Time Dependent Density Functional Theory Calculations of 4-Thiothymidine-containing DNA Dinucleotides

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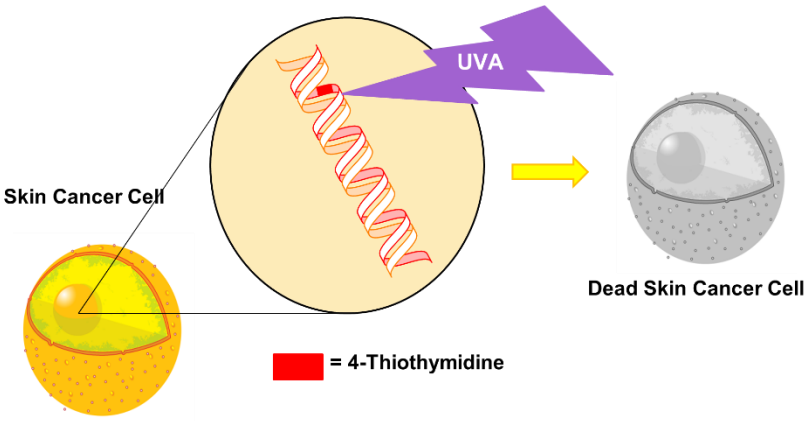
1. Introduction

The substitution of an oxygen atom in an exocyclic carbonyl group of the nucleobases by a sulfur atom in a nucleic acid base generates a thiobase. This substitution causes a redshift in the absorption spectrum of the thiobase with respect to the canonical nucleobase, moving the strongly allowed absorption band from the UVC to the UVA region of the electromagnetic spectrum. [1] Due to this redshift and the efficient population of the triplet state, 4-thiothymidine (4tThd) can be selectively excited without exciting canonical DNA, making it a powerful UVA photosensitizer. The synergistic toxicity of 4tThd and UVA radiation allows for the enhanced killing of skin cancer cells. As a result, 4tThd has been proposed for use in conjunction with UVA radiation as potential photodynamic therapy agent, due to its photochemical properties and to a diminished cytotoxicity.[2]

Studies of the monomer 4tThd have been performed to explore the prospective use of 4tThd in photochemotherapeutic application with reduced phototoxic side effects. One study of 4tThd in aqueous solution proposed the main kinetic mechanism to consist of intersystem crossing from the S_2 state to the triplet manifold.[3] Vertical excitation energies were calculated using the optimized ground state of 4tThd in water and vacuum. These were found to be in good agreement with the values previously reported.[3] After studying the monomer, the next step is to understand what happens when 4tThd interacts with the DNA bases. Therefore, ground state optimizations and vertical excitation energies calculations were performed for a series of 4tThd-containing dinucleotides. These vertical excitation energy calculations predict the order electronic states and likely kinetic mechanisms when 4tThd is incorporated into DNA, which will greatly assist in the interpretation of planned time-resolved experiments.



Scheme 1. Photochemotherapy with 4tThd.

Computational Methods

All calculations were performed using the Gaussian 16 suite of programs and the density functional theory (DFT) with the either the M052X or PBE0 functional and either the 6-31G(d) or 6-31+G(d) standard basis set. In detail, the geometries of the ground state of 4tThd, T-4tThd, 4tThd-T, A-4tThd, 4tThd-A, C-4tThd, 4tThd-C, G-4tThd, and 4tThd-G were optimized in water and vacuum using the M052X/6-31G(d) level of theory. In the calculations for the dinucleotides, a Na+ counter ion was added. The geometries of the ground state of 4tThd was also optimized in vacuum at the PBE0/6-31G(d) level of theory. Vertical excitation energy calculations of the optimized ground states in water and vacuum were performed using the time dependent density functional theory (TD-DFT) at both the M052X/6-31G(d) and PBE0/6-31+G(d) level of theory. The dinucleotides were labeled first with the base in the 5' position and then with the base in the 3' position.

3. Results and Discussion

Table 1: Vertical excitation energies for 4tThd in water calculated using the M052X/6-31G(d) level of theory from ground state M052X/6-31G(d) optimization

State	Transitions	% Contribution	Primary Character	eV
S_1	H-1→L+0	100	$n\pi*$	3.3794 (0)
S_2	H-0→L+0	100	$\pi\pi*$	4.3005 (0.5816)
S_3	H-2→L+0	97.3	$CT, \pi\pi*$	5.2711 (0.04)
	H-0→L+1	2.7		
S_4	H-6→L+1	2.3	$\pi\pi*$	6.1066 (0.1016)
	H-2→L+0	3.5		
	H-0→L+1	94.3		
S_5	H-6→L+0	13.7	$n\pi*$	6.2259 (0.0162)
	H-6→L+1	14.7		
	H-3→L+0	41.6		
	H-3→L+1	2.5		
	H-1→L+1	20.1		
	H-0→L+1	7.4		
T_1	H-0→L+0	97.8	$\pi\pi*$	2.6582
	H-0←L+0	2.2		
T_2	H-1→L+0	97.9	$n\pi*$	3.0991
	H-1→L+1	2.1		
T_3	H-2→L+0	89.1	$\pi\pi*$	3.8784
	H-0→L+1	10.9		
T_4	H-2→L+0	11.3	$\pi\pi*$	4.8668
	H-2→L+1	35.0		
	H-0→L+1	53.7		
T_5	H-11→L+0	2.9	$n\pi*$	5.9286
	H-6→L+0	7.7		
	H-6→L+1	38.6		

	H-6→L+5	3.0
	H-3→L+0	21.9
	H-3→L+1	7.2
	H-1→L+1	15.8
	H-0→L+1	2.8

Table 2: Vertical excitation energies for 4tThd in vacuum calculated using the M052X/6-31G(d) level of theory from ground state M052X/6-31G(d) optimizations

State	Transitions	% Contribution	Primary Character	eV
S_1	H-1→L+0	97.8	$n\pi*$	3.1275 (0)
	H-1→L+1	2.2		
S_2	H-2→L+0	2.2	$\pi\pi*$	4.4602 (0.5037)
	H-0→L+0	97.8		
S_3	H-2→L+0	95.1	$CT, \pi\pi*$	5.319 (0.0263)
	H-0→L+0	2.5		
	H-0→L+1	2.4		
S_4	H-5→L+1	2.7	$CT, \pi\pi*$	6.0209 (0.0582)
	H-3→L+1	2.6		
	H-2→L+0	2.5		
	H-1→L+1	12.3		
	H-0→L+1	79.9		
S_5	H-5→L+1	4.0	$n\pi*$	6.0758 (0.0222)
	H-3→L+1	2.7		
	H-2→L+1	3.7		
	H-1→L+1	69.8		
	H-0→L+1	19.8		
T_1	H-0→L+0	97.2	$\pi\pi*$	2.5302
	H-0←L+0	2.8		
T_2	H-1→L+0	97.3	$n\pi*$	2.7995
	H-1→L+1	2.7		
T_3	H-8→L+0	2.2	$\pi\pi*$	3.8718
	H-2→L+0	84.2		
	H-0→L+1	13.6		
T_4	H-2→L+0	13.6	$\pi\pi*$	4.9763
	H-2→L+1	39.7		
	H-0→L+1	46.8		
T_5	H-5→L+1	14.1	$n\pi*$	5.8197
	H-4→L+1	8.9		
	H-3→L+1	14.1		
	H-2→L+1	21.0		
	H-1→L+1	20.8		
	H-0→L+1	21.2		

Table 3: Vertical excitation energies for 4tThd in water calculated using the M052X/6-31G(d) level of theory from ground state PBE0/6-31G(d) optimizations

State	Transitions	% Contribution	Primary Character	eV
S_1	H-1→L+0	100	$n\pi*$	3.3805 (0)
S_2	H-0→L+0	100	$\pi\pi*$	4.2781 (0.5816)
S_3	H-2→L+0	97.3	$CT, \pi\pi*$	5.2587 (0.0386)
	H-0→L+1	2.7		
S_4	H-2→L+0	3.5	$\pi\pi*$	6.0786 (0.1035)
	H-0→L+1	96.5		
S_5	H-6→L+0	15.3	$n\pi*$	6.2021 (0.0154)
	H-6→L+1	7.1		
	H-4→L+0	2.5		
	H-3→L+0	62.4		
	H-1→L+1	7.1		
	H-0→L+1	5.4		
T_1	H-0→L+0	97.8	$\pi\pi*$	2.6391
	H-0←L+0	2.2		
T_2	H-1→L+0	97.8	$n\pi*$	3.1026
	H-1→L+1	2.2		
T_3	H-2→L+0	88.9	$\pi\pi*$	3.8663
	H-0→L+1	11.1		
T_4	H-2→L+0	11.4	$\pi\pi*$	4.8455
	H-2→L+1	34.7		
	H-0→L+1	53.9		
T_5	H-12→L+0	4.4	$n\pi*$	5.912
	H-11→L+0	2.8		
	H-6→L+0	6.2		
	H-6→L+1	22.6		
	H-3→L+0	36.4		
	H-3→L+1	3.5		
	H-2→L+1	5.3		
	H-1→L+1	9.3		
	H-0→L+1	9.4		

Table 4: Vertical excitation energies for 4tThd in vacuum calculated using the M052X/6-31G(d) level of theory from ground state PBE0/6-31G(d) optimizations

State	Transitions	% Contribution	Primary Character	eV
S_1	H-1→L+0	97.7	$n\pi*$	3.1198 (0)
	H-1→L+1	2.3		
S_2	H-2→L+0	2.1	$\pi\pi*$	4.4337 (0.5042)
	H-0→L+0	97.9		
S_3	H-2→L+0	95.2	$CT, \pi\pi*$	5.3014 (0.0248)

	H-0→L+0	2.4		
	H-0→L+1	2.4		
S_4	H-5→L+1	2.2	$CT, \pi\pi*$	5.9928 (0.0646)
	H-2→L+0	2.7		
	H-1→L+1	9.8		
	H-0→L+1	85.3		
S_5	H-5→L+1	4.7	$n\pi*$	6.0487 (0.0165)
	H-4→L+1	2.3		
	H-3→L+1	4.0		
	H-1→L+1	73.6		
	H-0→L+1	15.4		
T_1	H-0→L+0	97.1	$\pi\pi*$	2.5021
	H-0←L+0	2.9		
T_2	H-1→L+0	97.3	$n\pi*$	2.7936
	H-1→L+1	2.7		
T_3	H-8→L+0	2.2	$\pi\pi*$	3.8514
	H-2→L+0	84.0		
	H-0→L+1	13.8		
T_4	H-2→L+0	13.6	$\pi\pi*$	4.9556
	H-2→L+1	39.2		
	H-0→L+1	47.2		
T_5	H-5→L+1	7.1	$\pi\pi*$	5.7974
	H-4→L+1	3.0		
	H-3→L+1	7.2		
	H-2→L+0	2.8		
	H-2→L+1	37.3		
	H-1→L+1	9.1		
	H-0→L+1	33.6		

Table 5: Vertical excitation energies for 4tThd in water calculated using the PBE0/6-31+G(d) level of theory from ground state M052X/6-31G(d) optimizations

State	Transitions	% Contribution	Primary Character	eV
S_1	H-1→L+0	15.5	$n\pi*$	3.2281 (0.0001)
	H-0→L+0	84.5		
S_2	H-1→L+0	84.4	$\pi\pi*$	4.0683 (0.5184)
	H-0→L+0	15.6		
S_3	H-2→L+0	100	$CT, \pi\pi*$	4.8597 (0.0515)
S_4	H-3→L+0	100	$n\pi*$	5.0583 (0.0042)
S_5	H-1→L+1	10.9	$n\pi*$	5.3456 (0.0004)
	H-0→L+1	89.1		
T_1	H-1→L+0	83.9	$\pi\pi*$	2.585
	H-0→L+0	16.1		

T_2	H-1→L+0	16.2	$n\pi*$	2.9542
	H-0→L+0	83.8		
T_3	H-3→L+0	4.8	$CT, \pi\pi*$	3.6546
	H-2→L+0	88.5		
	H-1→L+1	6.7		
T_4	H-2→L+0	11.9	$\pi\pi*$	4.4519
	H-2→L+1	19.4		
	H-1→L+1	57.5		
	H-0→L+1	11.2		
T_5	H-3→L+0	93.4	$n\pi*$	4.942
	H-2→L+0	2.6		
	H-1→L+1	4.0		

Table 6: Vertical excitation energies for 4tThd in vacuum calculated using the PBE0/6-31+G(d) level of theory from ground state M052X/6-31G(d) optimizations

State	**Transitions**	**% Contribution**	**Primary Character**	**eV**
S_1	H-0→L+0	100	$n\pi*$	3.0236 (0.0001)
S_2	H-2→L+0	3.8	$\pi\pi*$	4.1996 (0.4252)
	H-1→L+0	96.2		
S_3	H-2→L+0	96.7	$CT, \pi\pi*$	4.8578 (0.0278)
	H-1→L+0	3.3		
S_4	H-0→L+2	17.5	$n\pi*$	5.1282 (0.0001)
	H-0→L+3	82.5		
S_5	H-0→L+1	97.0	$n\pi*$	5.2841 (0.0022)
	H-0→L+4	3.0		
T_1	H-1→L+0	97.7	$\pi\pi*$	2.449
	H-1←L+0	2.3		
T_2	H-0→L+0	100	$n\pi*$	2.6947
T_3	H-8→L+0	2.2	$CT, \pi\pi*$	3.6194
	H-2→L+0	88.8		
	H-1→L+3	9.0		
T_4	H-2→L+0	13.1	$\pi\pi*$	4.5508
	H-2→L+2	3.0		
	H-2→L+3	18.8		
	H-1→L+2	9.4		
	H-1→L+3	55.8		
T_5	H-0→L+2	17.1	$n\pi*$	5.0543
	H-0→L+3	82.9		

Table 7: Vertical excitation energies for 4tThd in water calculated using the PBE0/6-31+G(d) level of theory from ground state PBE0/6-31G(d) optimizations

State	**Transitions**	**% Contribution**	**Primary Character**	**eV**

S_1	H-1→L+0 H-0→L+0	62.9 37.1	$n\pi*$	3.2241 (0.0001)
S_2	H-1→L+0 H-0→L+0	37.0 63.0	$\pi\pi*$	4.0489 (0.5203)
S_3	H-2→L+0	100	$CT, \pi\pi*$	4.8474 (0.0481)
S_4	H-3→L+0	100	$n\pi*$	5.0351 (0.004)
S_5	H-1→L+1 H-0→L+1	55.5 44.5	$n\pi*$	5.3246 (0.0005)
T_1	H-1→L+0 H-0→L+0	36.4 63.6	$\pi\pi*$	2.5675
T_2	H-1→L+0 H-0→L+0	63.7 36.3	$n\pi*$	2.953
T_3	H-3→L+0 H-2→L+0 H-1→L+1 H-0→L+1	4.7 87.3 3.0 5.1	$\pi\pi*$	3.644
T_4	H-2→L+0 H-2→L+1 H-1→L+1 H-0→L+1	12.0 19.1 24.9 44.0	$\pi\pi*$	4.4289
T_5	H-3→L+0 H-2→L+0 H-0→L+1	93.1 2.6 4.3	$n\pi*$	4.9184

Table 8: Vertical excitation energies for 4tThd in vacuum calculated using the PBE0/6-31+G(d) level of theory from ground state PBE0/6-31G(d) optimizations

State	**Transitions**	**% Contribution**	**Primary Character**	**eV**
S_1	H-0→L+0	100	$n\pi*$	3.0112 (0.0001)
S_2	H-2→L+0 H-1→L+0	3.4 96.6	$\pi\pi*$	4.1769 (0.427)
S_3	H-2→L+0 H-1→L+0	97.0 3.0	$CT, \pi\pi*$	4.84 (0.0246)
S_4	H-0→L+2 H-0→L+3	18.1 81.9	$n\pi*$	5.1027 (0.0001)
S_5	H-3→L+0 H-1→L+3	97.9 2.1	$n\pi*$	5.2695 (0.0006)
T_1	H-1→L+0 H-1←L+0	97.7 2.3	$\pi\pi*$	2.4232
T_2	H-0→L+0	100	$n\pi*$	2.6847
T_3	H-8→L+0 H-2→L+0 H-1→L+3	2.2 88.7 9.1	$\pi\pi*$	3.6017

T_4	H-2→L+0	13.3	$\pi\pi*$	4.528
	H-2→L+2	3.1		
	H-2→L+3	18.3		
	H-1→L+2	9.8		
	H-1→L+3	55.6		
T_5	H-0→L+2	17.7	$n\pi*$	5.0292
	H-0→L+3	82.3		

Table 9: Vertical excitation energies for T-4tThd in water using the M052X/6-31G(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-2→L+0	6.9	$n\pi*$	3.3143 (0.0001)
	H-1→L+0	93.1		
S_2	H-0→L+0	100.0	$\pi\pi*$	4.2672 (0.4311)
S_3	H-2→L+0	92.6	$CT, \pi\pi*$	5.0008 (0.0059)
	H-1→L+0	7.4		
S_4	H-13→L+1	5.1	$n\pi*$	5.1645 (0.0001)
	H-10→L+1	3.5		
	H-7→L+1	53.6		
	H-7→L+4	5.6		
	H-6→L+1	5.0		
	H-5→L+1	13.0		
	H-4→L+1	14.2		
S_5	H-3→L+0	97.1	$CT, \pi\pi*$	5.2643 (0.0324)
	H-0→L+2	2.9		
T_1	H-0→L+0	97.7	$\pi\pi*$	2.6093
	H-0←L+0	2.3		
T_2	H-2→L+0	7.2	$n\pi*$	3.0268
	H-1→L+0	92.8		
T_3	H-2→L+1	90.5	$\pi\pi*$	3.5182
	H-1→L+1	9.5		
T_4	H-3→L+0	88.2	$\pi\pi*$	3.8537
	H-0→L+2	11.8		
T_5	H-3→L+0	12.7	$\pi\pi*$	4.8362
	H-3→L+2	33.0		
	H-0→L+1	3.7		
	H-0→L+2	50.7		

Table 10: Vertical excitation energies for T-4tThd in vacuum using the M052X/6-31G(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-1→L+0	13.3	$n\pi*$	3.0519 (0.0001)

	H-0→L+0	86.7		
S2	H-1→L+0	86.4	$\pi\pi*$	4.3993 (0.3529)
	H-0→L+0	13.6		
S3	H-8→L+1	11.9	$n\pi*$	4.9952 (0.0001)
	H-4→L+1	79.4		
	H-4→L+6	8.7		
S4	H-2→L+0	100	$CT, \pi\pi*$	5.0589 (0.0062)
S5	H-3→L+0	97.8	$CT, \pi\pi*$	5.283 (0.0213)
	H-1→L+4	2.2		
T1	H-1→L+0	82.1	$\pi\pi*$	2.5014
	H-0→L+0	15.4		
	H-1←L+0	2.5		
T2	H-1→L+0	15.3	$n\pi*$	2.7215
	H-0→L+0	82.4		
	H-0→L+4	2.3		
T3	H-2→L+1	100	$\pi\pi*$	3.5204
T4	H-3→L+0	87.5	$\pi\pi*$	3.8513
	H-1→L+4	12.5		
T5	H-11→L+1	2.4	$n\pi*$	4.562
	H-8→L+1	12.7		
	H-4→L+1	75.4		
	H-4→L+6	9.5		

Table 11: Vertical excitation energies for 4tThd-T in water using the M052X/6-31G(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S1	H-1→L+0	100	$n\pi*$	3.3587 (0.0002)
S2	H-0→L+0	100	$\pi\pi*$	4.258 (0.4839)
S3	H-12→L+1	12.6	$n\pi*$	5.1379 (0.0074)
	H-8→L+1	54.4		
	H-8→L+4	6.2		
	H-7→L+1	8.9		
	H-4→L+1	6.6		
	H-3→L+0	5.7		
	H-2→L+0	2.6		
	H-0→L+1	3.0		
S4	H-8→L+1	4.0	$CT, \pi\pi*$	5.1638 (0.0502)
	H-3→L+0	56.2		
	H-2→L+0	39.8		
S5	H-3→L+0	36.8	$CT, \pi\pi*$	5.2224 (0.0276)
	H-2→L+0	45.7		
	H-2→L+1	17.5		

