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CHEMICAL STUDIES ON PURSLANE AND WHITE MULBERRY LEAVES

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ABSTERACT

Purslane and white mulberry leaves were analyzed for their proximate composition, minerals, fatty acids, phenolic compounds, flavonoids, and amino acids content to evaluate their importance in human nutrition. The results showed that purslane and white mulberry leaves contain appreciable amount of essential nutrients. Moisture, carbohydrate, crude protein, ether extract, crude fiber and ash content in purslane were 4.93, 52.86, 23, 6.34, 10 and 17.80, respectively, and 12.02, 59.75, 21, 4.83, 15 and 14.42 for mulberry leaves(on dry weight basis).also high concentration of macro minerals Ca were(1.2 and 3.6%), K (5.6 and 1.16 %) and Na (2 and 0.79%) , high concentration of micro minerals' Fe were (0.255 and 0.260 %) and Zn (0.030 and 0.022 %) ,respectively . The dietary purslane oil consisted mainly of oleic acid, linoleic acid, plamitic acid, arahidic acid and capric. On the other hand fatty acids composition of white mulberry leaves consisted mainly of arahidic, plamitic, linoleic, and capric and oleic. High concentration of phenolic compounds and flavonoids were recorded. White mulberry leaves contain essential amino acids lysine and non-essential amino acids glutamic and aspartic. From this study, it could be concluded that consumption of purslane and white mulberry leaves in different quantities could provide a reasonable daily recommended amount of essential nutrients for maintenance of healthy life and normal body functions.

INTRODUCTION

The plant purslane, in Arabic *'Rejlah', (Portulaca oleraceae* L.) occurs in the Arabian Peninsula and adjacent areas including, United Arab Emirates and Oman. Purslane is also consumed as a vegetable in some provinces of China (Chan *et al*., 2000; Hu, *et al.,* 2003).

It is also used as an antibacterial and anti-viral agent, as well as for the treatment of viral hepatitis and diabetes management in China (Meng & Wu, 2008).Purslane is reported to be rich in *á* linolenic acid and *ß*-carotene and used as a health food for patients with cardiovascular diseases (Liu *et al.*, 2000). It contains several types of vitamins and minerals (Mohammad, *et al.,* 2004), fatty acids (Xin *et al.*, 2008), and amino acids glutathione, glutamic acid, and aspartic acid. Other constituents include a mucilage composed of a neutral fraction with structure determined, dopamine and dopa, coumarins, flavonoids, alkaloids, saponins, and anthocyanin (Peksel, *et al.,* 2006). Recently, Hussein &Abdel-Gawad (2010) studied the potential hepato-protective effect of ethanolic and aqueous extracts of air-dried leaves of purslane against paracetamol-induced hepato-toxicity and showed that the ethanolic and aqueous extracts of purslane leaves can generate antioxidants. The effect was more pronounced in ethanolic extract compared to aqueous extract. Large amounts of phenolic compounds (coumarins, flavonoids,

alkaloids, and saponins) in ethanolic extract may contribute towards the antioxidant properties (Sakai *et al.,* 1996).

Purslane has been described as a 'power Food' of the future because of its high nutritive and anti-oxidant properties (Al-Howiriny, 2008).
In continuation of my research on Therapeutic evaluation of plants of medical importance (Hussein & Abdel-Gawad, 2010).

Medicinal plants play an important role in Indian ayurvedic system of medicine and many active compounds were isolated from the plants which used as medicines. These active compounds are chemically in nature which is known as phytochemical or secondary plant products. Mulberry plant is one of conventional herbs which are used in medicine from centuries ago due to its chemical composition and Pharmacological functions. Most of the parts of mulberry plants are used as medicine in Chinese and Indian medicine.

According to Singh, Amritpal. (2008) active principles which are isolated from medicinal plants may influence health and inhibited the bacterial or fungal pathogens. Mulberry is a fast-growing deciduous plant that grows under various conditions i.e., tropical, subtropical and temperate (Srivastava *et al.,* 2003).

