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Antioxidative Activity of Flavonoids Isolated from Jindalrae Flowers (*Rhododendron mucronulatum* Turcz.)

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Abstract : Seven antioxidative flavonoids were isolated from Jindalrae flowers (*Rhododendron mucronulatum* Turcz.), an edible plant in Korea. These compounds were identified as afzelin, ampelopsin, catechin, myricetin, myricitrin, quercetin and quercitrin on the basis of IR, UV, FAB-MS, ^{1}H NMR, and ^{13}C NMR data. These compounds were consisted of two flavonols, three flavonol glycosides, a flavane, and a dihydroflavonol. The flavonol glycosides (14.4 g) present in both ethyl ether and ethyl acetate fractions comprised up to 82% of their total flavonoid amount (17.6 g) finally recovered by means of polyamide C-200 column chromatography, preparative TLC, recrystallization, and Sephadex LH-20 column chromatography. The antioxidant activities were measured in an ethanol solution of linoleic acid in the presence of ferric thiocyanate. The antioxidant efficiency increased in the order of afzelin<α-tocopherol<catechin<quercitrin<quercetin<myricitrin<ampelopsin<myricetin(Received May 16, 1996; accepted June 21, 1996).

Introduction

A large number of plants, which have been used as food and medicine in Asia, often based on the traditional prescriptions in literature, have been supplying spices, beverages and medicines to the other parts of the world. Many of flavonoids and their related compounds produced by these plants, have been important sources for the development of many modern medicines. Flavonoids are widely distributed in edible plants, primarily in the form of flavonol glycosides.[1)] It has been estimated that humans consuming high fruit and vegetable content diets ingest up to 1 g of these compounds daily.[2)]

Jindalrae flowers and their leaves have long been known to have high pharmacological potency such as tonic, diuretic and stomachic in Chinese medicine.[3,4)] For this reason, Jindalrae flowers have also been used for raw materials of colorful Korean traditional dishes.[5)] Home-made Jindalrae flower wine showed a significant antioxidative activity in an *in vitro* fatty acid peroxidation assay, so that we have started in an investigation concerning antioxidants.

Although many of the flavonoids and their related compounds isolated from the flowers of the other members of *Rhododendron* were already investigated,[6-9)] no investigation has been made on flavonoids of Jindalrae flowers. In the course of our investigation on antioxidative constituents, we isolated and characterized seven flavonoids as antioxidative principles in Jindalrae flowers.

Materials and Methods

General procedure and materials

Melting points were measured on a Büchi 535 melting point apparatus and are uncorrected. IR spectra were recorded on a JASCO FT/IR-5000. ^{1}H and ^{13}C Nuclear magnetic resonance (NMR) spectra were measured 500 and 125 MHz, respectively, on a Bruker AM-500. Fast atom bombardment mass spectrometry (FAB-MS) was carried out with a VG ZAB-2F and electrospray mass spectrometry (ES-MS) on a VG Quattro-BQ. UV/visible spectra were recorded on a Shimadzu UV-2100.

Fresh Jindalrae flowers were collected at Tongyoung, Kyoungsangnam-do, Korea, in April 1993. Solvents for extraction and shift reagents for UV measurement, and polyamide C-200 for column chromatography were purchased from Wako Pure Chemical Industries Ltd. (Osaka, Japan) and then α-tocopherol, ammonium thiocyanate, and linoleic acid from Aldrich Chemical Company (Milwaukee, WI, USA). Catechin, myricetin and quercetin were obtained from Roth (Karlsruhe, Germany) and quercitrin from Sigma Chemical Company (St. Louis, MO, USA).

Extraction and Isolation

Jindalrae flowers (20.5 kg) were extracted twice with

Key words : Antioxidants; flavonoids; Jindalrae; Korean azalea flower; *Rhododendron mucronulatum;* Ericaceae
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40 L of 80% MeOH and of 50% MeOH at room temperature, respectively. The aqueous methanol extracts were combined and concentrated under the reduced pressure to remove most of MeOH. The remaining aqueous solution was successively partitioned with n-hexane, ethyl ether and ethyl acetate, resulting in four fractions of differing polarity, namely, n-hexane (0.12 g), ethyl ether (8.12 g), ethyl acetate (41 g), and water fraction (245.92 g).

