Electronic circular dichroism spectrum of uridine studied by the SAC–CI method

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Abstract

Symmetry-adapted cluster–configuration interaction (SAC–CI) method was applied to calculate electronic CD spectrum of a nucleoside, uridine. Based on the theoretical CD and absorption spectra, the observed peaks in the experimental spectra were assigned. The excited states of uracil, the base part of uridine, were also calculated for comparison. The origin of CD rotational strength for the low-lying π–$\pi*$ and n–$\pi*$ excited states was analyzed. Rotational strength of the π–$\pi*$ transition depends on the magnitude of the electric and magnetic transition dipole moments, while that of the n–$\pi*$ originates from the angle between the two transition moments.
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1. Introduction

Nucleic acids, DNA and RNA, treat genetic information and play indispensable roles in living organisms. Photochemistry of nucleic acids is also very important, because the irradiation of UV light to DNA causes skin cancer [1–3]. Electron conduction of nucleic acids got attention in material science [4]. To understand the fundamental photochemical processes of DNA or RNA, many experimental and theoretical studies focused on the excited states of nucleotides, the basic unit of DNA and RNA (for review, see Refs. [2,3]).

A popular and simple approach to study excited states would be UV–vis photo-absorption spectroscopy. In the case of the simplest nucleoside, uridine, absorption spectrum shows two peaks (260 and 205 nm) [5]. Since nucleosides are chiral molecules, circular-dichroism (CD) spectroscopy is also useful to find the excited states having very small intensity in the absorption spectrum. CD spectrum of uridine shows four peaks at 267, 240, 210, and

190 nm [5], which illustrates that CD spectrum can detect the excited states being difficult to identify only with the absorption spectrum. This is because the rotational strength, intensity of CD spectrum, depends not only on the electric transition dipole moment (ETDM) but also on the magnetic transition dipole moment (MTDM). Although CD spectrum contains more information, the assignment of the peaks often becomes more difficult than that in the absorption spectrum. This is because CD signal is the result of the cancellation between positive and negative peaks. Since there has been so far no assignment for the CD spectrum of the nucleosides, accurate theoretical calculation provides useful information for understanding the spectrum.

Another point of interest is to figure out the reason why nucleosides are optically active. Based on the observation of the experimental CD spectrum, π–$\pi*$ transitions of nucleosides have strong rotational strength. This fact is actually puzzling, because both π and $\pi*$ orbitals are localized within the base part of nucleosides (see Fig. 1). Since the base part is composed of planner aromatic ring and has mirror symmetry, there should be no intrinsic origin of the optical activity within the base part. It is not clear why π–$\pi*$ transition shows large rotational strength. In

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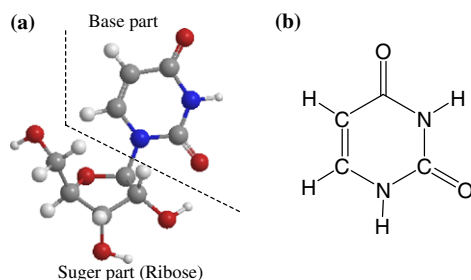
(a) Base part **(b)**

Suger part (Ribose)

Fig. 1. Structures of: (a) uridine and (b) uracil. The H, C, N, and O atoms are colored by white, grey, blue, and red, respectively. (For interpretation of the references in colour in this figure legend, the reader is referred to the web version of this article.)

addition, we found in the present study that n–$\pi*$ transitions also show significant contributions in the CD spectrum, although these transitions are very difficult to detect with the absorption spectrum.

In this study, we studied CD and UV–vis absorption spectra of the simplest nucleoside, uridine, by SAC–CI (symmetry-adapted cluster–configuration interaction) [6–9] method. It is an established method for studying the excited states of molecules, spectroscopy, photochemistry [9]. Recently, we have developed new routines for CD rotational strength [10]. In the next section, we briefly explain CD rotational strength and compare with oscillator strength in the absorption spectrum. In Section 4, the result for uridine is explained and compared with that for uracil. The origin of the optical activity is also discussed in Section 4.

2. Rotational strength of CD spectroscopy

We briefly describe rotational strength of CD spectroscopy. For the detailed explanation, the readers are recommended to refer previous review articles [11–13].

