International Food Research Journal 25(6): 2292-2300 (December 2018)
Journal homepage: *http://www.ifrj.upm.edu.my*

Evaluation of antioxidant, antimicrobial activities and phytochemical content of some Egyptian plants

[1*]Rashed, K., [2]Ivanišová E. and [3]Kačániová M.

[1]*Pharmacognosy Department, National Research Centre, 33 El-Bohouth st.-Dokki, Giza, Egypt-P.O.12622.*
[2]*Department of Storing and Processing Plant Products, Slovak University of Agriculture in Nitra, Tr. A. Hlinku 2, Nitra, SK-949 76, Slovak Republic*
[3]*Department of microbiology, Slovak University of Agriculture in Nitra, Tr. A. Hlinku 2, Nitra, SK-949 76, Slovak Republic*

Article history

Received: 4 December 2017
Received in revised form: 18 January 2018
Accepted: 5 February 2018

Keywords

Egyptian plants
Stems
Antioxidant
Antimicrobial
Phytoconstituents

Abstract

This study evaluated antioxidant and antimicrobial activities of methanol extracts of *Artocarpus heterophyllus, Parkia africana, Derris rubosta* and *Cichorium intybus* stems and also phytochemical analysis of the four plants was detected. Antioxidant activity was evaluated by radical scavenging assay and phosphomolybdenic assay. Total phenolics and flavonoids were estimated by Folin-Ciocalteu reagent and aluminium chloride method. The antimicrobial screenings were carried out via disc diffusion method and minimal inhibition concentration against three species of gram negative bacteria: *Escherichia coli* CCM 3988, *Salmonella enterica* subsp. *enterica* CCM 3807, *Yersinia enterocolitica* CCM 5671 and three grampositive bacteria: *Bacillus thuringiensis* CCM 19, *Listeria monocytogenes* CCM 4699, *Stapylococcus aureus* subsp. *aereus* CCM 2461. All the plants tested had some antioxidant activity. *Parkia africana* had the highest antioxidant activity values of 47.99 µg TEAC/g and 1036.56 µg TEAC/g in the DPPH• and phosphomolybdenum assays respectively; along with the highest amount of total flavonoids (0.165 µg QE/g). The inhibition zones against three gram-negative strains of different tested plants were ranged from 10-24 mm. From our results the P*arkia africana* extract showed the strongest action against *E. coli* (24 mm), *S. enterica* subsp. *enterica* (21 mm) and *Y. enterocolitica* (22 mm). The minimum inhibitory affect of plants ranged from 16 to 256 µg/ml. The best antibacterial activity was found at *Parkia africana* against gram-negative bacteria strains. The results indicated that the plants tested may be potential sources for isolation of natural antioxidant and antimicrobial compounds.

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Introduction

In recent years, the number of multi–drug resistant microbial strains and the appearance of strains with reduced susceptibility to antibiotics are continuously increasing in alarming rate. This increase has been attributed to the abusive and indiscriminate use of broad-spectrum antibiotics, immunosuppressive agent, intravenous catheters, organ transplantation and ongoing epidemics of HIV infection (Graybill, 1988, Dean and Burchard, 1996). Medicinal plants are important sources of natural products. They have been screened for their potential uses as alternatives remedies for the treatment of many infectious diseases. Medicinal plants are of great interest as sources of natural products. They have been screened for their potential uses as alternatives remedies for the treatment of many infectious diseases. During the last decade, many reports focused on the use of several medicinal herbs for improving the healthy lifestyle of humans and suggested that the health benefits provided by these plants are attributable to the presence of various bioactive compounds (Dzoyem *et al*., 2014). Phenolic compounds mainly flavonoids and phenolic acids exhibit strong biological activities (Wijekoonetal, 2013). Antioxidants from natural sources, predominantly phenolic compounds, are also considered as important factors for inhibiting oxidative stress in human body and many authors reported that the antioxidant properties of plants could be correlated with oxidative stress defense (Costantino *et al.*, 1992, Rice-Evans *et al.*, 1997). From chemical point of view phenolic compounds belong to a secondary plant metabolites, which comprising a wide variety of molecules with a polyphenol structure, but also molecules with one phenol ring, such as phenolic acids and phenolic alcohols. Phenolic compounds are a very important and wide group of phytochemicals that can be found in plants. Nowadays, the identification and

*Corresponding author.
Email: *khalednabih2015@yahoo.co.uk*

development of polyphenols or extracts from various plants considered strong antioxidants in vitro has become a major area of human health, pharmacy and medical related research (Ramkissoon *et al*., 2013).

