

Total Phenolics, Flavonoids, and Anthocyanin Contents of Six *Vireya Rhododendron* from Indonesia and Evaluation of their Antioxidant Activities

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ABSTRACT

Six species of *Vireya Rhododendron* from Indonesia, i.e. *Rhododendron javanicum*, *R. jasminiflorum*, *R. konori*, *R. malayanum*, *R. retusum* and *R. seranicum* were analyzed for total phenolics, flavonoids, anthocyanin and antioxidant activities of their ethanolic extract of leaves and twigs. Total phenolics, flavonoids and anthocyanin contents varied from 11.98–48.11 mg equivalent gallic acid/g dried powder, 0.78–8.92 mg equivalent quercetin/g dried powder and 0.12–0.29 mg equivalent cyanidin-3-glucoside/g dried powder, respectively. Antioxidant activities were investigated by using 2,2-diphenyl-1-picrylhydrazyl (DPPH), ferric reducing antioxidant power (FRAP), and cupric ion reducing antioxidant capacity (CUPRAC). From the twig bark extracts, *R. retusum* has the highest antioxidant capacity by DPPH method of about 36.68 $\mu\text{mol trolox/g}$ dry weight. Among the leaves extracts, *R. konori* has the highest antioxidant capacity of about 941.76 and 1793.62 $\mu\text{mol trolox/g}$ dried powder by FRAP and CUPRAC method, respectively. From the results obtained, only total phenolics content correlated with the antioxidant measured by DPPH, FRAP, and CUPRAC. Consequently, the antioxidant activity of *Vireya Rhododendron* is not only attributed to the flavonoids and anthocyanin, but also from other phenolics compounds.

INTRODUCTION

The genus *Rhododendron* belongs to the family of Ericaceae and comprising of 8 subgenera with the number of species around 900 species worldwide (Bhattacharya, 2011). The species are widely distributed from Europe, North America, Asia, to northern Australia (Popescu and Kopp, 2013). Many of *Rhododendron* species are known to have many health benefits

(Rezk *et al.*, 2015). Traditionally, plants from this genus are used to treat inflammation, pain, influenza, skin diseases and digestive disorders (Popescu and Kopp, 2013).

Several *Rhododendron* species have been analyzed for their effects as antimicrobial agents, antifungal, antidiabetic, antidiarrheal, and antioxidants (Jaiswal *et al.*, 2012; Lin *et al.*, 2014). Antioxidant and antimicrobial are the most significant biological activities from *Rhododendron* species. The antioxidant has been believed to have major roles in the body's defense system against reactive oxygen species that can induce oxidative stress. From the previous studies, many *Rhododendron* plants throughout Asia showed antioxidant activities, including *R. ponticum*, *R.*

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luteum, *R. smirnovii*, *R. caucasicum*, *R. arboreum*, *R. lepidotum*, *R. campanulatum*, *R. anthopogon*, *R. setosum*, and *R. brachycarpum* (Popescu and Kopp, 2013).

Indonesia has around 200 species of *Rhododendron* that grow from Sumatra to Papua and some are endemic. This plant needs cold weather, so most of *Rhododendron* species in Indonesia grow in the cool areas, such as in the highland or mountain (above 1000 m from the sea level). However, some species can be found in the lowland area and as epiphytes in the mangrove forests. *Rhododendron* from Indonesia come mostly from the *Vireya* group, with Kalimantan and Papua become the two hotspots for the distribution of *Rhododendron* species in Indonesia (Argent, 2006).

About 208 phytochemical compounds from various *Rhododendron* species have been successfully identified (Qiang *et al.*, 2011). Flavonoids as their aglycons or glycosides and diterpenoids are known to be the major class of phytochemical compounds found in *Rhododendron* species. As we have known earlier, most of the flavonoids exhibit antioxidant activities. Harbilas *et al.* (2009) showed that there was a positive correlation between phenolics content and antioxidant activity of *R. tomentosum* leaves. Also, isolation of flavonoid compound, i.e., myricitrin-5-methyl ether, in *R. yedonense* flower showed antioxidant activity (Jung *et al.*, 2007).

