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Research Article

Studies on rhizosphere fungi and allelopathic potential of *Santalum album* L.

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Abstract

Sandal (*Santalum album* L.) popularly known for its wide medicinal and economic importance belonging to the family Santalaceae. Present research work was divided into two parts. A: Rhizosphere fungi: Serial dilution plate technique was used for isolation of rhizosperic fungi of *S. album*. In the present investigation total 71 isolates of 11 species of rhizosperic fungi were recorded. Out of 11 species 10 species belongs to Class Hyphomycetes *viz. Aspergillus niger* Van Tieghem, *Aspergillus terricola* Marchal, *Penicillium* spp., *Aspergillus terreus* Thom, *Aspergillus flavipes* Thom & Church *Aspergillus funiculosus* Smith, *Aspergillus fumigatus* Fresenius, *Aspergillus flavus* Link, *Aspergillus restrictus* Smith, *Fusarium oxyporum* Schlechtendahl *ex* Fries and one species i.e. *Mycelia sterilia* belongs to Basidomycetes. During the investigation *Aspergillus niger, Aspergillus terricola and Penicillium* spp. were frequently observed and recorded during study. B: Allelopathic potential: Laboratory experiment on the effect of leaf leachates of *S. album* on the germination of *Trigonella foenum* L. revealed that stronger the concentration of leachates greater the allelopathic effect on germination. No germination was recorded in the leachates from 25 gm (except 25 gm DLP for 4 hrs) and 50 gm of dry leaf powder (DLP). All the treatments showed significant effect of delayed in germination. The leachates of 5gm DLP for 4hrs to 24hrs reduced the growth of plumule from 4.59 cm to 3.95 cm respectively. Length of radical reduced from 0.45 to 0.20cm in the treatment 3 days after sowing (DAS) while in control, highest plumule and radicle growth was recorded i.e. 4.88 cm and 0.65 cm respectively.

INTRODUCTION

Santalum album L. is one of the most demanded trees for its heart wood, incredible flavor and fragrance commonly known as Sandal wood or White sander belonging to family Santalaceae. The wood is commercially known as "East Indian Sandalwood" and internationally reflects as "Dollar earning parasite" (Durairaj and Kamaraj, 2013).

In last 2-3 decades the demand of *S. album* has increased in domestic as well as International market due its wide array of application range. *S. album* has more than 5000 years history which witnessed by literature. India has been the traditional head of sandalwood oil production for pharmaceuticals, cosmetics and perfumery (Hansda, 2009). Sandal oil has distinctive soft, warm, smooth, creamy and milky precious–woody scent. Sandalwood oil has been widely used in traditional medicine for treatment of bronchitis, common colds, general weakness, skin disorders, heart illness, fever, urinary tract related diseases, redness of the mouth and pharynx, liver and gall bladder

complaints and other ailments (Misra, 2003).

The term Rhizosphere was first introduced by Hiltner (1904). Rhizosphere is generally defined as the narrow zone of soil directly adjacent to and affected by plant root (Jeffery *et al*., 2002). Rhizosphere is a zone of greater microbial activity than in root free soil, the relation of microbial behavior to root development and plant health is logically studied most in this region. Fungal abundance is 10–20 times higher in the rhizosphere than in the bulk soil (Morgan *et al.*, 2005). Rhizosphere fungi are more potential than rhizobacteria because of spreading through the soil (Ortíz-Castro *et al.*, 2009). *S. album* is the commercially important medicinal plant species and has considerable cultural importance.

The term allelopathy was first coined by the Australian professor Hans Molisch in 1937. 'Allelopathy' has seen considered as one component of crop interference and plant species may affect each other's growth through this mechanism (Rice, 1974)."Allelopathy" refers to the harmful effect of one plant on another plant from the release of biochemical, known as allelochemicals. Allelochemical can be present any part of plants. These allelochemicals release from plants part by leaching, residue decomposition, root exudation, volatilization and other processes in both natural and agriculture system (Willis, 1999 and Batish *et al*., 2007).

