad Mahamed Baka 2014) from the result are observed that the treatments were possotively growth of all fungi tested, compared to the control.

A leaf extract of ocimum tenuifolia retards the growth of Penicillium chrysoginum . similar type of result was reported by (Mmdouh salem serag 2014) from the result indicate that the treated seeds and grains with plant extract gave very good effects on their germination and also reducing seed born infection.

From the present study it was observed that. Rhizopus stolonifer and Aspergillus flavus show their dominance on sorghum seeds. Seeds collected from market places yield more number of fungi. Compare to seed of ware house our results were well supported by Zakaria Awad Mohamed Baka (2014). They reported the Aspergillus flavus and Rhizopus stolonifers.

Summary and conclusion-

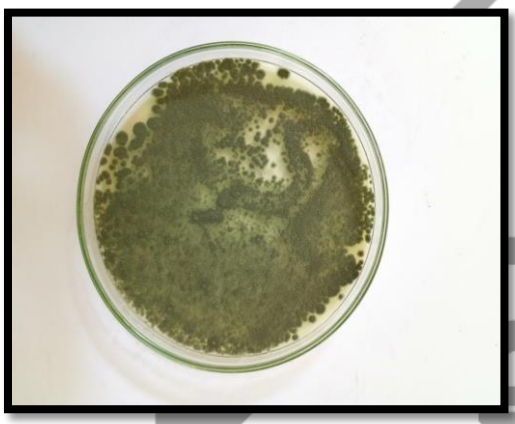
The infected sorghum sample collected from different places used for isolation of pencillium species as well as aspergillus flavus, alternaria alternata, rhizopus stolonifer. The isolation was carried out by agar-plate, blotter plate method. The identification was made by preparing slides of fungal growth and with the help of manuals. Pure culture of these fungi was prepared to maintain on potato dextrose agar slants. Two penicillium species, aspergillus, rhizopus and alternaria species isolated all the selected species was carried out by the pigmentation. Study the different colour are found in this different species. The antifungal activity studied by using syzygium cumini, Tinospora cordifolia, ocimum tenuiflorum, Zingiber officinale, Citrus limon etc. plant leaves extract the fungal growth are control.

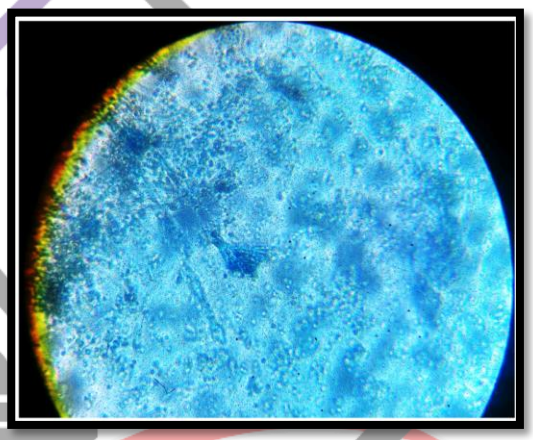
Rhizopusstolonifer severaly affect the sorghum panicle. These rhizopus species shows diversity in pigmentation. It ranges from yellow, black, green, brown.

To control these rhizopus species effect citrus, gulvel, tulsi, zingiber,jamun etc.

Leaf extract was tested against these pathogenic fungi.It was observed that in treated flask growth of these Rhizopus,penicillum species was retarded as compare to control.It is concluded that leaf extract of citrus, gulvel, tulsi, zingiber,jamun is good source to control the growth of selected different species of fungi.

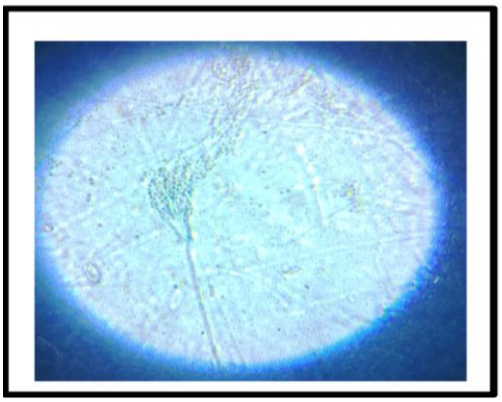
Aspergillus flavus





Penicillium citrinum

Alternaria alternata



Penicillium chrysogenum

Rhizopus stolonifer

Penicillium citrinum

penicillium chrysoginum

Aspergillus flavus

Penicillium chrysoginum

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