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Food and Bioproducts Processing

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Review

A review of the antioxidant potential of medicinal plant species

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A B S T R A C T

Some researchers suggest that two-thirds of the world's plant species have medicinal value; in particular, many medicinal plants have great antioxidant potential. Antioxidants reduce the oxidative stress in cells and are therefore useful in the treatment of many human diseases, including cancer, cardiovascular diseases and inflammatory diseases. This paper reviews the antioxidant potential of extracts from the stems, roots, bark, leaves, fruits and seeds of several important medicinal species. Synthetic antioxidants such as butylated hydroxytoluene (BHT) and butylated hydroxyanisole (BHA) are currently used as food additives, and many plant species have similar antioxidant potentials as these synthetics. These species include *Diospyros abyssinica*, *Pistacia lentiscus*, *Geranium sanguineum* L., *Sargentodoxa cuneata* Rehd. Et Wils, *Polyalthia cerasoides* (Roxb.) Bedd, *Crataeva nurvala* Buch-Ham., *Acacia auriculiformis* A. Cunn, *Teucrium polium* L., *Dracocephalum moldavica* L., *Urtica dioica* L., *Ficus microcarpa* L. fil., *Bidens pilosa* Linn. Radiata, *Lea indica*, the *Lamiaceae* species, *Uncaria tomentosa* (Willd.) DC, *Salvia officinalis* L., *Momordica Charantia* L., *Rheum ribes* L., and *Pelargonium endlicherianum*. The literature reveals that these natural antioxidants represent a potentially side effect-free alternative to synthetic antioxidants in the food processing industry and for use in preventive medicine.

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Keywords: Antioxidant; Oxidative stress; Medicinal species; Different countries

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Abbreviations: ABTS, 2,2'-azinobis(3-ethylbenzthiazoline-6-sulphonic acid); ATC, ammonium thiocyanate; BHA, butylated hydroxyanisole; BHT, butylated hydroxytoluene; BR, Briggs Rauscher; DAD, diode array detector; DPPH, 1,1-diphenyl-2-picrylhydrazine; FRAP, ferric reducing antioxidant power; FTC, ferric thiocyanate; FTC, Folin-Ciocalteu; GA, gallic acid; GAE, gallic acid equivalents; GPx, glutathione peroxidase; HPLC, high performance liquid chromatography; LPO, lipid peroxidation; NBT, nitro blue tetrazolium; ORAC, oxygen radical absorbance capacity; PCL, luminol-photochemiluminescence; PEs, pyrocatechol equivalents; PG, propyl gallate; PMS-NADH, phenazine methosulfate-nicotinamide adenine dinucleotide-reduced; QEs, quercetin equivalents; Re eq., resorcinol equivalents; ROS, reactive oxygen species; SOD, superoxide dismutase; TBA, thiobarbituric acid; TEAC, trolox equivalent antioxidant capacity; TFA, total flavonoids; TFO, total flavonols; TP, total phenols; TPC, total phenolic content; TRAP, total radical-trapping antioxidant potential; WHO, World Health Organization.

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Received 12 October 2009; Received in revised form 28 April 2010; Accepted 29 April 2010

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doi:10.1016/j.fbp.2010.04.008

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1. Introduction

The adverse effects of oxidative stress on human health have become a serious issue. The World Health Organization (WHO) has estimated that 80% of the earth's inhabitants rely on traditional medicine for their primary health care needs, and most of this therapy involves the use of plant extracts and their active components (Winston, 1999). Under stress, our bodies produce more reactive oxygen species (ROS) (e.g., superoxide anion radicals, hydroxyl radicals and hydrogen peroxide) than enzymatic antioxidants (e.g., superoxide dismutase (SOD), glutathione peroxidase (GPx), and catalase) and non-enzymatic antioxidants (e.g., ascorbic acid (vitamin C), α -tocopherol (vitamin E), glutathione, carotenoids, and flavonoids). This imbalance leads to cell damage (Aruoma, 1998; Lefer and Granger, 2000; Smith et al., 2000; Bhatia et al., 2003; Peuchant et al., 2004) and health problems (Steer et al., 2002; Uchida, 2000). A lack of antioxidants, which can quench the reactive free radicals, facilitates the development of degenerative diseases (Shahidi et al., 1992), including cardiovascular diseases, cancers (Gerber et al., 2002), neurodegenerative diseases, Alzheimer's disease (Di Matteo and Esposito, 2003) and inflammatory diseases (Sreejayan and Rao, 1996). One solution to this problem is to supplement the diet with antioxidant compounds that are contained in natural plant sources (Knekt et al., 1996). These natural plant antioxidants can therefore serve as a type of preventive medicine.

Recent reports indicate that there is an inverse relationship between the dietary intake of antioxidant-rich foods and the incidence of human disease (Sies, 1993). However, synthetic antioxidants, such as butylated hydroxytoluene (BHT) and butylated hydroxyanisole (BHA), have been widely used as antioxidants in the food industry and may be responsible for liver damage and carcinogenesis (Grice, 1988; Wichi, 1986).

For this reason, interest in the use of natural antioxidants has increased.

Plants have been the basis of traditional medicines throughout the world for thousands of years and continue to provide new remedies to humankind; a great deal of effort has therefore focused on using available experimental techniques to identify natural antioxidants from plants. Several authors have reviewed the beneficial uses of these plant species (Speroni and Scartezzini, 2000; Matkowski, 2008). Recently, Ali et al. (2008) reviewed twenty-four medicinal Indian herbs that have great antioxidant potential. This review covers medicinal species from a variety of countries (Africa, Algeria, The United States of America, Australia, Brazil, Bulgaria, China, India, Iran, Italy, Japan, Malaysia, Poland, Portugal, Thailand and Turkey). The purpose of this review is to survey the antioxidant capacity and the total phenolic content of medicinal plants from around the world and to evaluate potential sources of natural antioxidants for food and medicinal purposes.

2. An overview of the assay methods used to estimate antioxidant content

Antioxidants, including phenolic compounds (e.g., flavonoids, phenolic acids and tannins), have diverse biological effects, such as anti-inflammatory, anti-carcinogenic and anti-atherosclerotic effects, as a result of their antioxidant activity (Chung et al., 1998). The antioxidant extracts were evaluated in terms of their total phenols (TP), total flavonoids (TFA), total flavonols (TFO), phenolic acids, catechins, lignans and tannins (Cai et al., 2004; Djeridane et al., 2006).

The antioxidant properties were evaluated using the following methods: 1,1-diphenyl-2-picrylhydrazine (DPPH) radical scavenging assay (Blois, 1958; Hatano et al., 1988; Navarro et al., 1992; Brand-Williams et al., 1995; Cotelle et al.,

1996), β -carotene linoleic acid bleaching assay (Miller, 1971; Koleva et al., 2002; Siddhuraju and Becker, 2003), inhibition of linoleic acid peroxidation (Osawa and Namiki, 1981), ferric reducing antioxidant power (FRAP) (Oyaizu, 1986; Benzie and Strain, 1996; Benzie and Szeto, 1999), total radical trapping antioxidant potential (TRAP) assay (Lissi et al., 1992; Krasowska et al., 2001; Leontowicz et al., 2002), oxygen radical absorbance capacity (ORAC) assay (Ou et al., 2001; Huang et al., 2002; Silva et al., 2007), 15-lipoxygenase inhibition (Lyckander and Malterud, 1992), lipid peroxidation (LPO) method (Ohkawa et al., 1979; Ramos et al., 2001), nitro blue tetrazolium (NBT) reduction assay or superoxide anion scavenging activity (Beauchamp and Fridovich, 1971; Nishimiki et al., 1972; Kirby and Schmidt, 1997), hydroxyl radical scavenging activity (Halliwell et al., 1987; Chung et al., 1997; Jodynis-Liebert et al., 1999) or non-site- and site-specific deoxyribose degradation assay (Halliwell et al., 1987; Arouma et al., 1987; Maulik et al., 1997), hydrogen peroxide scavenging activity (Ruch et al., 1989), enzymatic and non-enzymatic *in vitro* antioxidant assay, 2,2'-azinobis(3-ethylbenzthiazoline-6-sulphonic acid) (ABTS) radical scavenging method (Rice-Evans and Miller, 1994; Re et al., 1999; Baltrusaityte et al., 2007), reducing power assay (Oyaizu, 1986), 50% inhibition of a particular assay (IC_{50}), Briggs Rauscher (BR) method (Cervellati et al., 2002), Trolox equivalent antioxidant capacity (TEAC) method (Salah et al., 1995; Campos and Lissi, 1996; Rice-Evans et al., 1996; Re et al., 1999), phenazine methosulfate–nicotinamide adenine dinucleotide-reduced (PMS-NADH) system superoxide radical scavenging (Lau et al., 2002), linoleic acid peroxidation, ammonium thiocyanate (ATC) method (Masude et al., 1992), ferric thiocyanate (FTC) method, thiobarbituric acid (TBA) method (Mackeen et al., 2000) and luminol-photochemiluminescence (PCL) assay. Similarly, the phenolic concentration was determined using the Folin-Ciocalteu (FTC) method (Singleton and Rossi, 1965; Slinkard and Singleton, 1977; Singleton et al., 1999), while the total phenol content (Lamaison et al., 1991; Singleton et al., 1999; Djeridane et al., 2006), the total flavonoid content (Dai et al., 1995; Moreno et al., 2000; Sakanaka et al., 2005), the tannin content (Hagerman and Butler, 1978) and the total flavanol content (Butler et al., 1982) were also determined by known methods.

