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Antibacterial Activity of the *Chennopodium album* Leaves and Flowers Extract

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Abstract**—**Recent years have instance that there is a invigoration of interest in drug discovery from medicinal plants for the support of health in all parts of the world . This study was designed to examine the *in vitro* antimicrobial activities of the flowers and leaves methanolic and ethanolic extracts of *Chenopodium album* L. *Chenopodium album* Linn. flowers and leaves were collected from East Esfahan, Iran. The effects of methanolic and ethanolic extracts were tested against 4 bacterial strains by using disc,well-diffusion method. Results showed that flowers and leaves methanolic and ethanolic extracts of *C.album* don't have any activity against the selected bacterial strains. Our study has indicated that ,there are effective different factors on antimicrobial properties of plant extracts

Keywords**—***Chennopodium album*, antibacterial activity, extract

I. INTRODUCTION

MEDICINAL herbs represents one of the important fields of traditional medicine all over the world. Over the past 20 years, there has been an increased concern in the research of natural materials as sources of new antibacterial factors. Various extracts from traditional medicinal plants have been experimented to determine the source of the therapeutic effects [1]. new compounds with the property to act against multi-resistant bacteria [2]. Some medicinal plants used in traditional Iran medicine are effective in treating different ailments caused by bacterial and oxidative stress. New antioxidants such as plant phenolic compounds are sought for general health maintenance [3]. The term phenolics covers a very large and diverse group of chemical compounds.These compounds classified into groups comprise:flavonoids,lignins,tannins etc.[4]. Flavonoids are a large group of polyphenolic compounds that occur commonly in plants.In addition,flavonoid-based herbal medicines are available in different countries as anti-inflammatory,antispasmodic,antiallergic,antibacterial and antifungal remedies [5].

Chenopodium album L. (lamb's quarters, fathen, pigweed, white goosefoot) is one of the most important genera of the Chenopodiaceae family. This plant is widely found in different parts of Iran. *C.album* is a serious weed and does a salt-tolerant species inhabit semi-arid and light-saline environments in China [6].

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Thus. this is an annual weed of cultivated fields, especially on rich soils and old manure heaps [7]. Methanol extract of *C.album* leaves exhibited maximum antibreast cancer activity. This plant is a wild neglected herb which has various pharmacological properties such as antiviral, antifungal, anti-inflammatory, antiallergic, antiseptic and immunomodulating activity [8]. Methanol inflorescence extract of C. album exhibited highest antifungal activity resulting in up to 96% reduction in fungal biomass production [9]. *C.album* has antioxidant capacity, total phenol flavonoid glycosides (quercetin, rutin, kaempferol), thus, should be considered as a nutraceutical food and an alternative source for nutrients and free radical scavenging compounds [10]. Amjad 2009 have shown to *C.album* pollens had an allergenic effect, thus, During the skin prick test , the allergenic sensitivity was observed for *C.album* pollen grains , with an average wheal diameter of about 4 cm [11]. In previous studies showed that the electromagnetic fields reduction of pollen grains number and male sterility in *C.album* anthers , thus , the data presented suggest that prolonged exposures of plants to magnetic field may cause different biological effects at the cellular tissue and organ levels [12].

Thus, the principal aim of the present study was to screen ethanolic and methanolic extracts obtained from the *Chenopodium album* L. flowers and leaves in Iran for antibacterial activity against *Staphylococcus aureus*, *Bacillus cereus*, *Pseudomonas aeruginosa*, and *Escherichia coli*.

II. MATERIAL AND METHODS

Chenopodium album Linn. (Chenopodiaceae) flowers and leaves were collected locally in july 2011 from East Esfahan, Iran. The plant was identified in herbarium of Research Institute of Isfahan Forests and Rangelands. The plant materials were dried under shade .The dried flowers and leaves were homogenized to fine powder using electric blender and further subjected to extraction.

Powdered plant materials were extracted with different solvents and methods.