Nearly 80% of people living in the developing countries especially in Africa depend on herbal medicine for their health needs including wounds, infectious and metabolic diseases (Agyare *et al*., 2009). Interest in medicinal plant research has escalated, with the aim of identifying alternative antimicrobial therapies to overcome resistance (Aiyegoro and Okoh, 2009). There is, however, general consensus amongst the various studies, that plant derived antimicrobials possess a lower potency than conventional antimicrobials (Van Vuuren and Viljoen, 2011). Furthermore, antimicrobial resistance against conventional antimicrobials has been on the rise and has become a major public health concern. This has propelled research in the direction of combination therapies for enhanced efficacy. Many researchers have studied antimicrobial interactions between natural products, as well as combinations of natural products with conventional therapies (Hübsch *et al*., 2014). No previous reports on bioactivities of the stems of these plants and also their phytoconstituents and this part of the other species of these plants were very active as anticancer and antioxidant agents. The objective of this research was to screen some plant species, Artocarpus heterophyllus, Parkia africana, Derris rubosta and Cichorium intybus stems and to investigate their antioxidant and antimicrobial activities and their phytochemical profile.

Materials and Methods

Plants collection and identification

Plants were collected from Al-Zohiriya garden, Giza, Egypt in May 2012. The identification of the plants was by Dr. Mohammed El-Gebaly, National Research Centre (NRC), Giza, Egypt.

Chemicals

All chemicals were analytical grade and were purchased from Reachem (Slovakia) and Sigma Aldrich (USA).

Plants extracts preparation

Stems powdered of *Artocarpus heterophyllus, Parkia africana, Derris rubosta* and *Cichorium intybus* (200 g) were extracted with methanol until exhaustion. The dried extracts were 8 g, 7.2 g, 6.9 g, and 7.5 g, respectively. Phytochemical investigation was done according to Yadav and Agarwala, (2011).

Sample preparation for measurement

0.01 g of each herbal extract was dissolved with 10 mL of methanol used for measurement (DPPH method, phosphomolybdenum method, total phenolic content and total flavonoid content– all spectrophotometric analyses were carried out in quadruplicate).

Antioxidant acitivity

Radical scavenging activity

Radical scavenging activity of extracts was measured using 2,2-diphenyl-1-picrylhydrazyl (DPPH) (Sánchéz-Moreno *et al*., 1998). The sample (0.4 mL) was mixed with 3.6 mL of DPPH solution (0.025 g DPPH in 100 mL methanol). Absorbance of the reaction mixture was determined using the spectrophotometer Jenway (6405 UV/Vis, England) at 515 nm. Trolox (6-hydroxy-2, 5, 7, 8-tetramethylchroman-2-carboxylic acid) (10-100 $mg.L^{-1}$; R^2=0.989) was used as the standard and the results were expressed in $\mu g.g^{-1}$ Trolox equivalents.

Reducing power

Reducing power of extracts was determined by the phosphomolybdenum method of Prieto *et al*., (1999) with slight modifications. The mixture of sample (1 mL), monopotassium phosphate (2.8 mL, 0.1 M), sulfuric acid (6 mL, 1 M), ammonium heptamolybdate (0.4 mL, 0.1 M) and distilled water (0.8 mL) was incubated at 90°C for 120 min, then rapidly cooled and detected by monitoring absorbance at 700 nm using the spectrophotometer Jenway (6405 UV/Vis, England). Trolox (10-1000 $mg.L^{-1}$; R^2=0.998) was used as the standard and the results were expressed in $\mu g.g^{-1}$ Trolox equivalents.

Total polyphenol content

Total polyphenol content extracts was measured by the method of Singleton and Rossi (1965) using Folin-Ciocalteu reagent. 0.1 mL of each sample was mixed with 0.1 mL of the Folin-Ciocalteu reagent, 1 mL of 20% (w/v) sodium carbonate, and 8.8 mL of distilled water. After 30 min. in darkness the absorbance at 700 nm was measured using the spectrophotometer Jenway (6405 UV/Vis, England). Gallic acid (25-300 $mg.L^{-1}$; R^2=0.998) was used as the standard and the results were expressed in $\mu g.g^{-1}$ gallic acid equivalents.

