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EFFECT OF MIXING ANTIBIOTICS WITH SOME HERBAL COMPONENTS (*ALOE VERA*) ON THEIR ANTI-MICROBIAL ACTIVITIES PIYUSH VYAS, ARVIND SUTHAR, BHARGAV DAVE & MEDHA PRAJAPATI SHETH M N SCIENCE COLLEGE, PATAN

ABSTRACT:

In the present study we discuss about Phyto chemical Analysis of Herbal Drugs with brief work on Anti-Bacterial Activity of Herbal and Allopathy combinations By Disc, Broth and Streak Methods and its HPTLC with Bio autography. We report that Aloe vera has an antimicrobial activity against common pathogens, but when it is combined with Antibacterial drug like Roxythromycin, Cefixime and Levofloxacin the combinations help in - inhibiting growth of Staphylococcus aureus, Staphylococcus epidermis, Escherichia coli and Bacillus subtilis. However, these combinations have powerful effect against bacteria with less side- effects. The medicinal importance of the Aloe vera in the prevention of aerobic and anaerobic bacterial infections is obvious considering the growing number of these developing resistance organisms to conventional antibiotics. Phytochemical Analysis of the Aloe vera helps to find out the presence of chemical constituents in the plant extract. In the present paper, the status on the above mentioned combinations has been discussed.

KEY WORDS: Aerobic and anaerobic bacteria, Antibacterial activity, Plant extract, Aloe vera, Roxythromycin, Levofloxacin, Cefixime

INTRODUCTION:

A methanolic extract of Aloe vera showed significant in vitro antibacterial efficacy against Escherichia coli, Staphylococcus aureus, Staphylococcus epidermis, Basillus subtilis and Shigella flexneri(Dhar ML,1968).Roxythromycin (BP) ,the second generation antibiotic is a macrolide antibiotic(Eltenza JM et al,1994).with a broad and essentially bacteriostatic action(Godkar Praful B.,2003). against many gram positive and gram negative bacteria like staphylococci, streptococci, Bacilus spp., E coli, Neisseria spp. etc.(Jain PC,1991). Cefixime (USP), the third generation cephalosporin (Kokate C.K ,2001).gives broad spectrum cephalosporin effective against many gram negative bacteria like staphylococci, streptococci, Ecoil, Basillus spp., Klebsiella spp. etc.(Licata, et, al,1997). Levofloxacin (BP), the third generation antibiotic which gives bacteriostatic effect against gram positive and gram negative

bacteria(Langtry HD,1994). like staphylococci, streptococci, Bacilus, Klesiella, Proteus, E. coli etc.(Morrissey George J.,1997)(Mukharjee Kunal L.,2002).

MATERIALS AND METHODS:

Phytochemistry Of Aloe vera

The qualitative analysis tests were carried out by several methods.

1 Alkaloids

Dried powder of herb treated with 5% Ammonical Ethanol and the test carried out after 48 hours, The fraction was treated with Mayer's ,Wagner's and Dragndroff's reagent.

2 Tannin's & Saponins

5gmplant material in 50ml H2O and boiled for 45 min. in waterbath and 2% gelatin solution is added dropwise.

3 Proanthocyanidin 2gm of herb in 5ml. of 2N HCL and kept in water bath for 30minuts. Then the mixture was cooled and shaken with amyl alcohol.

4 Iridoids

1gm. Of herb in 5ml. of aqueous HCL and kept for 3-6 hours 0.1 ml. from the decant macrate and treated with trim Hill reagent.

5 Flavonoids

1gm of herb in 10ml. of 95% ethanol and kept in boiling waterbath for 15 minutes and after filtration mg ribbon were added along with 2-3 drops of HCL.

6 Steroids

1gm of herb was extracted with methanol for 15minuts and then Libermann Burchard reagent was added dropwise.

Plant extract :

0.5 gm of the herbal powder samples were separately taken in conical flasks along with 10ml of Methanol. The mixture was then allowed to stand overnight and after that the extract was filtered out. This procedure was respected thrice. The solids obtained were reconstituted such that the final concentration set was 100mg./ml. till complete extraction was ensured.

Preparation of combinations :

Antibiotic such as Roxythromycin, Cefixime and, Levofloxacin were dissolved in H2O. Various mixtures were made in combination with the plant extract. Details of which can be seen in the table -2.

