Journal of Tropical Pharmacy and Chemistry

Journalhomepage:https://jtpc.farmasi.unmul.ac.id

# Isolation and Evaluation of Antibacterial Potential Test of Plant *Carthamus oxycantha*

**Amir Hassan[1,*], Ibad Ullah[2], Waqas Ahmad[2]**

[1]Department of Chemistry, Government Post Graduate College Mardan, Abdul Wali Khan University, Mardan 23200 Khyber Pakhtunkhwa, Pakistan
[2]Department of Botany, Government Post Graduate College Mardan, Abdul Wali Khan University, Mardan 23200 Khyber Pakhtunkhwa, Pakistan
*corresponding author:amirhassan741@gmail.com

## Abstract

The present investigation was initiated to find a suitable alternative to synthetic antibiotics for the management of diseases caused by bacteria. *Carthamus oxycantha*.L locally known as wild safflower member of family *Asteraceae* that grows wildly. The study was conducted using as Agar well diffusion to trace the antibacterial potential for to evaluate the efficiency of ethanolic extract of *Carthamus oxycantha* with concentration of 05, 10, 15, and 20 mg/ml against gram-positive *Staphylococcus aureus* and gram-negative *Escherichia Coli* species and them compared with that of Clindamycin, Ampicillin and Kanamycin (10 mg). Zone of inhibition for the extracts were 10.667 to 20.00 mm as compared to standard drug Clindamycin, Ampicillin and kanamycin (15.00-20.00 mm). Antibacterial assays indicates that *Carthamus oxycantha* has potential natural antimicrobial agents against *E-coli* and *S. aureus*. The findings of the present study suggested that ethanolic extract of *C. oxycantha* has strong potential to serve as possible antibacterial.

**Keywords:** *Carthamus oxycantha*; Isolation; Antibacterial; Agar well diffusion; E-coli; *S. aureus*

**Submitted**: 05 May 2021 **Accepted**: 23 August 2021 **DOI**: https://doi.org/10.25026/jtpc.v5i4.343

## 1 Introduction

As the world population increase people's nature about diet becomes more conscious and sensible, that initiates a major change in approach towards healthy and balanced nutrition. Omnivorous nutrition uptake contains a variety of chemical compounds that have solution against number of long lasting diseases like cardiac disorders, cancer, helping in bone fitness, immune performance and also in regulating human body functions by curing such disease medicinal plants are take part [1]. It is noticed that the medicinal plants were

used centuries ago especially in rural parts of the developing and under developed countries like china, India and other Asian countries, to cure the infectious disease [2]. Almost 20% of plants yields are involved in pharmacological or biotic tests [3]. Plant constituents are important source against severe disease. From ancient times large number of medicinal plants are used as therapeutic. That contain some important chemically active molecules like alkaloids, tannins, flavonoids, and phenolic compounds producing positive biological actions on organism body [4]. Scientist have great attention in these biologically vigorous compounds found in medicinal plants. Which is essential for antimicrobial against various infectious disease [5-9].

Rise in population and an extreme progressions and developments in science, microbial disease replicate the death rate in many emerging countries. Use of these antibiotic on human's treatment evolved the numerous opposition in bacterial strains like efflux of antibiotics. About 10% indigenous population use flowering plants for to treat numerous infections. While slightly 1% are documented by recent researcher [10]. Antibiotics initiate a way for the treatment of microbe's damages. From the discovery the chemotherapeutic role create confidence in the medical fields also leads to ultimate extinction of contagious ailments. Misuse of these antibiotics tends to the appearance and distribution of resistant strains of microorganisms [11].

Microbial infectious disease involve in killing nearly 50,000 people per day. *E. coli, Vibrio cholera, Shigellaspp,* and *Staphylococcus aureus* are some common bacterial pathogens [12]. But resistance of these microbes to antibiotics drugs causing major health problems. About 70% of bacteria are almost resistance to one of the antibiotic drug that is frequently used against them [13]. Ancient medication are likely to enhanced in evolving countries as substitutes to health difficulties [14]. Particularly the developing regions of the world need inexpensive, reliable and innovative therapeutics to treat infectious disease [15]. Medicinal plant have incredible capacity to yield a diversity in secondary metabolites like alkaloids, terpenoids, saponins, steroids, flavonoids & tannins etc [16].

From thousand years nature provide foundation for therapeutic agents. An extensive known herbal tonics were explained in Holy books like Quran, Bible & Vedas. According to WHO world population with almost 80% use herbal medicines for a prim health issues [17]. Microbes are likely to cause diseases in unicellular as well as in multicellular organism. Commonly viruses, bacteria and fungi act as a key pathogenic creatures. In early 20$^{th}$ century the detection of Antibiotics initiate its significance in fighting against bacterial disease. By using Antibiotics in such a large scale, many bacterial strains become unaffected to these antibiotics. This obligation leads to investigate the potential of traditionally therapeutic medicinal plants as antibiotics. Plant extracts are broadly used to treat these bacterial disease [18]. As antibiotics discovered in the mid-twentieth century create a major breakthrough in upholding and treatment of bacterial diseases causes series infections. worldwide fatal diseases were now curable. Since then, antimicrobial agents saved the lives and reduced the bearing of millions of people. Today, antibiotics are crucial for bacterial infections as well as for curing of high risk patient's. Massive synthesis of penicillin in 1943 play a vital role in curing and reducing illness and death rates. However, within four years, bacteria began appearing that could resist the action of penicillin. Pharmaceutical businesses began for to develop new as well as more reactive antibiotics.

