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Review on - Recent Trends in Isolation of Antioxidantsfrom Spices and its Biological Effects ofEssential Oils

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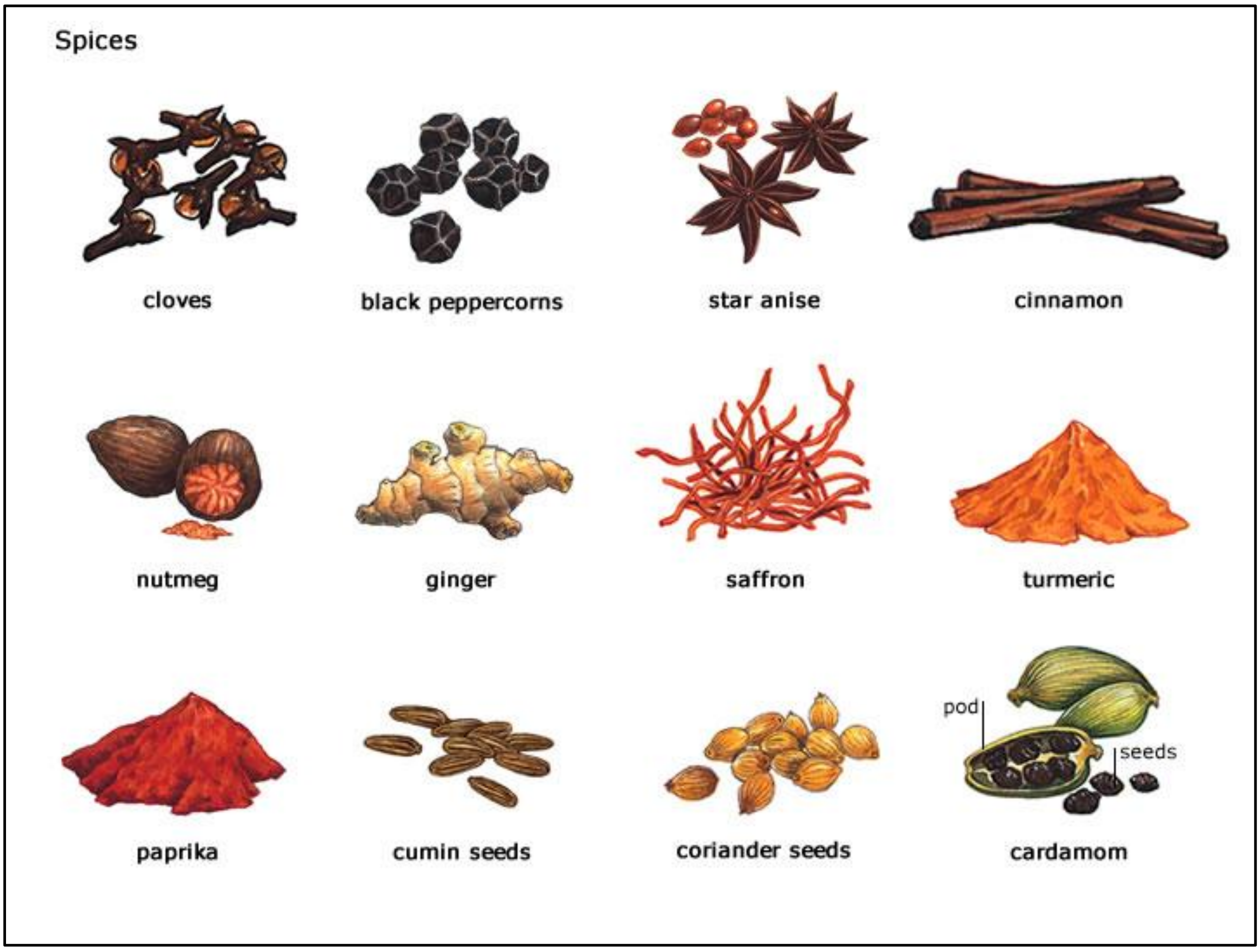
Abstract
Spices played a dramatic role in civilization andin the history of nations. The delightful flavour and pungency of spices make them indispensable in the preparation of palatable dishes. In addition, they are reputed to possess several medicinal and pharmacological properties and hence find position in the preparation of a number of medicines. Antioxidant compounds in food play important roles as health-protecting factors. Antioxidants are also widely used as additives infats and oils and in food processing to prevent or delay spoilage of foods. Spices and some herbs have received increased attention as sources of many effective antioxidants.Since the middle ages, essential oils have been widely used for bactericidal, virucidal, fungicidal, antiparasitical, insecticidal, medicinal and cosmetic applications, especially nowadays in pharmaceutical, sanitary, cosmetic, agricultural and food industries. Because of the mode of extraction, mostly by distillation from aromatic plants, they contain a variety of volatile molecules such as terpenes and terpenoids, phenol-derived aromatic components and aliphatic components. In vitro physicochemical assays characterise most of them as antioxidants. This review presents some information about the most common and most-used spice antioxidants and its essential oils, biological effects and describes their isolation of antioxidant properties.
Keywords: Spices, Flavour, Antioxidants, Essential Oil, Volatile, Isolation and Pharmaceuticals.

