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Allosteric anion binding controlled by infrared irradiation: Light-triggered chromogenic sensing of iodide in aqueous solution

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ABSTRACT

Allosteric molecular sensing, which makes a synthetic receptor work like a biological signal transduction system, is a challenging topic attracting intense research interest. In this manuscript, we report the first allosteric chemosensor that can be activated and controlled by infrared (IR) irradiation. As a very important structural element in the maintenance and regulation of protein function, disulfide bond (DSB) was employed as the photoactivatable hinge of a homotropic anion receptor based on disulfide-bridged binuclear silver(I) complexes. Because of the light-driven rotation of DSB, this biomimetically designed receptor shows a unique dynamic color response to iodide in aqueous solution under IR irradiation, which is distinctly different from what occurs in the dark. As a consequence, specific colorimetric detection of iodide in aqueous solution can be well established at the micromolar concentration level. Our study exemplifies a brand-new molecular sensing mode for anions. Furthermore, it opens a new way to operate disulfide bond as an optical switch, which may lead to important applications in chemical biology.

1. Introduction

Precise regulation of the function of a protein by allosteric interaction between distinct active sites ubiquitously occurs in biological events [1–3]. Biomimetic design of allosteric molecular recognition systems offers an important strategy to regulate the complexation ability or catalytical activity of synthetic receptors and is therefore attracting much research interest [4–8]. An allosteric guest binding process enables possible high selectivity and sensitivity in molecular sensing. Furthermore, the resulting dynamic or multiple sensing responses are highly useful for establishing a reliable diagnosis. Although the photochemical control of molecular sensing and biological functions has motivated many researches because of its capability of probing in real time and real place [9–13], it remains challenging to manipulate allosteric molecular sensing by a similar external control. Herein, we report an allosteric anion receptor driven by IR irradiation and its application to chromogenic sensing of iodide in aqueous solution.

The synthetic receptor was designed to contain two anion binding sites linked by a disulfide bridge (Scheme 1). As a very important structural element in the maintenance and regulation of protein function [14–19], DSB can be cleaved by reductants or ultraviolet (UV) irradiation and thus act as an allosteric trigger. Indeed, the photolysis of DSB has been successfully utilized to regulate the conformations and bioactivities of proteins or synthetic peptides [20–28]. In addition to the photolysis reactivity, the optical activity of disulfide compounds arising from the rotation-induced different dihedral angles over DSB has been well studied [29–32]. The fact that the low rotation energy barrier of DSB can be matched by IR irradiation opens another theoretically possible entry to the optical regulation of protein conformation. Compared with the UV photolysis, IR control means minimal damage and deeper penetration in biological applications. However, conformation regulation of DSB-containing species by IR irradiation has never been reported. In our design, DSB was employed as a photoactive hinge of a binuclear silver(I) complex (**1**), which displays a unique allosteric response to iodide anions under irradiation.

2. Materials and methods

2.1. Reagents and apparatus

2,2′-Dithiosalicylic acid, taurine and silver nitrate were purchased from Sigma–Aldrich Co., Ltd. They were used without any further purification. All other reagents were of analytical grade or better and used without further purification.

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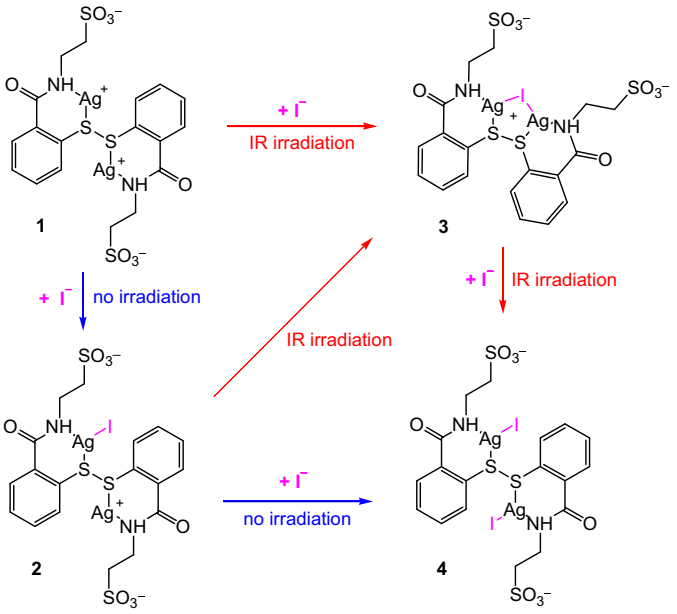


Scheme 1. Proposed working mechanism for allosteric sensing of iodide by **1**.