More than half century people use it as wonder drug but with variation in bacterial strains create a bit anti behavior against these antibiotics [19]. Solving this problems pharmacological industries synthesis a variety of antibiotics in last 30 years period. Just because that bacteria species have ability in transmitting and to modify their genetic makeup which tend to make it more resistive against these antibiotics [20]. Traditional practice of medication is quite necessary for possible future medicines. Almost 122 compounds are discovered that are involved in alternate as well as in modern medicines and also play an important role in world economy [21-22]. A number of plant families like Asteraceae especially in Pakistan have a variety

of medicinal plants. *Carthamus oxycantha* is one wed found in agricultural fields. Having ability in anti-inflammatory and wounds healing also help in irritation observe in *albino* rabbits Asteraceae, a leading dicotyledon angiosperm family that includes 1000 genera and about 10,000 species worldwide. The present investigation was initiated to find a suitable alternative to synthetic antibiotics for the management of diseases caused by bacteria. The findings of the present study suggested that ethanolic extract of *C. oxycantha* has strong potential to serve as possible antibacterial.

## 2 Materials and Methods

### 2.1 Collection and Preparation of Carthamus oxycantha

All the experimental procedure followed as performed in [23-33]. Total plant (except root) was used as plant materials. Diseased Free whole plant (fresh) sample of *Carthamus oxycantha* collected from its natural habitat located on the ground of Govt. Post Graduate Collage Mardan Pakistan, and were identified and authenticated at the Department of Botany (GPGC Mardan). The plants samples were brought to the laboratory and carefully washed to remove debris as well as dust corpuscles and rinsed with distilled water then spread on a table and air dried for 14 days at room temperature until the plant became crispy and ready for grinding and powdering After then the plant was ground into fine powder using grinding machine

### 2.2 Instruments and reagents used

Grinding machine, shaker, portable autoclave, drier, beakers, glass rod, crock borer, conical flasks, round-bottom flasks, round-mouth bottle, No. 1 Whatman filter paper, aluminum foil, Nutrient agar medium, ethanol, Dimethyl sulfoxide (DMSO) and distilled water.

### 2.3 Extraction of Plant Materials

Precisely 400 g of crushed plant powder was added in 1.5 liter of 99.9% pure ethanol as a solvent for 4 days in a large sized round mouth bottle. The mixture was commoved just after the addition of the solvent. At regular time interval of proper shaking for four days the solution was filtered with fine piece of cloth into a clean jar and also filtered it through No. 1 Whatman filter paper. The filtrate (plant extract) was dried at room temperature.

### 2.4 Preparation of Test Organisms

Clinical isolate of disease causing strains of Gram-positive *Staphylococcus aureus* and Gram-negative *Escherichia coli* were kindly provided by microbiology department ta Bacha Khan medical college, and were maintained further experiments in nutrient agar medium at 4 ˚C. It was sub-cultured into nutrient agar medium and incubated at 37˚Cfor 24 hour that to cover the agar medium surface properly.

### 2.5 Culture Media and Chemicals

Nutrient agar was used as media for conducting this Activity. Also ethanol and dimethyl sulfoxide (DMSO) was used as lab chemicals for extraction as well as solution preparation. The media and chemicals were provided by Department of Botany GPGC Mardan.

### 2.6 Agar Medium Preparation

19 gm nutrient agar were dissolved in 1500 ml distilled water. Sterilized glass rod was used for dissolving agar solution. After completely dissolving the solution, each of the petri dish was fill up to half of its original depth. Agar medium allowed to solidify for 10 min.

### 2.7 Extract Solution Preparation

Plant extract solution with concentration of 05, 10, 15 and 20mg/ml were used for conducting this experiment. 2ml of pure DMSO was dissolved in 18 ml of distilled water to prepare the 20% of DMSO solution. 5, 10, 15, 20mg of plants extract were taken in Eppendorf and add 1 ml of 20% DMSO solution using ml micro pipette for to prepare the concentration.

### 2.8 Antibiotics activity assay

Standard antibiotics clindamycin, ampicillin and kanamycin was used against tested organism. Antibiotic discs directly

placed on the surface of a nutrient agar medium that has been protected with test bacterial strains. Antibiotics diffuse outward through incubation from the discs creating a concentration gradient. After 24 hours, the inhibition zones diameter is measured. As the name indicate the standard antibiotics show a strong restriction against the test organisms. The values that are reported for *Escherichia coli* was given in Table 1 and data for *Staphylococcus aureus* Table 3.

### 2.9 Well diffusion Method and Plant Extract assay

Antibacterial activity was evaluated using method of agar-well diffusion as recommended by [23-33]. Four wells of 6 mm in diameter using a cork borer (sterile) were made in agar nutrient media plate that was previously seeded with standardized bacterium. Plant extracts with concentration of 05, 10, 15, and 20 mg/ml, A standard antibiotics clindamycin, ampicillin and kanamycin for positive control) was added into each well on the medium. Wait for 30 min as allowed it for to diffuse it properly. After incubation at 37 °C for 24hrs, the resulting zones of inhibition around the wells were measured using a transparent ruler centimeter and then convert the values in millimeters (mm). Numerical data of that zone of inhibition was represented in **Table 1** and **Table 2**.

