

Effect of herbal extract on growth of *Alternaria alternata*

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Abstract

Alternaria alternata an important fungal pathogen which mostly cause disease on aerial part of many plants worldwide. The leaf spot of soyabean caused by *Alternaria alternata* is becoming a common disease to soyabean crop. Therefore the present work undertaken to study cheaper and more environmental friendly bio compound for the control of plant disease using different herbal extract on growth of pathogen. The effect of plant extract was determined by colony diameter method using Martin Rose Bengal streptomycin agar medium. The *in vitro* antifungal activity of the aqueous and methanolic extract of five species Viz. *Ocimum sanctum* (Tulas), *Datura stromonium* (Dhotra), *Annona reticulata* (shitaphal), *Santalum album* (chandan) and *Cassia tora* (Tarota) were examined against *Alternaria alternata*. Methanolic extract of all the plants were found to have maximum antifungal activity in comparison to aqueous extract.

Key Words: *Alternaria alternata*, antifungal activity, Herbal extract.

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INTRODUCTION

In India soyabean (*Glycine max.* (L) Merrill) has been number one oil seed crop in terms of both area and production since 2005. It has shown unparallel growth over the last four decades from an area of only 30000ha and production of 14000ton In 1970. The area reached 9.95 million ha with total production of 12.57 million ton in 2011. With an average national yield of 1264kg/ha. Soyabean occupied 42% of India's total oil seed and 25% of edible oil production, the feasibility of growing soyabean crop with minimum input/management lead to the rapid expansion in area and production with the results that India now ranks 4th in terms of global Soyabean area sown and 5th in terms of soyabean production. In India soyabean is mainly grown in the state

of Madhya pradesh, Maharashtra, Rajeshtan, Karnataka, Telangana, Chittisgarh, Nagaland and Gujrat as a rainfed crop during the rainy (Kharif) season. The crop has potential of mitigating rampant protein energy malnutrition as well as becoming ideal food of the country on account of a number of neutraceutical and functional compound. currently soyabean is severely attacked about half a dozen major disease and a dozen of insects pest and several major weed, yield losses due to individual disease (A.M Sharma.et.al2014) weed species ranges from 20 to 100 % (IPM Package for soyabean NCIPM Government of India 2014). *Alternaria alternata* causes leaf spot and blight on a large variety of agricultural crop such as tomato, potato, carrot, soyabean moreover many pathogen including *Fusarium oxysporum*, *R.solani* and *Alternaria alternata* are causing severe damage to agriculture crop (Hadizadeh et.al 2009). Biological screening of plant extract was carried out throughout the world for the determination of their antifungal activity. Synthetic chemical used to control plant disease not only pollute the environment, but are also harmful to human health. Because of environmental and economic considerations, plant scientist are involved to find the cheaper and more environmental friendly biocompound for the control of plant diseases using different forms of botanicals (Khare and Shukla 1998). Medicinal plant might represent an alternative treatment

in non severe cause of infection or Disease (Gonzalez 1980) we chose plants *Ocimum sanctum*, *Datura stramonium*, *Annona reticulata*, *Santalum album* and *Cassia tora* used in folk medicine, to determine their antifungal activity against clinical pathogen *Alternaria alternata*.

MATERIALS AND METHODS

Isolation of pathogen: The fungal strain *Alternaria alternata* were originally isolated from diseased soyabean plant collected from agricultural field of Dharmabad Maharashtra region. The fungus was cultured on Martin Rose Bengal streptomycin agar plate and incubated at $28 \pm 2^{\circ}\text{C}$ for one week, purification of resulting isolates was done using the hyphal tip technique and single spore technique to obtain the pure culture. Detected isolates were then transferred into slants of Martin Rose Bengal streptomycin agar and kept at 4°C for further studies. It is then identified with microscopical examination (Ruksana et.al 2010).

Preparation of aqueous extract: Selected plants were collected from Dharmabad (Maharashtra) region and washed thoroughly with tap water and air dried. 10gm of fresh leaves were grind using mortar and pestle adding equal amount of sterilized distilled water. (1:1w/v). concentration were prepared in ranges of 5%, 10%, 15%, 20% and 25%.

