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Antimicrobial activity of methanolic extract from pod, root and bark of *Delonix regia* plant

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ABSTRACT KEYWORDS

This plant occurs all over the world having the following characteristics: Height 35-40 feet with regular outline like crown having fast growth rate, leaf arrangement is alternate and bippinate type with entire margine. Leaf shape is oblong and evergreen type of leaf. Flowers are orange red in colour and very showy. Fruits are elongated pod like about 12inches or more. Initially fruit covering is green is become dry, hard and turns brown. Bark of tree is vertical thick nothrons uniform. Root of this tree is not much deep with paste resistance. © 2010 Trade Science Inc. - INDIA

Antibacterial activity;
Polarity;
Delonix regia;
Bark;
Pod.

INTRODUCTION

Plant *delonix regia* belong to family fabacae is a plant occur in all over Asia. This plant occurs all over the world having the following characteristics: Height 35-40 feet with regular outline like crown having fast growth rate, leaf arrangement is alternate and bippinate type with entire margine. Leaf shape is oblong and evergreen type of leaf. Flowers are orange red in color and very showy. Fruits are elongated pod like about 12inches or more. Initially fruit covering is green is become dry, hard and turns brown. Bark of tree is vertical thick nothrons uniform. Root of this tree is not much deep with paste resistance.

To validate antiseptic property of plant material. The plant material is extracted and studied for antibacterial and antifungal activity.

EXPERIMENTAL

Plant material and preparation of extracts

Preparatin of various extract of *Delonix regia* bark

Around 1kg of fresh shade dried bark was powdered and around 800gm were extracted by hot percolation method by soxhalate apparatus. With five liter of each pet-ether (40-60°C) chloroform, acetone and methanol successively. All the extracts finally reduced to dryness at 40degree c by rotary evaporator (Rotavapour Buchhi Switzerland).

The quantity of each extract after the extraction was pet. ether (15gm), chloroform (20gm), Acetone (12gm), Methanol (40gm), aqueous extract (28gm).

Preparation of various extract of *Delonix regia* root

Around 1kg of fresh shade dried root was powdered and around 800gm were extracted by hot percolation method by soxhalate apparatus. With five liter of each pet-ether (40-60°C) chloroform, acetone and methanol successively. All the extracts finally reduced to dryness at 40degree c by rotary evaporator (Rotavapour Buchhi Switzerland).

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TABLE 1 : Antibacterial activity of methanolic extract from bark of *Delonix regia* plant

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Bacterial	Extract ethanolic		Cefotax	Penicil	Tetrax
G (+)	In mm	In mm	In mm	In mm	In mm
Staphylococcus pidermidis	9	8	10	10	10
Staphylococcus oureus	7	12	10	8	8
Bacillus paludis	10	7	10	11	9
Bacillus subtilis	7	6	11	10	9
G (-)	In mm	In mm	In mm	In mm	In mm
Escherichia Coli	3	2	4	3.5	5.5
Pseudomonus aeruginosa	4	2	4	3	3.5
Shigella flaxinely	3	4	4.5	3.5	4
Enterobacter aero genes	3	4	5	3	2.

TABLE 2: Antifungal activity of methanolic extract from bark of *Delonix regia* plant

Fungus	Extract ethanolic	Extract CCl ₄	Cefotax	Penicil	Tetrax
Candida albicans	5	6	7	5	4
Aspergillus fumigatus	6	8	5	6	4
Aspergillus niger	5	3	6	4	5

The quantity of each extract after the extraction was pet. Ether (7gm), chloroform (10gm), Acetone (6gm), Methanol (20gm), aqueous extracts (14gm).

Preparation of various extracts of *Delonix regia* pods

Around 1kg of fresh shade dried pods was powdered and around 800gm were extracted by hot percolation method by soxhalate apparatus . With five liter of each pet-ether (40-60°C) chloroform, acetone and methanol successively. All the extracts finally reduced to dryness at 40degree c by rotary evaporator (Rotavapour Buchhi Switzerland).

The quantity of each extract after the extraction was pet.ether (20gm), chloroform (30gm), acetone (25gm), Methanol (60gm), aqueous extract (38gm).

MICROORGANISMS

The test microorganism used for antimicrobial activity screening were & bacteria (2 gram +ve)-Enterococcus faecalis, Staphylococcus onerous, (2gram -ve)-Klebsiallea pneumaniae, Escherichia coli, and two fungi- Candida albicans and Aspergillus fumigants.