Although many methods are available to determine antioxidant activity, it is important to employ a consistent and rapid method. While each method has its own merits and drawbacks, it has been found that the most common and reliable methods are the ABTS and DPPH methods; these have been modified and improved in recent years.

2.1. DPPH method

The 1,1-diphenyl-2-picrylhydrazine (DPPH) radical scavenging assay was first described by Blois in 1958 and was later modified slightly by numerous researchers. It is one of the most extensively used antioxidant assays for plant samples. DPPH is a stable free radical that reacts with compounds that can donate a hydrogen atom. This method is based on the scavenging of DPPH through the addition of a radical species or an antioxidant that decolourizes the DPPH solution. The antioxidant activity is then measured by the decrease in absorption at 515 nm. In this method, a 0.1 mM solution of DPPH in methanol is prepared, and 4 ml of this solution are added to 1 ml of the sample solution in methanol at varying concentrations. Thirty minutes later, the absorbance was measured at 517 nm. A large decrease in the absorbance of the reaction mixture

indicates significant free radical scavenging activity of the compound.

2.2. ABTS method

The ABTS radical scavenging method was developed by Rice-Evans and Miller in 1994 and was then modified by Re et al. in 1999. The modification is based on the activation of metmyoglobin with hydrogen peroxide in the presence of ABTS^{•+} to produce a radical cation. This improved method generates a blue/green ABTS^{•+} chromophore via the reaction of ABTS and potassium persulfate and is now widely used. Along with the DPPH method, the ABTS radical scavenging method is one of the most extensively used antioxidant assays for plant samples.

The ABTS radical cation is generated by the oxidation of ABTS with potassium persulfate, and its reduction in the presence of hydrogen-donating antioxidants is measured spectrophotometrically at 734 nm. This decolourisation assay measures the total antioxidant capacity in both lipophilic and hydrophilic substances. The effect of the antioxidant concentration and the duration of the inhibition of the radical cation's absorption are taken into account when the antioxidant activity is determined. Trolox, a water-soluble analog of Vitamin E, is used as a positive control. The activity is expressed in terms of the Trolox-equivalent antioxidant capacity of the extract (TEAC/mg).

2.3. ORAC assay

The ORAC assay uses beta-phycoerythrin (PE) as an oxidizable protein substrate and 2,2'-azobis(2-amidinopropane) dihydrochloride (AAPH) as a peroxy radical generator or a Cu^{2+} - H_2O_2 system as a hydroxyl radical generator. To date, it is the only method that takes the free radical reaction to completion and uses an area-under-the-curve (AUC) technique for quantification, thereby combining both the inhibition percentage and the length of the inhibition time of the free radical's action into a single quantity. The assay has been widely used in many recent studies of plants.

2.4. PCL assay

The PCL assay measures the antioxidant capacity of a compound against the superoxide radical in lipid (ACL) and aqueous (ACW) phases. This method allows the quantification of the antioxidant capacity of both hydrophilic and lipophilic substances either as pure compounds or as a component in a complex matrix from various origins, including synthetic, vegetable, animal, or human sources. The PCL method is based on an approximately 1000-fold acceleration of the oxidative reactions *in vitro* when compared to normal conditions because of the presence of an appropriate photosensitizer. The PCL method is a very quick and sensitive method of measurement. Using the PCL assay, researchers have determined the antioxidant properties of marigold flowers.

2.5. β -Carotene linoleic acid bleaching assay

The β -carotene linoleic acid bleaching assay was first described by Miller (1971) and is one of the antioxidant assays suitable for plant samples. In this assay, the antioxidant capacity is determined by measuring the inhibition of the production of volatile organic compounds and the formation of

conjugated diene hydroperoxides arising from linoleic acid oxidation, which results in the discolouration of β -carotene. β -Carotene (0.5 mg) in 1 ml of chloroform is added to 25 μ l of linoleic acid and 200 mg of the Tween 40 emulsifier mixture. After evaporation of the chloroform under vacuum, 100 ml of oxygen-saturated distilled water is added with vigorous shaking. Next, 4 ml of this mixture is transferred into test tubes containing different concentrations of the sample. As soon as the emulsion is added to each tube, the zero time point absorbance is measured at 470 nm using a spectrophotometer. The emulsion is incubated for 2 h at 50 °C. A blank, devoid of β -carotene, is prepared for background subtraction. Quercetin, BHT and α -tocopherol are used as standards.

2.6. Reducing power assay

The reducing power of the samples is determined according to the method described by Oyaizu (1986). The sample in 1 ml of methanol is mixed with a phosphate buffer (5 ml, 0.2 M, pH 6.6) and potassium ferricyanide (5 ml, 1%), and the mixture is incubated at 50 °C for 20 min. Next, 5 ml of trichloroacetic acid (10%) are added to the reaction mixture, which is then centrifuged at 3000 RPM for 10 min. The upper layer of the solution (5 ml) is mixed with distilled water (5 ml) and ferric chloride (1 ml, 1%), and the absorbance is measured at 700 nm. A stronger absorbance indicates increased reducing power.

2.7. NBT assay or the superoxide anion scavenging activity assay

The superoxide anion scavenging activity assay was first described by Beauchamp and Fridovich (1971). The scavenging potential for superoxide radicals is analysed with a hypoxanthine/xanthine oxidase-generating system coupled with a nitroblue tetrazolium (NBT) reduction (measured spectrophotometrically). The reaction mixture contains 125 μ l of buffer (50 mM $\text{KH}_2\text{PO}_4/\text{KOH}$, pH 7.4), 20 μ l of a 15 mM Na_2EDTA solution in buffer, 30 μ l of a 3 mM hypoxanthine solution in buffer, 50 μ l of a 0.6 mM NBT solution in buffer, 50 μ l of xanthine oxidase in buffer (1 unit per 10 ml buffer) and 25 μ l of the plant extract in buffer (a diluted, sonicated solution of 10 μ g per 250 μ l buffer). Microplates (96 wells) are read at 540 nm 2.5 min after the addition of the xanthine oxidase using the series 7500 Microplate Reader. The superoxide scavenging activity is expressed as percent inhibition compared to the blank, in which buffer is used in place of the extract. When using this system, any inhibition by tannins in the plant extracts would have to be due to their antioxidant activity and not to their action upon the enzyme.

2.8. Total flavonoid content

Total flavonoid content has been discussed by several authors (Dai et al., 1995; Moreno et al., 2000; Sakanaka et al., 2005). The measurement of an extract's flavonoid concentration is based on the method described by Moreno et al. (2000) with a slight modification, and the results are expressed as quercetin equivalents. An aliquot of 1 ml of a methanol solution containing 1 mg of extract is added to test tubes containing 0.1 ml of 10% aluminium nitrate, 0.1 ml of a 1 M potassium acetate solution and 3.8 ml of methanol. After 40 min at room temperature, the absorbance is measured at 415 nm. Quercetin is used as a standard.

2.9. Folin-Ciocalteu method

The Folin-Ciocalteu reagent assay is used to determine the total phenolics content (Singlenton and Rossi, 1965). The sample (0.2 ml) is mixed with 0.5 ml of the Folin-Ciocalteu reagent previously diluted with 7 ml of deionised water. The solution is allowed to stand for 3 min at 25 °C before 0.2 ml of a saturated sodium carbonate solution is added. The mixed solution is allowed to stand for another 120 min before the absorbance at 725 nm is measured. Gallic acid is used as a standard for the calibration curve. The total phenolics content is expressed as mM gallic acid equivalents (GAE) per l of sample (mM/l).