Sample No.	Combination
A1	Aloe vera (0.500mg) + Methanol (10ML)
A2	A1 + 1ml Roxythromycin
A3	A1 + 1ml Cefixime
A4	A1 + 1ml Levofloxacin

Table – 1 :- Sample Preparation

Bacterial cultures

Four bacterial species namely Staphylococcus, aureus, Staphylococcus epidermis, Escherichia coli and, Bacillus subtilis were inoculated in Nutrient broth and allowed to grow overnight at 37^oC. This freshly grown cultures was used for analysis.

Antibacterial activity by Disc Method

The paper Disc plate method is the most commonly used technique for determining susceptibility of microorganisms to know concentration of antibiotics. Small paper Disc impregnated with combination extraction agents were placed upon the surface of pre inoculated plate.(Majumdar Dr. Ashok,2002). The plates were incubated at 37^{0} C for 24 hours. Susceptibility of effectiveness was observed by the diameter of the inhibition zone around the Disc. Organisms which grew up to the edge of the disc were resistant.

RESULTS AND DISCUSSION:

Table-2 :- Phytochemical Analysis

Phytochemical group	Inference
Alkaloids	+
	++
Tannins	+
Saponins	+
Proanthocyanidins	
Iridoids	
Flavonoids	
Steroids	+
+ Less presence	
++ Medium presence	

- +++ ---- Complete presence
- ____ ---- No presence

Sample No.	Zone mm	Interpretation	Zone mm	Interpretation	Zone mm	Interpretation	Zone mm	Interpretation
A1	6	R	8	R	6	R	6	R
A2	28	S	24	S	20	S	28	S
A3	24	S	28	S	12	I	22	S
A4	22	S	20	S	6	R	22	S
A1, A2, A3, A4 - as shown in Table- 3 Zone								

Table-3:-Antibacterial Activity By Disc Method

		A1, A2, A3, A4
<8	R -	Resistant
8 to 16	I-	Intermediate
>16	S-	Sensitive

Antibacterial activity by streak plate method

Petri plate containing Nutrient Agar was seeded with the combination mixture and allowed to solidify. Overnight grown culture were than taken one by one and streaked on the plate.(Neu HC,1987).

A control plate continuing only nutrient agar devoid of antibiotic mixture was simultaneously streaked and incubated.

The growth was observed against the control plate.

Sample No.	Organism	100mg/ml	250mg/ml	500mg/ml	750mg/ml	1000mg/ml
Al	1	++	++	++	++	++
	2	+++	+++	+++	+++	+++
	3			++		++
	4	+	++	++		++
A2	1	+++	+++	+++	+++	+++
	2	+++	+++	+++	+++	+++
	3	+++	+++	+++	+++	+++
	4	+++	+++	+++	+++	+++
A3	1	++	++	++	++	++
	2	+	++	++	+	+
	3	++	+	++	++	++
	4	++	+	+	++	++

Table – 4 :- Antibacterial Activity By Streak Plate Method

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PEER-REVIEWED Page | 373

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A4	1		++	+++	++	+++
	2	+	++	++	++	+++
	3	++	++	++	+++	++
	4	++	++	++	+++	++

- A1, A2, A3, A4 as shown in **Table-2**
- 1 Staphylococcus aureus
- 2 Staphylococcus epidemis
- 3 Escherichia coli
 - 4 Bacillus subtilis
- + Less inhibition++ Medium inhibition
- +++ Complete inhibition
 - --- No inhibition

Alternative complementary medicine now introduced in medical school curriculum teaches the benefits of non –traditional therapies such as herbal medicine(Patel Rakesh J. ;2000). According to phytochemical analysisTable-1, we can say that Aloe vera has more than 150 chemical constituents but the major chemical constituent is Aloin, which is a mixture of glucosides among which barbaloin is the chief constituent.(Ross Ivan A.2;000). Aloe vers has steroids like campesterol, lupeol, cholesterol, sotosterol.(Bogaard.M.P., ;1985).

According to table -3, Aloe vera gives slow effect on bacteria but when it is combined with Roxythromycin, Cefixime and Levofloxacin the mixture gives high zone of inhibition. Although Aloe vera gives high effect on Staphylococcus epidermis and Escherichia coli the effect become higher when combine.

The Table -4 shows different concentrations of single herbal and combination. It helps to find out the perfect concentration of drugs which gives highest inhibition of bacteria.