T_1	H-0→L+0 H-0←L+0	97.9 2.1	$\pi\pi*$	2.6773
T_2	H-1→L+0	100	$n\pi*$	3.0889
T_3	H-2→L+1	100	$\pi\pi*$	3.5183
T_4	H-3→L+0 H-0→L+3	91.4 8.6	$\pi\pi*$	3.8303
T_5	H-12→L+1 H-8→L+1 H-8→L+4 H-7→L+1 H-4→L+1	15.1 60.0 8.0 9.9 7.0	$n\pi*$	4.7174

Table 12: Vertical excitation energies for 4tThd-T in vacuum using the M052X/6-31G(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-1→L+0 H-0→L+0	28.3 71.7	$n\pi*$	3.0905 (0.0002)
S_2	H-3→L+0 H-1→L+0 H-0→L+0	2.4 70.0 27.5	$\pi\pi*$	4.3814 (0.4091)
S_3	H-11→L+1 H-6→L+1 H-6→L+6 H-4→L+0 H-4→L+1 H-4→L+6	6.8 21.8 2.8 2.5 59.5 6.7	$n\pi*$	4.9864 (0.0006)
S_4	H-2→L+0 H-2→L+1 H-1→L+1 H-0→L+1	34.0 59.8 3.2 3.0	$CT, \pi\pi*$	5.191 (0.0298)
S_5	H-3→L+0 H-2→L+0 H-2→L+1	36.8 45.7 17.5	$\pi\pi*$	5.3647 (0.1264)
T_1	H-1→L+0 H-0→L+0	57.7 42.3	$\pi\pi*$	2.5768
T_2	H-1→L+0 H-0→L+0	42.7 57.3	$n\pi*$	2.7847
T_3	H-2→L+0 H-2→L+1	3.9 96.1	$\pi\pi*$	3.5253
T_4	H-3→L+0 H-1→L+5 H-0→L+5	90.7 6.4 2.9	$\pi\pi*$	3.8012

T_5	H-11→L+1	7.8	$n\pi*$	4.5523
	H-6→L+1	22.2		
	H-6→L+6	3.2		
	H-4→L+1	59.0		
	H-4→L+6	7.8		

Table 13: Vertical excitation energies for T-4tThd in water using the PBE0/6-31+G(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-1→L+0	5.8	$n\pi*$	3.1817 (0.0002)
	H-0→L+0	94.2		
S_2	H-2→L+0	86.0	$CT, \pi\pi*$	4.0157 (0.028)
	H-1→L+0	14.0		
S_3	H-2→L+0	13.8	$\pi\pi*$	4.0571 (0.3526)
	H-1→L+0	79.5		
	H-0→L+0	6.7		
S_4	H-0→L+1	100	$n\pi*$	4.6763 (0.0004)
S_5	H-1→L+1	100	$CT, \pi\pi*$	4.7371 (0.0124)
T_1	H-1→L+0	91.9	$\pi\pi*$	2.5458
	H-0→L+0	8.1		
T_2	H-1→L+0	7.4	$n\pi*$	2.8965
	H-0→L+0	92.6		
T_3	H-2→L+1	96.4	$CT, \pi\pi*$	3.2898
	H-1→L+1	3.6		
T_4	H-6→L+0	21.6	$n\pi*$	3.6334
	H-5→L+0	27.0		
	H-4→L+0	5.1		
	H-3→L+0	35.8		
	H-2→L+0	2.5		
	H-1→L+2	7.9		
T_5	H-2→L+0	97.7	$\pi\pi*$	4.0118
	H-1→L+0	2.3		

Table 14: Vertical excitation energies for T-4tThd in vacuum using the PBE0/6-31+G(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-0→L+1	100	$n\pi*$	2.9545 (0.0002)
S_2	H-0→L+0	100	$n\pi*$	3.9368 (0)
S_3	H-2→L+1	100	$CT, \pi\pi*$	4.055 (0.0073)
S_4	H-1→L+0	100	$CT, \pi\pi*$	4.137 (0.0035)
S_5	H-1→L+1	100	$\pi\pi*$	4.1622 (0.2843)
T_1	H-1→L+1	97.6	$\pi\pi*$	2.4278

	H-1←L+1	2.4		
T_2	H-0→L+1	100	$n\pi*$	2.6203
T_3	H-2→L+2	100	$\pi\pi*$	3.273
T_4	H-5→L+1	3.1	$n\pi*$	3.6035
	H-4→L+1	43.3		
	H-3→L+1	45.5		
	H-1→L+6	8.1		
T_5	H-0→L+0	100	$n\pi*$	3.9368

Table 15: Vertical excitation energies for 4tThd-T in water using the PBE0/6-31+G(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-1→L+0	18.2	$n\pi*$	3.1962 (0.0005)
	H-0→L+0	81.8		
S_2	H-1→L+0	81.7	$\pi\pi*$	4.0253 (0.4038)
	H-0→L+0	18.3		
S_3	H-2→L+0	100	$CT, \pi\pi*$	4.1725 (0.0133)
S_4	H-1→L+1	44.4	$n\pi*$	4.6508 (0.006)
	H-0→L+1	55.6		
S_5	H-1→L+1	56.2	$CT, \pi\pi*$	4.7264 (0.0012)
	H-0→L+1	43.8		
T_1	H-1→L+0	74.7	$\pi\pi*$	2.5969
	H-0→L+0	25.3		
T_2	H-1→L+0	25.4	$n\pi*$	2.9314
	H-0→L+0	74.6		
T_3	H-2→L+0	3.7	$\pi\pi*$	3.2867
	H-2→L+1	96.3		
T_4	H-6→L+0	7.1	$n\pi*$	3.6049
	H-4→L+0	19.6		
	H-3→L+0	68.6		
	H-1→L+2	4.6		
T_5	H-2→L+0	96.6	$\pi\pi*$	4.1731
	H-2→L+1	3.4		

Table 16: Vertical excitation energies for 4tThd-T in vacuum using the PBE0/6-31+G(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-0→L+1	100	$n\pi*$	2.96 (0.0003)
S_2	H-0→L+0	100	$n\pi*$	4.0205 (0)
S_3	H-3→L+1	4.1	$\pi\pi*$	4.1237 (0.322)
	H-1→L+1	95.9		
S_4	H-1→L+0	100	$CT, \pi\pi*$	4.2251 (0.0013)

S_5	H-0→L+2	100	$n\pi*$	4.2682 (0.0023)
T_1	H-1→L+1	92.6	$\pi\pi*$	2.4878
	H-0→L+1	7.4		
T_2	H-1→L+1	7.3	$n\pi*$	2.6546
	H-0→L+1	92.7		
T_3	H-2→L+1	6.0	$\pi\pi*$	3.2744
	H-2→L+2	94.0		
T_4	H-3→L+1	91.6	$\pi\pi*$	3.5515
	H-2→L+1	2.6		
	H-1→L+8	5.8		
T_5	H-0→L+0	100	$n\pi*$	4.0205

Table 17: Vertical excitation energies for 4tThd-A in water using the M052X/6-31G(d) level of theory

State	**Transitions**	**% Contribution**	**Primary Character**	**eV**
S_1	H-2→L+0	100	$n\pi*$	3.3864 (0)
S_2	H-1→L+0	24.7	$\pi\pi*$	4.2731 (0.5085)
	H-0→L+0	75.3		
S_3	H-1→L+0	75.4	$CT, \pi\pi*$	4.9554 (0.0219)
	H-0→L+0	24.6		
S_4	H-4→L+0	11.6	$CT, \pi\pi*$	5.1879 (0.0483)
	H-3→L+0	88.4		
S_5	H-5→L+1	8.1	$\pi\pi*$	5.5322 (0.307)
	H-1→L+1	62.9		
	H-0→L+1	29.1		
T_1	H-1→L+1	26.1	$\pi\pi*$	2.6684
	H-0→L+0	73.9		
T_2	H-2→L+0	100	$n\pi*$	3.1116
T_3	H-1→L+0	2.6	$\pi\pi*$	3.7718
	H-1→L+1	69.3		
	H-0→L+1	28.1		
T_4	H-4→L+0	12.8	$\pi\pi*$	3.8227
	H-3→L+0	77.8		
	H-1→L+4	2.4		
	H-0→L+4	7.0		
T_5	H-1→L+0	8.3	$\pi\pi*$	4.7294
	H-1→L+3	62.4		
	H-0→L+0	2.9		
	H-0→L+3	26.4		

Table 18: Vertical excitation energies for 4tThd-A in vacuum using the M052X/6-31G(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-2→L+1	10.0	$n\pi*$	3.1183 (0.001)
	H-1→L+1	53.8		
	H-0→L+1	36.2		
S_2	H-1→L+1	36.0	$\pi\pi*$	4.378 (0.3445)
	H-0→L+1	64.0		
S_3	H-2→L+1	88.2	$CT, \pi\pi*$	4.8657 (0.0108)
	H-1→L+1	11.8		
S_4	H-5→L+1	3.7	$CT, \pi\pi*$	5.2459 (0.0176)
	H-3→L+1	96.3		
S_5	H-5→L+1	3.0	$n\pi*$	5.3338 (0.0036)
	H-5→L+3	21.4		
	H-4→L+1	2.2		
	H-4→L+3	45.4		
	H-4→L+9	3.7		
	H-3→L+1	2.4		
	H-3→L+3	3.4		
	H-2→L+3	4.2		
	H-0→L+3	14.2		
T_1	H-1→L+1	19.0	$n\pi*$	2.526
	H-0→L+1	81.0		
T_2	H-2→L+1	12.1	$n\pi*$	2.8035
	H-1→L+1	68.7		
	H-0→L+1	19.2		
T_3	H-2→L+1	4.8	$\pi\pi*$	3.741
	H-2→L+3	79.4		
	H-1→L+3	15.8		
T_4	H-5→L+1	6.3	$\pi\pi*$	3.8578
	H-3→L+1	82.4		
	H-1→L+6	2.5		
	H-0→L+6	5.6		
	H-0→L+7	3.2		
T_5	H-2→L+1	37.1	$\pi\pi*$	4.7507
	H-2→L+3	3.3		
	H-2→L+6	19.6		
	H-2→L+7	25.1		
	H-1→L+1	5.8		
	H-1→L+6	3.2		
	H-1→L+7	5.9		

Table 19: Vertical excitation energies for 4tThd-A in water using the PBE0/6-31+G(d) level of theory

State	Transitions	% Contribution	Primary Character	eV

S_1	H-1→L+0 H-0→L+0	80.1 19.9	$n\pi*$	3.221 (0.0001)
S_2	H-2→L+0 H-1→L+0 H-0→L+0	19.8 20.2 59.9	$CT, \pi\pi*$	3.9046 (0.0036)
S_3	H-2→L+0 H-0→L+0	79.4 20.6	$\pi\pi*$	4.0329 (0.4411)
S_4	H-4→L+0 H-3→L+0	10.3 89.7	$CT, \pi\pi*$	4.7394 (0.0499)
S_5	H-5→L+0 H-4→L+0 H-3→L+0	17.0 76.9 6.1	$n\pi*$	4.8332 (0.0084)
T_1	H-2→L+0 H-0→L+0	68.4 31.6	$\pi\pi*$	2.5877
T_2	H-2→L+0 H-1→L+0 H-0→L+0	2.2 79.6 18.2	$n\pi*$	2.9543
T_3	H-2→L+0 H-2→L+1 H-1→L+0 H-1→L+1 H-0→L+0 H-0→L+1	3.9 23.2 2.4 15.9 6.4 48.2	$\pi\pi*$	3.4977
T_4	H-7→L+0 H-5→L+0 H-3→L+0 H-2→L+3	2.7 16.4 76.9 4.0	$\pi\pi*$	3.5942
T_5	H-2→L+0 H-2→L+1 H-1→L+0 H-1→L+1 H-0→L+0 H-0→L+1	27.4 3.0 16.5 2.1 44.9 6.2	$\pi\pi*$	3.9281

Table 20: Vertical excitation energies for 4tThd-A in vacuum using the PBE0/6-31+G(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-1→L+1 H-0→L+1	2.37 97.6	$n\pi*$	3.0088 (0.0002)
S_2	H-2→L+1 H-1→L+1	92.1 7.9	$CT, \pi\pi*$	3.9356 (0.0017)
S_3	H-0→L+0	100	$n\pi*$	4.0208 (0.0001)
S_4	H-4→L+1	2.5	$\pi\pi*$	4.1086 (0.2692)

	H-2→L+1	7.0		
	H-1→L+1	87.3		
	H-0→L+1	3.1		
S_5	H-1→L+0	100	$CT, \pi\pi*$	4.1762 (0.002)
T_1	H-1→L+1	87.0	$\pi\pi*$	2.4462
	H-0→L+1	13.0		
T_2	H-2→L+1	2.5	$n\pi*$	2.6945
	H-1→L+1	11.4		
	H-0→L+1	86.0		
T_3	H-2→L+1	11.4	$\pi\pi*$	3.4658
	H-2→L+2	60.0		
	H-2→L+3	21.5		
	H-2→L+4	3.7		
	H-1→L+2	3.3		
T_4	H-5→L+1	5.5	$\pi\pi*$	3.6029
	H-4→L+1	67.1		
	H-3→L+1	20.8		
	H-1→L+8	6.6		
T_5	H-2→L+1	88.4	$\pi\pi*$	3.9512
	H-2→L+2	6.2		
	H-2→L+3	2.2		
	H-1→L+1	3.2		

Table 21: Vertical excitation energies for A-4tThd in water using the M052X/6-31G(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-2→L+0	91.8	$n\pi*$	3.3186 (0.0001)
	H-1→L+0	8.2		
S_2	H-1→L+0	25.3	$\pi\pi*$	4.1834 (0.3554)
	H-0→L+0	74.7		
S_3	H-2→L+0	9.6	$CT, \pi\pi*$	4.5941 (0.0621)
	H-1→L+0	65.4		
	H-0→L+0	25.1		
S_4	H-5→L+0	3.8	$CT, \pi\pi*$	5.2546 (0.024)
	H-4→L+0	89.4		
	H-3→L+0	4.4		
	H-1→L+0	2.4		
S_5	H-5→L+1	4.8	$\pi\pi*$	5.4699 (0.3071)
	H-2→L+1	2.2		
	H-1→L+1	13.4		
	H-0→L+1	79.6		
T_1	H-1→L+0	51.3	$\pi\pi*$	2.597

	H-0→L+0	48.7		
T2	H-2→L+0	91.4	$n\pi*$	3.0351
	H-1→L+0	8.6		
T3	H-2→L+1	4.9	$\pi\pi*$	3.7643
	H-1→L+1	29.1		
	H-0→L+1	66.1		
T4	H-5→L+0	4.2	$\pi\pi*$	3.8385
	H-4→L+0	84.0		
	H-3→L+0	3.1		
	H-1→L+2	5.0		
	H-0→L+2	3.8		
T5	H-2→L+0	7.4	$\pi\pi*$	4.5348
	H-1→L+0	38.8		
	H-0→L+0	49.2		
	H-0→L+4	4.6		

Table 22: Vertical excitation energies for A-4tThd in vacuum using the M052X/6-31G(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S1	H-2→L+1	100	$n\pi*$	3.1491 (0.0001)
S2	H-1→L+1	40.8	$\pi\pi*$	4.3351 (0.3099)
	H-0→L+1	59.2		
S3	H-1→L+1	59.6	$CT, \pi\pi*$	4.6518 (0.0221)
	H-0→L+1	40.4		
S4	H-5→L+1	61.7	$n\pi*$	5.2728 (0.0087)
	H-4→L+1	9.0		
	H-3→L+1	29.3		
S5	H-5→L+3	12.9	$n\pi*$	5.3344 (0)
	H-4→L+3	60.8		
	H-4→L+9	4.1		
	H-1→L+0	12.9		
	H-0→L+0	3.8		
	H-0→L+3	5.6		
T1	H-1→L+1	60.3	$\pi\pi*$	2.516
	H-0→L+1	39.7		
T2	H-2→L+1	97.6	$n\pi*$	2.831
	H-2→L+6	2.4		
T3	H-1→L+3	37.2	$\pi\pi*$	3.7578
	H-0→L+3	62.8		
T4	H-5→L+1	55.4	$n\pi*$	3.8249
	H-4→L+1	7.6		
	H-3→L+1	22.4		
	H-1→L+6	7.8		

	H-0→L+1	2.6		
	H-0→L+6	4.3		
T_5	H-1→L+1	36.0	$\pi\pi*$	4.639
	H-1→L+7	2.7		
	H-0→L+1	55.6		
	H-0→L+6	3.5		
	H-0→L+7	2.2		

Table 23: Vertical excitation energies for A-4tThd in water using the PBE0/6-31+G(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-2→L+0	2.1	$n\pi*$	3.1726 (0.0002)
	H-1→L+0	88.3		
	H-0→L+0	9.6		
S_2	H-2→L+0	11.7	$CT, \pi\pi*$	3.6431 (0.032)
	H-1→L+0	6.0		
	H-0→L+0	82.3		
S_3	H-2→L+0	86.9	$\pi\pi*$	4.0451 (0.3167)
	H-1→L+0	5.6		
	H-0→L+0	7.6		
S_4	H-5→L+0	2.6	$n\pi*$	4.7239 (0.0115)
	H-4→L+0	11.7		
	H-3→L+0	85.7		
S_5	H-5→L+0	5.5	$n\pi*$	4.7805 (0.006)
	H-4→L+0	86.9		
	H-3→L+0	7.7		
T_1	H-2→L+0	47.5	$\pi\pi*$	2.5222
	H-1→L+0	8.2		
	H-0→L+0	44.3		
T_2	H-2→L+0	2.1	$n\pi*$	2.8945
	H-1→L+0	91.2		
	H-0→L+0	6.8		
T_3	H-2→L+0	2.4	$\pi\pi*$	3.5059
	H-2→L+1	27.6		
	H-0→L+0	2.2		
	H-0→L+1	67.8		
T_4	H-6→L+0	9.8	$\pi\pi*$	3.5898
	H-5→L+0	50.6		
	H-2→L+0	17.0		
	H-2→L+2	3.3		
	H-0→L+0	14.3		
	H-0→L+1	2.7		
	H-0→L+2	2.3		

T_5	H-6→L+0	3.8	$\pi\pi*$	3.7333
	H-5→L+0	23.6		
	H-2→L+0	35.0		
	H-0→L+0	37.6		

Table 24: Vertical excitation energies for A-4tThd in vacuum using the PBE0/6-31+G(d) level of theory

State	**Transitions**	**% Contribution**	**Primary Character**	**eV**
S_1	H-0→L+1	100	$n\pi*$	3.0363 (0.0003)
S_2	H-2→L+1	10.4	$CT, \pi\pi*$	3.7026 (0.0052)
	H-1→L+1	89.6		
S_3	H-0→L+0	100	$n\pi*$	3.9896 (0)
S_4	H-2→L+0	6.8	$CT, \pi\pi*$	4.0261 (0.0023)
	H-1→L+0	93.2		
S_5	H-2→L+0	8.0	$\pi\pi*$	4.0927 (0.2414)
	H-2→L+1	81.8		
	H-1→L+1	10.3		
T_1	H-2→L+1	76.6	$\pi\pi*$	2.4398
	H-1→L+1	23.4		
T_2	H-0→L+1	100	$n\pi*$	2.7184
T_3	H-2→L+1	3.4	$\pi\pi*$	3.4738
	H-2→L+4	16.9		
	H-1→L+1	11.9		
	H-1→L+4	67.8		
T_4	H-5→L+1	55.3	$\pi\pi*$	3.5713
	H-4→L+1	19.1		
	H-2→L+8	4.5		
	H-1→L+1	11.6		
	H-1→L+4	9.5		
T_5	H-5→L+1	11.4	$\pi\pi*$	3.7549
	H-4→L+1	4.1		
	H-2→L+1	20.0		
	H-1→L+1	59.6		
	H-1→L+4	4.9		

