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Original Research Article

A comparative evaluation of antifungal activity of medicinal plant extracts and chemical fungicides against four plant pathogens

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Minimum Inhibitory Concentration, broad spectrum antifungal activity, *Pongamiapinnata*, *Calotropisprocera*, *Neriumindicum*and *Curcuma longa*

A B S T R A C T

Plants historically have served as models in drug development. The production of medicines and the pharmacological treatment of diseases began with the use of herbs. Leaves extract of *Pongamiapinnata*, *Calotropisprocera*, *Neriumindicum*and *Curcuma longa* were taken in aqueous solution, tested against plant pathogenic organisms - *Aspergillusfumigatus*, *Alternariasolani*, *Helminthosporium spp.* and *Fusariumsolani*. The antifungal activities of these medicinal plants were investigated. Antifungal activities of various plant extracts were compared with commercially available antibiotics. The antimicrobial potential of the above plant extracts were seen against the test organism using agar gel diffusion technique. The minimum inhibitory concentration was determined. It was observed that the latex of the plant had a broad spectrum antifungal activity against all the fungi in each of the tested extracts. These findings supported the traditional uses of the plants in the treatment of different infections.

Introduction

The use of synthetic chemicals as anti-microbial for the management of plant diseases has undoubtly increased crop protection but with considerable deterioration of environmental quality and human health (Cutler and Cutler1999).An attempt was made to employ plant extract against number of plant pathogens. A large number of plants have been reported to possess fungi toxic properties against plant pathogens which could be exploited commercially with practically no residual or toxic effect on ecosystem (Kumar *et al*., 2008).

The herbal products today symbolize safety in contrast to the synthetics that are regarded as unsafe to human and environment. A large number of plants have been reported by various researchers and practitioners of traditional medicine for the treatment of skin diseases (Bohlmann*et al*., 1964).Medicinal plants have nodoubt remained the major sources of traditional medicine worldwide. Accordingly, attention of scientists and researchers have been attracted towards developing new antibiotics that will curtail the increasing drug resistance among microorganisms (Edith *et al.*, 2005). Schimmer *et al*. (1994) reported that plants used for traditional medicine generally contain a number of compounds which may be a potential natural natural antimicrobial combination and which may serve as an alternative, effective, cheap and safe antimicrobial agents for treatment of common microbial infections (Mathur and Goyal, 2011).

Calotropisprocera belongs to the family *Asclepiadaceae* and is a soft wooded, evergreen perennial shrub. It is a xerophytic erect shrub, growing widely throughout the tropical and subtropical regions of Asia and Africa. This plant is popularly known because it produces large quantity of latex. This plant has potential antimicrobial properties against microbial infections (Mathur and Goyal, 2011).

Traditionally, *Calotropis* is used alone or with other medicines to treat common diseases such as fevers, rheumatism, indigestion, cough, cold, eczema, asthma, elephantiasis, nausea, vomiting and diarrhea. The leaves and the flowers *Neriumindium* (kanner*)* are cardio tonic, diaphoretic, and diuretic, emetic expectorant and sternutatory. A decoction of the leaves is applied externally in the treatment of scabies and to reduce swellings. This is a very poisonous plant, containing a powerful cardiac toxin and should only be used with extreme caution. The root is powerfully resolving. Because of its poisonous nature it is only used externally. Oil prepared from the root bark is used in the treatment of leprosy and diseases of scaly nature. The plant is used as a rat poison, aparasitic idée and an insecticide (Kavitha *et al.*, 2000). *Pongamiapinnata* (karanji) exhibits many pharmacological attributes. First, anti-Plasmodial characteristics make *P. pinnata* important to treat malaria caused by *Plasmodium falciparum*. Next, the leaves of the Pongam tree exhibit anti-inflammatory qualities as well as anti-diarrheal activity; also, the leaf extracts are antioxidants. *Pongamia* prevents ulcers by protecting damage from aspirin. It may also provide diabetic patients with a safer anti-hyperglycemic drug.