CONCLUSION :

We live in an era of rapidly emerging antimicrobial resistant pathogens and therefore a wide search for alternative remedies to prevent and cure infection are needed.

The allopathic forms of antibacterial drugs are available since long time but they have many side effects. For e.g. Roxythromycin gives adverse effects like gastrointestinal disturbances, increase liver enzymes and hepatitis, headache, weakness, etc.(Stone Jhons, 1987). Cefixime gives adverse effects like diarrhea, skin rashes, eosinophilia ,fever (Walters Kluke co, 2000). haemolytic

anaemia, etc. Levofloxacin gives adverse effects like nausea, vomiting(Williamson Elizabeth M.,2002), abdominal pain and dyspepsia.(Yasuda H et al ,1993).

Due to the high range of adverse effect, we need an antibacterial which has lower side effects and high effectiveness. The combination of high amount of herbal like Aloe vera and low amount of Allopathy drugs like Roxythromycin, Cefixime and Levofloxacin gives new path in medicinal world.

It is a new concept to combine herbal and Allopathy drugs to be known as herbo- Allopathy combinations.

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REFERENCES:

Ahmed V., Noor A., Hamdard Medicus, 40(4)5-9,1997

- Anon., Aloe Markets levels off. Herbal Gram, 17:3, 1988
- Bogaard M.P., Report on the analysis of Aloe vera gel. Friendship Aloe vera Pvt.Ltd.,1985
- Chopra R.N., Nayar S.L., Chopra I.C., Glossary of Indian Medicinal Plants, Council of Scientific and Industrial Research, 1992
- Dhar ML. Dhar MM. Dhawab BN, Screening of Indian medicinal plants for biological activity *American Botanical Journal*,232, 1968.
- Eltenza JM et al, Levofloxacin a review of its antibacterial activity, pharmaco kineties and therapeutic efficacy Drug, *Philosophical Analysis in latin America*, 677-700, 1994.
- Licata et al, comparision of postantibiotic and Staphylococcus aureus, Staphylococcus epidermis, *American Society For Microbiology*, 41, 1997.
- Langtry HD, Lamb HM Levofloxicn its use in infections of the respiratory trats, Martindale-The Complete Drug Refrence,33rd ed,219,1993
- Morrissey George J., The bacterial activity of Levofloxacin against bacteria, J Antimicrob Chemother, 39:719-23: 1997.
- Godkar Praful B., Textbook of Medicinal Laboratory Technology, Bhalani Publication House, 2nd ed 489-500, 2003.
- Grindlay D. Ans Reynolds, The Aloe vera Phenomenon: A review of the properties and modern use of leaf parenchyma gel . *J.Ethnopharma*, 16:117-151,1986

- Jain PC ,Ahmed S, Chemical Examination of Aloe vera, *Bulletin of Medico Ethnobotanical* research, 12(3-4):167,1991.
- Kokate C.K., Purohit A.P., Gokhale S.B., Pharmacognosy by C.K.kokate, Nirali Prakashan,181-183, 2001.

Majumdar Dr. Ashok, Home remedies in Ayurveda, Amar Granth Publications, Delhi, 390, 2002.

- Neu HC, Invitro activity of new broad spectrum cephalosporin, Cefixime, *Pediatr Infect Dis* J,6:954-7: 1987.
- Patel Rakesh J., Patel Kiran R., Experimental Microbiology ,Vol-2,Aditya publication, 72-96, 2000.
- Ross Ivan A., Medicinal plants of the world ,Totova,NJ;Humana Press, Vol-2, 321, 2000.
- Stone John clinical isolates in -vitro activity of cefixime, J Antimicrobe Chemother, 23:221-8, 1989.
- Walters Kluke co, The Review of Natural Products by facts and comparisons, St.Louis, MO, USA, Recent Progress in Medicinal Plants, Vol-6, 2000.

Williamson Elizabeth M., Major herbs of Ayurveda, Churchil Livingstone Publications, 40, 2002.

- Mukharjee Kunal L., Medicinal Laboratory Technology, Vol-2,McGraw-Hill Education(India) Pvt.Ltd, 600-604, 2002.
- Yasuda H et al , Interaction of antimicrobial agents, Biological Research Laboratory, Sankyo Co.Ltd, Tokyo, Japan ,37, 1993.