Bulgarian Chemical Communications, Volume 54, Special Issue B2 (pp. 61-65) 2022 DOI: 10.34049/bcc.54.B2.0399

DFT investigation of the radical-scavenging activity of biogenic amines

Y. Nacheva[1], A. Garkinin[2], I. Trenchev[2], Zh. Velkov[1*]

South-West University "Neofit Rilski", Blagoevgrad, Bulgaria
[2]University of Library Studies and Information Technologies (ULSIT), Sofia, Bulgaria

Received: November 25, 2021; Revised: June 07, 2022

It is known that some biogenic amines reduce the level of active radicals in the body. On the other hand, they have a catechol fragment in their molecule. The presence of this fragment is directly related to radical scavenging activity in flavonoids and phenolic acids. To date, however, there is no comprehensive theoretical investigation of the structural causes of such activity in biogenic amines. In this study, 13 biogenic amines were investigated using the DFT/UB3LYP functional and the orbital basis 6-311++G(d,p). It was found that their radical scavenging activity is comparable and in some cases greater than that of phenolic acids. The role of the side chain and of the amino group was evaluated.

Keywords: biogenic amines; radical-scavenging activity; DFT, enthalpy changes.

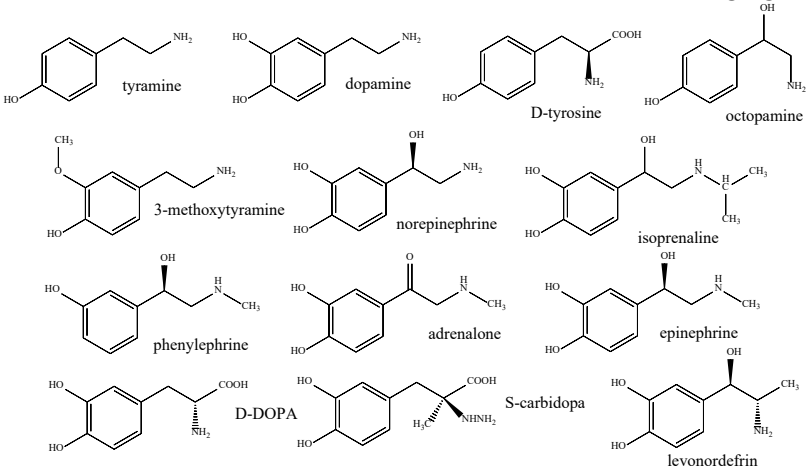
INTRODUCTION

Biogenic amines (BA) are polar or low-polar nitrogen bases (Scheme 1) comprising an aliphatic chain (putrescine, cadaverine, spermine, and spermidine), a benzene (tyramine, phenylethylamine) or heterocyclic ring (histamine, pyrrolidine) [1]. According to their physiological functions and the number of amino groups [2] they can be divided into (i) monoamines – containing one amino group and acting as neuromodulators or neurotransmitters [3], their reduced level in the organisms is a cause of neurodegenerative diseases [4]; and (ii) polyamines – which own two or more amino groups and are involved in physiological processes such as cell growth and differentiation [5].

At neutral pH they form ammonium cations which stabilize the structure of chromosomes and membranes by electrostatic interactions with negative charges of nucleic acids and phospholipids [6-8]. These compounds can be toxic when present in higher concentrations [9], but on the other hand, BAs are compounds that are crucial for maintaining cell viability, as well as the right direction of the organism's metabolic processes, such as protein synthesis, hormone synthesis and DNA replication.