3. Medicinal species with high antioxidant potential

Several authors have also reviewed medicinal species with great antioxidant potential (Speroni and Scartezzini, 2000; Matkowski, 2008; Ali et al., 2008). In this work, we have reviewed the antioxidant potential of a number of additional plants.

3.1. Diospyros abyssinica

Diospyros species have been used in many traditional medical systems around the world, including traditional Ayurvedic, African and Chinese medicine. Nearly every part of these plants has been used as a medicine in some way, for example as an astringent remedy and to cure biliousness (Mallavadhani et al., 1998). In India, a juice made from the bark and leaves of *Diospyros peregrine* combined with the root juice of *Albizia lebbek* is used as a remedy for snakebites. In Japan, the leaves of *Diospyros kaki* are used in combination with jasmine to make anti-smoking candies (Mallavadhani et al., 1998).

The most frequently-isolated compounds from *Diospyros abyssinica* are the triterpenoids betulin, betulinic acid and lupeol (Zhong et al., 1984; Recio et al., 1995). All of these compounds are well-known anti-inflammatory compounds. This species has a significant medicinal value demonstrated by its use in traditional medicine.

The root bark from *D. abyssinica* has been tested regarding its antioxidant activity (Maiga et al., 2006). It was extracted with a series of solvents, including petroleum ether, dichloromethane, chloroform, 80% aqueous ethanol, and water (at 50 °C and 100 °C). It was determined that the root bark from *D. abyssinica* is the richest source of extracted compounds; 36.7% of the weight of the plant material is composed of antioxidants. *D. abyssinica* exhibited the greatest radical scavenging activity and the greatest 15-lipoxygenase inhibition in the 80% ethanol and methanol extracts. Thus, this plant appears to be an excellent source of antioxidants (Maiga et al., 2006).

3.2. Pistacia lentiscus

Pistacia lentiscus is extensively used in folk medicine by rural populations in Algeria. Algeria is home to at least 3164 species of vascular plant, of which 7.9% are endemic. *P. lentiscus* is important because of its medicinal value. The reducing power and radical scavenging activity of the extracts from the leaves of *P. lentiscus* in solvents, such as ethanol, ethyl acetate, aqueous/ethyl acetate, hexane, aqueous/hexane, chloroform, and

aqueous chloroform has been studied *in vitro* (Atmani et al., 2009).

Using the DPPH scavenging activity assay, it was found that all of the *P. lentiscus* extracts, except for the chloroform extract, have a high radical scavenging activity (90%) equivalent to that of the standard, BHA (89%). The ethanolic and aqueous fractions from the ethyl acetate extract have high scavenging activities with values of $78 \pm 0.93\%$ and $90.29 \pm 0.29\%$, respectively. Overall, *P. lentiscus* exhibited outstanding reducing power, good radical scavenging activity against DPPH and H_2O_2 , slow inhibition of lipid peroxidation and richness in tannins; however, it also showed a lack of flavonoids (Atmani et al., 2009).

A strong correlation was found between reducing power and the total amount of phenols present in *P. lentiscus*, indicating that the phenol compounds play an important role in the beneficial effects of these medicinal plants. This finding is in agreement with the work of Chryssavgi et al. (2008), which demonstrated that the greatest phenolic content in *P. lentiscus* is 588 mg gallic acid/g of plant material and consists mainly of monoterpenes (81.6%).

3.3. *Geranium sanguineum* L

Geranium sanguineum L., commonly found in Bulgaria, has significant antioxidant activity and antiviral activity (Serkedjieva and Manolova, 1992). Its root extracts are used in traditional medicine to treat gastrointestinal disorders, infections and inflammatory conditions. It is also frequently used in folk medicine for the treatment of eruptive skin diseases and as a disinfectant bath and poultice for the affected area.

The polyphenolic compounds of this plant species include tannins (11.02%), flavonoids (0.14%), catechins and proanthocyanidines (2.1 mg/kg) (Ivancheva et al., 1992). Using three separate, complementary methods (the DPPH assay, the β -carotene-linoleic acid assay and the NBT-reduction assay), it was established that a polyphenol-rich extract from *G. sanguineum* L., which had a strong anti-influenza activity, possessed antioxidant and radical scavenging capacities. In this study, caffeic acid and the synthetic antioxidant BHT were used as positive controls.

The root extract of this plant exhibited a strong antioxidant capacity in the DPPH assay ($IC_{50} = 13.86 \pm 0.84 \mu\text{g/ml}$) when compared to BHT ($IC_{50} = 19.81 \pm 0.05 \mu\text{g/ml}$). In the β -carotene-linoleic acid test system, the root extract achieved 88–89% inhibition, which is as strong as BHT's inhibition. Furthermore, the total extract and ethyl acetate fraction exhibited a strong superoxide dismutase (SOD) activity, comparable to that of caffeic acid. In addition, the total methanol-soluble phenolic constituents were measured with the Folin-Ciocalteu reagent and were found to be 34.6% (w/w) (Sokmen et al., 2005). In another study, it was found that *G. sanguineum* L. reduced the accumulation of TBA-reactive products in rat liver microsomes *in vivo* in the induced LPO method, but the non-induced LPO method was not affected (Murzakhmetova et al., 2008).

3.4. *Sargentodoxa cuneata* Rehd. Et Wils

In the classification of Chinese medicinal plants, *Sargentodoxa cuneata* Rehd. Et Wils falls into the “heat-clearing” category. The plants in this category have significant anti-inflammatory, anti-tumour, anti-allergic, anti-viral and anti-bacterial activities.

China is the only country on Earth in which there are unbroken connections among tropical, subtropical, temperate and boreal forests. This unbroken connection has fostered the formation of rich plant associations rarely seen elsewhere in the world. China's plant life is enormously rich. Some 31,000 plant species are native to China, representing nearly one-eighth of the world's total plant species, including thousands found nowhere else on Earth. Chinese medicinal plants contain a wide variety of natural antioxidants, such as phenolic acids, flavonoids and tannins and possess more potent antioxidant activity than common dietary plants (Cai et al., 2004; Dragland et al., 2003). Sargentol, tyrosol, salidroside, methylprotocatechuate, vanillic acid, syringic acid, *p*-hydroxy benzoic acid, and ferulic acid have been identified in this plant (Li et al., 2008).

Using the FRAP and TEAC assays, *S. cuneata* Rehd. Et Wils was found to have the greatest antioxidant capacity with $453.53 \mu\text{mol Fe(II)/g}$ and $265.43 \mu\text{mol Fe(II)/g}$, respectively. In addition, *S. cuneata* Rehd. Et Wils had the highest phenolic content (52.35 mg GAE/g). A strong correlation was also found between the TEAC and FRAP values, which implies that the extracts from this plant are capable of scavenging free radicals and reducing antioxidants (Li et al., 2008). This study concludes that this medicinally-important species is a valuable source of natural antioxidants, both for the preparation of crude extracts and for the further isolation and purification of antioxidant components.

3.5. *Polyalthia cerasoides* (Roxb.) Bedd

Polyalthia cerasoides (Roxb.) Bedd. (*Annonaceae*) is a medium-sized tree distributed in almost all of the forests of Deccan India at elevations of up to 3000 ft. India is one of the richest countries in the world with respect to medicinal and aromatic plants. The plant life of India constitutes 11% of the world's total known flora that have medicinal properties. The number of plant species in India is estimated to be over 45,000.

The tribal people of Tamil Nadu and Andhra Pradesh (states of India) use the fruits of this plant, while tribes in Africa use the fruits, roots and leaves to treat rheumatism and toothaches, as an aphrodisiac, as a deparasitant and as an anti-inflammatory. Pharmacological studies confirmed that the stem bark of *P. cerasoides* reduces brain stress.

The antioxidative potential of the alcohol extract of *P. cerasoides* was evaluated using the DPPH, hydroxyl radical, superoxide anion scavenging, and reducing power assays. The methanol extract of *P. cerasoides* exhibited a significant dose-dependent inhibition of DPPH scavenging activity with 50% inhibition occurring at a concentration equivalent to $25 \mu\text{g/ml}$ of tannic acid. The total phenolic content of the alcohol extract of *P. cerasoides* was equivalent to $0.589 \mu\text{g}$ of tannic acid per mg of extract (Ravikumar et al., 2008). The phenolic compounds present in the extract may contribute directly to the antioxidative action of the plant, suggesting that the polyphenols present in the extract could be responsible for its beneficial effects.