Respectively, 60g and 90g of leaves and flowers powder were extracted with 300 ml methanol (CH_3OH) by Soxhlet extraction for 48 hours[13], [14]. The residues were dried over night and then evaporated by using a rotary evaporator and freeze dryer. The dried extracts were stored at -20°C until used. The extracts were dissolved in 5%, 10%, 20%, aqueous dimethylsulfoxide (DMSO) [15], [16]. The extracts were diluted in 5%, 10%, 20%, dimethylsulphoxide (DMSO) at the concentrations of 100 ,200,400,600,800 mg/ml.

60g of leaves and flowers powder were extracted with 200 ml methanol (CH_3OH) by Soxhlet extraction for 6 hours [15,16]. The residue were dried over night and then evaporated by using a rotary evaporator and freeze dryer. The dried extracts were stored at -20°C until used. The extracts were dissolved in %100 dimethylsulfoxide (DMSO)[1], [17]. The extracts were diluted in %100 dimethylsulphoxide (DMSO) at the concentration of 650 mg/ml.

In other method, Plant extracts were prepared by cold percolation method. 90 g of dried powder was soaked in 300 ml methanol (CH_3OH) and ethanol for 1 week with intermittent shaking [1], [18] - [20]. The residues were dried until a constant dry weight of each extract was obtained. The residues were dissolved in 5%, 10%, 20%, aqueous dimethylsulfoxide (DMSO). The extracts were diluted in 5%, 10%, 20%, dimethylsulphoxide (DMSO) at the concentrations of 100 ,200,400,600,800 mg/ml.

Microorganisms were obtained from the Institute of Scientific and Industrial Researchs, Iran. Two strains of gram-negative bacteria *Escherichia coli* (ATCC 25922), *Pseudomonas aeruginosa* (ATCC 27853), and two strains of gram-positive bacteria *Bacillus cereus*(ATCC 1274) and *Staphylococcus aureus* (ATCC 25923) were used. The cultures of bacteria were maintained in their appropriate agar slants at 4°C throughout the study and used as stock cultures.

For the study of antibacterial activity, total count of isolated strains were standardized to equivalent a 0.5 MacFarland Nephelometer standard (1×10^8 cfu/ml) by Mueller-Hinton broth and then diluted as 1:10. Antibacterial activity was studied by the agar well diffusion method [5]. Mueller Hinton agar was used as the bacteriological medium. The Mueller Hinton agar was melted and cooled to 48 – 50ºC and a standardized inoculum (1.5×10^8 CFU/mL, 0.5 McFarland) was then added aseptically to the molten agar and poured into sterile Petri dishes to give a solid plate.

The disc diffusion and agar well diffusion methods were used to screen the antimicrobial activity. Wells were prepared in the seeded agar plates. The test compound (50 µl) was introduced in the well (6 mm) and (30µl) was introduced in the disc. The plates were incubated overnight at 37ºC. The antimicrobial spectrum of the extract was determined for the bacterial species in terms of zone sizes around each well and disc. The diameters of zone of inhibition produced by the agent were compared with those produced by the commercial control antibiotics, ciproflocxacin (30 µl). The experiment was performed three times to minimize the error and the mean values are presented.

III. Results

The different methods of extraction were investigated with various concentrations for antibacterial effects against four strains of bacteria using the disc diffusion and agar well diffusion method. But, flowers and leaves methanolic and ethanolic extracts of *Chenopodium album* L. don't have any activity against the selected bacterial strains [*Escherichia coli* (ATCC 25922), *Pseudomonas aeruginosa* (ATCC 27853), *Bacillus cereus* (ATCC 1274) and *Staphylococcus aureus* (ATCC25923)].

IV. Discussion

Plant extracts have been used for many thousands of years in food preservation and pharmaceuticals. It is necessary to survey those plants theoretically which have been used in traditional medicine to modify the quality of healthcare. The plant extracts are potential sources of modern antimicrobial compounds especially against bacterial pathogens. *In vitro* studies in this work showed that the extacts have Variable effects [21].

