

Article

Insight into the Strong Antioxidant Activity of Deinoxanthin, a Unique Carotenoid in *Deinococcus Radiodurans*

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Abstract: Deinoxanthin (DX) is a unique carotenoid synthesized by *Deinococcus radiodurans*, one of the most radioresistant organisms known. In comparison with other carotenoids, DX was proven to exhibit significantly stronger reactive oxygen species (ROS)-scavenging activity, which plays an important role in the radioresistance of *D. radiodurans*. In this work, to gain deeper insights into the strong antioxidant activity of DX, the parameters characterizing ROS-scavenging potential were calculated by means of quantum chemical calculations. It was found that DX possesses lower lowest triplet excitation energy for its unique structure than other carotenoids, such as β -carotene and zeaxanthin, which endows DX strong potential in the energy transfer-based ROS-scavenging process. Moreover, the H-atom donating potential of DX is similar to zeaxanthin according to the theoretical homolytic O-H bond dissociation enthalpy. Thus, the large number of conjugated double bonds should be crucial for its strong antioxidant activity.

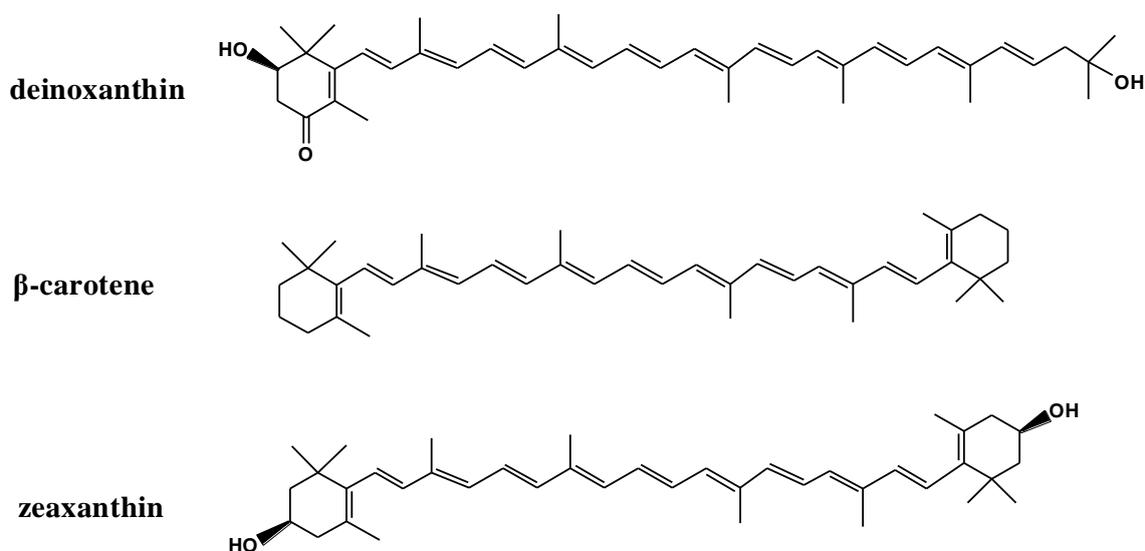
Keywords: deinoxanthin; lowest triplet excitation energy; bond dissociation enthalpy; density functional theory

1. Introduction

Deinococcus radiodurans is a red-pigmented, nonphotosynthetic bacterium well known for its resistance to ionizing radiation [1–3]. It has been demonstrated that cellular antioxidants make important contributions to the radioresistance of *D. radiodurans* besides the efficient and accurate

DNA repair strategy [4,5]. Among nonenzymic antioxidants, carotenoids possess efficient reactive oxygen species (ROS) scavenging capacity [6]. It is interesting to note that *D. radiodurans* synthesizes a unique ketocarotenoid, deinoxanthin (DX, Figure 1), as its major carotenoid [7–9]. DX was proven to exhibit significantly stronger ROS scavenging ability than other known carotenoids, such as β -carotene (BC, Figure 1) and zeaxanthin (ZX, Figure 1) [9], and the strong antioxidant effect of DX plays an important role in the radioresistance of *D. radiodurans* [9]. Therefore, it is interesting to explore the mechanistic underpinnings underlying the higher antioxidant potential of DX relative to other carotenoids. In the present work, by means of quantum chemical calculations, the parameters to characterize the antioxidant potential of DX, including the lowest triplet excitation energy (E_{T1}) and homolytic O-H bond dissociation enthalpy (BDE), were estimated. The theoretical results further our understanding of the higher ROS-scavenging activities of DX compared to BC and ZX.