### 2.10 Sterilization Process

All the instruments, equipment and solutions was sterilized for 30 min at 150$^{o}$C with constant pressure of 15 psi, using portable autoclave machine.

## 3 Results and Discussion

The Ethanolic plant extract of plant species named as *Carthamus oxycantha.* L, show significant ability against bacterial strains listed as *Staphylococcus aureus* and *Escherichia coli,* gram positive and gram negative respectively. Plant extract used in agar well diffusion varied in antibacterial activeness. The highest inhibitory effect to the extract was observed on *Staphylococcus aureus* (Table 3; Table 4; and Figure 2) with a zone of inhibition of 15 mm with a solution concentration of 20mg/ml, while at the same concentration of ethanol plant extract of 20mg/ml against *Escherichia coli* was the least inhibited with a zone of 09 mm. Three standard antibiotics were also applied against the bacterial strains and as result indicate a complete inhibition against bacterial strains. Numerical data are shown in Table 1 and Table 2.

The data regarding with mean zone of inhibition of *Carthamus Oxycantha* against *Escherichia coli* given in (Table 1 & Figure 1). The maximum mean inhibition zone was recorded using kanamycin in T3 is 26. This value is followed by T1 which was 20. The minimum zone of inhibition is recorded in T7.This valvue is followed by T4 and T6 with a value of 12. So the result show that standerd antibiotic kanamycin have strong affection against bacterial strains. Statistical analysis for *E.coli* in Table 2. While *S. aureus* in Table 4.

Table. 1: Standard deviation and mean for inhibition zones of Carthamus oxycantha against E. coli

| S.No | Treatment | Concentration | Zone of inhibition (Mean value) | Standard Deviation (SD) |
|---|---|---|---|---|
| 1 | T1 | Clindamycin (Standard) | 20.00$^{b}$ | 1.5275 |
| 2 | T2 | Ampicillin (Standard) | 15.00$^{c}$ | 1.5275 |
| 3 | T3 | Kanamycin (Standard) | 26.00$^{a}$ | 1.5275 |
| 4 | T4 | 5mg | 11.333$^{d}$ | 1.5275 |
| 5 | T5 | 10mg | 12.333$^{d}$ | 1 |
| 6 | T6 | 15mg | 11.333$^{d}$ | 1 |
| 7 | T7 | 20mg | 10.667$^{d}$ | 1 |

LSD value at 0.05 level of significance = 2.3245. Mean followed by the same English letter are not significantly different.

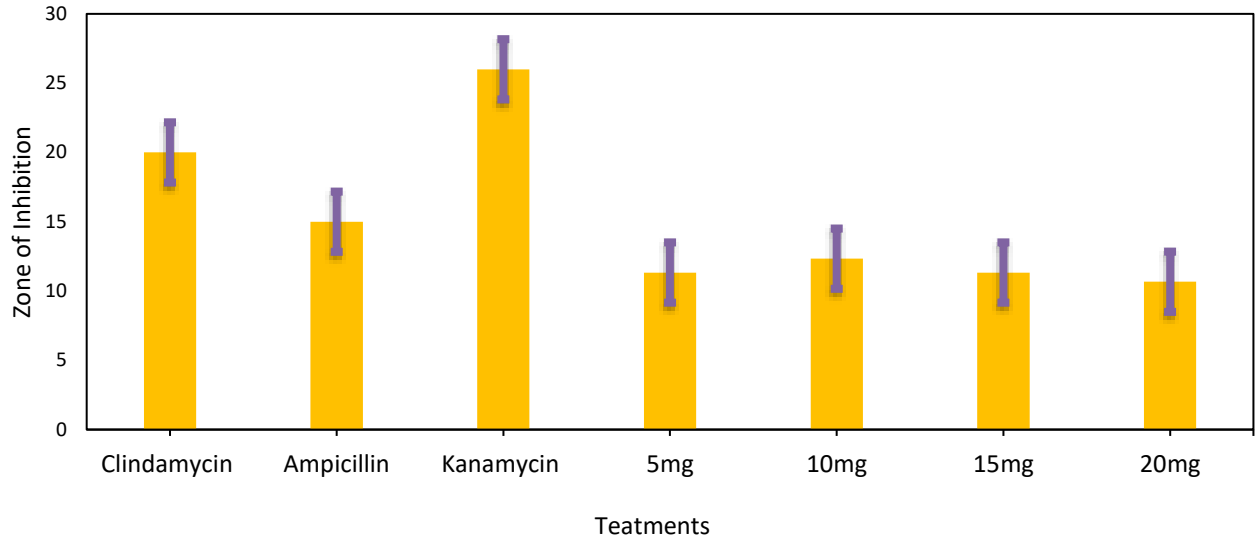


**Figure 1:** Graph for mean zone of inhibition of *Carthamus oxycantha* against *E. coli.*

Table. 2: ANOVA table for antibacterial activity of Carthamus oxycantha against *E. coli*:

| Source | Degree of freedom (DF) | Sum of squares (SS) | Mean square (MS) | F-value | P-value |
|---|---|---|---|---|---|
| Concentration | 6 | 595.143 | 99.1905 | 56.30 | 0.0000 |
| Error | 14 | 24.667 | 1.7619 | | |
| Total | 20 | 619.810 | | | |

Co-efficient of variation: 8.71

Table. 4: ANOVA table for antibacterial activity of *Carthamus oxycantha* against *Staphylococcus aureus*.