Table 25: Vertical excitation energies for 4tThd-C in water using the M052X/6-31G(d) level of theory

State	**Transitions**	**% Contribution**	**Primary Character**	**eV**
S_1	H-1→L+0	100	$n\pi*$	3.3949 (0)
S_2	H-0→L+0	100	$\pi\pi*$	4.3043 (0.5113)
S_3	H-4→L+0	28.1	$\pi\pi*$	5.1564 (0.1521)
		29.8		

	H-2→L+0 H-2→L+1	42.1		
S_4	H-4→L+0 H-3→L+0 H-2→L+1	59.2 3.5 37.4	$CT, \pi\pi*$	5.2111 (0.014)
S_5	H-4→L+0 H-3→L+0 H-2→L+0 H-2→L+1	5.9 7.8 69.6 16.8	$CT, \pi\pi*$	5.3433 (0.0512)
T_1	H-0→L+0 H-0←L+0	97.9 2.1	$\pi\pi*$	2.6851
T_2	H-1→L+0	100	$n\pi*$	3.1191
T_3	H-2→L+1 H-2→L+4	96.8 3.2	$\pi\pi*$	3.7692
T_4	H-4→L+0 H-3→L+0 H-0→L+3	87.7 2.9 9.5	$\pi\pi*$	3.839
T_5	H-4→L+1 H-3→L+1 H-3→L+4 H-2→L+4	3.0 90.5 2.9 3.6	$\pi\pi*$	4.5755

Table 26: Vertical excitation energies for 4tThd-C in vacuum using the M052X/6-31G(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-2→L+1 H-1→L+1 H-0→L+1	8.3 79.3 12.4	$n\pi*$	3.1334 (0.0001)
S_2	H-3→L+1 H-2→L+1 H-1→L+1 H-0→L+1	2.5 7.7 17.7 72.1	$\pi\pi*$	4.3835 (0.3837)
S_3	H-5→L+1 H-4→L+1 H-3→L+1 H-2→L+1 H-1→L+1 H-0→L+1	6.6 4.6 10.8 68.7 3.7 5.8	$CT, \pi\pi*$	4.9493 (0.0447)
S_4	H-2→L+2 H-2→L+3 H-0→L+2 H-0→L+3	9.8 58.2 4.6 27.4	$\pi\pi*$	5.0646 (0.1174)
S_5	H-5→L+1	22.6	$CT, \pi\pi*$	5.213 (0.0142)

	H-4→L+1	11.3		
	H-3→L+1	37.7		
	H-2→L+1	17.1		
	H-0→L+1	11.2		
T_1	H-2→L+1	19.3	$\pi\pi*$	2.5736
	H-1→L+1	13.7		
	H-0→L+1	67.0		
T_2	H-2→L+1	6.5	$n\pi*$	2.8223
	H-1→L+1	85.3		
	H-0→L+1	8.2		
T_3	H-2→L+2	10.5	$\pi\pi*$	3.7206
	H-2→L+3	62.6		
	H-0→L+2	3.9		
	H-0→L+3	23.0		
T_4	H-5→L+1	46.2	$\pi\pi*$	3.791
	H-4→L+1	20.0		
	H-3→L+1	23.3		
	H-2→L+1	3.2		
	H-0→L+6	7.3		
T_5	H-5→L+5	12.0	$\pi\pi*$	4.5068
	H-3→L+3	10.9		
	H-3→L+3	65.0		
	H-3→L+3	5.2		
	H-1→L+1	3.8		
	H-0→L+0	3.1		

Table 27: Vertical excitation energies for 4tThd-C in water using the PBE0/6-31G+(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-0→L+0	100	$n\pi*$	3.2327 (0.0001)
S_2	H-1→L+0	100	$\pi\pi*$	4.0535 (0.4284)
S_3	H-2→L+0	100	$CT, \pi\pi*$	4.2386 (0.0158)
S_4	H-5→L+0	2.7	$CT, \pi\pi*$	4.6355 (0.0063)
	H-3→L+0	97.3		
S_5	H-2→L+1	2.2	$CT, \pi\pi*$	4.7059 (0.0105)
	H-1→L+1	97.8		
T_1	H-1→L+0	100	$\pi\pi*$	2.5999
T_2	H-0→L+0	100	$n\pi*$	2.963
T_3	H-5→L+1	2.2	$\pi\pi*$	3.5468
	H-2→L+0	3.7		
	H-2→L+1	91.7		
	H-2→L+5	2.4		

T_4	H-7→L+0	3.6	$\pi\pi*$	3.6082
	H-5→L+0	4.2		
	H-4→L+0	80.6		
	H-3→L+0	5.6		
	H-1→L+3	5.9		
T_5	H-3→L+0	4.6	$\pi\pi*$	4.2198
	H-3→L+1	15.9		
	H-2→L+0	76.1		
	H-2→L+1	3.4		

Table 28: Vertical excitation energies for 4tThd-C in vacuum using the PBE0/6-31G+(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-1→L+1	2.1	$n\pi*$	3.0126 (0.0003)
	H-0→L+0	9.4		
	H-0→L+1	88.6		
S_2	H-2→L+0	2.3	$CT, \pi\pi*$	3.8893 (0.0211)
	H-2→L+1	18.2		
	H-1→L+0	8.7		
	H-1→L+1	68.8		
	H-0→L+1	2.0		
S_3	H-1→L+0	5.4	$n\pi*$	4.1253 (0.0001)
	H-0→L+0	85.5		
	H-0→L+1	9.1		
S_4	H-5→L+1	2.6	$\pi\pi*$	4.1477 (0.2894)
	H-2→L+0	9.9		
	H-2→L+1	68.5		
	H-1→L+0	2.9		
	H-1→L+1	16.1		
S_5	H-2→L+0	7.4	$CT, \pi\pi*$	4.2009 (0.0011)
	H-1→L+0	76.1		
	H-1→L+1	10.2		
	H-0→L+0	6.3		
T_1	H-2→L+0	5.3	$\pi\pi*$	2.4966
	H-2→L+1	50.7		
	H-1→L+0	3.9		
	H-1→L+1	37.6		
	H-0→L+1	2.6		
T_2	H-1→L+1	4.3	$n\pi*$	2.7028
	H-0→L+0	9.1		
	H-0→L+1	86.7		
T_3	H-2→L+2	35.3	$\pi\pi*$	3.4937
	H-2→L+3	6.0		

	H-1→L+2	47.5		
	H-1→L+3	8.1		
	H-0→L+2	3.3		
T_4	H-5→L+0	4.6	$\pi\pi*$	3.5139
	H-5→L+1	43.9		
	H-4→L+1	16.4		
	H-3→L+1	13.0		
	H-2→L+1	5.7		
	H-2→L+8	3.4		
	H-1→L+1	6.8		
	H-1→L+2	3.5		
	H-1→L+8	2.8		
T_5	H-5→L+1	5.3	$\pi\pi*$	3.9322
	H-3→L+1	3.3		
	H-2→L+0	4.4		
	H-2→L+1	35.2		
	H-1→L+0	5.9		
	H-1→L+1	45.9		

Table 29: Vertical excitation energies for C-4tThd in water using the M052X/6-31G(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-2→L+0	19.6	$n\pi*$	3.3326 (0.0001)
	H-1→L+0	80.4		
S_2	H-2→L+0	6.7	$\pi\pi*$	4.2547 (0.4266)
	H-1→L+0	2.2		
	H-0→L+0	91.1		
S_3	H-4→L+0	5.2	$CT,\pi\pi*$	4.8089 (0.0136)
	H-2→L+0	68.4		
	H-1→L+0	15.9		
	H-0→L+0	10.5		
S_4	H-2→L+1	58.5	$\pi\pi*$	5.1397 (0.1677)
	H-1→L+1	19.0		
	H-0→L+1	22.5		
S_5	H-5→L+0	6.4	$CT,\pi\pi*$	5.2555 (0.0211)
	H-4→L+0	60.9		
	H-3→L+0	24.5		
	H-2→L+0	5.5		
	H-1→L+0	2.6		
T_1	H-2→L+0	8.4	$\pi\pi*$	2.6363
	H-1←L+0	2.8		
	H-0→L+0	88.8		

T_2	H-2→L+0 H-1→L+0	20.2 79.8	$n\pi*$	3.0517
T_3	H-4→L+0 H-2→L+0 H-2→L+1 H-2→L+3 H-1→L+1 H-0→L+1	4.5 2.9 53.3 3.2 19.4 16.7	$\pi\pi*$	3.7417
T_4	H-5→L+0 H-4→L+0 H-3→L+0 H-2→L+0 H-2→L+1 H-1→L+1 H-0→L+3	6.5 63.6 6.5 5.5 6.9 2.4 8.5	$\pi\pi*$	3.801
T_5	H-3→L+1 H-3→L+5 H-1→L+1 H-0→L+1	90.1 3.4 3.4 3.1	$\pi\pi*$	4.5687

Table 30: Vertical excitation energies for C-4tThd in vacuum using the M052X/6-31G(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-2→L+0 H-1→L+0 H-0→L+0	44.1 48.7 7.2	$n\pi*$	3.2015 (0.0004)
S_2	H-2→L+0 H-1→L+0 H-0→L+0	11.2 35.6 53.2	$\pi\pi*$	4.3204 (0.3314)
S_3	H-2→L+0 H-1→L+0 H-0→L+0	42.5 14.4 43.2	$CT, \pi\pi*$	4.6102 (0.0141)
S_4	H-5→L+0 H-2→L+2 H-1→L+2 H-0→L+2	2.3 32.3 13.4 52.0	$\pi\pi*$	5.0763 (0.1082)
S_5	H-5→L+0 H-4→L+0 H-2→L+0 H-1→L+0	63.6 30.1 3.2 3.0	$n\pi*$	5.2559 (0.0241)
T_1	H-2→L+0 H-1←L+0 H-0→L+0	17.0 27.7 55.4	$\pi\pi*$	2.584

T_2	H-2→L+0	39.2	$n\pi*$	2.904
	H-1→L+0	57.1		
	H-0→L+0	3.7		
T_3	H-5→L+0	5.1	$\pi\pi*$	3.649
	H-4→L+0	2.5		
	H-2→L+0	5.8		
	H-2→L+2	26.9		
	H-1→L+2	12.1		
	H-0→L+0	3.8		
	H-0→L+2	43.8		
T_4	H-5→L+0	49.7	$n\pi*$	3.7554
	H-4→L+0	22.6		
	H-2→L+0	4.1		
	H-2→L+2	6.9		
	H-1→L+2	4.5		
	H-1→L+5	2.7		
	H-0→L+2	4.2		
	H-0→L+5	5.3		
T_5	H-4→L+2	2.5	$\pi\pi*$	4.4914
	H-3→L+0	4.5		
	H-3→L+2	82.0		
	H-3→L+7	4.7		
	H-2→L+2	3.0		
	H-1→L+2	3.4		

Table 31: Vertical excitation energies for C-4tThd in water using the PBE0/6-31G+(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-2→L+0	2.6	$n\pi*$	3.182 (0.0004)
	H-1→L+0	8.5		
	H-0→L+0	88.9		
S_2	H-2→L+0	40.5	$CT, \pi\pi*$	3.9041 (0.0841)
	H-1→L+0	48.8		
	H-0→L+0	10.7		
S_3	H-2→L+0	58.5	$\pi\pi*$	4.0542 (0.2888)
	H-1→L+0	41.5		
S_4	H-3→L+0	100	$CT, \pi\pi*$	4.5295 (0.0076)
S_5	H-2→L+1	7.0	$CT, \pi\pi*$	4.7056 (0.0733)
	H-1→L+1	71.7		
	H-0→L+1	21.3		
T_1	H-2→L+0	13.8	$\pi\pi*$	2.5621
	H-1←L+0	81.8		
	H-0→L+0	4.4		

T_2	H-2→L+0	3.2	$n\pi*$	2.9063
	H-1→L+0	8.2		
	H-0→L+0	88.6		
T_3	H-7→L+0	3.2	$\pi\pi*$	3.5079
	H-6→L+0	3.6		
	H-5→L+0	5.1		
	H-2→L+0	12.8		
	H-2→L+1	48.1		
	H-2→L+2	2.7		
	H-1→L+1	12.7		
	H-0→L+1	11.7		
T_4	H-7→L+0	12.6	$\pi\pi*$	3.5683
	H-6→L+0	15.4		
	H-5→L+0	22.0		
	H-4→L+0	4.3		
	H-3→L+0	6.0		
	H-2→L+0	10.1		
	H-2→L+1	21.5		
	H-1→L+2	4.8		
	H-0→L+1	3.2		
T_5	H-7→L+0	3.2	$\pi\pi*$	3.9878
	H-6→L+0	4.8		
	H-5→L+0	7.5		
	H-3→L+0	2.9		
	H-2→L+0	66.9		
	H-1→L+0	8.2		
	H-0→L+0	6.4		

Table 32: Vertical excitation energies for C-4tThd in vacuum using the PBE0/6-31G+(d) level of theory

State	**Transitions**	**% Contribution**	**Primary Character**	**eV**
S_1	H-1→L+0	28.9	$n\pi*$	3.0663 (0.0008)
	H-0→L+0	71.1		
S_2	H-2→L+0	17.7	$CT, \pi\pi*$	3.7168 (0.0143)
	H-1→L+0	56.9		
	H-0→L+0	25.4		
S_3	H-5→L+0	3.4	$\pi\pi*$	4.0668 (0.2492)
	H-2→L+0	79.5		
	H-1→L+0	13.9		
	H-0→L+0	3.2		
S_4	H-2→L+1	8.0	$n\pi*$	4.3267 (0.0005)
	H-1→L+1	41.1		
	H-0→L+1	50.8		

S_5	H-4→L+0	14.9	$n\pi*$	4.3757 (0.0227)
	H-3→L+0	85.1		
T_1	H-2→L+0	66.8	$\pi\pi*$	2.5076
	H-1←L+0	19.3		
	H-0→L+0	13.9		
T_2	H-1→L+0	35.3	$n\pi*$	2.773
	H-0→L+0	64.7		
T_3	H-5→L+0	12.1	$\pi\pi*$	3.3663
	H-2→L+0	14.1		
	H-2→L+2	10.0		
	H-1→L+0	16.0		
	H-1→L+2	22.8		
	H-0→L+0	7.3		
	H-0→L+2	17.6		
T_4	H-8→L+0	2.6	$\pi\pi*$	3.4988
	H-5→L+0	46.4		
	H-2→L+0	4.7		
	H-2→L+2	14.0		
	H-1→L+0	4.0		
	H-1→L+2	15.5		
	H-0→L+2	12.8		
T_5	H-5→L+0	29.8	$\pi\pi*$	3.8537
	H-2→L+0	17.6		
	H-1→L+0	29.4		
	H-1→L+2	4.4		
	H-0→L+0	15.2		
	H-0→L+2	3.5		

Table 33: Vertical excitation energies for 4tThd-G in water using the M052X/6-31G(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-2→L+0	93.8	$n\pi*$	3.4173 (0.0006)
	H-1→L+0	2.1		
	H-0→L+0	4.1		
S_2	H-1→L+0	95.6	$\pi\pi*$	4.224 (0.4257)
	H-0→L+0	4.4		
S_3	H-2→L+0	4.9	$CT, \pi\pi*$	4.3862 (0.0068)
	H-1→L+0	2.9		
	H-0→L+0	92.2		
S_4	H-4→L+0	6.8	$CT, \pi\pi*$	5.2126 (0.0388)
	H-3→L+0	93.2		
S_5	H-0→L+1	56.3	$\pi\pi*$	5.3491 (0.1506)
	H-0→L+2	37.0		

	H-0→L+4	6.7		
T_1	H-1→L+0	100	$\pi\pi*$	2.7068
T_2	H-2→L+0	96.2	$n\pi*$	3.1589
	H-0→L+0	3.8		
T_3	H-4→L+0	6.6	$\pi\pi*$	3.8341
	H-3→L+0	74.8		
	H-1→L+2	3.1		
	H-1→L+4	4.4		
	H-0→L+0	8.2		
	H-0→L+1	3.0		
T_4	H-12→L+1	2.7	$\pi\pi*$	3.9892
	H-3→L+0	5.8		
	H-0→L+0	11.8		
	H-0→L+1	79.7		
T_5	H-0→L+0	3.4	$\pi\pi*$	4.1969
	H-0→L+2	64.3		
	H-0→L+4	32.4		

Table 34: Vertical excitation energies for 4tThd-G in vacuum using the M052X/6-31G(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-2→L+0	93.0	$n\pi*$	3.3403 (0.0006)
	H-1→L+0	3.8		
	H-0→L+0	3.2		
S_2	H-1→L+0	91.2	$\pi\pi*$	4.2618 (0.3175)
	H-0→L+0	8.8		
S_3	H-2→L+0	4.9	$CT,\pi\pi*$	4.3174 (0.016)
	H-1→L+0	5.9		
	H-0→L+0	89.3		
S_4	H-5→L+0	2.6	$CT,\pi\pi*$	5.2025 (0.0289)
	H-3→L+0	97.4		
S_5	H-0→L+1	100	$CT,\pi\pi*$	5.3506 (0.0003)
T_1	H-1→L+0	100	$\pi\pi*$	2.6565
T_2	H-2→L+0	97.0	$n\pi*$	3.0709
	H-0→L+0	3.0		
T_3	H-4→L+0	3.1	$\pi\pi*$	3.8035
	H-3→L+0	85.6		
	H-1→L+2	4.0		
	H-1→L+4	7.3		
T_4	H-3→L+0	4.2	$\pi\pi*$	4.0307
	H-0→L+0	23.6		
	H-0→L+3	10.6		
		15.7		

	H-0→L+4 H-0→L+5	45.9		
T_5	H-0→L+3 H-0→L+4 H-0→L+5 H-0→L+6 H-0→L+7	24.5 27.3 15.9 22.6 9.8	$\pi\pi*$	4.1493

Table 35: Vertical excitation energies for 4tThd-G in water using the PBE0/6-31G+(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-2→L+0 H-1→L+0 H-0→L+0	45.6 32.0 22.5	$n\pi*$	3.2222 (0.0009)
S_2	H-2→L+0 H-1→L+0 H-0→L+0	14.3 8.0 77.7	$CT, \pi\pi*$	3.4733 (0.0029)
S_3	H-2→L+0 H-1→L+0	40.0 60.0	$\pi\pi*$	4.0132 (0.3673)
S_4	H-5→L+0 H-4→L+0 H-3→L+0	6.9 45.3 47.8	$n\pi*$	4.8024 (0.051)
S_5	H-8→L+0 H-7→L+0 H-6→L+0 H-5→L+0	20.0 5.5 61.3 13.3	$n\pi*$	4.9146 (0.0001)
T_1	H-2→L+0 H-1→L+0	29.5 70.5	$\pi\pi*$	2.6293
T_2	H-2→L+0 H-1→L+0 H-0→L+0	59.4 28.5 12.1	$n\pi*$	2.9807
T_3	H-4→L+0 H-2→L+0 H-0→L+0 H-0→L+1	2.8 10.8 80.7 5.7	$\pi\pi*$	3.3944
T_4	H-8→L+0 H-5→L+0 H-4→L+0 H-3→L+0 H-0→L+0 H-0→L+1	2.8 13.3 44.5 26.7 2.8 9.8	$n\pi*$	3.662
T_5	H-4→L+0 H-0→L+0	3.2 8.1	$\pi\pi*$	3.7428

