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Phytochemical analysis, *in-vitro* screening for antimicrobial and anthelmintic activity of combined hydroalcoholic seed extracts of four selected folklore indian medicinal plants

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ABSTRACT

Antimicrobial properties of medicinal plants are being increasingly reported from different parts of the world. The main aim of present research work is to investigate Invitro antimicrobial activity, anthelmintic activity and Preliminary screening for phytochemical constituents of the hydroalcoholic extract of Coriandrum sativum, Cassia occidentalis, Carica papaya, Moringa foetida). Anti-microbial assay was evaluated using Nutrient agar medium and Disc diffusion method at different concentrations (50,100,150,500mg/ml) and the results were compared with therapeutically used antibiotics. Rapid formation of Inhibition zones within 24 hours of incubation was obtained with Hydroalcoholic extract 50mg/ml. Anthelmintic activity was evaluated using adult Indian earthworms, Pheretima posthuma and Tubifex tubifex, having anatomical and physiological resemblance with intestinal round worms parasite. The earthworms were washed in normal saline solution before they were released in to 10ml of respective formulation as follows, vehicle (2%v/v Tween 80 in normal saline) and piperazine citrate (10mg/ml)and prototypes(10,20,50mg/ml). All the Investigations on Hydroalcoholic extract exhibited Antimicrobial and Anthelmintic activity at 20mg/ml with significant activity (P < 0.05). The MIC of Hydroalcoholic was determined using the parameters like time of paralysis and time of death with the extract at concentrations of 10,20,50mg/ml.The extract exhibited significant anthelmintic activity at highest concentrations of 50mg/ml as compared with piperazine citrate (10mg/ml) as standard reference and distilled water as control. Herbal drugs and synthetic drugs were equally effective in helminthic infestations but Hydroalcoholic extract of Indian medicinal plants exhibits potentiality and have maximum anthelmintic activity.

Key words: Combined hydroalcoholic seed extract, Antimicrobial, Anthelmintic, Bioassay.

INTRODUCTION

Parasitic infections are common in the tropical regions. Parasites that infect humans are of various protozoans and helminthes. Parasitic diseases disable their hosts and render them incapable of leading normal lives. In certain cases they cause mortality of the affected human hosts. Bioactive plant metabolites may offer cheap, cost effective and easily affordable drugs against parasitic infections.combined Hydro alcoholic extracts of four selected Indian medicinal plants were screened for their Antibacterial and Anthelmintic activities.

Since ancient times, plants and herbal preparations have been used as medicine. Research carried out in last few decades has certified several such claims of use of several plants of traditional medicine. Traditional system of

medicine and folklore claiming that medicinal plants as a whole or their parts are being used in all types of diseases successfully including antibacterial and anthelmintic, anti-inflammatory etc. As we know very well, now a days the medicinal preparation available in the market from which most of them either not effective up to the mark or has to develop resistance resulting in reoccurrence again. Plant derived drug serve as a prototype to develop more effective and less toxic medicines.Helminthiasis is among the most important animal diseases inflicting heavy production losses. The disease is highly prevalent particularly in third world countries due to poor management of Helminthiatic practices . A number of medicinal plants have been used to treat parasitic infections in man and animals.The plants are known to provide a rich source of botanical anthelmintics. The anthelmintic assay was carried as per the method of Ajaiyeoba *et al.* with minor modifications[1]. The assay was performed on adult Indian earthworm, *Pheretima posthuma* and *Tubifex tubifex* due to its anatomical and physiological resemblance with the intestinal roundworm parasite of human beings. Because of easy availability, earthworms have been used widely for the initial evaluation of anthelmintic compounds *in vitro*. Current research aims to highlight and prove traditional anthelmintic medicinal properties of plants with a view to focus on future studies.