Turmeric is an ancient spice derived from the rhizomes of *Curcuma longa*, which is a member of the ginger family (*Zingiberaceae*). Also known as 'Golden Spice of India' turmeric has been used in India for medicinal purposes for centuries. Turmeric, derived from the rhizomes of *Curcuma longa*, (family- Zingiberaceae) is a perennial plant having short stem with large oblong leaves, and bears ovate, pyriform or oblong rhizomes, which are often branched and brownish-yellow in colour (Rathaur et al. 2012). *Curcuma longa* (Turmeric) is taken as the blood purifier and is very useful in the common cold, leprosy, intermittent, affections of the liver, dropsy, inflammation and wound healing. The rhizome of the turmeric plant is highly aromatic and antiseptic. It is even used for contraception, swelling, insect stings, wounds, whooping cough, inflammation, internal injuries, pimples, injuries, as a skin tonic. Sweetened milk boiled with the turmeric is the popular remedy for cold and cough. It is given in liver ailments and jaundice. The most important fungi causing post-harvest diseases include: *Helminthosporium spp*, *Aspergillus spp*, *Alternar*ia *spp*, *Fusarium spp*. Many fruits are prone to damage caused by insects, animals, early splits, and during mechanical harvesting. This damage predispose the fruits to the wound invading pathogen *Aspergillus fumigatus*, and other fungi, that causes decay on stored citrus fruits. *Aspergillus fumigatus* can pose a health problem; especially it produces aflatoxin, a group of toxic, carcinogenic compounds (Diener*et al*., 1987). *Alternaria solani* is a fungal pathogen that produces a disease in tomato and potato plants called early blight. *Alternaria solani* infects stems, leaves and fruits of tomato (*Solanumly copersicum* L.), potato (*S. tuberosum*), eggplant (*S. melongena* L.), bell pepper and hot pepper (*Capsicum* spp.), and other members of the *Solanum* family. Distinguishing symptoms of *A. solani* include leaf spot and defoliation, which are most pronounced in the lower canopy. In some cases, *A. solani* may also cause damping off. *A. fumigatus*is the second most common agent of aspergillosis, the first being *Aspergillus fumigatus*. *A. fumigatus* may invade arteries of the lung or brain and cause infarction. Neutropenia predisposes to *Aspergillus* infection. *Aspergillus fumigatus* also produces a toxin, aflatoxin, which is one of the aetiological agents for hepatocellular carcinoma. In humans whose immune systems are weakened in a particular way, (neutropenia, *i.e.*, very low neutrophils count), aggressive fusarial infections penetrating the entire body and bloodstream (disseminated infections) may be caused by members of the *Fusarium solani* complex (Bruce and Miller, 2003). *Helminthosporium* attack essentially all turf grasses as well as numerous pasture, wild and weed grasses, and small grains. This work reports the antimicrobial effect of *Curcuma longa, Pongamia pinnata, Neriumi ndicum, Calotropisprocera* and their various combinations on some bacterial species. This is inpursuance of the efforts to search for drugs from plants and the verification of the scientific basis of some known practices in traditional medicine. A few reports have focused on the immense potential of this plant which has antimicrobial, wound healing, anti-inflammatory and immuno-modulatory properties.

Potato (*Solanum tuberosum* L.) is the worlds fourth kept in dark between the filter papers at room temperature important food crop after wheat, rice and maize because of till completely dry. Each plant sample was individually its great yield potential and high nutritive value for grounded into powder for preparation of extract. The extract was allowed to internationally important disease of potato resulting in stand for some time and decanted off into the flask and about 25 to 60% loss in yield in different countries and final volume was raised to 100ml by adding boiled distilled attempts have been made to manage the disease by water. The supernatant was used for assay. The treating with chemical compounds, biological agents as antifungal activity of each plant part extract was reported by Wharton and Kirk. Therefore, the investigation was preceded with the objective of antifungal activity determination of the selected medicinal value plants against plant pathogens and compare with the chemical fungicides.

Materials and Methods

Place of sample collection

The plant sample of *Calotropisprocera, Pongamiapinnata*, *Curcuma longa* and *Neriumindicum* was collected from Sam Higginbottom Institute of Agriculture and Sciences, Allahabad.

Procurement of Microorganism

The plant pathogens*Altrenariasolani, Fusariumsolani*, *Aspergillusfumigatus and Helminthosporium spp.* and antifungal disc of Bavistin and Mancozebwas procured from the Microbial Culture Collection (MCCB) , Department of Microbiology and Fermentation Technology.Sam Higginbottom Institute of Agriculture and Science, Allahabad.

Organism	Strain no.
Aspergillusfumigatus	0207
Alternariasolani	0079
Fusariumsolani	0068
Helminthosporium spp.	0243

Extraction of Plant Material

Fresh flower of *Nerium indicum* and leaves of *Calotropisprocera*, *Pongamia, Pinnata,Curcuma longa*wre collected, washed under tap water, then in distilled water, and then kept between folds offilter paper to remove excess of water from external surface. On drying 10g of fresh leaves of *Curcuma longa,Pongamia pinnata, Calotropisprocera* and *Neriumindicum,* was chopped well, and 100 ml of distilled water was added separately (1:1w/v) respectively. These were ground to fine powder and paste. respectively, soaked for 24 h. Soaked materials was then filtered through muslin clothes and leaf extract of *Curcuma longa, Pongamiapinneta,Calotropisprocera* and flowerof *Neriumindicum,* was collected with the help of funnel in separate conical flask. Finally filtrate obtained was used against plant pathogens.

Antifungal activity of Medicinal plant extract and Chemical fungicides against plant pathogens

Sabouraud dextrose agar was taken in culture tubes and broth culture was mixed in tubes by vortex mixture and poured into the plates. The plates were allowed to solidify for an hour. Wells (10 mm diameter) was made with the aid of flamed cork borer on the surface of the agar plates. 0.1ml of the leaf extracts of the plants and chemical fungicides suspension (Bavistin and Mancozeb) filled in the wells and the plates were incubated at 25°C for 5 days to observed zone of inhibition.

Determination of Minimum Inhibitory Concentration

To measure the Minimum Inhibitory Concentration values, micro-broth dilution method was used. The reconstituted extract was serially diluted 2-fold in SDA broth medium to obtain various concentrations (200,100, 50, 25, 12.5, 6.25, 3.125, and 1.5625 µl/ml) of the stocksand were assayed against the reconstituted organisms. The minimum inhibitory concentration is defined as the lowest concentration of extract which inhibited the growth of microorganism (Prescott *et al*., 1999).