Preparation of methanolic extract

The leaves of collected plants cleaned in tap and sterilized distilled water were dried at 38°C . The dried leaves were crushed to make dry powder soxhlet extraction was

made using methanol as solvent, concentration were prepared in ranges of 5%, 10%, 15%, 20% and 25%.

Antifungal activity of plant extract: The effect of plant extract taken into study against the mycelia growth of *Alternaria alternata* tested by poisoned food technique (Dhingra and Sinclair 1985). An appropriate quantity of each extract was incorporated in sterilized Martin Rose Bengal streptomycin agar to reach desired concentration of aqueous and methanolic extract of 5%, 10%, 15%, 20% and 25% were used in assessment, mycelia disc of 5mm diameter from ten days old pure culture is inoculated and different plant extract of different concentration. The plants were taken incubated at $28 \pm 2^{\circ}\text{C}$. The measurements of the mycelia growth dynamics of the fungus were recorded on a daily basis. the whole experiment was carried out for ten days until the control colonies reach the margin of the petriplate. Inhibition of radial growth was counted based on colony diameter on control plates (Sundar et.al1995).

RESULT AND DISCUSSION

Extract of all five different plants significantly inhibited mycelia growth of *Alternaria Alternata* at the tested concentration. Methanolic plant extract was found more effective than aqueous. methanolic extract of *Cassia tora* possessed potent antifungal activity amongst all the methanolic extract of other plants (61.53%) and antifungal activity of *Datura stramonium* (59.61%) *Ocimum sanctum* (56.84%) *Santalum album* (55.76%) and least found in *Annona reticulata* (36.53%).(Table 1)

Table 1: The effect of Aqueous extracts on *Alternaria alternata*

Sr. No.	Plant Extracts	Percentage of Inhibition of mycelia growth (%) respective to extract concentration				
		5%	10%	15%	20%	25%
1	<i>Ocimum sanctum</i>	26.92	22.84	36.53	44.27	50.00
2	<i>Datura stramonium</i>	23.07	26.92	30.76	45.15	55.76
3	<i>Annona reticulata</i>	34.61	36.53	40.38	42.30	44.23
4	<i>Santalum album</i>	30.76	40.38	50.00	53.84	54.76
5	<i>Cassia tora</i>	7.69	13.46	17.30	25.00	36.53

The aqueous plant extract was found less effective than methanolic. The maximum antifungal activity found in *Datura stramonium* (55.76%) followed by *Santalum album* (54.96%), *Ocimum sanctum* (50.00%) *Annona reticulata* (44.23%) and least in *Cassia tora* (36.53%). (Table 2)

Table 2: The effect of methanolic extracts on *Alternaria alternata*

Sr. No.	Plant Extracts	Percentage of Inhibition of mycelia growth (%) respective to extract concentration				
		5%	10%	15%	20%	25%
1	<i>Ocimum sanctum</i>	30.76	32.69	40.38	48.07	56.84
2	<i>Datura stramonium</i>	26.92	30.76	34.61	50.00	59.61
3	<i>Annona reticulata</i>	7.00	13.40	16.30	24.00	36.53
4	<i>Santalum album</i>	30.76	40.38	51.52	53.84	55.76
5	<i>Cassia tora</i>	36.83	44.23	53.84	57.69	61.53

The antifungal activity and mycelia inhibitory concentration of plant extract in methanolic solvent such as Soxhlet extract of plants traditionally used as medicine were as leaf extract evaluation against the clinical pathogen (*Inampudi sailaja* 2014). The present study thus states that all the plants are effective against fungal infection caused by *Alternaria alternata*.

CONCLUSION

The result obtained in this work showed that the herbal extract of different plants exhibit antifungal effect against *Alternaria alternata*. Further studies are needed to determine the chemical identity of the bio active compounds responsible for the observed antifungal activity. Natural plant derived fungicides are environment friendly and may be a cheaper source of new alternative active compounds than synthetic chemicals in particular disease control.

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