



## Antimicrobial Activity of *Citrullus colocynthis* Extracts Against Soil Bacteria

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### Abstract

The study aim to evaluate the ability of *Citrullus colocynthis*, to reducing soil microbial load. To prepared extracts using water, ethanol and chloroform as solvent. The extracts concentration 0.001, 0.001 and 0.1mg/l and were subjected to preliminary phytochemical screening and tested against the gram negative and positive rhizosphere bacteria by disc diffusion. Among these extracts the chloroform was found to be most effective in all three strains. Maximum inhibition zone was 15.6mm, and minimum inhibition zone was 9.1mm when ethanol extract was followed by the activities of extracts of three strains. Maximum inhibition zone was 14 mm and minimum inhibition zone was 8.8 mm. The water aqueous extract was less activity against tested organisms.

**Keywords:** reduce, soil ,bacteria , , *Citrullus colocynthis* , rhizosphere bacteria

### Introduction

*Citrullus colocynthis* is a desert plant that grows in sandy arid soils (Gurudeeban, S *et al* 2010). It is native to the Mediterranean Basin and Asia and is distributed among the west coast of northern Africa, eastward through the Sahara, Egypt until India and reaches also the north coast of the Mediterranean and the Caspian seas (Borhade P,*et al*, 2013). It has a great survival rate under extreme xeric conditions. In fact, it can tolerate annual precipitation of 250 to 1500 mm and an annual temperature of 14.8 to 27.8 °C, pH range between 5.0 and 7.8. (John uri lloyd, and cincinnati,O.,1989). Plants are continuously in contact with different microorganisms, including viruses, bacteria and fungi. The relationships established with some of them are beneficial for the plants; thus, some bacteria known as rhizobia, form symbiotic association with leguminous plants by fixing atmospheric nitrogen in root nodules. Other bacteria found close to the plant root (rhizobacteria) are able to control plant diseases caused by soil pathogens (Bais ,*et al* 2006) .Both the plants and the bacteria benefit from the process of nitrogen fixation; the plant obtains the nitrogen it needs to synthesize proteins, while the bacteria obtain carbon from the plant and a secure environment to inhabit within the plant roots (lonela,*et al* ,2007).Soil bacteria are very important in biogeochemicalcycles and have been used for crop production for decades. Plant – bacterial interactions in the rhizosphere are the deter-minants of plant health and soil fertility (Rifat H,*et al* 2010).

### Material and Method

#### Extract preparation

*Citrullus colocynthis* was collected form hail area KSA. The plant extracts were prepared using the solvents water, ethanol and chloroform. 10g of the samples were taken and homogenized with 100ml of the respective solvents. The crude preparation was left overnight in the shaker at room temperature and then centrifuged at 4000rpm for 20mins. The supernatant containing the plant extract was transferred to a beaker and the extract concentrated by evaporating the solvent at 60°C. The crude extract weighed and dissolved in a known volume of distal water to obtain a final concentration of 0.001, 0.001, and 0.1 mg / ml

#### Test microorganisms

Three rhizospheric isolates (P-1,P-2 and G-12 and) were obtained from the culture collection from Department of biology, College of Science, University of Hail.

#### Antibacterial Assay

The effect of plant extracts on the several bacterial strains were assayed by gar well diffusion method and further confirmed by Disc diffusion method. The minimum concentrations of the plant extracts to inhibit the microorganisms were also determined by microdilution method using plant fractions serially diluted in sterile nutrient broth

#### Procedure

Petriplates containing 20ml Muller Hinton medium were seeded with 24hr culture of bacterial strains. Wells were cut and 15  $\mu$ l of the plant extracts (namely aqueous,water, ethanol and chloroform extracts) were added. The plates were then incubated at 37°C for 24 hours. The antibacterial activity was assayed by measuring the diameter of the inhibition zone formed around the (Gurusiddaiah *et al*. 1979). water, ethanol and chloroform disc were used as a positive control.

### Result

The data obtained showed that the water extract was give zero inhibition zone in strain P1, strain P2 and strain G12 .When ethanol extract was gives 0,0 and 8.8 in strain P1 , 12.8, 13.5 and 14 inhibition zone in strain P2 and 0 ,0 and 9.5

inhibition zone in strain G12 and chloroform extract was give 0,10.25 and 12.8 in strain P1,10.8,13 and 15.6 in P2 and 0,9.1 and 13.95 inhibition zone in strain G12 (shown in Tables 1,2,3).

