REACTIVITY AND RADICAL SCAVENGING ACTIVITY POTENTIAL OF MELATONIN AND ITS DERIVATIVES

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ABSTRACT

Melatonin (5-methoxy-N-acetyltryptamine) is hormone produced by human organism with a general function to regulate the processes of sleep and wakefulness, but it has also well proved antioxidant potential. In this study, the dissociation enthalpies of the most easily cleaved N-H and C-H bonds in melatonin and its three selected derivatives, formed during reactions with HO• and H_2O_2, were calculated. B3LYP/DFT quantum chemical calculations with a 6-311++G orbital basis set, accounting for the solvent effect of water using the SCRF/PCM approach, were performed. The results revealed that, after the transformation of melatonin, the resulting compounds exhibited radical-scavenging properties similar to those of the parent compound. This behavior is atypical for most radical scavengers, as they usually undergo conversion into reactive and more toxic derivatives when reacting with oxygen-containing species. Finally, these findings provide valuable insight into the critical role of melatonin in living organisms.

Keywords: melatonin and derivatives, radical-scavenging activity, DFT/B3LYP.

INTRODUCTION

Melatonin (N-acetyl-5-methoxytryptamine, Fig. 1) is an endogenous molecule in the pineal gland of the brain.

It has diverse key physiological functions such as regulation of the circadian rhythm, sleep induction, seasonal reproductive regulation, immune enhancement and the light-dark signal transduction. Most of them were discovered by the Reiter [1, 2]. The same author and co-workers also established the ability of melatonin to serve as a potent radical scavenger [3]. They have been demonstrated that melatonin directly scavenges the most reactive and cytotoxic reactive oxygen species - hydroxyl radical (HO•) [4]. Consequently, the free radical scavenging and antioxidant properties of melatonin have been documented in hundreds of publications using both in vitro and in vivo models.

The Reiter group also reported cyclic 3-hydroxymelatonin as the product formed upon the reaction of melatonin with HO• (C-3-OHM, Fig. 2). This product has been detected in the urine of both rats and humans [5]. The concentration of this compound

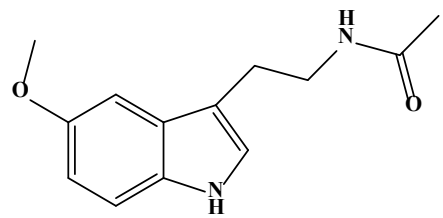


Fig. 1. Melatonin structure.

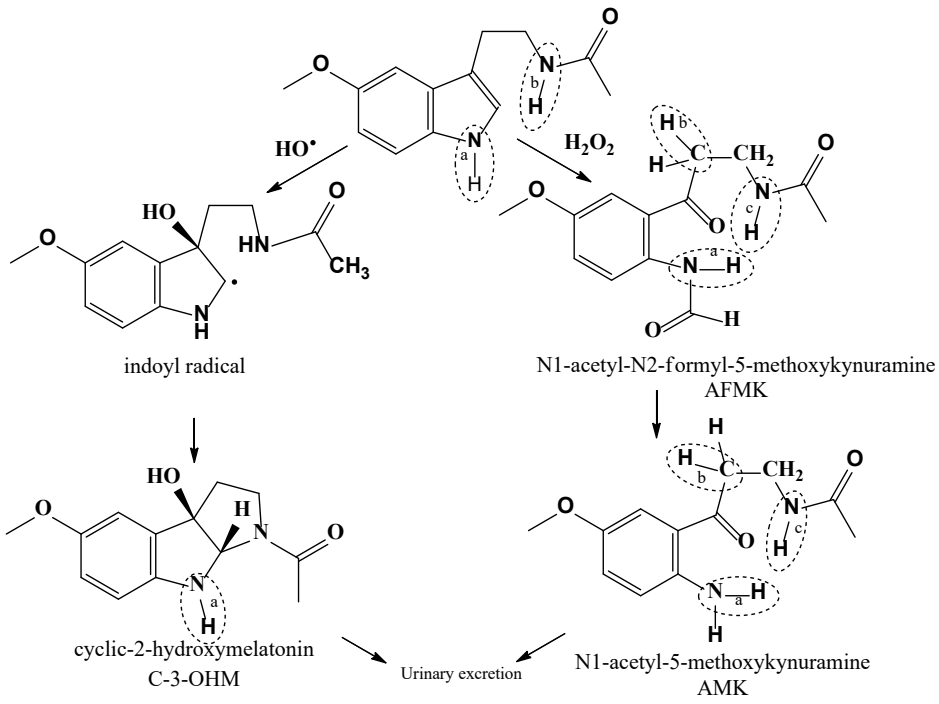


Fig. 2. The conversion of melatonin into multiple derivatives through its interaction with H_2O_2 and HO•.

in the urine rises when melatonin is administered by rats and in case when rats are exposed to whole-body ionizing radiation, which leads to the generation of large quantities of HO•. These discoveries align with the idea that melatonin scavenges HO• in vivo and further organism expelling the resulting product, namely C-3-OHM, through urinary excretion (fig. 2).

