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Determination Of phytochemical Screening And Antioxidant Activity Of Phenolic Compounds Extracting From Okra (Abelmoschus Esculentus) Cultivated In North Darfur State.

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Abstract

The aim of the present work is to investigate the phytochemicalprofilesscreening and antioxidant activity of phenolic compounds from okra, Abelmoschus esculentus L., (ladies finger). Which known as tropical vegetable, widely planted in North Darfur State Sudan.Maceration method was used to prepare the crude extracts with methanol and chloroform (1:1) to determine the contents of Abelmoschus esculentusparts by GC-MS,The phytochemical screening made upon the crude extracts of A. esculentus revealed the presence of tannins, flavonoids, reducing compounds, sterols and terpens. The antioxidant capacities in theforms of DPPH (2, 2-diphenyl-1-picrylhydrazyl) wasevaluated by spectrophotometric method.The results showed also presence of some bioactive principles such as total phenolic compounds content, flavonoids and tannins. Hence, the total phenolic compounds representa potential source of antioxidants. Okra is a nutritional source of power used throughout history for both medicinal and culinary purposesis good source of naturalantioxidants that are responsible for the health benefits.

Keywords: phytochemical screening, Abelmoschus esculentus, antioxidant capacity.

1. Introduction

Okra is one of the most important a food crop, and its elongated, edible pods are mostly harvested during the immature stages and eaten primarily as a vegetable dish[27]. Okra (Abelmoschus esculentus (L.) is one of futher important vegetable which is widely distributed from Africa and Asia having better dietary value with medicinal and industrial importance, is flowering plant in the mallow family. Even though, the plant is cultivated in tropical and warm temperate region [1]. It is one of the most widely

known and utilized species of the family Malvaceae [2] and an economically important vegetable crop grown in tropical and sub-tropical parts of the world.

Okra plays an important role in the human diet by supplying carbohydrates, minerals and vitamins[3].

At last time, okra has been used not only for its nutritional values but, also, for its nutraceutical and therapeutic properties, owing to the presence of various important bioactive compounds and their associated bioactivities. This review presents a summary of the nutritional significance of okra, as well as the possible pharmacological applications of okra bioactive components, and to explore the possible characteristics forthe development and formulation of nutraceuticals andfunctional food.

Many researchers indicate that vegetables may serve as an excellent dietary source of natural antioxidants for disease prevention and health promotion [4]. This potential is linked to their richness in secondary metabolites, among which are the phenolic compounds. Polyphenols are present in high amounts in most of foods plant and beverages [5]which cannot be synthesized by humans [6]. In the 1990s, several epidemiological studies demonstrated that dietary polyphenol consumption is associated with a reduced risk of cardiovascular disease [7,8]. As the basic and clinical research progressed, multiple functions ofpolyphenols contributing to human health were identified [9,10]. To help the indigenous population to fight against many diseases, it is necessary to identify and propose to them the plants food rich in phenolic compounds and accessible at a lower cost. The plant has many benefits, it is used as fresh leaves, buds, flowers, pods, stems and seeds [12] (Figure 1). On the other hand, eating too much okra can have adverse effects on some people as gastrointestinal problems because it is rich in fructans, a type of carbohydrates that can cause diarrhea, gas, cramping and bloating in people with bowel problems. It also can cause oxalates and kidney stones as it contains high oxalate contents.

Okra, Abelmoschus esculentus (L.), is an important vegetable crop grown mainly in the tropical or sub-tropical regions during summer and rainy season [13]. It is composed primarily of water, carbohydrates, and proteins with very little fat and a fair amount of dietary fiber. It is considered as a powerhouse of valuable nutrients, nearly half of which are soluble fibers in the form of gums and pectins which help to lower serum cholesterol and reducing the risk of heart diseases. The other fraction of okra is insoluble fiber,which helps to keep the intestinal tract healthy[13].

Figure 1. Abelmoschus esculentus L

Okra is rich in phenolic components which have remarkable pharmacological and antioxidant activities [14]. The flavonoid constituents of seed extracts showed enhanced cytotoxic effects on human-derived breast cancer cells (MCF-7) in comparison with hepatoma cells of human origin (HepG2) and human cervical cancer (HeLa) cells in a dose-dependent manner. These observations affirmed that the flavonoid isoquercitrin, in a synergistic association with other flavonoids, inhibited vascular endothelial growth factor (VEGF), resulting in the apoptosis of cancerous cells[26].

