Temperature and Solvent Effects on Radical Scavenging Ability of Phenols

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In this work we have demonstrated the free radical scavenging ability of two-hydroxy (catechol, hydroquinone, resorcinol) and three-hydroxy (phloroglucinol, pyrogallol, 1,2,4-benzenetriol) phenols against the diphenylpicrylhydrazyl radical at various temperatures (15−40 °C) and in different solvent media. Kinetic measurements, made by the stopped-flow method, showed that the phenols with OH groups in the *ortho* positions have the largest rate coefficients compared to those with OH groups in the *meta* and *para* positions at all temperatures and in all solvent media. Among the *ortho*-structured phenols catechol, pyrogallol, and 1,2,4-benzenetriol, pyrogallol (three OH groups *ortho* to each other) had the greatest radical scavenging ability. This suggested that intramolecular hydrogen bonding in phenols controlled the rate of radical scavenging ability. The radical scavenging ability of phenols was fastest in methanol and slowest in THF, which emphasized the importance of the interactive behavior of the phenolic OH with the solvent. We concluded from our kinetic data together with our theoretically calculated OH bond dissociation enthalpies of phenols that the OH position played a crucial role in addition to the temperature and nature of the medium in determining the rate of the radical scavenging ability of polyphenols.

1. Introduction

In today's world, ever-increasing levels of physical and mental stress, environmental pollution, and packed foods carry risks of generating free radicals which cause chronic and carcinogenic diseases. Recently, natural plant polyphenols have been the source for the search of new potent antioxidants to protect humans from such diseases by the scavenging of free radicals. More than 8000 compounds have been identified as polyphenols (ArOHs), which differ in structure depending on the multitude of combinations of hydroxyl, oxygen, and methyl groups attached on the two benzene rings (A and B) of the basic structure of flavonoids[1] (Figure 1). Such differences in the chemical structure determine their differences in activity.[2] Structure−activity relationship studies of polyphenols have therefore been more important to scientists in the 21st century. More details on the structure−activity by which natural polyphenols differ would be useful for producing therapeutic drugs, supplements, or food additives. For the design of potent synthetic antioxidants or potential utilization of natural antioxidants in formulating functional foods and nutraceuticals, it is essential to explore and clarify the contribution of key structural elements of antioxidants, principally the OH groups in polyphenols as OH-based moieties are common in most of the polyphenols (Figure 1).[3] As such, catechol, resorcinol, hydroquinone, pyrogallol, phloroglucinol, and 1,2,4-benzenetriol were chosen on the basis of the number and position of OH groups to help elucidate the structural requisites of OH groups for stronger activity of phenols (Figure 2).

The activity of polyphenols should be discussed from the point of view of their scavenging rate against free radicals, because in principle an effective antioxidant should scavenge the free radical at a much faster rate than the radical attacks the substrate. The free radical scavenging ability of polyphenolic antioxidants is governed by the rate of reaction[4] and the reaction environment in which the reaction occurs.[5,6] In the present work, we carried out a comprehensive kinetic study on phenols using the 2,2-diphenyl-1-picrylhydrazyl assay as not only is this a readily available free radical, i.e., it need not be generated using an oxidant, but also it is easy to control the exact quantity of the radical required for study. The radical scavenging ability may also be characterized by the hydrogen-donating ability of polyphenolic antioxidants to extinguish the free radical,[7] which is eventually controlled by the OH bond dissociation enthalpy (BDE) of phenols.[8] In recent years a theoretical method has been successfully employed to estimate this physiochemical parameter, and in this study the Gaussian 98 package that includes the density functional method was adopted to calculate the OH BDEs of phenols. The computed results were analyzed in combination with the experimental kinetic results—our aim being to investigate the structural importance of OH groups in active phenolic compounds for scavenging the radical.

2. Experimental Section

2.1. Reagents. 2,2-Diphenyl-1-picrylhydrazyl radical, catechol, resorcinol, hydroquinone, 1,2,4-benzenetriol, pyrogallol, and phloroglucinol were purchased from Sigma-Aldrich, Singapore. The purity of all compounds was greater than 98%. All solvents used were of HPLC grade, obtained from Fisher Scientific, Singapore.

2.2. Kinetic Method. Measurement of Kinetic Rate Coefficients for the Reaction of Phenols with DPPH$^\bullet$. A colored free radical assay using 2,2-diphenyl-1-picrylhydrazyl (DPPH$^\bullet$) has been successfully applied previously to demonstrate the radical scavenging ability of antioxidants.[9,10] The phenol reaction with DPPH$^\bullet$ leads to hydrogen donation from the phenol and the formation of DPPHH and a phenoxyl radical (ArO$^\bullet$). The reaction is observed as a decay in the absorbance of DPPH$^\bullet$.

$$\mathrm{DPPH}^\bullet + \mathrm{ArOH} \rightarrow \mathrm{DPPHH} + \mathrm{ArO}^\bullet \qquad (1)$$