Figure 1. Chemical structures of deinoxanthin, β -carotene and zeaxanthin.



2. Calculation Methods

The structures of DX, BC and ZX were fully optimized by hybrid density functional theory (DFT) [10,11] and B3LYP [12–14] functional with 6-31G(d) Gaussian basis set. The nature of the stationary point was ascertained by performing harmonic frequency calculations. The lowest triplet state energies (E_{T1S}) of DX, BC and ZX were calculated by time-dependent DFT (TD-DFT) formalism [15–17] with the same basis set. To ensure the accuracy of the results, the O-H BDEs of DX and ZX were estimated using a combined method labeled as (RO)B3LYP/6-311+G(2d,2p)//AM1/AM1, which takes advantage of accuracy and economy [18–21]. As the hydroxyls of DX and ZX are not conjugated with the polyene chain, which should influence little on the O-H bond dissociation reactions, only one double bond in the polyene chain is reserved while the rest was replaced by a methyl when estimating the O-H BDEs of DX and ZX. The two hydroxyl groups of ZX are equivalent and only one is considered. The O-H BDE was estimated according to the following equation, O-H BDE = $H_r + H_h - H_p$ [18–21], in which, H_r is the enthalpy of radical generated through H-abstraction reaction, H_h is the enthalpy of H-atom, -0.49765 hartree, and H_p is the enthalpy of parent molecule.

All calculations were performed using Gaussian 03 package of programs [22].

3. Results and Discussion

Carotenoids are efficient singlet oxygen ($^1\text{O}_2$) quenchers. Owing to the rather low E_{T1} of carotenoids, $^1\text{O}_2$ can be quenched through energy transfer (Equation 1); generating triplet excited state carotenoids and ground state oxygen ($^3\text{O}_2$).



The $^1\text{O}_2$ quenching capability is a good indicator of the E_{T1} . Table 1 lists the TD-B3LYP/6-31G(d) estimated E_{T1} of DX, BC and ZX. The theoretically predicted E_{T1} s of BC and ZX are close to the experimental value [23,24], which verifies the methodology.

The E_{T1} s of the three carotenoids are lower than the deactivation energy of $^1\text{O}_2$ (0.97 eV), which indicates that they are $^1\text{O}_2$ quenchers. Moreover, the E_{T1} of DX is approximately 0.1 eV lower relative to those of BC and ZX. The lower E_{T1} of DX will make the energy transfer process more favorable energetically, relative to the other two carotenoids, which is consistent with the experimental finding that DX possesses stronger $^1\text{O}_2$ quenching ability than BC and ZX [9]. Moreover, it was reported that the $^1\text{O}_2$ quenching enhanced as the number of conjugated double bonds in the polyene chain of carotenoids increased, by examining various naturally occurring carotenoids [25]. Thus, it can be inferred that the higher $^1\text{O}_2$ quenching ability of DX than that of other carotenoids should mainly arise from its extended conjugated double bonds system.

Table 1. Theoretically estimated lowest triplet excitation energies (E_{T1}) of deinoxanthin (DX), β -carotene (BC) and zeaxanthin (ZX) (in eV).

	DX	BC	ZX
Theoretical data	0.69	0.80	0.80
Experimental data	-	0.88 [23]	0.87 [24]

The direct H-atom transfer is one of the most important radical-scavenging processes [18–21]. Taking $\text{RO}\cdot$ as an example, the direct H-atom transfer process can be represented as follows.



Among the three carotenoids, DX and ZX possess hydroxyls as potential H-atom donors in their structures. O-H BDE acts as an appropriate parameter to characterize the H-atom donating ability [18–21]. The O-H BDE of the hydroxyl in the six-membered ring of DX is calculated to be about 101.92 kcal/mol, and that of the butyl hydroxyl at the end of the polyene chain is about 104.71 kcal/mol. This indicates that on thermodynamic grounds the hydroxyl in the six-membered ring should play a predominant role in the H-atom transfer-based ROS scavenging processes of DX. Moreover, the theoretical O-H BDE of ZX is about 101.74 kcal/mol. The close O-H BDEs between DX and ZX imply that they possess similar H-atom donating potential thermodynamically.

Collectively, the extended conjugated double bonds system of DX seems crucial for its strong ROS-scavenging activity. The larger number of conjugated double bonds means DX possesses lower E_{T1} , and thus higher $^1\text{O}_2$ quenching potential through energy transfer, in comparison with BC and ZX. The strong ROS scavenging ability of DX renders this unique carotenoid great potential in antioxidant therapy application.

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