I. Introduction

A **spice** is a dried seed, fruit, root, bark, or vegetable substance primarily used for flavoring, coloring or preserving food. Sometimes a spice is used to hide other flavors (Scully and Terence 1995). Spices are distinguished from herbs, which are parts of leafy green plants also used for flavoring or as garnish. According to Thomas *et al.*, 2012, many spices have antimicrobial properties. This may explain why spices are more commonly used in warmer climates, which have more infectious disease, and why use of spices is especially prominent in meat, which is particularly susceptible to spoiling.A spice may have other uses, including medicinal, religious ritual, cosmetics or perfume production, or as a vegetable. For example, turmeric roots are consumed as a vegetableand garlic as an antibiotic.



An **essential oil** is a concentrated hydrophobic liquid containing volatile aroma compounds from plants. Essential oils are also known as volatile oils, ethereal oils, or aetherolea, or simply as the "oil of" the plant from which they were extracted, such as oil of clove. An oil is "essential" in the sense that it carries a distinctive scent, or essence, of the plant. Essential oils do not form a distinctive category for any medical, pharmacological, or culinary purpose.

Essential oils are generally extracted by distillation, often by using steam. Other processes include expression or solvent extraction. They are used in perfumes, cosmetics, soaps and other products, for flavoring food and drink, and for adding scents to incense and household cleaning products.

Essential oils have been used medicinally in history. Medical applications proposed by those who sell medicinal oils range from skin treatments to remedies for cancer and often are based solely on historical accounts of use of essential oils for these purposes. Claims for the efficacy of medical treatments, and treatment of cancers in particular, are now subject to regulation in most countries.As the use of essential oils has declined in evidence-based medicine; one must consult older textbooks for much information on their use (Sapeika and Norman 1963). Modern works are less inclined to generalize; rather than refer to "essential oils" as a class at all, they prefer to discuss specific compounds, such as methyl salicylate, rather than "oil of wintergreen".

Interest in essential oils Gilman *et al*., (1990) and Klaassen*et al*., (1991) has revived in recent decades with the popularity of aromatherapy, a branch of alternative medicine that claims that essential oils and other aromatic compounds have curative effects. Oils are volatilized or diluted in a carrier oil and used in massage, diffused in the air by a nebulizer, heated over a candle flame, or burned as incense.