	H-0→L+1	80.9
	H-0→L+3	7.8

Table 36: Vertical excitation energies for 4tThd-G in vacuum using the PBE0/6-31G+(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-2→L+0	93.0	$n\pi*$	3.1571 (0.0009)
	H-1→L+0	3.8		
	H-0→L+0	3.2		
S_2	H-2→L+0	7.9	$CT, \pi\pi*$	3.3808 (0.0015)
	H-1→L+0	12.6		
	H-0→L+0	79.5		
S_3	H-0→L+1	100	$CT, \pi\pi*$	4.0222 (0.0002)
S_4	H-4→L+0	2.6	$\pi\pi*$	4.0364 (0.2673)
	H-2→L+0	64.6		
	H-1→L+0	32.9		
S_5	H-2→L+1	10.9	$n\pi*$	4.5412 (0)
	H-1→L+1	89.1		
T_1	H-2→L+0	52.5	$\pi\pi*$	2.5823
	H-1→L+0	47.5		
T_2	H-2→L+0	40.9	$\pi\pi*$	2.9042
	H-1→L+0	49.2		
	H-0→L+0	9.8		
T_3	H-4→L+0	3.2	$\pi\pi*$	3.3153
	H-2→L+0	6.6		
	H-1→L+0	3.1		
	H-0→L+0	84.2		
	H-0→L+6	2.9		
T_4	H-5→L+0	5.3	$n\pi*$	3.6164
	H-4→L+0	83.1		
	H-3→L+0	5.1		
	H-2→L+8	2.8		
	H-0→L+0	3.6		
T_5	H-0→L+4	66.4	$\pi\pi*$	3.7805
	H-0→L+6	23.3		
	H-0→L+8	10.3		

Table 37: Vertical excitation energies for G-4tThd in water using the M052X/6-31G(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-2→L+0	100	$n\pi*$	3.3427 (0)
S_2	H-1→L+0	71.3	$\pi\pi*$	4.2535 (0.4261)

	H-0→L+0	28.7		
S_3	H-1→L+0	28.7	$CT, \pi\pi*$	4.5875 (0.0849)
	H-0→L+0	71.3		
S_4	H-4→L+0	89.7	$CT, \pi\pi*$	5.2611 (0.0181)
	H-3→L+0	10.3		
S_5	H-0→L+1	60.5	$\pi\pi*$	5.3341 (0.1931)
	H-0→L+3	26.3		
	H-0→L+4	13.3		
T_1	H-1→L+0	91.8	$\pi\pi*$	2.6313
	H-0→L+0	6.0		
	H-1←L+0	2.2		
T_2	H-2→L+0	100	$n\pi*$	3.0589
T_3	H-4→L+0	80.6	$\pi\pi*$	3.8449
	H-3→L+0	8.6		
	H-1→L+3	7.2		
	H-0→L+0	3.6		
T_4	H-9→L+1	2.6	$\pi\pi*$	3.9814
	H-1→L+1	2.5		
	H-0→L+0	2.1		
	H-0→L+1	80.5		
	H-0→L+3	9.4		
	H-0→L+4	3.0		
T_5	H-16→L+8	2.6	$\pi\pi*$	4.2602
	H-0→L+3	21.1		
	H-0→L+4	76.3		

Table 38: Vertical excitation energies for G-4tThd in vacuum using the M052X/6-31G(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-2→L+1	2.7	$n\pi*$	3.0638 (0)
	H-1→L+1	97.3		
S_2	H-2→L+1	51.7	$\pi\pi*$	4.3766 (0.2811)
	H-0→L+1	48.3		
S_3	H-2→L+1	45.6	$\pi\pi*$	4.5637 (0.1533)
	H-0→L+1	54.4		
S_4	H-0→L+0	100	$CT, \pi\pi*$	5.0196 (0.0001)
S_5	H-5→L+1	2.2	$CT, \pi\pi*$	5.3285 (0.0107)
	H-4→L+1	92.5		
	H-2→L+4	2.2		
	H-0→L+5	3.1		
T_1	H-2→L+1	91.2	$\pi\pi*$	2.4866
	H-1→L+1	3.0		

	H-0→L+1	3.0		
	H-2→L+1	2.8		
T_2	H-2→L+1	3.0	$n\pi*$	2.7285
	H-1→L+1	94.6		
	H-1→L+4	2.4		
T_3	H-4→L+1	84.5	$\pi\pi*$	3.851
	H-2→L+4	12.6		
	H-0→L+1	2.9		
T_4	H-0→L+1	2.9	$\pi\pi*$	4.0691
	H-0→L+5	76.0		
	H-0→L+6	9.9		
	H-0→L+7	11.2		
T_5	H-0→L+4	9.5	$\pi\pi*$	4.1877
	H-0→L+5	15.1		
	H-0→L+7	75.3		

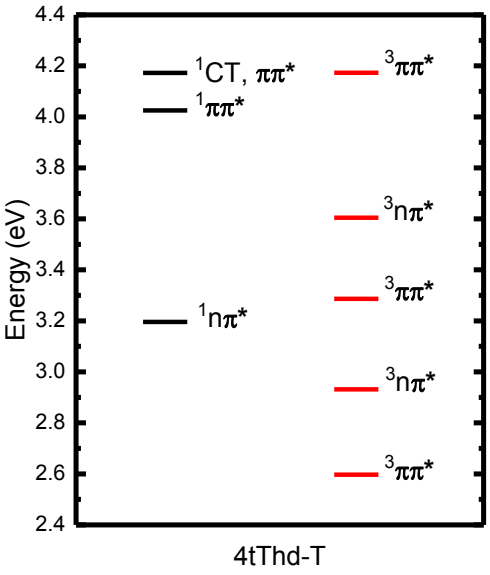
Table 39: Vertical excitation energies for G-4tThd in water using the PBE0/6-31+G(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-1→L+0	100	$n\pi*$	3.2028 (0.0001)
S_2	H-0→L+0	100	$CT, \pi\pi*$	3.4975 (0.0119)
S_3	H-2→L+0	100	$\pi\pi*$	4.082 (0.4274)
S_4	H-5→L+0	16.9	$CT, \pi\pi*$	4.8459 (0.0274)
	H-4→L+0	79.5		
	H-3→L+0	3.7		
S_5	H-0→L+1	84.1	$\pi\pi*$	4.9294 (0.1798)
	H-0→L+2	5.8		
	H-0→L+3	7.0		
	H-0→L+4	3.0		
T_1	H-2→L+0	92.8	$\pi\pi*$	2.5621
	H-0→L+0	7.2		
T_2	H-1→L+0	100	$n\pi*$	2.9248
T_3	H-5→L+0	3.9	$\pi\pi*$	3.4683
	H-4→L+0	6.8		
	H-2→L+0	5.5		
	H-0→L+0	79.0		
	H-0→L+1	4.7		
T_4	H-5→L+0	25.4	$\pi\pi*$	3.6582
	H-4→L+0	38.7		
	H-2→L+2	4.3		
	H-0→L+0	3.2		
	H-0→L+1	28.4		

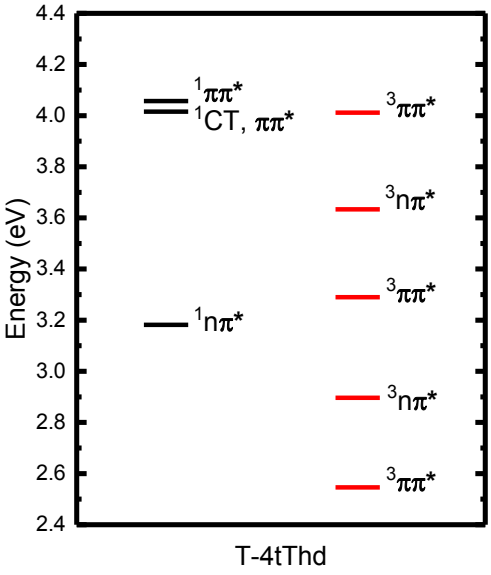
T_5	H-5→L+0	7.9	$\pi\pi*$	3.6997
	H-4→L+0	11.6		
	H-0→L+0	13.6		
	H-0→L+1	64.3		
	H-0→L+3	2.5		

Table 40: Vertical excitation energies for G-4tThd in vacuum using the PBE0/6-31+G(d) level of theory

State	Transitions	% Contribution	Primary Character	eV
S_1	H-1→L+0	70.4	$n\pi*$	2.9779 (0.0001)
	H-1→L+1	29.6		
S_2	H-0→L+0	72.1	$CT, \pi\pi*$	3.3992 (0.0046)
	H-0→L+1	27.9		
S_3	H-0→L+0	27.9	$CT, \pi\pi*$	3.7118 (0)
	H-0→L+1	72.1		
S_4	H-1→L+0	29.7	$n\pi*$	4.0976 (0)
	H-1→L+1	70.3		
S_5	H-5→L+0	2.4	$\pi\pi*$	4.1928 (0.3472)
	H-2→L+0	80.5		
	H-2→L+1	17.0		
T_1	H-2→L+0	68.5	$\pi\pi*$	2.4077
	H-2→L+1	29.1		
	H-0→L+0	2.4		
T_2	H-1→L+0	70.2	$n\pi*$	2.6385
	H-1→L+1	29.8		
T_3	H-5→L+0	3.3	$\pi\pi*$	3.3813
	H-0→L+0	69.4		
	H-0→L+1	27.3		
T_4	H-5→L+0	62.6	$\pi\pi*$	3.6274
	H-5→L+1	26.5		
	H-2→L+6	7.1		
	H-0→L+0	3.8		
T_5	H-0→L+0	28.5	$\pi\pi*$	3.7112
	H-0→L+1	71.5		



4tThd-T



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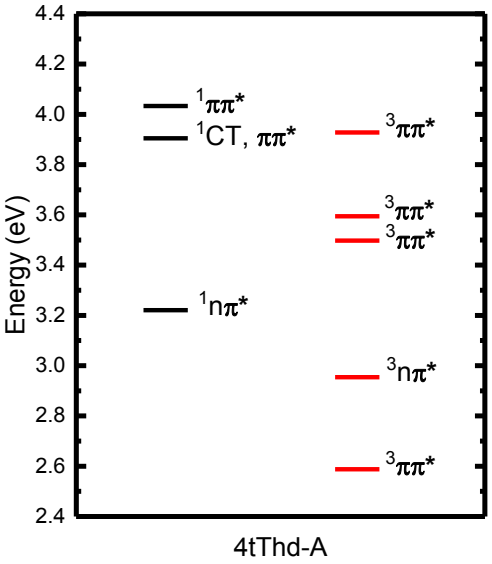
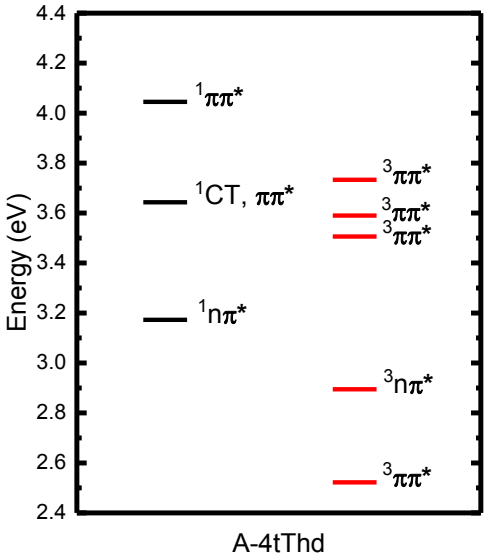


4tThd-A



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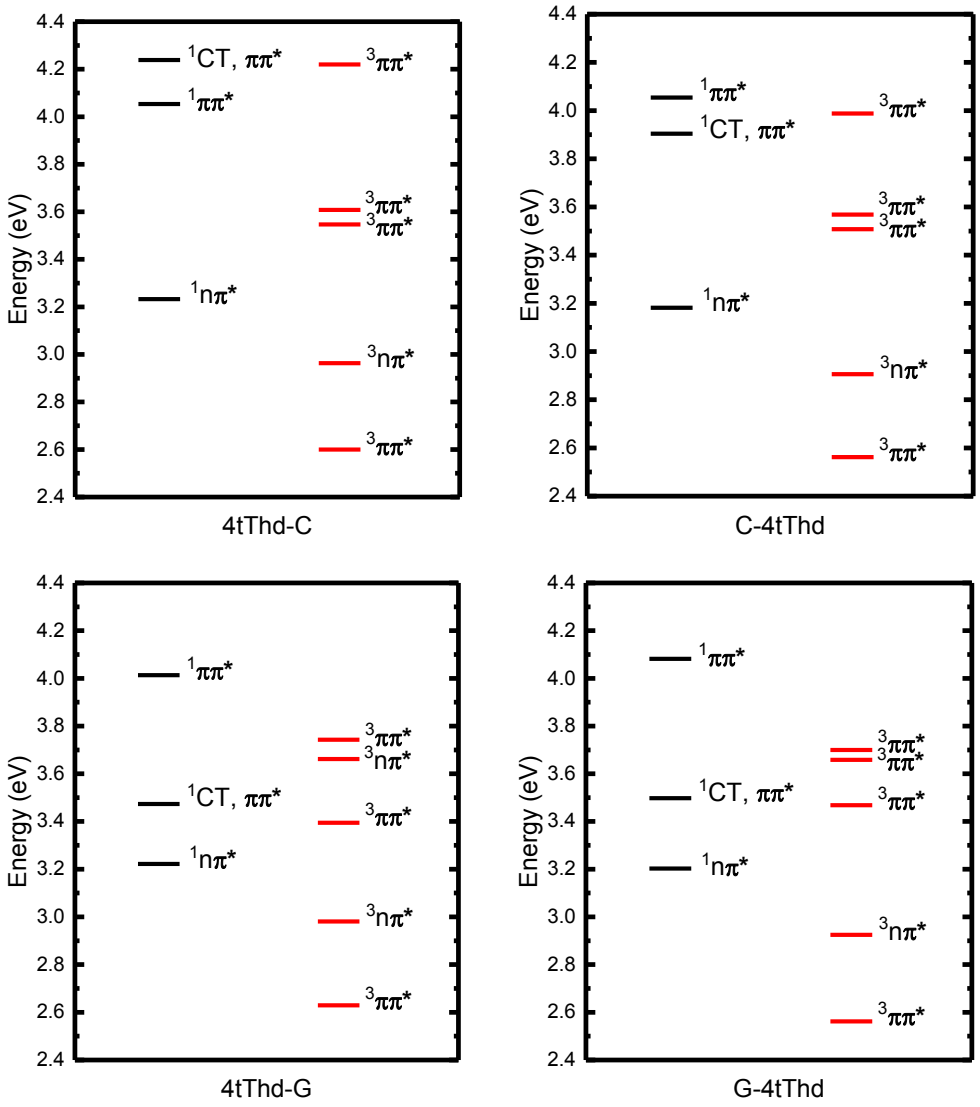
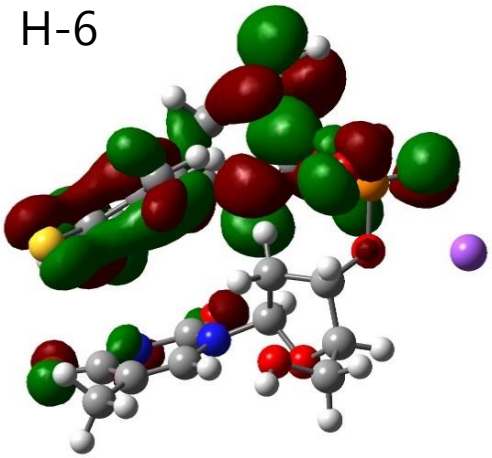


Figure 1. Vertical excitation energies for 4tThd-containing dinucleotides in water calculated at the PBE0/6-31+G(d) level of theory from ground state M052X/6-31G(d) optimiza

H-6



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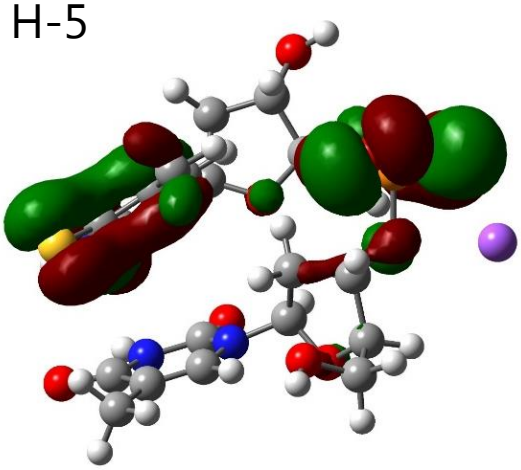

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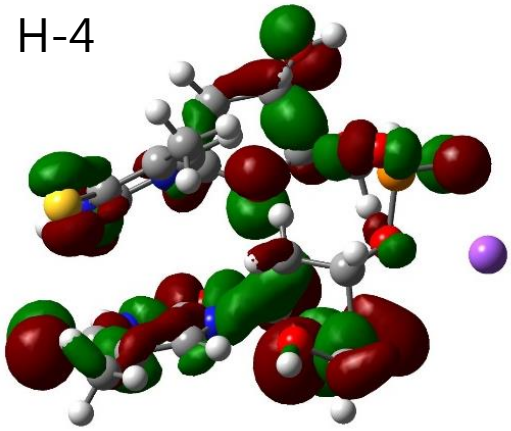

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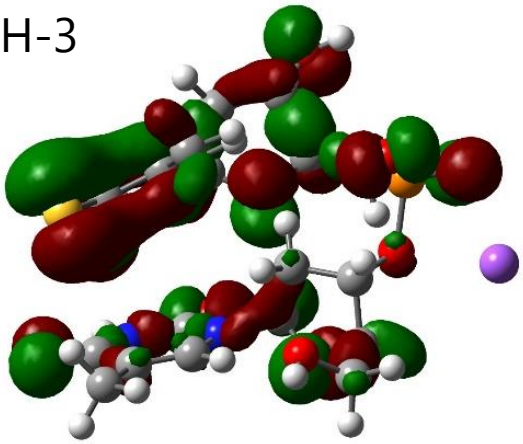

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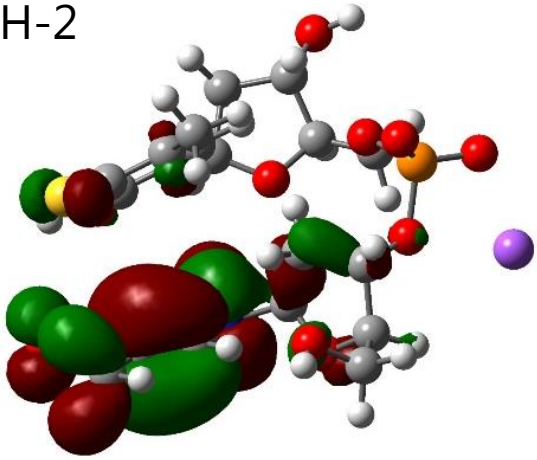

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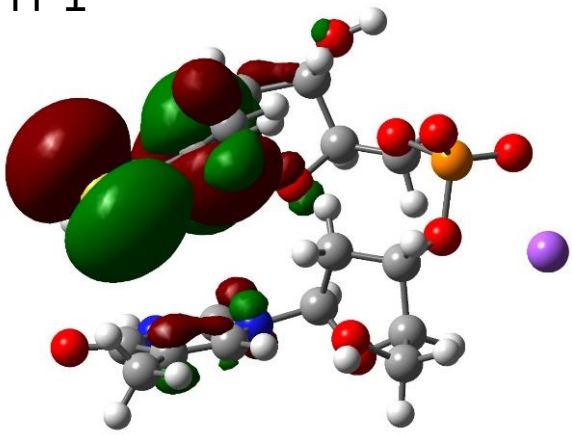


