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STUDY OF STRUCTURE-ANTIOXIDANT ACTIVITY RELATIONSHIP BY BDE ANALYSIS OF HYDROXYBENZALACETONE'S DERIVATIVES

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Abstract

Benzalacetone analogue that are easily synthesized compounds have some biological activities. The aims of this study is to analyze hydroxybenzalacetone's derivatives as antioxidant. The test has been done through deoxyribose degradation inhibition methods. The bond dissociation energy (BDE) was also determined using AM1 semi empirical method. BDE-OH on ROO-H is 88 kcal/mol, thus ArOH antioxidant compounds must less. If BDE-OH compound more than BDE ROOH, it will prefer to attack radical ROO-H to form peroxyl radicals and is followed by a radical chain reaction.

Hydroxybenzalacetones derivatives that have been studied are 2,2′-dihydroxydibenzalacetone (**1**), 3,3′-dihydroxydibenzalacetone (**2**), 3-hydroxydibenzalacetone (**3**), 3-methoxy-4-hydroxybenzalacetone (**4**), 2-hydroxybenzalacetone (**5**) and 3-hydroxybenzalacetone (**6**). Buthylated Hydroxy Toluene (BHT) was used as positive control. The previous research shows the IC_{50} of hydroxybenzalacetones were 8.44, 495.24, 429.68, 92.29, 97.91 and 137.09 µg/mL, while BDE value were 16.68, 106.05, 106.23, 84.62, 87.34 and 90.65 kcal/mol respectively. The most active antioxidant based on IC_{50} and BDE datas is compound **1.** Compounds **1, 4** and **5** are categorized into very active antioxidant because they have IC_{50} value lower than 100 µg/mL. Compounds **1, 4** and **5** have BDE-OH less than 88 kcal/mol. Compounds **4** and **5** have a BDE-OH slightly lower than ROOH and BHT as controls. If conclude from the BDE-OH data, compound **4** is more active than **5**. Compound **4** has two substituents, hydroxy and methoxy groups that are act as an electron donor in the ortho position. Electron donating groups on adjacent positions will increase the stability of phenolic radicals thus enhance its activity as an antioxidant.

Based on these study, deoxyribose degradation inhibition compared with BDE could be use to determine the structure-antioxidant activity of the benzalacetone analogue.

Key words: BDE, hydroxybenzalacetone, antioxidant

INTRODUCTION

Antioxidants are very important compounds. Antioxidants are compounds that can delay, prevent or eliminate oxidative damage to a target molecule (Halliwel, 2007). Based on the mechanism action, antioxidants are divided into primary and secondary antioxidants. Primary antioxidants are compounds that are directly related to free radical, donor hydrogen atom or radical scavenger substances. Radical scavenger compounds usually have phenolic group i.e. hydroxyflavone (Silva et al., 2002), divanillic acetone (Mousa, 2012), resveratrol (Lastra et al.,

2007) etc. Secondary antioxidants or known as preventive antioxidant, delay or prevent the rate of auto oxidation by a mechanism that is not directly associated with radicals such as metal binding or ultraviolet light absorption (Priyadarsini, 2005).

Based on the first function, antioxidants must have a certain structure that can donate hydrogen easily. In general, compounds that have weak CH bonds should not be used as an antioxidant because they will be carbon centered radicals. Carbon radicals react easily with oxygen to form a peroxide radical chain reaction (ROO•). Best antioxidants are those which have oxygen-centered radicals, especially ArO•. ArO• radical is relatively stable, so it cannot react with oxygen to form trioxyl radical ArOOO • (Hussain et al., 2003). If radical ArO• is stable, it will not form a chain reaction, then ArOH is qualified to be used as an antioxidant by donating a hydrogen atom.

The purpose of this study was to analyzed the structure-activity as antioxidant of some hydroxybenzalacetones derivatives that have been synthesized by Handayani et al. (2012a). Antioxidant activity test of several benzalacetones in previous research were done by using deoxyribose degradation inhibition compared to its BDE.

DISCUSSION

3-dimensionally molecular structure were drawn using HyperChem program. Compounds optimization was done by using Austin Model 1 (AM1) semi empiric methods, at Restricted Hartree-Fock (RHF) level. The convergence boundaries setting based on observations of the molecular structure orientation. Radical structure was made by removing a hydrogen atom in the hydroxyl group (-OH) for polyhydroxy molecules. Molecular single point energy was calculated by using radical semi empiric Austin Model 1 (AM1) method at unrestricted Hartree Fock (UHF) level. Spin multiplicity for radical molecules values set at 1 and the radical charge at 0. Bond enthalpy termination was determined by calculating the binding energy difference between the radical molecules with optimized binding energy of the neutral molecule (Handayani, et al. 2012b). Antioxidant activity data by deoxyribose degradation inhibition methods and BDE-OH from Handayani et al. (2012b) were presented in Table 1.