| Source | Degree of freedom (DF) | Sum of squares (SS) | Mean square (MS) | F-value | P-value |
|---|---|---|---|---|---|
| Concentration | 6 | 555.905 | 92.6508 | 41.40 | 0.0000 |
| Error | 14 | 31.333 | 2.2381 | | |
| Total | 20 | 587.238 | | | |

Co-efficient of variation: 9.64

**Table.3:** Standard deviation and Mean of zone of inhibition of *Carthamus oxycantha* against *S. aureus.*

| S.No | Treatment | Concentration | Zone of inhibition (Mean value) | Standard Deviation (SD) |
|---|---|---|---|---|
| **1** | T1 | Clindamycin (Standard) | 20.00$^{b}$ | 2 |
| **2** | T2 | Ampicillin (Standard) | 15.00$^{c}$ | 1.5275 |
| **3** | T3 | Kanamycin (Standard) | 26.00$^{a}$ | 2 |
| **4** | T4 | 5mg | 11.00$^{d}$ | 1.5275 |
| **5** | T5 | 10mg | 11.333$^{d}$ | 1 |
| **6** | T6 | 15mg | 12.00$^{d}$ | 1 |
| **7** | T7 | 20mg | 13.333$^{cd}$ | 1 |

LSD value at 0.05 level of significance = 2.6199 Mean followed by the same English letter are not significantly different.

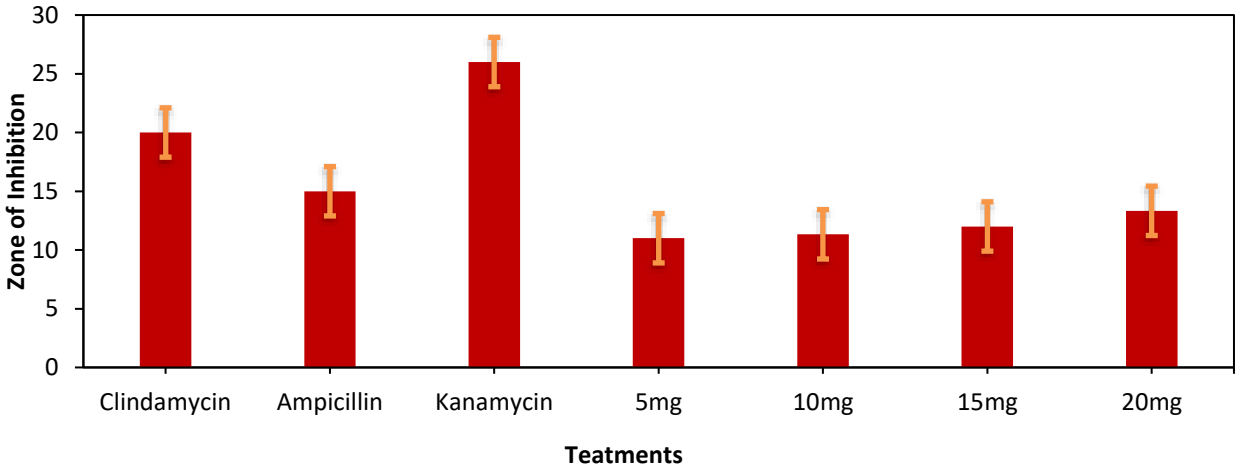


**Figure 2:** Graph for mean zone of inhibition of *Carthamus oxycantha* against *S. aureus*.

Plants, herbs, and spices are traditionally finest structures provide a rich foundation of naturally occurring compounds that hold antimicrobial attention against many bacterial pathogenic strains [34]. A massive worldwide increase specially in South Asia particularly in India, Pakistan, and Afghanistan labeled as under developed countries the rate of morbidity and mortality majorly caused by pathogenic multidrug resistant bacterial strains which reduce susceptibility to antibiotics, provide a way to investigate new varieties of antibacterial agents for to treat such diseases in plants as well as in [35-36].

Plants with a mixture of vital chemical constituents as secondary metabolites like flavonoids and terpenoids as well as Tannins, glycosides and alkaloids, now a days play a significant role in curing a number of infectious disease in human's life. Research study indicate a ratio of the presence of the plants in antibiotics is least about 25% [37]. The present study was recorded as a step in that chain of finding some new and efficient antibiotics. Two sample bacterial strains of *staphylococcus aureu s* (gram-positive) and *Escherichia coli* (gram negative) were screened against plants extract. Antimicrobial assay of *Carthamus oxycantha* by using agar well diffusion process, ethanolic plant extract with a concentration of 20mg/ml show a strong inhibition with a mean of 15mm zone against *staphylococcus aureus* and as same concentration of 20mg/ml, the mean inhibition zone of plants extract (*C. oxycantha*) against *Escherichia coli* is 9mm. Thus it leads a way that the selected plant have a must ability to contest against pathogenic bacterial strains. Some of the standard antibiotics (clindamycin, kanamycin and Ampicillin) were also tested against the sample bacterial strains. Result of this activity show that the plant (*C. oxycantha*) have a strong attention against bacterial strains and further research will find out some more specific constituents regarding with the antibacterial efficiency [38-41].

