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Effects of the residues on the excitation energies of protonated Schiff base of retinal (PSBR) in bR: A TD-DFT study

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Abstract

Effects of the residues on the excitation energies of protonated Schiff base of retinal (PSBR) in bacteriorhodopsin have been investigated by means of time-dependent density functional theory. The residues around PSBR are replaced by the point charges on atoms. The structures of PSBR and residues are referred from X-ray data. The atomic charges on the each residue were calculated the B3LYP/6-311G(d,p) level. The excitation energy of PSBR perturbed by the point charges on atoms of each residue was calculated at the B3LYP/6-31G(d,p) level. A total of 23 residues and five water molecules around PSBR were considered in the calculations. The large spectral shifts were caused by the Asp212 and Asp85. The origin of the spectral shifts was discussed on the basis of theoretical results.

Keywords: PSBR; Residue effects; Excitation energy; TD-DFT calculation; Point charge

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1. Introduction

The visual pigment rhodopsin [1,2] is a G-protein-coupled receptor containing 11-*cis* retinal chromophore bounded to a lysine residue (Lys296) via a protonated Schiff base linkage. Protonated Schiff base of retinal (PSBR) is one of the visual pigments in humans and also the light-driven *trans*-membrane proton pump in bacteriorhodopsin (bR) [1–3]. PSBR absorbs a light at ca. 440 nm in organic solvents, and its absorption maximum is drastically changed after the binding to apoprotein. Observed absorption maxima range from 360 to 635 nm. This is known as “opsin shift” [4,5]. The absorption spectra and excitation energies of PSBR are strongly affected by the environment around PSBR [6–13]. For the origin of the spectral shifts, several explanations have been attempted: for e.g. (1) electrostatic interaction from a part of amino acid residues of the opsin affects the excitation energy of PSBR; (2) the existence of counter ion near Schiff base; (3) deformation of polyene of PSBR from planar structure caused by steric effect. However, explanations for the spectral shift are speculative, and also the origin of the spectral shift is not clearly understood.

In the present study, to shed light on the origin of the opsin shift of PSBR, the effect of point charges of residues on the excitation energy of PSBR is investigated by means of time-dependent density functional theory (TD-DFT). The residues are replaced by point charges on atoms. It should be noted that this work is first attempt for elucidating the effect of point charges composed of residues on the excitation energy of PSBR.

2. Method of calculation

First, the structure of bR is constructed from X-ray data (PDB code: 1C3W). Second, the residues, by which

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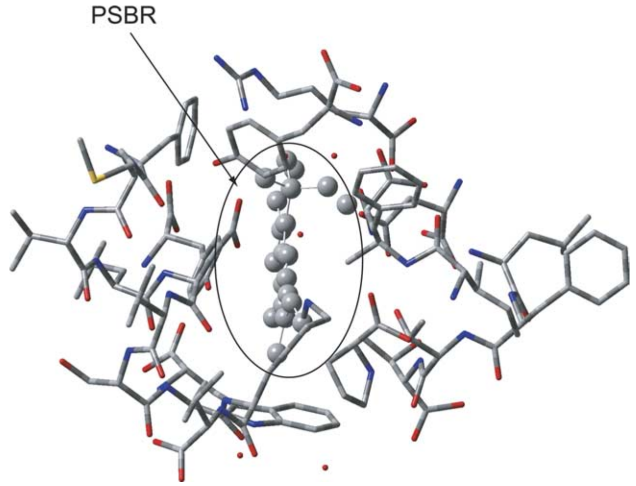

Fig. 1. Illustration of protonated Shiff base of retinal (PSBR) and residues around PSBR examined in the present study. Dot means oxygen atom of water molecule.

at least one of the atoms is contained in less than 8.0 Å from the N–H Schiff base of PSBR, are selected. Third, the similar selection is carried out for the water molecules near PSBR. Finally, 23 residues and five water molecules are selected. The structure of the PSBR–residues thus selected is illustrated in Fig. 1.