Available information on the evaluation of antioxidant activities and phytochemical contents of *Vireya* *Rhododendron* species grow in Indonesia is very limited. Therefore, in this paper, we evaluate the antioxidant activities and the correlation with the phenolics contents (total phenolics, flavonoids, and anthocyanin) from the leaves and twigs of several species of *Vireya* *Rhododendron* from Indonesia, namely *R. jasminiflorum*, *R. javanicum*, *R. konori*, *R. malayanum*, *R. retusum* and *R. seranicum* for the first time. We chose only the leaves and twigs because they are easy to collect and being the parts most widely studied in other *Rhododendron* species. Measurement of antioxidant activities was performed using 2,2-diphenyl-1-picrylhydrazyl (DPPH) method, ferric reducing antioxidant power (FRAP) and cupric ion reducing antioxidant capacity (CUPRAC).

MATERIALS AND METHODS

Plant materials and chemicals

Samples were collected from several locations in Indonesia. *R. jasminiflorum* was collected from Samosir, North Sumatera, *R. javanicum* and *R. retusum* from Gunung Gede-Pangrango National Park, West Java, *R. malayanum* from Gunung Halimun-Salak National Park, West Java, *R. konori* and *R. seranicum* from Eka Karya Botanical Garden, Bali. All samples were identified by Dr. Sri Rahayu and Dyan Meiningsasi Siswoyo, S.Si (authors). The leaves and twigs from all samples were dried, pulverized, and sieved before the extraction process. Ethanol p.a., Folin Ciocalteu reagent, sodium carbonate, aluminum chloride, potassium acetate, sodium acetate, hydrochloric acid, copper chloride, ammonium acetate, ferric chloride, acetic acid and potassium chloride were purchased from Merck (Darmstadt, Germany). Neocuproine, 2,2-diphenyl-1-picrylhydrazyl (DPPH), 2,4,6-tripyridyl-s-triazine (TPTZ), 6-Hydroxy-2,5,7,8-

tetramethylchroman-2-carboxylic acid (trolox), gallic acid, quercetin, and cyanidin-3-glucoside were obtained from Sigma Aldrich (St Louis, USA).

Preparation of extracts

About 20 g of powdered leaves or twigs of each *Rhododendron* species used in this study was weighed in a beaker glass and 200 ml of 80% ethanol was added. The extraction process was carried out for 24 h and repeated once. After filtration, the solvent was evaporated by using a rotary evaporator.

Determination of total phenolics

Total phenolics content in the sample was determined using Folin-Ciocalteu method as described by Kruawan and Kangsadalampai (2006). A total of 10 μ L extract solution, 160 μ L distilled water, 10 μ L of 10% Folin-Ciocalteu reagent and 20 μ L of 7.5% Na_2CO_3 were added to each well of a 96-well plate. The mixture was homogenized and incubated for 30 minutes in the dark. The absorbance was measured at 750 nm using microplate plate reader (Epoch-BioTek, Winooski, USA). Total phenolics content was calculated based on the calibration curve of gallic acid using the following equation: $y = 0.001x + 0.043$, with $R^2 = 0.981$. The results were expressed as gallic acid equivalent (GAE) in grams of dried powder sample (mg GAE/g dried powder). The measurement was carried out in triplicate.

Determination of total flavonoids

The aluminum chloride colorimetric method was used to determine the total flavonoids content following the procedure employed by Lee *et al.* (2011). 10 μ L Sample extract, 60 μ L ethanol, 10 μ L of 10% AlCl_3 , 10 μ L of 1 M potassium acetate and 120 μ L distilled water were added to each well of a 96-well plate. Then the mixture was homogenized and incubated for about 30 minutes. The absorbance of the solution was measured at a wavelength of 415 nm in a microplate plate reader. Total flavonoids content was calculated with quercetin calibration curve ($y = 0.001x - 0.006$, $R^2 = 0.998$) and the quantity was expressed as quercetin equivalent (QE) in a gram of dried powder (mg QE/g dried powder). Triplicate measurement was done for each sample extract.