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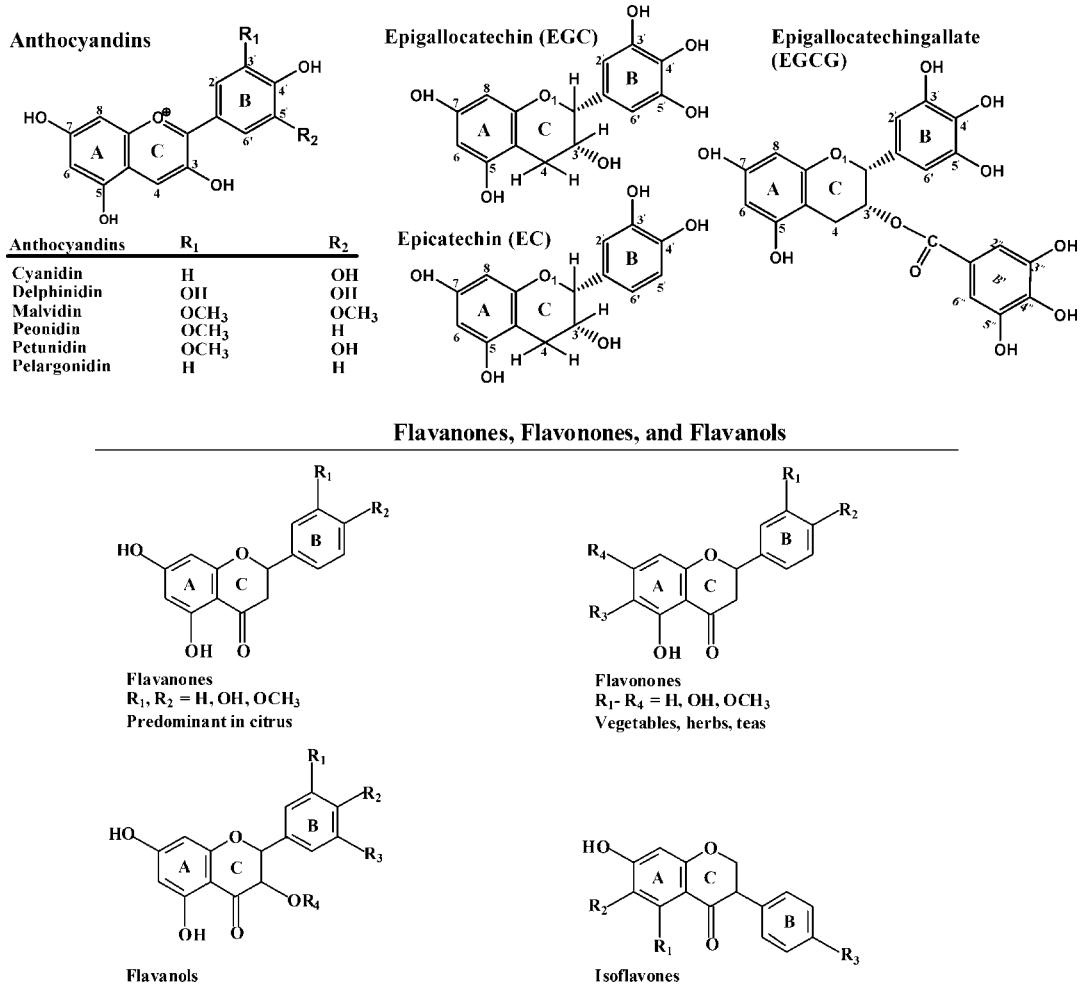


Figure 1. Chemical structure of major polyphenols.

A Biologic stopped-flow spectrophotometer (SFM) equipped with a 150 W xenon lamp was used to measure the decrease in absorbance of DPPH$^\bullet$ at 515 nm. The SFM possesses a three-syringe stopped-flow system capable of rapidly mixing three solutions at a time (Figure 3). One syringe was filled with phenols/solvent and the other one with DPPH$^\bullet$/solvent. Pure solvent was filled in the last syringe for washing the cuvette after each run. The dead-time volume for stopped-flow mixing was kept at 4.6 ms for all runs. The concentration of DPPH$^\bullet$ was kept constant at 0.025 mM for the entire study. The concentration of phenols was always in large excess compared to [DPPH$^\bullet$] to maintain pseudo-first-order kinetics. The pseudo-first-order rate coefficients k' were obtained for each phenol with the ratio of excess antioxidant to DPPH$^\bullet$ depending on the reactivity of the phenols (25−100 times for catechol, hydroquinone, pyrogallol, and 1,2,4-benzenetriol and 50−200 times for phloroglucinol and resorcinol). The solution temperature was controlled by the microprocessor-based digital controller (Polyscience) using ethylene glycol as the circulant. All kinetic measurements were carried out at six temperatures (15, 20, 25, 30, 35, and 40 °C), with an accuracy of ±0.2 °C. The measurements were performed in triplicate. The rate of the radical scavenging reaction (eq 1) is expressed as

$$r = -\frac{\mathrm{d[DPPH^\bullet]}}{\mathrm{d}t} = k[\mathrm{ArOH}]_0[\mathrm{DPPH^\bullet}] \qquad (2)$$

where [ArOH$^\bullet$]$_0$ and [DPPH$^\bullet$] are the concentrations of antioxidant and DPPH$^\bullet$ solution at time $t = 0$ and time t, respectively. The pseudo-first-order rate coefficient k' of the phenols was obtained after eq 2 was solved:

$$[\mathrm{DPPH^\bullet}] = [\mathrm{DPPH^\bullet}]_0 e^{-k't} \qquad (3)$$

The second-order rate coefficients k were then calculated from the slope of plots of k' against [ArOH] by a least-squares fit ($r^2 = 0.9-0.99$). Kinetic parameters are shown in Tables 1 and 2.

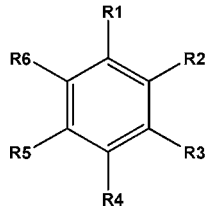
3. Results and Discussion

The reactions of phenols against DPPH$^\bullet$ were carried out in different media and at temperatures in the range of 15−40 °C. The obtained rate coefficients for phenols in methanol, acetonitrile, acetone, and THF are shown in Tables 1 and 2.

3.1. Effect of Temperature on Phenols. The rate coefficients of phenols were found to be larger upon increasing the temperature in all solvents (Tables 1 and 2). As more kinetic energy is imparted by increasing the temperature, the OH of the phenols become more labile and the H atom readily dissociates. As usual, the Arrhenius equation was used to quantify the effect of temperature:[11]

$$\ln k = \ln A - \frac{E_a}{RT} \qquad (4)$$

where k is the second-order rate coefficient (M^{-1} s^{-1}), A is the pre-exponential factor, R is the gas constant (8.314 J mol^{-1} K^{-1}),



Catechol, (Cat)	R1, R2 = OH;	R3, R4, R5, R6 = H
Resorcinol, (Res)	R1, R3 = OH;	R2, R4, R5, R6 = H
Hydroquinone, (HQ)	R1, R4 = OH;	R2, R3, R5, R6 = H
Phloroglucinol, (Phl)	R1, R3, R5 = OH;	R2, R4, R6 = H
Pyrogallol, (Pyr)	R1, R2, R3 = OH;	R4, R5, R6 = H
1,2,4-benzenetriol, (Benz)	R1, R2, R4 = OH;	R3, R5, R6 = H

Figure 2. Phenols chosen on the basis of the number and position of OH groups.