The evaluation of interactions between biologically active compounds and other components of the food matrix can be considered as the first action in the investigation of potential benefits of this annual herb, Okra is a nutritional source of power used throughout history for both medicinal and culinary purposesis good source of minerals, vitamins and nutrients that are responsible for the health benefits.It has various reported pharmacological properties like antidiabetic, antioxidant, and neurological disorders etc [15].A recent report elucidated that purified fractions of flavonoids from the flowers of okra plants had a significant antitumor effect on colorectal malignancy both in-vitro, as well as in vivo, exerting a strong antioxidant potency concomitantly with substantial antiproliferative effects on tumor growth. The antiproliferative effect of flavonoids within okra flowers induced the activation of p53, culminating into the ceasing of mitochondrial functions within colorectal tumor cells, ultimately resulting in apoptosis and restraining the autophagy [11]Okra is beneficial in normalizing blood sugar in the body by improving the rate as a result of which sugar is absorbed from the intestinal tract, okra is full of dietary fibers, which is essential for colon health and digestive health as a whole, plus it helps in promoting healthy skin. Vitamin C helps keeping the skin looking young and vibrant. Additionally, the

presence of these fibers together with other nutrition manifests its advantages of lowering sugar levels in the body, hence improves the concentration of the blood sugar level by reducing sugar absorption through the intestines [16]. Okra is a multipurpose crop due to its various uses of the pods, fresh leaves, buds, flowers, stems, and seeds. Okra immature fruits, which areconsumed as vegetables, can be used in salads, soups, and stews, fresh or dried, fried or boiled [17]. In addition, the plant has been used medicinally in treatment of several disorders. Anti-cancer, antimicrobial and hypoglycemic activities of plant are reported. The anti-ulcer activity of fresh fruits is recently reported [18].

2. Material and Methods

2.1. Plants Materials

Okra (Abelmoschus esculentus L.) samples were collected from a local market in El Fashir city (Umdafaso) in October 2020. The Okra washed properly with distilled water several times, to remove dust and other foreign particles and left on a clean surface to dry in the shade for 10-15 days. The dried material was grinded to fine powder using blender grinder and stored in sealed plastic bags at – 25 ^{0}C. The Powdered material was used further preparation of extracts for phytochemical screening and antioxidant studies.

2.2. Extraction method

150 g of fruit powder sample of Abelmoschus esculentus Okra were extracted by macerated with methanol, chloroform (1:1) and allowed to stand at room temperature for a period of 7 days with frequent agitation until the soluble matter has dissolved, the extract was filtered and evaporated by using rotary flask evaporator (IKA®RV10) (rotate 20 rpm heat 25 C^o) after 3hours remove the solvent and let it to dry, the residue extracts obtained was stored in container in the dark and kept at 20^{0}C until further tests to determine the total phenolic contents, antioxidant activityand GC-MS analysis.

2.3. Phytochemical Screening

Screening is a qualitative chemical analysis based on differential staining or precipitation reactions of the major chemical compounds groups contained in plants.

In the phytochemical tests; were screened find for presence of the active chemical constituents present in Abelmoschus esculentus Okra powder; such as flavonoid, phenolic compounds and tannins by the following procedures.

2.3.1 Test for Flavonoid

A few drops of diluted sodium hydroxide solution added to the stock solution ofOkra (0.5 mL). An intense yellow colour appeared in the plant crude extract, which became colorless upon the addition of a few drops of diluted H_2SO_4 acid,the test done according to[19].

2.3.2 Test for Phenolic Compounds (Ferric chloride test)

0.5 g crude plant extract of of Abelmoschus esculentus Okra was diluted in 5 ml of distilled water and filtered. To the filtrate, 5% Ferric chloride was added,the test done according.[20]

2.3.3 Test for Tannins

To 0.5 mL of extract solution of Abelmoschus esculentus Okra, 1 ml of water and 1- 2 drops of ferric chloride solution wad added,the test done according to.[21]

2.3.4 (GC/MS) Analysis

Gas Chromatography/Mass Spectrometry analyses were performed on an Agilent Technologies 7890A GC System, 5975C with Triple-Axis Detector mass spectrometer witha built-in- Auto sampler formed with the usage of HP-5 capillary column (30 m x 0.32 mm x 0.25 mm). GC/MS detection, electron ionization system and ionization energy was used. Helium carrier gas at a flow rate of 1 mL min-1, the column temperature program was the same as described above[22].

3. Results and Discussion

3.1. Phytochemical Screening

The biochemical screening ofchloroform and methanol extracts from dry powders of Abelmoschus esculentus were subjected to preliminary phytochemical analysis so as to find out the phytoconstituents present in the samples. The study revealed that the extract contained a blue colour precipitate immediately produced which indicates the presence of gallic tannins and green black for catecholic tannins. Yellow colour appeared which became colourless this shows the presence of flavonoids. Observed for dark green colour formation indicates for the Phenolic Compounds.

Table (1) shows the different metabolites identified in the plant materials studied. Various secondary metabolites have been identified in the powder extract of these varieties by a series of color and precipitation reactions more or less specific to each class of active ingredients. The phytochemical

screening of Abelmoschus esculentusextract studied showed the presence of active chemical constituents such as flavonoids, tannins and phenolic compounds.

(Table 1): Phytochemical analysis of extracts from Abelmoschus esculentus Okra.

Secondary metabolites		**varieties**				
		V1	V2	V3	V4	V5
Phenolic compounds		+	+	+	+	+
Flavonoids		+	+	+	+	-
Tannins	Cathechic	+	+	+	+	+
Gallic		-	-	-	-	-

+ =Presence, - =absence.