The Mulliken atomic charges on each residue are calculated at the B3LYP/6-311G(d,p) level, and then the atoms of each residue are replaced by point charges. Next, the excitation energy of 1:1 pair of PSBR–residue is calculated at the B3LYP/6-31G(d,p) level. The DFT calculations are carried out using GAUSSIAN-03 [14] program package.

3. Results

3.1. Excitation energies of PSBR

First, the excitation energies of PSBR without residues are calculated at the B3LYP/6-31G(d,p) level, and the results are given in Table 1. The first, second, and third excitation energies are calculated to be 2.183, 3.188 and 3.550 eV, respectively. The first excited state is composed of π–π* state in polyene part of PSBR.

Table 1
Excitation energies of PSBR (in eV)

	6-31G(d,p)	6-311G(d,p)	6-311 + G(d,p)
Ground	0.0	0.0	0.0
1st	2.183 (1.213)	2.107 (1.213)	2.136 (1.216)
2nd	3.188 (0.592)	3.164 (0.583)	3.132 (0.571)
3rd	3.550 (0.002)	3.534 (0.002)	3.505 (0.002)

Oscillator strengths are given in parenthesis. The values are calculated by means of the B3LYP method.

To check the basis set dependency, the excitation energies are also calculated using 6-311G(d,p) and 6-311 + G(d,p) basis sets. The results are also given in Table 1. The excitation energies are 2.107 eV (1st), 3.164 eV (2nd) and 3.534 eV (3rd) using 6-311G(d,p) and 2.136 eV (1st), 3.132 eV (2nd) and 3.505 eV (3rd) using 6-311 + G(d,p). These energies are close to those of 6-31G(d,p) basis set, indicating that the B3LYP/6-31G(d,p) calculation will give a reasonable feature on the excitation energy of PSBR and PSBR–residue system.

To test the structural relaxation of PSBR, the geometry optimization of PSBR is carried out at the B3LYP/6-31G(d,p) level, and the excitation energies are calculated using the optimized geometry. The excitation energies are 2.419 eV (1st) and 3.265 eV (2nd).

3.2. Effect of each residue on the excitation energies of PSBR

The excitation energies of PSBR perturbed by the point charges of each residue are calculated at the B3LYP/6-31G(d,p) level. The calculations are carried out for PSBR–residue 1:1 pair in order to elucidate the contribution of each residue to the spectral shift. The results are summarized in Table 2. The shifts of the excitation energies (ΔE_{shift}) are distributed from −0.022 to +0.207 eV.

In the present system, five water molecules are located around the Schiff base. Among them, W402 is the nearest neighboring water molecule from the N–H of Schiff base. The distance is 2.87 Å from the nitrogen atom of Schiff base. This water causes the spectral shift corresponding to the energy of +0.003 eV. The largest of the energy shift by water molecule is caused by W501 (ΔE_{shift} = +0.004 eV). Thus, the effect of water molecule on the energy shift is negligibly small.

The largest spectral shifts are caused by Asp85 and Asp212, which give ΔE_{shift} = +0.188 eV for Asp85 and +0.207 eV for Asp212. The shifts of the excitation energies caused by the other residues are distributed in the range 0.01–0.08 eV. For example, Ser214 and Ala215 are calculated to be +0.049 and +0.053 eV, respectively. The direction of the shifts is all positive except for that of Tyr185, indicating that the first excitation energy of PSBR is blue-shifted by the interaction with point charges for the residues. The shift of TRY185 is negative, but the small value (−0.022 eV).

As mentioned above, it is found that Asp85 and Asp212 give large spectral shifts. The structures of PSBR–Asp212 and PSBR–Asp85 are illustrated in Fig. 2. Asp85 and Asp212 are located above and blow the molecular plane of PSBR. Expanded view of PSBR–Asp85 and PSBR–Asp212 are given in Figs. 2(a) and (b), respectively. The geometrical data are given in Table 3. In PSBR–Asp85, the carbonyl group of Asp85 orients