## Discussion

The effect of the extracts obtained from *Citrullus colocynthis*, using different water ethanol and chloroform extracts the, against the tested organisms. Among these extracts the chloroform was highly inhibition zones, 10.25, 12.8, in strain P1 and 15.6 and 10.8mm in strain P2 and 9.1 and 13.95 mm strain G-12. When ethanol extract was followed by the activities of extracts of three concentration . The inhibition zones was 8.8 mm. Some studies on the antibacterial activities of the leaf extract of *Citrullus colocynthis* have possesses a broad spectrum of activity against a panel of bacteria and fungi responsible for the most common microbial diseases.this result is not simmiler the result obtained by (Tariq Hussain *et al* ,2011) who found that the *Citrullus colocynthis* showed no effect against *Staphylococcus aureus* and *Corynebacterium bovis* but showed some effect against all other tested bacteria. (Rasool Khatibi *et al* 2011) who found that Crude ethanolic extracts of fruits, leaves, stems and roots of *Citrullus colocynthis* Schrad were examined for their antibacterial potentialities against Gram positive and Gram negative bacilli. Ethanolic extracts of fruits, leaves, stems and roots were found to be active against Gram positive bacilli, viz., *Bacillus pumilus* and *Staphylococcus aureus*, while fruit and root extracts in double strength gave positive results against Gram positive bacillus (*Bacillus subtilis*). The Gram negative bacilli viz., *Escherichia coli* and *Pseudomonas aeruginosa* showed no response (Muhammad Sajid ,*et al* 2013) who study soil bacterium used Colocynth (*Citrullus colocynthis*) extract with concentration 10 % was effective.

## Conclusion

The study concluded that Chloroform extract was high effect when ethanol extract was followed by the activities of extracts. The water aqueous extract of two concentration 0.1 and 0.01 were active against some tested organisms, but the activity of the *Citrullus colocynthis* water extract was low.

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## References

Bais HP, Weir TL, Perry LG, Gilroy S, Vivanco JM. The role of root exudates in rhizosphere interactions with plants and other organisms. *Annu. Rev. Plant Biol.* 2006;57:233–266. [PubMed]

Gurudeeban, S., Rajamanickam,E., Ramanathan, T and Satyavani, K .Antimicrobial Activity Of *Citrullus Colocynthis* In Gulf Of Mammal. *International Journal of Current Research* , 2010, Vol. 2, pp. 078-081,

Gurusiddaiah, S., Winaward, L. D., Burger D. and Graham, S. O.Pantomycin: a new antimicrobial antibiotic. *Mycologica* . 1979,71:103-118.

Ionela. D and Ion I. Băra. Plant Products As Antimicrobial Agents. *Analele Științifice ale Universității „Alexandru Ioan Cuza”*, Secțiunea Genetica și Biologie Moleculară, TOM VIII. 2007.56.

Rifat H., Safdar A, Ummay A, Rabia K and Iftikhar A. Soil beneficial bacteria and their role in plant growthpromotion: a review. *Ann Microbiol* DOI,2010, 10.1007/s13213-010-0117-1.

Tariq Hussain, Muhammad Arshad, Sarzamin Khan, Hamid, Sattar And Muhammad Subhan Qureshi. *In vitro* screening of methanol plant extracts for their antibacterial activity. *Pak. J. Bot.*, 2011, 43(1): 531-538.

Rasool Khatibi, Mohammadreza DahmardehGhaleno and Akbar Fakhireh. Effect of alcoholics extract on rat livers and antibacterial screening of *Citrullus colocynthis*. *Journal of Horticulture and Forestry* ,2011,Vol. 3(13), pp. 386-391,

Muhammad Sajid, Abdul Rashid, Muhammad Ehetisham-ul-haq,Muhammad TalhaJaved, Humera Jamil, Muhammad Mudassir, Muhammad Farooq, Faqir Ahmad, Muhammad Latif, Munir Ahmad Chohan, Masood Ahmad8 and Ali Kamran1. *In vitro* evaluation of chemicals and plant extracts against colony growth of *Xanthomonas axonopodis* pv. *malvacearum* causing bacterial blight of cotton. *European Journal of Experimental Biology*, 2013, 3(1): 617-621

John uri lloyd, and cincinnati, O. *Citrullus Colocynthis*. The Western Druggist chicago,USA,1989.

Borhade P,Deshmukh T, Patil V and Khandelwal,K. *Citrullus Colocynthis*. *IJRPC*, 2013 3(1)46-53.