1.1 Occurrence, Botanical description ,Ethnopharmacology and Phytochemistry

Coriandrum sativum L. (Apiaceae) is an erect annual herb 20–70 cm tall with strong smell. It is widespread throughout the world as a result of cultivation for its aromatic seeds.Extracts from seeds of *Coriandrum sativum* have several pharmacological effects such as anti-fertility, anti-diabetic, antihyperlipidemic, antioxidant, and hypotensive activities [2].Earlier Phytochemical screening indicated the presence of chemicals such as quercetin 3-glucoronide, linalool, camphor, geranyl acetate, geraniol and coumarins. The major fatty acids were petroselinic acid (65.7% of the total fatty acid methyl esters) followed by linoleic acid, isocoumarins, coriandrones C–E, from whole plants of *Coriandrum sativum*. Two types of 2-C-methyl d- erythriol glycosides were also recently isolated from the seed of *Coriandrum sativum*.

Cassia occidentalis L. (Caesalpiniaceae) is an Ayurvedic plant with important medicinal values. It is known by various names, e.g. Coffee senna, fetid cassia, and Negro Coffee (English). In India it is known by its various vernacular names, the most commonly used ones are Kasamarda, Kaasaari (Ayurveda). Coffee senna grows throughout the tropics and subtropics.In India, C.*occidentalis* is a common weed found throughout India (up to an altitude of 1500 m) from Jammu and Kashmir to Kanyakumari. C.*occidentalis* is an erect, somewhat branched, smooth, semi-woody, fetid herb or shrub, 0.8–1.5 m tall, taproot, hard, stout, with a few lateral roots on mid section. This plant species varies from a semi-woody annual herb in warm temperate areas to a woody annual shrub or sometimes a short-lived perennial shrub in frost free areas .The main plant chemicals in C.*occidentalis* include: achrosin, aloe-emodin, emodin , anthraquinones , anthrones, apigenin, aurantiobtusin, campesterol, cassiollin, chryso-obtusin, chrysophanic acid, chrysarobin, chrysophanol , chrysoeriol, emodin, essential oils, funiculosin, galactopyranosyl, helminthosporine[3].Other chemical constituents like islandicine, kaempferol, lignoceric acid, linolenic acid, mannitol, mannopyranosyl, matteucinol, obtusifolin, obtusin, oleic acid, physcion , quercetin, rhamnosides, rhein, rubrofusarin, sitosterols, tannins, and xanthorine have been reported [4].

Carica papaya (Caricaceae) is a tree-like herbaceous plant, a member of the small family Caricaceae and widely cultivated for its edible fruits. It originates in the lowlands of all tropical countries and many subtropical regions of the world. Parts of the plant are used in tropical diets as a fruit or vegetable; it is sometime used as a therapeutic remedy for several of its medicinal properties. Papaya fruit is thought to contain some immuno-stimulating and anti-oxidant agents[5]. The immature fruits and roots are used for their abortifacient activity, the seeds are now being used as a potential post-testicular anti-fertility drug ,the pulp is used by African hospitals for treating wounds and burns ,the latex and the seeds are used in the care of gastrointestinal nematode infections and they have shown anthelmintic activity and the seeds and immature fruit have shown bacteriostatic activity against the human enteric pathogens .The leaves are used to relieve the symptoms of asthma and as a vermifuge, in the treatment of gastric problems, fever and amoebic dysentery. Methanolic leaf extract demonstrated vasodilatatory and anti-oxidant effects, both implicated in the reduction of cardiovascular risks. The aqueous extract showed beneficial effects for the acceleration of wound healing processes in rats .

Momordica foetida (*Cucurbitaceae*)is a climber commonly found in swampy areas. It has medicinal uses ranging from spiritual and psychiatric conditions to physical diseases. Drinking of aqueous leaf extracts of the plant for malaria treatment is reported in East and Central Africa [6]. Other medicinal uses of extracts of the plant include the treatment of hypertension, peptic ulcers, diabetes mellitus, and as a purgative. In India, various medicinal properties are claimed for *Momordica foetida* that include antidiabetic, abortifacient, anthelmintic, contraceptive, antimalarial

