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Antioxidant activities of extracts from selected culinary herbs and spices

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Abstract

Recently, interest in plant-derived food additives has grown, mainly because synthetic antioxidants suffer from several drawbacks. Furthermore, plant extracts have been shown to possess health-promoting properties. In the present study, hydrodistilled extracts from basil, laurel, parsley, juniper, aniseed, fennel, cumin, cardamom, and ginger were assessed for their total phenol content, and antioxidant (iron(III) reduction, inhibition of linoleic acid peroxidation, iron(II) chelation, 1,1-diphenyl-2-picrylhydrazyl radical-scavenging and inhibition of hydroxyl radical-mediated 2-deoxy-D-ribose degradation, site and nonsite-specific) activities. The extracts from basil and laurel possessed the highest antioxidant activities except for iron chelation. Although parsley showed the best performance in the iron chelation assay, it was less effective at retarding the oxidation of linoleic acid. In the linoleic acid peroxidation assay, 1 g of the basil and laurel extracts were as effective as 177 and 212 mg of trolox, respectively. Thus, both extracts are promising alternatives to synthetic substances as food ingredients with antioxidant activity.
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Keywords: Antioxidant activity; Culinary spices; Hydrodistilled extracts; Basil; Laurel; Parsley; Juniper; Aniseed; Fennel; Cumin; Cardamom; Ginger

1. Introduction

In recent years, interest in plant-derived food additives has grown. Deterioration of food quality occurs during processing and storage and is related to oxidative processes. These are often catalysed by, e.g., ferrous or copper ions. Degradation affects (above all) lipids, carbohydrates and proteins (Halliwell, 1997). Usually synthetic antioxidants such as butylhydroxyanisole (BHA) or butylhydroxytoluene (BHT) are used to decelerate these processes. Yet, these antioxidants suffer from the drawback that they are volatile and easily decompose at high temperatures. Additionally, it is still unclear whether chronic consumption can lead to health risks (Martinez-Tome et al., 2001).

Many herbs and spices, usually used to flavour dishes, are an excellent source of phenolic compounds which have been reported to show good antioxidant activity (Rice-Evans, Miller, & Paganga, 1996; Zheng & Wang, 2001). Therefore, they may serve as natural food preservatives. However, herbs and spices usually contain essential oils which show antioxidant activity but also carry flavour (Ruberto, Baratta, Deans, & Dorman, 2000; Teissedre & Waterhouse, 2000). Thus, extracts are prepared by hydrodistillation to remove the intrinsic flavour from the plant material. Furthermore, use of an aqueous solvent may prevent solubility problems and this avoids harmful residues from organic solvents. The hydrodistilled extracts may also have use in the functionalization of foods and beverages as phenolic

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Table 1
Characterization of the plant material and extraction yield for hydrodistilled extracts

Spice	Botanical family	Extraction yield (mg/g)	Total phenols (mg GA/g)
Basil (*Ocimum basilicum*)	Lamiaceae	246	147 ± 1.60a
Parsley (*Petroselinum crispum*)	Apiaceae	196	29.2 ± 0.44b,c
Laurel (*Laurum nobilis*)	Lauraceae	258	92.0 ± 2.45d
Juniper (*Juniperus communis*)	Cupressaceae	422	18.5 ± 0.62e
Cardamom (*Elettaria cardamomum*)	Zingiberaceae	88	24.2 ± 0.29b,f
Ginger (*Zingiber officinalis*)	Zingiberaceae	302	23.5 ± 1.26b,e
Aniseed (*Pimpinella anisum*)	Apiaceae	230	20.8 ± 0.62e,f
Fennel (*Foeniculum vulgare*)	Apiaceae	216	30.3 ± 0.76c
Cumin (*Carum carvi*)	Apiaceae	242	37.4 ± 0.32g

Lines with the same lowercase letter are not significantly different for total phenols ($P > 0.05$).

compounds have been ascribed health-promoting properties (Harborne & Williams, 2000).