The effective bioactive compoundsflavonoids, tannins and phenolic compounds were found in Abelmoschus esculentus Okra powderextracted by macerated with methanol, chloroform (1:1). Therefore, the detected different bioactive compounds may be responsible for the antioxidant activities. Several reports are available on phenolic compounds. According to [23], it has been reported that okra fruit has high amounts of total flavonoids and moderate amounts of total phenolics which both components are a good source of natural antioxidants. The flavonoid content of okra fruit which has biggest portion in phenolic compound, may affect the antioxidant activity of okra fruit.

GC-MS analysis (Table **2**) of Abelmoschus esculentus Okra powder extracts were performed antioxidant activities were obtained from powder extractsmight be due to the chemical profiles as studied in GC-MS Fig (2) Octadecadienoic acid, methyl ester2.18%Phytol 7.09%, andHexadecanoic acid, methyl ester6.45%.

okra extract has several hydroxyl groups in its structure, which can donate H+ atoms to stabilize free-radical reactions [25].

(**Table2**) GC-**MS determination of Abelmoschus esculentusMOH/$CHCl_3$ (1:1) Fruitpowder extracts.**

Compound number	RT (min)	%	Name
1	22.474	2.18	Octadecenoic acid (Z), methyl ester
2	24.73	3.04	7,10,13-Hexadecatrienoic acid, methyl ester

3	26.727	0.21	Phenol,2,4bis(1,1dimethylethyl)
4	28.455	6.45	Hexadecanoicacid, methylester
5	31.978	7.09	Phytol
6	32.959	1.2	Acetic acid, 2(dimethylamino) ethyl ester
7	32.317	1.2	Heptadecane
8	33.253	1.12	Tetradecanoicacid,12methyl,methylester
9	34.771	0.26	1,2Benzenedicarboxylicacid,dibutylester
10	31.712	6.54	9,12 Octadecadienoic acid, methyl ester

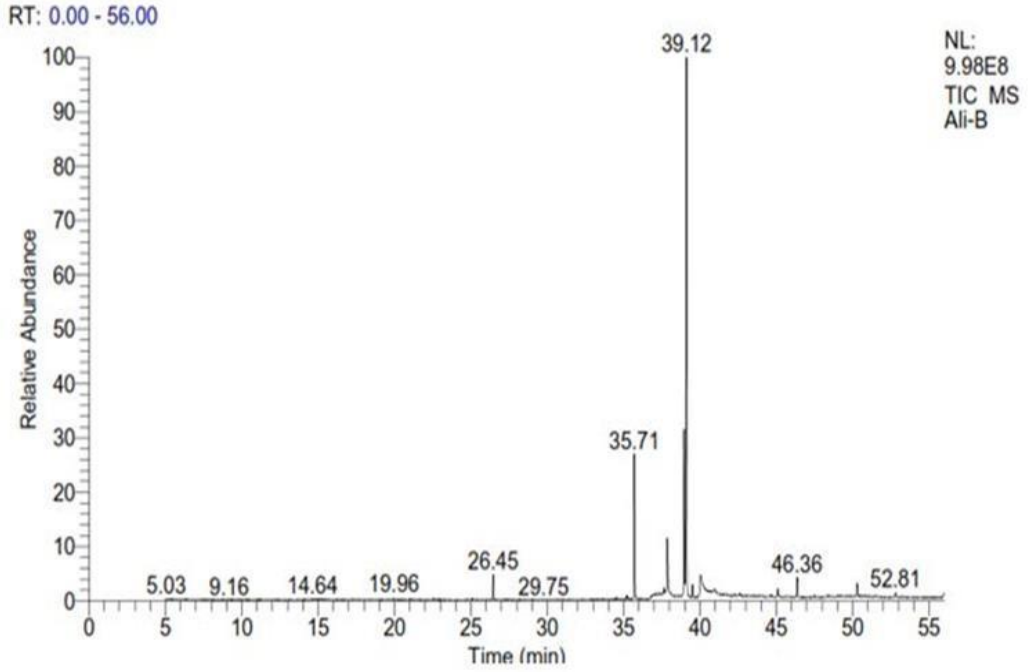

Figure (1)GC-MS of Abelmoschus esculentus L

4. Conclusion and recommendation

The main features of the okra, Abelmoschus esculentus L., (ladies finger) is a well-known tropical vegetable, widely planted in North Darfur State.Okra is a nutritional source of power used throughout history for both medicinal and culinary purposes. Okra is good source of minerals, vitamins and nutrients that are responsible for the health benefits. This effort is towards providing the evidence in support to encourage more scientific research to find out more pharmacological and nutritional potential of Abelmoschus esculentus that may be suggestive of new drug discovery.Okra, as well as its

specific mechanisms of action on different diseases, need more investigation.To focus how to complex etiology of diseases, along with different related factors aiding the diseases.

The results of this study clearly showed significantly different phenolic contents and antioxidant activities, source of phenolic compounds and potential antioxidants that could be recommended in pharmaceutical and food preparations.

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Abbreviations

GC: gas chromatography

MS : mass spectrometry

MOH/$CHCl_3$: chloroform methanol,

VEGF:vascular endothelial growth factor