Results and Discussion

Antifungal activity of *Curcuma longa*

The antifungal activity of aqueous extract of *Curcuma longa* was determined against *Aspergillus fumigatus, Alternariasolani, Fusarium solani* and *Helminthosporium spp.* at concentration of 100µl and zone of inhibition was found as a clear function of concentration. The antifungal activity of *Curcumalonga* was determined on subouraud dextrose agar medium. The aqueous extract of *Curcuma longa* showed better activity against *Aspergillus fumigates, Fusarium solani. Alternariasolani*, *Helminthosporium spp.* Similar study conducted by Fairs, *et al.* (2002) had reported zone of inhibition of aqueous extract of *Curcuma longa against Aspergillus spp.* with arecorded inhibition zones of 58.2%, 73.4%, 79.3%, and 90.1% at similar plant extract concentrations respectively. The inhibition zones observed in *Fusariumspp*. were 46.5%, 64.7%, 71.7% and 88.9% respectively. Rathaur*et al*. (2012) reported anti-inflammatory, antioxidant, anti-carcinogenic, anti-mutagenic, anticoagulant, antifertility, anti-diabetic, antibacterial, antifungal, antiprotozoal, antiviral, anti-fibrotic, anti-venom, antiulcer, hypotensive and hypocholesteremic activities of *Curcuma longa*. Kumar *et al*. (2008) concluded that*Curcuma longa* gives zone of inhibition of 25 mm to 30 mm against *Aspergillusfumigatus* and *Aspergillusflavus.*

Antifungal activity of *Neriumindicum*

The antifungal activity of *Neriumindicum* was determined by agar well diffusion at the concentration of 100 µl against the organisms (*Aspergillus fumigatus, Alternaria solani, Fusarium solani* and *helminthosporium spp. Neriumindicum* showed high activity against *Aspergillus fumigatus* (20mm), *Alternaria solani*(22mm) but did not show activity against *Fusarium solani* and *Helminthosporium spp.* Chauhan *et al*. (2013) concluded the inhibitory effect of *Neriumindicum* (19 mm) against some bacterial spp. Parastoo *et al*. (2012) concluded that the methanol extract of *Neriumindicum* showed inhibition of *Bacillus sp., Escherichia coli, Yersinia sp., Staphylococcus sp.* at 500ppm and 1000ppm, whereas no inhibitory effect was observed against *Pseudomonas sp., Lactobacillus sp., Enterococcus sp.,* and *Klebsiella sp.*

Antifungal activity of *Pongamiapinnata*

The antifungal activity of aqueous extract of *Pongamiapinnata* determined at concentration of 100 µl against *Aspergillus fumigatus, Alternaria solani, Fusarium solani* and *Helminthosporium spp. Pongamiapinnata* showed better activity against *Aspergillus fumigatus* (20mm) and *Alternaria solani* (21mm),but did not showed activity against *Helminthosporium spp.*and *Fusarium solani.* Sharma *et al*. (2012)reported that the *Pongamiapinnata* leaf extracts (Chloroform) produced zone of 22, 18, 16, 16, and 15 mm against *T. mentagrophytes, M. fulvum, T.rubrum, T. tonsurans* and *M. gypseum*, and methanol extracts produces maximum zones of 17, 16, 12, 12 and 11mm for *M. fulvum, T. mentagrophytes,*

Antifungal activity of *Calotropisprocera*

The antifungal activity of aqueous extract of *Calotropisprocera* was determined against *Aspergillus fumigatus, Alternaria solani, Fusarium solani* and *Helminthosporium spp.* at concentration of 100 µl and zone of inhibition was found to be clear function of concentration. The antifungal activity of *Calotropisprocera* was determined on subouraud dextrose agar medium. The aqueous extract of *Calotropisprocera* showed better activity against *Aspergillus fumigatus (*21mm) and *Alternaria solani* (30mm), but not showed activity against *Fusarium solani* and *Helminthosporium spp.* Saadabi*et al*. (2012) reported that *Calotropisprocera* showed inhibition of 25mm against *Aspergillus fumigatus*. Velmurugan*et al.* (2013) concluded that *Calotropisprocera* showed inhibitory activity of 15.1 mm against fungi *Fusarium spp*. Bharthi *et al.* (2011) examined the highest zone of inhibition (21mm) by *Calotropisprocera* against fungal pathogens was noted at the concentration of 100 µl (50mg/1ml). Analysis of variance revealed that the calculated value of f-test due to organism was greater then the tablulated value of F on 3,9 degree of freedom and at 5 % probability level, therefore it can be conclude from the data that there was significant effect of different organism on zone of inhibition. The average value due to *Curcuma longa* was highest followed by the average value of *Calotropisprocera* and *Neriumindicum*. Minimum value was recorded due to *Pongamiapinnata*. But the calculated value of F due to extract was smaller than its table value at 5% level of significance; hence there was no significant difference.