We chose nine commercially available herbs and spices to test antioxidant activity. The plant material was from aromatic plants used in different kinds of dishes as listed in Table 1. In hydrodistilled extracts from these herbs and spices, antioxidant activity has yet not been studied. In the present study, we have investigated the antioxidant activity (iron reduction, inhibition of lipid peroxidation, iron chelation, 1,1-diphenyl-2-picrylhydrazyl (DPPH$^{\bullet}$)-scavenging, and inhibition of hydroxyl radical-mediated degradation of deoxyribose). Furthermore, the total phenols content and the extraction yield is evaluated.

2. Material and methods

2.1. Materials

Dried plant materials were obtained from Paulig Oy, Finland. Ultrapure water was prepared using a Millipore Milli-RO 12 plus system (Millipore Corp., Bedford, MA). All reagents and solvents were either of analytical or HPLC grade and were obtained from Sigma Chemical Co. (St. Louis, MO).

2.2. Preparation of freeze-dried extracts

Fifty gramme of ground plant material were suspended in 500 ml ultrapure water and hydrodistilled for 2 h in a European Pharmacopoeian hydrodistillation apparatus. This process was repeated for 1 h with fresh solvent, and the combined aqueous extracts were then filtered, reduced in volume in vacuo (45 °C), freeze-dried, and stored at 4 °C. The extracts were dissolved in ultrapure water prior to use.

2.3. Total phenols

The total phenols were estimated according to the Folin-Ciocalteu method (Singleton, Orthofer, & Lamuela-Raventós, 1999). To 50 μl sample were added 250 μl of undiluted Folin-Ciocalteu-reagent. After 1 min, 750 μl of 20% (w/v) aqueous Na_2CO_3 were added, and the volume was made up to 5.0 ml with H_2O. The controls contained all the reaction reagents except the extract. After 2 h of incubation at 25 °C, the absorbance was measured at 760 nm and compared to a gallic acid calibration curve. Total phenols were determined as gallic acid equivalents (mg gallic acid/g extract), and the values are presented as means of triplicate analyses.

2.4. Iron(III) to iron(II)-reducing activity

The ability of the extracts to reduce iron(III) was assessed by the method of Oyaizu (1986). A 1-ml aliquot of each extract, dissolved in water, was mixed with 2.5 ml of phosphate buffer (0.2 M, pH 6.6) and 2.5 ml of a 1% aqueous potassium hexacyanoferrate [$K_3Fe(CN)_6$] solution. After a 30 min incubation at 50 °C, 2.5 ml of 10% trichloroacetic acid were added, and the mixture was centrifuged for 10 min. A 2.5-ml aliquot of the upper layer was mixed with 2.5 ml of water and 0.5 ml of 0.1% aqueous $FeCl_3$, and the absorbance was recorded at 700 nm. Iron(III) reducing activity was determined as ascorbic acid equivalents (mmol ascorbic acid/g extract). The values are presented as the means of triplicate analyses.

2.5. Inhibition of linoleic acid peroxidation

The antioxidant activity was determined as the degree of inhibition on the hemoglobin-catalysed peroxidation of linoleic acid according to Kuo, Yeh, and Sun Pan (1999). Ten microlitres of sample were added to 0.37 ml of 0.05 M phosphate buffer (pH 7.0), containing 0.14% Tween 20 and 4 mM linoleic acid, and then equilibrated at 37 °C for 3 min. The peroxidation of linoleic acid in the above reaction mixture was initiated by adding 20 μl of 0.035% hemoglobin (in water), followed by incubation at the same temperature for 10 min and stopped by adding 5 ml of 0.6% HCl (in ethanol). The hydroperoxide formed was assayed according to a ferric thiocyanate method with mixing, in order, of 30% ammonium thiocyanate (0.1 ml) and 0.02 M ferrous

chloride (0.1 ml). The absorbance (A_s) was measured at 480 nm after 5 min. The absorbance blank (A_0) was obtained without adding hemoglobin to the above reaction mixture; the absorbance of the control (A_{100}) was obtained by replacing the sample by buffer.

$$AA(\%) = (1 - (A_s - A_0)/(A_{100} - A_0)) \cdot 100. \quad (1)$$

The antioxidant activities of the samples were calculated as trolox equivalents (mg trolox/g extract) according to Eq. (1). The values are presented as the means of triplicate analyses.