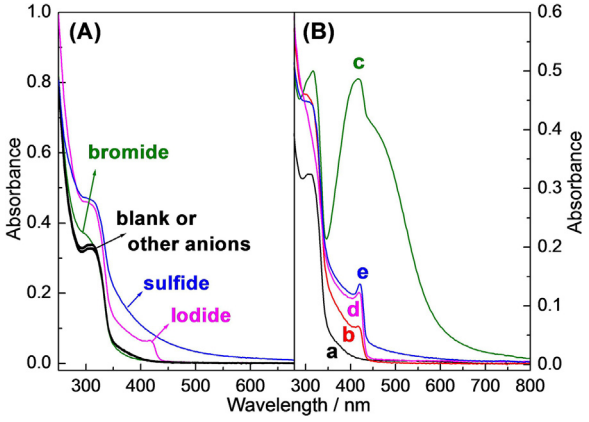
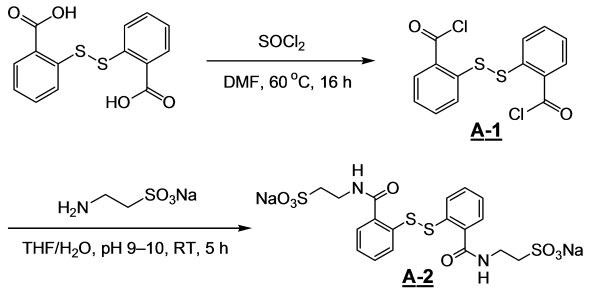


Fig. 1. (A) Absorption responses of **1** (50 μM) upon addition of different species (1.0 equiv.) including NaF, NaCl, NaBr, NaSCN, Na_2S, Na_2SO_3, Na_2SO_4, NaH_2PO_4, $NaNO_2$, $NaNO_3$, $NaHCO_3$, NaAcO, and $Na_2C_2O_4$ in the dark. (B) Absorption responses of **1** (50 μM) upon addition of different amounts of iodide: a, **1** blank; b, **1** + I^- (1.0 equiv.), no irradiation; c, **1** + I^- (1.0 equiv.), IR irradiation; d, **1** + I^- (2.0 equiv.), no irradiation; e, **1** + I^- (2.0 equiv.), IR irradiation. The IR irradiation was provided by a common IR lamp (0.76–5.0 μm, λ_{max} = 4.0 μm, 275 W).

^1H and ^{13}C NMR spectra were recorded on a Bruker Avance II 400 MHz NMR spectrometer. Electrospray ionization (ESI) mass spectra were recorded on a Bruker ESQUIRE-3000$^+$ mass spectrometer. Matrix-assisted laser desorption/ionization time-of-flight (MALDI-TOF) mass spectra were recorded on a Bruker-Dalton Reflex III mass spectrometer. X-ray photoelectron spectroscopy (XPS) spectra were acquired with a VG ESCA LAB MK-2 instrument. Absorption and fluorescence spectra were recorded on a Hitachi U-3900 ultraviolet–visible spectrophotometer and a Hitachi F-7000 fluorophotometer, respectively.

2.2. Preparation of the disulfide ligand

To a solution of 2,2′-dithiosalicylic acid (3.06 g, 10 mmol) in thionyl chloride (10 mL) was added one drop of DMF. The mixture was stirred at 60 °C for 16 h to give a brown solution. The residual thionyl chloride was evaporated under vacuum to produce compound **A-1** as a brown solid. The resulting solid was dissolved in



Scheme 2. Synthesis of the disulfide ligand.

