



Phytochemical analysis of some Iraqi medicinal plants

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ABSTRACT

Fourteen medicinal plants *Clerodendron inerme*, *Chorisia speciosa*, *Eucalyptus melliodora*, *Dodonaea viscosa*, *Myosotis sp.*, *Olea europaea*, *Myrtus communis*, *Rhamnus sp.*, *Sesbania sesban*, *Hevea brasiliensis*, *Callistemon viminalis*, *Carissa macrocarpas*, *Thymus vulgaris* and *Peganum harmal* were selected from University of technology gardens from April – June 2013. Qualitative and quantitative analysis was conducted for aqueous and alcoholic extract. Total protein, carbohydrates, alkaloids, phenols, steroids and terpenes were determined in both extracts beside of pH value and extraction rates that investigated. From findings the pH value in watery extracts were higher than ethanol extracts in mean 6.4 ± 1.2 while in ethanol extracts reach to 5 ± 0.56 , the extraction ratio mean in the watery was 11.7 ± 3.7 while in ethanol was 16.7 ± 4.7 . Total protein in both type of extraction also varies in aqueous extraction the mean reach to 438 while in ethanol was 260.14 in significant difference at $P \leq 0.05$. from findings we can also note that ethanol 70% has high ability to release most component of examined plants while aqueous extraction have high ability to release the protein components of studied plants.

Keywords: phytochemical, medicinal plants, Extraction ratio, alkaloids, University of Technology.



INTRODUCTION

The University of Technology Gardens accounted about 30% of total area and the plants cultivated or grow naturally, both types of plants take their share of attention by irrigation and fertilization during agricultural season. More than 200 species belong to 18 plant family grow within the gardens of UT some of them are known as medicinal plants like *Rhamnus*, *Myrtus*, *Nerium oleander* and *catharanthus vinca*, but many are unknown. Plants are capable of synthesizing a wide variety of low-molecular-weight organic compounds called secondary metabolites, usually with unique and complex structures. Presently 100,000 such compounds have been isolated from higher plants [1]. Numerous plant secondary metabolites possess interesting biological activities and find applications, such as pharmaceuticals, insecticides, dyes, flavors, and fragrances. Many metabolites have been found to protect plants against viruses, bacteria, fungi, and most importantly against herbivores. Many secondary metabolites such as cyanogenic glycosides, glucosinolates, terpenes, saponins, tannins, anthraquinones, and polyacetylenes also act as allelochemicals,

influencing the growth and development of neighboring plants [2]. For example, monoterpene limonene has shown deterrent and insecticide properties and carvone is used as sprouting inhibitors [3, 4]. Although it has become clear in the last decade that jasmonic acid (JA) is a key regulator in the development, physiology, and defense of plants [5]. There is also strong evidence supporting a central role of JA in plant defense [6-8].

Alkaloid-containing plants constitute an extremely varied group both taxonomically and chemically group, they are basic, they contain one or more nitrogen atoms (usually in a heterocyclic ring) and they usually have a marked physiological action on man or other animals. In the plant, alkaloids may exist in the Free State, as salts or as amine or alkaloid N -oxides. For examples Colchicine from *Colchicum spp.* and related genera (*Liliaceae*), taxol (a modified diterpene pseudo alkaloid) *Taxus brevifolia* (*Taxaceae*). Other major plants derivative groups are glycosides like *digitoxin* from *Digitalis purpurea* that have effects on myocardial contraction. In the present work, qualitative and quantitative phytochemical analysis were carried

out in Fourteen evergreen plant samples were collected during flowering time (April to June) to analyze the chemical compounds for aqueous and alcoholic extracts (*Clerodendron inerme*, *Chorisia speciosa*, *Eucalyptus melliodora*, *Dodonaea viscosa*, *Myosotis sp.*, *Olea europaea*, *Myrtus communis*, *Rhamnus sp.*, *Sesbania sesban*, *Hevea brasiliensis*, *Callistemon viminalis*, *Carissa macrocarpas*, *Thymus vulgaris* and *Peganum harmal*).