Mulberry belongs to the family of moraceae and genus morus usually cultivated to feed silkworm for manufacturing of silk. *Morus alba* (white mulberry) and *Morus indica* (Indian mulberry) are the most popular species of mulberry .According to Zou and Chen (2003), mulberry leaves contain N-containing sugars, rutin, quercetin, volatile oil, amino acids, vitamins and microelements, which have so many pharmacological activies such as reducing blood glucose, antihyperlipidemia, hypertensive, bacteriostasis and antivirus. Andallu *et al.,* (2001) and Andallu and Varadacharyulu (2002) have reported many different medicinal properties of mulberry leaves. According to (Maria *et al.,* 2008) root extract of mulberry plants is also having antimicrobial activity. Bio active compounds in different species of mulberry can enhance life (Venkatesh and Chauhan, 2008). Different pharmaceutical properties of mulberry plants are reviewed by Singhal *et al.,* (2010). They found that many biochemical compounds such as Moranoline, Albafuran, Albanol, Morusin, Kuwanol, Calystegin and Hydroxymoricin are isolated from mulberry plants which play an important role in pharmaceutical industry. The medicinal properties of mulberry plants are identified for their profitable medicinal value and therefore attracted the attention of the pharmaceutical industry. The main objective of present review is to discuss the active principles of Mulberry plants relating to its pharmacokinetic activity to human diseases.

MATERIALS AND METHODS

1. Purslane and White Mulberry Leaves:

They were obtained from the local performer field of Kafrelsheikh city, Egypt. Plants were cleaned from dust and from forgein matters, washed with tap water and then plants were dried in a hot air oven maintained at 55ºC, milled and kept in polyethylene bags until used.

Methods:-

Methods of analysis:-

Determination of moisture:-

Moisture, protein, ether extract, ash, crude fibers were determined according to A. O. A.C (2000). Total carbohydrates content was calculated by difference. Mineral contents were determined according to the method of A.O.A.C. (2000) using atomic absorption spectrophotometer Perken Elmer Model 2180.

Energy value:-

The energy value was calculated according to James (1995).

Energy value = (% carbohydrate × 4.1) + (% protein × 4.1) + (% fat× 9.1).

Purslane and white mulberry leaves oil extraction.

Purslane and mulberry leaves powder were extracted with hexane (40-60ºC) using soxhelt extractor for 6-8 hours. The solvent was removed by rotary vacuum evaporation and the oil was collected. The percentage yield was calculated on a dry weight basis (Papageorgiou *et al.,* 1996).

Fatty acids composition of purslane and white mulberry leaves powder:-

Fatty acids composition of Purslane and white mulberry leaves powder oil were determined in Faculty of Agriculture, Alexanderia University, using gas chromatography(GC Modle,Shimadzu-8A,equipped with a FID Chromo Q, Detector temperature 270 ºC,H2 flow rate 75ml/min, Sensitivity 16x10, Column temperature 150-180 ºC at rate 2 c/min,N2 flow rate 20ml/min,Air flow rate 0.5ml/min and Start speed 2.5 mm/min according to the method described by Radwan(1978).

Determination of amino acids.

Amino acids content of white mulberry leaves was analyzed in National Research Center, Giza, Egypt. , as follows:

Samples were subjected to acid hydrolysis using 6N Hcl.. The resulted amino acids were analyzed using amino acid analyzer. (LC 3000 amino acid analyzer, High performance system, a product of LC biochrom Eppdrop, Germany). Flow rate 0.2 ml/min, Pressure of buffer form 0 to 2 bars, Pressure of reagent to 0-150 bar and reaction temperature 123 ºC. Amino acids were analyzed according to

A. O. A. C. (2000).

Determination of polyphenols:

Phenolic compounds from purslane and white mulberry leaves powder were extracted according to the methods of Rodriguez de Sotlillo , *et al*., (1994) with methanol 95% under cooling (4 ºC) as follows : five grams of purslane and mulberry leaves (powder)were homogenized for 4 min. (in adulrang Osterizer belender) with 29 ml of cold methanol . The resulting slurry was centrifuged (Hettich, mikro rapid /K type 1306) at 3000 × 9 for 10 min. at 5 ºC. The supernatant liquid was filtered through whatman no. 4, filter paper the filtrate was collected for quantitative analysis.