Total of the ethyl ether fraction was adsorbed onto polyamide C-200 (4.5×50 cm) and continuously eluted by using a gradient of 45~100% MeOH in toluene to provide six fractions : A (45% MeOH), B (50% MeOH), C (65% MeOH), D (75% MeOH), E (85% MeOH) and F (100% MeOH). These fractions were spotted on TLC plates precoated with silica gel 60 F_{254} (particle size, 250 μm, Merck). The plates were developed with a chloroform/methanol/acetic acid (80/20/1) or a ethyl acetate/methyl ethyl ketone/formic acid/water/benzene mixture (40/30/10/10/20) and then completely air-dried in a draft chamber. Flavonoid zones (compound **1** from F, **2** from E, **3** from C, **4** from D and E, **5** from E, **6** and **7** from E) separated on the plates were detected under UV light (350 nm) without and with ammonia vapour (UV/NH_3) and subsequently scraped off with a spatula. Each scraped-off zone (UV : dark purple, UV/NH_3 : yellow fluorescence) was placed in a 300 m*l* round bottom flask containing an adequate amount of MeOH and gently heated for 20 min, occasionally swirling it. Subsequently, the hot mixture was filtered on a Büchner funnel, followed by washing with MeOH. The resulting filtrate was subsequently concentrated to one-third of its original volumn *in vacuo* with a rotary evaporator and precipitated by adding a small amount of nonpolar solvent such as chloroform, followed by filtration on a Büchner funnel. The precipitate was washed with cold-methanol, followed by purification on a column packed with Sephadex LH-20 (3×25 cm, Pharmacia Biotech., Sweden).

8.2 g portions of the ethyl acetate fraction were applied to five of the numbered polyamide C-200 columns (4.5×50 cm). The columns were eluted stepwise with distilled water, 20% MeOH, 40% MeOH, 60% MeOH, 80% MeOH, and 100% MeOH and then by using a gradient of 20~100% benzene in MeOH to provide five fractions : A-1 (20% benzene : compound **4**, **5** and **6**), B-1 (40% benzene : compound **4** and **5**), C-1 (60% benzene), D-1 (80% benzene) and E-1 (100% benzene). Each separated fraction was evaporated *in vacuo*, followed by purification with a preparative TLC, recrystallization, and Sephadex LH-20 column chromatography as same methods mentioned above.

Antioxidative Assay in Solution

The antioxidative activities of n-hexane, ethyl ether and ethyl acetate extracts, and seven purified flavonoids were tested by ferric thiocyanate assay according to Nakatani *et al.*[10] Each sample dissolved in 120 μ*l* of ethanol was added to a reaction mixture in a screw-cap test tube. Each reaction mixture consisted of 2.88 m*l* of 2.51 % linoleic acid in ethanol and 9 m*l* of 40 mM phosphate buffer (pH 7.0). The known natural antioxidant, α-tocopherol, was used as a comparative standard to evaluate the antioxidative activity of each sample. The test tube was covered with aluminum foil to eliminate the influence of the light upon the lipid peroxidation and then placed in an incubator at 40℃. At daily intervals during an incubation period, a 0.1 m*l* aliquot of the mixture was diluted with 9.7 m*l* of 75% ethanol, followed by addition of 0.1 m*l* of 30% ammonium thiocyanate. Precisely 3 min after the addition of 0.1 m*l* of 20 mM ferrous chloride in 3.5% hydrochloric acid to the reaction mixture, the absorbance was measured at 500 nm.

Results and Discussion

Structural Analysis of Compounds 1~7.

Physicochemical properties of seven compounds isolated from Jindalrae flowers were as follows.

Compound **1** (0.6 g) : Yellow crystal; mp 317~318℃; FAB-MS *m/z* 303 $[M+H]^+$. The other spectral data were identical with those reported on the literature.[11,12] The original spectra and physical data are available from the corresponding author upon request.