In the UV–vis spectroscopy, oscillator strength for electronic transition from states a to b is proportional to the square of the ETDM.

$$D_{ab} \propto |\bar{\mu}_{ab}|^2 = |\langle \Psi_b \mid \hat{\mu} \mid \Psi_a \rangle|^2 \tag{1}$$

Ψ_a and Ψ_b are wave functions for the states a and b, respectively. $\hat{\mu}$ is the electric dipole operator, $\hat{\mu} = (\hat{x}, \hat{y}, \hat{z})$.

On the other hand, CD spectroscopy measures difference of the absorption ($\Delta\varepsilon$) between right- and left-circularly polarized light. Theoretical definition of the CD signal is given by the rotational strength (R). The R can be calculated as imaginary part of the inner product of the ETDM and MTDM.

$$R_{ab} = \text{Im}[\langle \Psi_a \mid \hat{\mu} \mid \Psi_b \rangle \langle \Psi_b \mid \hat{m} \mid \Psi_a \rangle] \tag{2}$$

$\hat{m}$ is the magnetic dipole moment operator which is proportional to the angular momentum operator. Since the rotational strength includes the MTDM, the CD spectrum can detect excited states having little oscillator strength in the absorption spectrum.

Rotational strength can also be expressed by using the angle θ between the ETDM and MTDM.

$$R_{ab} = \text{Im}[\| \bar{\mu}_{ab} \| \mid \bar{m}_{ab} \mid \cos\theta] \tag{3}$$

This expression indicates that the rotational strength becomes zero if the ETDM and MTDM are orthogonal to each other. This holds for symmetric molecules having no optical activity. Once the symmetry is broken, the R is not zero any longer, and the molecule becomes optically active. This expression is useful for analyzing the source of the rotational strength as we show in Section 4.

3. Computational details

Nucleosides are composed of two parts, base and sugar ring (ribose) as shown in Fig. 1. For computational model, the OH and hydroxymethyl groups in the sugar ring are substituted by the H atoms. Geometry of the ground state of uridine was optimized with DFT(B3LYP) with 6-31G(d) sets using GAUSSIAN03 program [14].

For calculating the excited states and CD spectrum, the basis functions employed were TZ [15] with double polarization functions [16] plus double Rydberg functions [17] for every C, N and O atoms in the base part. DZ [17,18] sets were used for the other atoms. In addition, double Rydberg d-functions [17] were placed on the center of the base ring.

In the SAC–CI calculation, 1s orbitals of the C, O and N atoms were treated as the frozen orbitals. All singles and selected doubles were included in the SAC/SAC–CI wave functions. Perturbation selection [19] was carried out with the 'LevelTwo' sets of thresholds: 5×10^{-5} and 5×10^{-6} hartree for the SAC and SAC–CI calculations, respectively. As described in the previous section, the rotational strength depends on the MTDM, which is gauge dependent. Since the states of our interests took place on base ring, the origin of magnetic dipoles moments was set to center of the base ring.

4. Results and discussion

The low-lying absorption peaks of uridine originate mostly from the base part of uridine, uracil. Therefore, we start with explaining the SAC–CI result for the excited states of uracil. In Table 1, the calculated excited states and some one-electron properties are summarized. The present SAC–CI result consistently explains the observed absorption peaks. In the experimental absorption spectrum of uracil [20], there are two peaks at 5.08 and 6.63 eV and two shoulders at around 6.0 and 6.4 eV. Based on the present calculation, the two peaks are assigned to the $2^1A'$ (5.09 eV) and $4^1A'$ (6.81 eV) states, respectively. The $2^1A'$ state is π–$\pi*$ transition having strong oscillator strength (0.16 a.u.). Calculated second moment is 106.93 (bohr)^2, which indicates the valence character. As for the $4^1A'$ state, the main configuration is mixed π–$\pi*$ and π–Rydberg excitations. Second moment is calculated to be 133.77 (bohr)^2,