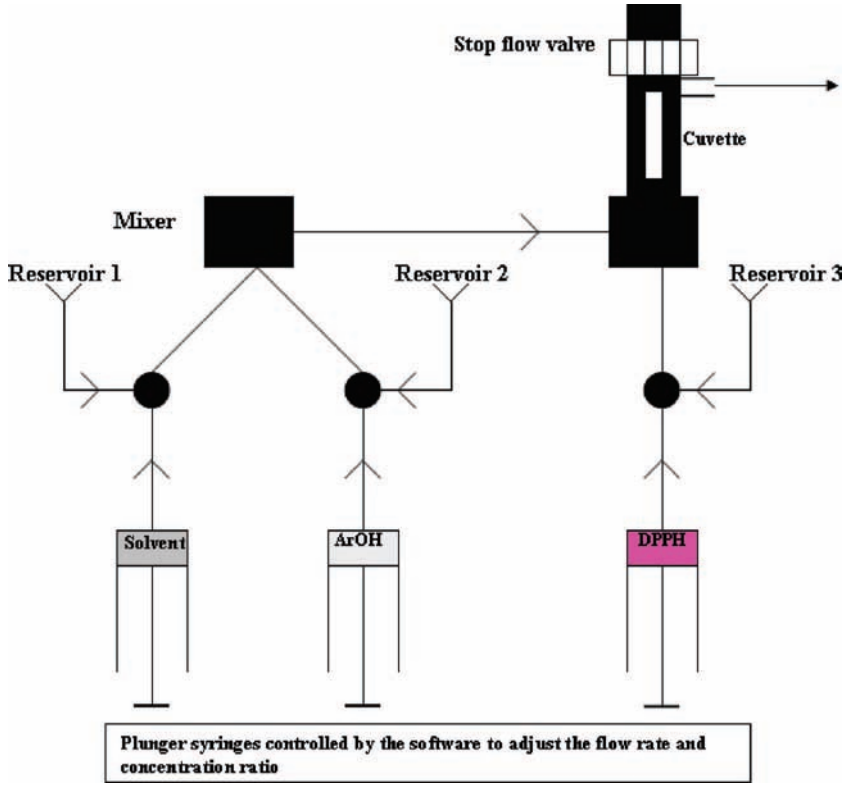


Figure 3. Schematic of the stopped-flow UV−vis spectrophotometer.

E_a is the activation energy (kJ mol^{-1}), and T is the temperature (K). The determined activation energies, E_a, for all phenols are given in Tables 1 and 2. The kinetics and thermodynamics (of the transition state) for chemical reactions are intimately related as a change in enthalpy accompanies the chemical reaction during the breaking and forming of bonds.[12] Such a relationship for phenols was examined using the Eyring transition-state theory:[13]

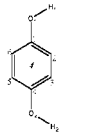
$$\ln \frac{k}{T} = -\frac{\Delta H^{\ddagger}}{R}\frac{1}{T} + \ln \frac{k_B}{h} + \frac{\Delta S^{\ddagger}}{R} \qquad (5)$$

where k is the second-order rate coefficient (M^{-1} s^{-1}), T is the temperature (K), $\Delta H^{\ddagger}$ is the activation enthalpy (kJ mol^{-1}), $\Delta S^{\ddagger}$ is the activation entropy (J mol^{-1} K^{-1}), k_B is the Boltzmann constant (1.3807×10^{-23} J K^{-1}), and R is the gas constant (8.314 J mol^{-1} K^{-1}). A plot of $\ln(k/T)$ versus $1/T$ produced a straight line with a slope of $-\Delta H^{\ddagger}/R$ and intercept of $\ln(k_B/T) = \Delta S^{\ddagger}/R$ to obtain the activation enthalpy ($\Delta H^{\ddagger}$) and activation entropy ($\Delta S^{\ddagger}$), respectively. Our results are presented in Tables 3 and 4.

3.2. Effect of Two-OH Phenols. Catechol possesses two OH groups *ortho* to each other, i.e., C(1,2) positions in the benzene ring. This feature appears as the catecholic moiety in the B ring of most of the flavonoids such as quercetin, rutin, taxifolin, catechin, epicatechingallate, and cyanidin. Resorcinol has the structure of two OH groups positioned *meta* to each other, i.e., C(1,3) in the benzene ring, whereas hydroquinone possesses two OH groups in *para* positions, C(1,4). Resorcinol is observed as the hydroxylation pattern in the A ring of flavonoids. Our kinetic experimental studies showed that catechol had the largest rate coefficient among the two-OH phenols at all temperatures and in all solvents, followed by hydroquinone and resorcinol. The activation barrier (E_a) for the catechol−free radical reaction was noted to be ca. 30−50 kJ mol^{-1}, which is 8−12 kJ mol^{-1} lower than that of resorcinol and hydroquinone. This highlights the dominant role of *o*-OH groups in free radical scavenging reactions. The rate coefficient for H-atom abstraction of phenols depends on the degree of stabilization of the aroxyl radical.[14] The *ortho* arrangement of OH groups in catechol exerts an intramolecular H-bond (IHB) between the two neighboring OH groups, providing more stability to the phenoxyl radical derived from catechol upon hydrogen donation. The hydroquinone free radical reaction (Table 1) possessed a lower E_a by about 25−50 kJ mol^{-1} than that of resorcinol. These results indicated the importance of the *p*-OH positions. The kinetic study on two-OH phenols revealed that the reactivity order of phenols against the free radical was *ortho* > *para* ≫ *meta*.

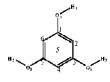
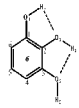
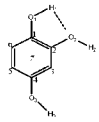
3.3. Effect of Three-OH Phenols. Pyrogallol has the structure of three OH groups attached to the benzene in the vicinal position, i.e., C(1,2,3), and is illustrated as the B ring

TABLE 1: Rate Coefficients (k) and Activation Energies (E_a) of Two-OH Phenols in Solvents

phenols	solvent[a]	Second order rate coefficient, k ($M^{-1}s^{-1}$)						Arrhenius parameters	
		15°C	20°C	25°C	30°C	35°C	40°C	E_a[b] kJ mol^{-1}	lnA[b] $M^{-1}s^{-1}$
Cat	meoh	66	98	204	300	455	800	74.8 ±3.1	35.4 ±1.3
	acn	16	30	45	90	126	300	84.1 ±5.0	37.9 ±2.0
	acet	10	21	45	78	105	200	87.4 ±4.9	38.9 ±2.0
	thf	0.7	1.8	4.0	7.0	9.5	18	93.6 ±7.0	38.9 ±2.8
Res	meoh	0.25	0.60	1.50	2.70	5.0	9.0	106.7 ±4.3	43.3 ±1.7
	acn	0.042	0.13	0.21	0.70	0.92	2.0	113.2 ±7.8	44.3 ±3.1
	acet	0.03	0.10	0.2	0.475	0.7	1.8	116.6 ±6.8	45.4 ±2.7
	thf	0.004	0.01	0.03	0.06	0.13	0.8	150 ±12.1	56.9 ±4.9
HQ	meoh	26	50	80	137	238	430	82.4 ±1.9	37.7 ±0.7
	acn	5.0	8.0	16	31	54	100	91.5 ±2.5	39.7 ±1.0
	acet	3.0	5.0	9.0	18	40	58	93.1 ±4.2	39.9 ±1.7
	thf	0.4	1.1	3.2	5.0	7.0	13	100.7 ±9.0	41.4 ±4.0

[a] Abbreviations: meoh, methanol; acn, acetonitrile; acet, acetone; thf, tetrahydrofuran. [b] Data are presented as the mean ± standard error.