Total flavonoid content

Total flavonoids were determined using the modified method of Willett (2002). 0.5 mL of sample was mixed with 0.1 mL of 10% (w/v) ethanolic

solution of aluminium chloride, 0.1 mL of 1 M potassium acetate and 4.3 mL of distilled water. After 30 min. in darkness the absorbance at 415 nm was measured using the spectrophotometer Jenway (6405 UV/Vis, England). Quercetin (0.5-20 $mg.L^{-1}$; R^2=0.989) was used as the standard and the results were expressed in $\mu g.g^{-1}$ quercetin equivalents.

Antimicrobial activity

Microbial tests

Six strains of microorganisms were tested in this research, including three gram-negative bacteria (*Escherichia coli* CCM 3988, *Salmonella enterica* subsp. *enterica* CCM 3807, *Yersinia enterocolitica* CCM 5671) and three gram-positive bacteria (*Bacillus thuringiensis* CCM 19, *Listeria monocytogenes* CCM 4699, *Stapylococcus aureus* subsp. *aereus* CCM 2461). All tested strains were collected from the Czech Collection of Microorganisms. The bacterial suspensions were cultured in the nutrient broth (Imuna, Slovakia) at 37°C.

Preparation of plant extracts

For the antimicrobial assays, each herbal extract was dissolved in dimethyl sulfoxid (DMSO) (Penta, Czech Republic) to 102.4 mg/ml as stock solution, while for chemical analysis methanol was used as solvent. Stock solutions of plant extracts were stored at -16°C in refrigerator until use (Hleba *et al*., 2014).

Disc diffusion method

The agar disc diffusion method was used for the determination of antimicrobial activities of the plant extract. Briefly, a suspension of the tested microorganism (0.1 ml of 10^5 cells per ml) was spread on the solid media plates. Filter paper discs (6 mm in diameter) were impregnated with 15 µl of the plant extract and placed on the inoculated plates. They were inoculated onto the surface of Mueller Hinton Agar (MHA, Oxoid, Basingstoke, United Kingdom). These plates, after remaining at 4°C for 2 hours, were incubated aerobically at 37°C for 24 h. The diameters of the inhibition zones were measured in millimeters. All the tests were performed in duplicate.

Minimal inhibitory concentration

The minimum inhibitory concentration (MIC) is the lowest concentration of the sample that will inhibit the visible growth of microorganisms. Plant extracts dissolved in DMSO were prepared to a final concentration of 102.4 mg/ml by dissolving stock solution with 102.4 mg/100 ml. MICs were determined by the microbroth dilution method in Mueller Hinton broth (Biolife, Italy) for bacteria. Briefly, the DMSO plant extracts solutions were prepared as serial two-fold dilutions, in order to obtain a final concentration ranging between 0.5-512 µg/ml. Each well was then inoculated with microbial suspension at the final density of 0.5 McFarland. After 24 h incubation at 37°C for bacteria, the inhibition of microbial growth was evaluated by measuring the well absorbance at 450 nm in an absorbance microplate reader Biotek EL808 with shaker (Biotek Instruments, USA). The 96 microwell plates were measured before and after experiment. Differences between both measurements were evaluated as growth. Measurement error was established for 0.05 values from absorbance. Wells without plant extracts were used as positive controls of growth. Pure DMSO was used as negative control. This experiment was done in eight-replicates for a higher accuracy of the MICs of used medical plant extracts (Hleba *et al*., 2014).

Results and Discussion

This study evaluated antioxidant, antimicrobial activities and chemical constituents of *Artocarpus heterophyllus, Parkia africana, Derris rubosta* and *Cichorium intybus* stems.