II. Isolation of Antioxidants from spices

The number of contributions to isolation methods,techniques and activity testing of plant-origin antioxidants has significantly increased in recent years. Oxidation is one of the major causes of chemical spoilage, resulting in rancidity and/or deterioration of the nutritional quality, color, flavour, texture and safety of foods (Antolovich*et al.,* 2002). There is at present increasing interest both in the industry and in scientific research for spices and aromatic herbs because of their strong antioxidant and antimicrobial properties, which exceed many currently used natural and synthetic antioxidants. These properties are due to many substances, including some vitamins, flavonoids, terpenoids, carotenoids, phytoestrogens, minerals, etc. and render spices and some herbs or their antioxidant components as preservative agents in food (Calucci*et al*., 2003).

Except basic plant antioxidants some specific ones are characteristic for some important aromatic herbs and spices. Some examples of specific antioxidants are pimento from allspice; gallates, biflorin, its isomer eugenol and eugenyl acetate in clove (Anon, 1997; Lee and Shibamato,2001; Peter, 2000); carnosol, carnosic acid, rosmanol, rosmaridiphenol, rosmadial and rosmariquinone, and various methyl and ethyl esters of these substances in rosemary (Bandoniene*et al.*, 2002a, b; Pizzale*et al*., 2002); diarylheptanoids, gingerol and zingerone in ginger (Kikuzaki and Nakatani, 1993; Peter, 2000); curcumin and tetradehydrocurcumin in turmeric (Relajakshmi and Narasimhan, 1996); flavonides, ferulicacid, piperine, phenolic amide feruperine in black pepper (Peter, 2000; Shahidi and Wanasundara, 1992; Nakatani*et al*., 1986); derivatives of phenolic acids, flavonoids, tocopherols, rosmarinic acid and carvacrol in oregano (Peter, 2000; Pizzale*et al*., 2002); etc.

According to a phytochemical database (USDA, 2003), the number of different antioxidants in some plants can reach up to 40 (soybean 42, tea 36, fennel 35, onion 32, etc.). In this database, plants with the highest contents of antioxidants are walnut, betel nut, guava, coconut, and other less known plants. A list of some known substances with antioxidant activity in some verycommon spices is reported in Table 1. More information isavailable for examples on USDA food antioxidant database (USDA, 2003).

Table 1: Antioxidant active chemicals isolated from some of the most common and used spices (USDA, 2003)

Spices (vernacular and scientific names)	Antioxidants (name, part of plant, quantity in ppm)
Black pepper (Piper nigrum)	Ascorbic-acid fruit 0–10, beta-carotene fruit 0.114–0.128, camphene fruit, carvacrol fruit, eugenol fruit, gammaterpinene fruit, lauric-acid fruit 400–447, linalyl-acetate fruit, methyl-eugenol fruit, myrcene fruit, myristic-acid fruit 700–782, myristicin fruit, palmitic-acid fruit 12,200–13,633, piperine fruit 17,000–90,000, terpinen-4-ol fruit, ubiquinone fruit
Chilli pepper (Capsicum frutescens)	Alanine fruit 820–6691, ascorbic-acid fruit 350–19,992, beta-carotene fruit 1–38, caffeic-acid fruit 0–32, campesterol fruit, capsaicin fruit 100–17,900, capsanthin fruit, chlorogenic-acid stem, hesperidin fruit, histidine fruit 410–3346, kaempferol anther, lauric-acid resin, exudate, sap, lutein fruit, methionine fruit 240–1958, myrcene fruit, myristic-acid fruit 10–82, myristic-acid seed, p-coumaric-acid fruit 0–540, palmitic-acid fruit 150–1224, palmitic-acid seed, pentadecanoic-acid fruit, quercetin fruit 0–63, scopoletin fruit, stigmasterol fruit, terpinen-4-ol fruit, tocopherol fruit 0–24, tryptophan fruit 260–2122
Coriander (Coriandrumsativum L.)	Apigenin fruit, ascorbic-acid leaf 780–6290, beta-carotene leaf 29–228, beta-carotene seed, beta-sitosterol fruit, caffeic-acid fruit, caffeic-acid leaf, camphene fruit 2–155, chlorogenic-acid plant 305–320, gamma-terpinene fruit 762–2626, isoquercitrin fruit, myrcene fruit 13–169, myristic-acid fruit 200–219, myristicin fruit, p-hydroxybenzoic- acid fruit 0–960, p-hydroxy-benzoic-acid plant 252–333, palmitic-acid fruit 5000–16,800, protocatechuicacid fruit 0–760, protocatechuic-acid plant 167–179, quercetin fruit, rhamnetin fruit, rutin fruit, scopoletin fruit, tannin fruit, terpinen-4-ol fruit 6–80, trans-anethole fruit 1–2, vanillic-acid fruit 0–960, vanillic-acid plant 221–347
Ginger (Zingiberofficinale)	6-Gingerol rhizome 130–7138, 6-shogaol rhizome 40–330, alanine rhizome 310–1793, ascorbic-acid rhizome 0–317, beta-carotene rhizome 0–4, beta-sitosterol plant, caffeic-acid rhizome, camphene rhizome 28–6300, capsaicin plant, chlorogenic-acid plant, curcumin plant, delphinidin plant, ferulic-acid plant, gamma-terpinene rhizome 0.4–25, rhizome 300–1738, kaempferol plant, lauric-acid rhizome 390–3630, methionine rhizome 130–737, myrcene 2–950, myricetin plant, myristic-acid rhizome 180–1650, p-coumaric-acid rhizome 0–19, p-hydroxy-benzoic-acid plant, palmitic-acid rhizome 1200–11,220, quercetin plant, selenium rhizome 10, shikimic-acid leaf, sucrose rhizome, terpinen-4-ol rhizome, tryptophan rhizome 120–693, vanillic-acid plant, vanillin plant