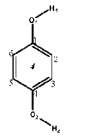
TABLE 2: Rate Coefficients (k) and Activation Energies (E_a) of Three-OH Phenols in Solvents

Phenols	solvent[a]	Second order rate coefficient, k ($M^{-1}s^{-1}$)						Arrhenius parameters	
		15°C	20°C	25°C	30°C	35°C	40°C	E_a[b] kJ mol^{-1}	ln$A$[b] $M^{-1}s^{-1}$
Phl	meoh	0.2	0.5	1.9	3.0	4.2	7.8	108.3 ±11.6	43.9 ±4.6
	acn	0.009	0.02	0.05	0.15	0.4	1.0	143.9 ±6.0	55.3 ±2.4
	acet	0.008	0.014	0.044	0.15	0.3	0.9	145.7 ±7.3	55.8 ±2.9
	thf	0.001	0.011	0.023	0.06	0.15	0.26	157.5 ±18.3	59.6 ±7.3
Pyr	meoh	620	990	1700	2185	3600	6000	66.2 ±2.8	34.0 ±1.1
	acn	291	450	740	1250	1900	2700	68.5 ±1.6	34.3 ±0.6
	acet	202	336	560	892	1400	2200	71.5 ±0.3	35.2 ±0.1
	thf	16	51	63	145	190	250	79.7 ±9.6	36.4 ±3.9
Benz	meoh	364	560	998	1444	2325	3427	67.9 ±1.5	34.3 ±0.6
	acn	191	360	522	928	1434	2100	71.6 ±2.2	35.2 ±0.9
	acet	223	340	565	919	1478	2338	71.3 ±1.2	35.1 ±0.5
	thf	12	32	54	105	145	202	83.2 ±7.4	37.4 ±3.0

[a] Abbreviations: meoh, methanol; acn, acetonitrile; acet, acetone; thf, tetrahydrofuran. [b] Data are presented as the mean ± standard error.

of tea catechins, myricetin, and delphinidin. Phloroglucinol possesses three OH groups attached *meta* to each other, i.e., C(1,3,5) positions in the benzene ring, whereas 1,2,4-benzenetriol has two OHs in C(1) and C (2) positions but the third OH in the C(4) position of the benzene ring. Among 3-OH phenols, the largest rate coefficients were obtained for pyrogallol

TABLE 3: Activation Enthalpies ($\Delta H^{\ddagger}$), Entropies ($\Delta S^{\ddagger}$), and Free Energies ($\Delta G^{\ddagger}$) and OH BDEs of Two-OH Phenols in Solvents

Phenols	Solvents[a]	Eyring Parameters			Computed thermodynamic parameter
		$\Delta H^{\ddagger}$[b] kJ mol^{-1}	$\Delta S^{\ddagger}$[b] J mol^{-1} K^{-1}	[c]$\Delta G^{\ddagger}$[b] (25 °C) kJ mol^{-1}	BDE_{solv}[d] kJ mol^{-1}
Cat	meoh	72.3 ±3.1	41.13 ±1.9	60.07	310.933
	acn	81.6 ±4.9	61.09 ±1.8	63.36	312.075
	acet	84.9 ±6.9	70.26 ±1.7	63.95	311.363
	thf	91.1 ±4.9	70.54 ±1.8	70.09	308.948
Res	meoh	104.4 ±4.2	106.72 ±1.8	72.62	343.612
	acn	110.7 ±7.8	114.66 ±1.7	76.53	342.998
	acet	114.1 ±6.8	124.03 ±1.8	77.13	343.392
	thf	147.5 ±12.1	220.11 ±1.6	81.89	344.038
HQ	meoh	79.9 ±1.8	59.98 ±1.9	62.04	318.536
	acn	88.9 ±2.5	76.99 ±1.9	66.03	318.447
	acet	90.6 ±4.2	78.21 ±1.8	67.27	317.690
	thf	98.2 ±9.9	90.96 ±1.6	71.07	319.392

[a] Abbreviations: meoh, methanol; acn, acetonitrile; acet, acetone; thf, tetrahydrofuran. [b] Data are presented as the mean ± standard error. [c] $\Delta G^{\ddagger}(25\ ^{\circ}C) = \Delta H^{\ddagger} - T\Delta S^{\ddagger}$. [d] Self-consistent reaction field−B3LYP/6-311++G (3df, 3pd) in Gaussian 98.

(Table 2), which showed an E_a ca. 3 kJ mol^{-1} lower than that of 1,2,4-benzenetriol. Upon analyzing the structure with the experimental results of pyrogallol and 1,2,4-benzenetriol, the following dominant role of the double *ortho* position was proposed. The two OH groups located at the C(2) and C(3) positions of pyrogallol exert two IHBs, which we expect provide more stability to the phenoxyl radical generated upon hydrogen donation from pyrogallol and in turn lead to effective free radical scavenging action. In pyrogallol, the hydrogen H2 atom of the O2−H2 group can be donated easily due to the presence of intramolecular hydrogen bonds with neighboring OH groups and dissociates first during the radical scavenging reaction. In our recent paper involving a computational study we also predicted that H2 of the O2−H2 group dissociates to scavenge the free radical on the basis of the OH BDEs.[15]