Table 1
Singlet excited states of Uracil calculated by the SAC–CI method

State	SAC–CI				Experimental[a]
	Nature	E_{ex}[b]	Second[c]	Oscillator[d]	E_{ex}[b]
X^{1}A′	Ground state		107.67		
1^{1}A″	n–$\pi*$	4.69	103.04	0.0002	
2^{1}A′	π–$\pi*$	5.09	106.93	0.1628	5.08
2^{1}A″	π–Ryd, π–(Ryd, $\sigma*$)	5.86	144.10	0.0072	6.0(sh)
3^{1}A″	n–$\pi*$	6.22	103.04	0.0000	
3^{1}A′	π–π	6.40	108.92	0.0554	6.4(sh)
4^{1}A′	π–$\pi*$, π–Ryd	6.81	133.77	0.1438	6.63
4^{1}A″	π–Ryd, π–$\sigma*$	6.88	169.55	0.0146	
5^{1}A″	π–Ryd, π–$\sigma*$	6.90	153.10	0.0050	
5^{1}A′	π–(Ryd, $\pi*$), π–$\pi*$	6.91	193.55	0.0712	

[a] Ref. [22].
[b] Excitation energy in eV.
[c] Electronic second moment in bohr^2.
[d] Oscillator strength in bohr.

which is relatively larger than the 2^{1}A′ state. The two shoulders are assigned to the 2^{1}A″ (5.86 eV) and 3^{1}A′ (6.40 eV) states. The 2^{1}A″ state is characterized as π–Rydberg excitation to which π–$\sigma*$ excitation strongly mixes. The 3^{1}A′ state has valence π–$\pi*$ character as indicated by the second moment (108.92 (bohr)^2). The present result also indicates the n–$\pi*$ transitions at 4.69 and 6.22 eV, which is difficult to identify only with the experimental absorption spectrum due to their very small intensity.

Next, we studied CD and absorption spectra of uridine. In Fig. 2, the SAC–CI theoretical spectra (blue lines) are compared with the experimental ones (red lines). Excitation energy, second moment, oscillator strength, and rotational strength are summarized in Table 2.

As seen in Fig. 2a, the experimental spectrum shows two peaks at 260 (4.77 eV) and 205 nm (6.05 eV) [5]. In addition, a tail of an absorption peak starts at 190 nm (6.5 eV). These peaks are so broad that several absorptions would contribute to the two peaks. On the other hand, the experimental CD spectrum shown in Fig. 2b has four peaks at around 267 nm (peak I, 4.64 eV), 240 nm (peak II, 5.17 eV), 210 nm (peak III, 5.90 eV), and 190 nm (peak IV, 6.53 eV) [5]. Comparing these two experiments, there are two different features. First, the peak positions observed in the absorption spectrum shift around 0.13–0.15 eV in the CD spectrum. The peak at 4.77 and 6.05 eV in the absorption spectrum shift to 4.64 and 5.90 eV in the CD spectrum, respectively. Second, the new peaks II and IV appear in the CD spectrum. It is difficult to find the absorption peaks corresponding to the peaks II and IV seen in the CD spectrum.

The 2^{1}A state is assigned to the peak at 260 nm (4.77 eV) in the absorption spectrum. Calculated excitation energy is 4.64 eV, and oscillator strength is largest in the calculated excited state. The 2^{1}A state is characterized as valence π–$\pi*$ excited state. The second moment (171 bohr^2) is very close to that of the ground state.

The 3^{1}A state is at 4.74 eV, which is very close to the 2^{1}A state. The oscillator strength is very small (0.0001 bohr),
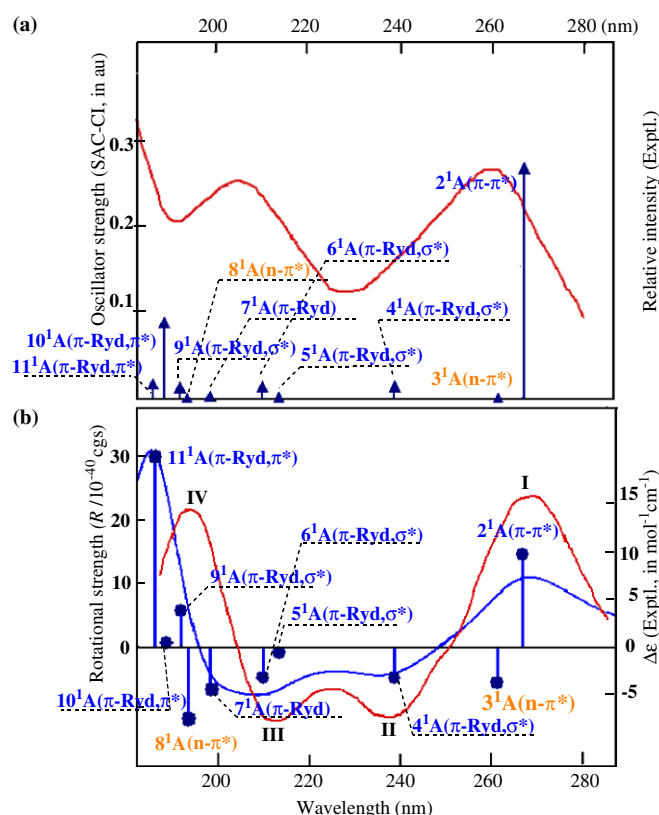