Nutmeg (Myristicafragrans)	Camphene seed 80–640, cyanidin plant, eugenol seed 40–320, gamma-terpinene seed 580–4640, isoeugenol seed 140–320, kaempferol plant, lauric-acid seed 375–1600, methyl-eugenol seed 20–900, myrcene seed 740–5920, myristic-acid seed 60–304,000, myristicin leaf 410–620, myristicin seed 800–12,800, oleanolic-acid seed, palmiticacid seed 25,000–128,000, quercetin plant, terpinen-4-ol seed 600–4800
Red (sweet) pepper (Capsicum annuum)	Alanine fruit 350–4774, alpha-tocopherol fruit 22–284, ascorbic-acid fruit 230–20,982, beta-carotene fruit 0–462, beta-sitosterol plant, caffeic-acid fruit 0–11, campesterol fruit, camphene fruit, capsaicin fruit 100–4000, capsanthin fruit, chlorogenic-acid fruit, eugenol fruit, gamma-terpinenefruit, hesperidin fruit, histidine fruit 170–2319, lupeol seed, lutein fruit, methionine fruit 100–1364, myrcene fruit, myristic-acid fruit 10–136, pcoumaric- acid fruit 0–79, palmitic-acid fruit 500–6820, palmitic-acid seed, pentadecanoic-acid fruit, scopoletin fruit, selenium fruit 0.001–0.002, stigmasterol fruit, terpinen-4-ol fruit, tocopherol fruit 0–24, tryptophan fruit 110–1500
Rosemary (Rosemarinusofficinalis)	Apigenin plant, ascorbic-acid plant 612–673, beta-carotene plant 19–21, beta-sitosterol plant, caffeic-acid plant, camphene leaf 0–23, camphene leaf 0–145, camphene plant 23–2350, camphene shoot 355–1435, camphene shoot 620–1260, camphene shoot 1035–2280, carnosic-acid plant, carnosol leaf 530–9803, carvacrol leaf 0–5, carvacrol leaf 0–6, carvacrol leaf 5–6, carvacrol plant, chlorogenic-acid plant, gamma-terpinene leaf 0–4, gamma-terpinene plant 4–400, gamma-terpinene shoot 25–50, gamma-terpinene shoot 37–225, gamma-terpinene shoot 105–300, hesperidin leaf, hispidulin plant, isorosmanol flower 0–17, labiatic-acid plant, luteolin leaf, luteolin plant,
Turmeric (Curcuma domestica)	Ascorbic-acid rhizome 0–293, beta-carotene rhizome, caffeic-acid rhizome 0–5, curcumin rhizome 9–38,888, eugenol essential oil 0–2100, p-coumaric-acid rhizome 0–345, protocatechuic-acid leaf, syringic-acid leaf, vanillicacid leaf