**Table.1.Test extracts against Rhizosphere bacteria P1**

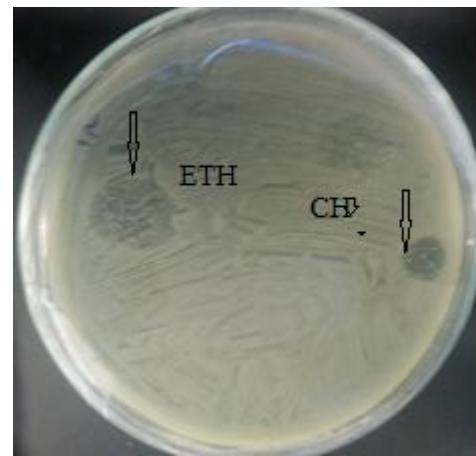
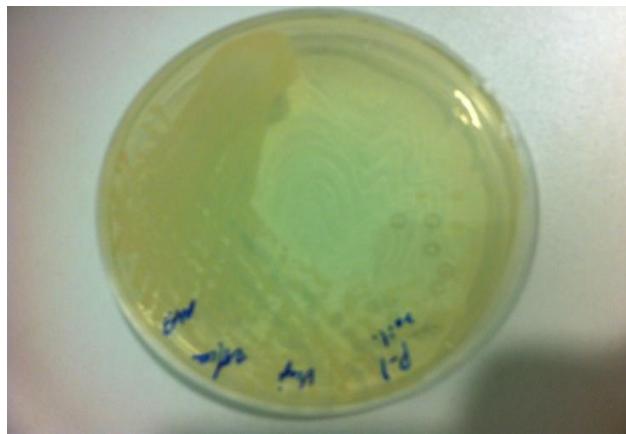
Concentration/mg/ml	Water extract	Ethanol extract	chloroform
0.001	0	0	0
0.01	0	0	10.25
0.1	0	8.8	12.8

**Table.2.Test extracts against Rhizosphere bacteria P2**

Concentration/mg/ml	Water extract	Ethanol extract	chloroform
0.001	0	12.8	10.8
0.01	0	13.5	13
0.1	0	14	15.6

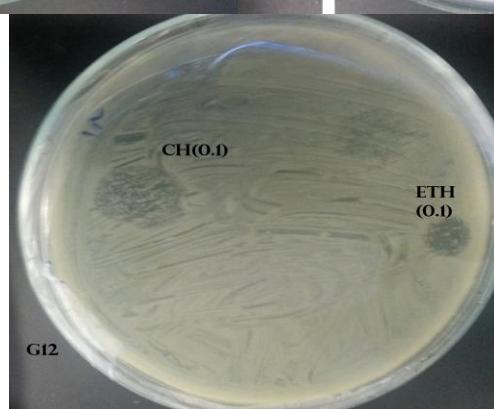
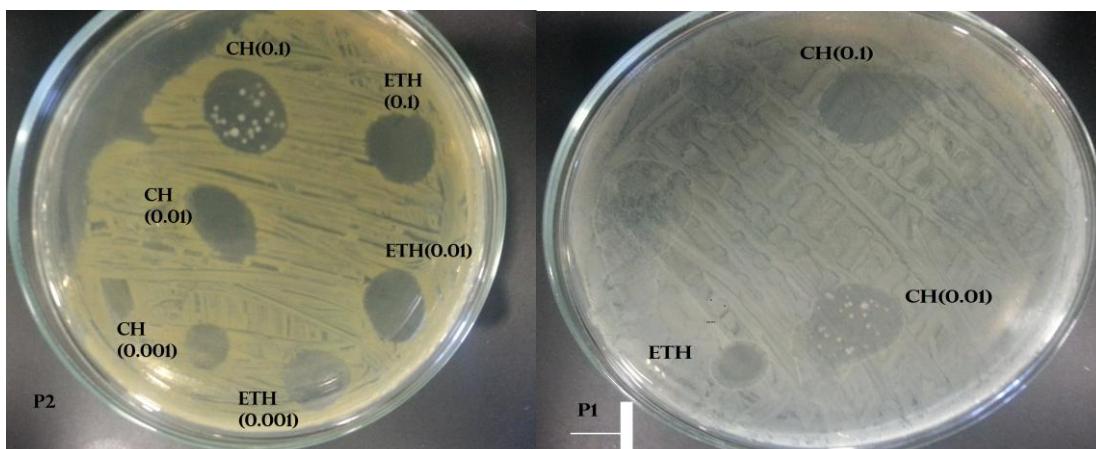
Table.3. Test extracts against Rhizosphere bacteria G-12

Concentration/mg/ml	Water extract	Ethanol extract	chloroform
0.001	0	0	0
0.01	0	0	9.1
0.1	0	9.5	13.95



Rhizosphere

bacteria positive control(CH= chloroform, ETH =ethanol ) negative control



Test extracts against Rhizosphere bacteria