3.4. Comparison of Two- and Three-OH Phenols. Phloroglucinol and resorcinol possess three OH groups and two OH groups positioned *meta* to each other and showed the highest E_a in all solvents. This indicated that regardless of the number of OH groups in the *meta* position no enhancement of the radical scavenging ability of phenols occurs. The E_a of pyrogallol−free radical reaction is ca. 10 kJ mol^{-1} lower in comparison to that of catechol, suggesting that the greater the number of *o*-OH groups in the phenolic structure, the lower the energy barrier E_a and the faster the radical scavenging reaction. Ideally both catechol and hydroquinone exert an equal number of resonance structures in their radical form; nevertheless, the larger rate coefficient was observed for catechol. Similarly, despite the fact that 1,2,4-benzenetriol exhibits the same number of resonance structures as pyrogallol, pyrogallol showed the lower activation barrier for free radical scavenging. In catechol, pyrogallol, and 1,2,4-benzenetriol an IHB exists due to the presence of *o*-OH (Figure 4). In comparison with 1,2,4-benzenetriol, pyrogallol has an additional IHB attraction at H2 from O3 due to the presence of a second *o*-OH in its structure, which facilitates the donation of a hydrogen atom. A greater number of *o*-OH groups may bring a greater stability to phenoxyl radical via more IHBs, and this may account for the high radical scavenging ability of pyrogallol. In all phenols, the lower $\Delta G^{\ddagger}$ was associated inversely with larger rate coefficients. The activation enthalpy, $\Delta H^{\ddagger}$, also followed a trend similar to that of the activation energy, E_a. Positive values of the activation entropy ($\Delta S^{\ddagger}$) were obtained for all the phenol−radical reactions, which indicated a higher rigidity of the reactant state compared to the transition state. Linear dependences between enthalpy and entropy were observed for the phenol−radical reaction (Figure 5) within 15−40 °C in all solvents, which suggested that a single mechanism operates in all the solvents. From the thermokinetic study of both two and three-OH phenols, it is clear that the rate of the radical scavenging reaction significantly depends on the position of the OH groups of the phenols, but not the number of OH groups.

3.5. Effect of Solvation. The plot of activation energy against the solvent medium (Figure 6) shows that the energy barrier of all phenol−radical reactions was the lowest in methanol and highest in THF despite the structural differences of phenols. The observed rate coefficient for the catechol−free radical reaction in methanol was found to be about 3 times higher than in acetonitrile and 4−6 times than in acetone, highlighting the increased radical scavenging ability of phenols in the presence

TABLE 4: Activation Enthalpies ($\Delta H^{\ddagger}$), Entropies ($\Delta S^{\ddagger}$), and Free Energies ($\Delta G^{\ddagger}$) and OH BDEs of Three-OH Phenols in Solvents

Phenols	Solvent	Eyring Parameters			Computed thermodynamic parameter
		$\Delta H^{\ddagger}$ [b] kJ mol^{-1}	$\Delta S^{\ddagger}$ [b] J mol^{-1} K^{-1}	$^{c}\Delta G^{\ddagger}$ [b] (25 °C) kJ mol^{-1}	BDE_{solv} [d] kJ mol^{-1}
Phl	meoh	105.75 ±11.6	111.28 ±1.6	72.59	343.728
	acn	141.45 ±4.23	206.11 ±1.8	80.02	343.623
	acet	143.14 ±7.3	210.37 ±1.7	80.45	342.987
	thf	155.03 ±18.3	241.90 ±1.4	82.94	344.298
Pyr	meoh	63.75 ±2.7	30.17 ±1.9	54.76	291.830
	acn	66.01 ±1.6	31.63 ±1.9	56.58	291.829
	acet	68.99 ±0.3	39.13 ±1.9	57.34	291.667
	thf	77.21 ±9.6	49.04 ±1.7	62.59	291.365
Benz	meoh	65.43 ±1.5	31.55 ±1.9	56.03	292.008
	acn	69.14 ±2.2	39.54 ±1.9	57.36	289.503
	acet	68.76 ±1.2	38.70 ±1.9	57.23	292.139
	thf	80.59 ±7.3	57.89 ±1.8	63.34	292.678

[a] Abbreviations: meoh, methanol; acn, acetonitrile; acet, acetone; thf, tetrahydrofuran. [b] Data are presented as the mean ± standard error. [c] $\Delta G^{\ddagger}(25\ ^{\circ}C) = \Delta H^{\ddagger} - T\Delta S^{\ddagger}$. [d] Self-consistent reaction field−B3LYP/6-311++G (3df, 3pd) in Gaussian 98.

of a methanol. Solvent effects are often successfully interpreted in terms of the dielectric constant. In our study, though methanol and acetonitrile have nearly the same dielectric constant (33 and 36.6, respectively), the reactivity of phenols in methanol was found to be the greatest. Das et al.[16] carried out a kinetic solvent effect study and highlighted that the lower reaction rates of phenols in acetonitrile were attributed to the formation of a PhOH···solvent interaction. Both methanol and acetonitrile, being polar solvents, are expected to form a PhOH···solvent bond and thus influence the hydrogen atom donation of phenols. However, methanol can potentially form hydrogen bonds with surrounding methanol molecules.[17] Such hydrogen-bonding interactions between methanol molecules intrinsically limit or eliminate the maximum possibility of formation of methanolic H-bonding interactions with the phenolic OH; thus, the hydrogen atom donating behavior of phenols to scavenge the free radical is least affected in methanol (Figure 7). As such, the phenols are expected to be fully involved in their activity against free radicals, which is reflected in the large rate coefficients in methanol (Tables 1−4). Hence, the polar protic nature of solvent plays an important role in facilitating the reactivity of phenols effectively against the free radical.

Rate coefficients of all phenol−free radical reactions were found to be smaller in acetone than in acetonitrile. The dielectric constant of acetone ($\varepsilon = 20.7$) is lower than that of acetonitrile ($\varepsilon = 36.6$), so there appears to be some correlation between the dielectric constant and rate coefficient. The differences in the rate coefficients of phenols in acetonitrile and acetone may also be due to the differences in the ability of the solvent to engage in intermolecular H bonding with the phenolic OH. The lower rate coefficients of phenol−free radical reaction in acetone