III. Methods of antioxidant isolation from spices

Spices can be added to foods in several forms: as whole spices, as ground spices, or as isolates from their extracts. Spices are aromatic and pungent food ingredients, therefore, their direct use as antioxidants is limited. The extraction procedure is determined by the types of antioxidant compounds to be extracted. Selection of a suitable extraction procedure can increase the antioxidant concentration relative to the plant material. For polyphenols and other antioxidants in plant materials three principal extraction techniques may be used: extraction using solvents, solid-phase extraction and supercritical extraction. It is advisable to complete the extraction using dry, frozen or lyophilized samples since some antioxidants are unstable or can be degraded by enzyme action in undried plant material. Several extraction techniques have been patentedusing solvents with different polarities, such aspetrol ether, toluene, acetone, ethanol, methanol, ethyl acetate, and water. In addition, supercritical CO2-extraction and medium-chain triglycerides as carrier in a mechanical extraction process have been applied (Schwarz *et al.*, 2001). Extraction using edible oil or fat is a very simple method. Natural material containing antioxidants, such as herbs and spices, is mixed with fats and/or oils, and the mixture is left at a room or moderately increased temperature for a defined time. The mixture is then filtered and used (Pokorny*et al*., 2001). For industrial purposes ethanol would probably be better than methanol as eventual solvent residues would be less toxic. A review of some extraction procedures used for preparation of some spice and herb antioxidant substances is given in Table 2.

Table 2: Extraction and isolation procedures of antioxidant chemicals from some of the most common and used spices

Spices	Process of extraction	References
Basil, black pepper, cinnamon,oregano, parsley, rosemary, sage	(1) Trichloacetic acid extract for ascorbate determination, centrifugation 8000 g, filtration (2) Methanol extract for carotenoids and capsaicinoids determination, vacuum concentration, filtration	**Calucci*et al.*, (2003)**
Allspice, clove	Methanol extract	**Anon (1997)**
Ginger, turmeric, cayenne pepper, rosemary, sage, thyme, oregano, green	Mechanical medium-chain triglyceride and propylene glycol extracts prepared by hydraulic laboratory press	**Schwarz *etal.*, (2001)**

tea, spice mixture		
Pepper (Capsicum species)	Methanol extract (50% methanol, 50% water)	**Howard *et al.*, (2000)**
Ginger, nutmeg, coriander	Ethanol extract (96% ethanol, 4% water)	**Takacsova *et al.*, (1999)**
Thyme, basil, rosemary, chamomile, lavender, cinnamon	Liquid–liquid continuous extraction following steam distillation under reduced pressure	**Lee and Shibamato, (2002)**
Rosemary	CO2 supercritical fluid extraction	**Ibanez *et al.*, (2001)**
Chilly, black pepper, ginger	Near-critical carbon dioxide, propane, and dimethyl ether	**Catchpole *et al.*, (2003)**
Chilly, black pepper, turmeric	Aqueous extract, homogenization, centrifugation, filtration	**Sharma *et al.*, (2000)**
Cinnamon	Fat-free cinnamon Soxhlet extraction with 80% methanol, then n-hexane and ethyl acetate	**Bozan*et al.*, (2003)**
Pepper (Capsicum sp.)	50% methanol	**Howard *et al.*, (2000)**

