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SCREENING OF ANTI BACTERIAL AND ANTI FUNGAL ACTIVITY OF GANDHAKA DRUTI

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ABSTRACT

Antimicrobial agents are commonly used nowadays for treating various bacterial and fungal infections. But still most of the physicians are unable to treat these infections appropriately due to hindrances like development of resistance, adverse effects, patient affordability etc. Gandhaka druti formulation mentioned in Ayurvedic Texts can be an ideal replacement for treating various infectious diseases. Assessment of its antibacterial and antifungal activity may provide scientific evidence for the study. Gandhaka druti prepared with classical reference was subjected for antibacterial and antifungal activity by cup plate method. The drug was tested against bacteria like Staphylococcus aureus, Pseudomonas auregenosa and E coli and fungi Aspergillus niger, Cryptococcus neoformans, Candida albicans and Trycophytum rubrum taking Fluconazole as standard for comparison. Gandhaka druti showed a significant zone of inhibition against three strains of bacteria and four strains of fungi when compared to Fluconazole standard. Gandhaka druti showed a significant antibacterial and antifungal activity. Antibacterial activity was significant compared to antifungal activity.

KEYWORDS: Gandhaka druti, Antimicrobial agents, Flucanozole.

INTRODUCTION

Amongst the massive world of medicines, antimicrobial agents are one of the widely used and misused drugs available to medical profession. Once upon a time they were called as the magic bullets for their great contribution in curing an infection, but recently these bullets also backfire in way of causing resistance, decreasing their effectiveness and causing side effects.[1] In recent years number of antimicrobial agents has been discovered, but unfortunately even with the advent of numerous antimicrobial agents; still most of the physicians are unable to treat these infection appropriately due to hindrances while treating

these infection like development of resistance, adverse effects and, patient affordability etc. Though the centre for diseases control and prevention has taken steps to curb antimicrobial resistance, still it is flaring at a rampant rate due to over prescribing of these drugs. This over prescribing is driven largely by patient demand, time pressure on clinicians and diagnostic uncertainty, fuelling an ever-increasing need for newer drugs.[2] Making antibacterial drug therapy effective safe and affordable has been the focus of interest during recent years.[3] Some of the Ayurvedic formulations may be considered as an ideal replacement for treating certain bacterial and fungal infection. Gandhaka drutiis one such drug mentioned in Ayurvedic text having a wide range of therapeutic properties as well as prophylactic activity. It is extensively used by various physicians in different diseases like Kustha, Kasa, Shwasa, Kshaya.[4]

MATERIALS AND METHODS

To screen the antibacterial and antifungal activity of gandhaka druti the following materials were used Materials as follows:

A) Drugs:

1. Gandhaka druti
2. Fluconazole

B) Micro organisms

Bacteria		Fungi	
a)	Staphylococcus aureus	a)	Candida albicans
b)	Pseudomonas auregenosa	b)	Cryptococcus neoformans
c)	Escheria coli	c)	Trycophytum rubrum
		d)	Aspergillus niger

C) Chemicals & solvents

1. Nutrient broth
2. Nutrient agar
3. Surgical spirit
4. Distilled water

D) Equipments and Glasswares

Equipments			Glasswares	
a)	Water bath		a)	Petri dish
b)	Loops and loop holder		b)	Conical flask
c)	Borer		c)	Test tubes
d)	Hot air oven		d)	Beakers

e)	Inoculation hood		e)	Funnel
f)	Autoclave		f)	Stirrer
g)	Incubator			

MATERIALS AND METHODS

Antibacterial and antifungal activity was carried by cup-plate method.[5,6]

Identification of cultures was done under microscopic examination

Methods

A. Preparation of solution

a. test drug

5 ml samples of Gandhaka druti was taken as sample 1.

2.5 ml samples of Gandhaka druti was taken as sample 2.

1.25 ml samples of Gandhaka druti was taken as sample 3.

b. Preparation of standard solution

100 mg Fluconazole tablet was dissolved in 100ml distilled water and used as standard drug for this study.

B. Preparation of Growth Media

Nutrient broth was used for the preparation of growth media. Nutrient broth 13 gm was dissolved in 1000ml of distilled water, boiled for 15min and allowed to cool. 100ml was then transferred to each conical flask and sterilized in autoclave at 15 lbs pressure (i.e 121^0c) for 20min.

