Received: 14 October 2015,

Revised: 17 December 2015,

(wileyonlinelibrary.com) DOI 10.1002/bio.3095

Published online in Wiley Online Library

Properties, theoretical study and crystal structure of 3-benzothiazole-9-ethyl carbazole

Accepted: 21 December 2015

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ABSTRACT: The title compound of 3-benzothiazole-9-ethyl carbazole was synthesized by the reaction of 3-aldehyde-9-ethyl carbazole and 2-aminothiophenol. The compound was characterized by ¹H nuclear magnetic resonance (NMR) and mass spectrometry (MS). Its crystal structure was obtained and determined by single crystal X-ray diffraction. The results showed that the crystal belongs to the orthorhombic crystal system and the cell parameters of space group P2(1)2(1)2(1) were a = 5.6626 (12) Å, b = 12.606 (3) Å, c = 22.639 (5) Å, $\alpha = 90^{\circ}$, $\beta = 90^{\circ}$, $\gamma = 90^{\circ}$, V = 1616.0 (6) Å³, Z = 4, Dc = 1.350 mg/m³. The UV-vis and fluorescence spectra were also studied preliminarily. The fluorescence spectra of the title compound with bovine serum albumin (BSA) showed that BSA could be marked with the compound and the stability constant between them was 0.82×10^7 M⁻¹. Meanwhile, the crystal and molecule were theoretically surveyed by density functional tight-binding (DFTB). The results showed that there was an orbital overlap for lowest unoccupied molecular orbital (LUMO) between the neighbouring molecules for the crystal, which is different from the molecule structure. It was also showed that the crystal structure is a non-conductor. Copyright © 2016 John Wiley & Sons, Ltd.

Keywords: 3-benzothiazole-9-ethyl carbazole; spectra properties; crystal structure; theoretical study; stability constant

Introduction

The carbazole nucleus can be easily functionalized at its 3, 6 and 9 positions and covalently linked with other molecular groups. Up to now, carbazole derivatives have been widely utilized as a functional building block in the fabrication of the organic medicine, pesticide, material, etc. because of their excellent solubility, stability, excellent hole-transporting capability (the electron-donating properties) and biological activity (1–6). In addition, carbazole and its derivatives can be used as a strong electro-donating chromophore, which is the main structure of fluorescent dyes.

Recently, serious novel carbazole derivatives have been synthesized as efficient emitting materials in organic devices.

Benzothiazole derivatives are widely used and are of good potential in the preparation of organic photoconductors and other fields because of the thick heterocyclic system and electronacceptor moieties and strong fluorescence effect (7,8). For this reason, using benzothiazole ring as the electron acceptors (A) and carbazole ring as the electron donor (D), researchers have combined carbazole with benzothiazole to prepare many carbazole–benzothiazole derivatives with different fluorescent properties (9–13). Because of their large electron delocalization lengths, they are used to experienced intramolecular charge transfer (ITC) from A to D of the molecules.

So, it is very important for the applications of carbazoles as optical materials to understand the relationship between optical properties and molecule structure, especially the effect of π -conjugated structure on the optical properties. In order to understand the energy, geometry structure and properties of carbazole–benzothiazole derivative in detail, we report the crystal structures, theoretical calculation and the spectra properties of 3-benzothiazole-9-ethyl carbazole in this article.

Experimental

Materials and methods

All the reagents and solvents were commercial and used without further purification. Melting points were measured by using a melting point apparatus made by Beijing Tech. Instrument Co. UV–Vis absorption spectra were recorded on a Perkin Elmer LAMBDA 35 spectrometer. Fluorescence spectra were recorded on a Hitachi F-7000 spectrometer. Mass spectral analyses were obtained using an electrospray ionization (ESI) mass spectrometer on MICROMASS QUATTRO micro API (Waters Corporation, America). ¹H NMR spectra were recorded on a Bruker (400 MHz) spectrometer. Chemical shifts were reported in parts per million (ppm) downfield from