IV. Surprising Source of Antioxidants from Spices and Herbs:

SPICES AND HERBS

A SURPRISING SOURCE OF ANTIOXIDANTS

Spice	Serving Size	ORAC (umol TE/serving)
CIMMAMON, GROUND	1 tsp	6956
CLOVES, GROUND	1 tsp	6603
OREGANO	1 tsp	3602
CUMIN	1 tsp	1613
CURRY POWDER	1 tsp	970
CHILI POWDER	1 tsp	615
BLACK PEPPER	1 tsp	580
GINGER, GROUND	1 tsp	519
THYME*	1 tsp	407
PAPRIKA	1 tsp	376
ROSEMARY	1 tsp	364
GARLIC POWDER	1 tsp	187

SOURCE: Oxygen Radical Absorbance Capacity (ORAC) of Selected Foods – 2007. Nutrient Data Laboratory USDA, November 2007. www.ars.usda.gov/nutrientdata/ORAC.

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The extracts obtained using organic solvents may be further concentrated, for instance, by molecular distillation. Essential oils present in spice extracts, are responsible for the characteristic aroma of the spices and can be removed by steam distillation at normal atmospheric pressure or in a vacuum, but antioxidant activity may be partially lost. Commercial antioxidant extracts from spices are available in powder form or as oily oleoresins (Pokorny*et al*., 2001).

V. Essential Oils

Essential oils are liquid products of steam or water distillation of plant parts (leaves, stems, bark, seeds, fruits, roots and plant exudates). Expression is used exclusively for the extraction of citrus oil from the fruit peel, because the chemical components of the oil are easily damaged by heat. Citrus oil production is now a major by-product process of the juice industry. An essential oil may contain up to several hundred chemical compounds and this complex mixture of compounds gives the oil its characteristic fragrance and flavour. The plant parts can be extracted with organic solvents to produce oleoresins, concretes and absolutes or extracted with a near or supercritical solvent such as carbon dioxide to produce very high quality extracts. These oleoresins and extracts contain not only the volatile essential oil but also the concentrated non-volatile flavor components and these have wide application in the food and pharmaceutical industries. Essential oils have been largely employed for their properties already observed in nature, i.e. for their antibacterial, antifungal and insecticidal activities. At present, approximately 3000 essential oils are known, 300 of which are commercially important especially for the pharmaceutical, agronomic, food, sanitary, cosmetic and perfume industries.

Essential oils or some of their components are used in perfumes and make-up products, in sanitary products, in dentistry, in agriculture, as food preservers and additives, and as natural remedies. For example, d-limonene, geranyl acetate or d-carvone are employed in perfumes, creams, soaps, as flavour additives for food, as fragrances for household cleaning products and as industrial solvents.

Moreover, essential oils are used in massages as mixtures with vegetal oil or in baths but most frequently in aromatherapy. Some essential oils appear to exhibit particular medicinal properties that have been claimed to cure one or another organ dysfunction or systemic disorder (Silva *et al.,* 2003; Hajhashemi*et al*., 2003; Perry *et al*., 2003).

Owing to the new attraction for natural products like essential oils, despite their wide use and being familiar to us as fragrances, it is important to develop a better understanding of their mode of biological action for new applications in human health, agriculture and the environment.

Some of them constitute effective alternatives or complements to synthetic compounds of the chemical industry, without showing the same secondary effects (Carson and Riley, 2003).

The most important spices traditionally traded throughout the world are products of tropicalenvironments. The major exceptions to this group are the capsicums (chilli peppers,paprika), and coriander which are grown over a much wider range of tropical and non-tropicalenvironments. Production of spices and essential oils in these wet and humidenvironments brings special difficulties for crop and product management. Drying the cropto ensure a stable stored product is of particular importance, and in wet humidenvironments this creates the need for efficient and effective drying systems.