The natural population of *S. album* is decreased in several folds due to anthropogenic activities and uncontrolled exploitation from the natural habitats. It is essential to cultivate this tree on large scale following good agricultural practices (GAP). The demand for sandal wood and sandal oil has increased extensively in day today life and it will increase by several folds in future. The review of literature revealed that survival rate and essential oil yield of *S. album* is not remarkable. It is verified that, rhizosphere fungi play an important role in enhancing the essential oil content in *Ocimum tenuiflorum* L. (Khare and Pandey, 2014) and in *Mentha arvensis* L. (Ratnekumari *et al*, 2014). Host plant screening and Allelopathic study will useful for cultivation programed of *S. album.* Meager work has been done in the determine objectives. Therefore the preliminary present investigation was undertaken to study the rhizosphere mycoflora, host identification and allelopathic effect of *S. album.*

MATERIALS AND METHODS

Rhizosphere fungi:

Collection of soil samples from rhizosphere region:

For the collection of soil sample initially the plant was identified and authentified by experts in the Savitribai Phule Pune University campus. Soil samples from rhizosphere of *S. album* were collected by following the method of Dongmo and Oyeyiola, 2006. Each rhizosphere soil sample were collected by carefully uprooting the plant and shaking the soil adhering to the roots into a sterile polythene bag and brought to the laboratory within 12 hours.

Serial Dilution:

The rhizosphere mycoflora were isolated by serial dilution plate method (Johnson & Curl, 1972). One ml of each dilution was poured into each of the Petri dishes containing the above sterilized Potato Dextrose Agar (PDA) media supplemented with Streptomycin Sulphate (200µg/Lit). The cultured Petri plates were incubated at 26°C in an incubator and the colony growth was monitored.

Isolation and identification of rhizosphere fungi:

The colonies of fully-grown fungi were isolated individually and the identifications were made with the help of the relevant literature (Onions, Allsopp & Eggins, 1981 and Khulbe, 2001).

Allelopathic potential:

Experimental design and Preparation of leaf leachates-

Experimentation was conducted in randomized block design. Four replications were taken with nineteen treatments including control. The leaves of *S. album* were collected and dried at 42°C in oven. These dried leaves pulverized in grinder to made fine powder. Leachates were prepared from 5 gm, 25gm and 50 gm DLP in 250 ml of DW for 4, 8, 12, 16, 20 and 24 hrs. Effects of nineteen treatments including control were studied on germination of *T. foenum* seeds at 3DAS and 5DAS. The allelopathic effects of leachates of *S. album* were also studied on the growth rate of plumule and radical at 3, 5 and 11 DAS of seeds.

Seed Incubation

Germinating trials were conducted in autoclaved petridishes. Twenty seeds of *T. foenum* were kept on germinating paper in each petridish. From all nineteen treatments germination percentage, length of plumule and length of radical were recorded. Observations were recorded from 3rd day to 11thof soaking. The data were statistically analyzed.

RESULTS AND DISCUSSION

Rhizosphere fungi:

Isolation of rhizosphere fungi of *S.album* serial dilution was done following Johnson and Curl (1972) method. Our results showed total 71 isolates of 11 species of rhizosphere fungi. Out of 11 species 10 species belongs to class Hyphomycetes *viz. Aspergillus niger* Van Tieghem, *Aspergillus terricola* Marchal, *Penicillium spp.*,*Aspergillus terreus* Thom, *Aspergillus flavipes* Thom & Church *Aspergillus funiculosus* Smith, *Aspergillus fumigatus* Fresenius, *Aspergillus flavus* Link, *Aspergillus restrictus* Smith ,*Fusarium oxyporum* Schlechtendahl ex Fries and one species i.e. *Mycelia sterilia* belongs to Basidomycetes. (Fig.1). *Aspergillus niger* Van Tieghem, *Aspergillus terricola* Marchal and *Penicillium* spp. were frequently observed during the investigation.Wahid *et al*.(1997) were observed that *Aspergillus* and *Penicillium* are the richest amongst all the genera of class hyphomycetes found in soils of tomato field.

Allelopathy potential:

The leachates were prepared from 25gm DLP (except only in 4 hrs concentration) and 50gm DLP were recorded strong allelopathic effect on seed germination of *T. foenum.* No seed germination