As [42-45] conducted an experiment to observed the antibacterial ability of *Eucalyptus camaldulensis* (leaf extracts) for bacterial strains of *Klebsiellaspp, Salmonella typhi, Yersinia enterocolitica, Pseudomonas aeruginosa (Gram-negative), Staphylococcus aureus* and *Bacillus subtilis.* Dichloromethane fraction, methanol residue and methanol extract were applied using agar-well diffusion method. At least 10 mg/ml extract show significant result against tested strains while petroleum ether fraction showed no inhibition zones. A standard antibiotic drug (gentamycin) were also used. *Bacillus subtilis*with extract amount of 10mg/ml show inhibition zone at least 0.04 mm. chemical analysis of plants extract indicate the active components of tannins, saponins and cardiac glycosides. The result initiate that *Eucalyptus camaldulensis*leaves have potential against bacteria.

While [46-49] carry out a research on antibiotic resistance of *Parquetinani grescens* against *Pseudomonas aeruginosa,*

*Staphylococcus aureus, Klebsiella pneumoniae, Enterococcus feacalis* and *Escherichia coli.* Plant extract of two types aqueous and ethanol were used conducting this activity. Aqueous extract show low inhibition against most of the bacterial strains while *E. coli* show inhibition toward methanol extract. Aqueous extract with concentration of 100 mg/ml and ethanol extract of 300 mg/ml show inhibition of >12 mm and >20 mm respectively against *S. Aureus.* A standard antibiotics methicillin were also used which show 17-26 mm zone of inhibition for bacterial strains. For MIC aqueous extract concentration of 25-50 mg/ml while methanol extract of 12.5-50 mg/ml were taken.

In [50-55] tested the antibacterial capability of methanolic plant extract of *Marrubiumvulgare* against *Escherichia coli, Bacillus subtilis, Staphylococcus aureus, S. epidermidis, Pseudomonosaeruginosa* and *Proteus vulgaris*. Extract were applied with concentration of 50, 100, 200, 400 and 600 mg/ml by Disk diffusion method. A standard antibiotics ciprofloxacin with amount of 10 µg/ml were also used. The study comprised that methanolic crude extract have strong inhibition against *B. subtilis*, *S. epidermidis* and *S. aureus* while fairly effective against *P. vulgaris* and *E. coli*. But *P. aeruginosa*extract don't show any effect.

While [56-58] stated tha potential of Acacia aroma towards the bacterial strains like methicillin-resistant *Staphylococcus aureus*, methicillin sensitive *Staphylococcus aureus*as well as Methicillin-resistant *Staphylococcus epidermidis.* Leaf extract were obtained using ethyl acetate and ethanol and are applied by agar well diffusion method. (MIC) of acacia leaf extract are in between 2.5 to 10 mg/ml as well as from 2.5 to 5 mg/ml individually. Beside organic extracts aqueous show less restriction ability towards tested bacterial strains. The result show clear indication toward future planes in antibacterial field for *Acacia aroma.*

## 4 Conclusions

It is concluded from present study that ethanolic plant extract of *Carthamus oxycantha*. L show significant result and initiate strong antibacteriual inhibition against sample bacterial strains. In the antibacterial potential and the efficiency of ethanolic extract of *Carthamus oxycantha* with concentration of 05, 10, 15, and 20 mg/ml against gram-positive *Staphylococcus aureus* and gram-negative *Escherichia Coli* species and them compared with that of Clindamycin, Ampicillin and Kanamycin (10 mg). Zone of inhibition for the extracts were 10.667 to 20.00 mm as compared to standard drug Clindamycin, Ampicillin and kanamycin (15.00-20.00 mm). Antibacterial assays indicates that *Carthamus oxycantha* has potential natural antimicrobial agents against *E-coli* and *S. aureus*. Plant extract with a positive sign toward antibacterial ability shows enhancement in field the of Antibiotics.

## 5 Acknowledgments

The authors thanks (Waqas Ahmad and Ibad Ullah) to his supervisor Prof. Muhammad Israr of Botany Department Government Post Graduate College Mardan Khyber Pakhtunkhwa Pakistan and also acknowledges Amir Hassan of Chemistry department for assembling and Submission and Publishing of the Manuscript.

## 6 Conflicts of Interest

The authors declare no conflict of interest.

## 7 References

[1] Srivastava, J.; Lambert, J.; Vietmeyer, N., *Medicinal plants: An expanding role in development*. World Bank Publications: **1996**; Vol. 320.

[2] Zhang, X.; Organization, W. H., Traditional medicine strategy 2002 2005. **2002**.

[3] Suffredini, I.; Sader, H. S.; Gonçalves, A. G.; Reis, A. O.; Gales, A. C.; Varella, A. D.; Younes, R. N., Screening of antibacterial extracts from plants native to the Brazilian Amazon Rain Forest and Atlantic Forest. *Brazilian journal of medical and biological research* **2004,***37* (3), 379-384. https://doi.org/10.1590/S0100-879X2004000300015

[4] Edeoga, H. O.; Okwu, D.; Mbaebie, B., Phytochemical constituents of some Nigerian medicinal plants. *African journal of biotechnology* **2005,***4* (7), 685-688.

[5] Kaushik, R.; Garg, G.; Sharma, G.; Arora, C., Antibacterial activity of plant extracts from Uttaranchal Hills, India. *Allelopathy Journal* **2003,***12* (2), 205-214.

[6] Hassan, A.; Akmal, Z.; Khan, N., The Phytochemical Screening and Antioxidants Potential of Schoenoplectustriqueter L. Palla.