VI. Biological effects of Essential Oils:

6.1 Cytotoxicity

As typical lipophiles, they pass through the cell wall and cytoplasmic membrane, disrupt the structure of their different layers of polysaccharides, fatty acids and phospholipids and permeabilize them. Cytotoxicity appears to include such membrane damage. Essential oils can coagulate the cytoplasm (Gustafson *et al*., 1998) and damage lipids and proteins (Ultee*et al.*, 2002; Burt, 2004). Damage to the cell wall and membrane can lead to the leakage of macromolecules and to lysis (Juven*et al*., 1994; Gustafson *et al.*, 1998; Cox*et al*., 2000; Lambert *et al*., 2001; Oussalah*et al.*, 2006). A Permeabilizationof outer and inner mitochondrial membrane leads to cell death by apoptosis and necrosis (Yoon *et al.*, 2000; Armstrong, 2006). It seems that chain reactions from the cell wall or the outer cell membrane invade the whole cell, through the membranes of different organelles like mitochondria and peroxisomes. Analyses of the lipid profiles by gas chromatography and of the cell envelope structure by scanning electron microscopy of several bacteria treated by some essential oil constituents showed a strong decrease in unsaturated and an increase in saturated fatty acids, as well as alterations of the cell envelopes (Di Pasqua*et al*., 2007). The induction of membrane damages has been also confirmed by a microarray analysis showing that Saccharomyces*cerevisiae*genes involved in ergosterol biosynthesis and sterol uptake, lipid metabolism,cell wall structure and function, detoxification and cellulartransport are affected by a treatment with a-terpinene, a monocyclic monoterpene (Parveen*et al*., 2004). Until now, because of their mode of action affecting several targets at the same time, generally, no particular resistance or adaptation to essential oils has been described. However, a resistance to carvacrol of Bacillus cereus has been observed after growth in the presence of a sublethalcarvacrol concentration. Pre-treatment with carvacroldiminished the fluidity of the membrane by changing its fatty acid ratio and composition (Ultee*et al*., 2000; DiPasqua*et al.*, 2006). However, Rafii and Shahverdi (2007) have found a potentiation of the antibiotic nitrofurantoin at a sub-inhibitory concentration by essential oils against enterobacteria. Probably, given the effect of essential oils on cell membranes, the bacterial susceptibility or resistance depend on the mode of application and may suggest that the antibiotic has to be first in contact with the cells (Rafii and Shahverdi, 2007).

6.2 Phototoxicity

Some essential oils contain photoactive molecules likefurocoumarins. Dijoux*et al.*, (2006)have shown that Fusanusspicatuswood essential oil was notphototoxic but was very cytotoxic. In other words, cytotoxicityseems rather antagonistic to phototoxicity. In the caseof cytotoxicity, essential oils damage the cellular and organellemembranes and can act as prooxidants on proteinsand DNA with production of reactive oxygen species(ROS), and light exposures do not add much to the overallreaction. In the case of phototoxicity, essential oils penetratethe cell without damaging the membranes or proteins andDNA. Obviously, cytotoxicity or phototoxicitydepends on the type of molecules present in the essential oilsand their compartmentation in the cell, producing differenttypes of radicals with or without light exposure. Thus, when studying anessential oil, it may be of interest to determine systematically it's cytotoxic as well as its possible phototoxic capacity.

6.3 Nuclear mutagenicity

Several studies with various essential oils or their maincomponents have demonstrated that, generally, most ofthem did not induce nuclear mutations, whatever theorganism, i.e. bacteria, yeast or insect, with or withoutmetabolic activation and whatever form of essential oils, For example, mentone of the peppermentha essential oil gave positive results in the Ames test (Andersen and Jensen, 1984) Mentone was also foundgenotoxic in SMART test (Franzios*et al*., 1997). Anetholfrom fennel and anise essential oils was active in the Amestest (Nestman and Lee, 1983; Hasheminejad and Caldwell, 1994), however, the oxidized metabolic intermediates of thesetwo molecules, trans-anethole oxide and Trans-asaroneoxide, were genotoxic in the Ames test and induced liverand skin cancers (Kim *et al*., 1999).