Phenolic compounds of methanolic extracts from purslane and mulberry leaves powder were determined using HPLC Hewllet Packared (series 1050) equipped with auto sampling injector, solvent degasser, ultra violet (UV) detector set at 210 nm and quarter HP pump (series1050). The

column temperature was mainted at 35ºC .An isocratic separation was carried out with 0.01N H2SO4 as a mobile phase at flow rate of 1 ml/min. The phenolic compounds standard from Fluka Co. Were dissolved in a mobile phase and inject into HPLC. Retention time and peak area were used to calculate phenolic compounds concentration by the data analysis of Hewllet Packared software according to the method described by Anderson and Pederson, (1983).

RESULTS AND DISCUSSION

The chemical composition of purslane and white mulberry leaves (g/100g on dry weight basis).

The chemical compositions of purslane and white mulberry leaves were studied. The results found in Table (1) reveal that purslane and white mulberry leaves can be considered as rich source for crude protein since it contained 23 and 21 %, respectively.

Table (1): The chemical composition of purslane and white mulberry leaves (g/100g on dry weight basis).

Component	purslane	white mulberry leaves
Moisture	4.93	12.02
Ether extract	6.34	4.83
Crude protein	23.00	21.00
Ash	17.80	14.42
Total carbohydrates ***	52.86	59.75
Crude fibers	10.00	15.00
Available carbohydrates**	42.86	44.75
Energy value cal/100gm	327.72	313.53

*****Total carbohydrates werer calculated by difference**
****Available carbohydrates = Total carbohydrates – Crude fibers.**

In addition, it could be noticed that moisture content was 4.93 and 12.02% in purslane and white mulberry leaves. Results in Table (1) also showed that the total carbohydrates were higher in white mulberry leaves 59.75%, comparing with purslane 52.86%. On the other hand crude fibers is higher in white mulberry leaves (15 %), comparing with purslane (10%). while mean ether extract were 6.34 and 4.83 %, finally ash was recorded 17.80 % in purslane and 14.42 % in white mulberry leaves.

With respect to the energy value of purslane it was slightly higher than that of white mulberry leaves. The obtained results agree with

El-Zaawely (1999) who stated that chemical composition content of purslane collected from Tanta, El-Hamool and El- Borollus districts. He found that moisture content ranged from 89.83 to 91.89 %. Crude fibers ranged from 5.89 to 11.58 %. Carbohydrates 34.42 to 42.94 %. Protein 23.52 to 29.04 %. Fat 3.48 to 5.08 % .ash 19.88 to 23.01 % .On the other hand, the obtained results agree with Andallu *et al.,*(2003) who found that, protein, fat, ash, crude fiber and total carbohydrates content were value were

23.10%,7.92%,15.43%,13.85% and 39.70%.respectively, for white mulberry leaves.

Minerals content of purslane and white mulberry leaves.

Data listed in Table (2) reveals that purslane and white mulberry leaves appear to be poor in phosphors (0.32 and 0.05%), rich in potassium(5.6 and 1.16%) also calcium higher in white mulberry leaves comparing purslane and contain considerable amounts of magnesium (0.53 and 0.55%), respectively. On the other hand five trace elements, iron, zinc, manganese, chromium and copper were detected for their contents in the purslane and white mulberry leaves. The results are listed in Table (2). Data reveals that, the purslane and white mulberry leaves contained higher concentration (0.225 and 0.260%) of iron and contain considerable amounts of zinc, manganese and chromium. The obtained results agree with El-Zaawely (1999) who stated that content of purslane collected from Tanta, El-Hamool and El- Borollus districts. He found that nitrogen content ranged from (3.76 to 4.65 %), P (0.36 to 0.46 %), K (5.19 to 6.38 %), Ca (0.38 to 0.52 %), Fe (0.35 to 0.55 %) and Na (1.45 to 2.27 %) (Dry weight basis). On the other hand, the obtained results agree with Andallu *et al.,* (2003) who found that mulberry leaves contain calcium, phosphorus, magnesium and iron.