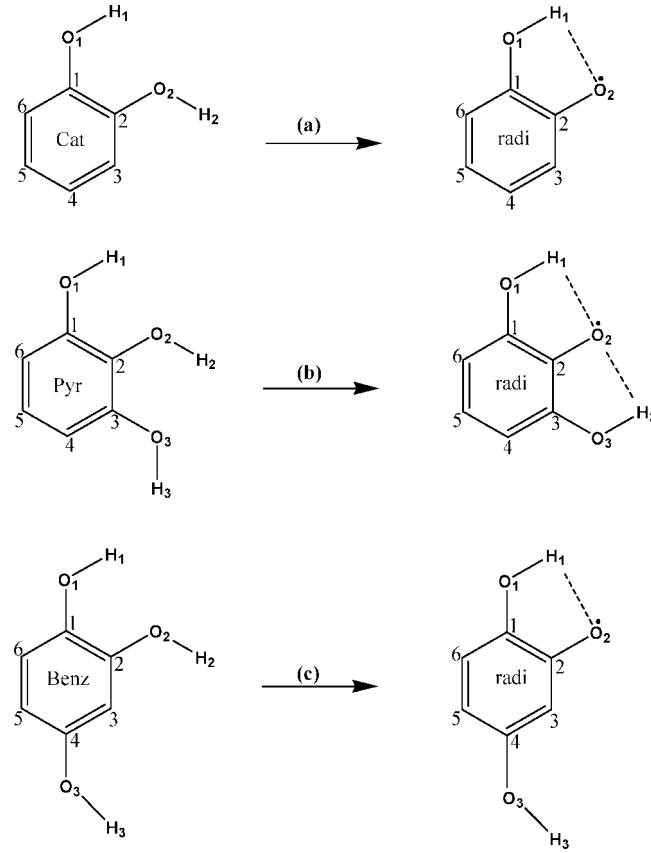


Figure 4. IHB exerted stability of aroxyl radicals derived from (a) catechol, (b) pyrogallol, and (c) 1,2,4-benzenetriol.

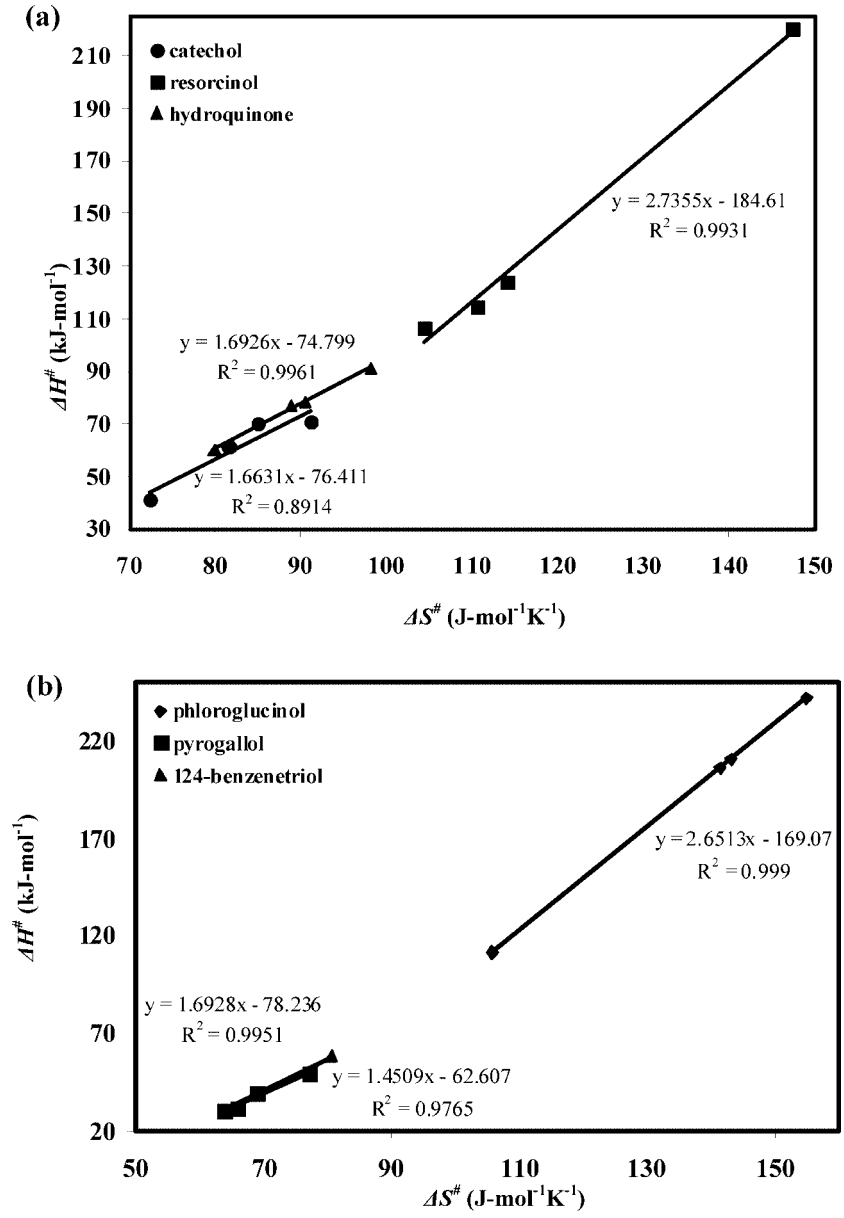


Figure 5. Activation enthalpy and entropy compensation for phenolics with (a) two OH groups and (b) three OH groups.

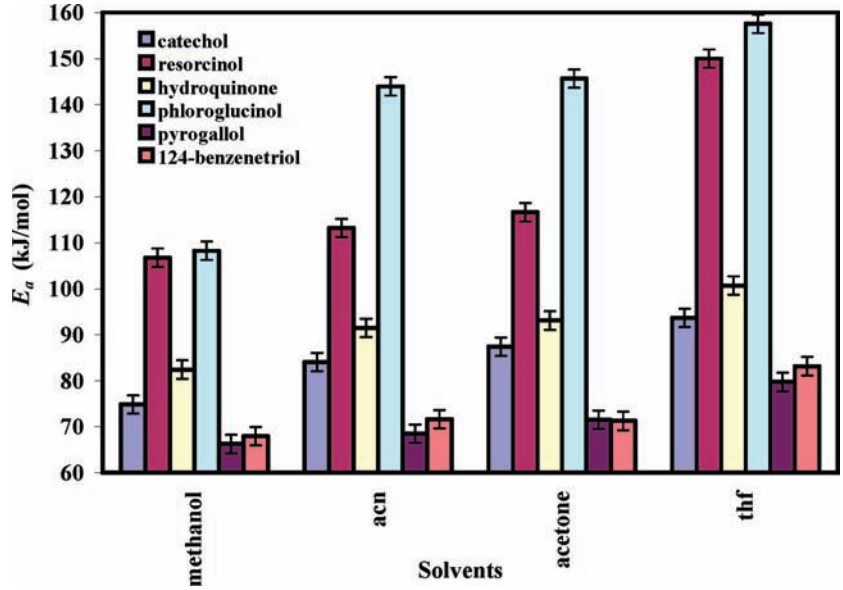


Figure 6. Experimental activation energies E_a against the reaction media.

