
Sweet switching: development of photoswitchable glycoconjugates

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Abstract

Photochromism, a reversible photo-induced transformation of a molecule between two isomers having distinguishably different absorption spectra, has found increasing applications in materials science, chemistry, biology and pharmacology. In this review, we summarize our recent effort on the development of photoswitchable carbohydrates and their conjugates. Synthesis, photophysical properties and potential applications of azobenzene-functionalized glycomacrolactones and glycolipids, as well as multichromophoric sugar bearing diarylethene photochrome and dicyanomethylene-4*H*-pyran fluorophore will be discussed and compared.

Keywords: azobenzenes • diarylethenes • glycoconjugates • macrolactones • photochromism

1. INTRODUCTION

Photochromic molecules are bistable molecules featuring different structural and/or electronic properties which may be reversibly isomerized by light, with the possibility to cycle up to one million “round trips”.^[1] They offer numerous opportunities for reversibly photomodulating chemical, biological or pharmacological activities or properties, with various applications in optical materials,^[1,2] chemistry^[3] and supramolecular chemistry^[4], as well as in biology^[2] and photopharmacology.^[5,6] Light is generally non-invasive and orthogonal toward most elements of living systems. It can be easily and precisely controlled in time, location, wavelength and intensity, thus enabling the precise activation or deactivation of chemical and biological function. It also offers the potential to change the properties of defined molecules with minimal disturbance to the rest of the biological system.

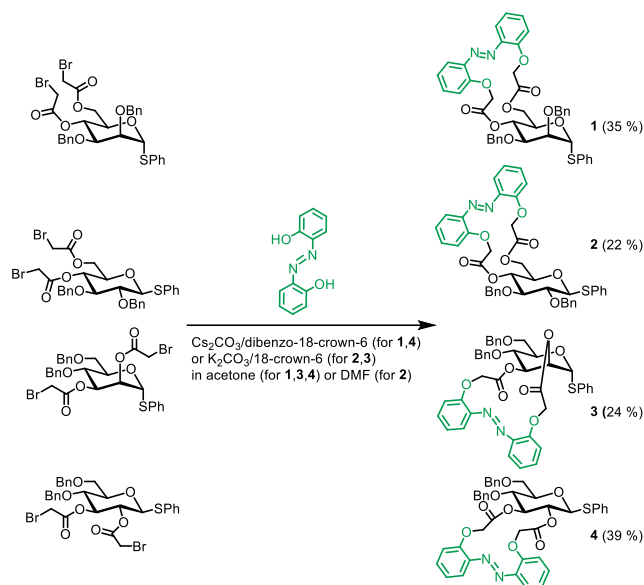
Carbohydrates are polyfunctional natural compounds which provide chiral building blocks for chemistry. Carbohydrates and glycoconjugates are involved in a myriad of important biological, physiological and pathological processes, such as cell-cell interactions, viral and bacterial infections, immune response, autoimmune disorders and chronic inflammations, cancer progression, etc.^[7] Spatiotemporally controlling their structure and functions by light would be particularly interesting since carbohydrates are optically transparent. Introducing a photochromic unit on the carbohydrates represents a recent research

field to develop new chemical tools for studying light-controlled carbohydrate-protein interactions,^[8] protein conformation,^[9] etc. In these studies, azobenzene, diarylethene and spiropyran have been used as molecular photoswitches. Azobenzene and spiropyran are T-type photochrome, allowing using light and temperature to stimulate the isomerization, while diarylethene is a P-type photochrome, insensitive to the temperature. This review focuses on our recent work on the development of photoswitchable glycomacrocyclic compounds, glycolipids, and multichromophoric sugar derivatives.

2. AZOBENZENE-FUNCTIONALIZED GLYCOMACROLACTONES

Macrocyclic compounds represent an interesting class of molecules because of their unique structural, physicochemical and biological properties as well as their potential applications in nanotechnology, biology and medicine.^[10] Natural and synthetic glycomacrocycles, especially carbohydrate-based macrolactones, have attracted increasing interest in chemistry and biology.^[11] Development of photo-responsive glycomacrocycles is appealing to control their various properties by light. We have decided to use azobenzene as molecular photoswitch because of its small size, its excellent photostability and the large changes in molecular size, shape and dipole moment induced by the photoisomerization between the thermodynamically favored *E*-isomer

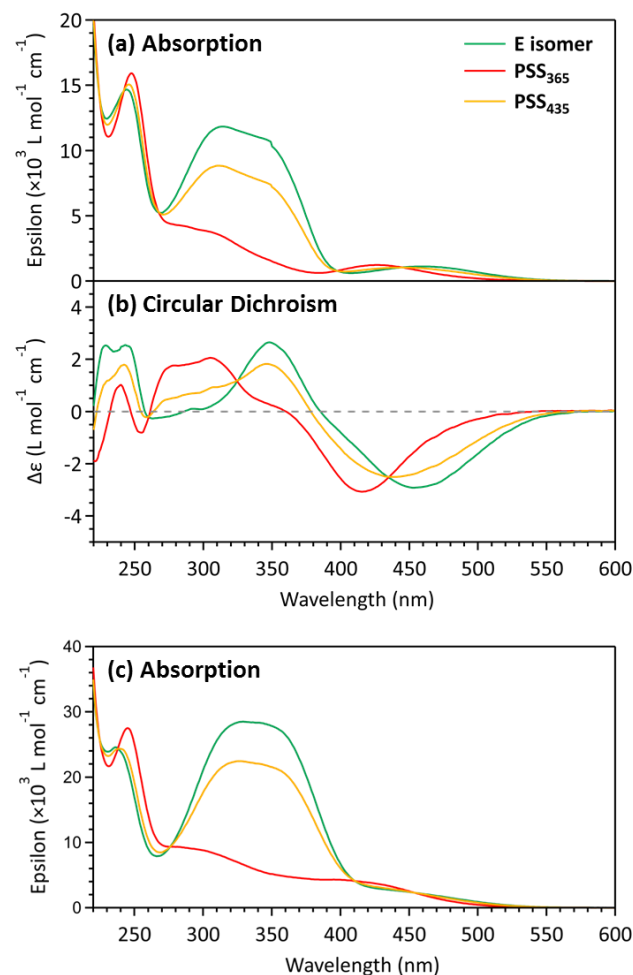
and the energetically higher *Z*-isomer.^[12] Four azobenzene-embedded glycomacro lactones with the di-*o*-hydroxy azobenzene linked to 4,6 or 2,3-position of *manno* (**1,3**) and *gluco* (**2,4**) derivatives have been prepared through the one-pot *O*-alkylation mediated macrocyclization reaction between di-*O*-bromoacetyl thio- α - or β -D-glycopyranosides and 2,2'-dihydroxy azobenzene in 22 to 39% yield (Scheme 1).^[13,14]