Antioxidant activity

The antioxidant properties evaluated by two differential method (DPPH and phosphomolybdenum method) of each herbal extract are summarized in Figure 1. The highest activity by DPPH method was found in *Parkia africana* (47.99 µg TEAC.g^{-1}), followed by *Derris rubosta* (43.61 µg TEAC.g^{-1}), *Artocarpus heterophyllus* (29.64 µg TEAC.g^{-1}) and *Cichorium intybus* (21.93 µg TEAC.g^{-1}). DPPH is a protonated radical having the characteristic absorption maxima at 517 nm which decreases with the scavenging of the proton radical by plant extracts. Hence, DPPH finds applications in the determination of the radical scavenging activity of plant materials (Narayanaswamy and Balakrishnan, 2011). *Parkia africana* possessed highest DPPH radical scavenging activity. A study reported antioxidant properties of *Parkia* genus and confirmed strong activity which was higher than that of the standards (rutin, ascorbic acid, BHA) (Adaramola *et al.*, 2012). Higher activity by DPPH method was also detected in *Derris rubosta.* A study showed that flavonoids and tannis were extracted from *Derris* roots and determined strong antioxidant and also antimicrobial activity of these extracts (Sharif *et al.*, 2014)

By phosphomolybdenum method (reducing power) was the highest antioxidant activity (reducing

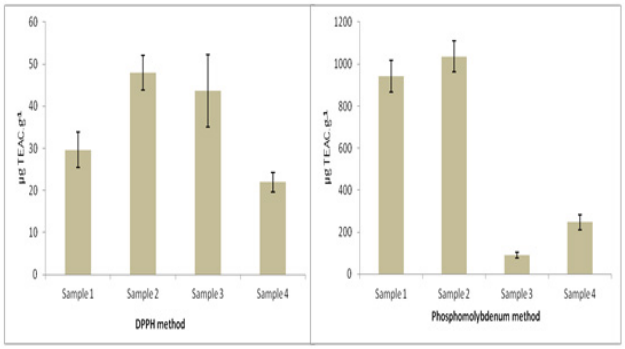

Figure 1. Antioxidant activity of plant extracts tested by DPPH and phosphomolybdenum method (sample 1 - *Artocarpus heterophyllus*, sample 2 - *Parkia africana,* sample 3 - *Derris rubosta*, sample 4 - *Cichorium intybus*; TEAC – Trolox equivalent antioxidant capacity)

power) detected in *Parkia africana* (1036.56 µg TEAC.g^{-1}) followed by *Artocarpus heterophyllus* (942.28 µg TEAC.g^{-1}), *Cichorium intybus* (246.87 µg TEAC.g^{-1}) and *Derris rubosta* (89.84 µg TEAC.g^{-1}). The reducing ability of a plant extracts can be as a significant marker of its potential antioxidant activity. The reducing power of a compound generally attributes on the presence of reductones, which have indicated antioxidative efficiency by breaking the free radical chain and donating a hydrogen atom (Huang *et al*., 2007). As shown in Figure 1, all tested plant extracts possessed considerable reducing properties as demonstrated by its ability to reduce Mo^{6+} to Mo^{5+} suggesting that the antioxidants present in the plant extracts probably act as reductones by donating electrons to free radicals and terminating the free radical mediated chain reactions. The propensity for metal chelation, particularly iron and copper, supports the role of antioxidants, mainly phenolic compounds as important antioxidants in terms of inhibiting transition metal-catalyzed free radical formation because it reduces the concentration of the transition metal that catalyzes lipid peroxidation (Komolafe *et al.*, 2014). Similar like DPPH, the best result was recorded in *Parkia africana* extracts. Extract from *Artocarpus heterophyllus* also shown strong antioxidant activity (reducing power). Previous report indicated that the extract from *Artocarpus heterophyllus* seeds had a strong reducing power (67 µM in 1 g of dry sample) tested by FRAP method, in which antioxidants from extract reduce Fe^{3+} to Fe^{2+} (Jagtap and Bapat, 2010).

Previous report determined strong reducing power by FRAP method in *Artocarpus heterophyllus* leaves water extract (565.8 µM Fe(II)/g) and also reported that this plant can be use more not only in pharmacy but also food industry (Baliga *et al.*, 2011). The antioxidant activity of tested plant extract may be due to the hydrogen donating ability of phenols and flavonoids present in it.