Fig. 2. (a) Absorption and (b) CD spectra of uridine. SAC–CI spectra (shown by blue lines) compared with the experimental spectrum [5] (shown by red lines). (For interpretation of the references in colour in this figure legend, the reader is referred to the web version of this article.)

because of the n–$\pi*$ character. However, the rotational strength is calculated to be -6.42×10^{-40} cgs, which is comparable to that of the 2^{1}A state (17.00×10^{-40} cgs). It should be noted that the rotational strength of n–$\pi*$ transition can be large, even though the oscillator strength is very small. Since the rotational strengths of the 2^{1}A and 3^{1}A states are positive and negative, respectively, the two peaks cancel each other. As a result, the residual positive contribution

Table 2
Singlet excited states of uridine calculated by the SAC–CI method

State	Nature	SAC–CI				Experimental[a]		
		E_{ex}[b]	Second[c]	Oscillator[d]	Rotational strength[e]	$E_{ex}(abs)$[f]		$E_{ex}(CD)$[g]
X^{1}A	Ground state	–	−170	–	–	–		
2^{1}A	π–π*	4.64	−171	0.2875	17.00	4.77	}	4.64(+)
3^{1}A	n–π*	4.74	−169	0.0001	−6.42			
4^{1}A	π–(Ryd,σ*)	5.19	−228	0.0153	−5.42			5.17(−)
5^{1}A	π–(Ryd,σ*)	5.80	−241	0.0008	−1.00		}	
6^{1}A	π–(Ryd,σ*)	5.90	−266	0.0144	−5.46	6.05		5.90(−)
7^{1}A	π–Ryd	6.24	−282	0.0026	−7.83			
8^{1}A	n–π*	6.40	−167	0.0004	−13.12		}	
9^{1}A	π–(Ryd,σ*)	6.45	−276	0.0132	6.84			
10^{1}A	π–(Ryd,π*)	6.57	−240	0.0944	0.75	>6.5		6.53(+)
11^{1}A	π–(Ryd,π*)	6.66	−261	0.0182	34.57			

[a] Reference [5].
[b] Excitation energy in eV.
[c] Electronic second moment in bohr^2.
[d] Oscillator strength in bohr.
[e] Rotational strength in 10^{-40} cgs unit.
[f] Peak maximum in the absorption spectrum [5].
[g] Peak maximum in the CD spectrum [5]. Sign in the parenthesis denotes the sign of the rotational strength.

from the 2^{1}A state is observed as the positive peak I in the CD spectrum. This cancellation would be a reason why the peak in the absorption spectrum shifts to the lower-energy region in the CD spectrum.

The 4^{1}A state is calculated at 5.19 eV. The nature of the state is one-electron excitation from π orbital to mixed σ* and Rydberg orbital. This transition could be assigned to the shoulder in the high-energy side of the 260 nm peak (4.77 eV) in the absorption spectrum. In the CD spectrum, the 4^{1}A state is assigned to peak II, because of its negative rotational strength (-5.42×10^{-40} cgs).

Three excited states, 5–7^{1}A states, were calculated in the energy range from 5.8 to 6.2 eV. The 5^{1}A and 6^{1}A states are characterized to be excitations from π orbital to mixed σ* and Rydberg orbital. The 7^{1}A state is π to Rydberg transition. These three states would compose the peak centered at 205 nm (6.05 eV) in the absorption spectrum. Due to the negative rotational strength, these states are attributed to the peak III in the CD spectrum.