6.4 Cytoplasmic mutagenicity

Most of the mutagenicity (and anti-mutagenicity) studieson essential oils were performed on bacteria (Salmonella*typhimurium*with Ames test, Escherichia coli with SOSChromotest, and Bacillus*subtilis*with DNA Repair test). In this test system, it is impossible to distinguishthe mode of action of essential oils and their targets. Usually,cytotoxicity, mutagenicity or anti-mutagenicity isassessed without being able to take into account possible defects in energy metabolism and respiration as direct orindirect causes. In this respect, tests in yeast (Saccharomyces*cerevisiae)* have been shown to be potentially very useful.

Taking advantage of the yeast system, it is possible to showthat, among others, mitochondria are very important cellulartargets for essential oils. Indeed, a relation between thedeterioration of mitochondria and immediate changes ofrespiratory metabolism was demonstrated after treatmentof yeast cells (Saccharomyces *cerevisiae*) with the tea treeessential oil (Schmolz*et al*., 1999). Cells of Saccharomyces*cerevisiae* showed a delay in ethanol production in the presenceof cinnamon, clove, garlic, onion, oregano and thymeessential oils, as estimated by the measure of the CO2 volumeproduced (Conner *et al*., 1984).

6.5 Carcinogenicity of the essential oils

Since most essential oils have been found to be cytotoxicwithout being mutagenic, it is likely that most of them arealso devoid of carcinogenicity.
However, some essentialoils or rather some of their constituents may be consideredas secondary carcinogens after metabolic activation (Guba, 2001). For example, essential oils like those from Salvia sclareaand Melaleucaquinquenerviaprovoke estrogen secretionswhich can induce estrogen-dependent cancers. Someothers contain photosensitizing molecules like flavins,cyanin, porphyrins, hydrocarbures which can cause skinerythema or cancer.Pulegone, a component of essentialoils from many mint species, can induce carcinogenesisthrough

metabolism generating the glutathione depletory p-cresol (Zhou *et al*., 2004).

6.6 Antimutagenic properties of essential oils

Anti-mutagenicproperties may be due to inhibition of penetration of themutagens into the cells (Kada and Shimoi, 1987; Shankel*et al*., 1993), inactivation of the mutagens by direct scavenging,antioxidant capture of radicals produced by a mutagenor activation of cell antioxidant enzymes (Hartman andShankel, 1990; Sharma *et al*., 2001; Ipek*et al*., 2005), or activation of enzymatic detoxification ofmutagens for instance by plant extracts. Hernandez-Ceruelos*et al*. (2002) showed that Matricariachamomilla essentialoil inhibits SCEs induced by daunorubicine and methyl methane sulfonatein mouse bone marrow cells. In a more recentstudy, they showed in the same system that Origanumcompactumessential oil and some of its sub-fractions and constituentsare antimutagenic against the indirect-actingmutagen urethane and also against the direct-acting mutagenmethyl methanesulfonate (Mezzoug*et al.*, 2007).

VII. Conclusion

In this view of the diversity of methods used for the isolation of antioxidants from spice and herb, their activity determination and biological effects of essential oils, there is a great need to standardize them for both these measurements. The search for more specific assays that give us chemical information, which could be related directly to oxidative deterioration of foods and biological systems, should be the objective of future research. Modern consumers ask for natural products, free of synthetic additives. Therefore, the application of natural antioxidants and essential oils will probably continue even the future, and it will be necessary to study their changes and interactions in more details. Scientists will look for new effective herbal sources with potent antioxidants and essential oils from natural herbal plants. All these plants extract their mixtures, isolates and concentrates with antioxidant and biological effects of essential oils have to meet all the requirements of human health safety.

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