2.6. Iron(II) chelation activity

The chelation of iron(II) ions by the different extracts was carried out as described by Carter (1971). Two hundred microlitres of each extract were added to 100 μl of 2.0 mM aqueous $FeCl_2$ and 900 μl methanol. The controls contained all the reaction reagents except the extract or positive control substance. After a 5 min incubation, the reaction was initiated by 400 μl of 5.0 mM ferrozine. After a 10 min equilibrium period, the absorbance at 562 nm was recorded. The iron chelation activities were calculated from the absorbance of the control (A_c) and of the sample (A_s) using Eq. (2) and expressed as Na_2EDTA equivalents (mg Na_2EDTA/g extract). The values are presented as the means of triplicate analyses.

$$\text{inhibition } (\%) = \frac{A_c - A_s}{A_c} \cdot 100. \quad (2)$$

2.7. 1,1-Diphenyl-2-picrylhydrazyl radical-scavenging

The ability of the extracts to scavenge DPPH$^{\cdot}$ free radicals was determined by the method of Gyamfi, Yonamine, and Aniya (1999). A 50-μl aliquot of each extract was mixed with 450 μl of Tris–HCl buffer and 1.0 ml of 0.1 mM 1,1-diphenyl-2-picrylhydrazyl in methanol. The controls contained all the reaction reagents except the extract or positive control substance. After a 30 min incubation in darkness and at ambient temperature, the resultant absorbance was recorded at 517 nm. The percentage inhibition values were calculated from the absorbance of the control (A_c) and of the sample (A_s) using Eq. (2), and the IC_{50} values were estimated by a nonlinear regression algorithm (SigmaPlot 2001 version 7.0). Ascorbic acid, BHA, BHT, and gallic acid were used as positive controls. The values are presented as the mean of triplicate analyses.

2.8. Nonsite-specific hydroxyl radical-mediated 2-deoxy-D-ribose degradation

The ability of the extracts to inhibit nonsite-specific hydroxyl radical-mediated peroxidation was carried out according to Halliwell, Gutteridge, and Aruoma (1987) with some adaptions. The reaction mixture contained 100 μl of extract dissolved in water, 500 μl of 5.6 mM 2-deoxy-D-ribose in KH_2PO_4–NaOH buffer (50 mM, pH 7.4), 200 μl of premixed 100 μM $FeCl_3$ and 104 mM EDTA (1:1 v/v) solution, 100 μl of 1.0 mM H_2O_2 and 100 μl of 1.0 mM aqueous ascorbic acid. Tubes were vortexed and incubated at 50 °C for 30 min. Thereafter, 1 ml of 2.8% TCA and 1 ml of 1.0% TBA were added to each tube. The samples were vortexed and heated in a water bath at 50 °C for 30 min. The extent of oxidation was estimated from the absorbance of the solution at 532 nm. The percentage inhibition values were calculated from the absorbance of the control (A_c) and of the sample (A_s) using Eq. (2), where the controls contained all the reaction reagents except the extract or positive control substance. The antioxidant activities of the extracts were expressed as mannitol equivalents (μmol mannitol/g extract). The values are presented as the means of triplicate analyses.

2.9. Site-specific hydroxyl radical-mediated 2-deoxy-D-ribose degradation

The ability of the extracts to inhibit site-specific hydroxyl radical-mediated peroxidation was carried out as described in the nonsite-specific hydroxyl radical-mediated peroxidation inhibition procedure, except that EDTA was replaced by buffer. The antioxidant activity of the extracts were expressed as mannitol equivalents (μmol mannitol/g extract). The values are presented as the means of triplicate analyses.

2.10. Statistical analysis

All statistical analyses were carried out using inerSTAT-a (Vargas, Mexico) as add-in for Microsoft Excel 2000. Analysis of variance (ANOVA) was followed by Tukey's pairwise comparison test at a level of $P < 0.05$ for the determination of significant differences between means.

3. Results

3.1. Extraction yield and total phenols

Table 1 shows an overview of the spices and herbs used, as well as the extraction yields and total phenols, for the hydrodistilled extracts.