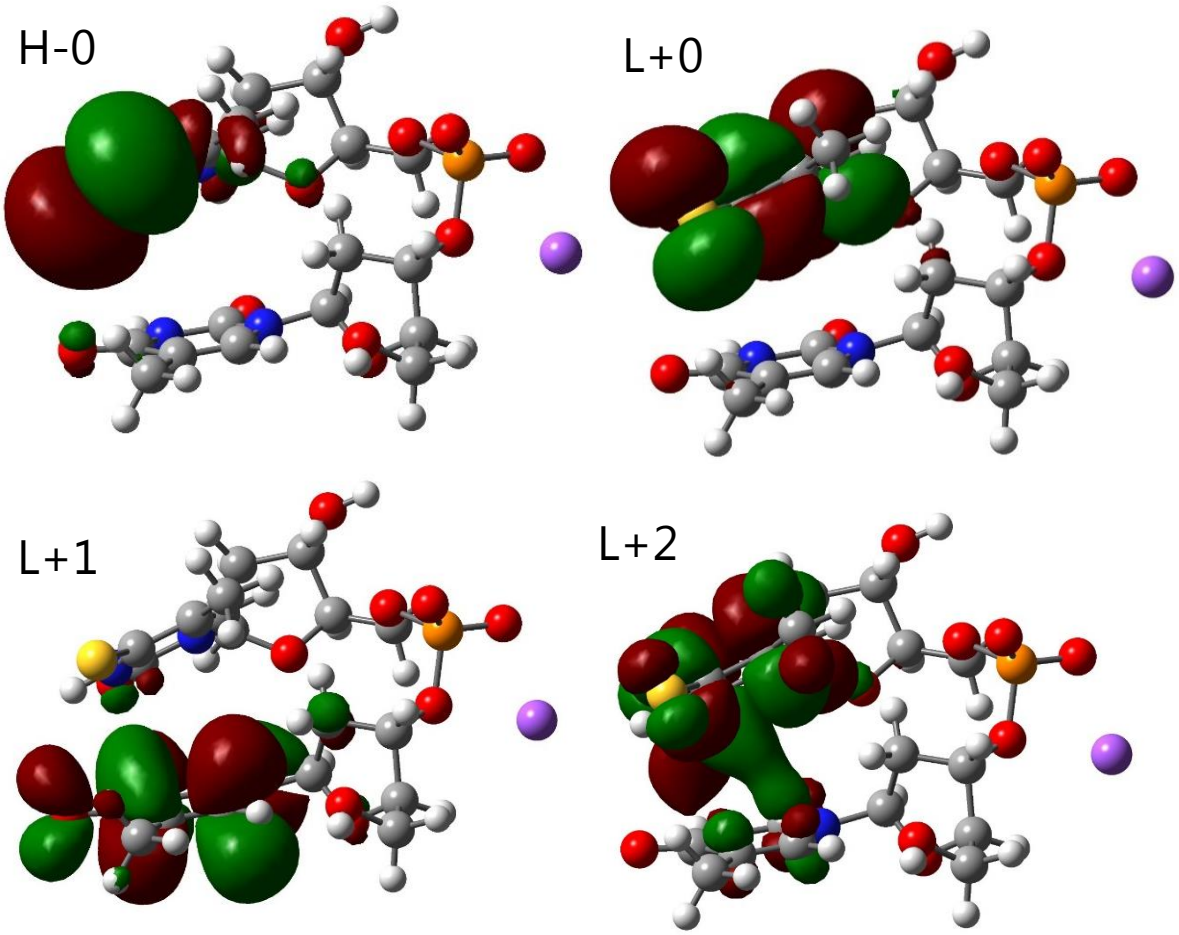
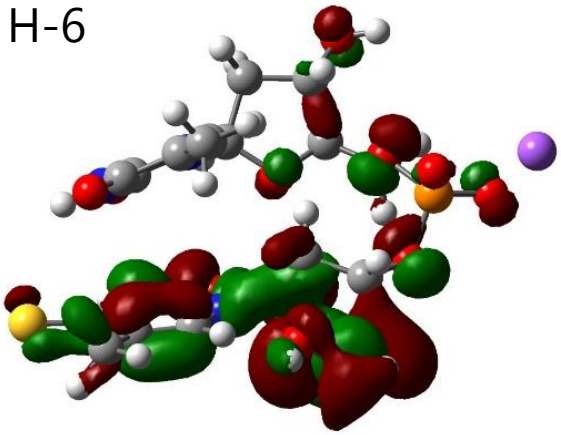


Figure 2. Primary Kohn-Sham orbitals of T-4tThd in water.

H-6



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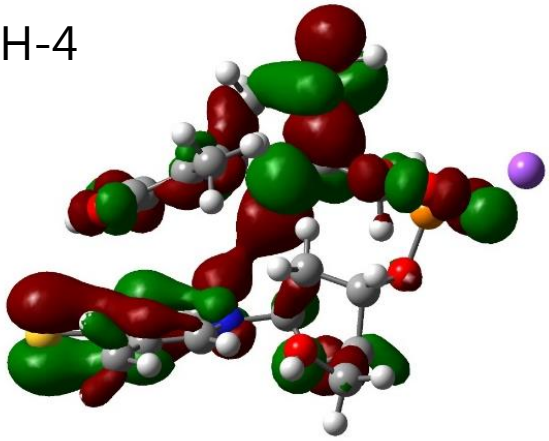

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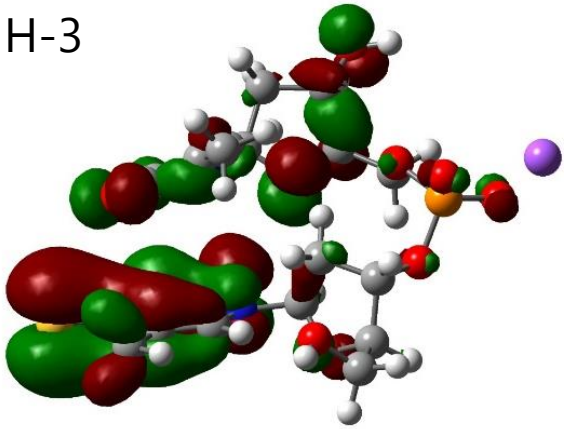

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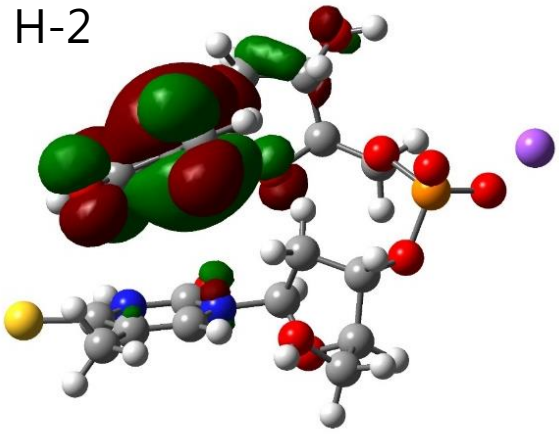

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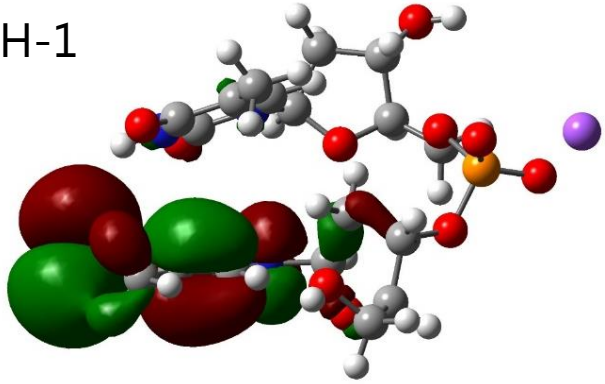

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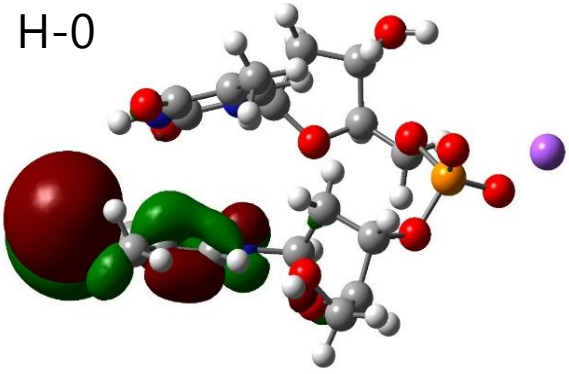


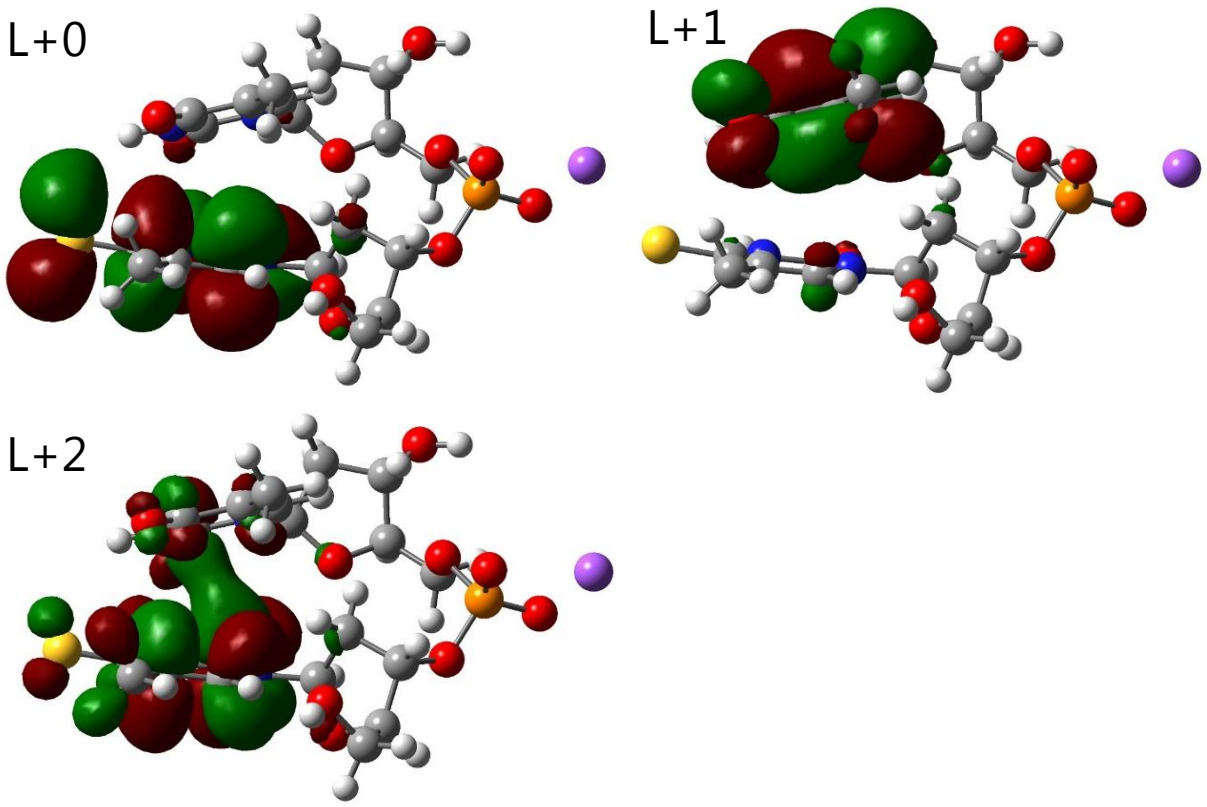
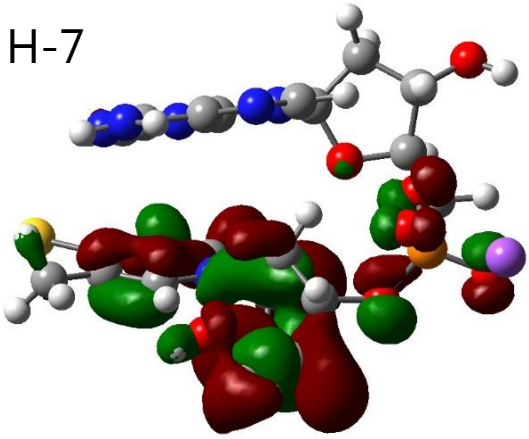


Figure 3. Primary Kohn-Sham orbitals of 4tThd-T in water.

H-7



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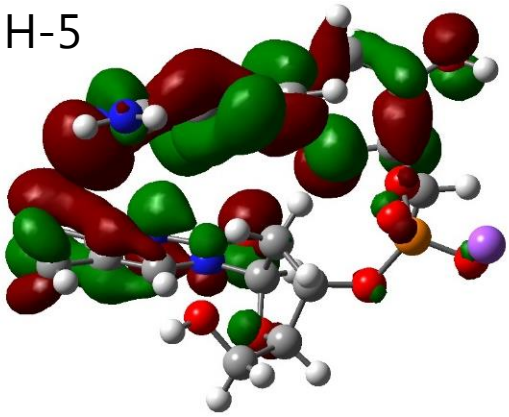

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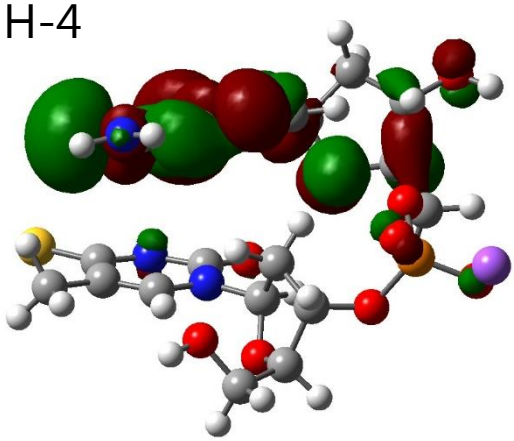

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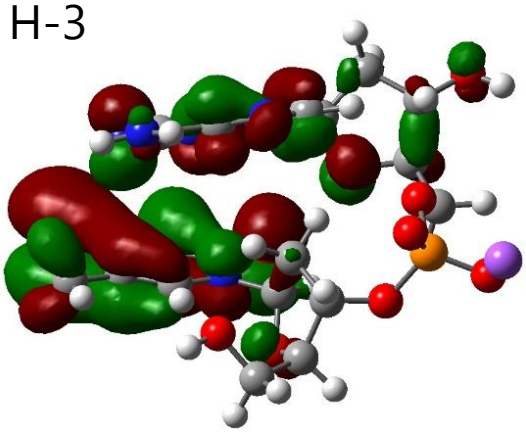

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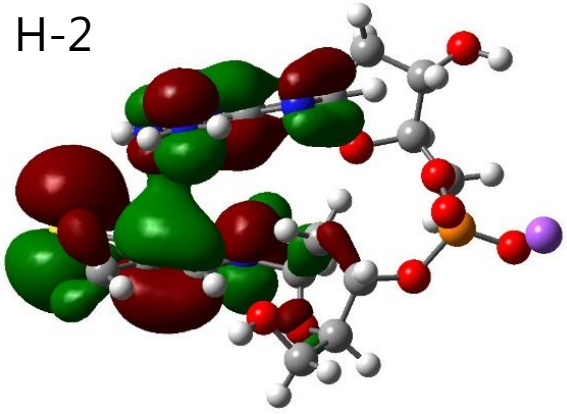

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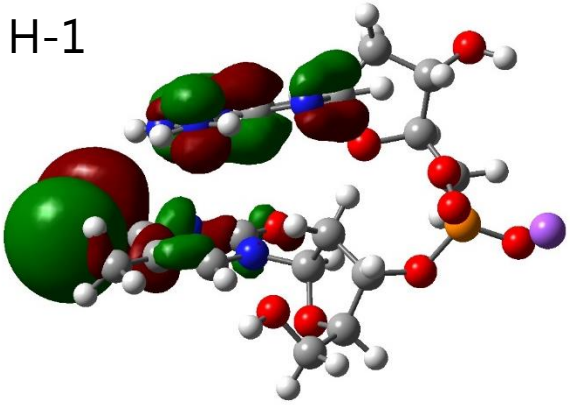


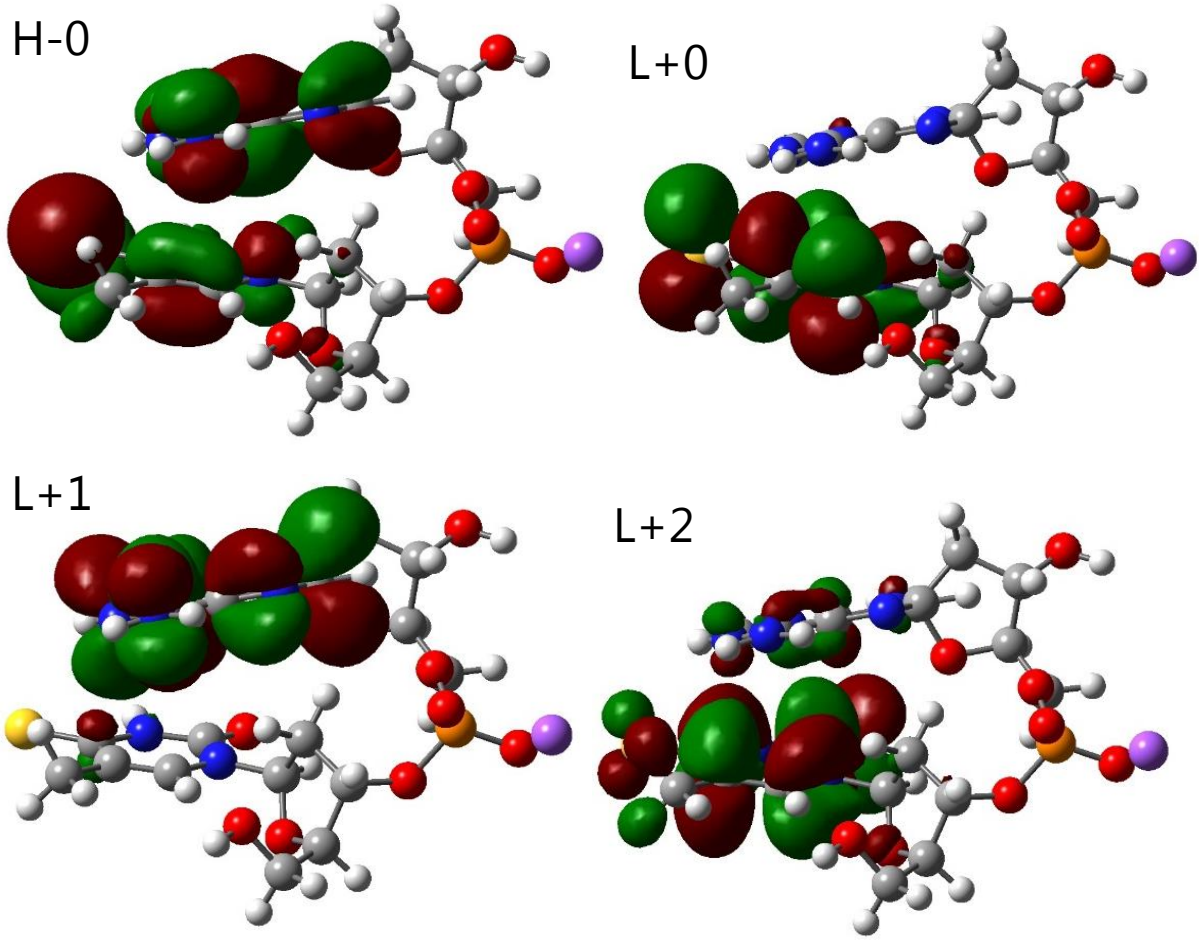
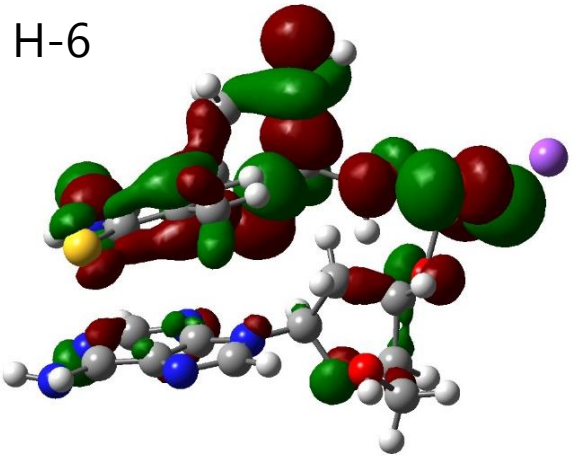


Figure 4. Primary Kohn-Sham orbitals of 4tThd-A in water.