Compound **2** (0.4 g) : Light-yellow crystal; mp 324℃ (dec.); IR ν_{max}^{KBr} cm^{-1} 3314 (OH), 1660 (>C=O), 1595 (conjugated diene), 1552 (phenyl), 1519, 1455, 1225 (C-O-C), 834; UV λ_{max}^{MeOH} nm 254, 272 (sh), 301 (sh), 373; (+NaOMe) 284 (sh), 325, 423 (dec.); (+$AlCl_3$) 235 (sh), 269, 456; (+$AlCl_3$+HCl) 265, 310 (sh), 358 (sh), 432; (+NaOAc) 254 (sh), 267, 324 (sh), 383; (+NaOAc+H_3BO_3) 259, 295 (sh), 389; FAB-MS *m/z* 319 $[M+H]^+$; ^1H NMR (CD_3OD) δ 7.35 (2H, *s*, H-2′ & H-6′), 6.37 (1H, *d*, *J*=1.8, H-8), 6.18 (1H, *d*, *J*=1.7, H-6); ^{13}C NMR (CD_3OD) δ 148.3 (*s*, C-2), 137.2 (*s*, C-3), 177.5 (*s*, C-4), 158.4 (*s*, C-5), 99.5 (*d*, C-6), 165.8 (*s*, C-7), 94.7 (*d*, C-8), 162.7 (*s*, C-9), 104.8 (*s*, C-10), 123.4 (*s*, C-1′), 108.8 (*d*, C-2′ & C-6′), 147.0 (*s*, C-3′ & C-5′), 137.6 (*s*, C-4′).

Compound **3** (1.6 g) : Yellow needle; mp 172~174℃; IR ν_{max}^{KBr} cm^{-1} 3413 (OH), 1652 (>C=O), 1604 (conjugated diene), 1584 (phenyl), 1440 (rhamnosyl CH_3), 1256 (ether), 831; UV λ_{max}^{MeOH} nm 265, 337; (+NaOMe) 273, 327, 389; (+$AlCl_3$) 273, 302, 346, 396; (+$AlCl_3$+HCl) 274, 300, 341, 392; (+NaOAc) 270, 290 (sh), 353; (+NaOAc+H_3BO_3) 265, 350; FAB-MS *m/z* 433 $[M+H]^+$; ^1H NMR (CD_3OD) δ 7.78 (2H, *d*, *J*=8.5, H-2′ & H-6′), 6.95 (2H, *d*, *J*=8.7, H-3′ & H-5′), 6.40 (1H, *d*, *J*=1.9, H-8), 6.22 (1H, *d*, *J*=1.8, H-6), 5.40 (1H, *d*, *J*=1.7, H-

1″), 4.28 (1H, *dd*, *J*=1.7, 3.2, H-2″), 3.75 (1H, *dd*, *J*=3.2, 9.3, H-3″), 3.38~3.34 (2H, *m*, H-4″ & H-5″), 0.96 (3H, *d*, *J*=6.1, H-6″); ^{13}C NMR (CD_3OD) δ 159.0 (*s*, C-2), 136.4 (*s*, C-3), 179.9 (*s*, C-4), 163.2 (*s*, C-5), 99.6 (*d*, C-6), 165.2 (*s*, C-7), 95.1 (*s*, C-8), 159.6 (*s*, C-9), 106.4 (*s*, C-10), 123.2 (*s*, C-1′), 132.0 (*d*, C-2′ & C-6′), 116.8 (*d*, C-3′ & C-5′), 161.9 (*s*, C-4′), 103.5 (*d*, C-1″), 73.5 (*d*, C-2″), 72.5 (*d*, C-3″), 72.3 (*d*, C-4″), 69.6 (*d*, C-5″), 17.9 (*q*, C-6″).