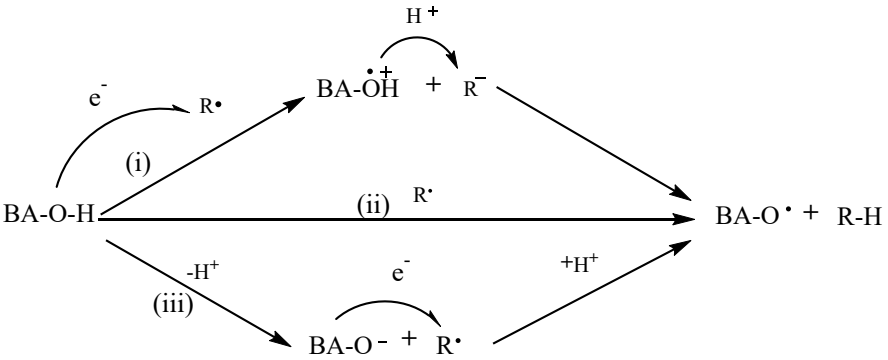
A possible antioxidant role of BA has been reported in the literature. Catecholamines and their metabolites appear to play a key role in the redox balance for the formation of new synapses and the removal of old ones [10], while melatonin and its metabolites are involved in the reduction of oxidative stress [11]. Among polyamines, it has been found that spermine acts as a free radicals' scavenger in nuclei, mitochondria and brain [12-14] and as a radical-scavenger in lipoperoxidation *in vivo* [15].

The ability of phenols to react directly with radicals can be assessed by the enthalpy change of the dissociation of its readily breakable O-H bonds. Calculating the enthalpies of dissociation of the O-H bond by different mechanisms shows the most preferred among them and the proclivity of compounds to participate in reactions with active radicals – radical-scavenging activity.



Scheme 1. Investigated BA

* To whom all correspondence should be sent:
E-mail: *jivko_av@abv.bg*



Scheme 2. Mechanisms of O-H bond dissociation

The reaction between phenolic antioxidants and active radicals can proceed following different mechanisms, but three of them are most often discussed in the literature [6–9] (Scheme 2): (i) electron transfer from the phenol to the active radical, which produces a cation-radical and the radical is converted into an anion; the electron transfer is followed by a proton transfer from the cation-radical to the anion (SET-PT); (ii) direct hydrogen atom transfer between the antioxidant and the active radical (HAT); (iii) deprotonation of the antioxidant followed by an electron transfer from the resulting anion to the active radical; the next step is the protonation of the anion produced by the active radical (SPLET).

Since the selected BAs possess phenolic hydroxyl groups, their radical scavenging potential could be evaluated by the same way as the other phenolic type radical-scavengers.

Computational details

The calculations were carried out using the density functional theory (DFT) [16], as implemented in the Gaussian09 program package [17]. The optimization of the geometry was performed with the Becke 3-parameter hybrid exchange functional combined with the Lee-Yang-Parr correlation functional (B3LYP) [18, 19] with the standard 6-311++G(d,p) basis set [20]. All possible intramolecular interactions were taken into account in the initial geometries. For all structures the harmonic vibrational frequencies were computed to confirm the true minima on the calculated potential surface.

Solvent effects on the calculated structures were investigated with the self-consistent reaction field (SCRF) method *via* the polarized continuum method (PCM) [21].

The total enthalpies of the species X are usually estimated from the equation:

$$H(X) = E_0 + ZPE + \Delta H_{trans} + \Delta H_{rot} + \Delta H_{vib} + RT, \quad (1)$$

where E_0 is the calculated total electronic energy, ZPE stands for zero-point energy, ΔH_{trans}, ΔH_{rot}, and ΔH_{vib} are the translational, rotational and vibrational contributions to the enthalpy. Finally, RT represents the pV-work term added to convert the internal energy into enthalpy. The total enthalpies were calculated at T = 298 K. The ZPE values were not scaled.