Antifungal activity of chemical fungicides against plant pathogens

The antifungal susceptibility test was determined by suspension of Bavstin and Mancozeb against (*Fusarium solani, Aspergillus fumigatus, Alternaria solani* and *Helminthosporium spp.*) both showed high activity against *Fusarium solani, Aspergillus fumigatus, Alternaria solani*, and *Helminthosporium spp.* Shirurkar and Wahegaonkar (2012) studied the antifungal activity of the eight different plant derived oils and two known antifungal synthetic chemicals available in market tested against the eight fungal species *Aspergillus flavus, A. niger, A.terreus, A. oryzae, A. fumigatus, Fusarium moniliforme., F. solani, Penicillium sp.* isolated from maize grains. The results revealed that Bavistin, Mancozeb and some of the oils tested exhibited different degrees of antifungal activity against different seed borne fungi of maize. Kiran *et al.* (2011) studied the antifungal activity of aqueous and solvent extract of seeds of *P.corylifolia*against five seed borne fungi of maize *viz*., *Curvularialunata, Dreschslerahalodes, Alternaria alternata, Cladosporium cladosporioide s*and *Rhizopus*sp. were tested *in vitro*. In aqueous extract, maximum inhibition was observed in *A.alternata*and recorded 95.4% inhibition at 50% concentration followed by *C.lunata*(86.0%), *Rhizopus*sp.(82.3%), *D. halodes*(68.0%) and *C.cladosporioides* (57.7%). Significant activity was also observed in 10, 20, 30 and 40% concentration. In solvent extracts tested at 250, 500, 750 and 1000 μl concentration, maximum inhibition was observed in petroleum ether extract and moderate activity was observed in methanol extract. Compared to synthetic fungicide Bavistin and Thiram, complete inhibition was observed against all the test fungi tested at 2% recommended concentration.

Comparative analysis of antifungal activity of plant extract/chemical fungicides against plant pathogens

After comparing the antifungal activity of plant extract with chemical fungicides against plant pathogens showed that the aqueous extract of plant leaves were more susceptible then the chemical fungicides. In plant extract the leaves of *Curcuma longa* were more active against pathogenic fungi.

Determination of Minimum Inhibitory Concentration

Further using themacro-dilution broth method, the aqueous extract of the leaves of *Curcuma longa, Neriumindicum, Pongamiapinnata* and *Calotropisprocea* were subjected to MIC determination. The extracts were serially diluted to obtain a decreasing concentration of 200 to 1.56 μl of the SDA broth.

The aqueous extract of *Curcuma longa* exhibited inhibition at the concentration of

6.25 µl/ml against *Aspergillus fumigatus*, at a concentration of 12.5 µl/ml against *helminthosporium spp*.The final MIC value was found to be two-fold higher than the initial MIC value.

The extract of *Pongamiapinnata* exhibited at the concentration of 6.25 µl against *Alternariasolani* and *Aspergillus fumigatus*and even the lowest concentration the MIC was not found against *Fusarium solani* and *Helminthosporium spp.* The final MIC value was found to be two-fold higher than the initial MIC value.

The extract of *Neriumindicum* exhibited against *Alternariasolani* at the concentration of 12.5 µl, and for *Aspergillus fumigatus*it exhibited at the concentration of 6.25 µl. but MIC was not found against *Helminthosporium spp.* and *Fusarium solani* even at the lowest concentration. The final MIC value was found to be two-fold higher than the initial MIC value.

The extract of the *Calotropisprocera* exhibited against *Alternariasolani* at the concentration of 12.5µl, *for Aspergillus fumigatus* it exhibited at the concentration of 3.125µl.but no MIC was found even at the lowest concentration *Helminthosporium spp.* and *Fusarium solani*. The final MIC value was found to be two-fold higher than the initial MIC value. Similer study was shown by Saadabi*et al.* (2012), who determined the minimum inhibitory concentration value of the different extracts of *Calotropisprocera* showed that the highest activity was recorded against *Aspergillus fumigatus*, *Candida albicans* and *Bacillus subtilis* (10mg ml^{-1}) in chloroform extract of the latex. The lowest activity obtain against *Candida albicans* (25mg ml^{-1}) in methanol and in aqueous (25mg ml^{-1}) against *Escherichia coli*. It was clearly noticed that the latex of the plant have a broad spectrum antimicrobial activity against all of the bacteria and fungi in all of the tested Extract. Lalsangluaii*et al*. (2013) determined the minimum inhibitory concentration (MIC) of the oil of *Curcuma longa* was recorded at 1.9 mg/ml against *T. mentagrophytes* and 2.1 mg/ml against *T. rubrum*. However, it was fungicidal at 2.4 mg/ml against *T.mentagrophytes* and 2.5 mg/ml against *T. rubrum* respectively. The effective concentration contains heavy doses of inoculums (25 discs of 5 mm each).

When modern drugs of choice become ineffective in the treatment of various diseases, it is anticipated that this study would lead to the establishment of some compounds that could be used to formulate new and more potent antimicrobial drugs of natural origin for the treatment. It will also help in alleviating the health problems of the people especially living in far flung areas having no access to modern health care systems.