3.6. *Crataeva nurvala* Buch-Ham

Crataeva nurvala Buch-Ham. is used extensively in traditional medicine as a blood purifier. The bark of *C. nurvala* is used in herbal powders to treat urinary stones, thyroid disorders, obesity and cancer.

C. nurvala has a higher total antioxidant capacity than catechin (Kumari and Kakkar, 2008). In this study,

C. nurvala showed the highest SOD mimetic activity (122.53 unit/min/mg), which was determined spectrophotometrically by measuring inhibition in the nicotinamide adenine dinucleotide (reduced)-phenazine methosulfate-nitroblue tetrazolium reaction system. *C. nurvala* was found to have the highest LPO inhibitory potential. *C. nurvala* was more efficient at scavenging peroxide radicals than catechin.

Using the ABTS assay, the total antioxidant potential of *C. nurvala* was found to be 0.39 mmol/l TEAC/mg of extract. In addition, a study of this plant's phytochemicals revealed that the stem bark of *C. nurvala* contained triterpenoids such as phragmalin triacetate and lupeol.

3.7. *Acacia auriculiformis* A. Cunn

Acacia auriculiformis A. Cunn is a vigorously-growing deciduous or evergreen tree. It can reach heights of up to 30 m and belongs to the family *Mimosaceae*. It is rich in methylglucuronic acid, glucuronic acid, galactose, arabinose, and rhamnose.

Tannins and triterpenoid saponins are present in the species (Parkashi et al., 1991; Ghosh et al., 1993; Garai and Mahato, 1997). In addition, extracts from the *Acacia* species are rich in phenols and polyphenols and have strong antimutagenic and antioxidant activities (Kaur et al., 2002, 2005; Singh et al., 2004).

The hydroxyl radical scavenging potency of the extracts of *A. auriculiformis* increased with solvent polarity and was greatest in the water fraction, followed by the ethyl acetate fraction, and the crude extract. The water fraction had a higher phenolic content (720 mg) than the ethyl acetate fraction (600 mg) or the crude ethyl acetate extract (390 mg) when expressed as GAE/g of extract/fraction (Singh et al., 2007).

3.8. *Teucrium polium* L

Teucrium polium L. is a wild flower species belonging to the *Lamiaceae* family, which is composed of numerous species with exploitable antioxidant activity (Del Bano et al., 2003). An infusion of the leaves and flowers of the plant is consumed as a refreshing beverage. This infusion is also used for liver ailments, gastrointestinal diseases, fevers, colds, diarrhoea, stomach pains and fevers.

In this study, the aerial part of the plant was extracted with petroleum ether, chloroform, methanol and water. The antioxidant flavonoids were separated from the methanol extract and were identified as rutin, apigenin, 3',6-dimethoxy apigenin and 4',7-dimethoxy apigenin; their IC₅₀ values in the DPPH assay were found to be 23.7 ± 1.9 µg/ml, 30.3 ± 2.1 µg/ml, 31.5 ± 3.4 µg/ml and 37.4 ± 3.4 µg/ml, respectively. The DPPH assay IC₅₀ value for the methanol extract was found to be 20.1 ± 1.7 µg/ml; this value is similar to the IC₅₀ value of the synthetic antioxidant, butylated hydroxytoluene (18.3 ± 1.9 µg/ml). The potential antioxidant activity and the rich flavonoid content of *T. polium* suggests that its extracts may be added to various food products in place of synthetic antioxidants (Shariffar et al., 2009).

The aqueous extract of *T. polium* can effectively inhibit oxidative processes and has substantial antioxidant activity *in vitro* (Ljubuncic et al., 2006). The ethanol extract prepared from *T. Polium* exhibited the same antioxidant activity as α-tocopherol. The antioxidant activity of *T. polium* was also demonstrated in a recent *in vivo* study of rats. Rats were treated with a *T. polium* extract that showed significant antioxidant activity in the DPPH test compared to the positive control (α-

tocopherol). The *T. polium* extract given to rats at doses of 50 and 100 mg/kg significantly increased the total antioxidant power (TAP) and decreased the thiobarbuteric acid reactive substances (TBARS) relative to the control (Hasani et al., 2007).

3.9. *Dracocephalum moldavica* L

The Moldavian balm (*Dracocephalum moldavica* L., *Lamiaceae*) is a perennial herb that is native to central Asia and is naturalised in eastern and central Europe. It is used as a food ingredient, a tea, and as an herbal drug used to treat stomach and liver disorders, headaches and congestion (Rechinger, 1986).

The antioxidant activity of this species has been studied by several researchers (Povilaityte and Venskutonis, 2000; Povilaityte et al., 2001; Dastmalchi et al., 2007). Dastmalchi et al. (2007) suggested that the components responsible for its activity were hydroxycinnamic acids and flavonoids, including caffeic acid, ferulic acid, rosmarinic acid, luteolin, luteolin-7-O-glucoside and apigenin. The extract yields ranged from 3.7 mg/g in the ethyl acetate and n-butanol extracts to 109.2 mg/g in the methanol extract; they increased in the following order: ethyl acetate and n-butanol, acetonitrile (ACN), dichloromethane, petrol, water and methanol. The total phenolic content of the extracts ranged from 0.0 ± 0.0 mg GA/g in the petrol extract to 488.4 ± 1.8 mg GA/g in the methanol extract and increased in the following order: petrol, dichloromethane, ACN, ethyl acetate, water, n-butanol and methanol.

The HPLC-determined total phenolic content of the raw material was found to be 476.59 ± 25.22 mg/g (sum of the individual extracts). Rosmarinic acid was the most abundant component identified (247.95 ± 24.78 mg/g), followed by chlorogenic acid (41.46 ± 2.76 mg/g) and apigenin-7-O-glucoside (26.55 ± 2.20 mg/g). The greatest quantities of phenolic substances were found in the n-butanol (39%), methanol (31.1%) and water (11.5%) fractions (Dastmalchi et al., 2007).

3.10. *Urtica dioica* L

Urtica dioica L. (*Urticaceae*) leaves have been used in Sardinia, Italy as a medicinal tea or decoction as diuretic and anti-diabetic therapies and to treat stomach disorders. The flora of Italy is the richest in Europe. As of 2004, 6759 species had been recorded in the data bank of Italian vascular flora, of which 700 are endemic. *U. dioica* L. leaves are also used to treat stomachaches in Turkish folk medicine (Yesilada et al., 1993).

The antioxidant capacity of this plant was evaluated using several *in vitro* methods (BR, TEAC, DPPH, and FC). The BR method determined that the antioxidant activity of *U. dioica* at an acidic pH was 0.013 ± 0.001 µg/ml resorcinol equivalents (Re eq.); the DPPH method in methanolic solutions determined that the antioxidant activity was 419 ± 10 µg/ml. The total phenolic content was found to be 0.35 ± 0.02 mg/l GAE (Dall'Acqua et al., 2008).

Concentrations of *U. dioica* L. extract of 50, 100 and 250 µg/ml showed 39%, 66% and 98% inhibition, respectively, of the peroxidation of a linoleic acid emulsion. However, α-tocopherol, positive control, at 60 µg/ml, exhibited only 30% inhibition (Gulcin et al., 2004). It can be concluded that *U. dioica* L. has powerful antioxidant activities.

3.11. *Ficus microcarpa* L. fil

Ficus microcarpa L. fil. (Chinese banyan tree, *Moraceae*) is a popular ornamental tree grown widely in many tropical regions. It is native to areas including Ceylon, India, southern China, the Ryukyu Islands, Australia and New Caledonia. It is also a popular ornamental plant in Taiwan (Chiang et al., 2005). Its dried leaves, aerial roots and bark have been used as folk remedies to decrease perspiration, alleviate fever and relieve pain in the Okinawa Islands. There are about 7000 species of vascular plants in Japan, and about 40% of these, approximately 2900 species, are recognized as endemic.