The extraction yields range from 88 mg/g plant material for cardamom to 422 mg/g plant material for juniper. No significant association could be found neither between the extraction yields and total phenols nor between the extraction yields and the results from the different antioxidant assays.

Phenolic substances have been shown to be responsible for the antioxidant activity of plant materials

(Rice-Evans et al., 1996). Therefore, the amount of total phenols in the extracts was investigated by the Folin-Ciocalteu method. The content of total phenols is expressed as gallic acid equivalents (mg gallic acid/g extract). Significantly highest results were found in the order basil > laurel > cumin > fennel ($P < 0.05$). Juniper had the lowest content of total phenols. The content of total phenolics shows a good correlation with most of the antioxidant assays, such as iron reduction ($r^2 = 0.8871$, $P < 0.001$), inhibition of lipid peroxidation ($r^2 = 0.7327$, $P < 0.01$), nonsite-specific ($r^2 = 0.6589$, $P < 0.01$) and site-specific ($r^2 = 0.6041$, $P < 0.05$) hydroxyl scavenging. No association could be found between total phenolics and $DPPH^{\cdot}$ reduction as well as between total phenolics and iron chelation. The total phenols/extractable compounds ratio ranked from 7.8% for ginger to 59.7% for basil.

3.2. Iron(III) to iron(II)-reducing activity

Fe(III) reduction is often used as an indicator of electron-donating activity, which is an important mechanism of phenolic antioxidant action (Yildirim, Mavi, & Kara, 2001). The iron(III) to iron(II)-reducing activity is expressed as ascorbic acid equivalents (mmol ascorbic acid/g sample). Fig. 1(a) shows that the best activities