The enthalpy changes in the three possible reaction mechanisms of an O-H bond dissociation were calculated according to the scheme:

HAT
BA-O-H → BA-O• + H• BDE = H(BA-O•)+H(H•) - H(BA-O-H)

SET-PT
BA-O-H → BA-O-H+ + e− IP = H(BA-O-H)+H(e−) - H(BA-O-H)
BA-O-H+ → BA-O• + H+ PDE = H(BA-O•)+H(H+) - H(BA-O-H)

SPLET
BA-O-H → BA-O− + H+ PA = H(BA-O−)+H(H+) - H(BA-O-H)
BA-O− → BA-O• + e− ETE = H(BA-O•)+H(e−) - H(BA-O−)

Scheme 3. Possible mechanisms of O-H bond dissociations and their corresponding enthalpy changes

The enthalpies of the hydrogen atom, proton and electron in water are taken from the literature [22], the used proton enthalpy (H_{H+}) in water is –1083.803 kJ.mol^{-1} (6.197 kJ.mol^{-1} in vacuum); the used enthalpy of an electron (H_{e^-}) in water is –232.676 kJ.mol^{-1} (3.145 kJ.mol-1 in vacuum), the used enthalpy of a hydrogen atom ($H_{H\cdot}$) in water is –1316.479 kJ.mol^{-1} (-1312.479 kJ.mol^{-1} in vacuum).

RESULTS AND DISCUSSION

In vacuum

BDE

According to the O-H BDE values, the hydroxyl groups of the studied BAs can be divided into three groups. In the first group are hydroxyl groups with low reactivity and BDE above 338 kJ.mol^{-1}. In the second group are hydroxyl groups with BDE from 310 to 338 kJ.mol^{-1} and in the third group are hydroxyl groups with high reactivity and BDE below 310 kJ.mol^{-1}.

Table 1. Calculated enthalpy changes in vacuum and in water (in kJ.mol^{-1})

Compound	Vacuum	Water				
	BDE	BDE	IP	PDE	PA	ETE
3-methoxytyramine	338.99	326.52	312.34	5.00	74.38	242.77
adrenalone (3)	324.48	331.39	324.85	-2.65	105.74	216.28
adrenalone (4)	321.29	330.13	324.85	-3.91	90.06	230.71
D DOPA (3)	305.23	315.88	331.20	-24.51	118.14	188.47
D DOPA (4)	303.80	312.81	331.20	-27.58	118.89	184.55
dopamine (3)	307.47	314.28	322.77	-17.68	119.92	185.00
dopamine (4)	303.64	309.94	322.77	-22.03	121.16	179.41
D-tyrosine	340.53	341.27	354.97	-22.89	141.39	190.51
epinephrine (3)	306.17	315.47	327.97	-21.69	118.42	187.68
epinephrine (4)	304.09	312.70	327.97	-24.45	118.08	185.26
isoprenaline (3)	305.97	315.36	327.52	-21.35	118.56	187.43
isoprenaline (4)	303.72	312.48	327.52	-24.22	118.34	184.78
levonordefrin (3)	303.70	313.24	325.11	-21.06	119.62	184.25
levonordefrin (4)	302.74	313.09	325.11	-21.21	115.97	187.75
norepinephrine (3)	308.82	317.22	333.55	-25.52	118.44	189.41
norepinephrine (4)	305.81	313.35	333.55	-29.39	119.58	184.4
octopamine	340.35	342.05	348.39	-15.53	138.49	194.20
phenylephrine	345.60	348.25	363.58	-24.52	136.99	201.90
S-carbidopa (3)	305.92	314.82	330.42	-24.79	118.84	186.61
S-carbidopa (4)	304.37	312.72	330.42	-26.89	118.45	184.90
tyramine	339.38	341.31	333.28	-1.16	144.86	187.08

In the first group is the hydroxyl group of phenylephrine, followed by octopamine, D-tyrosine, tyramine and 3-methoxytyramine. All these compounds own one phenolic hydroxyl group. Only in 3-methoxytyramine, there is a second substituent in the phenyl ring – a methoxy group at the 3rd position in the phenyl ring. In phenylephrine, the hydroxyl group is in the 3rd position relative to the aliphatic side chain, while in all other first-group BAs the hydroxyl group is in the 4th position.