H-6



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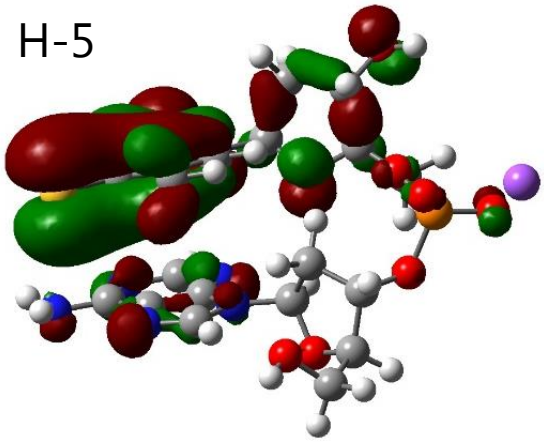

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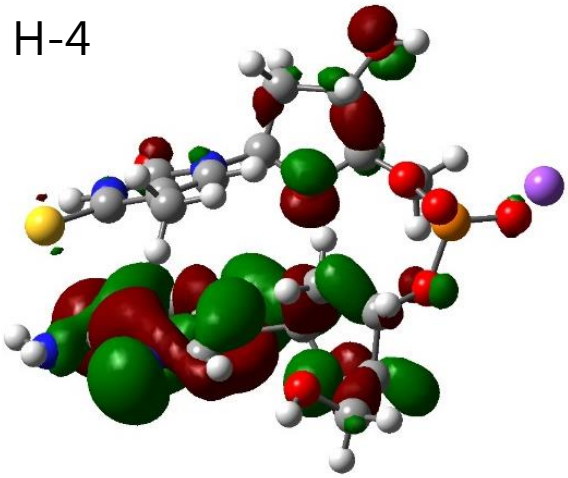

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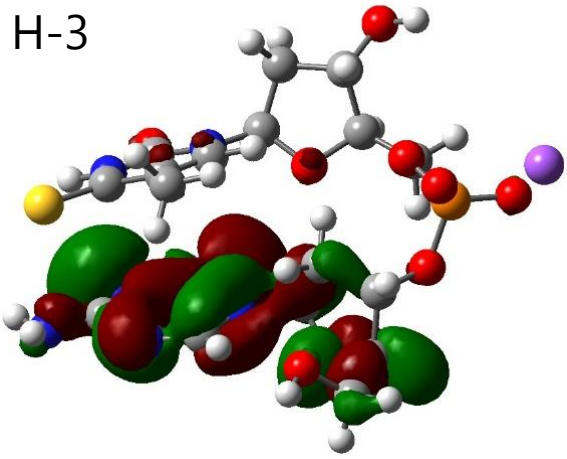

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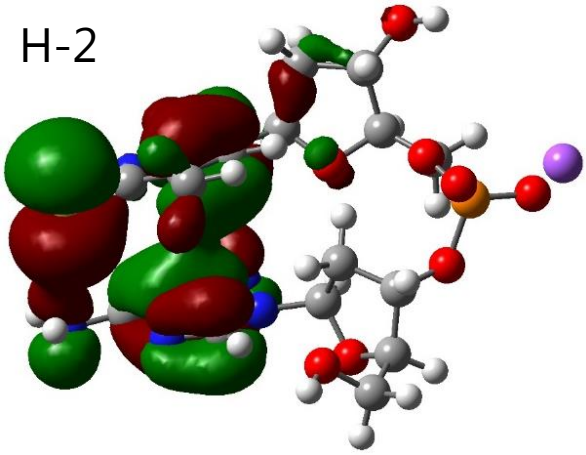

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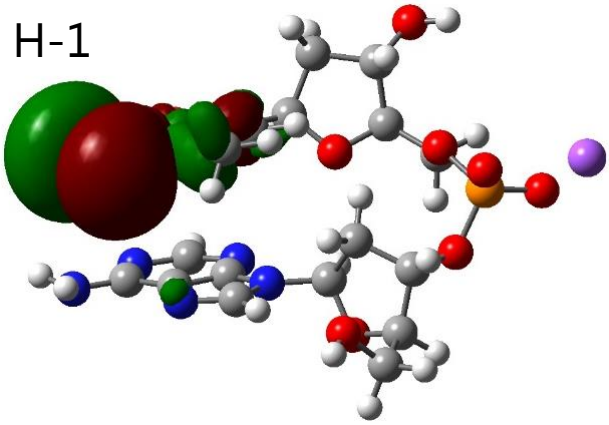


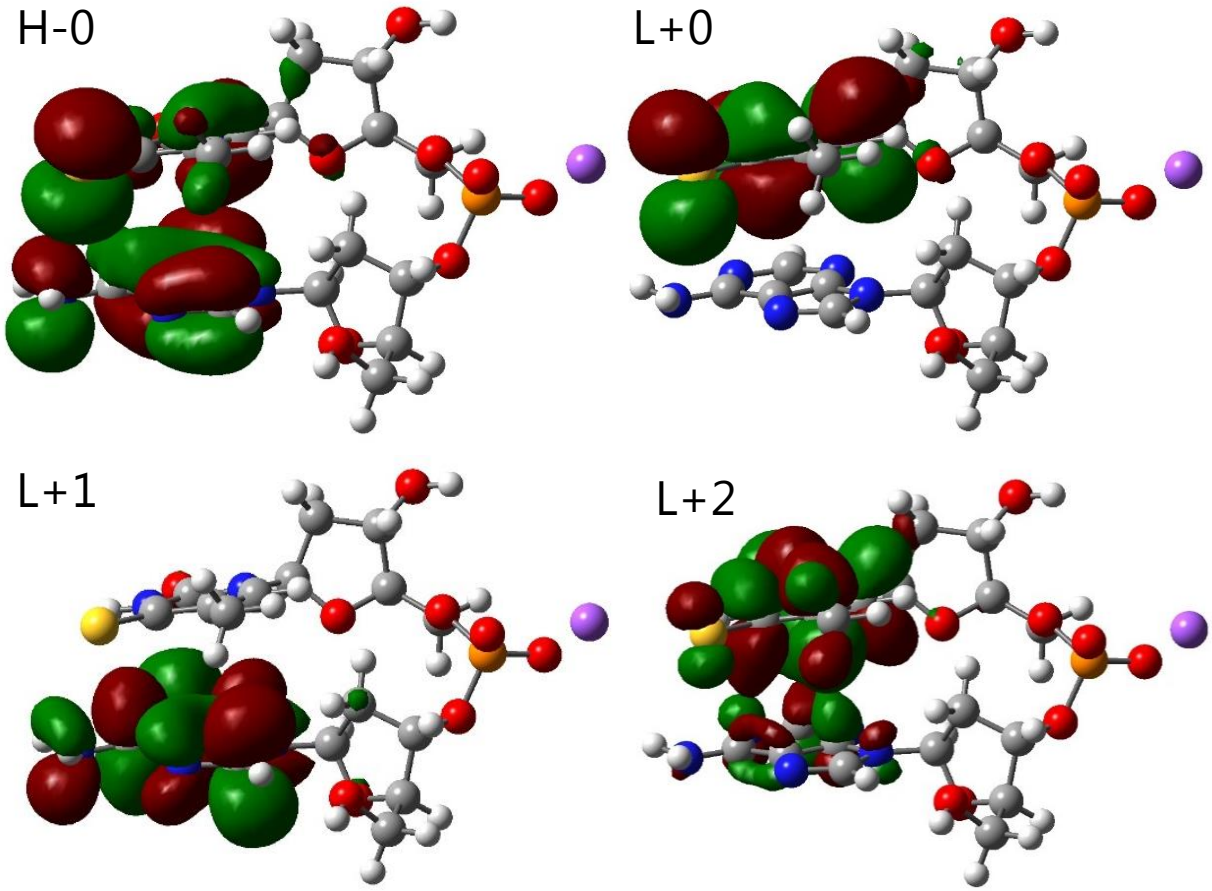
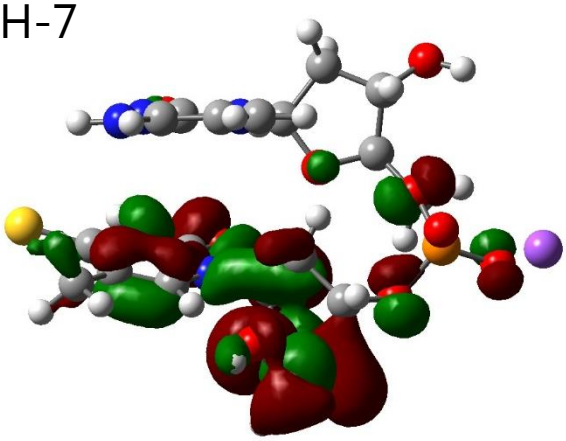


Figure 5. Primary Kohn-Sham orbitals of A-4tThd in water.

H-7



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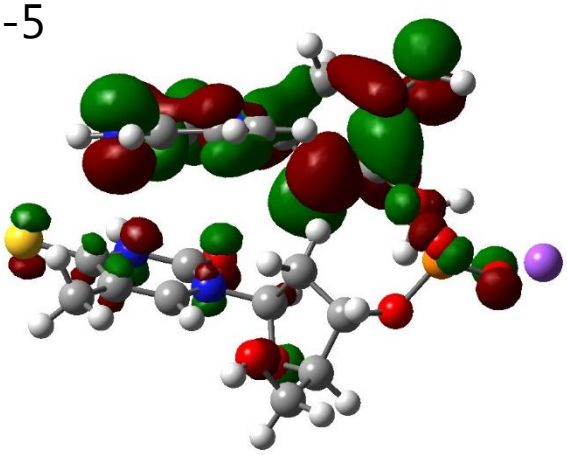

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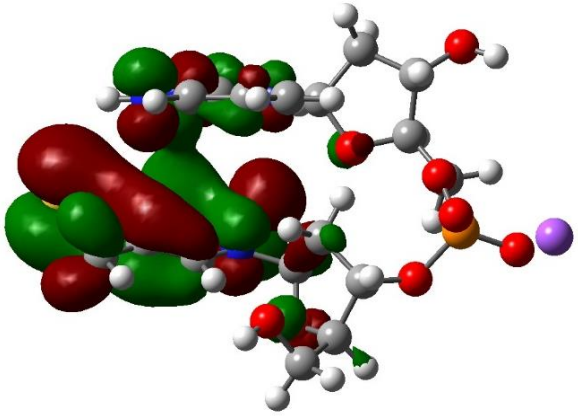

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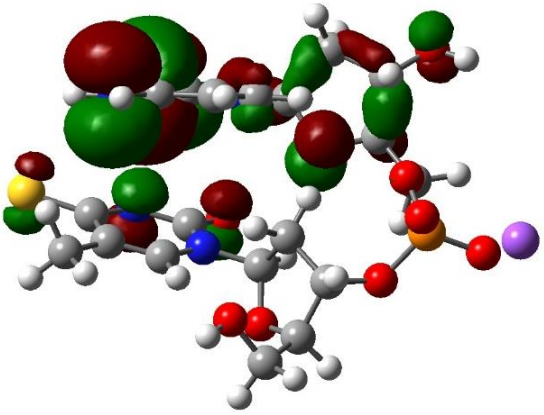
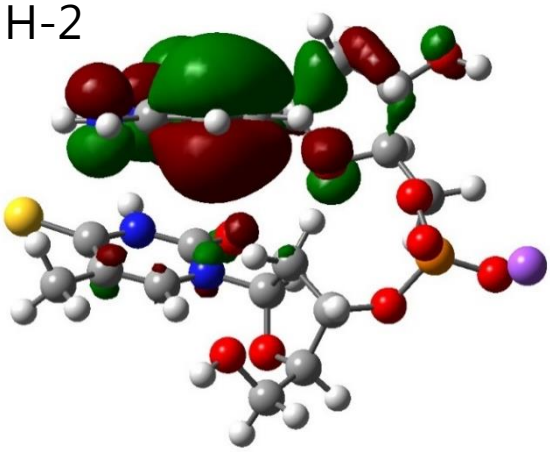


H-2



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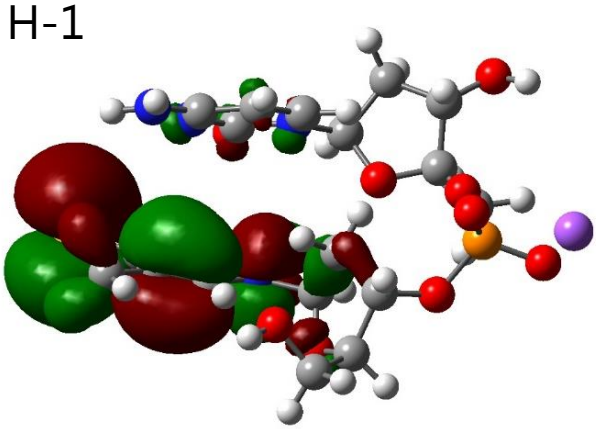


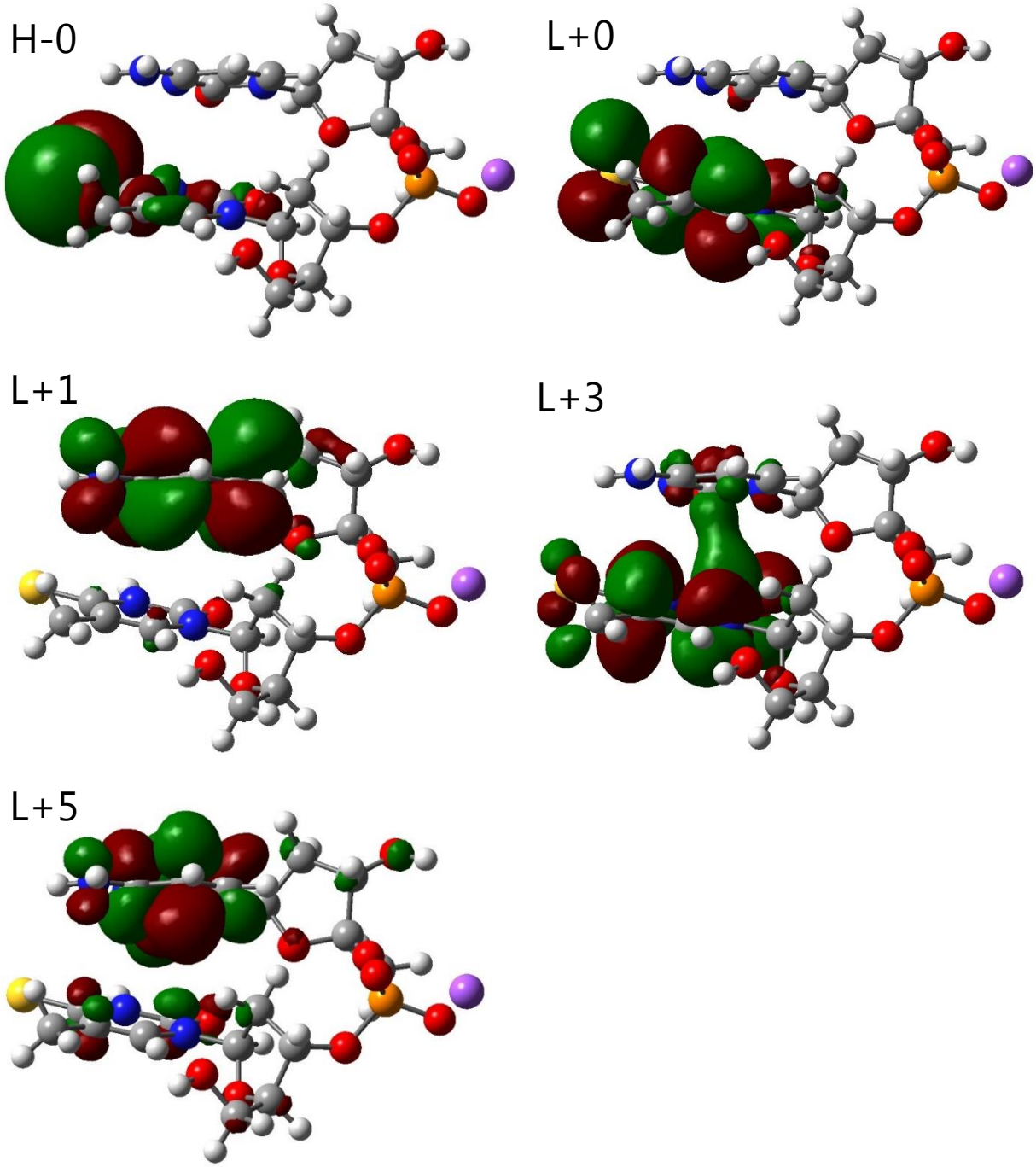
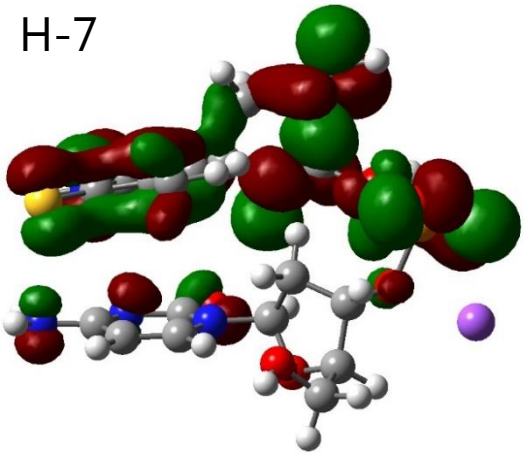


Figure 6. Primary Kohn-Sham orbitals of 4tThd-C in water.