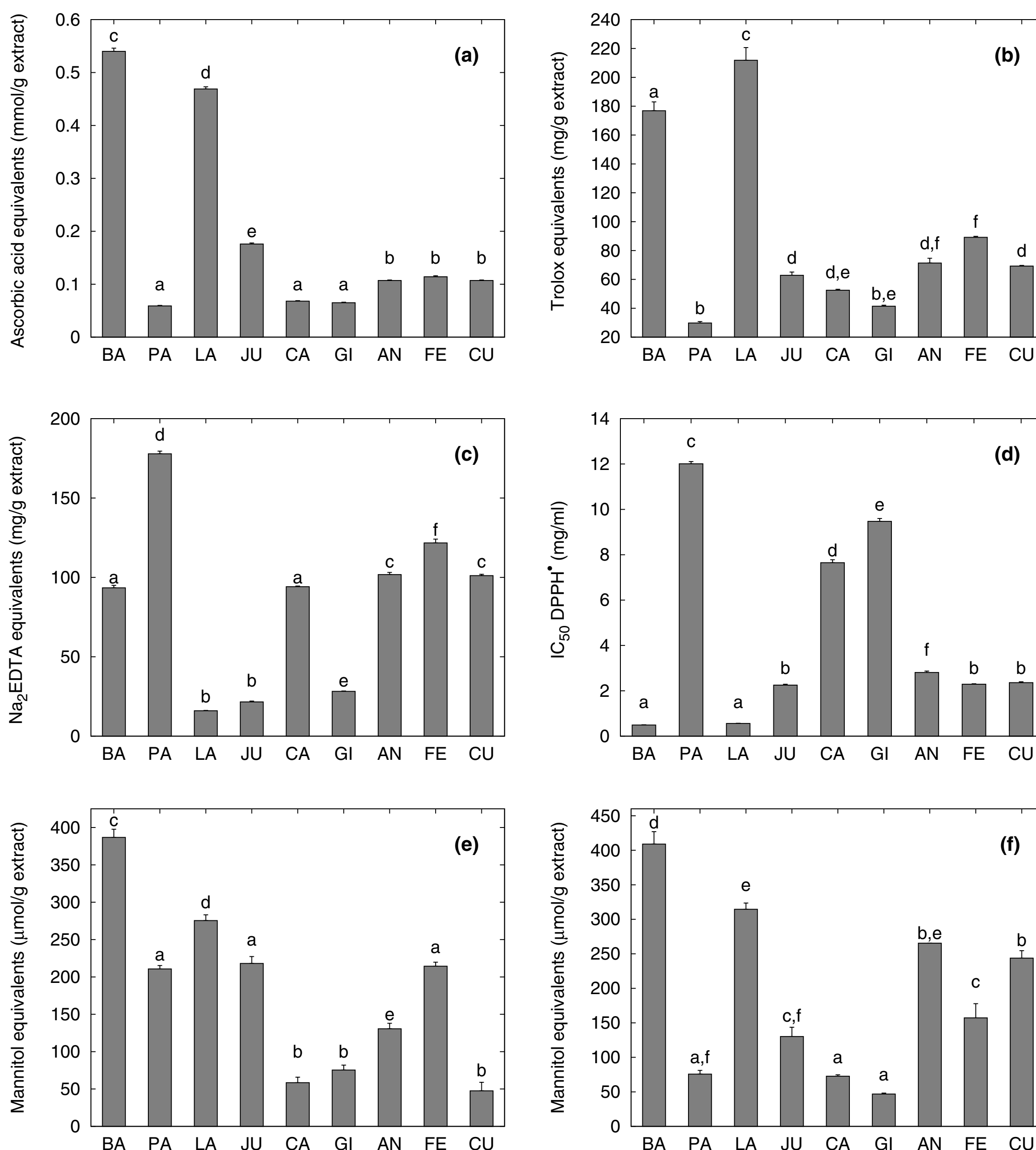

Fig. 1. Antioxidant activity of spice extracts in iron reduction (a), in lipid peroxidation (b), in iron chelation (c), against $DPPH^{\cdot}$ (d), in site-specific (e) as well as nonsite-specific (f) deoxyribose degradation assay. BA, basil; PA, parsley; LA, laurel; JU, juniper; CA, cardamom; GI, ginger; AN, aniseed; FE, fennel; CU, cumin. Bars with the same lowercase letter are not significantly different ($P > 0.05$).

were found for basil, laurel and juniper. For these three spices, the ascorbic acid equivalents were significantly higher than for the other extracts ($P < 0.05$). No significant difference could be found between aniseed, fennel and cumin, the extracts of which performed significantly better than parsley, ginger and cardamon. Ascorbic acid equivalents ranked from 0.54 mmol ascorbic acid/g extract to 0.06 mmol ascorbic acid/g extract.

The iron(III)-reducing activity correlated well with the content of total phenols ($r^2 = 0.8871$, $P < 0.01$), inhibition of lipid peroxidation ($r^2 = 0.8958$, $P < 0.001$) and deoxyribose degradation inhibition – site-specific ($r^2 = 0.6713$, $P < 0.01$) and nonsite-specific ($r^2 = 0.6883$, $P < 0.01$). No correlation was found with the DPPH· and the iron chelation assay.

3.3. Hemoglobin-catalysed peroxidation of linoleic acid

In the hemoglobin-catalysed peroxidation of linoleic acid assay, linoleic acid served as a model lipid. Peroxidation was induced by hemoglobin and the damage was assayed following the thiocyanate method Kuo et al. (1999). Trolox was used as reference substance. As can be seen in Fig. 1(b), basil and laurel were significantly better inhibitors of lipid peroxidation than the other spices ($P < 0.01$), whereas significantly lowest values were found for parsley, ginger and cardamom ($P < 0.05$). Trolox equivalents ranked from 29.8 ± 1.03 mg trolox/g extract for parsley to 212 ± 8.80 mg trolox/g extract for laurel.

The antioxidant activity in this assay correlated well with iron reduction ($r^2 = 0.8958$, $P < 0.001$), DPPH· scavenging ($r^2 = 0.5147$, $P < 0.05$), total phenols ($r^2 = 0.7327$, $P < 0.01$) and the inhibition of deoxyribose inhibition, site specific ($r^2 = 0.5051$, $P < 0.05$) as well as nonsite-specific ($r^2 = 0.6850$, $P < 0.01$). No correlation was found with iron chelation or extraction yield.

