

Screening of certain Ayurvedic plants extracts against *E. turcicum*

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The use of chemicals against pathogens is environmentally dangerous, so use of natural inhibitors for disease management is need of time. In this work we screen botanical extracts from ayurvedic plants for their antifungal properties against economically important plant fungal pathogen. As test fungal pathogen we select *E. turcicum*, a potent fungal pathogen responsible for Northern leaf corn blight of Maize This fungal pathogen was challenged by the leaf extract prepared from certain ayurvedic plants and these observation has shown a promising future in biocontrol of fungus by using such environment friendly antifungal agents.

India through its long history has accumulated a rich empirical knowledge of the use of herbs for the treatment of various diseases. Numerous studies have been carried out to extract various natural products for screening antimicrobial activity but proper attention has not been viewed intensively on evolving the competitive studies of these products for their antimicrobial activity. Traditionally, plant fungal diseases are controlled by synthetic fungicides, which increase agricultural costs and contaminate the environment with very toxic substances (Carvalho, 2004). A possible alternative to solve such problems is the use of plants able to produce antifungal substances (Miranda, 2003). Identification of indigenous plant extracts with anti-fungal activity against *P. griseola* would contribute substantially in development of an environmentally friendly control method for angular leaf spot. Extracts from plants such as *Azadirachta indica*, *Allium sativum*, *Lipkea javanicum*, *Urtica massaica*, *Satureia biflora*, *Warburgia ugandensis* and *Zingiber officinales* are reported to possess antimicrobial activity against a wide range of plant pathogens (Singh *et al.*, 1995; Otanga, 2005). The possible use of plant extracts with antifungal activity for the control of *P. griseola* is an area that has not been exploited. Many such trials have been made to discover new antimicrobial compounds from various kinds of sources such as microorganisms, animals and plants. One of such resources is traditional medicines. Systematic screening of them may result in the discovery of novel effective compounds. The present scenario of emergence of multiple drug resistance to human pathogenic microorganisms has been necessitated a search for antimicrobial substance from other sources including plants (John Britto *et al.*, 2004).

Northern leaf corn Blight of Maize

Maize (*Zea mays* L.) an important crop used as major food part for both human & animal consumption along with this it's a cereal with economic and industrial value. This crop threatens by many biotic stresses one of which *Turcicum* Leaf Blight (TLB) caused by a fungus *Exserohilum turcicum* (Pass) which is also known as Northern corn leaf blight in general. The pathogen and disease was first reported by Passerini (1876) from Italy. In India, it was first reported by Butler *et al.* (1920) is very important from the point of view of its management for better grain productivity (Grover and Pental, 2003). Although disease has wide spread distribution in cold humid regions with heavy dew(Dorothea *et al.*,1998;Juliana *et al.*, 2005) in Indian scenario, the disease is considered to be mostly appears in a sizeable form in Karnataka, Maharashtra, Andhra Pradesh and Himalayan regions and some other parts of the country. The disease causes leaf necrosis and premature death of leaves which can reduce the grain yield of maize by 28 to 91 per cent (Carlos 1997 and Harlapur *et al.*, 2000). Sample disease leaf is given below in fig.1.



Figure1- Leaf sample with *E. turcicum* infection

Practically many approaches have been developed for TLB management, like screening and development of resistance varieties by inheritance studies and characterization of qualitative and quantitative genes (Juliana *et al.*, 2005). Highly studied gene *Ht1*, *Ht2*, *Ht3*, *Htn*, and *Htm*, have profound phenotypic effect characterized by lesions without spore formation(Mwangi,1998, Singh *et al.*,2004).effect of *Ht* gene now being challenged by newly evolving strains of *E. turcicum*, with improved virulent capabilities. Use of fungicides and chemical agents against TLB, have proven their potential to control disease in most threaten areas. There are several potent fungicides has been screened against *E. turcicum* (Begum *et al.* (1993). The presence of naturally occurring substances in plants with antifungal properties have been reported and tested against wide range of fungi infecting many commercially

important crops (Shivapuri *et al.*, 1997). However, studies on control of *turcicum* leaf blight of maize by means of plant extracts are meager. In same series of attempt ethanol extracts of some plants like *Allium cepa*, *Azadirchata indica*, *Ocimum sanctum* and *Polyalthia longifolia*, give profound percentage of inhibition (Shivapuri *et al.* 1997; Singh *et al.* 1998).

Material & Methods

Plant samples were collected from Ayurvedic garden, Banaras Hindu University, Varanasi. The plants used in this experiment in this experiment are given in table-1 with their common Indian name and general properties.