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Abbreviations: BSA, bovine serum albumin; DFTB, density functional tightbinding; DOS, Disk Operation System; HOMO, highest occupied molecular orbital; HPLC, high pressure liquid chromatography; LUMO, lowest unoccupied molecular orbital; TICT, twisted intramolecular charge transfer. tetramethylsilane (TMS) using CDCl₃ or DMSO-d₆ as a solvent. The chemical purity is analyzed by high pressure liquid chromatography (HPLC) Lab Alliance PC4006A from the USA (flow rate: 1.0 mL/min; 4.6 mm × 250 mm, 5 μ m, C18 reversed-phase column).

Syntheses of 3-benzothiazole-9-ethyl carbazole

The title compound was synthesized according to the reference (14) and the synthetic route was shown in Scheme 1.

- The detailed synthetic processes are shown below:
- 15 mL of anhydrous N,N-dimethylformamide (DMF) was added slowly to 15.202 g of POCl₃ (0.1 mol) under stirring in an ice bath at room temperature until the solution became red. Then 3.155 g of 9-ethylcarbazole (16 mmol) dissolved in 25 mL 1,2-dichloroethane were added. The mixture was heated to 353 K and kept reacting at this temperature. The reaction was monitored by thin layer chromatography (TCL) (ethyl acetate: petroleum ether =1:10) and after 8 h, the mixture was cooled to room temperature. The resultant was poured into a mixture of ice-water and followed by extraction with dichloromethane $(3 \times 100 \text{ mL})$. The resultant dichloromethane solution was washed three times with 100 mL of water and dried over anhydrous magnesium sulphate. Afterwards the solvent was evaporated under reduced pressure and the residue was dissolved in a minimum amount of dichloromethane and then purified by silica-gel column chromatography to get 3-formyl-9-ethyl carbazole. M. p.: 84-86 °C, yield: 50%. Purity: 99.8% (HPLC, a solution of 80% CH₃OH and 20% H₂O was chosen as the mobile phase).
- 560 mg (2.5 mmol) of 3-formyl-9-ethyl carbazole were dissolvedin 25 mL of DMSO and then 0.27 mL (2.5 mmol) of

2-aminobenzenethiol was added. The mixture was slowly heated to 333 K. The reaction was monitored by TCL (ethyl acetate: petroleum ether =1:10) and after 4 h, the mixture was cooled to room temperature. The resultant was poured into a mixture of ice-water and faint yellow precipitate was obtained. After filtration, the crude product was purified by silica-gel column chromatography to get the title compound. M. p.: 142-145 °C, yield: 80%. Purity: 99.7% (HPLC, solution of 80% CH₃OH and 20% H₂O was chosen as the mobile phase). The brown single crystal were obtained by evaporation for petroleum ether and ethyl acetate (4:1 = v/v) after a few days. ¹H NMR (400 MHz,CDCl₃) δ:, 1.45–1.49 (t, J = 7.20 Hz, 3 H, 4.37–4.43 (m, 2 H), 7.28–7.38 (m, 2 H), 7.43–7.54 (m, 4 H), 7.91 (d, J = 8.00 Hz, 1 H), 8.08 (d, J = 8.00 Hz, 1 H), 8.21 (d, J = 7.60 Hz, 2 H), 8.86 (s, 1 H). ESI-MS (m/z): 329.9 [M⁺+1].

Structure determination and refinement

A brown crystal of the title compound was mounted on the bottom of a glass fiber. X-ray diffraction data were collected on a Bruker Smart-1000 CCD diffractometer equipped with a graphite-monochromatized MoK_a radiation (0.7170 Å) by using an Ω/ψ scan mode in the range of $1.80 \le \theta \le 27.88^{\circ}$ ($-7 \le h \le 7$, $-16 \le k \le 15$, $-29 \le l \le 29$) at 113 (2) K. In total, 16 575 reflections were collected with 3858 unique ones ($R_{int} = 0.0434$). The structure was solved by direct methods and refined on F² by full-matrix least-squares procedure with SHELXS-97 and SHELXL-97 (15a). The crystal data and other structure refinement parameters of the title compound (15b) are listed in Table 1.