MATERIALS AND METHODS

Site Description and plant Samples Collection:

Plant sample collected from University of Technology gardens, Baghdad, Iraq, from April to June 2013. The annual temperature averages 31.2°C and the annual rainfall averages 67.5 mm while annual average of relative humidity 42%. Fourteen evergreen plant samples (leaves) were collected for phytochemical analyzing, all samples putted in polyethylene clear bags capacity (1 and 2) Kg and transported to the lab within one hour, all plants were rinsed by running tap water and distilled water, placed on shelves in the shade with continuous air stream to dry by air during two weeks, all samples pulverized using mechanical blender and sieved by 2mm mesh kept in dark container with proper labeling.

Preparation of plant extracts:

Aqueous extraction: Ten grams of fine powdered plant material put in 250 ml conical flask with 100 ml of DDW, covered by aluminum foil and left for 24 hours at room temperature, cold water extract was filtered by Wattman filter paper NO.1. All extracts dried by incubator at 48°C for 24 hrs and weighed to determined ratio of extraction. Dry extracts were collected and kept in dark dry bottle at 4°C until use it.

Ethanol extraction: Ten grams of powdered plant material dissolved in 100 ml of 70% ethanol alcohol, covered and left for 24 hrs. at room temperature, infusion was filtered by Wattman filter paper No.1. Dried by incubator at 48°C for 24 hrs and weighed to determined ratio of extraction, dry extracts were collected and kept in dark dry bottle at 4°C until use it.

Qualitative phytochemical analysis: The extract was tested for the presence of bioactive compounds by using following standard methods [9-11].

Carbohydrates test: Carbohydrates were determined in the crud extracts of all plant materials by following Benedict's test by adding equal volume of plant extract to Benedict's reagent with boiling, a reddish brown precipitate refer to presence of carbohydrates.

Protein determination assay: Total protein was determined by following Biuret method according to [12].

Alkaloid determination assay: Alkaloid presence was examined by using Marquis reagent , black reddish colors indicate to positive result.

Phenols determination assay: Phenols are natural compounds present in the plants; these compounds react with Fe (III) (ferric chloride) to form purple color complex, evidence positive result.

Liebermann–Burchard test for steroids: 200 mg of powder sample was dissolved in 2 ml of acetic acid separately; solutions were cooled followed by the addition of few drops of conc. H₂SO₄. Color development from violet to blue or bluish-green was taken as positive test steroidal ring

Terpenoids assay: 100 mg of powdered sample was dissolved in 2ml of chloroform and evaporated to dryness, then, 2ml of concentrated H₂SO₄ was added and heated for about 2 minutes. A grayish color indicated the presence of terpenoids.

pH assay: 2.5 ml of crude extract was examined by pH meter to determine the pH value.

RESULTS AND DISCUSSIONS

Varieties in pH value and extraction ratio of fourteen plants were summarized in table (1), the results show that aqueous extraction gave value of pH higher than alcoholic extraction in mean (6.4±1.2 and 5±0.56) respectively, this means diversity in the active components that extracted from examined plants and ER of each plants, from table (1) that ethanol lead to high ER in mean 16.7±4.7 and 11.7±3.7 for aqueous, from findings that *Eucalyptus melliodora* has less pH value in watery extraction 4.6 associated with high ER 17.2%, while plant no.14 gave high ER 21.2% in alcohol. It should be noted that essential oil of *Eucalyptus* used in medicine, is obtained by aqueous distillation of the fresh leaves consists chiefly of a terpene and a cymene. *Eucalyptus* oil is used as a stimulant and antiseptic gargle, locally applied; it impairs sensibility and increases cardiac action [13].