3.4. Iron(II) chelation

Foods are often contaminated with transition metal ions which may be introduced by processing methods. Bivalent transition metal ions play an important role as catalysts of oxidative processes, leading to the formation of hydroxyl radicals and hydroperoxide decomposition reactions via Fenton chemistry (Halliwell, 1997). These processes can be delayed by iron chelation and deactivation. Therefore, the ability of the extracts to chelate iron(II) ions was evaluated and expressed as Na_2EDTA equivalents (mg Na_2EDTA/g extract). The results are presented in Fig. 1(c).

Parsley showed the significantly ($P < 0.01$) best iron chelation (178 ± 1.68 mg Na_2EDTA/g extract), followed by fennel. No superior activity was found for basil, as in the other antioxidant assays. Laurel and juniper showed no significant difference in their low Na_2EDTA equivalent values (16.0 ± 0.17 and 21.6 ± 0.51 mg Na_2EDTA/g extract).

No significant correlation was found between the iron chelation ability of the extracts and the other antioxidant activities.

3.5. DPPH· scavenging

DPPH· is a stable nitrogen-centred free radical the colour of which changes from violet to yellow upon reduction by either the process of hydrogen- or electron-donation. Substances which are able to perform this reaction can be considered as antioxidants and therefore radical scavengers (Brand-Williams, Cuvelier, & Berset, 1995). IC_{50} values ranked from 0.49 ± 0.01 mg/ml for basil to 12.0 ± 0.10 mg/ml for parsley (Fig. 1(d)). No significant difference could be found for basil and laurel or for juniper, fennel and cumin. Ascorbic acid, BHA, BHT and gallic acid were tested as references. The IC_{50} values were 0.09 ± 0.01 mg/ml for ascorbic acid and BHA. BHT showed 50% inhibition at 0.21 ± 0.01 mg/ml. Gallic acid performed best, with an IC_{50} value of 0.03 ± 0.01 mg/ml. The results for the reference substances were significantly different from those for all extracts ($P < 0.05$), except basil.

The IC_{50} values for DPPH·-scavenging correlated with the results from the nonsite-specific deoxyribose degradation assay ($r^2 = 0.6440$, $P < 0.01$) and linoleic acid peroxidation ($r^2 = 0.5147$, $P < 0.05$); for the other assays no association could be found.

3.6. Site- and nonsite-specific hydroxyl radical-mediated deoxyribose degradation

Deoxyribose degradation occurs by hydroxyl radicals generated by a Fenton reaction. The activities of the extracts were compared to that of mannitol which has been reported to be an effective hydroxyl radical scavenger (Halliwell et al., 1987).

In the site-specific assay, the mannitol equivalents ranked from 387 ± 10.9 μmol mannitol/g extract for basil to 47.5 ± 11.5 μmol mannitol/g extract for cumin which was not significantly different from the values found for cardamom and ginger (Fig. 1(e)). The order of performance was basil > laurel > parsley ≃ juniper ≃ fennel > aniseed > cardamom ≃ ginger ≃ cumin. In the nonsite-specific assay, basil and laurel performed best as well (Fig. 1(f)). Lowest ranks were shared by parsley, cardamom and ginger (not significantly different). Here, the order of performance was slightly different with basil > laurel > aniseed ≃ cumin > juniper > parsley ≃ cardamom ≃ ginger.

Correlations were found between the site-specific assay and total phenols ($r^2 = 0.6042$, $P < 0.05$), linoleic acid peroxidation ($r^2 = 0.5051$, $P < 0.05$) and iron reduction ($r^2 = 0.6713$, $P < 0.01$). The nonsite-specific assay

correlated with DPPH· scavenging ($r^2 = 0.6440$, $P < 0.05$), iron reduction ($r^2 = 0.6883$, $P < 0.01$), linoleic acid peroxidation ($r^2 = 0.6850$, $P < 0.01$) and total phenols ($r^2 = 0.6589$, $P < 0.01$). No significant correlation could be found with the other assays.

4. Discussion

In recent years, interest in plant-derived food additives has grown. Plant extracts might substitute synthetic food antioxidants, which may influence human health when consumed chronically (Martinez-Tome et al., 2001). Plant-derived food additives, especially polyphenolic compounds, have also been ascribed health-promoting properties, as for example in terms of prevention of chronic cardiovascular diseases (Harborne & Williams, 2000). Such food additives are required to be odour free and tasteless. Hydrodistilled extracts have the advantage that the essential oils which carry the intrinsic flavour of a spice have been removed and polyphenols, as the main compounds with antioxidant activity have been concentrated.