*Journal of Chemistry* **2020,***2020*.https://doi.org/10.1155/2020/3865139
[7] Hassan, A.; Ullah, H., Antibacterial and antifungal activities of the medicinal plant veronica biloba. *Journal of Chemistry* **2019,***2019*. https://doi.org/10.1155/2019/5264943
[8] Hassan, A., Traditional Chinese Herbal Medication for corona-virus 2019. *Travel medicine and infectious disease* **2020,***36*, 101810. https://doi.org/10.1016/j.tmaid.2020.101810
[9] Hassan, A.; Ashfaq, M.; Khan, A.; Khan, M. S., Isolation of Caffeine from Carbonated Beverages. *Journal of Tropical Pharmacy and Chemistry* **2020,***5* (1), 33-38. https://doi.org/10.25026/jtpc.v5i1.234
[10] Kafaru, E., Immense help formative workshop. *Essential pharmacology* **1994,***1*.
[11] Harbottle, H.; Thakur, S.; Zhao, S.; White, D., Genetics of antimicrobial resistance. *Animal biotechnology* **2006,***17* (2), 111-124. https://doi.org/10.1080/10495390600957092
[12] Namita, P.; Mukesh, R., Medicinal plants used as antimicrobial agents: a review. *Int Res J Pharm* **2012,***3* (1), 31-40.
[13] Dahiya, P.; Purkayastha, S., Phytochemical screening and antimicrobial potentials of Alangiumsalvifolium and Piper longum against multi-drug resistant bacteria from clinical isolates. *International Journal of Pharmacy and Pharmaceutical Sciences* **2011,***3* (5), 462-465.
[14] Riazunnisa, K.; Adilakshmamma, U.; Khadri, C., Phytochemical analysis and in vitro antibacterial activity of Soymidafebrifuga (Roxb.) Juss. and Hemidesmus indicus (L.). *Indian J. Appl. Res* **2013,***3*, 57-59. https://doi.org/10.15373/2249555X/DEC2013/14
[15] Awouafack, M. D.; McGaw, L. J.; Gottfried, S.; Mbouangouere, R.; Tane, P.; Spiteller, M.; Eloff, J. N., Antimicrobial activity and cytotoxicity of the ethanol extract, fractions and eight compounds isolated from Eriosemarobustum (Fabaceae). *BMC Complementary and Alternative Medicine* **2013,***13* (1), 1-9.
[16] Das, K.; Tiwari, R.; Shrivastava, D., Techniques for evaluation of medicinal plant products as antimicrobial agents: current methods and future trends. *Journal of medicinal plants research* **2010,***4* (2), 104-111.
[17] Ali, M. N.; Khan, M. M., Antibacterial activity of Herbal and Spice Extracts on Bacterial Clinical Isolates of Urinary Tract Infections. *Journal of Chemical, Biological and Physical Sciences (JCBPS)* **2017,***7* (3), 718.
[18] Bandaranayake, W., Traditional and medicinal uses of mangroves. *Mangroves and salt marshes* **1998,***2* (3), 133-148.
[19] Group, S. f. M. o. A. T. S., CD4+ count–guided interruption of antiretroviral treatment. *New England Journal of Medicine* **2006,***355* (22), 2283-2296. https://doi.org/10.1056/NEJMoa062360
[20] Nascimento, G. G.; Locatelli, J.; Freitas, P. C.; Silva, G. L., Antibacterial activity of plant extracts and phytochemicals on antibiotic-resistant bacteria. *Brazilian journal of microbiology* **2000,***31* (4), 247-256.
[21] Rehman, S.; Hasnat, A.; Hasan, C.; Rashid, M.; Ilias, M., Pharmacological evaluation of Bangladeshi medicinal Plant—A Review. *Pharmacol. Biol* **2001,***37*, 202-207. https://doi.org/10.1076/phbi.39.1.1.5939
[22] Fabricant, D. S.; Farnsworth, N. R., The value of plants used in traditional medicine for drug discovery. *Environmental health perspectives* **2001,***109* (suppl 1), 69-75. https://doi.org/10.2307/3434847
[23] Mocan, A.; Vodnar, D.; Vlase, L.; Crişan, O.; Gheldiu, A.M.; Crişan, G. Phytochemical characterization of Veronica officinalis L., V. teucrium L. and V. orchideaCrantz from Romania and their antioxidant and antimicrobial properties. *International journal of molecular sciences* **2015**, *16*, 21109-21127, https://doi.org/10.3390/ijms160921109
[24] Dunkic, V.; Kosalec, I.; Kosir, I.J.; Potocnik, T.; Cerenak, A.; Koncic, M.Z.; Vitali, D.; Muller, I.D.; Kopricanec, M.; Bezic, N. Antioxidant and antimicrobial properties of Veronica spicata L.(Plantaginaceae).*Current drug targets* **2015**, *16,* 1660-1670, https://doi.org/10.2174/1389450116666150531161820
[25] Sharifi-Rad, J.; Iriti, M.; Setzer, W.N.; Sharifi-Rad, M.; Roointan, A.; Salehi, B. Antiviral activity of Veronica persica Poir. on herpes virus infection. *Cellular and Molecular Biology* **2018**, *64*, 11-17, https://doi.org/10.14715/cmb%2F2018.64.8.2
[26] Stojković, D.S.;Živković, J.; Soković, M.; Glamočlija, J.; Ferreira, I.C.;Janković, T.; Maksimović, Z. Antibacterial activity of Veronica montana L. extract and of protocatechuic acid incorporated in a food system.*Food and Chemical Toxicology* **2013**, *55*, 209-213, https://doi.org/10.1016/j.fct.2013.01.005
[27] Sharifi-Rad, J.; Tayeboon, G.S.; Niknam, F.; Sharifi-Rad, M.; Mohajeri, M.; Salehi, B.; Iriti, M.; Sharifi-Rad, M. Veronica persica Poir. extract–antibacterial, antifungal and scolicidal

activities, and inhibitory potential on acetylcholinesterase, tyrosinase, lipoxygenase and xanthine oxidase. *Cellular and Molecular Biology* **2018**, *64*, 50-56, https://doi.org/10.14715/cmb/2018.64.8.8