and laxative and is used for treatment of dysmenorrhea, eczema, emmenagogue, galactagogue, gout, jaundice, kidney (stone), leprosy, leucorrhea, piles, pneumonia, psoriasis, rheumatism and scabies It contains biologically active chemicals that include glycosides, saponins, alkaloids, fixed oils, triterpenes, proteins and steroids [7]. The immature fruits are a good source of Vitamin C and also provide Vitamin A, phosphorus, and iron .Several phytochemicals such as momordenol, momordicilin, momordicins, momordicinin, momordiol, cryptoxanthin, cucurbitins, cucurbitacins, cucurbitanes, cycloartenols, diosgenin, elaeostearic acids, erythrodiol, galacturonic acids, gentisic acid, goyaglycosides, goyasaponins, multiflorenol, have been isolated [8]. These are reported in all parts of the plant. The hypoglycemic chemicals of plant are a mixture of steroidal saponins known as charantins, insulin-like peptides and alkaloids and these chemicals are concentrated in fruits, therefore fruit has shown more pronounced hypoglycemic/antihyperglycemic activity. However, two types of hypoglycemic substances have been differentiated with different time dependent effects-one with fast antihyperglycemic activity of around 1 h present in the aqueous and the residue after alkaline chloroform extraction of aqueous extract and another with a slow hypoglycemic activity in acidic wash of the chloroform extract remaining after an alkaline water wash. HIV inhibitory proteins like MRK29 (MW: 28.6 kDa), ssssssMAP30 (MW: 30,000 kDa) and lectin are documented [9]. The presence of trypsin inhibitors, elastase inhibitors , guanylate cyclase inhibitors and alphaglucosidase inhibitor like D-(+)-trehalose are also reported [10].

MATERIALS AND METHODS

Plant material

Fresh seeds of plants *Coriandrum sativum*, *Cassia occidentalis*, *Carica papaya*, *Moringa foetida* were collected from Aswini Herbal Garden, Hindu College of pharmacy, Guntur, Andhra Pradesh (A.P), India. Seeds were washed thoroughly with distilled water and then shade dried. All the dried seeds were wholly powdered with the help of mixer grinder and further used for

extract preparation.

2.1 Preliminary Phytochemical Screening

The extracts were subjected to preliminary phytochemical testing to detect for the presence of different chemical groups of compounds. Air-dried and powdered Seed materials were screened for the presence of tannins, alkaloids, flavonoids, triterpenoids, steroids, proteins, Aminoacids and glycosides [11-13].

2.2 Preparation of Plant Extract

Extraction

The extraction of the seeds of four prominent plants was carried out using known standard procedures[14]. The seeds were dried in shade and powdered in a mixer grinder. The powder (25.0 g) of the seeds were initially defatted with petroleum ether (60-80°C), followed by 900 ml of hydroalcohol (30:70) by using a Soxhlet extractor for 72 hours at a temperature not exceeding the boiling point of the solvent. The extracts were filtered using Whatman filter paper (No.1) while hot, concentrated in vacuum under reduced pressure using Rotary flask evaporator, and dried in a desiccator. The hydroalcoholic extract yields a dark brown solid residue weighing 5.750 g (23.0% w/w). More yields of extracts were collected by this method of extractions. The dry weight of the Seeds extract was obtained by the solvent evaporation and used to determine concentration in mg/ml. The extract was then kept in sterile bottles, preserved at 2- to 4°C under Refregirated conditions until further use . This crude hydroalcoholic extract was then used to investigate further for potentials of Antimicrobial and Anthelmintic properties.

3. Antimicrobial screening

Test Microorganisms and Growth Media

The following microorganisms *Staphylococcus aureus* (MTCC 96), *Streptococcus pyogenes* (MTCC 442), *Escherichia coli* (MTCC 443), *Pseudomonas aeruginosa* (MTCC 424) and fungal strains *Aspergillus niger* (MTCC 282), *Aspergillus clavatus* (MTCC 1323), *Candida albicans* (MTCC 227) were chosen based on their clinical and pharmacological importance[15-19]. The bacterial strains obtained from Institute of Microbial Technology, Chandigarh, were used for evaluating antimicrobial activity. The bacterial and fungal stock cultures were incubated for 24 hours at 37°C on nutrient agar and potato dextrose agar (PDA) medium, respectively, following refrigeration storage at 4°C. The bacterial strains were grown in Mueller-Hinton agar (MHA) plates at 37°C (the bacteria were grown in the nutrient broth at 37°C and maintained on nutrient agar slants at 4°C), whereas the yeasts and molds were grown in Sabouraud dextrose agar and PDA media, respectively, at 28°C. The stock cultures were maintained at 4°C.