In the study, two isoflavones comprised of 28 components were identified in the bark of the *F. microcarpa* tree (Kuo and Li, 1997). In addition, the methanol extracts of this tree's bark, fruits and leaves exhibited strong antioxidant activity when assayed by the DPPH method, the ABTS free radical scavenging method, the PMS-NADH system superoxide radical scavenging assay and the β -carotene-linoleic acid system. The methanol extract of the bark showed stronger antioxidant activity than the extracts of the leaves or fruits in the ABTS method, the PMS-NADH method and the β -carotene-linoleic acid system. However, no significant difference was found between the bark and the fruits in the DPPH assay. Furthermore, the bark contained a significantly higher amount of total phenolics (237 mg GAE/g extract) than the fruits (179 mg GAE/g extract) or the leaves (127 mg GAE/g extract).

Furthermore, the total phenolics in the bark were present in greater amounts in the ethyl acetate fraction than in the aqueous fraction and the hexane fraction; the values were 436, 194 and 41.7 mg GAE/g extract, respectively. The ethyl acetate fraction contained twelve phenolic compounds, of which seven were quantified by HPLC: protocatechuic acid (6.60 ± 0.20 mg/g extract), catechol (11.1 ± 0.00 mg/g extract), *p*-vinylguaiacol (4.40 ± 0.07 mg/g extract), syringol (173 ± 1.12 mg/g extract), *p*-propylphenol (10.5 ± 0.78 mg/g extract), vanillin (4.27 ± 0.02 mg/g extract) and syringaldehyde (8.96 ± 0.29 mg/g extract) (Ao et al., 2008).

3.12. *Bidens pilosa* Linn. *Radiata*

Bidens pilosa Linn. *Radiata* (family *Asteraceae*) is widely distributed in subtropical and tropical regions. It is 30–100 cm in height with yellow flowers and is commonly known as “hairy beggar ticks,” “sticks tight,” and “Spanish needles.” The plant is used in various folk medicines for its anti-inflammatory, antiseptic, liver-protective, blood-pressure lowering, and anti-hypoglycaemic effects (Dimo et al., 2002). The plant has been widely used in Taiwan as a traditional medicine and as a major ingredient of an herbal tea that is believed to prevent inflammation and cancer (Yang et al., 2006).

Phenylpropanoid glucosides, polyacetylenes, diterpenes, flavonoids and flavone glycosides have been identified as the bioactive components of this plant and are thought to be involved in its antioxidant activity (Chiang et al., 2004). The methanol extract of *B. pilosa* was shown to prevent the onset of hypertension and to reduce blood pressure in rats (Dimo et al., 2002). In addition, the fresh leaves and flowers of *B. pilosa* were subjected to steam-distillation, and colourless and yellowish essential oils were obtained in amounts of 0.08% and 0.06% (w/w), respectively. GC-MS analysis of these essential oils resulted in the identification of forty-four com-

pounds including the major essential oils, β -caryophyllene (10.9% and 5.1% in the leaves and flowers, respectively) and τ -cadinene (7.82% and 6.13% in the leaves and flowers, respectively). Both of these essential oils are terpenes. The other chemical components were α -pinene, limonene, β -trans-ocimene, β -cis-ocimene, τ -muurolene, β -bourbonene, β -elemene, β -cubebene, α -caryophyllene, caryophyllene oxide and megastigmatrienone.

The essential oils in the leaves and flowers were able to reduce the stable free radical DPPH to the yellow coloured diphenylpicrylhydrazine with IC_{50} s of 57 and 50 μ g/ml, respectively, whereas the synthetic and natural antioxidant activities were 21 and 36 μ g/ml, respectively. This study revealed that the flowers of *B. pilosa* have an antioxidant activity that is similar to that of synthetic antioxidants. In addition, the aqueous extracts of the flowers and leaves were found to be less efficient in radical scavenging and had IC_{50} values of 172 μ g/ml and 61 μ g/ml, respectively. Furthermore, the essential oils of the leaves and the aqueous extracts of the leaves and flowers exhibited higher antioxidant activities than did the flower oils. The lower activity of the essential oils of *B. pilosa*'s flowers may be due to their volatility at higher temperatures. The study showed that the antioxidant effects of essential oils depend not only on the temperature but also on other factors such as their structural features, the characteristics of the lipid system, and the binding of the fatty acids (Deba et al., 2008).

3.13. *Leea indica*

Leea indica, a member of the *Leeaceae* family, was studied for its antioxidant and nitric oxide inhibitory properties because of its traditional use for various medicinal purposes. It is commonly found in Malaysia. Malaysian tropical rainforests contain many species that are important sources of traditional medicines. About 10,000 species of higher plants and 2000 species of lower plants are available in Peninsular Malaysia; 16% of these are used for traditional medicinal purposes (Latif et al., 1984).

The leaves of *Leea indica* contain 23 relevant chemical compounds, including eleven hydrocarbons, phthalic acid, palmitic acid, 1-eicosanol, solanesol, farnesol, three phthalic acid esters, gallic acid, lupeol, beta-sitosterol and ursolic acid (Srinivasan et al., 2008).

This study used the FTC and TBA methods to demonstrate that methanol extracts of *Leea indica* had strong antioxidant activity that is comparable to, or higher than, that of α -tocopherol, BHT and quercetin. Saha et al. (2004) also confirmed that extracts from *Leea indica* had strong activity compared with the standards (i.e., vitamin C, quercetin and BHT). The high antioxidant activity of *Leea indica* extracts may be due to the presence of gallic acid.

3.14. *Lamiaceae* species

Six *Lamiaceae* species (i.e., *Leonurus cardiaca*, *Lamium album*, *Marrubium vulgare*, *Stachys officinalis*, *Lamium purpureum* and *Galeopsis speciosa*) are rich in antioxidant activity. *Leonurus cardiac* L. is a mild cardiac drug containing flavonoid and phenolic glycosides.

The chemical composition, therapeutic uses and pharmacological properties of these species have been reported. *Lamium album* L. (dead nettle) has antispasmodic, diuretic and haemostatic properties and is used to alleviate bladder, kidney and menstrual problems. *Lamium purpureum* L. is used

for similar medicinal purposes. *Marrubium vulgare* L., which contains diterpenoids, iridoids, flavonoids and terpenoid, is used to treat coughs and digestive disorders. *Stachys officinalis* Franch. is used for antiseptic, astringent, tonic, anthelmintic and digestive purposes. *Galeopsis speciosa*, which contains tannins, flavonoids, soluble silica and saponins, is used as an astringent, diuretic and expectorant.

The antioxidant activities of these six species have been studied by several authors (Mantle et al., 2000; Trouillas et al., 2003; Vander Jagt et al., 2002), but it is difficult to compare their results because of the methodological differences between the studies (Matkowski and Piotrowska, 2006). In addition, the antioxidative effects of the methanolic extracts from six wild European *Lamiaceae* species have been studied using three *in vitro* assays.

In the DPPH scavenging assay, the order of these species from strongest to weakest antioxidant activity was: *Leonurus cardiaca*, *Lamium album*, *Marrubium vulgare*, *Stachys officinalis*, *Lamium purpureum* and *Galeopsis speciosa*. In the LPO assay, *S. officinalis* and *M. vulgare* reached a maximum inhibition of 78%, *Lamium sp.* and *L. cardiac* slightly exceeded 70% while *G. speciosa* reached 65%. All of the extracts contained a considerable quantity of phenolic metabolites, ranging from 13.2% GAE in *S. betonica* to 20% in *L. cardiaca* (Matkowski and Piotrowska, 2006). *L. cardiaca* has significant antioxidant potential, as demonstrated by several authors (Mantle et al., 2000; Trouillas et al., 2003; Vander Jagt et al., 2002).

3.15. *Uncaria tomentosa* (Willd.) DC

Uncaria tomentosa (Willd.) DC., commonly known as cat's claw, belongs to the family *Rubiaceae* and is found in South and Central America. It is used for the treatment of asthma, cancer, cirrhosis, fevers, gastritis, diabetes, dysentery and inflammation of the urinary tract (Keplinger et al., 1999; Falkiewicz and Lukasiak, 2001; Heitzman et al., 2005). In addition, it is used as an anticancer remedy and has anti-inflammatory properties (Aguilar et al., 2002). Due to the chemical structure of its components, this plant is expected to have strong antioxidant activity (Deschmarchelier et al., 1997).

The active chemical constituents of this species are alkaloids, quinoic acid, glycosides, polyhydroxylated triterpenes and several steroidal components. The antioxidant properties of the aqueous and ethanolic extracts of *U. tomentosa* bark have been evaluated. A higher antioxidant activity and greater number of total phenolic compounds were detected in the alcoholic preparations (TEAC=0.57 mmol of Trolox/g and SOD=0.39 U/mg) than in the aqueous preparations (TEAC=0.34 mmol of Trolox/g and SOD=0.1 U/mg). This study revealed that five pentacyclic oxindole alkaloids, including uncarine F, speciophylline, mitraphylline, isomitraphylline and/or pteropodine and isopteropodine, were present in the bark.