C. Preparation of Inoculums

A loopful of the organisms was emulsified in 100ml sterile growth media under proper sterile conditions and incubated for 72 hrs at 37^0c in incubator.

D. Preparation of Agar Media

Nutrient agar was used for the preparation of agar media. Nutrient agar 28gm was dissolved in 1000ml of distilled water. It was boiled for 20min and allowed to cool. Then it was sterilized in autoclave at 15lbs pressure for 20min.

E. Preparation of Agar plates

5ml of inoculums prepared was added to 45ml of flask containing nutrient agar at 37^0 c. This was immediately poured into a dry sterile petridish to a depth of 5mm. The petridishes were

placed on a leveled surface to ensure that the layers of medium are of uniform thickness. Allow the plates to solidify at room temperature for 12hrs. Incubated some plates at 35^0c to check sterility. The surface of the agar layer was kept dry before use. With the help of sterile borer (diameter 8mm) cylinders were made in agar plates. A uniform volume of 3 samples of Gandhaka druti and standard drug Fluconazole were added to each cavity, sufficient enough to fill the holes. After 30min agar plates were incubated at 37^0c for 72hrs. Zone of inhibition was measured after 72hrs using mm scale. The diameter of the circular zone is the measurement of the zone of inhibition.

Interpretation of Results

A) In General

Results were interpreted by measuring the zone of inhibition shown by samples on test organisms.

a) Sensitive (S) Zone – Diameter wider than 8mm.

b) Intermediate (I) Zone – Diameter between 6mm to 8mm.

c) Resistant (R) Zone – No zone of inhibition or diameter less than 6mm.

OBSERVATIONS AND RESULTS

Test drugs	S.A	P.A	E.C	T.R	A.N	C.N	C.A
SAMPLE 1	30	28	26	22	25	20	20
SAMPLE 2	22	18	20	16	20	14	14
SAMPLE3	18	14	16	12	18	12	10
FLUCANOZOLE	-	-	-	20	22	22	20

S.A-Staphylococcus Aureus, P.A- Pseudomonas auregenosa, E.C- Escheria coli, T.R-Trycophytum rubrum, A.N- Aspergullus niger, C.N- Cryptococcus neoformans, C.A- Candida albicans.

DISCUSSION

Gandhaka druti sample1 showed similar antifungal activity compared to Fluconazole against Candida albicans and Cryptococcus neoformans, but it was more significant against Trycophytum rubrum and Aspergillus Niger than Fluconazole. sample 2&3 also shown significant zone of inhibition against bacterial and fungal organism Gandhaka druti consists of Sulphur along with other herbal ingredients. Gandhaka is mentioned as Krimighna in ancient Ayurvedic texts. Sulphur has been associated as an important constituent in Sulphonamides, which are used as antimicrobial agents. These groups of drugs have been proved to act by inhibiting Folic acid metabolism in the susceptible bacteria and preventing

their growth. Thus Sulphur in Gandhaka druti also might have same mechanism of antimicrobial activity. Sulphur ointment topically was widely used as a scabicide and pediculocide, because one of the metabolite of Sulphur i.e. Pentathionic acid, which was suppose to cause lysis of cuticle. This proves that Sulphur is effective both systematically and topically as a microbicidal agent.[7] Gandhaka present in druti has been detoxified with ancient process mentioned in Rasashastra text, so that the irritant and toxic effect of Sulphur is reduced. At the same time other ingredients like shundi maricha pippali, tila thaila. have attributed additional therapeutic properties and proved to have antimicrobial activity. Sulfonamides and Fluconazole act only as antimicrobial agents and may produce adverse effects on human beings but Gandhaka druti not only act asantimicrobial agent but have additional properties like rejuvenation and promotespositive health and vigour by increasing the immunity, thus making the body resistant against disease causing factors.

CONCLUSION

This study has contributed for the evidence base to rationality of using Gandhaka druti as antibacterial and antifungal drug (Krimighna). Sulfonamides and Fluconazole act only as antimicrobial agents and may produce adverse effects on human beings but Gandhaka druti not only act as antimicrobial agent but have additional properties like rejuvenation and promotes positive health and vigour by increasing the immunity, thus making the body resistant against disease causing factors. Gandhaka druti has demonstrated significant antibacterial and antifungal activity, which gives further scope for experimental and clinical study on various microorganisms.

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