Further this study indicates that the leaf extracts using solution in water were more effective supporting the use of “teas” to treat intestinal disease caused by worm parasites. *Curcuma longa*.is primarily an ornamental plant, although it has been used in traditional medicine to cure some

Table.4.1 Antifungal activity of plant extracts against plant pathogenic fungi

Extract	**Zone of inhibition (mm) of organism**			
	Aspergillusfumigatus	***Alternariasolani***	***Fusariumsolani***	***Helminthosporium spp.***
Curcuma longa	22	20	23	25
Neriumindicum	20	22	0	0
Pongamiapinnata	20	21	0	0
Calotropisprocera	21	30	0	0

Due to Organism $f_{(cal)}$ =6.611>f(tab)=3.86 at 5%,Significant C.D.=11.56
Due to Extract f (cal)=2.932<f(tab)=3.86,Non significant

Fig.4.2 Antifungal activities of chemical fungicides against plant pathogens

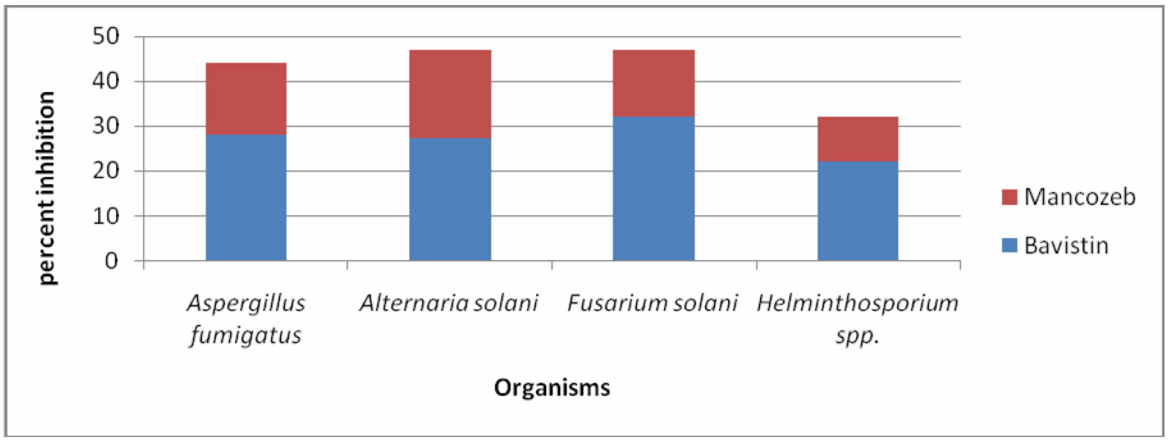
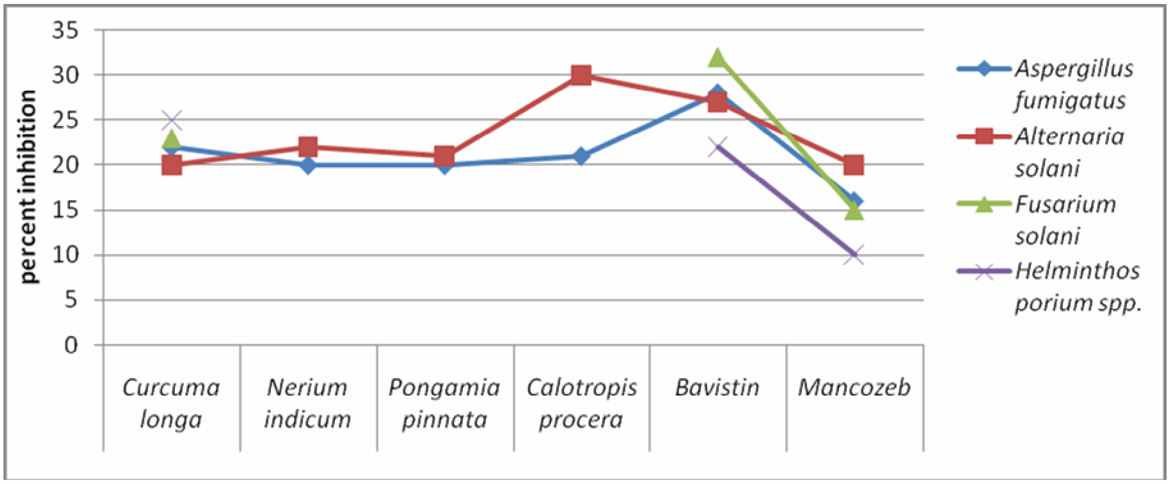


Table.4.3 Comparative analysis of antifungal activity of plant extract/chemical fungicides against plant pathogens

Organism	**Antifungal activity of plant extract/chemical fungicides against plant pathogens (Zone of inhibition in mm)**					
	Curcuma longa	*Neriumin dicum*	*Pongamia pinnata*	*Calotropisp rocera*	Bavistin	Mancozeb
Aspergillusf umigatus	22	20	20	21	28	16
Alternaria solani	20	22	21	30	27	20
Fusarium solani	23	0	0	0	32	15
Helmintho-sporium spp.	25	0	0	0	22	10

Fig.4.3 Comparative analysis of antifungal activity of plant extract/chemical fungicides against plant pathogens



diseases. Recent reports of various pharmaceutical properties of this plant established it as a valuable source of medicinal compounds. In this study, aqueous extract of *Neriumindicum*.leaves possessed broad spectrum antimicrobial activity against various clinical isolates of fungi. It is concluded that aqueous extract of *Calotropisprocera*leaves is a good source of antimicrobial compounds and in future, the bioactive molecule can be isolated and characterized using advanced analytical techniques.