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3.1 Determination of zone of inhibition

In vitro antibacterial and antifungal activities were examined for combined hydroalcoholic extract. Antibacterial and antifungal activities of seed extracts against four pathogenic bacteria (two Gram-positive and negative) and three pathogenic fungi were investigated by the agar disk diffusion method[20-23]. Antimicrobial activity testing was carried out by using agar cup method. Combined hydroalcoholic extract were dissolved in Dimethyl sulfoxide, sterilized by filtration using sintered glass filter, and stored at 4°C. For the determination of zone of inhibition, pure Gram-positive, Gram-negative, and fungal strains were taken and antibiotic as a standard for comparison of the results. The extract were screened for their antibacterial and antifungal activities against the Escherichia coli, Pseudomonas aeruginosa, Staphylococcus aureus, Streptococcus pyogenes and the fungi Candida albicans, Aspergillus niger, and Aspergillus clavatus[24-28]. The test compound (CHASE extract of CCCM) of different concentrations ranging from 50, 100, 150 and 200 μ g/6 mm disc was introduced into the well and the plates were incubated at 37 ⁰C for 12h. Control experiments were carried out under similar condition by using ampicillin, chloramphenicol, ciprofloxacin, and norfloxacin for antibacterial activity, nystatin and griseofulvin for antifungal activity as standard drugs [19,20]. The zones of growth inhibition around the disks were measured after 18 to 24 hours of incubation at 37°C for bacteria and 48 to 96 hours for fungi at 28°C. The sensitivities of the microorganism species to the plant extracts were determined by measuring the sizes of inhibitory zones (including the diameter of disk) on the agar surface around the disks, and values <8 mm were considered as not active against microorganisms[29-32]. Antibacterial and antifungal potential of extract were assessed in terms of zone of inhibition of bacterial growth. The results of the antibacterial and antifungal activities are presented in Table.I-IV

4. Anthelmintic Investigation

Drugs and Chemicals

Saline water (Claris life sciences Ltd, Ahmedabad) and Piperazine citrate (pure drug) used as standard and seed extract were prepared as per standard procedure vehicle (2% v/v Tween 80 in distilled water were used. The Prototype was dissolved in minimum quantity of 2% v/v Tween80 and then the volume was adjusted to 10 ml with normal saline for making the concentration of 10, 20 ,50 mg/ml).

4.1 Experimental Model

Indian earthworm *Pheretima posthuma*, were used to study the anthelmintic Activity (Annelida) were collected from the water logged areas of soil at Tadikonda, Guntur district *Tubifex tubifex* (Annelida) were collected from Aquarium of the local market. The average size of *Pheretima posthuma* and *Tubifex tubifex* were 6-8 cm and 1-1.5 cm respectively. Worms were authenticated by Dr.prakash, Parasitologist, Department of Microbiology, Acharya Nagarjuna university, Andhra Pradesh, India and washed with normal saline to remove all the fecal matter, waste surrounding their body. The earth worms (Pheretima posthuma) 6-8 cm in length and 0.1-0.2 cm in width weighing 0.8-3.04 g were used for all experiment protocols. The earthworms resembled the intestinal roundworm parasites of human beings both anatomically and physiologically and hence where used to study the anthelmintic activity[33-38].