Table (2): Minerals content of purslane and white mulberry leaves (g/100g on dry weight basis)

Major elements					
Samples	**Calcium (Ca)**	**Sodium (Na)**	**Potassium (K)**	**Magnesium (Mg)**	**Phosphors (p)**
Purslane	**1.2**	**2**	**5.6**	**0.53**	**0.32**
White mulberry leaves	**3.6**	**0.79**	**1.16**	**0.55**	**0.05**
Trace elements					
Samples	**Iron (Fe)**	**Zinc (Zn)**	**Manganese (Mn)**	**Chromium (Cr)**	**Copper (Cu)**
Purslane	**0.255**	**0.030**	**0.024**	**0.010**	**Trace**
White mulberry leaves	**0.260**	**0.022**	**0.024**	**0.007**	**Trace**

Phenolic compounds of purslane and white mulberry leaves

The results in Table (3) cleared that purslane powder extract had high amount of hsperidin, catchin, vanillic and caffeic acid (735, 97, 45 and 27 mg/100g) respectively, followed by chlorogenic, catechol, salicylic, procatchnic, hesperetin and chrisin. On the other hand, the data cleared that, catechin was the main phenolic compound in white mulberry leaves powder extract, catechin representing about (151mg/100g). Followed by salicylic acid 59 (mg/100g). Gallic acid was the third major phenolic compound 42(mg/100g), followed by, OH benzoic, ferulic, syringic, chrisin and hesperetin. Hussein (2010) explained that purslane extract in the form of ethanolic formulation is rich in polyphenols, flavonoids, anthocyanin, and melatonin.

Table (3): Phenolic compounds of Purslane and White mulberry leaves using (HPLC).

Identification phenolic compounds	Purslane mg/100g	White mulberry leaves mg/100g
Catechin	97	151
Protocatchnic	6	Trace
Chlorogenic	9	Trace
Catechol	6	Trace
Vanillic	45	Trace
Caffeic	27	Trace
Hesperidin	735	Trace
Salicylic	6	59
Hesperetin	2	Trace
Chrisin	1	7
Gallic	Trace	42
OH Benzoic	Trace	24
Syringic	Trace	16
Ferulic	Trace	22

Fatty acids composition of purslane and white mulberry leaves

Fatty acids composition of purslane and white mulberry leaves were given in Table (4).The dietary purslane oil consisted mainly of oleic acid ѡ-9(22.80%),linoleic acid ѡ-6(19.66%), plamitic acid (14.05%), arahidic acid (12.89%), capric (12.67%) with small concentrations of saturated fatty acid such as myristic (9.91%), stearic (4.41%), heptadecanoic (2.09%) and plamiticoleic (1.51%).On the other hand fatty acid composition of white mulberry leaves consisted mainly of arahidic (33.83%), plamitic (18.50%), linoleic ѡ -6(15.65%), capric (11.05%) and oleic ѡ-9 (7.17%) with small concentrations of saturated fatty acid such as myristic (6.16%), stearic (4.49%), heptadecanoic (1.03%), myristicoleic(0.60%), lauric (0.27%), plamiticoleic (0.21%) and heptadecenoic (0.17%).

Table (4): Fatty acids content of purslane and white mulberry leaves (%g total fatty acids).

Fatty acids	Symbol	Purslane	White mulberry leaves
Capric	**C10:0**	**12.67**	**11.05**
Lauric	**C12:0**	**Trace**	**0.27**
Myristic	**C14:0**	**9.91**	**6.61**
Myristicoleic	**C14:0**	**Trace**	**0.60**
Plamitic	**C16:0**	**14.05**	**18.50**
Plamiticoleic	**C16:1**	**1.51**	**0.21**
Heptadecanoic	**C17:0**	**2.09**	**1.03**
Heptadecenoic	**C17:1**	**Trace**	**0.17**
Stearic	**C18:0**	**4.41**	**4.49**
Oleic ϕ9	**C18:1**	**22.80**	**7.17**
Linoleic ϕ6	**C18:2**	**19.66**	**15.65**
Arahidic	**C20:0**	**12.89**	**33.83**
Total saturated fatty acids	**%**	**56.02**	**75.25**
Total Unsaturated fatty acids	**%**	**43.97**	**23.77**

These results were in agreement with those reported by *Liu et al.,*(2000), who showed that major fatty acids for purslane were oleic acid ,linoleic acid , plamitic acid , arahidic acid , capric ,with small concentrations of saturated fatty acid such as myristic , stearic , and plamiticoleic .