References

Ahmad, I. and Beg, A. Z. 2001.Antimicrobial and phytochemical studies on 45 Indian medicinal plants against multi drug resistant human pathogens. *Journal of Ethnopharmacology*.74:113–123.

Akanksha, Yadav, P. P., Srivastava, K. and Maurya R. 2011Synthesis of analogues of antihyperglycemic lead karanjin. *Medicinal Chemistry Research*, 209, 1465-1472,

Alavijeh, P. K., Alavijeh, P. K. and activity cycloart-23-ene-3, 25 diol B2 isolated from *Pongamiapinnata* L.

Sharma, D. 2012.Study of antimicrobial activity of few medicinal herbs.*Asian Journal of Plant Sciences Research*.2 4:496-502.

Alharbi, A.S., Arunachalam C., Murugan, M.A. and Wainwright, M. 2012.Antibacterial activity of *actinomycetes* isolated from terrestrial soil of Saudi Arabia. *Journal of Food, Agriculture & Environment Vol.10 2: 1093-1097. 2012*

Apak, R., Guclu, K., Ozyurek, M., and Kardemir, S. E. 2004.Novel Total Antioxidant Capacity Index for Detoxy Polyphenols and Vitamins C and E, using their Cupric Ion Reducing Capacity in the Presence of Neocuproine:CUPRAC method. *J. Agriculture Food Chemistry*. vol. 5 2: 7970-798

Arote, S. R. and Yeole, P. G. 2010.*Pongamiapinnata* : A Comprehensive Review. *International Journal of Pharmtech Research*.:2283-2290.

Badole, S. L., Zanwar, A. A., Khopade, A. N., and Bodhankar. S. L. 2011. *In vitro* antioxidant and antimicrobial

Pierre. *Asian Pacific Journal of Tropical Medicine*: 910-916.

Bharathi, B., Sundari, K. and Daniel, G. S. 2011. Effect of some medicinal plants on opportunistic bacterial and fungal pathogens associated with HIV. *Asian Journal of Pharmaceutical and Clinical Research*:149-154.

Bhardwaj, S. K. 2012. Evaluation of plant extracts as antifungal agents against *Fusariumsolani*Mart. Sacc.*World Journal of Agricultural Sciences* 8 4: 385-388.

Bhardwaj, S. K., 2012. Evaluation of plant extracts as antifungal agents against *Fusariumsolani* Mart. Sacc. *World Journal of Agricultural Sciences.* 8 4: 385-388.

Bohlamann, F., KIene. K. M., and Arndt, C. 1964.Naturally occurring thiophen-acetylene compounds. *Chemische Berichte:*.97-212.

Bruce, B. J., and Miller J. D. 2003.Mycotoxins as harmful indoor air contaminants.*Applied Microbiology and Biotechnolgy*.367-372.

Chandrashekar. K. S. and Prasanna, K. S., 2010. Antimicrobial activity of *Pongamiapinnata* leaves. *International Journal of Medicobiological Research*. 11: 18-20.

Chauhan, S., Singh, S., Thakur, A. and Dogra, M. S. 2013.Antibacterial activity of *Neriumindicum*gainst some Gram positive bacterial species.*International Journal of Drug Research and Technology*.3 1: 8-11.

Cikrikci, S., Mozioglu, E., Yılmaz, H. 2008.Biological activity of curcuminoids isolated from *Curcuma longa.Records of Natural Products.* 12: 19-24.

Cutler, H. G. and Cutler, S. J. 1999.Biological active natural product: *Agrochemical CRS Press, Boca Raton: USA:*299.

Damalas, C .A. 2011. Potential uses of turmeric C*urcuma longa* products as alternative means of pest management in crop production. *Plant Omics Journal.* 43:136-141.

Diener, U. L., Cole, R. J., Sanders, T. H., Payne, G. A., Lee, L. S., and Klich, M. A. 1987.Epidemiolgy of Aflatoxin formation by *Aspergillusflavus*. *Review Phytopathlogy*.25:249-270.

Edith, A., Mofolusho, F., Omonike, O., Larry, O., Dora, A. 2005. In vivo antimalarial and cytotoxic properties of Annonasenegalenisis extract. *Afr. J.Trad. Compl. Alt. Med.* 31:138-139.

Fairs, G., Singh, H., and Syarhabi, M. 2002.Bioefficacy of some plant derivatives against the *Aspergillus niger, Penicillium digitatum,* and *Fusarium sp in vitro*. *International Journal of Scientific and Research Publication*, Volume 2, Issue 3,497-502.

Goyal, M. and Mathur, R. 2011. Antimicrobial potential and phytochemical analysis of plant extracts of *Calotropisprocera. International Journal of Drug Discovery and Herbal Research.*13: 138-143.

Hegde, M. N., Shetty, S., Yelapure, M., and Patil, A. 2012. Evaluation of Antimicrobial activity of aqueous and hydro-alcoholic *Curcuma longa* extracts against endodontic pathogens.*Journal of Pharmacy.* 192-198.