Compound 4 (7.6 g) : Pale-yellow powder; mp 240℃ (dec.); IR ν_{max}^{KBr} cm^{-1} 3457 (OH), 1656 (>C=O), 1608 (conjugated diene), 1508 (phenyl), 1458 (rhamnosyl CH_3), 1065 (ether), 837; UV λ_{max}^{MeOH} nm 256, 265 (sh), 320 (sh), 352; (+NaOMe) 271, 327, 397; (+$AlCl_3$) 272, 295 (sh), 422; (+$AlCl_3$+HCl) 267, 291 (sh), 355, 400; (+NaOAc) 269, 326, 365; (+NaOA+H_3BO_3) 260, 278 (sh), 369; FAB-MS *m/z* 449 $[M+H]^+$; ^{1}H NMR (CD_3OD) δ 7.37 (1H, *d*, *J*=1.9, H-2′), 7.34 (1H, *dd*, *J*=1.9, 8.3, H-6′), 6.94 (1H, *d*, *J*=8.3, H-5′), 6.38 (1H, *d*, *J*=1.7, H-8), 6.22 (1H, *d*, *J*=1.7, H-6), 5.39 (1H, *d*, *J*=1.7, H-1″), 4.26 (1H, *dd*, *J*=1.7, 3.2, H-2″), 3.75 (1H, *dd*, *J*=3.2, 9.3, H-3″), 3.45~3.44 (1H, *m*, H-4″), 3.39~3.35 (1H, *m*, H-5″), 0.98 (3H, *d*, *J*=5.9, H-6″); ^{13}C NMR (CD_3OD) δ 158.1 (*s*, C-2), 136.5 (*s*, C-3), 179.4 (*s*, C-4), 162.9 (*s*, C-5), 100.5 (*d*, C-6), 165.7 (*s*, C-7), 95.4 (*d*, C-8), 158.7 (*s*, C-9), 106.1 (*s*, C-10), 122.4 (*s*, C-1′), 116.7 (*d*, C-2′), 146.8 (*s*, C-3′), 149.0 (*s*, C-4′), 117.2 (*d*, C-5′), 122.4 (*d*, C-6′), 103.8 (*d*, C-1″), 72.3 (*d*, C-2″), 72.4 (*d*, C-3″), 73.6 (*d*, C-4″), 72.0 (*d*, C-5″), 18.0 (*q*, C-6″).

Compound 5 (5.2 g) : Pale-yellow powder; mp 270℃ (dec.); IR ν_{max}^{KBr} cm^{-1} 3400 (OH), 1666 (>C=O), 1606 (conjugated diene), 1500 (phenyl), 1460 (rhamnosyl CH_3), 1066 (ether), 837; UV λ_{max}^{MeOH} nm 258, 325 (sh), 353; (+NaOMe) 267, 325, 398; (+$AlCl_3$) 267, 285 (sh), 426; (+$AlCl_3$+HCl) 270, 290 (sh), 398; (+NaOAc) 270, 325, 392; (+NaOAc+H_3BO_3) 258, 296 (sh), 389; FAB-MS *m/z* 465 $[M+H]^+$; ^{1}H NMR (CD_3OD) δ 7.16 (2H, *s*, H-2′ & H-6′), 6.39 (1H, *d*, *J*=1.9, H-8), 6.23 (1H, *d*, *J*=1.9, H-6), 5.35 (1H, *d*, *J*=1.7, H-1″), 4.25 (1H, *dd*, *J*=1.7, 3.2, H-2″), 3.74 (1H, *dd*, *J*=3.2, 9.3, H-3″), 3.39~3.34 (2H, *m*, H-4″ & H-5″), 0.99 (3H, *d*, *J*=6.0, H-6″); ^{13}C NMR (CD_3OD) δ 158.8 (*s*, C-2 & C-9), 136.5 (*s*, C-3), 179.9 (*s*, C-4), 163.5 (*s*, C-5), 99.9 (*d*, C-6), 166.3 (*s*, C-7), 95.0 (*d*, C-8), 106.1 (*s*, C-10), 121.4 (*s*, C-1′), 109.9 (*d*, C-2′ & C-6′), 147.2 (*s*, C-3′ & C-5′), 136.5 (*s*, C-4′), 103.8 (*d*, C-1″), 72.3 (*d*, C-2″), 72.4 (*d*, C-3″), 73.7 (*d*, C-4″), 72.2 (*d*, C-5″), 17.3 (*q*, C-6″).