In the energy region higher than 6.4 eV, total four states, 8–11^{1}A states, were calculated. In the absorption spectrum,

these four states would possibly contribute to the broad absorption in this part of the spectrum. The peak IV in the CD spectrum would be ascribed to the positive rotational strength from 9^{1}A and 11^{1}A states. The 8^{1}A state is characterized to be n–π* state. As in the 2^{1}A state, rotational strength is very large, although the oscillator strength is very small. The 10^{1}A and 11^{1}A states are excitation from π orbital to mixed Rydberg and π* orbitals.

To understand the origin of the rotational strength, we performed factorization analysis for the rotational strength of π–π* (2^{1}A) and n–π* (3^{1}A) transitions. This analysis is based on Eq. (3). The result is summarized in Table 3. The rotational strength is factorized into $|\vec{\mu}|$ (magnitude of the ETDM), $|\vec{m}|$ (magnitude of the MTDM), and $\cos\theta$ (cosine of the angle between these two transition moments). This analysis can classify the origin in terms of the magnitudes of the two transition moments and the structural distortion. The latter determines the selection rule of the optical activity. To illustrate this factorization analysis, we also included the result for π–π* (2^{1}A′) transition of uracil. Since uracil has C$_s$ symmetry, the transition

Table 3
Factorization analysis for the rotational strength of the CD spectra of uridine and uracil

	State	$|\vec{\mu}|$	$|\vec{m}|$	θ[a]	$\cos\theta$	$\vec{m}\cdot\vec{\mu}$	R[b]
Uridine	2^{1}A (π–π*)	1.59	1.41	89.07	0.02	0.04	17.00
	3^{1}A (n–π*)	0.04	0.64	127.08	−0.60	−0.01	−6.42
Uracil	2^{1}A′ (π–π*)	1.15	1.26	90.00	0.00	0.00	0.00

[a] Angle between the transition dipole and transition magnetic dipole moments.
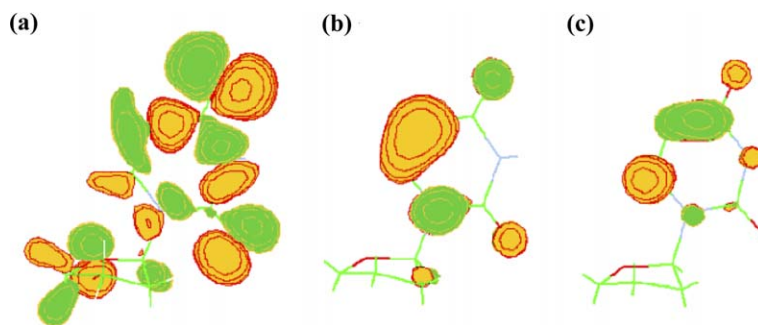[b] Rotational strength in 10^{-40} cgs unit.

Fig. 3. (a) $n + \sigma$, (b) π, and (c) $\pi*$ orbitals of uridine. Molden [21] was used for graphical representation of molecular orbitals.

dipole moment is orthogonal to the transition magnetic dipole moments. This orthogonality zeros the rotational strength, even though the magnitudes of the two transition moments are very large.

In the case of the π–$\pi*$ transition of uridine, $|\vec{\mu}|$ and $|\vec{m}|$ are similar to those of uracil. The angle between $\vec{\mu}$ and $\vec{m}$ is almost orthogonal (89.07°). However, this small deviation from 90° becomes important in the rotational strength. In the n–$\pi*$ transition, the ETDM is very small (0.04 bohr), as indicated by the weak oscillator strength in the absorption spectrum. The angle θ is 127.08°, which is significant deviation from 90°. This enlarges the rotational strength through $\cos\theta$. The reason of the deviation is in the character of the n-orbital. As shown in Fig. 3, the π and $\pi*$orbitals of uridine is localized in the uracil moiety. On the other hand, the n-orbital has small amount of amplitude in the sugar part of uridine. We summarize that the rotational strength of the π–$\pi*$ transition originates from the magnitude of the transition dipole moments, and that of the n–$\pi*$ transitions from the angle between $\vec{\mu}$ and $\vec{m}$.

Acknowledgements

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