Determination of total anthocyanin

Total anthocyanin was quantified using a pH differential method with a two-buffer system - potassium chloride buffer, pH 1.0 (0.025 M) and sodium acetate buffer, pH 4.5 (0.4 M) as described by Lako *et al.* (2007). About 0.4 mL of extract solution was added to 3.6 mL of the corresponding buffer. The absorbance of each solution was measured against a blank in a cuvette with 1 cm path length at 510 nm and 700 nm using UV-Vis spectrophotometer U-2800 (Hitachi, Kyoto, Japan). The absorbance used for quantifying the anthocyanin content was calculated as

$$A = (A_{510 \text{ nm, pH} = 1} - A_{700 \text{ nm, pH} = 1}) - (A_{510 \text{ nm, pH} = 4.5} - A_{700 \text{ nm, pH} = 4.5}).$$

Total anthocyanin content was expressed as cyanidin-3-glucoside (%w/w) equivalents, as follows: [Anthocyanin] mg/L = $A \times \text{MW} \times \text{DF} \times 1000 / (\epsilon \times 1)$, where A = absorbance, MW = molecular weight (449.2 g/mol), DF = dilution factor, ϵ

= extinction molar coefficient (26,900 L/cm mol). The final anthocyanin content was expressed as cyanidin-3-glucoside equivalent (CYE) in 100 g of dried powder (mg CYE/100 g dried powder). We measured the anthocyanin content in triplicate for each sample extract.

DPPH radical scavenging activity

Radical scavenging activity was measured using DPPH method developed by Blois *et al.* in 1958 with some modifications performed by Salazar *et al.* (2009). In a 96-well plate, 40 μ L of extract solution was mixed with 250 μ L of 125 μ M DPPH solution (dissolved in ethanol). The solution was immediately incubated for 30 minutes in the dark at room temperature. The absorbance of the solution was measured at 515 nm against ethanol as blank using microplate reader and the radical scavenging activity was determined using the following equation: $y = 0.004x + 0.001$, with $R^2 = 0.998$. Trolox was used as standard and the measurements were performed in triplicate. Radical scavenging activity was expressed as micromolar trolox equivalents in g of extract (μ M TE/g extract).

Cupric ion reducing antioxidant capacity (CUPRAC)

We used a procedure from Öztürk *et al.* (2011) to determine the antioxidant activity by CUPRAC method. About 40 μ L extract solution, 50 μ L of 10 mM CuCl_2 , 50 μ L of 7.5 mM neocuproine and 60 μ L of 1 M $\text{NH}_4\text{CH}_3\text{COO}$ (pH 7) were added to a 96-well plate with total volume of 200 μ L. Incubation of the mixture was carried out for 1 h, and after that, the absorbance was recorded at 450 nm using microplate plate reader. The antioxidant capacity was determined using the following equation: $y = 0.000x - 0.019$, with $R^2 = 0.998$. Trolox was used as a standard, and triplicate measurements were performed for each extract. Antioxidant capacity was expressed as micromolar trolox equivalents in g of extract (μ M TE/g extract).

Ferric ion reducing antioxidant power (FRAP)

The FRAP assay (Benzie and Strain, 1996) and a procedure from Settharaksa *et al.* (2014) were used to determine the antioxidant power for all samples. The FRAP working solution was made by mixing 300 mM acetate buffer pH 3.6, 10 mM TPTZ in 40 mM HCl, and 20 mM $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$ with ratio 10:1:1. In a 96-well plate, we added 30 μ L extract solution and 270 μ L FRAP reagent and let it stand for 30 minutes at room temperature. The absorbance was recorded at 593 nm using microplate plate reader. Total reducing capacity of FRAP was determined using the following equation: $y = 0.003x + 0.019$, with $R^2 = 0.986$, and calculated using trolox as standard and triplicate measurements were performed. Antioxidant power was expressed as micromolar trolox equivalent in g of extract (μ M TE/g extract).

Data analysis

The experimental data were reported as mean \pm standard deviation from triplicate measurements. To determine whether there is a significant difference, we analyzed the data using statistical one-way analysis of variance (ANOVA) followed with Duncan test, if $p < 0.05$. Significant differences were defined at 5% level ($p < 0.05$). ANOVA was performed using XLSTAT software version 2012.2.02 (Addinsoft, New York, United States).

RESULTS AND DISCUSSIONS

Extraction yield, total phenolics, flavonoids and anthocyanin contents

Maceration was used to extract the chemical component of *Rhododendron* species used in this study. We used 80% ethanol as the extraction solvent which gave varying yields ranging from 16.12–31.96% from leaves and 9.07–38.71% from twigs (Table 1). Differences in these yields were due to the differences in the origin of the plant and the species, which is known to give different contents of secondary and primary metabolites.

Table 1: Total phenolics, total flavonoids and total anthocyanin from ethanol extracts of leaves and twigs from six species of *Rhododendron*.