THF (15 mL) and the solution was added dropwise into a solution of taurine (3.5 g, 28 mmol) in 40% (v/v) THF/water (25 mL) at 0 °C. The reacting system was kept at pH 9–10 and stirred for 5 h at room temperature. THF was removed under vacuum. The residual solution was poured into ethanol (200 mL) to give a yellow precipitate, which was collected by filtration. The residual taurine was removed by ion exchange chromatography. The yellow effluent was collected and evaporated under vacuum to produce compound **A-2** as yellow powder. Yield: 1.50 g, 26.8%. ^1H NMR (400 MHz, D_2O, ppm): δ = 7.65 (d, 2H, J = 8.4 Hz, Ar*H*), 7.42–7.34 (m, 4H, Ar*H*), 7.27 (t, 2H, J = 7.9 Hz, Ar*H*), 3.56 (t, 4H, J = 7.0 Hz, CH_2), 3.04 (t, 4H, J = 7.0 Hz, CH_2). ^{13}C NMR (100 MHz, D_2O, ppm): δ = 170.54, 135.77, 135.02, 131.73, 131.06, 128.18, 127.76, 49.48, 35.60. ESI-MS m/z for $[M+Na]^+$: calc. 586.96; found 586.97 (Scheme 2).

2.3. Preparation of the disulfide-bridged binuclear silver(I) complex *1*

A mixture of **A-2** (56.4 mg, 0.10 mmol) and $AgNO_3$ (34.0 mg, 0.20 mmol) in water (10 mL) was stirred for 20 h in the dark at room temperature. The resulting solution was poured into ethanol to give a yellow precipitate. The precipitate was collected and dried under vacuum to produce compound **1** as yellow powder. Yield: 8.3 mg, 10.6%. XPS (eV): Na (2s, 63.8; Auger, 497.6; 1s, 1072.0), C (1s, 284.8; C=O, 294.4), O (1s, 532.8), N (1s, 398.4); S (2p, 182.4; loss, 256.0); Ag ($3d_{5/2}$, 369.6; $3d_{3/2}$, 377.6). ^1H NMR (400 MHz, D_2O, ppm): δ = 7.88 (d, 2H, J = 8.0 Hz, Ar*H*), 7.75–7.61 (m, 4H, Ar*H*), 7.43 (t, 2H, J = 7.5 Hz, Ar*H*), 4.21 (t, 4H, J = 6.9 Hz, CH_2), 3.28 (t, 4H, J = 6.9 Hz, CH_2). ^{13}C NMR (100 MHz, D_2O, ppm): δ = 166.93, 141.03, 132.66, 125.96, 125.61, 123.39, 120.89, 49.12, 40.20.

3. Results and discussion

3.1. Spectral responses of *1* to different anions

1 was a highly water-soluble receptor easily obtained from direct complexation of $AgNO_3$ with a 2,2′-dithiosalicylic acid derivative. The optical responses of **1** in 1:1 reaction with various inorganic anions were tested in aqueous solutions. In the dark, addition of iodide results in a new edge-shape absorption band at 419 nm which is characteristic of the I 5p → Ag 5s electronic transition [33–35], while other anions induce no obvious absorption responses except that the sulfide-titrated solution is slightly turbid because of the formation of Ag_2S (Fig. 1A). Under IR illumination, the iodide-titrated solution changes from yellow to deep brown gradually, while other anions cause no similar absorption responses. As shown in Fig. 1B, besides the Ag–I charge transfer absorption at 419 nm, an intense absorption band centered at 470 nm appears under IR illumination. The formation of I_3^- ions was precluded from the cause of this distinct response by spectral comparison (Supplementary Fig. S1). Like what is usually observed from J-aggregated dyes [36,37], the new absorption band is broad

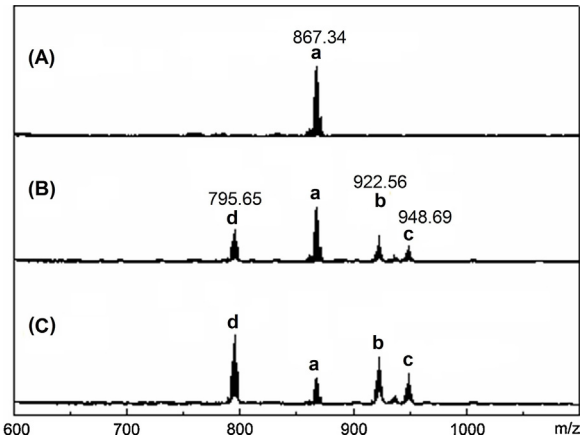


Fig. 2. MALDI-TOF mass spectra of **1** in the presence of different amounts (A, 0 equiv.; B, 1.0 equiv.; C, 2.0 equiv.) of iodide under infrared irradiation. The labeled signals a, b, c, d can be assigned to the related fragments of **1**, **3**, **2** and **4**, respectively, according to their characteristic isotopic distributions (Supplementary Fig. S2).

and obviously red-shifted, indicating the formation of the integrated Ag-I-Ag dimer (**3**) through the light-driven rotation of the disulfide bridge.