These organisms were identified a procured from

TABLE 3: Antibacterial activity of methanolic extract from root of *Delonix regia* plant

Bacteria	Extract ethanolic	Extract CCl ₄	Cefotax	Penicil	Tetrax
G (+)	In mm	In mm	In mm	In mm	In mm
Staphylococcus epidermidis	12	11	10	10	10
Staphylococcus oureus	12	10	10	8	8
Bacillus paludis	10	9	10	11	9
Bacillus subtilis	12	5	11	10	9
G (-)	In mm	In mm	In mm	In mm	In mm
Escherichia Coli	8	9	4	3.5	5.5
Pseudomonus aeruginosa	10	9	4	3	3.5
Shigella flaxinely	11	9	4.5	3.5	4
Enterobacter aero genes	12	4	5	3	2

TABLE 4: Antifungal activity of methanolic extract from root of *Delonix regia* plant

Fungus	Extract ethanolic	Extract CCl ₄	Cefotax	Penicil	Tetrax
Candida albicans	13	5	7	5	4
Aspergillus fumigatus	10	9	5	6	4
Aspergillus niger	11	8	6	4	5

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Antimicrobial activity

The agar diffusion method^[11] was used to evaluate the antimicrobial activity. Bacteria were cultured overnight at 37°C in Mueller Hinton 10µl Broth (MHB, Oxoid) and fungi at 28°C for 72h in Potato Dextrose Broth (PDB, Oxide) and used as inoculums. A final inoculums, using 100µl of suspension containing 10⁸ CFV/ml of bacteria 10⁴ spore/ml of fungi spread on Mueller Hinton Agar (MHA) and Potato Dextrose Agar (PDA) medium respectively.

The disc (6 mm in diameter) was impregnated with $10\mu l$ of $75\mu l/ml$, $50\mu l/ml$, $25\mu l/ml$, $10\mu l/ml$ and $5\mu l/ml$ of each extracts and for each organism placed on seeded agar. Ciprofloxacin and Fluconazole ($75\mu l/ml$, $50\mu l/ml$, $25\mu l/ml$, $10\mu l/ml$ and $5\mu l/ml$) were used as positive control bacteria and fungi respectively. The test plates were incubated at $37^{\circ}C$ for 24h for bacteria and at $28^{\circ}C$ for 72h for fungi depending on the incubation time required for a visible growth.

MIC values were also studied for microorganisms by turbidimetric method, which were determined as sensitive to the extracts in cup plate method. MIC was defined as the lowest concentration of extract that inhibit visible growth.

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TABLE 5: Antibacterial activity of methanolic extract from pod of *Delonix regia* plant

Bacteria	Extract ethanolic	Extract CCl ₄	Cefotax	Penicil	Tetrax
G (+)	In mm	In mm	In mm	In mm	In mm
Staphylococcus epidermidis	12	9	10	10	10
Staphylococcus oureus	11	9	10	8	8
Bacillus paludis	12	6	10	11	9
Bacillus subtilis	7	6	11	10	9
G (-)	In mm	In mm	In mm	In mm	In mm
Escherichia Coli	10	5	4	3.5	5.5
Pseudomonus aeruginosa	9	6	4	3	3.5
Shigella flaxinely	9	5	4.5	3.5	4
Enterobacter aero genes	9	6	5	3	2.

A comparison is shown between the antibiotic activities of the plant extract with reference antibiotics (Cefotax). Na-cefotaxime, (Penicil) benzyl penicillin sodium and (tetrac) tetracycline R=absence of inhibition even at the hihest concentration used ($100\mu g/ml$).

Tested material

Methanolic extract was obtained after the degatting with petroleum ether $(40^{\circ}-60^{\circ})$ and CHCl₃.

Studied activities

Antibacterial activity was studied using the MIC broth dilution method^[8,9].

Organism used

For the antibacterial activity standard bacterial strains, obtained from the Nikhil Analytical Laboratory, Sangli (Maharashtra).

RESULT AND DISCUSSION

The results of Antimicrobial activity were done for all the five, pet ether, chloroform, acetone, and methanol and aqueous extracts. During antimicrobial study methanolic extracts showed maximum zone of inhibition against almost all organisms in cup plate method.

TABLE 6: Antifungal activity of methanolic extract from pod of *Delonix regia* plant

Fungus	Extract ethanolic	Extract CCl ₄	Cefotax	Penicil	Tetrax
Candida albicans	10	9	7	5	4
Aspergillus fumigatus	9	6	5	6	4
Aspergillus niger	10	5	6	4	5

CONCLUSION

The methanolic extract pods of delonix regia showed a good inhibition against all the bacterial Strains tested (MIC between $10\&80\mu g/ml$). The gram (+) bacteria were sensitive with gram (-) bacteria and some common fungi.

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