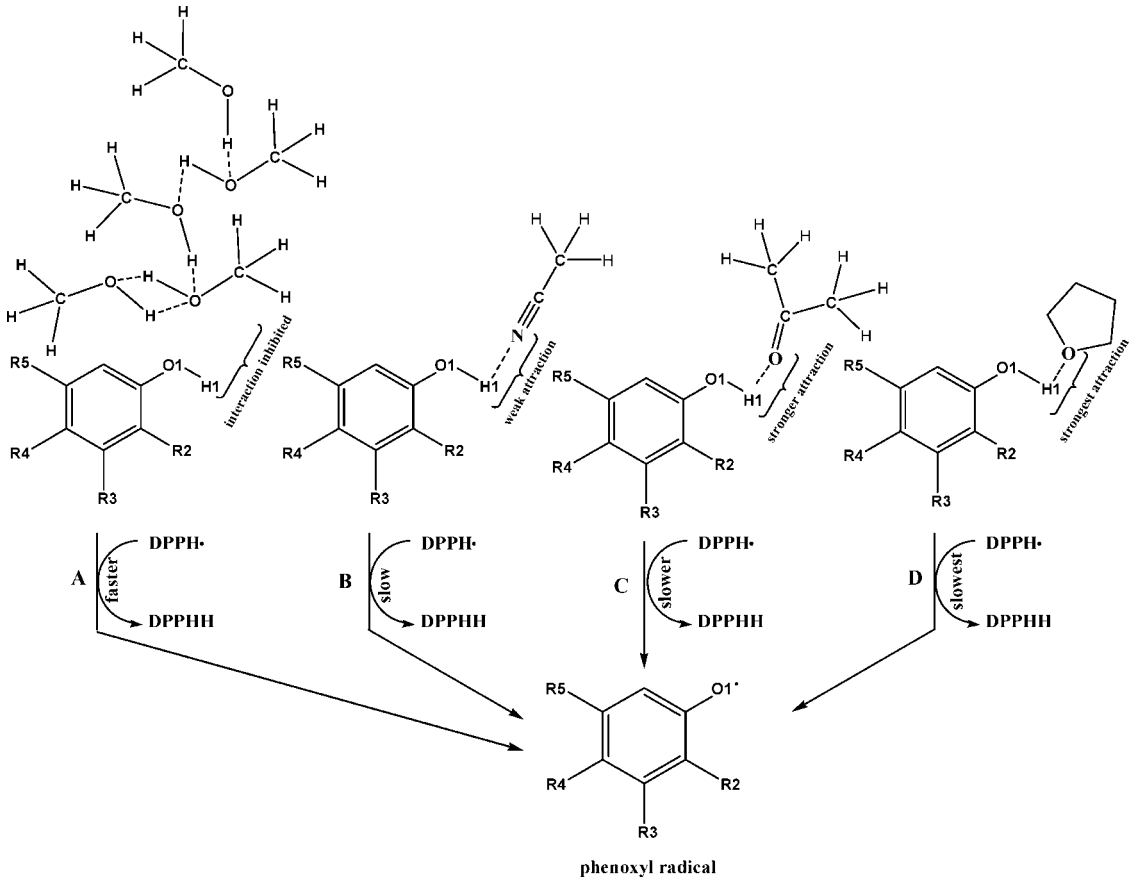


Figure 7. Effect of solvents: DPPH radical scavenging ability of phenols in methanol (A), acetonitrile (B), acetone (C), and THF (D).

were attributed to the strength of electronegativity of the PhOH···solvent hydrogen bond. The interaction between phenols and acetonitrile is weaker compared to that between phenols and acetone due to the weaker electronegative nature of the N atom (Figure 7); thus, phenols release their H atom at a considerably faster rate in acetonitrile than in acetone and THF. The rate coefficients of all phenol−free radical reactions were found to be smallest in THF, which is reflected in the higher E_a. The dielectric constant of THF ($\varepsilon = 7.52$) is the lowest of all the solvents studied here, indicating again that the dielectric constant plays a significant role. Furthermore, the higher electronegative nature of the O atom in THF exerts a stronger interaction with the OH of the phenols, restricting the hydrogen atom donation of phenols and thus decreasing the rate coefficients. Since hydrogen abstraction is retarded, the rate of free radical scavenging is reduced in the THF medium, and in turn, smaller reaction rate coefficients are obtained (Tables 1 and 2). In the literature, a similar decrease in the reactivity of 2,6-dibutyl-4-methylphenol against DPPH$^\bullet$ was observed by Litwinienko and Ingold.[18]

3.6. Comparison of Experimental and Computational Results. The rate of phenol−free radical reactions depends on the intrinsic reactivity of the two reactants, which is largely governed by the BDE. The BDE may be computed theoretically and thus used as a parameter to characterize radical scavenging activity.[8] In particular, it is the BDE of the hydroxyl groups that is used to interpret the free radical scavenging ability of polyphenols.[19−21] In this study, OH BDEs of phenols were computed using the self-consistent reaction field solvent model (SCRF)[22] with the B3LYP method and the basis set 6-311++G (3df, 3pd), as implemented in the Gaussian 98 suite of programs.[23] The solute radius was computed using a gas-phase molecular volume calculation using the volume = tight option in the Gaussian program suite and was used for the frequency calculations. Full geometry optimizations and frequency calculations were performed using the restricted B3LYP method for the parent molecule and unrestricted B3LYP for the radical as stated in our previous work.[15] The OH BDEs were calculated at 25 °C using the following equation for the most stable ArOH conformer and the weakest ArOH bond:

$$\mathrm{BDE(OH)} = H(\mathrm{Ar{-}O^\bullet}) + H(\mathrm{H}) - H(\mathrm{ArOH}) \qquad (6)$$

$H(j)$ in eq 6 is the enthalpy of chemical j at 25 °C and 1 atm. As had been done in previous work,[24] we used the exact H(H) of the hydrogen atom (−0.5 au) for the BDE calculations. Calculated OH BDEs are shown in Tables 3 and 4.