Fig.1: Total no. of rhizosphere fungal isolates

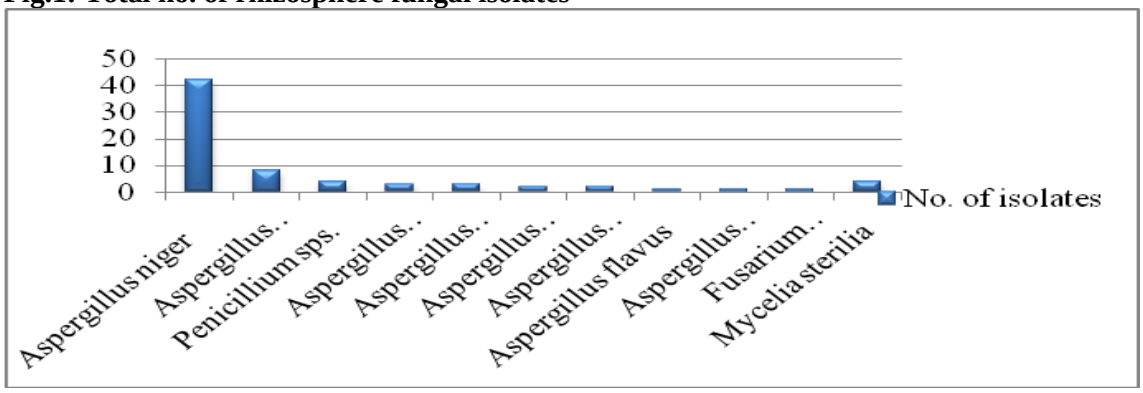

Table.1: Allelopathic effect of *S. album* leaf leachates on seed germination of *T. foenum-graecum*

Treatment	Germination%		3DAS		5DAS		11DAS	
	3DAS	5DAS	Length of Plumule (cm)	Length of Radicle (cm)	Length of Plumule (cm)	Length of Radicle (cm)	Length of Plumule (cm)	Length of Radicle (cm)
T1 (control)	85	86.25	4.88	0.65	5.08	1.18	5.88	2.02
T2 (5g DLP+250 ml 4 h)	57.5	58.75	4.59	0.45	4.98	0.62	5.66	1.17
T3 (5g DLP+250 ml for 8h)	48.75	56.25	4.53	0.34	4.58	0.52	5.01	0.83
T4 (5g DLP+250 ml for 12h)	33.75	52.50	4.24	0.33	4.35	0.48	4.86	0.7
T5 (5g DLP+250 ml for 16h)	31.25	40.00	4.12	0.24	4.15	0.41	4.68	0.64
T6 (5g DLP+250 ml for 20h)	27.5	36.25	3.99	0.22	4.14	0.38	4.48	0.57
T7(5g DLP+250 ml for 24 h)	26.6	32.61	3.95	0.20	4.08	0.37	4.17	0.53
T8 (25g DLP+250 ml for 4h)	7.5	8.75	2.73	0.20	2.926	0.34	3.023	0.41
T9 (25g DLP+250 ml for 8h)	-	-	-	-	-	-	-	-
T10(25g DLP+250 ml 12h)	-	-	-	-	-	-	-	-
T11(25g DLP+250 ml 16h)	-	-	-	-	-	-	-	-
T12 (25g DLP+250 ml 20 h)	-	-	-	-	-	-	-	-
T13 (25g DLP+250 ml 24h)	-	-	-	-	-	-	-	-
T14 (50g DLP+250 ml 4 h)	-	-	-	-	-	-	-	-
T15 (50g DLP +250 ml 8h)	-	-	-	-	-	-	-	-
T16 (50g DLP+250 12h)	-	-	-	-	-	-	-	-
T17 (50g DLP+250 ml 16h)	-	-	-	-	-	-	-	-
T18 (50g DLP+250 ml 20h)	-	-	-	-	-	-	-	-
T19 (50g DLP+250 ml 24h)	-	-	-	-	-	-	-	-
S.E.			0.654	0.285	0.236	0.095	0.203	0.184

was found in both these concentrations. Leachates from 5gm DLP in 250 ml of distilled water for 4, 8, 12, 16, 20 and 24 hrs showed decreased in the length of plumule and radical as compare to control in all three observations i.e. 3rd , 5th and 11th DAS. The seed germination recorded in control was 85% and 86.25% at 3DAS and 5DAS respectively while seed germination percentage significantly reduced in other treatments as compare to control. It was revealed that higher concentrations of leachates of *S. album* had stronger allelopathic effect of germination of *T. foenum.* (Table.1).The present results correlated with the Patil and Khade (2015). They revealed that aqueous leaf extract of *Parthenium hysterophorus* showed significant effect on germination of seed of *T. foenum* that 20% leaf extract showed lowest germination as compare to control. Mokat *et al.,* in 2005 observed that stronger the concentration of leachates of *Hyptis suaveolens*, greater the allelopathic effect on seed germination of rice.

The findings of the present investigation will be useful for further research, conservation, propagation, production and other aspects associated with *S. album.* Additionally scientific research is required to explore the maximum potential of commercially important "Dollar earning parasite".

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