Scheme 1. The synthetic route of 3-benzothiazole-9-ethyl carbazole.

Table 1. Crystal structure data for the compound				
Empirical formula	C21 H16 N2 S	Formula weight	328.42	
Temperature	113 (2) K	Wavelength	0.71073 A	
Crystal system	Orthorhombic	Space group	P2 (1)2 (1)2 (1)	
a (Å)	5.6626 (12)	b (Å)	12.606 (3)	
c (Å)	22.639 (5)	α (°)	90	
β (°)	90	γ (°)	90	
Volume (Å ³)	1616.0 (6)	Z	4	
Calculated density (mg/m ³)	1.350	Absorption coefficient (mm ⁻¹)	0.204	
F (000)	688	Crystal size (mm)	$0.20 \times 0.18 \times 0.12$	
θ range for data collection (°)	1.80 to 27.88	Limiting indices	$-7 \le h \le 7, -16 \le k \le 15,$	
			$-29 \le I \le 29.$	
Reflections collected/unique	16575/3858 [R(int) = 0.0434]	Refinement method	Full-matrix least-squares on F ²	
Data/restraints/parameters	3858/0/219	Goodness-of-fit on F ²	0.970	
Final R indices $[I > 2\sigma(I)]$	$R_1 = 0.0291, wR_2 = 0.0750$	R indices (all data)	R1 = 0.0311, wR2 = 0.0770	
Max. and min. Transmission	0.9760 and 0.9604	Largest diff. Peak and hole (e $Å^{-3}$)	0.194 and -0.210	



Results and discussion

Crystal structure

The selected bond lengths and bond angles are listed in Table 2, and the selected torsion angles for non-hydrogen atoms are given in Table 3. The molecular structure of the title compound is shown in Fig. 1. The packing diagram of the title compound in a unit cell is shown in Fig. 2.

The angles of C4-C5-C6-N1 (-178.54 (12)°), S1-C1-C2-C3 (179.02 (10)°), C7-S1-C1-C2 (-178.15 (13)°), C7-N1-C6-C5 (179.42 (12)°) show that C4,C5,C6,C1,C2,C3,C7,N1,S1 are in the same plane. The angles of N1-C7-C8-C9 (-2.18 (18)°), S(1)-C(7)-C(8)-C(19) (-2.92 (16)°), C(6)-N (1)-C(7)-C(8) (-179.33 (11)°), C(1)-S(1)-C(7)-C(8) (179.33 (10)°) show that there is an angle of 2–3° between the thiazole ring and the carbazole ring. The angle of C(9)-C(10)-C(17)-N(2) is 177.81 (11), C(16)-N(2)-C(17)-C(18) is 177.49 (13), C(12)-C(11)-C(16)-N(2) is -179.16 (11) respectively, indicating that C9, C10, C17, N2, C16, C11 and C12 are in a plane.



Figure 1. X-ray crystal structure of the title compound.

So, the two aromatic rings are of an almost coplanar structure linked by the single bond from the selected torsion angles. The structure elucidation was achieved by X-ray diffraction, and proved cleanly that the reaction occurs to form the title compound.

Theoretical calculation

In this study, the energy and geometry structure of the crystal and molecule structure was calculated by density functional tightbinding (DFTB) method and complemented by an empirical London dispersion energy term (DFTB-D) method. The structures