Table 1. Total polyphenol and flavonoid contents of the plants extracts

Sample	Total polyphenol content (µg GAE.g^{-1})	Total flavonoid content (µg QE.g^{-1})
Artocarpus heterophyllus	6.389 ±1.111	0.001 ±0.000
Parkia africana	44.173 ±4.157	0.165 ±0.003
Derris rubosta	2.585 ±0.786	0.006 ±0.001
Cichorium intybus	124.868 ±4.037	0.084 ±0.001

GAE – gallic acid equivalen, QE – quercetin equivalent

Total polyphenol and flavonoid content

Antioxidant activity of the plant extract is often associated with the polyphenol compounds present in them. Plant polyphenols constitute the major group of compounds that act as primary antioxidant (Hatano *et al*., 1989). They can inhibit the lipid peroxidation by their reaction with active oxygen radicals. Nowadays plant materials rich in polyphenols are increasingly being used mainly in the food industry because they can protect lipids from oxidation and increase nutritional value of food (Kähkönen *et al*., 1999). In this study was evaluated total polyphenols and flavonoid content in methanol plant extracts. A report indicated that methanol extract exhibits the highest total polyphenols content, with compare to aqueous extract, in which is much smaller total polyphenols content (Ao *et al*., 2008). The highest total polyphenol content (Table 1) was found in *Cichorium intybus* (124.868 µg GAE.g^{-1}), followed by *Parkia africana, Artocarpus heterophyllus* and *Derris rubosta.* The phytochemical analysis of *Cichorium intybus* showed, that this plant contains coumarins, flavonoids and tannins, compounds from polyphenol class. Tannins are potent antioxidant, because many studies confirmed their antimicrobial and anticancer activities. Antioxidant activity of tannins is attributed to ability act by iron sequestration, hydrogen bonding or specific reactions with proteins such as enzymes (Adaramola *et al.*, 2012). A study determined compositions of *Cichorium intybus* root and found that from polyphenols are predominat in root chlorogenic and dicaffeoylquinic acids (Milala *et al*., 2009). In *Parkia africana* was also detected high amount of total polyphenol. A study was published that *Parkia* bitter bean and leaves are rich for polyphenols, mainly gallic acid, catechin, ellagic acid and quercetin (Huey-Jiun *et al*., 2014, Adaramola *et al.*, 2012).

Flavonoids are phytochemicals and belong to the polyphenols. A wide variety of beneficial factors has been attributed to their mode of action. Some of their activities concern the inhibition of inflammatory pathways and the down regulation of genes involved in chronic inflammatory disease states (Hoensch and Oertel, 2015). Flavonoids are present in most plants with high concentrations found in fruit peels, leaves and flowers (Pei-Dawn *et al*., 2002). In this study total flavonoid content in plant extract with aluminium chloride method was determined. As demonstrate in Table 1 the highest content was found in *Parkia africana* (0.165 µg QE.g^{-1}), followed by *Cichorium intybus, Derris rubosta* and *Artocarpus heterophyllus. Parkia africana* showed the best results, not only in flavonoid content but also in antioxidant activity. Our findings are with accordance to results from several authors (Cook and Samman, 1996; Atanassova *et al.*, 2011) which reported that flavonoids are responsible for the radical scavenging activity and chelating processes.

Antimicrobial activity

Natural herbal medicine practitioners in many African nations have employed different parts e.g. seed, stem bark, root, leaves, pod, pulp, seed and flower of the locust bean tree as remedy to many human infections and ailments. *Artocarpus heterophyllus* leaf and bark showed the presence of glycosides, terpenoids and in addition alkaloids, saponins were also found in the bark extract. While the methanol extract of leaf showed flavanoids, phenols, glycosides, and terpenoids and its bark showed above all these compounds alkaloids, tannins, steroids, saponins and anthraquinone except cardiac glycosides. From the screening experiment, methanol extracts of *A. heterophyllus* extracts showed the best antibacterial activity; and hence they can be further subjected to isolation of the therapeutic antimicrobials and for the further pytochemical and pharmacological studies that may open the possibility of finding new clinically effective antimicrobial compounds (Binumol and Sajitha, 2013).