In our study, flowers and leaves methanolic and ethanolic extracts of *Chenopodium album* L. do not have any activity against the selected bacterial strains. A previous study in China has shown that leaves %95 ethanolic extract of *Chenopodium album* have inhibition effect on the *Escherichia coli* and *Staphylococcus aureus* [22]. The results of Adedapo et al. of South Africa showed that the leaves methanolic extract of *Chenopodium album* do not have any activity against the *Escherichia coli*, *Pseudomonas aeruginosa* and *Staphylococcus aureus* ,while,this methanolic extract showed antibacterial activity against *Bacillus cereus* [18].Thus, Yasmin et al. of Banglades reported that the carbon tetrachloride soluble fraction, chloroform soluble fraction and petroleum ether soluble fraction of methanolic extract of *Chenopodium album* demonstrated antibacterial activity against *Bacillus cereus*, but do not have any activity against the *Escherichia coli*, *Pseudomonas aeruginosa* and *Staphylococcus aureus* [19]. Several studies have shown that *Chenopodium album* L. leaves have class of phytoconstituents includes non-polar lipid, phenols, l ignins, alkaloids, flavonoids, glycosides and saponins [23].

There are reports showing that alkaloids and flavonoids are the responsible compounds for the antibacterial activities in higher plants . In addition, secondary metabolites such as tannins and other compounds of phenolic nature are also classified as active antimicrobial compounds [16]. Thus, Chludil et al. studies have shown that antioxidant capacity, total phenolic content and flavonoid glycosides profile of *Chenopodium album* were varied in intensively cultivated (IC) and nondisturbed (ND) soils and antioxidant activity relationships were established.Three known major flavonoid were isolated of *Chenopodium album* comprise: quercetin; rutin; kaempferol [10].

Although there have been rather few studies into the mechanisms basic flavonoid antibacterial activity, information from published writing indicates that different compounds within this class of phytochemicals may target different components and functions of the bacterial cell [24]. However, it may be that exclusive antibacterial flavonoids have multiple cellular targets, Instead of one specific site of action. Alternatively, these common structural features may simply be necessary for flavonoids to gain presence to or uptake into the bacterial cell. It may be that flavonoids are not killing bacterial cells but singly inducing the

formation of bacterial aggregates and thereby reducing the number of CFUs in viable counts [24]. However, there are different antibacterial mechanisms of action of various flavonoids: 1) Inhibition of nucleic acid synthesis; 2) Inhibition of cytoplasmic membrane function; 3) Inhibition of energy metabolism [23]. So, Comparison of the data obtained in this study with previously published results is problematic. Our interpretation of these results is include:

First, the composition of extracts is known to vary according to local climatic and environmental conditions [10], [23] .

Secondly, the method used to evaluation antimicrobial activity, and the choice of test organisms, varies between publications . The results obtained by different methods may differ as many factors vary between assays. These include : volume of broth or agar, type of broth or agar, size of wells , size of paper disks, strains of a particular bacterial species used, incubation period and microbial growth [22], [23].

Tertiary, the hydrophobic nature of most plant extracts prevents the uniform diffusion of these substances through the agar medium [23]. It will remain necessary to consider carefully additional variables such as the solvent used to solve test flavonoids. It has already been shown that sedimentation occurs when selected flavonoids are dissolved in organic solvents and diluted with neutral polar solutions. Sedimentation of flavonoids in a minimum inhibitory concentration assay is likely to cause diminished contact between bacterial cells and flavonoid molecules and may lead to false negative reports of antibacterial activity. Also, in false controlled experiments, flavonoid sedimentation could be misinterpreted as bacterial growth and further false negative results may be recorded as a consequence. Thus, if flavonoid salts are formed and these have increased or decreased potency compared with the parent structure, this may lead to false positive/negative reports of antibacterial activity [23].

Therefore, our study has indicated that perhaps, the compounds with antimicrobial effect such as phenols, flavonoids, glycosides and saponins with compounds without antimicrobial effect such as non-polar lipids have together drug antagonism effects.

V. Conclusion

The results of this study indicate the flowers and leaves methanolic and ethanolic extracts of *Chenopodium album* L. don't have any antimicrobial activity. However, more studies are needed to investigate the antimicrobial activity of different extracts with different solvents kind (chloroform, n-hexane,petroleum ether) in this plant and their potential for use in clinical situations.

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