The methoxy group in 3-methoxytyramine increases the electron density in the aromatic system, which usually reduces the O-H BDE of phenolic hydroxyl groups, but on the other hand, the hydrogen of the dissociable hydroxyl group is engaged in a hydrogen bond with the oxygen of the methoxy group, which stabilizes the compound and additional energy is needed to be detached. Ultimately, the positive mesomeric effect is in practice offset by the negative effect of the hydrogen bond on the ability of the O-H bond to dissociate to a radical and a hydrogen atom.

The compound adrenalone has two phenolic hydroxyl groups at adjacent positions in the phenyl ring (catechol structure) and occupies an intermediate place according to BDE. Their BDEs in vacuum are 321.29 and 324.48 kJ.mol^{-1}, which is significantly less than the BDE of the first-group compounds. At the same time, this value is much higher than the BDE values for the other catechol-containing compounds. However, only adrenalone has a carbonyl group attached directly to the aromatic ring. Therefore, it is the only compound in which the π-electron system is extended to the side chain. Among the other compounds studied, there is no one in which a functional group so effectively draws electron density from the aromatic system. The expansion of the system has a favorable effect on reactivity, when anions are formed in the reaction. This is not the case when radicals are formed during the reaction. On the other hand, however, the charge transfer in the aromatic system stabilizes the input compound and increases the BDE. This is the reason BDE in adrenalone to have an intermediate value.

All other amines have a catechol ring. The presence of such a fragment in the organic compounds is sufficient for the manifestation of radical scavenging properties. The least reactive among the third group compounds is the hydroxyl group at the 3rd position in norepinephrine (308.82 kJ.mol^{-1}). The small differences in the reactivity of these compounds are due to the influence of the side

chain: the more efficiently it donates electron density to the aromatic system, the lower the BDE value is.

Seven of the compounds have O-H BDE about 308 kJ.mol^{-1} and the most reactive hydroxyl group is that at the 4th position in the phenol ring of levonordefrine (302.74 kJ.mol^{-1}). The side chain is composed of three sp3-hybrid carbon atoms and an amine group. All other compounds have a side chain of two carbon atoms and one amine group. In the structure of S-carbidopa and D-DOPA there is a carboxyl group at the end of the side chain and there is no hydroxyl group adjacent to the aromatic ring. As a result, the O-H BDE of these compounds are greater than the others.

In water

BDE

The polarizing effect of water affects the BDE of the O-H bonds in different ways. There is a significant decrease of BDE upon transition from vacuum to an aqueous medium in 3-methoxytyramine with 12.47 kJ.mol^{-1}. In all other compounds there is an increase of the BDE, which is the expected change. Typically, the decrease in BDE in any environment is due to greater radical stabilization than the input compound. However, the large decrease of O-H BDE in 3-methoxytyramine is due to a decrease in the stability of the input compound.

In an aqueous medium, the strength of the intramolecular hydrogen bond in the compound decreases. Hydrogen bonds in an aqueous medium are weaker than in a vacuum, which reduces the stability of the catecholic radicals (where the stabilization effect of the hydrogen bond is more essential) resulting in an increase of the BDEs. The water alters the BDE of these compounds by the strength of the hydrogen bonds in the compounds and in the corresponding radicals.

The increase in BDE of the hydroxyl group at the 4th position of adrenalone by about 4 kJ.mol^{-1} is due to the additional polarization of the carbonyl group and the stronger charge transfer from the aromatic ring to this group. Withdrawal of electron density from the aromatic ring always increases the BDE of the OH bonds. Despite the significant difference in the structure of this amine - the phenyl ring-conjugated carbonyl group, there is no significant difference in its properties.

The other amine that differs from the others is the phenylephrine. It has one hydroxyl group at the 3rd position relative to the side chain. Probably this is why the phenylephrine has the highest BDE in vacuum (345.60 kJ.mol^{-1}) and in water (348.25 kJ.mol^{-1}), and the highest IP (363.58 kJ.mol^{-1}).