Indian adult earth worm 6-8 cm in length and 0.1 - 0.2 cm in width were used for the invitro anthelmintic bioassay of combined Hydroalcoholic seed extracts of four medicinal plants. The worms were divided into the respective groups containing six-earth worms in each group[39-45]. The extract was dissolved in minimum quantity of 2% v/v Tween 80 and the volume was adjusted to 10 ml with normal saline for making the concentration of 10, 20and 50mg/ml. All the prototypes and the standard drug solution were freshly prepared before commencement of the experiments. All the earthworms were washed in normal saline solution before they were released into 10ml respective formulation as follows, vehicle (2% v/v Tween 80 in normal saline), and Piperazine Citrate (10 mg/ ml) and prototypes (10, 20 and 50 mg/ml) the anthelmintic activity was determined. 10 ml formulations containing three different concentrations of Hydroalcoholic extract (10,20 and 50 mg/ml in double distilled water) were prepared and taken in different petridishes and six earthworms (same type) were placed in the solutions respectively. Similarly lump of *Tubifex* worms were placed in the test solutions. They were observed for their spontaneous motility and evoked responses. Observations were made for the time taken to paralysis and death of individual worms. Paralysis was said to occur when the worms do not revive even in normal saline. Death was concluded when the worms lost their motility followed with fading away of their body color. Time for death of worms were recorded after ascertaining that the worms neither moved when shaken vigorously nor when dipped in warm water at 50°C.

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5. Statistical Analysis

All results are expressed as mean \pm SEM Groups of data was compared with analysis of variance (ANOVA). Values would be considered statistically significant, when P<0.05.

RESULTS

Anti bacterial activity

The antimicrobial activity of plants is related to their zone of inhibition against the some of the pathogenic organisms. showed their activity against infectious bacterial species The antimicrobial activity of combined Hydroalcoholic extract of four medicinally important plants were studied in different concentrations (50,100,150,200 µg/6 mm disc) against four pathogenic bacterial strains, two Gram-positive (*Staphylococcus aureus* MTCC 96, *Streptococcus pyogenes* MTCC 442) and two Gram-negative (*Escherichia coli* MTCC 443, *Pseudomonas aeruginosa* MTCC 424), and three fungal strains (*Aspergillus niger* MTCC 282, *Aspergillus clavatus* MTCC 1323, *Candida albicans* MTCC 227). Combined Hydro alcoholic extract shown inhibition against all four selected pathogenic organism. The seed extract showed highest zone of inhibition to a distance of 20.0 \pm 0.5 mm at 200µg concentration.

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Antibacterial activity (Zone of inhibition in mm.)						
Micro organism	Hydro alcoholic extract plants		Concentration in $\mu g/ml$			
	50	100	150	200		
E.coli	12	15	16	20		
P.aeruginosa	12	15	17	20		
S.pyogenes	10	11	13	18		
S.aureus	11	13	15	18		
Values are mean \pm SD of three parallel measurements						

Table-II Antibacterial activity of standard drugs against bacterial test organism

Antibacterial activity

			-					
Drava		Zone of inhibition in mm						
Drug	μg/ml	E.coli	P.aeruginosa	S.pyogenes	S.aureus			
Ampicillin	50	14	14	11	10			
-	100	15	15	14	13			
	150	17	15	16	14			
	200	20	20	19	18			
Chloramphe	enicol 50	14	14	10	12			
	100	17	17	13	14			
	150	28	23	19	19			
	200	28	27	22	22			
Ciprofloxad	in 50	20	20	16	16			
-	100	23	23	19	19			
	150	23	23	19	19			
	200	28	27	22	22			
Norfloxacir	ı 50	22	18	18	19			
	100	25	19	19	22			
	150	27	23	21	26			
	200	29	23	21	28			

The Hydroalcoholic crude extract containing 10, 20, and 50 mg/mL, produced dose-dependent paralysis ranging from loss of motility to loss of response to external stimuli, which eventually progressed to death. As evident from the available literature, Anthelmintic activities of four well known Indian medicinal plants were tested in this

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bioassay at various concentrations of 10, 20 and 50 mg/ml (Table V). The investigational extract acquired the anthelmintic activity at minimal dose of 10 mg/ml. its significant activity (P<0.05) at 10 mg/ml for time taken to paralysis and death when compared to the standard drugs Piperzine citrate used at 10 mg/ml. [6]

Table-III Antifungal activitiy of hydroalcoholic extract against fungal test organism

	Antif	ungal activity		
Microorganism	Hydro alcoholic extract concentration in µg/ml			
-	50	100	150	200
A.niger	13	14	16	20
A.clavatus	11	17	19	20
C.albicans	11	17	19	20

Table-IV Antifungal activity of standard drugs against fungal test organism

Antifungal activity

initungai activity					
Drug Concentration	zone of inhibition in mm				
in µg/ml	A.niger	A.clavatus	C.albicans	Griseofulvin	
	50	19	18	18	
	100	23	21	21	
	150	25	22	22	
	200	28	26	24	
Nystatin	50	18	19	18	
	100	19	21	21	
	150	29	26	26	
	200	29	27	26	

Table – V Anthelmintic activity of Combined Hydroalcoholic seed extract (CHASE) of Coriandrum sativum, Cassia occidentalis, Carica papaya, Momordica foetida.