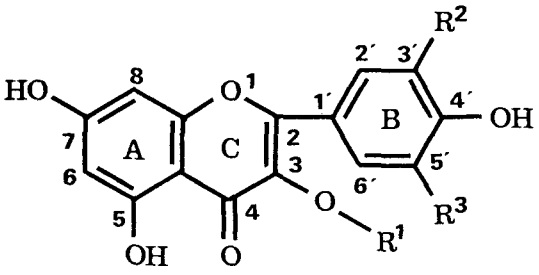
Compound 6 (2.0 g) : Greenish-yellow powder; mp 173~175℃; IR ν_{max}^{KBr} cm^{-1} 3411 (OH), 1612 (alkene), 1515 (phenyl), 1460 (methylene), 1021, 832; UV λ_{max}^{MeOH} nm 280; FAB-MS *m/z* 291 $[M+H]^+$; ^{1}H NMR (CD_3OD) δ 6.86 (1H, *d*, *J*=1.8, H-2′), 6.79 (1H, *dd*, *J*=1.6, 8.1, H-6′), 6.74 (1H, *d*, *J*=8.2, H-5′), 5.96 (1H, *d*, *J*=2.1, H-8), 5.88 (1H, *d*, *J*=2.1, H-6), 4.59 (1H, *d*, *J*=7.5, H-2), 3.99 (1H, *ddd*, *J*=5.4, 7.5, 8.2, H-3), 2.87 (1H, *dd*, *J*=5.4, 16.1, H-4(ax)), 2.53 (1H, *dd*, *J*=8.2, 16.1, H-4(eq)); ^{13}C NMR (CD_3OD) δ 83.1 (*d*, C-2), 69.0 (*d*, C-3), 28.7 (*t*, C-4), 157.8 (*s*, C-5), 96.6 (*d*, C-6), 157.1 (*s*, C-7), 95.8 (*d*, C-8), 158.0 (*s*, C-9), 101.1 (*s*, C-10), 132.4 (*s*, C-1′), 115.5 (*d*, C-2′), 146.5 (*s*, C-3′ & C-4′), 116.4 (*d*, C-5′), 120.4 (*d*, C-6′).

Compound 7 (0.2 g) : Needles; mp 245~246℃; IR ν_{max}^{KBr} cm^{-1} 3300 (OH), 1650 (>C=O), 1480 (methylene), 1170 (C-CO-C), 1090 (C-O-C), 830 (phenyl); UV λ_{max}^{MeOH} nm 291, 330 (sh); (+NaOMe) 246 (sh), 323; (+$AlCl_3$) 275 (sh), 351; (+$AlCl_3$+HCl) 252 (sh), 316; (+NaOAc) 326, 336 (sh); (+NaOAc+H_3BO_3) 314, 328 (sh); FAB-MS *m/z* 321 $[M+H]^+$; ^{1}H NMR (CD_3OD) δ 6.62 (2H, *s*, H-2′ & H-6′), 6.00 (1H, *d*, *J*=1.8, H-8), 5.94 (1H, *d*, *J*=1.8, H-6), 4.59 (1H, *d*, *J*=10.8, H-3), 4.85 (1H, *d*, *J*=10.8, H-2); ^{13}C NMR (CD_3OD) δ 84.5 (*d*, C-2), 73.4 (*d*, C-3), 198.8 (*s*, C-4), 161.0 (*s*, C-5), 97.4 (*d*, C-6), 168.7 (*s*, C-7), 96.5 (*d*, C-8), 164.6 (*s*, C-9), 102.2 (*s*, C-10), 129.4 (*s*, C-1′), 108.8 (*d*, C-2′ & C-6′), 146.9 (*s*, C-3′ & C-5′), 135.7 (*s*, C-4′).

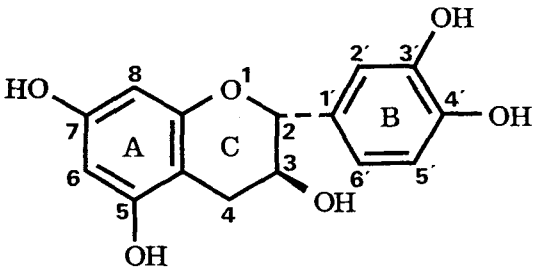
Compound **1** was identified as quercetin by matching most spectral data of authentic sample.