The content of TPC in the ethanol extract from *U. tomentosa* bark (292 mg/g D-catechin units) was two times higher than in the aqueous extract (111 mg/g). These values are very high compared to other TPC-containing cereals (from 0.481 to 0.896 mg/g), vegetables (e.g., 11.7 mg/g for broccoli, 9.9 mg/g for garlic and 7.6 mg/g for pepper) and fruits (e.g., 23.1 mg/g for blackberries) (Vinson et al., 1998; Wang and Lin, 2000). The ethanol extract showed higher superoxide radical scavenging activity (0.39 U/mg) than did the aqueous extract (0.10 U/mg) (Pilarski et al., 2006).

3.16. *Salvia officinalis* L

Common sage (*Salvia officinalis* L., *Lamiaceae*) is an aromatic and medicinal plant of Mediterranean origin commonly found in Portugal and Lithuania and well known for its antioxidant properties that are mainly due to its phenolic-rich composition.

Methanolic and aqueous extracts were prepared from the aerial parts of *S. officinalis* and analysed for phenolic compounds by HPLC/DAD. Eight phenolic compounds were identified, including five phenolic acids (i.e., rosmarinic acid, caffeic acid, ferulic acid, 3-caffeoylquinic acid and 5-caffeoylquinic acid) and three flavonoids (i.e., luteolin-7-glucoside; 4',5,7,8-tetrahydroxyflavone; apigenin-7-glucoside). The methanolic extract had a higher content of these compounds than did the aqueous extract.

The main phenolic compound in the methanolic extract was rosmarinic acid (132.2 µg/mg extract), while the main compounds in the aqueous extract were rosmarinic acid (52.0 µg/mg extract) and luteolin-7-glucoside (19.7 µg/mg extract). The methanolic extract had a higher content of phenolic compounds and a higher anti-radical activity in the DPPH assay (IC₅₀ = 13.5 ± 0.5 µg/ml) and a higher anti-radical efficiency than the aqueous extract, which had an IC₅₀ of 14.9 ± 0.3 µg/ml. The activity of both extracts was lower than the positive control, quercetin. In the superoxide radical scavenging assay, the aqueous extract had a greater anti-radical activity (14.4 ± 1.4 µg/ml) than the methanolic extract (162 ± 39 µg/ml) (Lima et al., 2007). A separate study showed that replacing the drinking water of rats and mice with *S. officinalis* infusions for 14 days led to improved liver antioxidant status (Lima et al., 2005).

3.17. *Momordica Charantia* L

The bitter melon (*Momordica Charantia* L.) or *Mara* (in Thai) belongs to the family *Cucurbitaceae* and has long been used in foods and medicines (El Batran et al., 2006). The bitter melon is known by different names, such as balsam pear and karela, and it grows in tropical and sub-tropical regions of India, Malaysia, China, Africa, the Middle East, USA and Thailand (El Batran et al., 2006). Thailand is home to a wide range of herbal plant species. Medicinal plants and herbs have long been a part of everyday life in Thailand; many are used as spices in various Thai dishes. The therapeutic efficacy of Thai medicinal plants and traditional herbal medications has been scientifically proven and described in the literature by both Thai and non-Thai scientists. The bitter melon can be used to treat diabetes mellitus and appears to be a safe alternative to reduce blood glucose (Virdi et al., 2003).

In the DPPH radical scavenging assay, the activity of the positive control, ascorbic acid, was the highest (200 mg/ml), followed by BHT, the leaf, the green fruit, the stem and the ripe fruit fractions of the bitter melon. The IC₅₀ values were lowest in the leaf fraction (9.72 ± 0.25 mg/ml), followed by the green fruit fraction (11.00 ± 0.76 mg/ml), the stem fraction (17.8 ± 0.66 mg/ml) and the ripe fruit fraction (27.6 ± 0.23 mg/ml). In the hydroxyl radical scavenging assay, the activity of the leaf fraction was greater than that of the other fractions but lower than that of ascorbic acid and BHT. The green fruit had the highest IC₅₀ value (119 ± 0.34 mg/ml), followed by the leaf (167 ± 0.96 mg/ml), the stem (267 ± 0.72 mg/ml) and the ripe fruit (173 ± 0.23 mg/ml). In the β-carotene-linoleate bleaching assay, the antioxi-

Table 1 – Worldwide distribution of medicinal plants with superior antioxidant potential.

| Land mass | Plant species | | | | | | | | | | | | | | | | | | |
|-----------|----------------------|--------------------|-----------------------|------------------------------------|-------------------------------------|----------------------------|--------------------------------|--------------------|----------------------------|------------------|--------------------------|-----------------------------|-------------|-------------------|--------------------------------|-----------------------|------------------------|----------------|----------------------------|
| | Diospyros abyssinica | Pistacia lentiscus | Geranium sanguineum L | Sargentodoxa cuneata Rehd. Et Wils | Polyalthia cerasoides (Roxb.) Bedd. | Crataeva nurvala Buch-Ham. | Acacia auriculi-formis A. Cunn | Teucrium polium L. | Dracocephalum moldavica L. | Urtica dioica L. | Ficus microcarpa L. fil. | Bidens pilosa Linn. Radiata | Leea indica | Lamiaceae species | Uncaria tomentosa (Willd.) DC. | Salvia officinalis L. | Momordica Charantia L. | Rheum ribes L. | Pelargonium endlicherianum |
| Africa | + | | | | + | | | | | | | | | | | | | | + |
| Algeria | | + | | | | | | | | | | | | | | | | | |
| USA | | | | | | | | + | | | | | | | | | | | + |
| Australia | | | | | | | | | | | + | | | | | | | | + |
| Brazil | | | | | | | | | | | | | | | + | | | | + |
| Bulgaria | | | + | | | | | | | | | | | | | | | | |
| China | + | | | + | + | | | | + | + | + | + | + | | | | | | + |
| India | + | | | | + | + | + | | + | + | + | + | + | | | | | | + |
| Iran | | | | | | | | | + | + | + | | | | | | | | + |
| Italy | | + | + | | | | | | + | + | | | | | | + | | | |
| Japan | + | | | | | | | | | | | + | + | | | | | | |
| Malaysia | | | | | | | | | | | + | + | + | | | | | | + |
| Poland | | | + | | | | | | | | | | | | + | | | | |
| Portugal | | + | + | | | | | | | | | | | | | + | | | |
| Thailand | | | | | + | | | | | | + | | | | | | | | + |
| Turkey | | + | + | | | | | | | | | | | | | | | | + |

Table 2 – Extraction methods, main components and antioxidant potential of medicinal plant species.

