Spectrophotometric analysis of some metals and phytochemical screening of *Digera muricata* (Leaves and stems)

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Abstract: The current research deals with estimation of the spectrophotometric analysis of some metals and phytochemical screening of the plant *Digera muricata*. The importance plant study arises from the fact that it has both medicinal and nutritional values. The plant were collected from Albaha mountains in Albaha area. The parts of the plant that were studied are stems and leaves. Spectrophotometric and proximate analysis showed that the percentage of moisture 7.24% and 7.12% for leaves and stems respectively, ash content 25.3% for leaves and 23.9% for the stems, while iron content for leaves and stems was 208.3 and 75.0mg/kg respectively, and for copper content both parts have the same amount which is 11.5mg/kg. Phtychemical analysis of the plant parts revealed the presence of alkaloids, flavonoids and saponin content in both plant parts while tannins present in the leaves but absent in the stems. The results obtained from this study can be used to standardize different medicinal plants. Also the bioactive compounds found in the plant under study may be a source of investigations of promising herbal medicines.

Keywords: Spectrophotometry, phytochemical, proximate compositions, *Digera muricata.*

INTRODUCTION

Digera muricata is a medicinal plant which belong to the family amaranthaeae and the life form is herbal. Its nativity is south west Asia (Aravindhan *et al*., 2014). Most of the plant parts of *Digera muricata* possess fundamental medicinal uses (Sharma *et al*., 2013), the flowers is used to treat liver disorders (Tiwari *et al*., 2014). It is useful in renal disorders in traditional medicine by the action of reactive radicals generated which lead to for the elimination of the CCl_4 -induced nephrotoxicity that is resposible for kidney diseases (Khan *et al*., 2009). The extract of the leaves of the plant was used against fungal diseases (Chandra *et al*., 2013). The extract of *Digera muricata* with petroleum ether was used against bacteria *Vibrio cholerae* which is cause cholera disease (Pratima *et al*., 2010). Nowadays, great attention has been focused to use biologically friendly plant derived materials for the protection and care of different human being diseases, because of the harmful effects of formulated drugs, so people are looking for naturally occurring compounds for treatment , which are less danger and effective (Shah *et al*., 2011). The overlapping importance of plants has both medicinal and nutritional values to promote health and respond to illness (Moeller *et al*., 2000). The intake of phytochemicals exist in traditional foods has direct benefits on the health of people (Palozza *et al*., 1992). The plant of *Digera muricata* treatment support the antioxidant activities, defence against CCl_4-induced toxicity and emphasis the fact that it may have therapeutic importance in free radical mediated diseases (Usmani *et al*., 2013). The roots of *Digera muricata* contain sugar and phenols, while the leaves contain proteins, starch and lipids. And it has antimicrobial activities against *Escherichia coli* and *Fusarium oxysporum* in the leaves extract. The leaves extract is used to treat kidney stone (Sharma *et al*., 2011).

In this work, we initiated this study in order to determine the phytochemical composition of *Digera muricata* (leaves and stems) includes tannins, alkaloids, flavonoids, saponins and some proximate compositions such as moisture content, ash value and some trace metals (Fe and Cu) which play a vital role in the metabolic processes and general health of human body. The study outcome is very necessary for investigating and obtaining the bioactive compounds which may serve as medicinal and nutritional values.

MATERIALS AND METHODS

Samples area and preparations

Leaves and stems of *Digera muricata* were collected from the mountains of a local area in Albaha area of Southwest of Saudi Arabia. The plant (*Digera muricata*) was identified as a common plant in Albaha area in Saudi Arabia. Leaves and stems were thoroughly cleaned with water to remove dust and unwanted particles. The two parts (leaves and stems) were separately cut in pieces, dried at 115ºC and ground in a porcelain mortar and pistol into powder before being used for analysis. The grinded samples were stored in containers at room temperature.

Moisture and ash content

Moisture and ash content of the samples (leaves and stems) of *Digera muricata* were determined by using the Official Method of the Association of Official Analytical Chemists (AOAC) (AOAC, 1990) as stated by Asibey-Berko (Asibey-Berko., 2009) (table 1).