Computed OH BDEs in solution also showed that the phenols with *o*-OH groups have lower BDEs compared to other phenols. It is of interest to correlate the theoretical OH BDEs with the kinetic parameters as any correlation between the OH BDEs and the radical scavenging ability may provide a deeper insight into the phenol−radical mechanism.[25−27] It is of note that the OH BDEs of all phenols were computed at 25 °C, while the experimental activation energy was the result of measurements made in the temperature range 15−40 °C. Wright et al.[28] reported that the OH BDE of the phenols varied by less than 0.09 kJ mol^{-1} over the temperatures 25−37 °C and hence carried out their OH BDE calculations at 25 °C to explain the antioxidant activity of polyphenols at 37 °C. Likewise, it was assumed in this study that the OH BDE of phenols does not vary significantly over the temperatures considered. Thus, we plotted our computed OH BDEs against experimentally calcu-

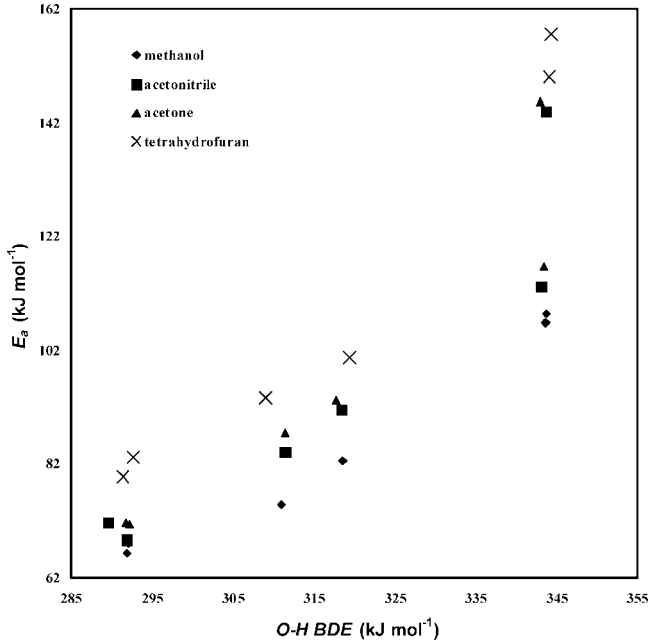


Figure 8. Correlation between theoretical OH BDEs and experimental activation energies E_a of phenols.

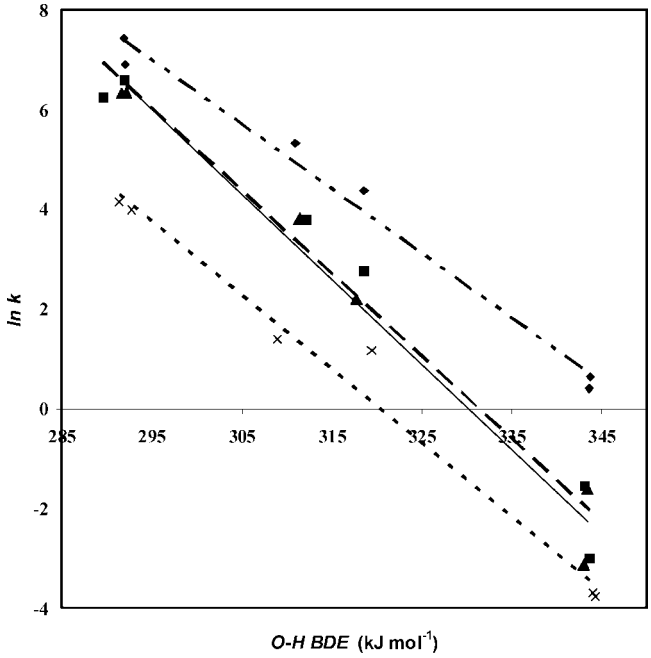


Figure 9. Correlation between experimental ln k values and OH BDEs at 25 °C for phenols.

lated activation energies of phenol−radical reactions (Figure 8) and the OH BDEs versus the logarithm of experimental rate coefficients k obtained at 25 °C (Figure 9). Both figures clearly show that the rate coefficients for the phenol−radical scavenging reactions are dependent upon the BDEs. Both the kinetic and computational studies highlighted that catechol, pyrogallol, and 1,2,4-benzenetriol were effective in scavenging the free radicals, which suggested that any phenolic antioxidant that possesses a OH in an *ortho* position can scavenge the free radicals more readily than one that possesses a OH in another position. As such, moiety B that consists of catecholic and pyrogallol structures in epicatechin and epogallocatechin, respectively (Figure 1), is expected to have a predominant role in radical scavenging reactions at all temperature and in all solvents. Such predictions are also supported by researchers who have performed NMR conformational analysis and electron spin resonance analysis on tea catechins using DPPH$^{\bullet}$.[29]

4. Conclusions

Our kinetic and computational studies demonstrated that the position of the OH functional group significantly influences the rate of the radical scavenging reaction of phenols. The kinetic results from resorcinol and phloroglucinol clearly indicated that OH groups in the *meta* positions of the phenolic ring do not contribute to effective scavenging of free radicals. Results from pyrogallol, 1,2,4-benzenetriol, and catechol distinctly signify that the rate of radical scavenging is mainly contributed to by the *ortho* arrangement of OH groups in phenols. The rate of the radical scavenging reaction of phenols is also affected by the nature of the solvent and the strength of the phenolic OH···solvent interaction. The intermolecular hydrogen bonding between methanol solvent molecules prevents the interaction between methanol and the phenolic OH, which ensures that more phenolic OH groups are available for scavenging the free radical. Overall, it can be concluded that (i) three OH groups in the vicinal position (*ortho* to each other) impart the largest radical scavenging ability to phenols, (ii) ΔH and E_a are reliable parameters to explain the free radical scavenging ability of phenols, and (iii) the polarity and protic nature of a solvent play vital roles in the radical scavenging ability. Our detailed results can be summarized as follows:

rate coefficient: pyrogallol > 1,2,4-benzenetriol > catechol > hydroquinone > resorcinol ≈ phloroglucinol

solvent reactivity: methanol > acetonitrile > acetone > THF

Consequently, we believe that this experimental and computational approach will ultimately not only provide the possibility of explaining the radical scavenging ability of existing phenols but also be of value for the design of new synthetic antioxidants.

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