| S. No. | Species | Extraction method | Solvent(s) used | Main components (or groups) | Antioxidant assay methods | | | | Reference(s) |
|--------|--|--|--|--|--|--|---|---|---|
| 1 | <i>Diospyros abyssinica</i> (root bark) | Soxhlet extraction | Petrol, ether, dichloro methane, chloroform, 80% ethanol, methanol, and water (50 °C and 100 °C) | Triterpenoids, betulin, betulinic acid and lupeol | DPPH assay: (80% ethanol) EC ₅₀ = 16 ± 2 µg/ml | 15-lipoxygenase inhibition (80% ethanol): IC ₅₀ = 21 ± 2 µg/ml | NA | NA | Maiga et al., 2006; Zhong et al., 1984; Recio et al., 1995 |
| 2 | <i>Pistacia lentiscus</i> (leaves) | Solvent extraction | Ethanol, ethyl acetate, aqueous ethyl acetate, hexane, aqueous hexane, chloroform and aqueous chloroform | Monoterpenes | Reducing power assay (aqueous hexane): 0.91 ± 0.03 | DPPH assay (aqueous chloroform): IC ₅₀ = 4.24 µg/ml | Inhibition of linoleic acid peroxidation (aqueous hexane): 98.77% | NA | Atmani et al., 2009; Chiang et al., 1993; Chryssavgi et al., 2008 |
| 3 | <i>Geranium sanguineum</i> L (root) | Solvent extraction | Methanol | Tannins, flavonoids, catechins and proanthocyanidines | DPPH assay: IC ₅₀ = 13.86 ± 0.84 µg/ml | β-carotene- linoleic acid assay: 88–89% inhibition | NBT-reduction assay: IC ₅₀ = 26 µg/ml | NA | Sokmen et al., 2005; Ivancheva et al., 1992 |
| 4 | <i>Sargentodoxa cuneata</i> Rehd. Et Wils (plant) | Hot water extraction | Methanol | Sargentol, tyrosol, salidroside, methylprotocatechuate, vanillic acid, syringic acid, p-hydroxy benzoic acid, and ferulic acid | FRAP assay: 453.53 µmol Fe (II)/g | TEAC assay: 265.43 µmol Fe (II)/g | NA | NA | Li et al., 2008 |
| 5 | <i>Polyalthia cerasoides</i> (Roxb.) Bedd. (stem bark) | NA | Ethanol | NA | DPPH assay: IC ₅₀ = 25 µg/ml | Hydroxyl radical scavenging assay: IC ₅₀ = 50 µg/ml | Superoxide anion scavenging activity: IC ₅₀ = 80 µg/ml | NA | Ravikumar et al., 2008 |
| 6 | <i>Crataeva nurvala</i> Buch-Ham. (stem bark) | Cold reflux | Ethanol | Triterpenoids, such as phragmalin triacetate and lupeol; tannin, saponin, friedelin, and diosgenin | SOD mimetic activity: 122.53 unit/min/mg | LPO inhibitory potential: 83.3% LPO inhibition/10 µg of extract | ABTS assay: 0.39 mmol/l TEAC/mg of extract | NA | Kumari and Kakkar, 2008 |
| 7 | <i>Acacia auriculiformis</i> A. Cunn (bark) | Extraction by maceration of bark powder by increasing and decreasing the order of solvent polarity | Ethyl acetate fraction and water fraction | Tannins and triterpenoid saponins | DPPH assay – water fraction decreasing order of polarity: 67.14% | Reducing power assay – water fraction decreasing order of polarity: 1.717 Fe ³⁺ to Fe ²⁺ | Deoxyribose degradation assay – water fraction increasing order of polarity: 75.63% | TBA assay – Water fraction decreasing order of polarity: 71.62% | Singh et al., 2007; Parkashi et al., 1991; Ghosh et al., 1993; Garai and Mahato, 1997 |
| 8 | <i>Teucrium polium</i> L. (aerial parts) | NA | Methanol | Rutin; apigenin; 3',6-dimethoxy apigenin; 4',7-dimethoxy apigenin | DPPH assay: IC ₅₀ = 20.1 ± 1.7 µg/ml | β-Carotene bleaching test: 25.8 ± 1.2 mm inhibition | NA | NA | Shariffar et al., 2009 |

| | | | | | | | | | |
|----|---|--------------------|---|---|--|---|---|--|---|
| 9 | <i>Dracocephalum moldavica</i> L. (aerial parts) | Soxhlet | Petrol, dichloromethane, acetonitrile, ethyl acetate, methanol, n-butanol and water | Caffeic acid, ferulic acid, rosmarinic acid, luteolin, luteolin-7-O-glucoside and apigenin | DPPH assay (methanol extract): $89.5 \pm 0.2\%$ | ABTS assay (ethyl acetate): 0.81 ± 0.03 mm Trolox | β -Carotene linoleic acid bleaching (ethyl acetate): $19.2 \pm 3.1\%$ | Iron (III) reducing assay (water extract): $444.5 \pm 6.8 \mu\text{mol/g}$ | Dastmalchi et al., 2007; Povilaityte and Venskutonis, 2000; Povilaityte et al., 2001 |
| 10 | <i>Urtica dioica</i> L. (leaves) | Ultrasound bath | Methanol | NA | BR method: $0.013 \pm 0.001 \mu\text{g/ml}$ Re. | TEAC assay: 0.46 ± 0.07 mm Trolox | DPPH assay: $419 \pm 10 \mu\text{g/ml}$ | Total phenolics: $0.35 \pm 0.02 \text{ mg/l}$ GA | Dall' Acqua et al., 2008 |
| 11 | <i>Ficus microcarpa</i> L. fil. (bark, fruit and leaves) | Extraction | Methanol | Triterpenoids (lupenyl acetate, friedelin, glutinol, epifriedelinol, β -amyirin acetate and β -amyirin); phenolic compounds (protocatechuic acid, catechol, syringol and vanillin) | DPPH assay (bark): $\text{EC}_{50} = 7.9 \pm 0.1 \mu\text{g/ml}$ | ABTS assay (bark): $\text{EC}_{50} = 4 \pm 0.0 \mu\text{g/ml}$ | PMS- NADH system superoxide radical scavenging assay (bark): $\text{EC}_{50} = 97.5 \pm 2.8 \mu\text{g/ml}$ | NA | Ao et al., 2008; Kuo and Li, 1997 |
| 12 | <i>Bidens pilosa</i> Linn. Radiata (leaves and flowers) | Steam distillation | Diethyl ether | Terpenes (β -caryophyllene and τ -cadinene), α -pinene, limonene, β -trans-ocimene, β -cis-ocimene, τ -muurolene, β -bourbonene, β -elemene, β -cubebene, α -caryophyllene, caryophyllene oxide and megastigmatrienone | DPPH assay (leaves and flowers): $\text{IC}_{50} = 61$ and $172 \mu\text{g/ml}$, respectively | NA | NA | NA | Deba et al., 2008 |
| 13 | <i>Leea indica</i> (plant) | Solvent extraction | Methanol | Phthalic acid, palmitic acid, 1-eicosanol, solanesol, farnesol, three phthalic acid esters, gallic acid, lupeol, beta-sitosterol and ursolic acid | DPPH assay: $\text{IC}_{50} = 25 \mu\text{g/ml}$ | NA | NA | NA | Saha et al., 2004; Srinivasan et al., 2008 |
| 14 | Lamiaceae species (<i>Leonurus cardiaca</i> , <i>Lamium album</i> , <i>Marrubium vulgare</i> , <i>Stachys officinalis</i> , <i>Lamium purpureum</i> and <i>Galeopsis speciosa</i>) (plant material) | Reflux-extracted | Methanol | <i>Leonurus cardiaca</i> contains flavonoid and phenolic glycosides | DPPH assay: $\text{EC}_{50} = 0.7$ (L. cardiaca), 1 (L. album), 1.15 (M. vulgare) $\mu\text{g/ml}$ | Linoleic acid peroxidation assay $78.7 \pm 5.6\%$ (M. vulgare) $77.8 \pm 5.6\%$ (S. officinalis) $73.3 \pm 8.6\%$ (L. cardiaca) | NA | NA | Matkowski and Piotrowska, 2006; Mantle et al., 2000; Trouillas et al., 2003; Vander Jagt et al., 2002 |

Table 2 (Continued)