On the basis of IR and UV spectral data, compound **2** suggested the presence of a 3, 5, 7, 3′, 4′, 5′-hexahydroxyl system flavone. The ^{1}H NMR showed the presence of 2 aromatic protons on the A and B ring, respectively. The FAB-MS indicated an M+1 ion peak at 319, suggesting that the molecular weight is greater an oxygen atom portion than that of compound **1**. The ^{13}C NMR also exhibited signals suitable for 15 carbon atoms. Since all of the spectral data agreed with the structure, compound **2** was confirmed as myricetin having a molecular formula of $C_{15}H_{10}O_8$. On the other hand, these data were mostly identical with those reported on the literature.[13]

The IR and UV spectral data of compounds **3, 4** and **5** suggested the presence of 5, 7, 4′-trihydroxylflavone, 5, 7, 3′, 4′-tetrahydroxylflavone, and 5, 7, 3′, 4′, 5′-pentahydroxylflavone, respectively. The NMR data of all these compounds commonly indicated the presence of *O*-glycosidic linkages due to rhamnopyranoside at the C-3 position. In confirmation of the glycosylation site, the ^{13}C NMR data were more informative than the other spectral data. In the case of flavonol-3-*O*-glycosides, their glycosylation sites were confirmed by downfield shifts (about 10.3, 1.95, 0.6 and 1.1 ppm, respectively) of C-2, C-4, C-5 and C-10 and the upfield shift (about 1.05 ppm) of the C-3 as compared with those corresponding to their aglycones.[14,15] The molecular formulas of compounds **3, 4** and **5** were suggested to be $C_{21}H_{20}O_{10}$, $C_{21}H_{20}O_{11}$ and $C_{21}H_{20}O_{12}$, respectively, by FAB-MS in conjunction with the ^{1}H and ^{13}C NMR data. Therefore, these compounds were identified as afzelin (kaempferol-3-*O*-rhamnopyra-



	R^1	R^2	R^3
1	H	OH	H
2	H	OH	OH
3	Rha	H	H
4	Rha	OH	H
5	Rha	OH	OH



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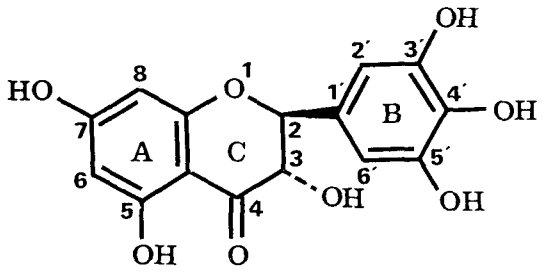

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Fig. 1. Chemical structures of flavonoids isolated from Jindalrae flower.

noside), quercitrin (quercetin-3-*O*-rhamnopyranoside), and myricitrin (myricetin-3-*O*-rhamnopyranoside), respectively, by various spectral data as mentioned above.[16-19]

The IR and UV spectral data of compound **6** suggested the presence of a chroman ring. These reasons were based on the disappearance of the characteristic absorption at about 1660 and 1600 cm^{-1} and nearly a stationary band in various shift reagents. The ^1H and ^{13}C NMR showed that a carbonyl group on the γ-pyrone ring is replaced by a methylene group and that the 2, 3-double bond is reduced by hydrogenation. Therefore, this compound was suggested to be a flavane on sufficent evidence by the NMR data. On the basis of FAB-MS and the NMR data, this compound was identified as catechin having a molecular formula of $C_{15}H_{14}O_6$.[20-22]

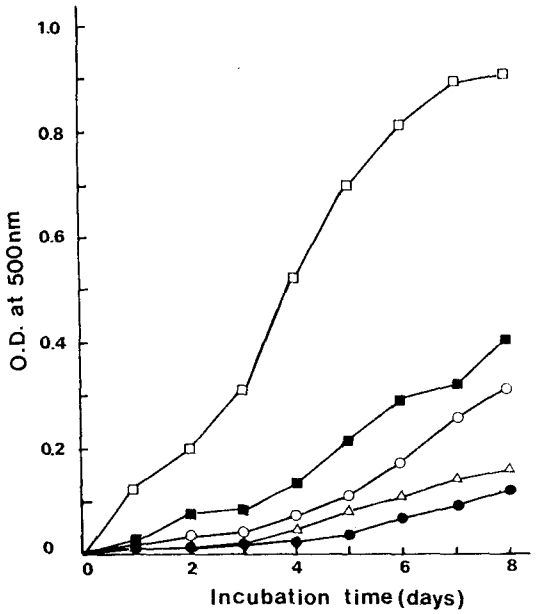

Fig. 2. Antioxidative activity of three different solvent extracts on α-tocopherol at the level of 100 ppm. (●—●, ethyl acetate ext.; ○—○, ethyl ether ext.; ■—■, hexane ext.; △—△, α-tocopherol; □—□, control).