Amino acids composition of white mulberry leaves.

The amino acids composition of white mulberry leaves was given in Table (5). The results showed that white mulberry leaves protein was considered a poor source of cystine 1% and methionine 1.74%. On the other hand, leucine, phenylalanine, lysine, valine, isoleucine and theronine were the predominant indispensable amino acids represented 8.66, 6.5, 6.30, 5.93, 4.79 and 4.56%, respectively. While tyrosine and histidine were 2.75 and 2.32% for indispensable amino acids. Dispensable amino acids contained glutamic and aspartic were the predominate of dispensable amino acids which reached 11.17 and 10.27% followed by arginine 6.04% while glycine, alanine, proline and serine represented 5.27, 5.17, 4.39 and 4.09%, respectively. These results agree with Yao *et al.,* (2000) who reported that protein concentration from mulberry leaves was characterized by higher levels of lysine for essential amino acids and glutamic acid followed by aspartic for non-essential amino acids.

Table (5): Amino acids composition of white mulberry leaves. (g / 100g protein).

Type	Amino acids	White mulberry leaves	FAO/WHO/UNU, 1985 Pattern.
Indispensable amino acids	Lysine	6.30	5.80
	Isoleucine	4.79	2.80
	Leucine	8.66	6.60
	Phenylalanine	6.5	6.30
	Tyrosine	2.75	
	Histidine	2.32	1.90
	Valine	5.93	3.50
	Threonine	4.56	3.40
	Methionine	1.74	2.50
	Cystine	1.00	
Dispensable amino acids	Aspartic	10.27	
	Glutamic	11.17	
	Serine	4.09	
	Proline	4.39	
	Glycine	5.27	
	Alanine	5.17	
	Arginine	6.04	

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جمال سعد الحديدي ، مسعد عبد العزيز ابو رية* ، مني محمود خليل*، فاتن يوسف ابراهيم* و عبد التواب سعد بركات****
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****معهد بحوث تكنولوجيا الأغذية – مركز البحوث الزراعية**

التركيب الكيماوي لكل من الرجلة و ورق التوت وكذلك المعادن والأحماض الدهنية والمركبات الفينولية والفلافونيدات تم تقديرها وذلك لأهميتها في تغذية الإنسان. واوضحت النتائج ان الرجلة وورق التوت يحتويان علي كميات مناسبة من المغذيات الضرورية وكانت نسبة الرطوبة والكربوهيدرات والبروتين الخام والمستخلص الإثيري والألياف الخام وكذلك الرماد في الرجلة (٤,٩٣ ، ٥٢,٨٦، ٢٣، ٦,٣٤، ١٠، ١٧,٨) علي التوالي وفي ورق التوت كانت (١٢,٠٢، ٥٩,٧٥، ٢١، ٤,٨٣، ١٥، ١٤,٤٢) علي التوالي، وكميات مرتفعة من المعادن الكبري مثل الكالسيوم والبوتاسيوم والصوديوم، وكميات مرتفعة من المعادن الصغري مثل الحديد والزنك. ووجد أن أهم الأحماض الدهنية السائدة في زيت الرجلة هي الأوليك واللينوليك والأرشيدك والبالميتك، بينما كانت الأحماض الدهنية السائدة في ورق التوت هي الأرشيدك والبالميتك واللينوليك. ووجد أن الرجلة و ورق التوت يحتويان علي المركبات الفينولية والفلافونيدات بتركيزات مرتفعة، واحتواء ورق التوت علي الأحماض الأمينية الأساسية منها الليسين وبعض الأحماض الامينيه غير الأساسية مثل الجلوتاميك والأسبارتيك. ومن هذه الدراسة نخلص إلي استهلاك الرجلة و ورق التوت بكميات مختلفة يزود الجسم بالكميات المسموح بها يوميا من المغذيات الضرورية.

قام بتحكيم البحث

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