3.2. Influence of IR irradiation on the iodide binding process

The IR irradiation induced rotation of DSB in reaction of **1** with iodide anions also explains our further observations well. Because **1** bears two Ag(I)-based binding sites for iodide, second equivalent of iodide was added into the 1:1 iodide-titrated solutions to test the absorption responses (Fig. 1B). In the dark, the edged absorption at 419 nm increases by one time, indicating that each of the separated Ag(I) centers binds an iodide anion efficiently to form **4**. Under IR irradiation, the titrated solution displays a color change from deep brown to yellow green and finally results in an absorption spectrum near to the titration result in the dark. It is worthy noting that this color change is greatly inhibited without IR irradiation. Apparently, the photoinduced rotation of the disulfide bridge enables the competitive binding of the second iodide anion to form **4**. On the contrary, this competitive binding is sterically precluded when the rotation of DSB is shut off in the dark. A light-driven allosteric anion binding with remarkable color changes is therefore realized by **1**. Indeed, the 1:1 iodide-titrated product in the dark (**2**) also turns brown under IR irradiation. Spectral traces indicate that this photochromic process reaches the equilibrium much faster (*ca.* 1.0 h) under IR irradiation but very slow (>125 h) under irradiation of an incandescent lamp (400–780 nm, 100 W). It was also noticed that heating the 1:1 iodide-titrated solution in the dark induced no obvious absorption responses (Supplementary Fig. S6). These observations further confirm that the light-driven rotation of DSB is the key to the allosterism of **1**.

The iodide binding process under IR irradiation was *in situ* explored by mass spectra (Fig. 2). The signal of **1** decreases gradually with the addition of iodide and the proposed 1:1 and 2:1 binding adducts are found in the resulting spectra. When 2.0 equivalences of iodide is added, the 2:1 adduct becomes the dominant product. These observations further confirm that the light-driven allosteric metal–ligand interaction between the two unsaturated Ag(I) coordination centers and iodide anions, but not other possible factors such as formation of AgI and the photosensitivity of Ag(I), enables the dynamic chromogenic response of **1** to iodide anions under IR irradiation. In-depth investigation on the allosteric binding process by NMR titration experiments was also carried out, whereas no reliable spectral data were obtained because of the formation of AgI in the presence of relatively high concentration of **1** and iodide.

Table 1
Equilibrium times of the transformations described in Scheme 1 at a receptor concentration of 50 μM.

Reaction condition	Equilibrium time				
	1 → 2	**1 → 3**	**2 → 3**	**2 → 4**	**3 → 4**
IR irradiation	–	60 min	60 min	–	100 min
No irradiation	5 min	–	–	5 min	>30 h

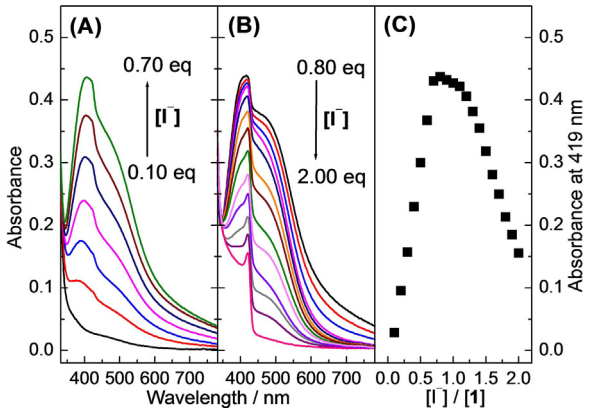


Fig. 3. Absorption spectral traces of **1** (50 μM) upon addition of increasing amount of iodide under infrared irradiation.