Sample name	Total phenolics (mg GAE/g dried powder)		Total flavonoids (mg QE/g dried powder)		Total anthocyanin (mg CYE/100 g dried powder)	
	Leaves	Twigs	Leaves	Twigs	Leaves	Twigs
<i>R. jasminiflorum</i>	48.11 \pm 0.79 ^d	29.38 \pm 0.54 ^e	8.92 \pm 0.34 ^d	4.32 \pm 0.47 ^d	1.04 \pm 0.03 ^e	1.45 \pm 0.05 ^d
<i>R. retusum</i>	26.53 \pm 0.29 ^b	43.24 \pm 1.61 ^c	4.57 \pm 0.10 ^b	3.67 \pm 0.42 ^e	0.92 \pm 0.09 ^e	1.17 \pm 0.08 ^c
<i>R. konori</i>	41.17 \pm 0.96 ^d	37.25 \pm 0.10 ^d	3.59 \pm 0.32 ^a	3.94 \pm 0.08 ^{cd}	0.76 \pm 0.07 ^b	0.24 \pm 0.02 ^a
<i>R. javanicum</i>	40.39 \pm 0.50 ^{cd}	11.39 \pm 0.06 ^a	6.15 \pm 0.14 ^c	1.33 \pm 0.03 ^{ab}	0.90 \pm 0.09 ^{bc}	0.25 \pm 0.02 ^a
<i>R. seranicum</i>	39.08 \pm 0.27 ^c	11.98 \pm 0.51 ^a	5.90 \pm 0.45 ^c	0.78 \pm 0.02 ^a	2.09 \pm 0.09 ^d	0.25 \pm 0.02 ^a
<i>R. malayanum</i>	19.78 \pm 0.54 ^a	24.10 \pm 0.83 ^b	4.38 \pm 0.11 ^b	1.66 \pm 0.07 ^b	0.12 \pm 0.04 ^a	0.80 \pm 0.04 ^b

The reported values are mean \pm SD of the triplicate assay for each sample. The mean \pm SD within each *Rhododendron* species in the same column followed with different lowercase letters represent significant differences at $p < 0.05$.

Rhododendron species are rich in phenolics compounds, such as flavonoids and anthocyanin, which are known to have antioxidant activities. Total phenolics content can also describe the antioxidant capacity of a sample because the reaction of the method used also involves the transfer of electrons. Total phenolics contents in the samples were varied ranging from 19.78–48.11 mg

GAE/g dried powder of the leaves and 11.39–43.24 mg GAE/g dried powder of the twig. From the results of statistical tests, it is shown that the levels of total phenolics, flavonoids, and anthocyanin in leaves and twigs of all samples were significantly different, except for *R. seranicum* that gave no significant difference ($P < 0.05$) for the total phenolics and flavonoids contents.

Flavonoids and anthocyanin belong to the phenolic compound class, but in this study high level of phenolics compounds do not always produce high levels of flavonoids and anthocyanin. It indicates that flavonoids and anthocyanin are not the major contributors to the presence of phenolics in the leaves and twigs of *Rhododendron* species since anthocyanin

are more common found in flowers. Flavonoids levels in some *Rhododendron* species ranging from 3.59–8.92 and 0.78–4.32 mg QE/g dried powder of the leaves and twigs, respectively. *R. jasminiflorum* was found to have flavonoids content of both leaves and twigs higher than the other samples used in this study.

Table 2: Antioxidant capacities of six *Vireya Rhododendron* leaves and twigs ethanolic extract.