[28] Alabri, T.H.A.;AlMusalami, A.H.S.;Hossain, M.A.;Weli, A.M.;Al-Riyami, Q. Comparative study of phytochemical screening, antioxidant and antimicrobial capacities of fresh and dry leaves crude plant extracts of Datura metel L. *Journal of King Saud University-Science* **2014**, *26*, 237-243, https://doi.org/10.1016/j.jksus.2013.07.002

[29] Alghazeer, R.; El-Saltani, H.; Saleh, N.; Al-Najjar, A.; Hebail, F. Antioxidant and antimicrobial properties of five medicinal Libyan plants extracts. *Natural science* **2012**,*4,* 324, https://doi.org/10.4236/ns.2012.45045

[30] Edziri, H.; Mastouri, M.; Cheraif, I.; Aouni, M. Chemical composition and antibacterial, antifungal and antioxidant activities of the flower oil of Retama raetam (Forssk.) Webb from Tunisia.*Natural product research* **2010**, *24*, 789-796, https://doi.org/10.1080/14786410802529190

[31] Cushnie, T.T.; Lamb, A.J. Antimicrobial activity of flavonoids. *International journal of antimicrobial agents* **2005**, *26,* 343-356, https://doi.org/10.1016/j.ijantimicag.2005.09.002

[32] Hernández, N.E.; Tereschuk, M.; Abdala, L. Antimicrobial activity of flavonoids in medicinal plants from Tafı del Valle (Tucuman, Argentina).*Journal of Ethnopharmacology* **2000**, *73,* 317-322, https://doi.org/10.1016/s0378-8741(00)00295-6

[33] Epand, R.F.; Savage, P.B.; Epand, R.M. Bacterial lipid composition and the antimicrobial efficacy of cationic steroid compounds (Ce ragenins). *Biochimica et Biophysica Acta (BBA)-Biomembranes***2007**, *1768*, 2500-2509, https://doi.org/10.1016/j.bbamem.2007.05.023

[34] Kumar, V. P.; Chauhan, N. S.; Padh, H.; Rajani, M., Search for antibacterial and antifungal agents from selected Indian medicinal plants. *Journal of ethnopharmacology* **2006,***107* (2), 182-188. https://doi.org/10.1016/j.jep.2006.03.013

[35] Koochak, H.; Seyyednejad, S. M.; Motamedi, H., Preliminary study on the antibacterial activity of some medicinal plants of Khuzestan (Iran). *Asian Pacific Journal of Tropical Medicine* **2010,***3* (3), 180-184. https://doi.org/10.1016/S1995-7645(10)60004-1

[36] Dwivedi, S.; Dudey, R.; Tyagi, R.; Masand, M.; Advani, U., Medicinal Bioactives as antimicrobial agents: an overview. *Int. J. Pharm. Dev* **2011,***3*, 24-30.

[37] Malesh, B.; Satish, S., Antimicrobial Activity of some important medicinal plant against plant and human pathogen. *World Journal of Agriculture Sciences* **2008,***4* (5), 839-843.

[38] Sanyal, B.; Sharma, A. Cytological Studies in Indian Cyperaceae. *Cytologia* **1972,** *37*, 13-32, https://doi.org/10.1508/cytologia.37.13

[39] D'abrosca, B.; Dellagreca, M.; Fiorentino, A.; Isidori, M.; Monaco, P.; Pacifico, S. Chemical constituents of the aquatic plant Schoenoplectus lacustris: evaluation of phytotoxic effects on the green alga Selenastrum capricornutum. *Journal of chemical ecology* **2006,** *32*, 81-96, https://doi.org/10.1007/s10886-006-9354-y

[40] Liu, X.; Wang, Z.; Zhang, X.; Wang, J.; Xu, G.; Cao, Z.; Zhong, C.; Su, P. Degradation of diesel-originated pollutants in wetlands by Scirpus triqueter and microorganisms. *Ecotoxicology and environmental safety* **2011,** *74*, 1967-1972, https://doi.org/10.1016/j.ecoenv.2011.06.005

[41] Jiménez-Mejías, P.; Luceno, M.; Castroviejo, S. Schoenoplectus corymbosus: a tropical Old-World sedge (Cyperaceae) discovered in Spain and Morocco. *Nordic Journal of Botany* **2007,** *25*, 70-74, https://doi.org/10.1111/j.0107-055X.2007.00007_26.x

[42] Roalson, E.H. A synopsis of chromosome number variation in the Cyperaceae. *The Botanical Review* **2008,** *74*, 209-393, https://doi.org/10.1007/s12229-008-9011-y

[43] Chang, A.K.T.; Frias Jr, R.R.; Alvarez, L.V.; Bigol, U.G.; Guzman, J.P.M.D. Comparative antibacterial activity of commercial chitosan and chitosan extracted from Auricularia sp. *Biocatalysis and agricultural biotechnology* **2019,** *17*, 189-195, https://doi.org/10.1016/j.bcab.2018.11.016