Groups	Drug Treatment	Concentration (mg/ml)	Pheritima posthuma		Tubifex tubifex	
		(υ),	Paralyzing	Death	Paralyzing	Death
			Time	Time	Time	Time
Ι	Control					
П	Piperazine citrate	10	21 67 - 1 67	56 02 +1 17	22 cc + 1 3	7 52 22 + 1 202
	CHASE		21.07	30.23 1 1 <i>I</i>	22.00 <u>1</u> 1 10	55.55 <u>1</u> 1.202
ш	(CCCM) A	10	28 33 +1.87	62.86 ± 1.37	32 48 +1.8	9 69.67 + 1.77
	CHASE	10	20.33	00100 - 2107	52.10 <u>.</u> 2121	
IV	(CCCM) B					
		20	20.94+1.07	49.53 + 1.09	22.36 +1.5 5	3 44.87 + 1.53
			2017			
V	CHASE	50				
	(CCCM) C1		16.48 ±1.01	33.75 ± 0.66	19.63 ±1.0 9	9 38.36 ± 1.24

CHASE----- Hydro alcoholic extract., (CCCM)---Coriandrum sativum, cassia occidentalis, carica papaya, Momordica foetida Results are expressed as mean ±SEM. The results were analyzed by Analysis of variance (ANOVA). P< 0.05



Figure-1: Anthelmintic activity of Combined Hydroalcoholic Seed Extracts of CCCM Plants

DISCUSSION

Antimicrobial properties of medicinal plants are being increasingly reported from different parts of the world. The World Health Organization estimates that plant extract or their active constituents are used as folk medicine in traditional therapies of 80% of the world's population. In the present work, the Hydro alcoholic extracts obtained from Coriandrum sativum, Cassia occidentalis, Carica papaya, Momrdica foetida shows strong activity against most of the tested bacterial and fungal strains. The results were compared with standard antibiotic drugs. In this screening work, extracts were found to be not inactive against any organism, such as Gram-positive, Gram-negative, and fungal strains were resistant to Hydroalcoholic extract. The assay of biological activity, of combined Hydro alcoholic extracts were used to evaluate anthelmintic activity, has shown dose dependant activity. The Mean ± S.E.M. values were calculated and reported. The result of anthelmintic activity on earthworm *pheretima* posthuma and Tubifex tubifex was given in Table-V reveals that, the different concentrations used has shown paralysis and death of worms and it was compared in the same concentration with Piperazine citrate as reference drug. Piperazine cause hyperpolarization of worms muscle by GABA agonistic action opening Cl⁻ channels that cause relaxation and depresses responsiveness to contractile action of A.Ch. (Acetylcholine). By increasing chloride ion conductance of worm muscle membrane produced hyperpolarization and reduced excitability that led to muscle relaxation and flaccid paralysis. The seed extracts of coriandrum sativum, cassia occidentalis, carica papaya, Momordica foetida not only demonstrated paralysis, but also caused death of worms especially at higher

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concentration of 50 mg/ml, in shorter time. Possible mechanism for anthelmintic effect of seed extract of plants are due to presence of secondary metabolites that they can bind to free proteins in the gastrointestinal tract of host animal or glycoprotein on the cuticle of the parasite and may cause death. In addition, Tannins or their metabolites have a direct effect on the viability of the pre- parasitic stages of helminthes and other phytochemicals may be responsible for an anthelmintic effect include essential oils, flavonoids and terpenoids .This speculation is supported by the varying rates of effectiveness of medicinal plants. The above results show that the activity of hydroalcohol extracts shows significant antibacterial and antifungal activities. This study also shows the presence of different phytochemicals with biological activity that can be of valuable therapeutic index. The result of phytochemicals in the present investigation showed that the plant contains more or less same components like triterpenoids, steroids, glycosides, anthraquinone, flavonoids, proteins, and amino acids. Results show that plant rich in tannin and phenolic compounds have been shown to posses antimicrobial activities against a number of microorganisms.