Table 2. Selected bond length(Å) and bond angles (°)					
Bond	Dist.	Bond	Dist.	Bond	Dist.
S (1)-C (1)	1.7304 (14)	N (2)-C (16)	1.3881 (18)	C (3)-C (4)	1.398 (2)
S (1)-C (7)	1.7577 (13)	N (2)-C (20)	1.4607 (17)	C (8)-C (19)	1.4112 (18)
N (1)-C (7)	1.3002 (17)	C (1)-C (2)	1.3982 (18)	C (10)-C (11)	1.4516 (17)
N (1)-C (6)	1.3921 (16)	C (1)-C (6)	1.4034 (18)	C (20)-H(20 A)	0.9900
N (2)-C (17)	1.3802 (16)	C (2)-C (3)	1.381 (2)	C (20)-C (21)	1.518 (2)
C (7)-C (8)	1.4687 (17)	C (8)-C (9)	1.3963 (17)	C (21)-H(21 A)	0.9800
Angle	(°)	Angle	(°)	Angle	(°)
C (1)-S (1)-C (7)	89.08 (6)	C (7) -N (1)-C (6)	110.60 (11)	C (17)-N (2)-C (16)	108.54 (11)
C (17)-N (2)-C (20)	125.62 (12)	C (16)-N (2)-C (20)	125.40 (11)	C (2)-C (1)-C (6)	121.54 (12)
C (2)-C (1)-S (1)	128.93 (11)	C (6)-C (1)-S (1)	109.51 (9)	N (1)-C (6)-C (5)	125.19 (12)
N (1)-C (6)-C (1)	115.15 (12)	C (8)-C (7)-S (1)	119.86 (9)	N (1)-C (7)-C (8)	124.49 (11)
N (1)-C (7)-S (1)	115.65 (10)	C (19)-C (8)-C (7)	120.20 (11)	N (2)-C (16)-C (15)	128.44 (14)
C (9)-C (8)-C (7)	119.85 (11)	C (17)-C (10)-C (11)	106.22 (11)	N (2)-C (17)-C (18)	128.71 (13)
C (9)-C (10)-C (11)	134.23 (13)	C (16)-C (11)-C (10)	106.52 (12)	N (2)-C (20)-C (21)	111.48 (12)
C (12)-C (11)-C (10)	133.97 (13)	N (2)-C (16)-C (11)	109.36 (12)	N (2)-C (17)-C (10)	109.36 (11)

Table 3. Selected torsion angles (°)

Angle	(°)	Angle	(°)
C (7)-S (1)-C (1)-C (2)	-178.15 (13)	C (7)-S (1)-C (1)-C (6)	0.39 (10)
C (7)-N (1)-C (6)-C (5)	179.42 (12)	C (1)-S (1)-C (7)-C (8)	179.33 (10)
C (6)-N (1)-C (7)-C (8)	-179.33 (11)	S (1)-C (7)-C (8)-C (9)	177.93 (9)
N (1)-C (7)-C (8)-C (9)	-2.18 (18)	S (1)-C (7)-C (8)-C (19)	-2.92 (16)
N (1)-C (7)-C (8)-C (19)	176.97 (12)	C (8)-C (9)-C (10)-C (11)	175.53 (13)
C (7)-C (8)-C (9)-C (10)	-178.86 (11)	C (17)-C (10)-C (11)-C (12)	178.54 (14)
C (9)-C (10)-C (11)-C (12)	1.3 (2)	C (16)-C (11)-C (12)-C (13)	1.89 (19)
C (9)-C (10)-C (11)-C (16)	-176.89 (14)	C (17)-N (2)-C (16)-C (15)	-175.49 (13)
C (10)-C (11)-C (12)-C (13)	-176.06 (14)	C (20)-N (2)-C (16)-C (11)	-172.05 (12)
C (20)-N (2)-C (16)-C (15)	11.7 (2)	C (12)-C (11)-C (16)-N (2)	-179.16 (11)
C (14)-C (15)-C (16)-N (2)	177.27 (13)	C (10)-C (11)-C (16)-C (15)	175.82 (12)
C (12)-C (11)-C (16)-C (15)	-2.65 (19)	C (20)-N (2)-C (17)-C (18)	-9.7 (2)
C (16)-N (2)-C (17)-C (18)	177.49 (13)	C (9)-C (10)-C (17)-N (2)	177.81 (11)
C (20)-N (2)-C (17)-C (10)	172.29 (12)	N (2)-C (17)-C (18)-C (19)	-176.16 (13)
C (11)-C (10)-C (17)-C (18)	-178.09 (12)	C (17)-N (2)-C (20)-C (21)	-89.40 (17)
C (7)-C (8)-C (19)-C (18)	-179.86 (12)	C (16)-N (2)-C (20)-C (21)	82.19 (16)