The studies of the same research group revealed that melatonin serves as a scavenger for H_2O_2, resulting in the formation of N1-acetyl-N2-formyl-5-methoxykynuramine (AFMK), which itself may possess significant scavenging properties (Fig. 2) [6].

The role of melatonin and its derivatives in scavenging reactions is still under investigation. [7-9]. Melatonin and its derivatives have not the typical structural features of phenolic acids, flavonoids, coumarins, or carotenoids. However, it has been revealed to be a potent radical scavenger, safeguarding one of the most intricate and highly susceptible to oxidative damage organ - the human brain. To explain this phenomenon herein, we are focused on quantum chemical study of the reactivity and radical-scavenging potential of melatonin and its derivatives, which are generated during their reaction with hydroxyl radical and hydrogen peroxide.

EXPERIMENTAL

Computational details

The calculations were carried out using DFT functional B3LYP as implemented in the Gaussian16 program package with the standard 6-311++G(d, p) basis set [10 - 14]. Solvent effects on calculated structures were investigated with the self-consistent reaction filed, via the polarized continuum method [15]. Solvation calculations with full geometry optimization were performed for the water at the same level as in the gas phase.

The ability of compounds to react directly with radicals can be assessed by the enthalpy changes in the dissociation of their most readily breakable N-H and C-H bonds. It is sufficient to consider the enthalpy changes of bonds dissociation via three different mechanisms (Fig. 3): SET-PT((i) Sequential Electron-Transfer and Proton-Transfer), HAT((ii) Hydrogen Atom Transfer) and SPLET ((iii) Sequential Proton

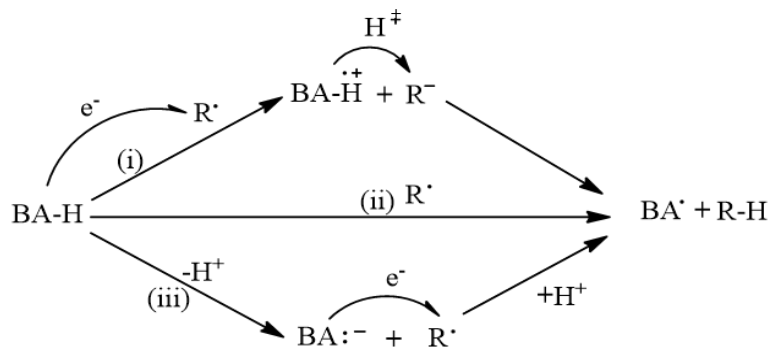


Fig. 3. Mechanisms of bond dissociation.

Lost Electron Transfer) - ionization potential (IP), proton detachment enthalpy (PDE), bond dissociation enthalpy (BDE), proton affinity (PA), electron transfer enthalpy (ETE).

The total enthalpies of the species X are usually estimated from the Eq. (1):

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

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.

Reaction enthalpies are calculated as a difference between the enthalpies of the resulting entities and the enthalpies of the input products.

The enthalpies of the hydrogen atom, proton and electron in water are taken from the literature, the used proton enthalpy (H_{H+}) in water is -1083.803 kJ mol^{-1} [16]. The used enthalpy of an electron (H_{e-}) in water is -232.676 kJ mol^{-1} and the used enthalpy of a hydrogen atom ($H^{H\cdot}$) in water is -1316.479 kJ mol^{-1}.

RESULTS AND DISCUSSION

Table 1 presents the calculated enthalpies of compounds, cation-radicals, anions, and neutral radicals obtained during the cleavage of selected N-H to C-H bonds, as shown in Fig. 2, along with enthalpy changes, descriptors of the propensity for each of the three hydrogen atom abstraction mechanisms, as illustrated in Fig. 3.

Propensity to SET-PT mechanism of bond dissociation

According to the calculated ionization potential (IP) (Fig. 3), first stage of mechanism (i)) - a general descriptor of a molecule's ability to act as a reducing agent - the most reactive among the four investigated compounds is cyclic C-3-OHM (265.026 kJ mol^{-1}). AMK (280.541 kJ mol^{-1}) ranks after C-3-OHM in this indicator property, and the melatonin (287.648 kJ mol^{-1}) takes the third place. The weakest reducing agent is AFMK (its IP is 334.57 kJ mol^{-1}).