Table (1): PH value and extraction ratio of fourteen medicinal plants

plant	Plant family	Aqueous extraction		Alcoholic extraction		
		PH	Extraction ratio%	PH	Extraction ratio%	
1	<i>Clerodendron inerme</i>	<i>Lamiaceae</i>	6.7	15.8	4.9	12.5
2	<i>Chorisia speciosa</i>	<i>Bombacaceae</i>	7.2	12.5	4.9	15.7
3	<i>Eucalyptus melliodora</i>	Myrtaceae	4.6	17.2	4.5	18.5
4	<i>Dodonaea viscosa</i>	<i>Sapindaceae</i>	5.8	4.7	4.8	27
5	<i>Myosotis sp.</i>	<i>Boraginaceae</i>	8.4	11.4	5	7
6	<i>Olea europaea</i>	<i>Oleaceae</i>	5.6	8.5	4.8	15.7
7	<i>Myrtus communis</i>	Myrtaceae	4.8	14.9	4.3	12.5
8	<i>Rhamnus sp.</i>	<i>Rhamnaceae</i>	7	10.4	5.1	19
9	<i>Sesbania sesban</i>	<i>Fabaceae</i>	7.5	6.9	5.1	13.6
10	<i>Hevea brasiliensis</i>	<i>Euphorbiaceae</i>	6.6	10.2	5.5	18.7
11	<i>Callistemon viminalis</i>	Myrtaceae	5.3	7.7	5.2	17.6
12	<i>Thymus Vulgaris</i>	<i>Lamiaceae</i>	8.4	13.1	6.8	21.9
13	<i>Carissa macrocarpa</i>	<i>Apocynaceae</i>	5.5	15.7	4.9	14
14	<i>Peganum harmal</i>	Apocynaceae	7.5	15.4	4.8	21.2
Mean ± SD			6.4±1.2	11.7±3.7	5±0.56	16.7±4.7

Phytochemical analysis of fourteen examined plants were summarized in table 2 and 3 which show that examined plants gave positive results of most medical components like carbohydrates, total proteins, alkaloids, phenols, steroids and terpenoides in the aqueous extraction. Carbohydrates and steroids appeared in all plant samples, while alkaloids appeared in five plants and phenols appeared in eight plants and

terpenoides were present in ten samples of plants and should be noted that aqueous extractions have high concentration of total proteins ranged from 162 µg/ml in *C. viminalis* to 750 µg/ml in both *C. inerme* and *C. macrocarpa* in mean 438 µg/ml, while total protein in alcohol extraction ranged from 75 µg/ml in *Myosotis sp.* to 750 µg/ml in *C. macrocarpa* in mean 260.14 µg/ml.

Table (2): Phytochemical analyses of aqueous extraction for fourteen medicinal plants

Plant	Aqueous extraction						
	Carbohydrates	Protein concentration µg/ml	Alkaloids	Phenols	Steroid	Terpenoides	
1	<i>C. inerme</i>	+	750	+	-	+	+
2	<i>C. speciosa</i>	+	550	-	+	+	+
3	<i>E. melliodora</i>	+	300	+	+	+	+
4	<i>D. viscosa</i>	+	600	-	-	+	+
5	<i>Myosotis sp.</i>	+	425	-	-	+	-
6	<i>O. europaea</i>	+	175	-	+	+	-
7	<i>M. communis</i>	+	425	+	+	+	+
8	<i>Rhamnus sp.</i>	+	250	-	+	+	-
9	<i>S. sesban</i>	+	500	-	-	+	+
10	<i>H. brasiliensis</i>	+	725	+	-	+	-
11	<i>C. viminalis</i>	+	162	-	+	+	+
12	<i>Thymus vulgaris</i>	+	550	-	-	+	+
13	<i>C. macrocarpa</i>	+	750	+	+	+	+
14	<i>Peganum harmal</i>	+	600	-	+	+	+
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Table (3): Phytochemical analyses of alcoholic extraction for fourteen medicinal plants

plants		alcoholic extraction					
		Carbohydrates	Protein concentration µg/ml	Alkaloids	Phenols	Steroid	Terpens
1	<i>C. inerme</i>	+	150	+	-	+	+
2	<i>C. speciosa</i>	+	200	-	+	+	+
3	<i>E. melliodora</i>	+	212	+	+	+	+
4	<i>D. viscosa</i>	+	275	-	-	+	+
5	<i>Myosotis sp.</i>	+	75	-	-	+	-
6	<i>O. europaea</i>	+	400	-	+	+	-
7	<i>M. communis</i>	+	250	+	+	+	+
8	<i>Rhamnus sp.</i>	+	180	-	+	+	-
9	<i>S. sesban</i>	+	200	-	-	+	+
10	<i>H. brasiliensis</i>	+	400	+	-	+	-
11	<i>C. viminalis</i>	+	250	-	+	+	+
12	<i>Thymus vulgaris</i>	+	125	-	-	+	+
13	<i>C. macrocarpa</i>	+	750	+	+	+	+
14	<i>Peganum harmal</i>	+	175	-	+	+	+
Mean ± SD			260.14				

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