Table 1. BDE-OH and IC_{50} data of hydroxybenzalacetone's derivatives

Compund No	Compound Name	Resonance No	BDE (kcal/mol)	IC_{50} (µg/mL)
1	2,2′-dihydroxydibenzalacetone	12	16.68	8.44
2	3,3′-dihydroxydibenzalacetone	4	106.05	495.24
3	3-hydroxydibenzalacetone	4	106.23	429.68
4	3-methoxy-4-hydroxybenzalacetone	6	84.62	92.29
5	2-hydroxybenzalacetone	6	87.34	97.91
6	3-hydroxybenzalacetone	4	90.65	137.09
7	BHT		87.73	343

Substituents with electron donor will stabilize the radical by resonance. Theoretically, the more resonances are formed, the higher the antioxidant activity. Resonance structures of some hydroxybenzalacetone compounds of synthesis results were presented in Fig. 1-6. Compounds that have six radical resonance structures are estimated as more active antioxidant than compounds that only have 4-5. Therefore, based on the number of resonance radical, compounds **1, 5** and **6** are more active as an antioxidant than **2, 3** and **6**.

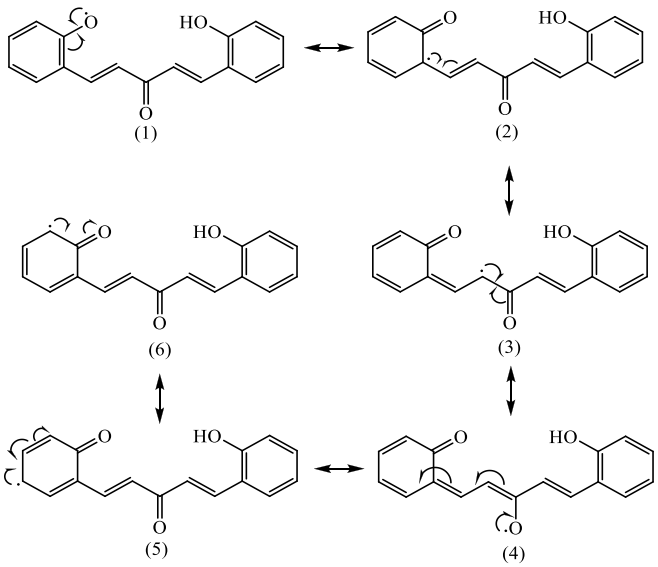

Fig 1. Resonance radical structure of compound 1

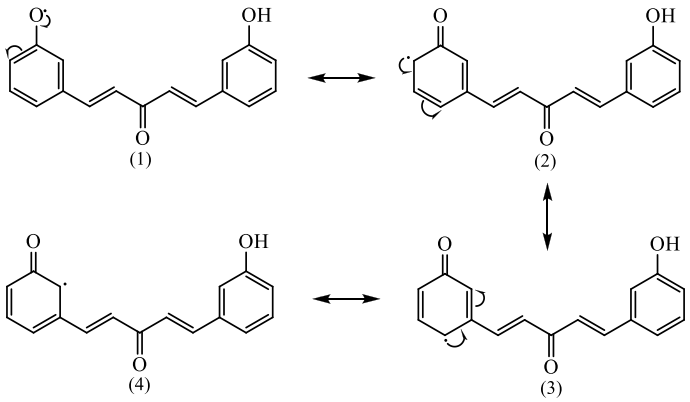

Fig 2. Resonance radical structure of compound 2

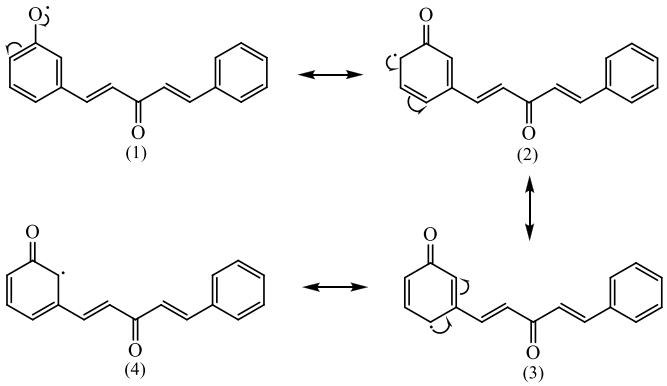

Fig 3. Resonance radical structure of compound 3

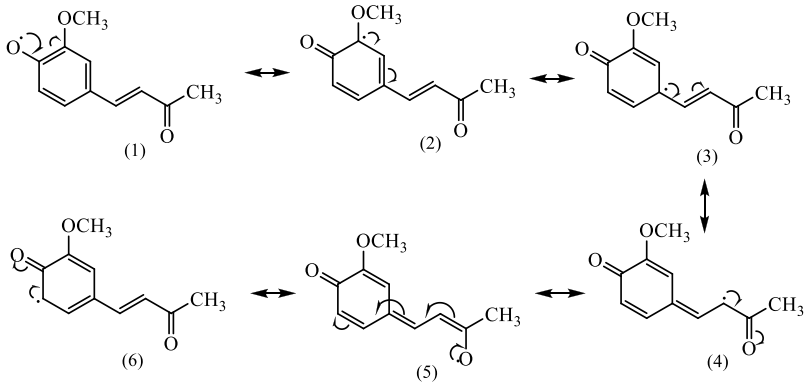

Fig. 4. Resonance radical structure of compound 4

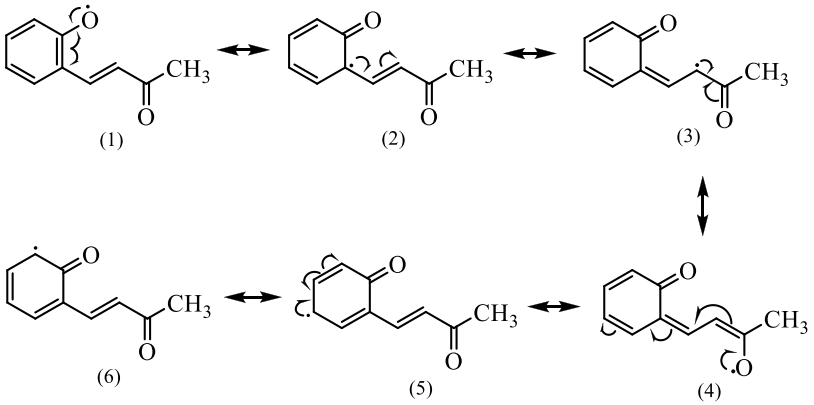

Fig. 5. Resonance radical structure of compound 5

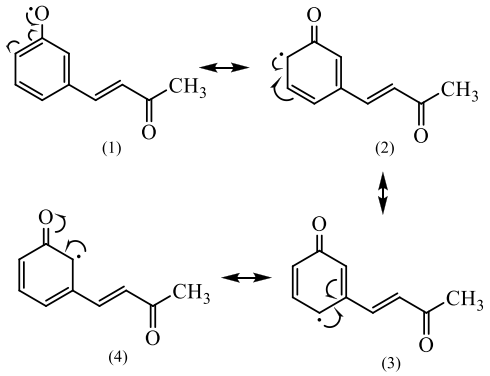

Fig 6. Resonance radical structure of compound 6

BDE-OH (Bond dissociation enthalpy-OH) could be used as other parameter of antioxidant activity. BDE-OH or termination of bond enthalpy is the energy required to break RO-H bond. The weaker the hydrogen bond, the easier separation of O-H. Primary antioxidant activity determined by their ability to donate hydrogen to free radicals, so there is a correlation between the antioxidant activity and BDE. The less energy required to remove the hydrogen, the greater the compound to donate hydrogen. Reducing BDE value is equivalent to the increasing

antioxidant activity.