IP

The ionization potential is a descriptor of the compounds ability to donate electrons: to be a reducing agent. However, oxy-reduction reactions take place in polar solvents and the calculation of IP in vacuum has less practical value. In fact, our calculations show that the IP values of the investigated compounds are by about 200 kJ.mol^{-1} lower in water than in vacuum.

The three compounds with the highest IP in the aqueous medium are: phenylephrine (363.58 kJ.mol^{-1}), followed by D-tyrosine (354.97 kJ.mol^{-1}) and octopamine (348.39 kJ.mol^{-1}). These are the weakest reducing agents among the studied amines.

The most reactive compound is 3-methoxytyramine (312.34 kJ.mol^{-1}), followed by dopamine (322.77 kJ.mol^{-1}), adrenalone (324.85 kJ.mol^{-1}), levonordefrine (325.11 kJ.mol^{-1}), isoprenaline (327.52 kJ.mol^{-1}) and epinephrine (327.97 kJ.mol^{-1}). The only compound with an intermediate IP is the norepinephrine (333.55 kJ.mol^{-1}). The SET-PT mechanism was found to be more probable than the HAT mechanism in aqueous media for three of them: 3-methoxytyramine, adrenalone and tyramine. For the other compounds, the opposite is true - the HAT mechanism is more probable.

PDE

Hence, the proton abstraction from the cation-radical is not the rate determining step. In this step of the SET-PT mechanism, the detachment of a proton from the cation-radical implies a significantly smaller positive (3-methoxytyramine) or negave (all other compounds) change in enthalpy.

PDE reflects the electron density distribution of the cation-radical and the uncharged radical that is obtained from it after proton detachment. PDE is a descriptor of the acidity of the cation-radical, but because this process takes place with significantly less change in enthalpy, it is much more probable.

Lowest acidity possesses the cation-radical of 3-methoxytyramine (5.00 kJ.mol^{-1}), followed by the cation-radical or tyramine (-1.16 kJ.mol^{-1}) and adrenalone (-2.65 kJ.mol^{-1}). The strongest proton acids are the cation-radicals of norepinephrine, D-DOPA and S-carbidopa (See Table 1).

With the exception of PDE of 3-methoxytyramine (4.99 kJ.mol^{-1}), all others have negative values for PDE, which indicates a spontaneous deprotonation process at room temperature.

PA

The strongest acids among the test compounds are 3-methoxytyramine (74.38 kJ.mol^{-1}) and adrenalone ((4) 90.06 kJ.mol^{-1} and (3) 105.74

kJ.mol^{-1}). They are followed by a series of compounds with intermediate acidity (between 118 and 122 kJ.mol^{-1}) and four compounds with PA above 135 kJ.mol^{-1}, all of them featuring a single O-H-phenolic group.

It can be argued on the significance of this descriptor, but we believe that the acidity of hydroxyl groups is decisive for the behavior of a phenolic radical scavenger. If the acidity of a phenol is high enough and it maintains a sufficiently high concentration of deprotonated phenolic hydroxyl groups, this will almost certainly direct the reaction to the SPLET mechanism instead the SET-PT mechanism. The oxidation of an anion in the second step is always easier than the oxidation of the compound itself.

According to this logic, the listed compounds are also candidates for interaction with radicals by the SPLET mechanism. Other compounds may also interact by this mechanism.

ETE

The anions are too unstable in a vacuum. When the molecules are immersed in water (implicitly), the anions they produce are much more stable. This is the reason for a strong decrease in PA in water and an increase in ETE to the range of 179.41 to 242 kJ.mol^{-1}.

It turns out that from the strongest acid (PA=74.38 kJ.mol^{-1}) in water (3-methoxytyramine) is the most difficult to tear off an electron (242.77 kJ.mol^{-1}). The next compound in this ranking is with 12 kJ/mol less ETE. All other compounds have an ETE below 200 kJ.mol^{-1}. Three of them have an ETE above 190 kJ.mol^{-1}, and the others, the most reactive, have an ETE below 190 kJ.mol^{-1}.