H-7



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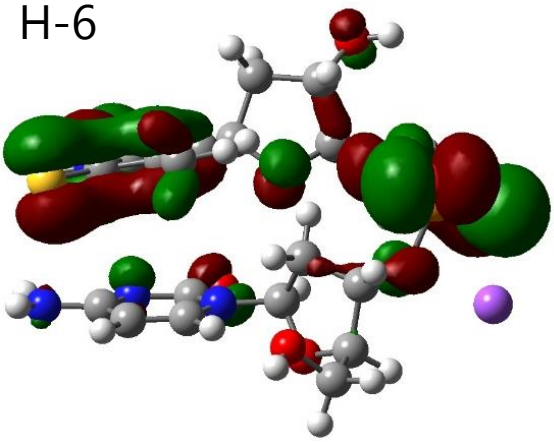

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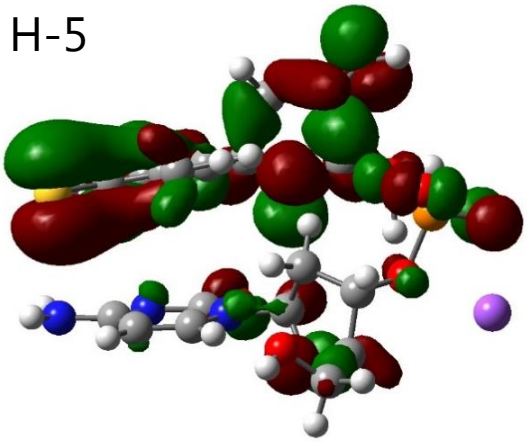

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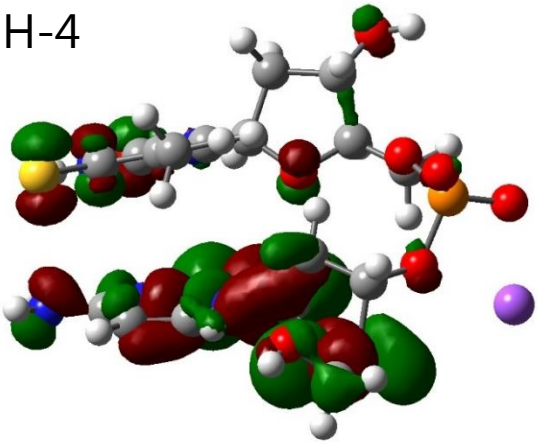

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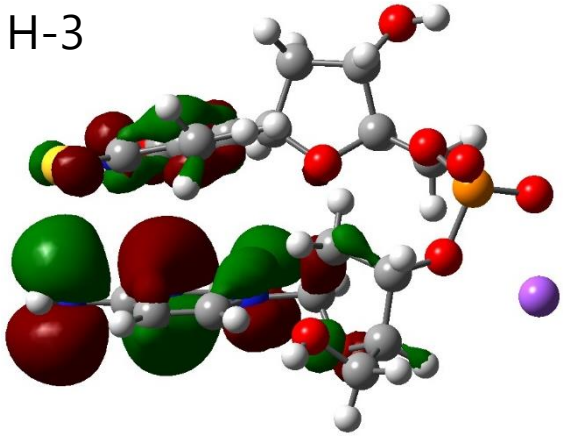

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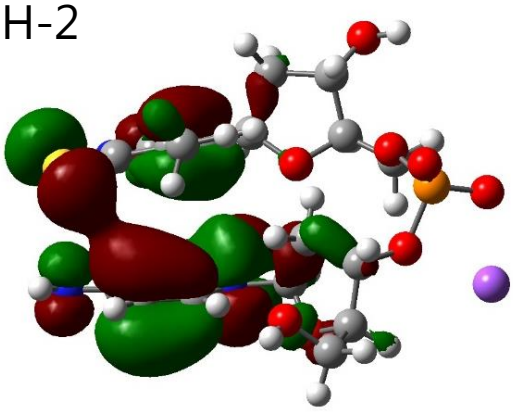


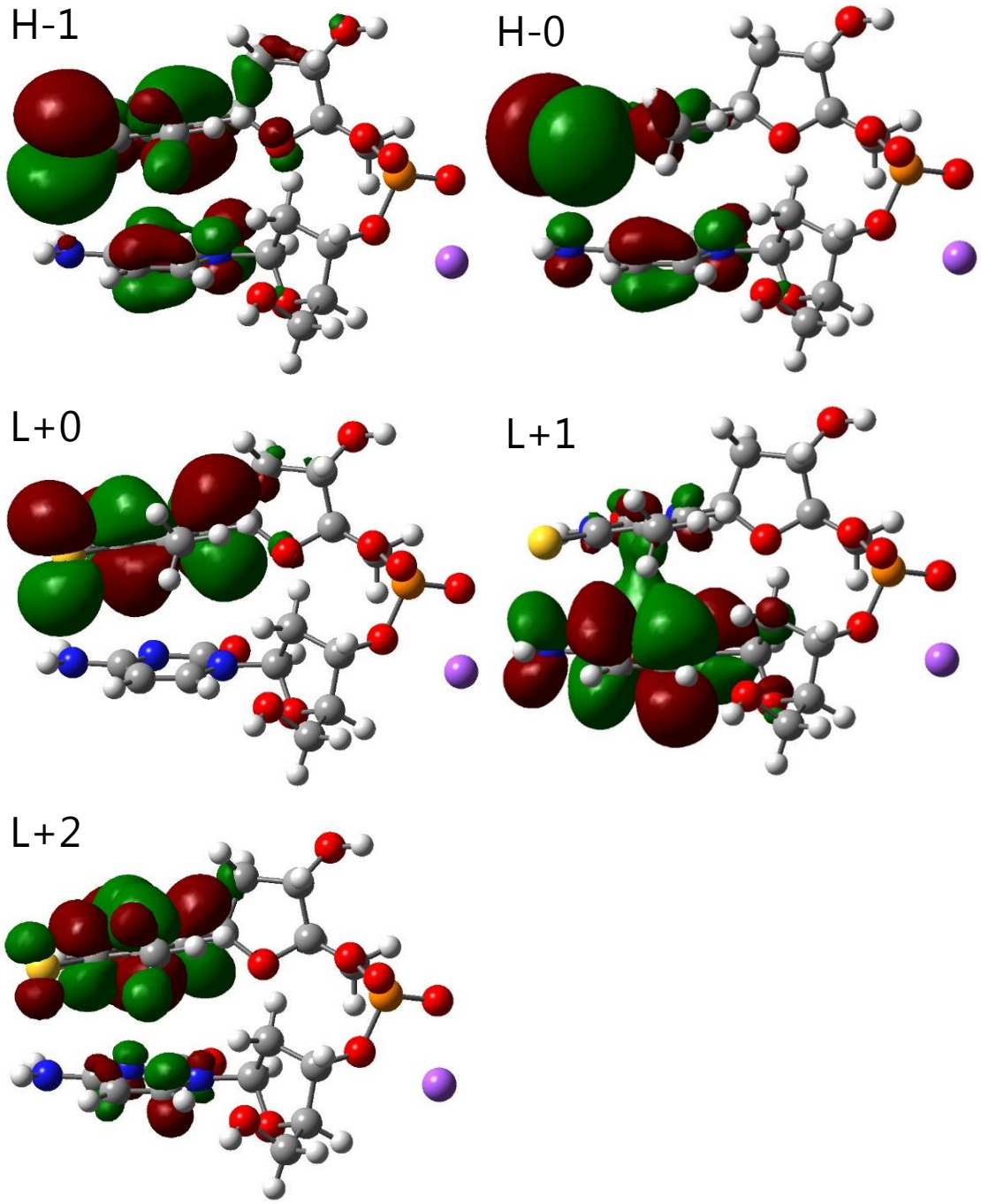
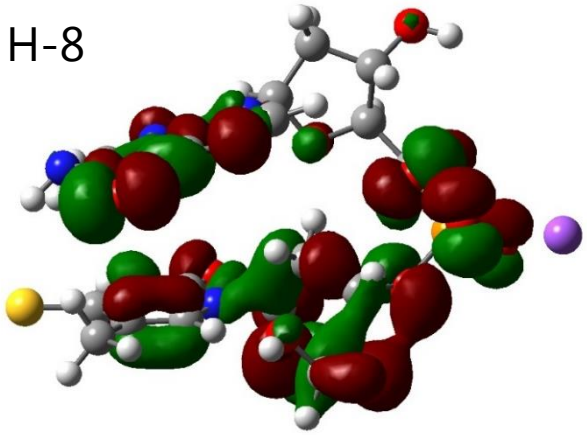


Figure 7. Primary Kohn-Sham orbitals of C-4tThd in water.

H-8



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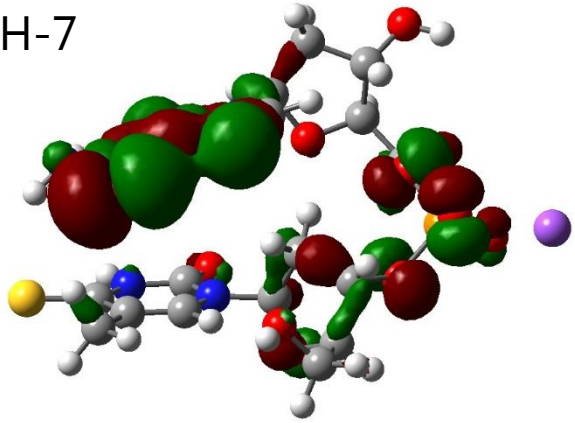

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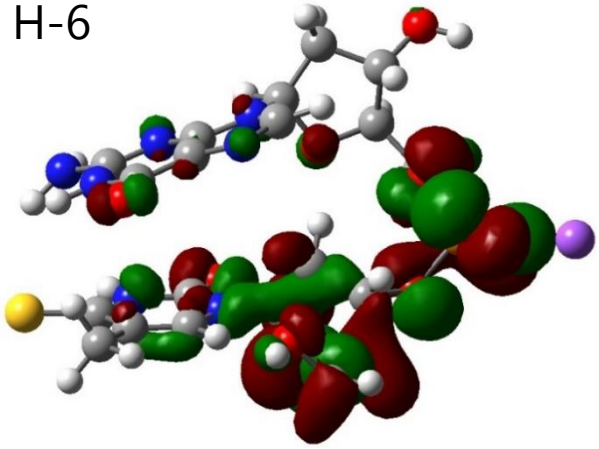

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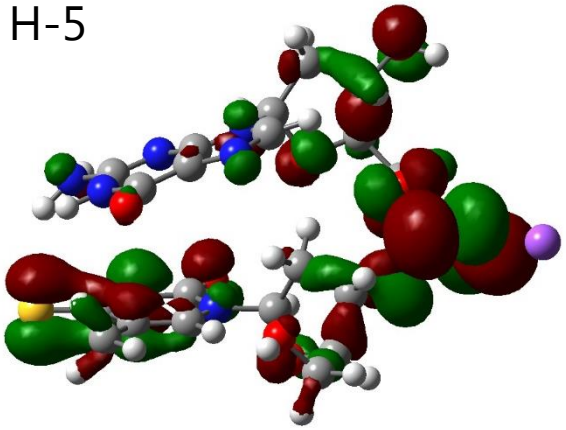

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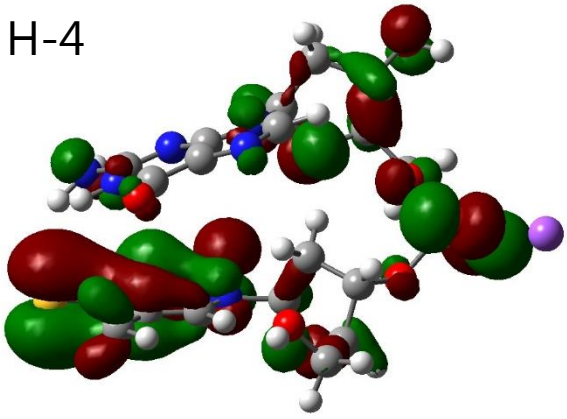

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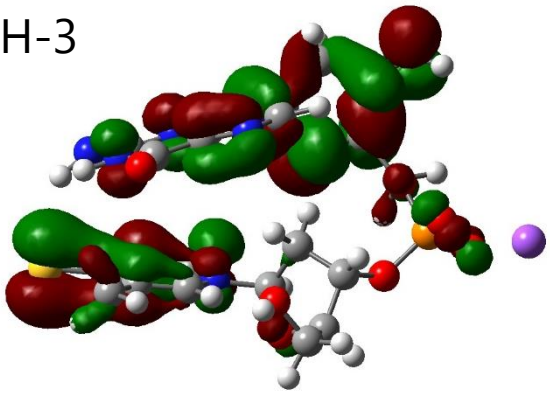


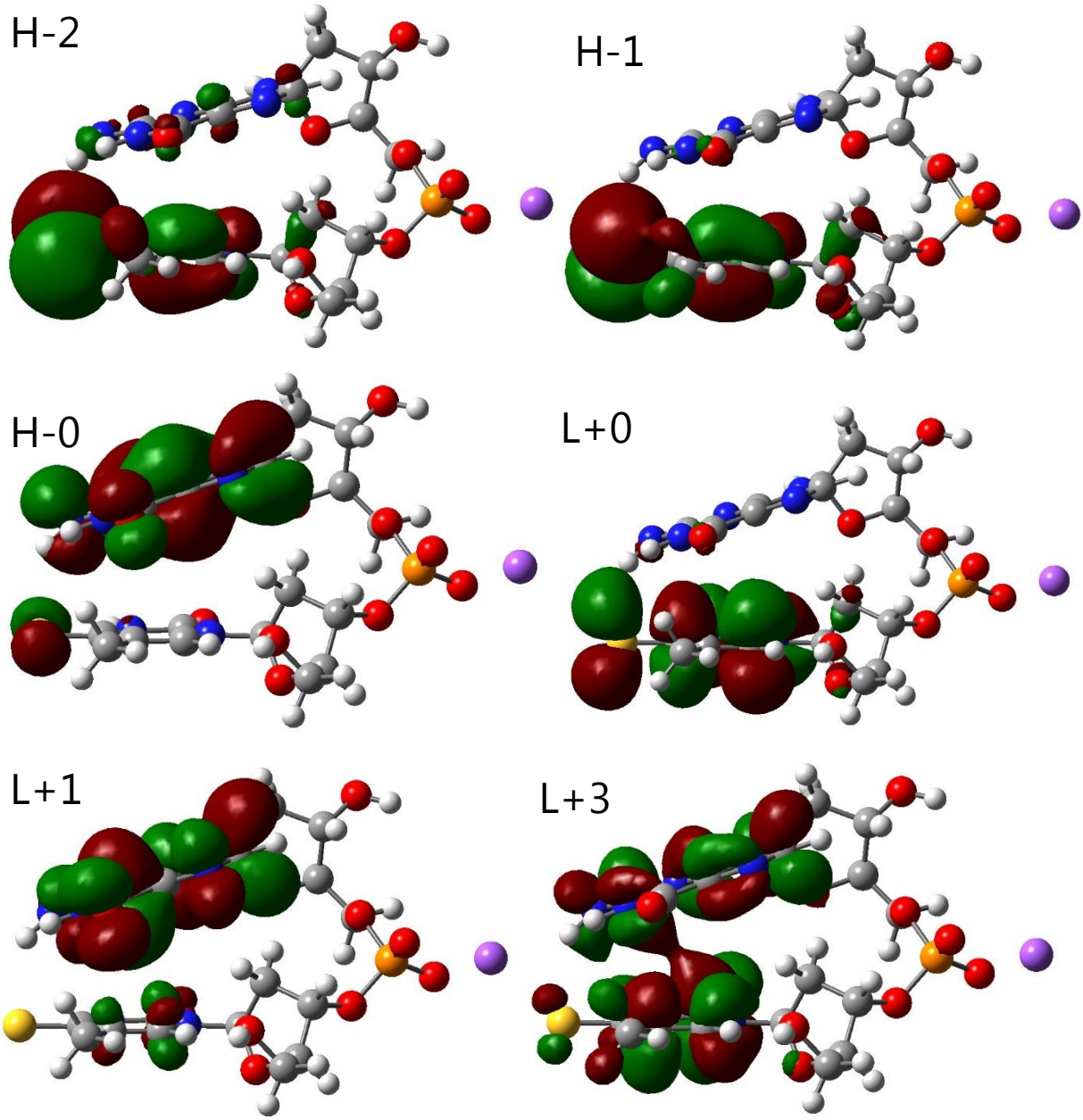
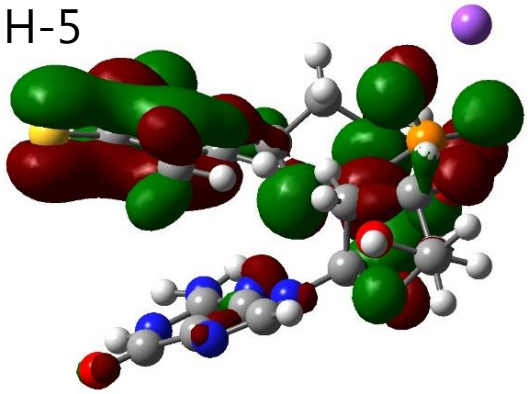


Figure 8. Primary Kohn-Sham orbitals of 4tThd-G in water.

H-5



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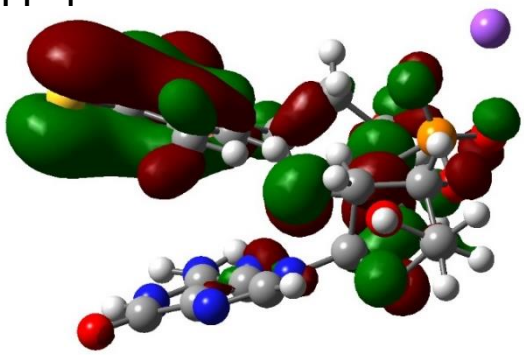

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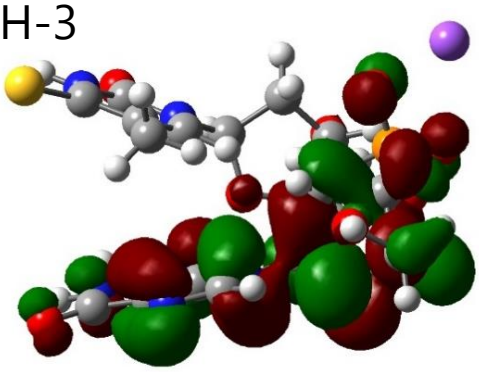

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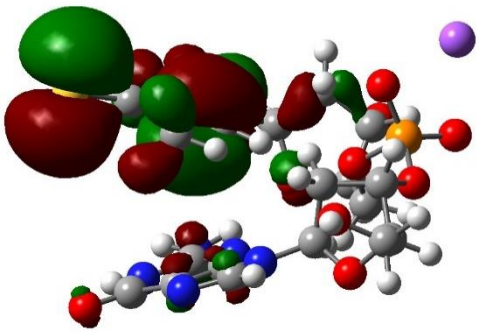
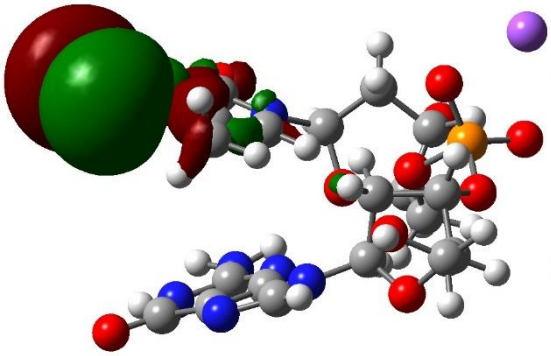
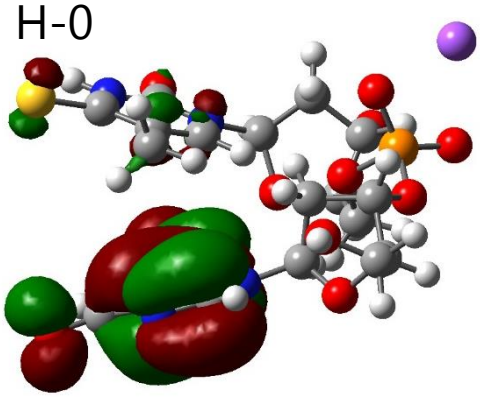


H-1



H-0



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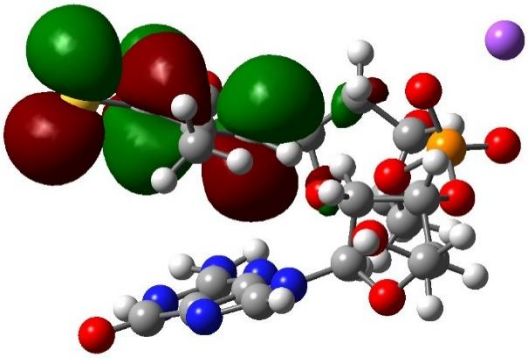

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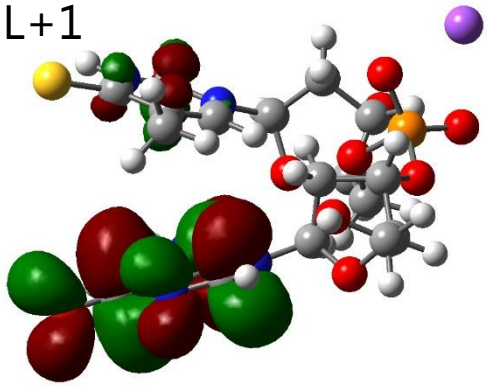

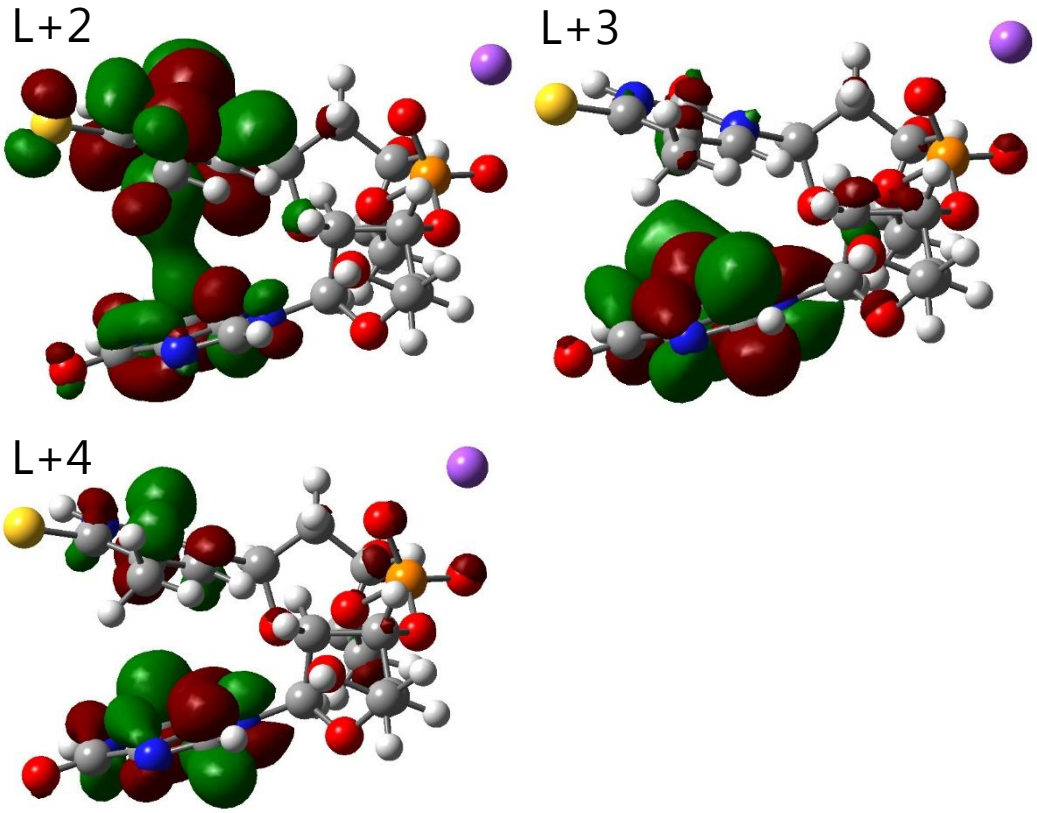

Figure 9. Primary Kohn-Sham orbitals of G-4tThd in water.