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Tests for tannin, saponin, alkaloids and flavonoids

The queous extracts for each of the two samples (leaves and stems) of *Digera muricata* were prepared by adding 1g of the powdered samples in 70ml of distilled water and let them overnight, then were filtered. Chemical analyses were carried out on the extracts and on the powdered samples. The analysis for the presence of tannin in the tested samples was carried out using ferric chloride test described by Harbone (Harbone, 1973) as reported by Osagie (Osagie, 2011) by the formation of a very dark precipitate indicated the presence of tannin upon addition 2g of the powdered sample into 10mls of distilled water, then was shaken for 30 minutes and the filterate used as aqueous extract. 2mls of the extract was added into to 3mls of distilled water and shaken well a few drops of Ferric chloride solution was added to the mixture, as seen in table 2. For the presence of saponin in the tested samples was done using the Harbone (Harbone, 1973), as reported by Osagie (Osagie, 2011). By using the Froth test in which 2mls of the aqueous extracts were added to 6mls of distilled water in a test tube, the mixture was vigoursly shaken and the froth formed indicate the presence of saponins (table 2). The presence of alkaloids in samples was tested using the procedure described by Okwu (Okwu, 2005). By formation of reddish brown precipitate upon addition of 2g of sample to 5ml of 2% HCl on a steam bath and filtered with filter paper. 1ml of filtrate were added to 0.5ml of Wagner's reagent which was prepared by addition of 3g of potassium iodine to 2g of iodine and then dissolved in 20ml of distilled water and completed to 100ml with distilled water. The precipitate indicates the presence of alkaloids (table 2). The analysis for the presence of flavonoids in the samples was carried out using the test described by Osagie (Osagie, 2011). By the formation of a yellow colour upon addition of 2mls of the extract to a few drops of concentrated ammonia. The colouration formed shows the presence of flavonoids (table 2).

Elemental analysis

Some metals of the two samples of *Digera muricata* were estimated by using the official method of the Association of official Analytical Chemists AOAC (AOAC., 1990), upon ash drying of 1g of each dried sample separately with a porcelain crucible in a muffle furnace at 570ºC for 8 hours. The resulting ash was put in a desiccator and weighed. 10ml of diluted HNO_3 were added to the ash, and then completed to 100ml distilled water. The quantification was carried out using UV-VIS Spectrophotometer (1cm quartz cell, SP- 3000 Plus model, Optima, Tokyo, Japan) by using ammonium thiocayanate as a chromogenic reagent for Fe determination and dimethylglycoxime as a chromogenic reagent for Cu determination. Stock solution of 1000 ppm for each of iron (III) and copper were prepared, and the scan for the wavelength of maximum absorbance for each of the two elements was carried out (table 3). A series of 5 dilutions of Fe(III) (1.0, 5.0, 10.0, 15.0 and 20.0ppm and of copper (1.0, 5.0, 10.0, 15.0 and 20.0ppm) were prepared from the stock solutions separately and the absorbance at the wavelength of maximum absorbance were measured for both elements. Absorbance versus concentration were plotted (figs. 1 and 2). Unknown samples solutions were treated in the same way for each elements. The absorbance of unknown sample solutions were measured. These absorbance values were referred to their related concentrations in the calibration curves (tables 4 and 5).

RESULTS

The results of the phytochemical and spectrophotometric analysis of the plant *Digera muricata* were presented in tables 1-5 and figs. 1-2.