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Figure 2. A packing diagram for the crystal compound.

were optimized using the conjugate gradient algorithm, and the force converge criterion and the charge convergence criterion were set to 10^{-4} a.u. and 10^{-5} electrons, respectively. All the calculations were performed using the DFTB+ 1.2 program (16–21). To make a comparison, we optimized both the crystal of 3-benzothiazole-9-ethyl carbazole and the molecular structure in gas phase. For the crystal, the lattice parameters were set to be 20.8470 × 8.4679 × 20.8720 for the lengths. The crystal cell and single molecule structures are shown in Fig. 3.

Side view and dihedral angles of conformer. The side view and dihedral angles of conformer of 3-benzothiazole-9-ethyl carbazole in crystal and gas phase were also calculated (Fig. 4). We can see that the structure torsion occurred when the molecule formed crystal structure. The bond angles and dihedral angle changed. The torsional energy curves indicate that the dihedral angle of the molecule is more stable for $\theta = 179.9^\circ$, which is the same

as the data reported previously (21). But for the crystal, the $\theta = 177.3^{\circ}$, indicating that there is a large deformation and a distortion between the benzothiazole ring and carbazole ring to form a stable interaction. The results also indicate that the crystal structure is almost planar.

Frontier molecular orbitals. The frontier molecular orbitals, including the highest occupied molecular orbital (HOMO) and the lowest unoccupied molecular orbital (LUMO), can provide a reasonable qualitative indication of the excitation properties and the ability of electron or hole transport. Information about the HOMO and LUMO distributions of the crystal and molecule in gas phase respectively are shown in Fig. 5.

For 3-benzothiazole-9-ethyl carbazole in gas phase, we found that the HOMO remains on the carbazole ring moiety with similar distributions to that of previous theoretical studies of dipolar molecules (11, 22), while LUMO is localized on the benzothiazole ring and little on the $-C_2H5$ group, which indicated that the electron was transferred from the $-C_2H5$ group to the carbazole ring. For the crystal, The HOMO is similar to that of single molecule, while there is an orbital overlap for LUMO between the neighbouring molecules, meaning that there is a significant interaction between them.

Net charge. Meanwhile, the net charge of the molecule and the crystal structure were also calculated and some important atom and their net charges are shown in Table 4.

It is seen that when the molecule was formed to be crystral structure, the net charges of the atoms changed. For example, the net charges of atoms 1, 3, 4, 13 and 14 on benzothiazole decreased, which indicated that the electron transfer occurred. The title charges of the atoms on the benzothiazole rings (except H) is -0.40359 in the molecule and -0.41758 in the crystal structure, title charges of the atoms on the carbazole rings (except H) in the molecule is -0.58681 and -0.64787 in the crystal structure, which indicated that the negative charges on the both ring (except H) increased in the crystal structure.

The density of states of the crystal. The Disk Operation System (DOS) of the crystal was also calculated and is shown in Fig. 6. The energy shown by the dashed line is the Fermi energy.

We can see that there is a wide band gap ($\Delta E = 1.6 \text{ eV}$), which indicated that the crystal structure is a non-conductor. The two peaks of the wide pseudogap indicated strong covalent bonding in the crystal molecule.



Figure 3. Optimized structures of the ground state of (a) crystal cell; and (b) single molecule.

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Figure 4. Side view, bond angles and dihedral angle of (a) crystal; and (b) molecule.



Figure 5. (a) HOMO and (b) LUMO diagrams of the crystal.