[44] de Aragao, A.P.; de Oliveira, T.M.; Quelemes, P.V.; Perfeito, M.L.G.; Araujo, M.C.; Santiago, J.D.A.S.; Cardoso, V.S.; Quaresma, P.; de Almeida, J.R.D.S.; da Silva, D.A. Green synthesis of silver nanoparticles using the seaweed Gracilaria birdiae and their antibacterial activity. *Arabian Journal of Chemistry* **2019,** *12*, 4182-4188, https://doi.org/10.1016/j.arabjc.2016.04.014

[45] Gummuluri, S.; Kavalipurapu, V.T.; Kaligotla, A.V. Antimicrobial efficacy of Novel Ethanolic Extract of Morinda Citrifolia Against Enterococcus Feacalis by Agar Well Diffusion Method and Minimal Inhibitory Concentration-An Invitro Study. *Brazilian Dental Science*

**2019,** *22*, 365-370, https://doi.org/10.14295/bds.2019.v22i3.1731

[46] Majumder, R.; Dhara, M.; Adhikari, L.; Ghosh, G.; Pattnaik, S. Evaluation of in vitro Antibacterial and Antioxidant Activity of Aqueous Extracts of Olax psittacorum. *Indian Journal of Pharmaceutical Sciences* **2019,** *81*, 99-109, https://doi.org/10.4172/pharmaceutical-sciences.1000484

[47] Raj, S.R.G.; Ng, C.H.; Mohd-Said, S.; Yu, K.X. Antibacterial Potential of Aqueous Extracts and Compounds from Selected Brown Seaweeds. *INNOSC Theranostics and Pharmacological Sciences* **2019,** *2*, https://doi.org/10.26689/itps.v2i1.804

[48] Tlili, H.; Marino, A.; Ginestra, G.; Cacciola, F.; Mondello, L.; Miceli, N.; Taviano, M. F.; Najjaa, H.; Nostro, A. Polyphenolic profile, antibacterial activity and brine shrimp toxicity of leaf extracts from six Tunisian spontaneous species. *Natural product research* **2019**, 1-7, https://doi.org/10.1080/14786419.2019.1616725

[49] Aghraz, A.; Albergamo, A.; Benameur, Q.; Salvo, A.; Larhsini, M.; Markouk, M.; Gervasi, T.; Cicero, N. Polyphenols contents, heavy metals analysis and in vitro antibacterial activity of extracts from Cladanthus arabicus and Bubonium imbricatum of Moroccan Origin. *Natural product research* **2020,** *34*, 63-70, https://doi.org/10.1080/14786419.2019.1573424

[50] Fernández-Agulló, A.; Pereira, E.; Freire, M.S.; Valentao, P.; Andrade, P.; González-Álvarez, J.; Pereira, P. Influence of solvent on the antioxidant and antimicrobial properties of walnut (Juglans regia L.) green husk extracts.*Industrial crops and products* **2013**, *42*, 126-132, https://doi.org/10.1016/j.indcrop.2012.05.021

[51] Roby, M.H.H.;Sarhan,M.A.; Selim, K.A.H.; Khalel, K.I. Evaluation of antioxidant activity, total phenols and phenolic compounds in thyme (Thymus vulgaris L.), sage (Salvia officinalis L.), and marjoram (Origanum majorana L.) extracts.*Industrial Crops and Products* **2013**, *43*, 827-831, https://doi.org/10.1016/j.indcrop.2012.08.029

[52] Schmidley, J. Free radicals in central nervous system ischemia. *Stroke* **1990**, *217*, 1086-1090, https://doi.org/10.1161/01.str.21.7.1086

[53] Hertog, M.G.; Feskens, E.J.; Kromhout, D.; Hertog, M.; Hollman, P.; Hertog, M.; Katan, M. Dietary antioxidant flavonoids and risk of coronary heart disease: the Zutphen Elderly Study. *The Lancet* **1993**, *342,* 1007-1011, https://doi.org/10.1016/0140-6736(93)92876-U

[54] Holiman, P.C.; Hertog, M.G.; Katan, M.B. Analysis and health effects of flavonoids. *Food Chemistry* **1996**, *57*, 43-46, https://doi.org/10.1016/0308-8146(96)00065-9

[55] Moure, A.; Cruz, J.M.; Franco, D.; omıng e , J.M.; Sineiro, J.; omıng e , H.; José Núñez, M.A.; Parajó, J.C. Natural antioxidants from residual sources. *Food Chemistry* **2001**, *72*, 145-171, https://doi.org/10.1016/S0308-8146(00)00223-5

[56] Halliwell, B. The role of oxygen radicals in human disease, with particular reference to the vascular system.*Pathophysiology of Haemostasis & Thrombosis* **1993**, *23*, 118-126, https://doi.org/10.1159/000216921

[57] Meyer, A.S.; Heinonen, M.; Frankel, E.N. Antioxidant interactions of catechin, cyanidin, caffeic acid, quercetin, and ellagic acid on human LDL oxidation.*Food Chemistry* **1998**, *61*, 71-75, https://doi.org/10.1016/S0308-8146(97)00100-3

[58] Hunt,E.J.; Lester, C.E.; Lester, E.A.; Tackett, R.L. Effect of St. John's wort on free radical production.*Life sciences* **2001**, *69,* 181-190, https://doi.org/10.1016/S0024-3205(01)01102-X