In Tables 1 and 2, the vertical excitation energies were calculated by first optimizing the ground-state of 4tThd in water and vacuum, respectively, using the M052X/6-31G(d) level of theory. The optimized geometries were then used to conduct vertical excitation energies calculations at the M052X/6-31G(d) level of theory. The values in Tables 3 and 4 were calculated following the same methodology except the molecule was optimized using the PBE0/6-31G(d) level of theory. In Tables 5 and 6, the vertical excitation energies were calculated using the PBE0/6-31+G(d) level of theory from ground state M052X/6-31G(d) optimizations in water and vacuum, respectively. The values in Tables 7 and 8 were calculated following the same methodology except the molecule was optimized using the PBE0/6-31G(d) level of theory.

The vertical excitation energies for 4tThd in Tables 5-8 were found to be in good agreement with values previously reported.[3] The calculations in tables 5-8 used the same functional as the one previously used for the 4tThd,[3] PBE0, however, the basis set used, 6-31+G(d), was smaller. When calculating vertical excitation energies with the PBE0 functional, the values from the ground state M052X/6-31G(d) optimizations (table 5 and 6) were very similar to the values from the ground state PBE0/6-31G(d) optimizations (table 7 and 8). For those calculations, a smaller basis set and different functional was used for ground state optimizations compared to the basis set and functional used for the calculations in reference 3. Even with a smaller basis set and different ground state optimizations, the vertical excitation energies were still found to be comparable when using PBE0 as the functional. However, when comparing the vertical excitation energies for 4tThd

calculated using the M052X functional, a larger deviation was found between those values and previously reported values.[3] This indicates that the vertical excitation energy calculations using the PBE0/6-31+G(d) level of theory are more accurate.

To predict how 4tThd behaves in DNA, vertical excitation energy calculations for dinucleotides containing 4-thiothymidine and a DNA base were conducted. Since the dinucleotides contained a negatively charged phosphate, a Na+ counter ion was added. In Tables 9-30, vertical excitation energies for 4tThd stacked with a DNA base were calculated using the M052X/6-31G(d) and PBE0/6-31+G(d) level of theory from ground state M052X/6-31G(d) optimizations. The M052X functional was used to optimize the geometries of the dinucleotides because it is a hybrid functional that takes into consideration the stacking interactions between the two bases. For the vertical excitation energy calculations using the M052X functional, 6-31G(d) was the largest basis set that would converge within the 36-hour time limit, while 6-31+G(d) was the largest basis set that would converge for the PBE0 functional. Due to the higher accuracy of the calculations using the PBE0 functional, discussion of the results will be referring to the vertical excitation energy calculations using the PBE0/6-31+G(d) level of theory from ground state M052X/6-31G(d) optimizations.

The character of the excited states was determined from analyzing the Kohn-Sham orbitals of the principal configuration interaction transitions. The most relevant Kohn-Sham orbitals for the dinucleotides in water are shown in Figures 2-9. The lowest-energy singlet and triplet states of 4tThd in water have approximately 16% ππ*and 84% nπ* character for the S_1 state, and 84% ππ*and 16% nπ* character for both the T_1 and S_2 state. The vertical excitation energies for the 4tThd in water are predicted to be 3.2281 (384 nm), 2.585 (480 nm), and 4.0683 eV (305 nm) for the S_1, T_1, and S_2 states, respectively, in good agreement with the results reported in reference 3. The vertical excitation energies for 4tThd in vacuum are predicted at 3.0236 (410 nm), 2.449 (506 nm), and 4.1996 eV (295 nm) for the S_1, T_1, and S_2 states, respectively. For 4tThd in water and vacuum, intersystem crossing can occur from the S_2 ππ* state to the T_2 nπ*. This agrees with the proposed mechanism in reference 3, in which the initial excited-state population in the S_2 state decays by ultrafast intersystem crossing to the triplet state.

For all of the dinucleotides, the S_1 state was found to come from 4tThd. In addition, the charge transfer states for all the dinucleotides show the electron density moving from the natural DNA base to 4tThd.

The vertical excitation energies for the T-4tThd dinucleotide in water are predicted at 3.1817, 4.0157, 2.5458, 2.8965, 3.2898, 3.6334, and 4.0118 eV for the S_1, S_2, T_1, T_2, T_3, T_4, and T_5 states, respectively. S_2 is a charge transfer, ππ* state with an oscillator strength of 0.028. Intersystem crossing can occur from the S_2 ππ* state to either the T_4 nπ* state or T_2 nπ* state or from the S_1 nπ* state to the T_1 ππ*state. The excited state population in the S_2 state should be more likely to decay to the T_4 state because of the smaller difference between their corresponding vertical excitation energies (0.3823 eV).

For the T-4tThd dinucleotide in vacuum, vertical excitation energies are predicted to be 2.9545, 3.9368, 4.055, 2.4278, 2.6203, 3.273, 3.6035, and 3.9368 eV for the S_1, S_2, S_3, T_1, T_2, T_3,

T_4, and T_5 states, respectively. Unlike when T-4tThd was in water, when in vacuum, S_2 is an $n\pi^*$ state instead of a $\pi\pi^*$ state. However, S_3 is a charge transfer, $\pi\pi^*$ state with an oscillator strength of 0.0073, similar to S_2 for T-4tThd in water. It is also observed that the S_2 and T_5 states are isoenergetic. Intersystem crossing can occur from the S_3 $\pi\pi^*$ state to either the T_2, T_4, or T_5 $n\pi^*$ state. Due to the small difference between the vertical excitation energies of S_3 and T_5 (0.1182 eV), the S_3 state should be more likely to decay to the T_5 state. Intersystem crossing can also occur from the S_2 $n\pi^*$ state to the T_3 or T_1 $\pi\pi^*$state or from the S_1 $n\pi^*$ state to the T_1 $\pi\pi^*$state.

The vertical excitation energies for the 4tThd-T dinucleotide in water are predicted at 3.1962, 4.0253, 4.1725, 2.5969, 2.9314, 3.2867, and 3.6049 eV for the S_1, S_2, S_3, T_1, T_2, T_3, and T_4 states, respectively. S_3 is a charge transfer, $\pi\pi^*$ state with an oscillator strength of 0.0133. Intersystem crossing can occur from the S_3 or S_2 $\pi\pi^*$ state to either the T_4 or T_2 $n\pi^*$ state or from the S_1 $n\pi^*$ state to the T_1 $\pi\pi^*$state. For the 4tThd-T dinucleotide in vacuum, vertical excitation energies are predicted to be 2.96, 4.0205, 4.1237, 2.4878, 2.6546, 3.2744, 3.5515, and 4.0205 eV for the S_1, S_2, S_3, T_1, T_2, T_3, T_4, and T_5 states, respectively. In addition to T-4tThd in vacuum, S_2 and T_5 of 4tThd-T in vacuum are also isoenergetic. Unlike 4tThd-T in water, in vacuum, there are no charge transfer states and S_2 is an $n\pi^*$ state instead of a $\pi\pi^*$ state. Intersystem crossing can occur from the S_3 $\pi\pi^*$ state to either the T_2 or T_5 $n\pi^*$ state. Due to the small difference between the vertical excitation energies of S_3 and T_5 (0.1032 eV), the S_3 state should be more likely to decay to the T_5 state. Intersystem crossing can also occur from the S_2 $n\pi^*$ state to the T_4, T_3, or T_1 $\pi\pi^*$state or from the S_1 $n\pi^*$ state to the T_1 $\pi\pi^*$state.

The vertical excitation energies for the 4tThd-A dinucleotide in water are predicted at 3.221, 3.9046, 4.0329, 2.5877, 2.9543, 3.4977, 3.5942, and 3.9281 eV for the S_1, S_2, S_3, T_1, T_2, T_3, T_4, and T_5 states, respectively. S_2 is a charge transfer, $\pi\pi^*$ state with an oscillator strength of 0.0036. Intersystem crossing can occur from the S_3 or S_2 $\pi\pi^*$ state to the T_2 $n\pi^*$ state or from the S_1 $n\pi^*$ state to the T_1 $\pi\pi^*$state. For the 4tThd-A dinucleotide in vacuum, vertical excitation energies are predicted to be 3.0088, 3.9356, 4.0208, 2.4462, 2.6945, 3.4658, 3.6029, and 3.9512 eV for the S_1, S_2, S_3, T_1, T_2, T_3, T_4, and T_5 states, respectively. In vacuum, S_2 is also a charge transfer, $\pi\pi^*$ state with an oscillator strength of 0.0017. Unlike when 4tThd-A was in water, when in vacuum, S_3 is an $n\pi^*$ state instead of a $\pi\pi^*$ state. Intersystem crossing can occur from the S_3 $n\pi^*$ state to the T_5, T_4, T_3, or T_1 $\pi\pi^*$ state, from the S_2 $\pi\pi^*$ state to the T_2 $\pi\pi^*$state, or from the S_1 $n\pi^*$ state to the T_1 $\pi\pi^*$state. When the dinucleotide is in water compared to in vacuum, the primary character of all the states, except for S_3, remain the same.

For the A-4tThd dinucleotide in water, vertical excitation energies are predicted to be 3.1726, 3.6431, 4.0451, 2.5222, 2.8945, 3.5059, 3.5898, and 3.7333 eV for the S_1, S_2, S_3, T_1, T_2, T_3, T_4, and T_5 states, respectively. S_2 is a charge transfer, $\pi\pi^*$ state with an oscillator strength of 0.032. Intersystem crossing can occur from the S_3 or S_2 $\pi\pi^*$ state to the T_2 $n\pi^*$ state or from the S_1 $n\pi^*$ state to the T_1 $\pi\pi^*$state. Due to the small difference between the vertical excitation energies of S_2 and T_2 (0.7486 eV), the S_2 state should be more likely to decay to the T_2 state. The vertical excitation energies for the A-4tThd in vacuum are predicted at 3.0363, 3.7026, 3.9896, 2.4398, 2.7184, 3.4738, 3.5713, and 3.7549 eV for the S_1, S_2, S_3, T_1, T_2, T_3, T_4, and T_5 states, respectively. S_2 is a charge transfer, $\pi\pi^*$ state with an oscillator strength of 0.0052. Unlike when A-4tThd was

in water, when in vacuum, S_3 is an nπ* state instead of a ππ* state. Intersystem crossing can occur from the S_3 nπ* state to the T_5, T_4, T_3, or T_1 ππ* state, from the S_2 ππ* state to the T_2 ππ*state, or from the S_1 nπ* state to the T_1 ππ*state. These same mechanisms are also predicted for 4tThd-A in vacuum.

The vertical excitation energies for the 4tThd-C dinucleotide in water are predicted at 3.2327, 4.0535, 4.2386, 2.5999, 2.963, 3.5468, 3.6082, and 4.2198 eV for the S_1, S_2, S_3, T_1, T_2, T_3, T_4, and T_5 states, respectively. S_3 is a charge transfer, ππ* state with an oscillator strength of 0.0158. Intersystem crossing can occur from the S_3 or S_2 ππ* state to the T_2 nπ* state or from the S_1 nπ* state to the T_1 ππ*state. For the 4tThd-C dinucleotide in vacuum, vertical excitation energies are predicted to be 3.0126, 3.8893, 4.1253, 2.4966, 2.7028, 3.4937, 3.5139, and 3.9322 eV for the S_1, S_2, S_3, T_1, T_2, T_3, T_4, and T_5 states, respectively. S_2 is a charge transfer, ππ* state with an oscillator strength of 0.0211. Unlike when 4tThd-C was in water, when in vacuum, S_3 is an nπ* state instead of a ππ* state. Intersystem crossing can occur from the S_3 nπ* state to the T_5, T_4, T_3, or T_1 ππ* state, from the S_2 ππ* state to the T_2 ππ*state, or from the S_1 nπ* state to the T_1 ππ*state. These same mechanisms are also predicted for 4tThd-A and A-4tThd in vacuum.

For the C-4tThd dinucleotide in water, vertical excitation energies are predicted to be 3.182, 3.9041, 4.0542, 2.5621, 2.9063, 3.5079, 3.5683, and 3.9878 eV for the S_1, S_2, S_3, T_1, T_2, T_3, T_4, and T_5 states, respectively. S_2 is a charge transfer, ππ* state with an oscillator strength of 0.0841. Intersystem crossing can occur from the S_3 or S_2 ππ* state to the T_2 nπ* state or from the S_1 nπ* state to the T_1 ππ*state. Due to the small difference between the vertical excitation energies of S_2 and T_2 (0.9978 eV), the S_2 state should be more likely to decay to the T_2 state. The vertical excitation energies for the C-4tThd in vacuum are predicted at 3.0663, 3.7168, 4.0668, 2.506, 2.773, 3.3663, 3.4988, and 3.8537 eV for the S_1, S_2, S_3, T_1, T_2, T_3, T_4, and T_5 states, respectively. S_2 is a charge transfer, ππ* state with an oscillator strength of 0.0143. Intersystem crossing can occur from the S_3 or S_2 ππ* state to the T_2 nπ* state or from the S_1 nπ* state to the T_1 ππ*state. Due to the small difference between the vertical excitation energies of S_2 and T_2 (0.9438 eV), the S_2 state should be more likely to decay to the T_2 state. The mechanisms for the same dinucleotide remain the same when in water or vacuum.

The vertical excitation energies for the 4tThd-G dinucleotide in water are predicted at 3.2222, 3.4733, 4.0132, 2.6293, 2.9807, 3.3944, 3.662, and 3.7428 eV for the S_1, S_2, S_3, T_1, T_2, T_3, T_4, and T_5 states, respectively. S_2 is a charge transfer, ππ* state with an oscillator strength of 0.0029. Intersystem crossing can occur from the S_3 or S_2 ππ* state to the T_2 nπ* state, from the S_3 ππ* state to the T_4 nπ* state, or from S_1 nπ* state to the T_1 ππ*state. Due to the small difference between the vertical excitation energies of S_2 and T_2 (0.4926 eV) and of S_3 and T_4 (0.3512 eV), the S_2 state should be more likely to decay to the T_2 state while the S_3 state should be more likely to decay to the T_4 state. For the 4tThd-G dinucleotide in vacuum, vertical excitation energies are predicted to be 3.1571, 3.3808, 4.0222, 2.5823, 2.9042, 3.3153, 3.6164, and 3.7805 eV for the S_1, S_2, S_3, T_1, T_2, T_3, T_4, and T_5 states, respectively. S_3 and S_2 are charge transfer, ππ* states with oscillator strengths of 0.0002 and 0.0015, respectively. Intersystem crossing can occur from the S_3 ππ* state to the T_4 nπ* state or from the S_1 nπ* state to the T_2 or T_1 ππ*state.

For the G-4tThd dinucleotide in water, vertical excitation energies are predicted to be 3.2028, 3.4975, 4.082, 2.5621, 2.9248, 3.4683, 3.6582, and 3.6997 eV for the S_1, S_2, S_3, T_1, T_2, T_3, T_4, and T_5 states, respectively. S_2 is a charge transfer, $\pi\pi^*$ state with an oscillator strength of 0.0119. Intersystem crossing can occur from the S_3 or S_2 $\pi\pi^*$ state to the T_2 $n\pi^*$ state or from the S_1 $n\pi^*$ state to the T_1 $\pi\pi^*$state. Due to the small difference between the vertical excitation energies of S_2 and T_2 (0.5727 eV), the S_2 state should be more likely to decay to the T_2 state. The vertical excitation energies for the G-4tThd in vacuum are predicted at 2.9779, 3.3992, 3.7118, 2.4077, 2.6385, 3.3813, 3.6274, and 3.7112 eV for the S_1, S_2, S_3, T_1, T_2, T_3, T_4, and T_5 states, respectively. S_3 and S_2 are charge transfer, $\pi\pi^*$ states with oscillator strengths of 0 and 0.0046, respectively. Intersystem crossing can occur from the S_3 or S_2 $\pi\pi^*$ state to the T_2 $n\pi^*$ state or from the S_1 $n\pi^*$ state to the T_1 $\pi\pi^*$state. Due to the small difference between the vertical excitation energies of S_2 and T_2 (0.7607 eV), the S_2 state should be more likely to decay to the T_2 state. The mechanisms for the same dinucleotide remain the same when in water or vacuum.

4. Conclusions

When comparing the dinucleotides, it can be seen that some of the dinucleotides share the same proposed kinetic mechanisms. Similar to the 4tThd monomer mechanisms, a majority of the proposed kinetic mechanisms for the dinucleotides consist of intersystem crossing from the S_3 or S_2 $\pi\pi^*$ state to a triplet state, usually T_2. Intersystem crossing can occur from the S_3 or S_2 $\pi\pi^*$ state to the T_2 $n\pi^*$ state or from the S_1 $n\pi^*$ state to the T_1 $\pi\pi^*$state for A-4tThd in water, C-4tThd in both water and vacuum, G-4tThd in both water and vacuum, 4tThd-A in water, and 4tThd-C in water. A-4tThd in vacuum, 4tThd-A n vacuum, and 4tThd-C in vacuum also share the same proposed kinetic mechanisms. Electron transfer pathways between the DNA base and 4tThd seem to be in competition with intersystem crossing to populate the T_1 state of 4tThd. For A-4tThd, 4tThd-A, and 4tThd-C, changing the solvent from water to vacuum results in the S_3 state changing from $\pi\pi^*$ to $n\pi^*$, however the first two singlet states remain the same. The primary character for the first three singlet states are the same among the dinucleotides with the canonical DNA base in the 5' position in water. Primary character of states as well as the proposed kinetic mechanisms show to be more consistent among the dinucleotides in water than in vacuum. Further investigations are needed in order to better understand how 4tThd behaves in DNA.

5. Personal Experience

The Project SEED program has provided me with opportunities that I would not have been able to receive without it. With the guidance of Dr. Crespo and his graduate students, I have been able to learn so much about photochemistry. This summer, I performed computational chemistry to predict the order electronic states and likely kinetic mechanisms for 4tThd-containing dinucleotides. Analyzing this data at the end of the summer is the part that I enjoyed the most. I was able to learn about what all of the numbers mean and how they helped in describing the behavior of 4tThd. I am glad that not only was I able to learn about what it's like to be a chemist, I was also able to help in the process of researching a potential photodynamic therapy agent that may help others in the future.

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