The IR and UV spectral data of compound **7** suggested the presence of a chromanone ring. There was enough evidence that compound **7** is a dihydroflavonol, because a chromanone ring shows the characteristic absorption at 1650, 1480, 1170 and 1090 cm^{-1} on the IR spectrum and the unique shift on UV spectra used various shift reagents. On the basis of FAB-MS and the NMR data, this compound was confirmed as ampelopsin (dihydromyricetin) having a molecular formula of $C_{15}H_{12}O_8$. This compound showed greater 2 hydrogen atoms portion than that of compound **2**. The chemical structures of all compounds identified above were as shown in Fig. 1. These compounds consisted of 82% flavonol glycoside, 11% flavane, 6% flavonol, and 1% dihydroflavonol.

Antioxidant Activities of Crude Extracts and Compound 1 through 7

Antioxidant activities of three crude extracts were investigated by measuring the hydroperoxidation of linoleic acid *via* radical chain reaction. Fig. 2 shows the results obtained from antioxidant testing of the crude extracts. Although the ethyl acetate extract showed better activities than those of α-tocopherol, the ethyl ether extract exhibited slightly less efficiency than those of α-tocopherol. On the other hand, the hexane extract showed pro-oxidative activity, suggesting the presence of pro-oxida-

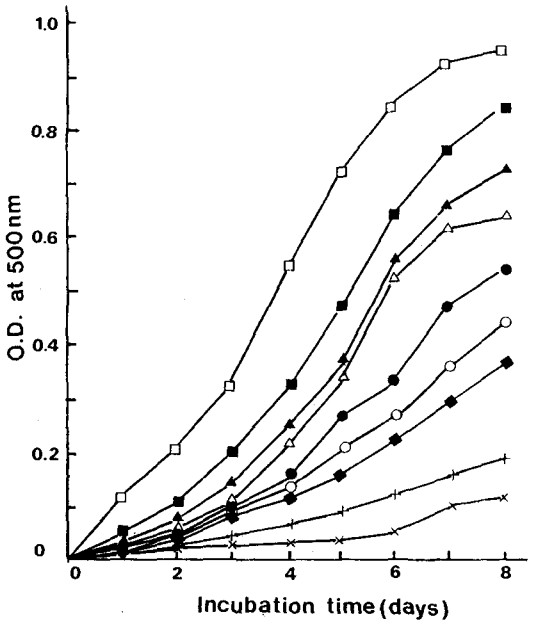

Fig. 3. Antioxidative activity of flavonoids isolated from Jindalrae flower on α-tocopherol at the level of 10 ppm. (×−×, myricetin; +−+, ampelopsin; ◆−◆, myricitrin; ○−○, quercetin; ●−●, quercitrin; △−△, catechin; ▲−▲, α-tocopherol(10 ppm); ■−■, afzelin; □−□, control).

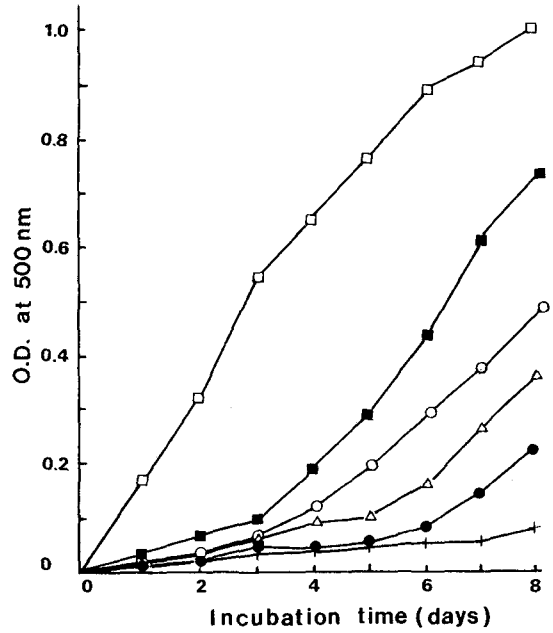

Fig. 4. Antioxidative activity of quercitrin on α-tocopherol at the level of 1, 3, 5 and 10ppm. (+−+, 10; ●−●, 5; △−△, 3; ○−○, 1; □−□, control; ■−■, α-tocopherol(10 ppm)).