Hussain, T., Arshad, M., Khan, S., Sattar, H. and Qureshi, M. S. 2011.*In vitro* screening of methanol plant extracts for their antibacterial activity. *Pakistan Journal of Botany*.431: 531-538.

Jabeen K. 2013.Antifungal Activity of *AzadirachtaIndica* against *AlternariaSolani*.*Journal of Life Sciences and Technologies. Vol. 1, No. 1.89;93*

Kareem. S. O., Akpan. I. and Ojo, O. P.

2008. Antimicrobial activities of *Calotropisprocera* on selected pathogenic microorganisms. *African Journal of Biomedical Research*.11:105 – 110.

Kavitha, Hilds, A. and Ramesh, V. M. 2000.Fungicidal activity of plant extract against the growth of health risk causing fungi.*Geobis*.27:81-84.

Khan, R., Pandey, M., Dan, S. Trivedi, N. and Chaudhary,B. 2012. Anti-arthritic activity of *Calotropisgigantean* root bark extract in experimental rats. I*nternational Journal of Pharmaceuticals and Chemical Sciences*. 11:417-434.

Kiran, B. Lalitha, V. and Raveesha, K. A., 2011.Antifungal activity of aqueous and solvent extracts of seeds of *Psoraleacorylifolia* L. against seed borne fungi of maize *International Journal of Pharmacy & Life Sciences.*Vol. 2, Issue 10:, 1133-1136.

Kopacki ,M. and Wagner, A. 2006.Effect of some fungicides on mycelium growth of *Fusariumavenaceum*sacc.Pathogenic to *Chrysanthemum.Agronomy Research* 49special issue, 237-240.

Kumar, A., Shukla, R., Singh, P., Prasad, C. S., and Dubey, N. K. 2008. Assessment of *Thymus vulgaris L.* essential oil as a safe botanical preservative against post harvest fungal infestation of food commodities. *Food Science.Emerg.*,4: 575-580.

Lalsangluaii, F., Kumar, A., Shukla, C. A. and Dikshit, A. 2013. Tradition to technology: an approach to drug development against human pathogenic fungi.*Science Vision*. Vol 13 Issue No 2;49-57.

Martins, C. V. B., daSilva, D. L., Neres, A. T. M., Magalha, T. F. F., Watanabe, G. A., Modolo, L. V., Sabino, A. A., de Fatima, A. and de Resende, M. A. 2009.*Curcumin* as a promising antifungal of clinical interest.*Journal of Antimicrobial Chemotherapy*. 63:337–339.

Masih, H., Singh, A. K., Kumar, Y., Srivastava, A., Singh, R. K., Mishra, S. K., Shivam, K., 2011. Isolation and optimization of metabolite production from mutant strain of *Bacillus sp*. with antibiotic activity against plant pathogenic agents. *Journal of Pharmaceutical and Biomedical Sciences.*1110:1-4.

Mathur, R. and Goyal, M. 2011.Antimicrobial Potential and Phytochemical Analysis of Plant Extracts of *CalotropisProcera.International Journal of Drug Discovery and Herbal Research* IJDDHR.13: 138-143.

Namita, P. and Mukesh, R. 2012.Medicinal plants used as antimicrobial agents.*International Research Journal of Pharmacy*.31:31-40

Nashwa and Kamal, A. M Abo-Elyousr., 2012.Evaluation of Various Plant Extracts against the Early Blight Disease of Tomato Plants under Greenhouse and Field Conditions. *Plant Protect. Sci.* 482: 74–79

Parastoo. K. A., Parisa K. A. and Sharma. D. 2012.A study of antimicrobial activity of few medicinal herbs. *Asian Journal of Plant Science and Research*, 2 4:496-502

Patel, J. D., Shrivastava, A. K. and Kumar, V. 2009. Evaluation of some medicinal plants used in traditional wound healing preparations for antibacterial property against some pathogenic bacteria. *Journal of Clinical Immunology and Immunopathology Research.*11 : 7-12.

Pathak, K., supekar, S. Pardeshi, K. and Karen, K. 2011. Study of *in vitro* Antibacterial activity of some medicinal plants against the bacterial strain – *Alcaligens*.*International Journal of Basic and Applied Research* Vol. 01, 88-92.

Prescott, M. L., Harley, J., Donald, P. and Klein, A., 1999. In: Antimicrobial chemotherapy.' Microbiology 2nd edition published by C. Brown Publishers, U.S.A:325.

Pundir, R. K. and Jain, P. 2010. Comparative studies on the antimicrobial activity of black Pepper *Piper nigrum* and Turmeric *Curcuma longa* extracts. *International Journal of Applied Biology and Pharmaceutical Technology*.493-501.

Ramya, V., Dhayalan, V. D. and Umamaheswari, S., 2010.*In vitro* studies on antibacterial activity and separation of active compounds of selected flower extracts by HPTLC. *Journal of Chemical and Pharmaceutical Research.* 26:86-91.

Ranjit, P. M., Krishna, P. M., Silpa, P., Nagalakshmi, V., Anjali, M., Girish, K., and Chowdary, Y. A. 2012.*In vitro* cytotoxic activities of *Calotropisprocera* latex and flower extracts against Mcf-7 and Hela cell line cultures.*International Journal of Pharmacy and Pharmaceutical Sciences*.41:66-70.