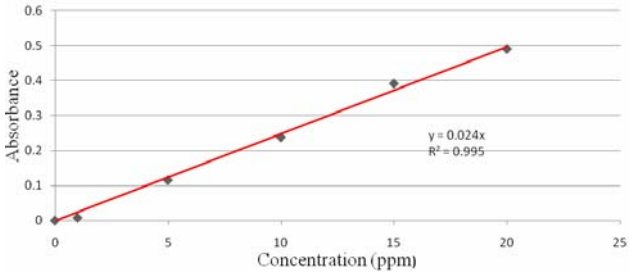

Fig. 1: Absorbance of solutions of Iron as a Function of Concentratios

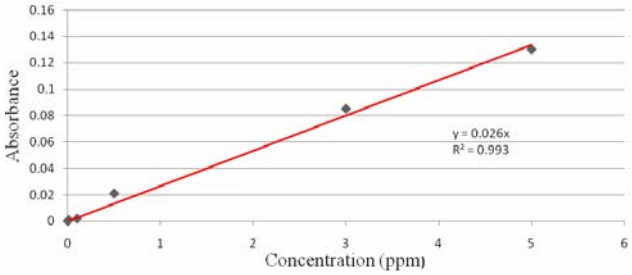

Fig. 2: Absorbance of solutions of Copper as a Function of Concentratios

DISCUSSION

The treatment of many diseases by herbs is well-known for many hundreds years ago. The plant *Digera muricata* is one of medicinal plant as well as its nutritional values. The chemical constituents of the plant can reveal by proximate and phytochemical analysis. *Digera muricata* has good values of moisture and organic constiuents which are making the plant as a good source of food. The plant (dry weight basis) was investigated to different quality analysis assessment and revealed moisture and ash content. The moisture content was found to be 7.24 and 7.12% for leaves and stems respectively and the ash content was found 25.3% for leaves and 23.9% for stems as shown in table 1.

Table 1: Proximate composition of *Digera muricata* (leaves and stems)

Parameters	*Digera muricata* (leaves) (%)	*Digera muricata* (stems) (%)
Moisture	7.24	7.12
Ash content	25.3	23.9

Table 2: Qualitative analysis of phytochemicals present in *Digera muricata* plant parts

Phytochemical	*Digera muricata* (leaves)	*Digera muricata* (stems)
Tannins	++	-
Saponin (Forth test)	++	+
Alkaloid	++	+
Flavonoids	++	+

Table 3: Wavelengths (λ max) of maximum absorption of Iron and Copper.

Element	λ max
Fe	370 nm
Cu	372 nm

Table 4: Absorbance of solution of *Digera muricata* (leaves and stems).

Parameters	*Digera muricata* (leaves)	*Digera muricata* (stems)
Fe (at 370 nm)	0.050	0.018
Cu (at 372 nm)	0.003	0.003

The phytochemical analysis of *Digera muricata* was carried out by applying the standard methods (Harbone, 1973, Osagie, 2011 and Okwu, 2005). The results showed the presence of variuos phytochemicals contituents like alkaloids, flavonoids, saponins and tannins which are presented in table 2. These exisiting bioactive compounds are very usefull in medicine uses for the treatment of several diseases. And also they can be a source of investigations of promising herbal medicines. Alkaloids and tannins have been shown to be active against bacterial diseases (Dhale *et al*., 2011 and Jeeshna *et al*., 2011). The saponins content may help in the liver functions, concerning the metabolite of cholesterol (Kabir *et al*., 2005). Flavonoids can be considered as antioxidants that may act as anti-inflammatory inhibitors (Houghton *et al*., 1999). As shown in table 2, the mentioned secondary metabiltes are present in higher levels in the leaves of *Digera muricata* as compared with the stems.

The metals like Fe and Cu were determined spectrophotometrically at the wavelengths of maximum absorption shown in table 3, using ammonium thiocayanate and dimethylglycoxime as a chromogenic reagent for Fe and Cu determination respectively. The calibration curves for both Fe and Cu by series of dilution of the standard solution of metals under study was constructed as in fig. 1 and 2. The absorbances of samples were measured and the content of metal were determined as shown in tables 4 and 5. The iron content in the leaves of *Digera muricata* is higher than that of the stems, while the copper content seems to be the same amount in both leaves and the stem of the plant. These metals are very necessary for biological, chemical and molecular levels of cell functions (Prashanth *et al*., 2015).

CONCLUSIONS

We can conclude that the current study revealed the presence of various biologically active compounds in the leaves and stems of *Digera muricata* which are served as antibactrial, antioxidand and untimicrobial activities. And also can be served as food source due to its nutritional qualities and proximate contents. This findings can help us to choose *Digera muricata* with greater quantity and quality of medicinally and nutrionally fundamental phytochemicals. *Digera muricata* (leaves and stems) have proven to be considered as a major source of iron and copper.. Recommendations that could be drawn from this investigation are that further studies should be carried out on the plant *Digera muricata* for the other chemical compounds and metals. Finding more about the useful effects of *Digera muricata* for health is of great importance in all over the world as it has a very little harm of toxicity and the ease of availability in all over the world.

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Table 5: Trace metals content of *Digera muricata* (leaves and stems)

Parameters	*Digera muricata* (leaves)		*Digera muricata* (stems)	
	(mg/L)	(mg/kg)	(mg/L)	(mg/kg)
Fe	2.083	208.3	0.750	75.0
Cu	0.115	11.5	0.115	11.5

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