nts. The antioxidant activity of these extracts increased in the order of hexane<ethyl ether<α-tocopherol<ethyl acetate. However, this result indicated that ethyl acetate extract may play an important role in the prevention of lipid peroxidation. The antioxidative activities of all compounds identified above are shown in Fig. 3. The antioxidant efficiency of these compounds increased in the order of afzelin<α-tocopherol<catechin<quercitrin<quercetin<myricitrin<ampelopsin<myricetin. This result showed that afzelin exhibited activities almost equivalent to those of α-tocopherol. On the other hand, the remaining compounds were more effective than α-tocopherol used as a comparative standard. All compounds identified in the present study commonly possess meta-dihydroxyl groups at both the 5 and 7 positions of chroman, chromanone and chromone systems. Since these compounds have two hydroxyl groups at the same positions, no conclusions can be drawn from the data presented concerning the importance of these groups. In the case of flavonol-3-*O*-glycosides, the antioxidant activities increased in the order of afzelin<quercitrin<myricitrin. The hydroxylation patterns and an increase in the level of hydroxylation on the B ring may cause the variation of the antioxidant efficiency.[23-25] However, this result suggested that the B ring definitely acts as an important factor in the estimation of the antioxidant activity. On the other hand, aglycones were more potent in their antioxidative activity than their corresponding glycosides in the present study. Flavonoids which have the ability to chelate metal ions by means of either the 3-hydroxy, 4-keto grouping or the 5-hydroxy, 4-keto grouping, may reduce the pro-oxidative activity of trace metals. Ampelopsin was found to have definitely less antioxidant activity than myricetin, indicating that the 2, 3-double bond is of major importance to antioxidant activity. Summarizing the relations between structure and antioxidant activity from the limited number of comparisons available in the present study, we concluded that the following structural characteristics were correlated with antioxidant activity: 1) dihydroxylation of the B ring increases antioxidant activity, 2) unsaturation of the γ-pyrone ring is essential for activity, 3) 5, 7-dihydroxylation of the chromone system did not decrease activity, and 4) 3-glycosylation of 3-hydroxyflavone altered its activity. The influence of glycosylation at other hydroxyl groups in the flavone structure need to be investigated further.

Fig. 4 shows the evaluation of antioxidative activity of quercitrin at the level of 1, 3, 5 and 10 ppm, respectively. This result suggested that quercitrin at the level of 1 ppm has greater antioxidative activities than those of α-tocopherol at the level of 10 ppm. However, flavonol-3-*O*-glycosides are the major components so that they may play an important role in antioxidant activity of Jindalrae flowers. Since flavonoids are widely distributed in various foods, the detailed investigations on the biological effects of the compounds would be quite valuable. Furthermore, characterization of unidentified phenolic compounds is necessary and is currently in progress.

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진달래꽃으로부터 분리된 플라보노이드 화합물의 항산화성에 관한 연구

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초록 : 우리나라에서 식용으로 이용되는 진달래꽃으로부터 7성분의 항산화성이 있는 플라보노이드 화합물이 분리 동정되었다. 이들 화합물의 구조는 IR, UV, FAB-MS, ^{1}H NMR 및 ^{13}C NMR에 의해서 얻어진 분광학적인 결과에 근거하여 afzelin, ampelopsin, catechin, myricetin, myricitrin, quercetin 및 quercitrin인 것으로 밝혀졌다. 이들 화합물은 2개의 flavonol, 3개의 flavonol glycoside, 1개의 flavane 및 1개의 dihydroflavonol로 이루어졌다. 에틸에테르 및 초산에틸 구분에 존재하는 flavonol glycoside (14.4 g)는 polyamide C-200 관 크로마토피법, 분취용 박층크로마토그래피법, 재결정화법, sephadex LH-20 관 크로마토그래피법을 통해서 양쪽 구분으로부터 최종적으로 회수된 총 flavonoid량 (17.6 g)의 82%에 달하였다. 항산화성은 티오시안산철의 존재하에서 리놀레산의 에타놀용액 중에서 측정되었다. 항산화 효능성은 afzelin<α-tocopherol<catechin<quercitrin<quercetin<myricitrin<ampelopsin<myricetin의 순서로 증대되었다.

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