Sample name	DPPH ($\mu\text{mol trolox/g dried powder}$) ^a		FRAP ($\mu\text{mol trolox/g dried powder}$) ^a		CUPRAC ($\mu\text{mol trolox/g dried powder}$) ^a	
	Leaves	Twig	Leaves	Twig	Leaves	Twig
<i>R. jasminiflorum</i>	29.00 \pm 0.20 ^a	18.69 \pm 0.05 ^d	889.14 \pm 6.87 ^d	725.11 \pm 1.78 ^d	1,603.28 \pm 10.89 ^c	1,477.78 \pm 22.86 ^d
<i>R. retusum</i>	21.83 \pm 0.06 ^b	36.68 \pm 0.09 ^f	463.03 \pm 0.86 ^b	887.82 \pm 3.95 ^c	818.53 \pm 18.37 ^b	1,726.62 \pm 11.42 ^c
<i>R. konori</i>	25.46 \pm 0.24 ^a	20.95 \pm 0.24 ^c	941.76 \pm 1.45 ^c	891.56 \pm 5.27 ^c	1,793.62 \pm 7.97 ^d	1,724.26 \pm 14.56 ^c
<i>R. javanicum</i>	26.81 \pm 0.12 ^d	8.62 \pm 0.02 ^a	885.75 \pm 2.62 ^d	149.07 \pm 0.63 ^a	1,599.28 \pm 55.55 ^c	239.50 \pm 1.73 ^a
<i>R. seranicum</i>	28.86 \pm 0.07 ^c	10.22 \pm 0.08 ^b	680.61 \pm 6.67 ^c	222.81 \pm 1.73 ^b	1,583.08 \pm 18.59 ^c	578.56 \pm 5.51 ^b
<i>R. malayanum</i>	15.92 \pm 0.04 ^a	15.51 \pm 0.04 ^c	366.82 \pm 4.16 ^a	397.19 \pm 1.89 ^c	540.98 \pm 4.72 ^a	762.73 \pm 6.24 ^c

The reported values are mean \pm SD of the triplicate assay for each sample. The mean \pm SD within each *Rhododendron* species in the same column followed with different lowercase letters represent significant differences at $p < 0.05$. *, # and ¥ showed significant different from Duncan test ($p < 0.05$).

Similarly, anthocyanin level does not always follow with the total phenolics content in the extract of all samples. For example, *R. seranicum* leaves have the highest content of anthocyanin, but the total phenolics content is below that of *R. jasminiflorum* leaves whose total phenolics content is higher than *R. seranicum* leaves. So, flavonoids and anthocyanin are not the major contributors to the presence of phenolic compounds in *Rhododendron* extracts used in this study. However, the presence of flavonoids and anthocyanin were known to give an antioxidant activity in *Rhododendron* or other plant extracts (Lin *et al.*, 2014; Saha and Verma, 2016).

Antioxidant activity

Determination of antioxidant capacity of the leaves and twigs extracts of each *Rhododendron* species by using DPPH, FRAP, and CUPRAC assay were expressed as trolox equivalent. We expressed the antioxidant activity as trolox equivalent because trolox is a compound with the highest antioxidant capacity index that we could use it to evaluate how high the antioxidant potency of one sample expressed as trolox equivalent (Apak *et al.*, 2004). The antioxidant capacity of all samples was varied by using the three methods shown in Table 2. With DPPH method, *R. retusum* twig extract gives the highest antioxidant capacity of about 36.68 $\mu\text{mol trolox/g dried powder}$, and the lowest is *R. javanicum* twig extract about 8.62 $\mu\text{mol trolox/g dried powder}$. The FRAP method showed that the value of antioxidant capacity is in the range of 149.07–941.76 $\mu\text{mol trolox/g dried powder}$ with *R. konori* leaves extract is the highest. By using the CUPRAC method, *R. konori* leaves extract gives highest antioxidant capacity compared to the other samples.

The antioxidant capacities from DPPH, FRAP, and CUPRAC methods of the leaves and twigs from the six *Rhododendron* species give different results. One-way ANOVA was used to show any differences among the mean values of each antioxidant capacity. From the results of the analysis of variance, it is found that F_{exp} is greater than the critical values for F (0.05,

2, 6). Therefore, the null hypothesis (H_0) is rejected, which means that the three methods give different results in the antioxidant capacity values and further analysis, Duncan test, is needed. The results from the Duncan test showed that Duncan's critical values (Sy) are greater than the differences among the methods, so it may be concluded that DPPH, FRAP, and CUPRAC are significantly different.