CONCLUSION

In the current investigation, the hydroalcohol extract in the ratio of 30:70 has been selected after study of such a selected plant with water extracts and methanol extracts, hydroalcohol extract gave higher yield of chemical constituents expected for this research work. The originality of this work is that good results have been found with hydroalcohol ratio, and it will be helpful to carry out other data with MIC and other formulation study, because in comparison of methanol or water extracts, hydroalcohol is more suitable for clinical study. The hydroalcoholic extracts of four plant seeds were found to be active on most of the clinically isolated microorganism and fungi, as compared with standard drugs. The present study justified the claimed uses of seeds in the traditional system of medicine to treat various infectious disease caused by the microbes. However, further studies are needed to better evaluate the potential effectiveness of the crude extracts as the antimicrobial agents. The present results will form the basis for selection of plant species for further investigation in the potential discovery of new natural bioactive compounds.

Biological parameter can be concluded that the plants *Coriandrum sativum*, *Cassia occidentalis*, *Carica papaya*, *Momordica foetida* has significant anthelmintic activity. Further studies using *invivo* model are required to find out and to establish effectiveness and pharmacological rationale for the use of seeds as anthelmintic drug. In the light of above mentioned pharmacological effects, it may be concluded that combined Hydroalcoholic extract has the maximum anthelmintic activity.

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REFERENCES

[1] Ajaiyeoba EO, Onocha PA, Olarenwaju OT. Pharm Biol 2001; 39:217-220.

[2] Al-Said, M.S., Al-Khamis, K.I., Islam, M.W., Parmar, N.S., Tariq, M., Ageel, A.M., **1987**. Journal of Ethnopharmacology 21, 165–173.

- [3] Kudav NA, Kulkarni AB. Ind. J. Chem 1974;12:1042-4.
- [4] Ginde BS, Hosangadi BD, Kudav NA, Nyak KV, Kulkarni AB. I. Chem. Soc. 1970:1285-9.

[5] Aruoma, O.I., Colognato, R., Fontana, I., Gartlon, J., Migliore, L., Koike, K., Coecke, S., Lamy, E., Mersch-Sundermann, V., Laurenza, I., Benzi, L., Yoshino, F., Kobayashi, K., Lee, M.C., **2006**. *Biofactors* 26 (2), 147–159.

- [6] Hakizamungu, E., Van Puyvelde, L., Wery, M., 1992. Journal of Ethnopharmacology 36, 143–146.
- [7] Raman, A., Lau, C., 1996. Phytomedicine 2, 349-362.
- [8] Husain, J., Tickle, I.J., Wood, S.P., **1994**. FEBS Letters 342, 154–158.
- [9] Putnam, C.D., Tainer, J.A., 2000. Natural and Structural Biology 7, 17-18.
- [10] Miura, S., Funatsu, G., 1995. Bioscience Biotechnology and Biochemistry 59, 469–473.
- [11] Khandelwal KR. 2nd ed. Pune: Nirali Prakashan; 2009. Practical Pharmacognosy; pp. 149-156.
- [12] Kokate CK. Delhi: New Gyan Offset Printers; 2000. Practical Pharmacognosy; pp. 107–109.
- [13] Kumar A, Ilavarasan R, Jayachandran, Decaraman M, Aravindhan P. Pak J Nutr. 2009;8:83-85.
- [14] Harborne JB. Chapman and Hall. Newyork: **1973**. Phytochemical methods: A guide to modern techniques of plant analysis; pp. 279–19.
- [15] Maidment C, Dyson A, Haysom I. Nutr. Food Sci 2006; 36 (4): 225 230.