3.3. Quantitative description of the light-driven allosterism

The equilibrium times of the transformations depicted in Scheme 1 were acquired. As shown in Table 1 (see the *supplementary data* for detailed absorption spectral traces), the direct coordination of iodide anions on the separated Ag(I) centers occurs quickly in the dark, while the light-driven allosteric binding of iodide anions is much slower. At the concentration level of 50 μM, the 1:1 reaction between iodide and **1** under the IR irradiation takes about one hour to form the Ag-I-Ag chromogenic dimer. After that, the coordination of second iodide anion on the receptor becomes difficult. The transformation from **3** to **4** is almost forbidden in the dark and takes about 100 min under the IR irradiation. All the above results clearly establish a negative homotropic allosterism [6] that can be controlled by IR irradiation. Importantly, this allosterism can be easily tracked by distinct color changes.

3.4. Chromogenic sensing of iodide in aqueous solution

The described allosteric response is specific for iodide, enabling its application to colorimetric detection of iodide in aqueous solution. The iodide content is often required in nutritional studies because of the important roles of iodide in neurological activity and thyroid function [38,39]. Therefore, highly selective and sensitive colorimetric chemosensors capable of straightforward detection of iodide in aqueous solution are highly desirable [40–42]. The iodide sensing reaction was carried out under IR irradiation to obtain a remarkable and dynamic color change. Upon interacting with iodide, **1** displays a unique nonlinear absorption response (Fig. 3). The absorbance at 419 nm increases in a good linearity with the concentration of iodide when the titration ratio (r_t), which is defined as the molar ratio of iodide to **1**, is lower than 0.7. The maximal response appears at $r_t \approx 0.8$. After that, because considerable amount of **3** has been formed in the solution and the transformation from **3** to **4** is accelerated, the absorption decreases with increasing addition of iodide. Although the dimer-dominated broad absorption is dramatically reduced, the edged I → Ag charge transfer absorption is continuously enhanced until r_t reaches 2.0, indicating the formation of Ag—I bond accompanied by the break-up of the

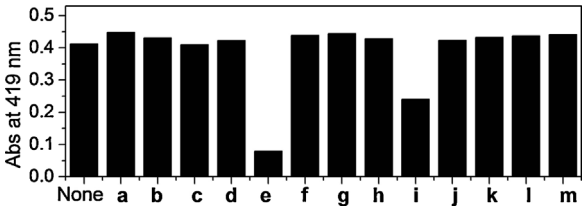


Fig. 4. Absorption response of **1** (50 μM) to iodide (50 μM) under infrared irradiation in the presence of coexisting species (500 μM) including (a) NaF, (b) NaCl, (c) NaBr, (d) NaSCN, (e) Na_2S, (f) Na_2SO_3, (g) Na_2SO_4, (h) NaH_2PO_4, (i) $NaNO_2$, (j) $NaNO_3$, (k) $NaHCO_3$, (l) NaAcO, and (m) $Na_2C_2O_4$.

Ag—I—Ag linkage. Taking advantage of the light-triggered allosteric response, colorimetric detection of iodide in aqueous solution can be established at 10^{-5} M level with a detection limit lower than 5 μM, which is comparable to that of sensitive colorimetric iodide sensors based on functional nanoparticles [40].

The iodide sensing performance of **1** was further evaluated in the presence of other anions (Fig. 4). The coexistence of most of common inorganic anions or carboxylate anions causes no or minor interference. Although nitrite itself is an inactive guest for **1**, the coexistence of nitrite reduces the sensing response because of the redox reaction between nitrite and iodide [43]. The only noteworthy interference is caused by sulfide which is able to destroy the receptor (Supplementary Fig. S10). Nevertheless, the high tolerance of other Ag(I)-philic anions including chloride, bromide and thiocyanate in this iodide sensing system confirms the contribution of allosterism to the improvement in guest binding selectivity.

4. Conclusions

In summary, we have developed the first IR irradiation-driven allosteric receptor. A photoactive DSB was integrated into the homotropic anion receptor to bridge two Ag(I)-based binding sites. Because of photoinduced rotation of the DSB, activation of the synthetic receptor by IR irradiation induces an allosteric iodide binding behavior which is greatly different from the occurrence in the dark. The resulting remarkable color changes were utilized to establish sensitive colorimetric detection of iodide in aqueous solution. Our study opens a new way to operate DSB as an optical switch, which may lead to important applications in remote and noninvasive control of protein functioning.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.snb.2015.08.102.

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