Table 4. Net charges of some atoms				
Atom	Net charge in molecule	Net charge in crystal structure	Change of charge	
1	-0.11215019	-0.09103415	Decrease	
2	-0.30231969	-0.30805490	Increase	
3	-0.01599018	-0.00580141	Decrease	
4	0.02536549	0.01933700	Decrease	
5	-0.08646399	-0.09094260	Increase	
7	-0.09100618	-0.10178281	Increase	
9	-0.07775094	-0.08726501	Increase	
11	-0.11444038	-0.11740788	Increase	
13	0.16165198	0.15879021	Decrease	
14	0.20951352	0.20658310	Decrease	
15	-0.01399070	-0.01751352	Increase	
16	-0.07273426	-0.08511343	Increase	
18	-0.04184887	-0.05033290	Increase	
19	-0.03812864	-0.04781313	Increase	
20	-0.06855822	-0.08047183	Increase	
22	-0.10357274	-0.11279878	Increase	
24	-0.06554401	-0.07901990	Increase	
26	-0.14078787	-0.14320401	Increase	
28	0.08932942	0.08640278	Decrease	
29	0.09239369	0.09383171	Increase	
30	-0.14131108	-0.13192956	Decrease	
32	-0.08205945	-0.07990573	Decrease	

Spectral properties

Spectral properties in different solvents. The UV–Vis and fluorescence spectra were measured. The sample was measured at room temperature in the range of 300–400 nm in the UV–Vis spectrum and the fluorescence spectrum was excited at 295 nm in the range of 350–550 nm

The UV–Vis and fluorescence spectra in different solvents were studied and the results are shown in Fig. 7 and Table 5.

The compound has two absorption peaks at ca. 307 nm and ca. 345 nm respectively in different solvents. The peak at ca. 307 nm is originated from the π - π * transition of the conjugated system in the carbazole moiety and the peak is at ca. 345 nm from the π donator-acceptor structure formed by the benzothiadiazole moiety.

The absorption peak shifted blue and the absorption intensity enhanced with the higher polarity of the solvent. The energy of the antibonding orbital and bonding orbital of the molecular both reduced with the polarity of the solvent. Because the polarity of the ground state is low, the energy of bonding orbital reduced less, resulting in the energy difference between the antibonding orbital and bonding orbital enlarged and the absorbing peak shifted blue.

Figure 7(b) and Table 5 showed that compound has different maximum emission wavelength (λ_{em}) in different solvents, and it obtained the largest λ_{em} in protonic solvent (methanol). In the aprotic solvents, it has the largest λ_{em} in DMSO, which is because DMSO has the maximum polarity. The fluorescence peak shifted

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red and the fluorescence intensity reduced with the higher polarity. We further explored the relationship between dipole moment of the solvents and λ_{em} . We can see that the λ_{em} and Stokes shift both increased with the increasing dipole moment and dielectric constant. The possible reason can be explained by twisted intramolecular charge transfer (TICT). The solvent with the larger polarity could result in the reduction of energy difference between excited state and ground state, which as a result led to a red-shift in the compound (22–26).

Spectral properties of compound 6 at different pH. The UV-vis and fluorescence spectra were also studied with the concentration of 10^{-6} M in aqueous solution with different pH and the results are shown in Fig. 8.

From Fig. 8, we can see that the absorbing intensity increased until the pH was 7 and decreased after that. The fluorescence intensity increased with the increases of the pH value, which is different from absorbing intensity. There is always a λ_{em} at ca. 437 nm and λ_{max} at ca. 355 nm regardless of the pH value of the solutions. Interestingly, a new peak was observed at ca. 375 nm. The intensity ratio of peaks at ca. 355 nm and ca. 375 increased when the pH value increased. The possible explanation is that under acid conditions, there is protonation of N on the benzothiazole ring. With the decrease in the pH value, there is more protonation occurring, resulting in the relative intensity increase at ca. 375 nm.