It is easiest to detach an electron from the anion which is obtained after deprotonation of the hydroxyl group at the 4th position in dopamine (179.41 kJ.mol^{-1}). And this is the strongest radical-scavenger among the studied BAs.

CONCLUSION

Thirteen BA have been investigated using the reliable DFT functional and a high theoretical level orbital basis. It has been found that their radical scavenging activity is comparable and in some cases greater than that of phenolic acids from our previous investigations. The role of the side chain amino group was evaluated. It has the strongest impact on their reducing properties and makes the SET-PT mechanism much more likely compared to most of the phenolic acids. However, after the deprotonation of a hydroxyl group, the SPLET mechanism provides the least change in the enthalpy of electron detachment and this is the preferred mechanism for all BAs.

REFERENCES

1. M. Papageorgiou, D. Lambropoulou, C. Morrison, E. Kłodzinska, J. Namiesnik, J. Płotka-Wasylka, *Trends Anal. Chem.,* **98,** 128 (2018).
2. R. Stevanato, M. Bertelle, S. Fabris, *Pharmacology & Pharmacy,* **4,** 696 (2013).
3. S. H. Snyder, *Science,* **224,** 22 (1984).
4. S. Surendran, S. Rajasankar, *Neurol. Sci.,* **31**, 531 (2010).
5. C. Tabor, H. Tabor, *Ann. Rev. Biochem.,* **53,** 749 (1984).
6. R. Kaur-Sawhney, A. Altman, W. Galston, *Plant Physiol.,* **2**, 158 (1978).
7. R. Stevanato, A. Wisniewska, F. Momo, *Arch. Biochem. Biophys.,* **346,** 203 (1997).
8. F. Momo, S. Fabris, R. Stevanato, *Arch, Biochem. Biophys.,* **382,** 224 (2000).
9. A. Shashank, A. K. Gupta, Sh. Singh, R. Ranjan, *Curr. Nutr. Food Sci.*, **17**, 1 (2021).
10. J. Smythies, *Antioxid. Redox Signalings*, **2**, 575 (2000).
11. A. Galano, D. Xian Tan, R. J. Reiter, *J. Pineal Res.*, **51**, 1 (2011).
12. H. C. Ha, N. S. Sirisoma, P. Kuppusamy, J. L. Zweier, P. M. Woster, R. A. Casero, *Proc. Natl. Acad. Sci. U. S. A.*, **95**, 11140 (1998).
13. I. G. Sava, V. Battaglia, C. A. Rossi, M. Salvi, A. Toninello, *Free Radical Biol. Med.*, **41**, 1272 (2006).
14. N. A. Velloso Bellé, G. Dalmolin, G. Fonini, M. A. Rubin, J. B. Teixeira Rocha, *Brain Res.*, **1008**, 245 (2004).
15. S. M. Hernandez, M. S. Sanchez, M. N. Schwarcz de Tarlovsky, *Acta Trop.*, **98**, 94 (2006).
16. W. Kohn, A. D. Becke, R. G. Parr, *J. Phys. Chem.*, **100**, 12974 (1996).
17. http://gaussian.com/.
18. A. D. Becke, *Phys. Rev*. A, **38**, 3098 (1988).
19. C. Lee, W. Yang, R. G. Parr, *Phys. Rev. B,* **37**, 785 (1988).
20. R. G. Parr, *Horiz. Quant. Chem.,* **3**, 5 (1980).
21. J. B. Foresman, T. A. Keith, K. B. Wiberg, *J. Phys. Chem.,* **100**, 16098 (1996).
22. J. Rimarčík, V. Lukeš, E. Klein, M. Ilčin, *J. Mol. Struct.: THEOCHEM,* **952,** 25 (2010).