Table-1 showing plant used in experiment

S.n o.	Plant	Indian Name	Properties
1	<i>Picrorhiza kurroa</i>	Katuka	Anti-allergic, anti-inflammatory, Immunostimulant
2	<i>Tinospora cordifolia</i>	Guduchi	anti-diabetic, anti-inflammatory, anti-arthritis, anti-oxidant, anti-allergic, anti-stress, anti-malarial, hepatoprotective, immunomodulatory
3	<i>Andrographis paniculata</i>	Kalmegh	antipyretic and hepatoprotective, anti-typhoid, antifungal
4	<i>Crinum latifolium</i>	Sudarshan	Used in hypoxia, inflammation, detoxification, tissue regeneration, and hormone balancing. Leaf juice is used for earache, rheumatic pain, and sprain
5	<i>Phyllanthus urinaria</i>	Bhumyamalaki	Enteritis and diarrhea, infantile marasmus, swollen and inflamed rectum. Asthma, bronchitis, cough, tuberculosis
6	<i>Terminalia chebula</i>	Haritaki	oral ulcers, sore throat, loose gums, bleeding and ulceration in gums

Preparation of Test fungus

Test fungus used was *E. turcicum* responsible for Turcicum leaf Blight of maize. The fungus was isolated from infected leaf sample. The part of sample with clear visible symptoms washed thoroughly with distilled water. A small portion of diseased tissues along with a portion of adjacent healthy tissue was trimmed around 1mm in length. Then surface sterilized with chemicals such as HgCl₂ (0.01%) solution.

The pieces were then rinsed thrice with sterilized distilled water and inoculated aseptically on sterilized Potato Dextrose Agar (PDA) plates. These cultured PDA in incubator at 28⁰ C.

Plant Extract preparation-

Plant extract was prepared as fresh leaf sample were taken with sterile condition, and left at room temperature. Now crush 5 g. of Plant sample with 10 ml 40% ethanol using mortar- pestle and filter through muslin cloth. Now centrifuged at 10,000rpm for 10 min. all this preparation was stored at 4⁰C.

Calculation of percentage inhibition-

The percentage inhibition was calculated by given formula

$$I = \frac{C-T}{C} \times 100$$

Where I = percentage inhibition

C = Radial Growth in control

T = Radial Growth in Treatment

Results & Discussion-

In the present investigation all the plant extracts were evaluated under in vitro condition against *E. turcicum* to know the antagonistic nature of these extracts. Though complete inhibition (100% inhibition) of the pathogen was not observed but significant amount of inhibition was noticed in some of them. These inhibitory result are given in Table-2

S.No.	Plant	T(cm.)	Part used	Inhibition
1	<i>Phyllanthus urinaria</i>	1.5	Leaf	63.3%
2	<i>Tinospora cordifolia</i>	1.6	Leaf	64.4%
3	<i>Crinum latifolium</i>	1.25	leaf	72.2%
4	<i>Terminalia chebula</i>	1.6	leaf	66.6%

Table-2 Showing percentage inhibition on *E. turcicum*

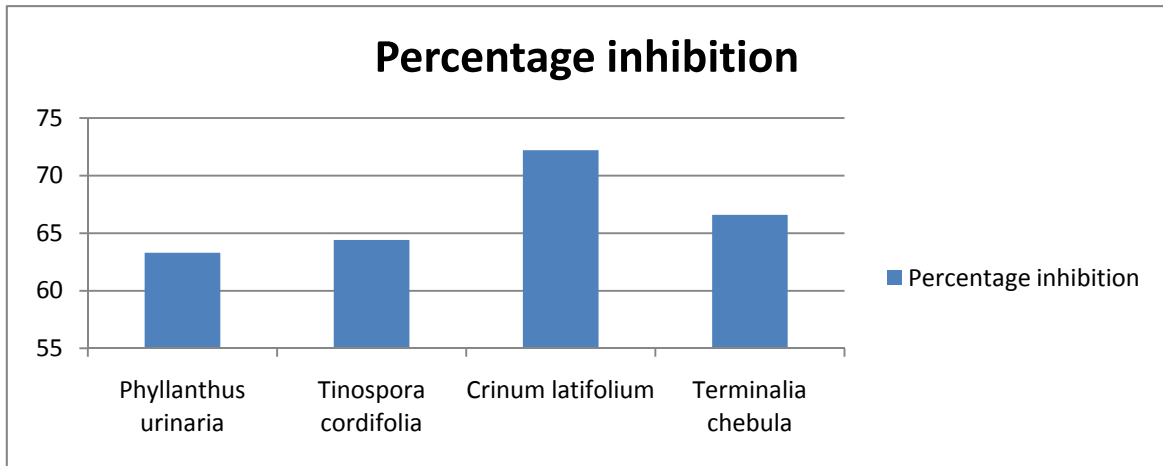


Figure-2 Percentage inhibition by plant extracts

It's quite clear from this graph plotted in figure 2, & clearly demonstrating the inhibitory effect of different plant extracts used, among all maximum controlling effect over *E. turcicum* was shown by *Crinum latifolium*. These demonstrations for inhibitory effects are given below



Control



Crinum latifolium



Four out of six Plant extracts prepared from ayurvedic plants has shown a significant controlling effect against plant fungal pathogens and also proved the usefulness of these common plants in control of Plant pathogens. This would make possibility to try these plant extract as antifungal preparation in poly house trials, so that field viability of this approach could be checked along with no harmful effect on environment and without any ecosystem interference. This experiment proves the significance of traditional Indian plants for plant disease control.

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