Melatonin and its derivatives have proven to be markedly more potent reducing agents compared to any phenolic compounds (our data 365-500 kJ mol^{-1}) we have examined thus far. Additionally, it is noteworthy that melatonin ranks third in its propensity to participate in a radical-trapping reaction through the SET-PET mechanism.

Propensity for the HAT mechanism in bond dissociation

The Bond Dissociation Energy (BDE) emerges as the most energetically demanding mechanism for bond dissociation among the compounds analysed

Table 1. Enthalpies of compounds, radicals and ions, as well as the N-H and C-H bonds dissociations enthalpies.

Compound		Cation Radicals	Radicals	Anions	Compound	BDE	IP	PDE	PA	ETE
Melatonin	a*	-764.722	-764.285	-764.442	-764.920	360.660	287.648	63.824	172.302	179.170
	b		-764.258	-764.428		429.939		133.103	208.977	211.773
C-3-OHM	a	-839.960	-839.522	-839.656	-840.149	337.858	264.026	64.643	211.474	117.195
AMK	a		-801.427	-801.568		380.013		90.284	233.214	137.614
	b		-801.429	-801.583		373.922		84.193	193.172	171.562
	c	-801.874	-801.403	-801.580	-802.069	442.802	280.541	153.072	201.114	232.499
AFMK	a	-915.211	-914.779	-914.949	-915.428	397.128	334.574	53.367	173.525	214.415
	b		-914.788	-914.952		372.541		28.779	166.090	197.263
	c		-914.761	-914.939		443.411		99.65	198.250	235.973

**The letters denote the dissociation sites indicated on the Fig. 2.*

in this study in an aqueous environment. The C-3-OHM exhibits the greatest propensity for homolytic cleavage (337.858 kJ mol^{-1}) of the N-H bond, higher than melatonin (360.660 kJ mol^{-1}). In the other two compounds (AMK and AMFK) there is one bond each, whose BDE is almost as much greater than melatonin (C-H bond in AMFK with BDE 372.54 kJ mol^{-1} and in AMK - 373.92 kJ mol^{-1}).

Propensity for the SPLET mechanism in bond dissociation and acidity-related processes

SPLET mechanism for the N-H or C-H bond dissociation takes place when the acidity of the bond is significant. Anions can undergo oxidation more readily than uncharged compounds, as evidenced in the Table 1. Therefore, the realization of this mechanism is largely determined by the acidity of the compounds in question. In addition, some of the considered substances have a PA that is less than the ETE. In this case, SPLET is determined solely by the acidity of the compounds. In the other case, ETE may prove to be a defining descriptor for the SPLET mechanism.

According to the obtained data, the order of propensity for the SPLET mechanism is as follows: melatonin (PA 172.302 and ETE 179.170 kJ mol^{-1}), followed by AMFK (166.090 and 197.263 kJ mol^{-1}), AMK (193.172 and 171.562 kJ mol^{-1}), and the least reactive is C-3-OHM (221.474 and 117.195 kJ mol^{-1}).

CONCLUSIONS

The enthalpy changes in all stages of the N-H and C-H bond breaking in melatonin and its derivatives were calculated. They describe the radical scavenging potential of melatonin and its metabolites.

The results suggest that HAT is the least likely mechanism for N-H and C-H bond dissociation in an aqueous environment for the studied compounds. The SET-PT mechanism is more likely than HAT, but the most probable mechanism of all is SPLET. This means that homolytic processes involving these compounds are excluded, and their reducing ability is not high enough to directly determine electron donation. The radical-scavenging potential of these compounds is determined by their ability to release a proton (N-H or C-H acidity) and subsequently for the resulting anion to donate an electron.

It demonstrates strong radical-scavenging potential,

comparable to that of phenolic radical scavengers. What distinguishes melatonin from phenols is that the products of its reaction with reactive radicals exhibit almost the same radical-scavenging activity as melatonin itself: AMFK, AMK, and C-3-OHM display remarkably similar radical scavenging potential. This is not the case with the products of phenolic radical scavengers. This may be one of the reasons why evolution assigned it the role of safeguarding the human brain.

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Authors' contributions

V.K.: Contributes to the discussion and interpretation of quantum-chemical results and to the calculation of target descriptors; S.Y.: Provides biochemical expertise to support the discussion and interpretation of the results; Zh.V.: Responsible for the quantum-chemical calculations and contributed to the interpretation and discussion of the results.

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