Table 5. The solvent properties and fluorescent characters of compounds					
Solvent	Methanol	Ethanol	DCM	DMF	DMSO
nD ^{20a}	1.33	1.36	1.42	1.43	1.48
μ ^b	1.70	1.69	1.60	3.82	3.96
ε ^c	32.7	24.5	9.08	38.3	47.2
сР ^d	0.59	1.20	0.44	0.92	2.00
λ max/nm	345	344	345	345	341
λ em/nm	424	419	410	417	421
Stokes shift	79	75	65	72	80
^a Refractive index. ^b Dipole moment. ^c Dielectric constant.					

Labeling of bovine serum albumin (BSA) protein. BSA was added into the compound solutions at the concentration of 10^{-5} mol/L to give compound–BSA solutions with protein concentrations of 0 μ M, 0.18 μ M, 0.54 μ M, 0.72 μ M, 1.44 μ M or 1.62 μ M, the fluorescence emission spectra results are shown in Fig. 9.

The fluorescence intensity increases with increasing BSA concentration, accompanied by a blue shift of the λ_{em} . The increase in intensity is because the hydrogen bond between the molecule and BSA (the amino and thiol) made the molecule inlayed into the helical structure of BSA. The protection from the BSA reduced the polarization of the solvent, and made a fluorescence enhancement. With the increase of protein concentration, the α -helix structure changed from a stretching state to a folding state and steric hindrance to the molecule made it easier to form a rigid flat structure (27). The blue shift of the λ_{em} maybe caused by the reduced ground state energy of the title compound caused by the interaction between the compound and protein.

To quantify the interaction between the compounds and BSA, we determined the stability constants (K_s). The stability constants (K_s) were determined according to a method in the literature (28,29). The interaction between the dye (D) and the protein (P) expressed as D + P = DP can be quantified by the stability constant K_s:

$$K_s = \frac{[DP]}{[D][P]}$$



Figure 7. The UV–Vis (a) and fluorescence (b) spectra of the title compound with the concentration of 10^{-6} M in MeOH.





Figure 8. The UV–Vis (a) and fluorescence (b) spectra of the title compound with the concentration of 10⁻⁶ M in water at different pH.



Figure 9. Fluorescence of the compound–BSA solution with different BSA concentrations.

K_s was determined according to the equation:

$$\frac{1}{F} = \frac{1}{k[D]} + \left(\frac{1}{k[D]Ks}\right)\frac{1}{[P]}$$

where F is the measured fluorescence intensity of the dye in the protein mixture; k is an instrumentation dependent constant, and all other variables are as previously defined. Thus, a plot of 1/F(y) versus 1/[P](x) should yield a straight line, with $K_s = -x_0$.

According to this described method, the K_s between BSA and the dye was $8.2\times10^6~(M^{-1})$, which showed that the BSA can be marked with the dye in the paper.

Conclusion

The compound of 3-benzothiazole-9-ethyl carbazole was synthesized and characterized by ¹H NMR and MS and single crystal X-ray diffraction. The results showed that the crystal belongs to the orthorhombic crystal system and the crystal structure was almost planar. Meanwhile, the crystal and molecule were theoretically surveyed and the results showed that there was a distortion between the two rings and an orbital overlap for LUMO between the neighbouring molecules for the crystal, which were different from those of the molecule structure. The UV–vis and fluorescence spectra were also studied in different solvents and pH. The results showed that the synthesized compound was sensitive to both solvent and pH value. The protein labeling experiment and the stability constant result showed that BSA could be marked by the compound with enhanced fluorescence intensity and the stability constant between them was $0.82 \times 10^7 \text{ M}^{-1}$. The findings suggested that the title compound could be used in protein labeling, and will give guidance to study the design, structure and properties of other carbazole compound with benzothiazole moieties.

Acknowledgements

The present study was financially supported by the National Natural Science Foundation of China (grant nos. 51578360, 21502137, 11447222) and Technology Development Foundation Plan Project of Tianjin Colleges (20130505).

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