- [16] Farnsworth, N.R., (1966). J. Pharm Sci. 55(3), 225-276.
- [17] Kuhnt et al., 1994 M. Kuhnt, A. Probestle, H. Rimpler, R. Bauer and M. Heinrich, (1994). Planta Medica, 61, 227-232.
- [18] Afolayan AJ, JJM Meyer (1997). J. Ethnopharmacol, 57, 177-181.
- [19] Haslam, E., Lilley, T.H., YaCai, Martin, R., Magnolato, D., (1989). Planta Medica, 55, 18.
- [20] Klaudija Carovic´-Stanko, Sandi Orlic, Olivera Politeo, Frane Strikic, Ivan Kolak, Mladen Milos, Zlatko Satovic. Food Chem 2010; 119, 196 201.
- [21] Rajan S, Baburaj DS, Sethuraman M, Parimala S. Ethnobotany. 2001;6:19-24.
- [22] Morimoto S, Nonaka G, Chen R. Chem Pharmacol Bull. 1988;36:39-47.
- [23] Bauer AW, Kirby WMM, Sherris JC, Turck M. Am J Clin Pathol. 1966;36:493-6.
- [24] Alzoreky NS, Nakahara K. Int J Food Microbiol. 2003;80:223-30.
- [25] Rios JL Recio MC, Villar A. J Ethnopharmacol. 1988;23:127-49.
- [26] Jawetz, E., Melnick, J.L., Adelberg, E.A., Brooks, G.F., Batel, J.S., Ornston, L.N., (1995). Medical Microbiology, vol. 20th ed. Appleton and Lang, New York
- [27] Cerrutti, P., & Alzamora, S. M. (1996). International Journal of Food Microbiology, 29, 379-386.
- [28] National Committee for Clinical Laboratory Standards (NCCLS), (1993). Performance standards for antimicrobial disk susceptibility tests, NCCLS, Pennsylvania, USA, , M2-A5
- [29] Koroch A.R, Juliani R, Zygadlo J.A. 2007. Bioactivity of essential oils and their components.
- [30] Lahlou M 2004. Phytotherapy Rresearch. 18, 435-448.
- [31] Tatsadijeu N L, Jazet Dongmo P M, Nagassoum M B, Etoa F X, Mbofung c M F, 2009. Foodcontrol 20, 161-166.
- [32] Chang C-W, Chang W-L, Chang S-T, Cheng S-S, 2008. Water Research 42. 278-286.
- [33] Pillai LS, Nair BR. Indian J Pharm Sci 2011; 73(1):98-100.
- [34] Das SS, Dey M, Ghosh AK. Indian J. Pharm .Sci 2011; 73(1):104-7.
- [35] KD Chatterjee, Parasitology, Protozoology and Helminthology, 6th ed., In Guha Ray Sree Saraswathy Press Ltd. Calcutta, 1967.
- [36] S Athnasiaduo, I Kyriazakis, F Jackson, RL Coop, Vet parasitol, 2001, 99, 205-219.
- [37] DP Thompson, TG Geary, The structure and function of helminth surfaces, in: J.J. Marr [Ed.], Biochemistry and Molecular Biology of Parasites. 1st ed. Academic Press, New York, pp. 1995, 203-232.
- [38] RJ Martin, Vet J, 1997, 154, 11-34.
- [39] Bundy DA. Trans Royal Soc Trop Med Hyg 1994; 8:259-61
- [40] Idika IK, Okonkwo EA, Onah DN, Ezeh IO, Iheagwam CN, Nwosu CO. Parasitol Res. 2012. [ahead of print]
- [41] Lukhoba C W, Simmonds MSJ, Paton AJ, Ethnopharmacol 2006; 103: 1-24.
- [42] Gbolade AA, Adeyemi AA. Fitoterapia 2008; 79: 223-5.
- [43] Ong HC, Nordiana M. Fitoterapia 1999; 70: 502-13.
- [43] Das SS, Dey M, Ghosh AK. Indian J Pharm Sci 2011; 73(1):104-7.
- [44] Jain ML, Jain SR. Planta Med 1972; 22:66-70.
- [45] Pillai LS, Nair BR. Indian J Pharm Sci 2011; 73(1):98-100.