Differences in the antioxidant capacity values were due to the different mechanisms in the measurement of antioxidant activities of each method. In the DPPH method, the mechanism of measurement of antioxidant capacity relies on the electron and hydrogen transfer from antioxidant molecules to the DPPH radical and depends on the type of solvent and redox potential of the compounds involved. The DPPH radical has a dark purple color, exhibiting a maximum wavelength around 515 nm and the color will change to yellow if the radical receive an electron or hydrogen from the antioxidant molecules, and resulting in a stable DPPH molecule (Carmona-Jiménez *et al.*, 2014). In the FRAP method, the reduction power of antioxidant compounds in reducing Fe^{3+} to Fe^{2+} is analogous to the ability of the compound in reducing free radicals via electron transfer. The reaction in the FRAP method is based on the reduction of iron (III) to iron (II) and the formation of iron(II)-tripirydyltriazine (Fe (II)-TPTZ) complex at low pH (Limongkon *et al.*, 2017). While in the CUPRAC method, the reduction strength to convert Cu^{2+} into Cu^+ was used to measure the antioxidant activity. Neocuproine (Nc, 2,9-dimethyl-1,10-phenanthroline) was used to form Cu-Nc complexes. The antioxidant compounds will reduce the blue Cu^{2+} -neocuproine ($\text{Cu}(\text{Nc})_2^{2+}$) complexes to yellow Cu^+ -neocuproine ($\text{Cu}(\text{Nc})_2^+$) (Apak *et al.*, 2007).

The three methods used in this study have their typical selectivity as well as advantages and disadvantages in the determination of antioxidant activities, resulting in different antioxidant capacities for the samples tested. DPPH radical scavenging method gives the lowest value of antioxidant capacity compared to the other methods. This may be due to the stability of

DPPH radicals that can be affected by many factors and also the DPPH reagent only dissolve in organic solvents (Özyürek *et al.*, 2011). The antioxidant capacity of the CUPRAC method is higher than DPPH and FRAP. The CUPRAC method is measured at pH 7, which is close to physiological pH, and this could stimulate the antioxidant action further. The CUPRAC method can also react with thiol and glutathione antioxidants that cannot react with the FRAP reagent. Charged chromophores in the CUPRAC method, i.e. $(\text{Cu}(\text{Nc})_2)^+$, are water and organic soluble allowing to determine lipophilic and hydrophilic antioxidants simultaneously. The CUPRAC method can determine the lipophilic antioxidant capacity in hydrophobic media. The CUPRAC reagents are selective because they have a lower redox potential than $\text{Fe}^{3+}/\text{Fe}^{2+}$ in FRAP method and the reactants used are more stable than DPPH (Özyürek *et al.*, 2011).

Correlation between total phenolics, flavonoids, and anthocyanin and antioxidant activity

In this study, the antioxidant activity is known to increase proportionally with the phenolics level because the methods used to determine the antioxidant activities and the phenolics content have the same mechanism through electron transfer in the reaction (Leopoldini *et al.*, 2004; Rasinen *et al.*, 2008). Quantitatively, the relationship between total phenolics, flavonoids and anthocyanin contents with the antioxidant activities of six *Rhododendron* species could be modeled using a linear regression equation. From the correlation analysis, it is shown that the total phenolics contents and the antioxidant activities measured by DPPH, FRAP, and CUPRAC methods had a positive relationship and showed significant values with correlation coefficient about 0.9194, 0.9522, 0.9324, respectively. Correlation coefficient values close to one indicates that the antioxidant activities in *Rhododendron* extracts mainly caused by the phenolics compounds.

The correlation between total flavonoids and anthocyanin contents with antioxidant activities of the samples using linear regression showed a positive correlation, but not as high as with total phenolics contents. The correlation coefficients obtained were 0.6688, 0.6488 and 0.5915 for total flavonoid contents and 0.6232, 0.4324 and 0.5429 for total anthocyanin contents with DPPH, FRAP, and CUPRAC method, respectively. Thus, some flavonoids and anthocyanins may give some contribution to the antioxidant activities of *Rhododendron* leaves and twigs extracts. Correlation with DPPH method showed higher correlation than the two other methods, presumably hydrophilic antioxidant contributes to the antioxidant capacity.

CONCLUSION

From the results obtained we found that the six *Vireya* *Rhododendron* species used in this study were rich in phenolics compounds. Flavonoids and anthocyanin levels were not the primary contributors to the phenolics contents in all *Rhododendron* species used in this study. Significant differences in the antioxidant capacities of *Rhododendron* extracts measured with DPPH, FRAP, and CUPRAC methods were observed. In DPPH method, *R. retusum* twigs extract was found to give the highest antioxidant capacity, while in the FRAP and CUPRAC methods, *R. konori* leaves extract had the highest antioxidant capacity. Total phenolics content gave strong correlation with antioxidant capacity, while

for flavonoids and anthocyanin the correlations were not so strong. Accordingly, for the *Rhododendron* species used in this work, the antioxidant compounds are not mainly come from flavonoids and anthocyanin.

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