A study conducted in Togo confirmed the medicinal values of *P. biglobosa* and its popularity in the nation's traditional healthcare services (Karou *et al.*, 2011). The efficient wound-healing property of the plant in the southwestern region of Nigeria was reported by (Adetutu *et al.*, 2011) while (Traore *et al*., 2013) reported that the plant extracts was used as anti-malaria in Guinea. The anti-bacterial property of the plant was observed by (Abioye *et al*., 2013) to compare with synthetic streptomycin in action and potency. Similarly, the crude extract from different parts of *P. biglobosa* showed positive inhibitory effect on the growth of methicillin resistant *Staphylococcus aureus* (MRSA). Stem bark at various concentrations (10 – 25 mg/ml) was observed to be most active against MRSA isolates from orthopedic patients (Ajaiyeoba, 2002) suggesting strong antimicrobial potentials.

Gram-positive bacteria than gram-negative bacteria as evident from the fact that MIC value of active fraction of *Cichorium intybus* is less in case of gram-positive bacteria than gram-negative bacteria. It indicates that outer envelope of gram-negative bacteria to some extent protecting the gram-negative bacteria from inhibitory action of *Cichorium intybus* extract. As chicory root also have fungal inhibitory actions so the mechanism of action of active principle is as applicable on both prokaryotes and eukaryotes. However hexane extract of the chicory root has neither antibacterial nor antifungal activities indicating that active principle of chicory root is certainly a polar compound. This extract can be used for the preparation of effective antimicrobial agents. The present work shows that the compounds from chicory possess potent antimicrobial activity and suggesting that the chicory root extracts contains the effective active constituents responsible for eliminating the bacterial concentration (Koner *et al.*, 2011).

Disc diffusion method

The inhibition zones against three gram-negative strains of different tested plants were ranged from 10-24 mm. From our results the *Parkia africana* extract showed the strongest action against *E. coli* (24 mm), *S. enterica* subsp. *enterica* (21 mm) and *Y. enterocolitica* (22 mm) (Figure 2). The inhibition zones against *E. coli* strain of different tested plants were ranged from 12 to 24 mm. This inhibition zones were *Parkia africana* (24 mm) > A*rtocarpus heterophyllus* (17 mm) > *Derris rubosta* (15 mm) > *Cichorium intybus* (12 mm). The inhibition zones against *S. enterica* subsp. *enterica* strain of different tested plants were ranged from 10 to 21 mm. This inhibition zones were *Parkia africana* (21 mm) > *Artocarpus heterophyllus* (15 mm) > *Derris rubosta* (13 mm) > *Cichorium intybus* (10 mm). The inhibition zones against *Y. enterocolitica* strain of different tested plants were ranged from 12 to 22 mm. This inhibition zones were *Parkia africana* (22 mm) > *Artocarpus heterophyllus* (17 mm) > *Derris rubosta* (16 mm) > *Cichorium intybus* (12 mm).

The inhibition zones against three gram-positive strains of different tested plants were ranged from 2-84 mm. From our results the *Parkia africana*

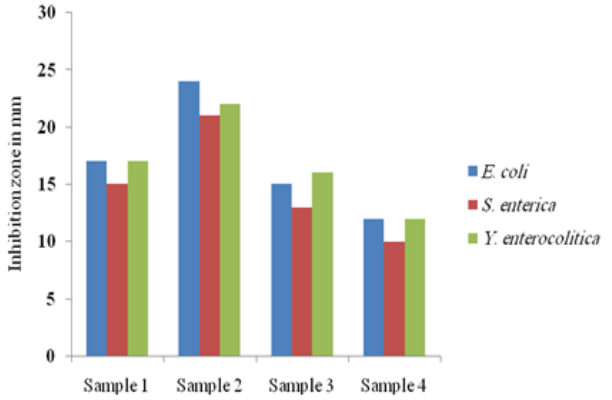

Figure 2. Antimicrobial activity in mm against gram-negative bacteria (sample 1- *Artocarpus heterophyllus,* sample 2 - *Parkia africana,* sample 3 - *Derris rubosta,* sample 4 - *Cichorium intybus*)