The lower the BDE, the higher the radical stability level. Molecules can react with peroxyl radicals (ROO•) quickly. BDE-OH on ROO-H is 88 kcal/mol, thus ArOH antioxidant compounds must have less. If BDE-OH compound more than BDE ROOH, it will prefer to attack radical ROO-H to form peroxyl radicals and followed by a radical chain reaction (Hussain et al., 2003). The BDE parameters could be used to conclusively predict the antioxidant activity of a compound. Determination of BDE can be evaluated using AM1 semi empiric computational chemistry method. BDE hydroxybenzalacetone that evaluated is resonance number 1, because ArOH which is releases hydrogen to form •ArO, has the lowest BDE (Aini et al., 2007).

Compounds **1, 4** and **5** have BDE-OH less than 88 kcal/mol. Compounds **4** and **5** have a BDE-OH slightly lower than ROOH and BHT as controls. If conclude from the BDE-OH data, compound **4** is more active than **5**. Compound **4** has two substituents, hydroxy and methoxy groups that are act as an electron donor in the ortho position. Electron donating groups on adjacent positions will increase the stability of phenolic radicals thus enhance its activity as an antioxidant. This is consistent with Yamagami's et al. (2004).

Compounds **2, 3** and **6** have 4 resonances. Resonance of the three compounds are only involves shifting electrons in aromatic compounds. Additional conjugated double bonds in the compound does not affect the stability of the radical. The longer the chain that do not contribute to the resonance, the more it will reduce the stability of the radical, so the antioxidant activity will decreased.

CONCLUSION AND SUGGESTION

The order of antioxidant activity based on BDE-OH data from the most active compound are **1, 4**, **5, 6, 2** and **3**, respectively. The order of antioxidant activity based on IC_{50} data, differ only in the last two compounds, which is **3** is more active than **2**. However, the value is not significantly difference. Based on the results of this study, antioxidant activity assay by using the deoxyribose degradation inhibition method compared with BDE could be suitable for structure-activity analysis as antioxidant for analog benzalacetone test.

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