Hydrodistillation was performed by means of the hydrodistillation apparatus described in European Pharmacopoeia. During the extraction process, problems were observed with some spices. For parsley leaves, the formation of a kind of jelly could be observed. The extraction of cardamom led to filtration problems due to the starch occurring in the spice. The low extraction yields for both spices might be attributable to these observations.

The results of total phenol analyses show that three of the top five total phenol spices belong to the family of Apiaceae (cumin, fennel, parsley) whereas, in the spices derived from Zingiberaceae, only a small amount of total phenols can be found. The ratio of phenols to extraction yield can serve as a measure of the suitability of the hydrodistillation method for concentrating phenols in the plant extracts. Hydrodistillation seems to be a favourable extraction method for basil and laurel, but not appropriate for ginger and aniseed. Data suggest that these spices contain water-soluble nonphenolic material to a great extent as their extracts have a rather high content of nonFolin-Ciocalteu-reactive substances.

Phenolic substances have been reported for most of the examined spices. Prevalent phenolics in basil are rosmarinic acid and other phenolic acids (Jayasinghe, Gotoh, Aoki, & Wada, 2003). Parsley contains apigenin glycosides (Justesen & Knuthsen, 2001). Main phenolics are quercetin and kaempferol glycosides in cumin and fennel, whereas in aniseed quercetin, luteolin and apigenin glycosides were found (Kunzemann & Herrmann, 1977). In laurel, quercetin glycosides have been reported (Skerget et al., 2005), such as isoquercitrin (Kang et al., 2002) as well as kaempferol glycosides (Fiorini, David, Fouraste, & Vercauteren, 1998). Quercetin glycosides occur as well in juniper (Stefova, Kulevanova, & Stafilov, 2001) whereas cardamom contains mainly phenolic acids, such as caffeic acid, *p*-coumaric acid, and protocatechuic acid (Variyar, Bandyopadhyay, & Thomas, 1998). No polyphenols have been reported in ginger, yet the main constituents, gingerol and zingerone, possess a monophenolic moiety and may therefore be Folin-Ciocalteu-reactive (Masuda, Kikuzaki, Hisamoto, & Nakatani, 2004).

The good correlation between the results from total phenols analysis and the antioxidative assays has been previously reported (Zheng & Wang, 2001). Yet, we found no association between total phenols and the activity of the extracts as DPPH· scavengers although this has been found for other plant materials (Dorman, Bachmayer, Kosar, & Hiltunen, 2004; Dorman, Kosar, Kahlos, Holm, & Hiltunen, 2003). In our case, the lack of correlation may be due to the variety of the plant materials as in the cited references the plant material was quite homogeneous (different *Mentha* species and *Lamiaceae* plants, respectively).

In complex systems, such as food and food preparations, various different mechanisms may contribute to oxidative processes, such as in Fenton reactions, where transition metal ions play a vital role, different reactive oxygen species might be generated and various target structures such as lipids, proteins and carbohydrates, can be affected. Therefore, it is important to characterize the extracts by a variety of antioxidant assays (Halliwell, 1997). In the iron(III)-reduction assay, the general ability of the extracts to donate electrons is tested whereas, in the DPPH assay, hydrogen atoms are involved as well. One important mechanism of antioxidant action may be the chelation of iron(II) ions which serve as catalysts in Fenton reactions. Linoleic acid, a polyunsaturated fatty acid, acts as model lipid whereas, in the deoxyribose assay, the ability to protect carbohydrate structures is tested.

The results from the antioxidant assays show that all extracts can act as radical scavengers to a certain extent. Basil showed the highest activity in the iron reduction assay and in the inhibition of deoxyribose degradation. It shared the top ranking with laurel in the DPPH· assay. Yet, the IC_{50} values for these two extracts were still 2.5–5.5 times higher than those of the tested reference antioxidants, ascorbic acid, BHT and BHA, although the differences between basil and BHT were not significant. Laurel performed best in protecting linoleic acid from peroxidation whereas parsley was the top iron chelator among the examined spices.