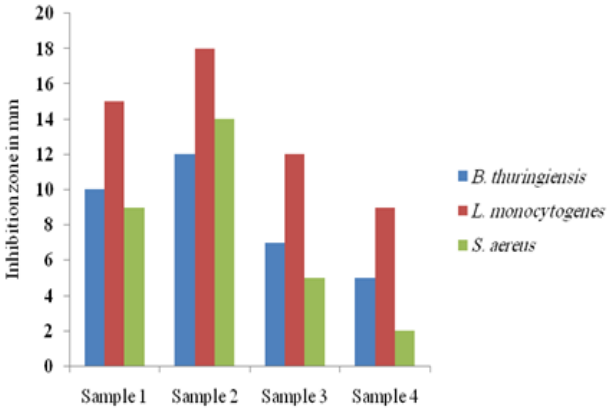

Figure 3. Antimicrobial activity in mm against gram-positive bacteria (sample 1- *Artocarpus heterophyllus,* sample 2 - *Parkia africana*, sample 3 - Derris rubosta, sample 4 - *Cichorium intybus*)

extract showed the strongest action against *L. monocytogenes* (18 mm), *S. aureus* subsp. *aereus* (14 mm) and *B. thurigiensis* (12 mm) (Figure 3). The inhibition zones against *L. monocytogenes* strain of different tested plants were ranged from 9 to 18 mm. This inhibition zones were Parkia africana (18 mm) > *Artocarpus heterophyllus* (15 mm) > *Derris rubosta* (12 mm) > *Cichorium intybus* (9 mm). The inhibition zones against *S. aereus* subsp. *aereus* strain of different tested plants were ranged from 2 to 14 mm. This inhibition zones were Parkia africana (14 mm) > *Artocarpus heterophyllus* (9 mm) > *Derris rubosta* (5 mm) > *Cichorium intybus* (2 mm). The inhibition zones against *B. thuringiensis* strain of different tested plants were ranged from 5 to 12 mm. This inhibition zones were *Parkia africana* (12 mm) > *Artocarpus heterophyllus* (10 mm) > *Derris rubosta* (7 mm) > *Cichorium intybus* (5 mm).

The results of recent study showed that unfiltered ethanol extracts of the *P. africana* stem recorded the highest zone of inhibition measuring 31 mm for *Staphylococcus aureus* while the unfiltered aqueous extract of the root had the least zone of inhibition of 9 mm diameter for *Pseudomonas aeruginosa*. In our study we found that more effective was extract of *P. africana* against gram-negative bacteria (Osemwegie and Dahunsi, 2015).

Table 2. Minimal inhibition concentration MIC (µg/ml) against gram negative and gram positive bacteria

	Artocarpus heterophyllus	***Parkia africana***	***Derris rubosta***	***Cichorium intybus***
E. coli	32	16	64	128
S. enteritidis	64	32	64	128
Y. enterocolitica	64	32	64	128
B. thurigiensis	128	64	128	256
L.monocytogenes	128	64	128	256
S. aereus	256	128	256	256

A study reported antibacterial activity of water extract of *A. heterophyllus* leaf showed inhibition zone of 8.5 mm in *B. subtilis* and 6.5 mm in *P. fluorescens.* Methanol extract showed inhibition zone of 9.5 mm in *B. subtilis* and 6.5 mm in *P. fluorescens* (Binumol and Sajitha, 2013). Extracts of *Derris trifolliata* L. were found to possess various degrees of antibacterial activity against both gram-positive and gram-negative bacteria. Among the tested extracts bioactive molecules extracted into methanol exhibited highest antibacterial activity (12.66-19.33 mm) against all the tested bacterial species irrespective of their Gram nature. Active constituents of ethyl acetate fraction specifically inhibited the growth of *E. coli, B. subtilis, En. faecalis.* Acetone solubles of *Derris trifolliata* L. exerted more or less similar activity against all the tested cultures except no activity against *En. cloacae* (Sharief *et al*., 2014). The maximum zone of inhibition 13.3 and 12.8 mm was exhibited by methanol root and leaf *Cichorium intybus* extracts respectively against *Pseudomonas aeruginosa.* On comparing the inhibitory activity of methanol extract of *Cichorium intybus* against *Escherichia coli* and *Pseudomonas aeruginosa* it was found that *E.coli* was less sensitive as compared to *P. aeruginosa* (Verma *et al*., 2013).