Rathaur, P., Raja, W., Ramteke, P. W. and John, S. A. 2012. Turmeric: The golden spice of life. *International Journal of Pharmaceuticals Sciences and Research.* 37:1987-1994.

Saadabi, A. M. A., Ali, N. M., Mohammed, H.I., Alsafi, F. N. and Mustafa, H. B. 2012.An *in vitro* antimicrobial activity of *Calotropisprocera*Ait.extracts on certain groups of pathogenic microorganisms.*Research Journal of Medicial Sciences*.61:13-17.

Sailaja, D., Srilakshmi, P., Bhanuteja, M. and Kumar, D. R., 2012. Physicochemical studies in *Pongamiapinnata* galls infected with fungus. *International Journal of Applied Biology and Pharmaceutical Technology*.3 2:205-207

Sangwan, S., Rao, D. V. and Sharma, R. A. 2010.A review on *Pongamiapinnata* L. Pierre: A great versatile leguminous plant. Nature and Science.811 130-139.

Sati, S. C. and Joshi, S., 2011.Aspects of antifungal potential ofethnobotanically know medicinal plants. *Research Journal of Medicinal Plant*.54:377-391.

Schimmer O., Kruger A., Paulin H., Haefele F. 1994. An Evaluation of 55 Commercially pants extract in the Ames Mutagenicity Test. *Pharmazie* 49: 448-451.

Senthil, K. Dr., Sivamani, P., Baskaran, C. and Dr. Md, J. 2012. Evaluation of Anti Microbial Activity and Phytochemical analysis of Organic Solvent extracts of *Calotropisgigantea* .*IOSR Journal of Pharmacy*. Vol. 2, Issue 3, 389-394.

Sharma, B., Fatima, A. and Agarwal, P., 2013.Phytochemistry and pharmacological potential of *Neriumindicum* Mill.*InternationalePharmaceuticaScie ncia*.31:39-45.

Sharma, K. K., Kotoky, J., Kalita, J. C. and Barthakur, R. 2012. Evaluation of antidermatophytic activity of *Ranunculus sceleratus*and *Pongamiapinnata* available in North Eastern Region of India. *Asian Pacific Journal of Tropical Biomedicine.*:808-811.

Shirurkar, D. and Wahegaonkar, K. N .,

2012. Antifungal activity of selected plant derived oils and some fungicides against seed borne fungi of maize.*European Journal of Experimental Biology*, 2 5:1693-1696.

Shivaji, Sharma, R., Pawar, P., Kashikar, V. and Sasanoor, M. 2012.Review on Anti-microbial activities of poisonous drugsdescribed in Ayurveda.*International Journal of Pharmaceutical Frontier Research*. April-June 2012; 22: 80-89.

Singh, S. P., Tanwer, B. S. and Khan, M.2010. Antifungal potential of ashwagandha against some pathogenic fungi.*International Journal of Biopharmaceutics.*12: 72-74.

Srilakshmi, P., Sailaja, D., Bhanuteja, M., Kumar, D. R. and Madhuri, M., 2012 Quantitative estimation of carbohydrates in insect induced and fungal infected leaf galls of *Pongamiapinnata*. *International Journal of Plant, Animal and Environmental Sciences* .203-205.

Velmurugan, S., Viji, V. T., Babu, M. M., Punitha, M. J. and Citarasu, T. 2013 Antimicrobial effect of *Calotropisprocera* active principles against aquatic microbial pathogens isolated from shrimp and fishes. *Asian Pacific Journal of Tropical Biomedicine.*812-817.

Wagh, P., Rai, P., Deshmukh, S. K. and Durate, M. C. T. 2007.Bio-activity of oils of *Trigonellafoenum-graecum* and *Pongamiapinnata.African Journal of Biotechnology.*6 13:1592-1596.

Wharton, P. and W. Kirk, 2007.*Fusarium* dry rot leaves. *J. Med. and Aroma. Plant Sci*., 25: 397-399. http://www.potatodiseases.org/dryrot.html.

Yadav, A. P. P., Srivastava, A. K. and Maurya, R. 2011.Synthesis of analogues of antihyperglycemic lead karanjin.*Medicinal Chemistry Research*. 209:1465-1472.

Yulia 2005.Antifungal activity of plant extracts and oils against fungal pathogens of pepper *Pippernigrum*, cinnamon *CinnamomumZeylanicumBlume* and turmeric *Curcuma domesticaval*..*Boreal Environment Research* 10;493-510.

Zahin, M., Aqil, F., Khan, M. S. A. and Ahmad, I. 2010.Ethnomedicinal plants derivedantibacterials and their prospects. *Asian Pacific Journal of Tropical Biomedicine.*149-178.

Zhang, H. and Zheng, X. 2005. Biological controlof post harvest blue mold of oranges by *Cryptococcus laurentii*Kufferath skinner. *Biocontrol.*50: 331-342.

Zuzarte, M., Gonçalves, M. J., Canhoto, J. and Salgueiro, L. 2011.Antidermatophytic activity of essential oils. *International Journal of Pharmtech Research:* 1167- 1178.