Different extracts from the tested spices have been previously evaluated for their antioxidant activity and similar results to our findings have been reported. Aqueous-methanolic extracts from basil and laurel showed good results in scavenging DPPH· and hydroxyl radicals

(Kosar, Dorman, & Hiltunen, 2005). For methanolic extracts from parsley leaves, Fejes et al. (2000) found considerable antioxidant activity against lipid peroxidation. Water extracts from aniseed showed a better inhibition of lipid peroxidation than alcohol extracts (Al-Ismail & Aburjai, 2004). Water and alcohol extracts from fennel performed nearly as well as BHT and BHA in antioxidant assays (Oktay, Gülcin, & Küfrevioglu, 2003). For laurel, antioxidant activity has been found for methanolic and ethanolic extracts (Kang et al., 2002; Simic, Kundakovic, & Kovacevic, 2003). Zheng and Wang (2001) found for water extracts from fresh bay leaves a higher antioxidant activity than for basil and parsley. Badei, El-Akel, Faheid, and Mahmoud (2002) showed that powdered cardamom and its essential oil both protect cookie lipids against peroxidation. A 10% alcohol extract from juniper berries decreased lipid peroxidation of rape-seed oil (Takacsova, Pribela, & Faktorova, 1995).

Ginger showed a lower total antioxidant activity than did basil and parsley in experiments by Wu et al. (2004). Rababah, Hettiarachchy, and Horax (2004) found a 60% ethanol extract from ginger to be less effective in the inhibition of lipid peroxidation than equivalent amounts of vitamin E. The IC_{50} value in lipid peroxidation was about 300 times higher for a 50% ethanol extract from ginger than for BHT (Shobana & Naidu, 2000). In investigations by Murcia et al. (2004) a 5% water extract from ginger yielded nearly the same antioxidant activity toward lipid peroxidation as did BHT, both applied in a dose of 0.1%. A similar result was found for a dichloromethane extract from ginger powder (Rehman, Salariya, & Habib, 2003). For ginger, it seems advisable to use extraction media which are able to extract the lipophilic antioxidant compounds. The results from Murcia et al. (2004) seem to contradict our findings, but they used an extraction method which might have preserved essential oils in the extracts. The antioxidant activity of essential oils from ginger has recently been shown (Dang, Takacsova, Nguyen, & Kristianova, 2001).

The results from the inhibition of deoxyribose degradation might also reveal some of the antioxidant mechanisms of the extracts. In the nonsite-specific assay, EDTA forms a complex with iron(III), and hydroxyl radicals are generated in solution. In the site-specific assay, EDTA is omitted. Therefore, iron(III) can bind to deoxyribose and produce hydroxyl radicals at this site. So iron chelating compounds can reduce the extent of deoxyribose degradation even if they are not effective hydroxyl radical-scavengers. In the nonsite-specific assay, this only influences the results if the compounds form a more stable complex with iron(III) than EDTA. Extracts which perform better in the site-specific than in the nonsite-specific assay are supposed to be better iron chelators than hydroxyl radical scavengers. Considerable divergences between the performance in the site-specific and the nonsite-specific assays were found for parsley, juniper, aniseed and cumin. Parsley and juniper performed better in the site-specific assay; thus they have a higher iron chelation ability than scavenging activity for hydroxyl radicals. With aniseed and cumin, the scavenging activity is higher than the iron chelation ability.

In this study, we present results on the antioxidant activity of hydrodistilled extracts from nine selected herbs and spices. The extracts from basil and laurel were found to have the highest antioxidant activity in the assays, with the exception of iron chelation. Although parsley showed the best performance in the iron chelation assay, it was less effective at retarding the oxidation of linoleic acid.

One gramme of laurel extract was as effective as about 212 mg of trolox in the prevention of lipid peroxidation. Basil extract offers the same protection as 177 mg trolox. An optimization of the extraction process could lead to an even better ratio. As the extracts are better soluble in aqueous media than are the synthetic antioxidants, they offer a promising alternative as food ingredients with antioxidant activity. In future experiments, it would be interesting to investigate the effectiveness of the extracts from laurel leaves and basil herb in different food systems.

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