Minimal inhibition concentration

The antimicrobial activity (expressed as µg/ml) of four methanolic extracts from *Artocarpus heterophyllus, Parkia africana, Derris rubosta, Cichorium intybus* against various strains of bacteria are summarized in Table 2. The organism *E. coli* was found to be more susceptible to the *P. africana* extract with a MIC value of 16 µg/ml. *S. enteritidis* subsp.

enteritidis and *Y. enterocolitica* was less susceptible to *P. africana* with MIC value of 32 µg/ml. The organisms *B. thurigiensis* and *L. monocytogenes* were less susceptible to the *P. africana* extract with higher MIC values 64 µg/ml. *S. aureus* subsp. *aureus* were less susceptible to the *P. africana* extract with higher MIC values 128 µg/ml. The organism *E. coli* was found to be more susceptible to the *Artocarpus heterophyllus* extract with a MIC value of 32 µg/ml. *S. enteritidis* subsp. *enteritidis* and *Y. enterocolitica* was less susceptible to *Artocarpus heterophyllus* with MIC value of 64 µg/ml. The organisms' *B. thurigiensis* and *L. monocytogenes* were less susceptible to the Artocarpus heterophyllus extract with higher MIC values 128 µg/ml. *S. aureus* subsp. *aureus* were less susceptible to the *Artocarpus heterophyllus* extract with higher MIC values 256 µg/ml.

The organism *E. coli, S. enteritidis* subsp. *enteritidis* and *Y. enterocolitica* was found to be more susceptible to the *Derris rubosta* extract with a MIC value of 64 µg/ml. The organisms *B. thurigiensis, L. monocytogenes* and *S. aureus* subsp. *aureus* were less susceptible to the *Artocarpus heterophyllus* extract with higher MIC values 128 µg/ml.

The organism *E. coli, S. enteritidis* subsp. *enteritidis* and *Y. enterocolitica* was found to be more susceptible to the *Cichorium intybus* extract with a MIC value of 128 µg/ml. The organisms' *B. thurigiensis, L. monocytogenes* and *S. aureus* subsp. *aureus* were less susceptible to the *Cichorium intybus* extract with higher MIC values 256 µg/ml.

Ethanol extracts of *P. africana* had wider MIC values as our study that range from 3.125 to 200 mg/ml for target bacterial isolates (Akintobi *et al.*, 2013, Osemwegie and Dahunsi. 2015).

A study reported the bacterial potency of *A. heterophyllus* in methanolic extract on gram-positive bacteria, *B. subtillis* showed that result 100 mg/ml larger diameter of clearance than that of the other ethanolic and chloroform extracts of gram-positive bacteria used in this study (Madhav *et al*., 2013). Similarly, *A. heterophyllus* methanolic extract showed a maximum zone of clearance in the gram-negative bacteria *P. aeruginosa* 100 mg/ml than that of other ethanolic and chloroform extracts of gram-negative bacteria. The MIC values of study ranged from 1.25-5 mg/100µl and varied from Derris trifolliata extract (Sharief *et al.*, 2014). The MIC of acetone and methanol extracts towards the tested cultures is 5 mg/100µl. Whereas, the MIC of methanol extract is 1.25 mg/100µl to *Escherichia coli* and *Bacillus subtilis,* while the MIC value to the remaining test culture is 2.5 mg/100µl. The results of the measurement of minimum inhibitory concentration (MIC) of study, indicated that 100% *Cichorium intybus* methanolic extract showed good activity against *E. coli* and *P. multocida,* showing the lowest MIC values (80.4 and 140 mg/ml) (Mehmood *et al*., 2012). Least activity was exhibited against *B. subtilis*, with the highest MIC values (198 mg/m.). Ethyl acetate fraction showed strong activity against *E. coli* and *S. aureus* with the lowest MIC values (50.3 and 60.2 mg/mL), respectively.

Conclusion

In conclusion, our results indicate that Egyptian plants are rich sources for bioactive compounds and can be used more in future in pharmacy, medicine and food industry. On the basis of these results we can included that the stems extracts of these Egyptian plants are very active as anticancer and antioxidant agents and so it is important to inform consumers on the benefits of varying plants consumption, and choosing those that have the highest antioxidant activity in order to promote a healthy life-style. Egyptian plants showed very good antimicrobial activity against gram-positive and gram-negative bacteria. Better antimicrobial activity of Egyptian plants was found against gram-negative bacteria.

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