| S. No. | Species | Extraction method | Solvent(s) used | Main components (or groups) | Antioxidant assay methods | | | | Reference(s) |
|--------|--|----------------------|-------------------------------------|--|--|---|--|---|-------------------------------|
| 15 | <i>Uncaria tomentosa</i> (Willd.) DC. (bark) | Hot water extraction | Ethanol extract and aqueous extract | Alkaloids (uncarine, speciophylline, mitraphylline, isomitraphylline, isoteropodine); quinoic acid, glycosides and polyhydroxylated triterpenes | TEAC assay (ethanol and aqueous): 0.57 and 0.34 mmol Trolox/g, respectively | SOD activity (ethanol and aqueous): 0.39 and 0.1 U/mg, respectively | NA | NA | Pilarski et al., 2006 |
| 16 | <i>Salvia officinalis</i> L. (aerial parts) | Ultrasonic bath | Methanol extract and water extract | Phenolic acids (rosmarinic acid, caffeic acid, ferulic acid, 3-caffeoylquinic acid and 5-caffeoylquinic acid); flavonoids (luteolin-7-glucoside, 4',5,7,8-tetrahydroxyflavone, apigenin-7-glucoside) | DPPH assay (methanol and water): IC ₅₀ = 13.5 ± 0.5 (methanol) and 14.9 ± 0.3 (water) µg/ml | Superoxide radical method (methanol and water): IC ₅₀ = 162 ± 39 (methanol) and 14.4 ± 1.4 (water) µg/ml | NA | NA | Lima et al., 2007 |
| 17 | <i>Momordica Charantia</i> L. (leaf, stem, green fruit and ripe fruit) | Extraction | Distilled water | Gallic acid, tannic acid, catechin, caffeic acid, p-coumaric acid, ferulic acid and benzoic acid | DPPH assay (leaf, stem, green fruit and ripe fruit): IC ₅₀ = 9.72 ± 0.25 (leaf), 17.8 ± 0.66 (stem), 11 ± 0.76 (green fruit) and 27.6 ± 0.23 (ripe fruit) mg/ml | Hydroxyl radical scavenging activity (leaf, stem, green fruit and ripe fruit): IC ₅₀ = 167. ± 0.96 (leaf), 267 ± 0.72 (stem), 119 ± 0.34 (green fruit) and 173 ± 0.23 (ripe fruit) mg/ml | β-Carotene bleaching assay (leaf, stem, green fruit and ripe fruit): 63.9 ± 0.71 (leaf), 36.2 ± 0.59 (stem), 79.9 ± 0.7 (green fruit) and 59 ± 0.44 (ripe fruit) mg/ml | FRAP assay (leaf, stem, green fruit and ripe fruit) 433 ± 0.007 (leaf), 39 ± 0.008 (stem), 43.8 ± 0.008 (green fruit) and 9.41 ± 0.007 (ripe fruit) µ.mol FeSO ₄ /g dry sample | Kubola and Siriamornpun, 2008 |
| 18 | <i>Rheum ribes</i> L. (roots and stems) | Extraction | Chloroform and methanol | Chrysophanol, physcion, emodin, quercetin, 5-desoxyquercetin, and quercetin-3-O-rhamnoside | β-Carotene bleaching method (chloroform extracts of root at 50 and 100 µg): 91.09 ± 0.8% and 93.14 ± 1.17%, respectively | DPPH assay (methanol extracts of stems and roots): 87.07 ± 0.54% and 60.6 ± 0.86%, respectively | NA | NA | Ozturk et al., 2007 |
| 19 | <i>Pelargonium endlicherianum</i> (aerial parts) | Soxhlet | Methanol | NA | DPPH assay: IC ₅₀ = 7.43 ± 0.47 µg/ml | β-Carotene linoleic acid: 72.6 ± 2.96% | NA | NA | Tepe et al., 2006 |

dant activity of the bitter gourd extracts of the green fruit (79.9 ± 0.70 mg/ml) was greater than the activity of the extracts of the leaf (63.9 ± 0.71 mg/ml), the ripe fruit (59.0 ± 0.44 mg/ml) and the stem (36.2 ± 0.59 mg/ml). The FRAP value for the extracts of the leaf was the greatest with 433 ± 0.007 μ mol FeSO₄/g dry sample, followed by the extracts of the green fruit (43.8 ± 0.008 μ mol FeSO₄/g dry sample), the stem (39 ± 0.008 μ mol FeSO₄/g dry sample) and the ripe fruit (9.41 ± 0.007 μ mol FeSO₄/g dry sample). The antioxidant activity was greatest in the leaf, followed, in decreasing order, by the green fruit, the stem and the ripe fruit. The TPC of the leaf extract was 474 ± 0.71 , the green fruit extract was 324 ± 1.63 , the stem extract was 259 ± 1.20 and the ripe fruit extract was 224 ± 0.86 , all in units of mg GAE/g dry sample.

In the four analysed fractions, seven phenolic compounds were identified: *p*-coumeric acid, tannic acid, benzoic acid, ferulic acid, gallic acid, caffeic acid and (+)-catechin. Gallic acid was the most predominant of the phenolic compounds in all parts of the bitter gourd, contributing from 72.8 mg/l in the extracts of the stem to 202 mg/l in the extracts of the ripe fruit. Caffeic acid was most concentrated in the leaf extract (7.77 ± 1.02 mg/l), while *p*-coumeric acid was most abundant in the stem extract (6.73 ± 0.21 mg/l). Ferulic acid was only found in the stem and green fruit extracts, while benzoic acid was not present in either the leaf or the stem extracts. The bitter gourd fractions are rich in phenolics and have strong antioxidant activity and radical scavenging action by all of the testing methods (Kubola and Siriamornpun, 2008). Semiz and Sen (2007) have studied the fruit extract of *M. Charantia* in rats (200 mg/kg of weight) and found that there is a significant increase in the activity of the hepatic antioxidant enzymes, including SOD, catalase and glutathione peroxidase.

3.18. *Rheum ribes* L

Rhubarb (*Rheum ribes* L.) belongs to the family Polygonaceae. It is used for medicinal purposes, and its fresh stems and petioles are also consumed as a vegetable. It is commonly found in eastern Turkey, Lebanon and Iran. In Turkey, 11,700 types of plants are available, of which nearly a thousand have aromatic and medicinal value. *R. ribes* is the only *Rheum* species growing in Turkey. Rhubarb roots have been used as a laxative and an antipsoriatic drug in Iran (Shokravi and Agha Nasiri, 1997). The roots of the species are also used to treat diabetes, hypertension, obesity and diarrhoea (Abu-Irmaileh and Afifi, 2003; Tabata et al., 1994). The young shoots and petioles of *R. ribes* are used against diarrhoea and as a stomachic and antiemetic treatment.

The medicinal properties of this species are due to its anthroquinone content. It was found using the β -carotene bleaching method that the chloroform extracts of the roots at concentrations of 50 and 100 μ g/ml ($91.09 \pm 0.8\%$ and $93.14 \pm 1.17\%$, respectively) were more active than the same concentrations of quercetin ($86.11 \pm 1.09\%$ and $86.21 \pm 1.10\%$, respectively). Furthermore, the DPPH assay showed that methanol extracts of both the stems and the roots exhibited higher activity than BHT at concentrations greater than 50 μ g/ml. The methanol extract of the stems showed the highest DPPH radical scavenging activity among all of the extracts tested ($87.07 \pm 0.54\%$), followed by the methanol extract of the roots ($60.60 \pm 0.86\%$) and the chloroform extract of the roots ($50.87 \pm 0.3\%$) at a concentration of 100 μ g/ml.

In addition, the chloroform extract of the roots (48.66 ± 1.23 μ g PEs/mg extract) had a higher phenolic content

than the other extracts, and the extract containing the lowest quantity of phenolics was the chloroform extract of the stems (22.68 ± 1.10 μ g PEs/mg extract). The most flavonoid-rich extract was found to be the chloroform extract of the roots (145.59 ± 0.22 μ g QEs/mg extract), while the methanol extract of the stems (13.66 ± 0.75 μ g QEs/mg extract) had the lowest flavonoid content (Ozturk et al., 2007).

3.19. *Pelargonium endlicherianum*

Pelargonium endlicherianum is commonly found in Turkey and has biological activities including antimicrobial, antifungal, anti-inflammatory and analgesic activities.

In this study, the *P. endlicherianum* extract exerted a two-fold greater antioxidant activity ($IC_{50} = 7.43 \pm 0.47$ μ g/ml) than the synthetic antioxidant BHT (18.0 ± 0.4 μ g/ml). In the β -carotene/linoleic acid test system, *P. endlicherianum* exhibited a $72.6 \pm 2.96\%$ inhibition rate (Tepe et al., 2006). The results of this study support the use of *P. endlicherianum* as an additive in food and as a traditional medicine for anti-aging remedies.

The Germplasm Resources Information Network (GRIN), maintained by the United States Department of Agriculture (USDA), was used to obtain information about the distribution of these potent medicinal species from around the world. Although some of the species are also available in other parts of the world, the major distribution of these species is presented in Table 1. Table 2 shows a comparison of the antioxidant potential, extraction methods and chemical composition of these species.

4. Conclusion

This review discussed medicinally significant plant species from around the world and showed that many have high antioxidant activity when compared to synthetic antioxidants. In addition, many of these species have a high phenolic content and a large amount of flavonoids and flavonols. However, an overall ranking of the antioxidant strength of these species cannot be determined because of the different experimental methods used in various studies.

We have focused on plants belonging to several different families from around the world to understand their therapeutic uses and their potential antioxidant activities. Unfortunately, most of the species that are claimed to contain potent antioxidant activity have not been studied *in vivo*. Screening with *in vitro* assays has little meaning if there is no clear evidence of the effectiveness of the extracts *in vivo*. Therefore, further *in vivo* studies of these species are required, and a systematic investigation of these antioxidant-rich species is needed before they can be used in the food processing industry and as preventive medicine.

Acknowledgements

The authors wish to acknowledge the financial support of MOSTI Malaysia. This work was carried out under e-science grant number SCF0049-IND-2007.

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