Scheme 1. One-pot synthesis of photoswitchable glycomacro lactones **1-4**.

Photophysical studies demonstrated that the glycomacro cycles **1-4** showed good photochromic properties in acetonitrile. Similar absorption spectra have been obtained, with a relatively strong $\pi \rightarrow \pi^*$ transition ($\lambda_{\text{max}} = 310\text{-}314\text{ nm}$) and a weaker forbidden $n \rightarrow \pi^*$ transition ($\lambda_{\text{max}} = 451\text{-}458\text{ nm}$) of azobenzene moiety (see example of compound **4**: Figure 1a, green line). Irradiation at 365 nm resulted in the decrease of the bands near 310, 345, 455 nm and appearance of a new band around 407-430 nm, revealing the *E* \rightarrow *Z* isomerization of the macrocycles (Figure 1a, red line). Two isosbestic points can also be observed at *ca.* 400 nm and *ca.* 440 nm. At the photostationary state (the equilibrium under saturating illumination at specific wavelength), *E*:*Z* ratio varied from 5:95 (for **1**), 10:90 (for **2**) and 15:85 (for **4**) to 20:80 (for **3**) at PSS₃₆₅, with higher *E* \rightarrow *Z* conversion for the 17-membered 4,6-*O*-linked azobenzene-embedded glycomacro lactones **1** and **2** (Table 1).

Irradiation at 435 nm led to the back *Z* \rightarrow *E* photoisomerization (Figure 1a, yellow line), with *E*:*Z* ratio varied from 53:47 to 70:30 at the PSS₄₃₅ (Table 1). This lower efficiency is due to the comparable level of molar absorption coefficient of both *E* and *Z* isomers at 435 nm. Nevertheless, the *Z*-azobenzenes can be converted totally back to the *E*-isomers by thermal relaxation through heating. The macrocycles *Z*-**1** to *Z*-**4** are stable at room temperature, with the *gluco* macro lactones **2** and **4** (half-life at 293 K: 72 and 56 days respectively) more stable than the corresponding *manno* derivatives **1** and **3** (51 and 37 days respectively) (Table 1). All macrocycles showed excellent fatigue resistance since photoswitching cycles have been repeated more than 15 times without any noticeable degradation.^[13,14]



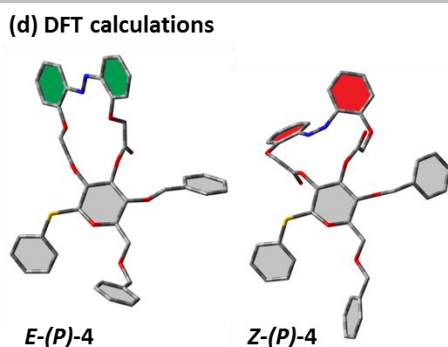


Figure 1. Normalized absorption and CD spectra of **4** (a, b), and absorption spectra of **6** (c) in MeCN; *E*-isomer (green line), PSS₃₆₅ (red line), PSS₄₃₅ (yellow line). (d) DFT-optimized geometries of *E*- and *Z*-(*P*)-**4**.^[13,14]

We have also observed chiroptical properties for macrocyclic glycoazobenzenes.^[13,14] As demonstrated by circular dichroism (CD) spectra, the 4,6-*O*-bridged *manno* derivative **1**,^[13] as well as the *gluco* macrolactones **2** and **4**^[14] showed a negative band at 454 nm and a positive band (~350 nm) corresponding respectively to $n \rightarrow \pi^*$ and $\pi \rightarrow \pi^*$ transitions of the *E*-azobenzene (Figure 1b, green line, Table 1), revealing a chirality transfer from the sugar ring to the azobenzene moiety. Irradiation at 365 nm led to a blue shift of all CD bands, without sign reversal (red line). For the 2,3-*O*-bridged *manno* derivative **3**, only the *Z*-azobenzene displayed a significant negative Cotton effect at 407 nm (Table 1). Large changes of specific rotation between *E*- and *Z*-isomers have also been observed, with the largest change for the macrocycle **1** (ca. 480°) (Table 1). As azobenzene could generate *P* or *M* helical chirality,

we have then realized DFT and TD-DFT calculations of *E*- and *Z*-(*P*),(*M*) isomers of glycomacrocycles to calculate the optimal geometry and the theoretical absorption and CD spectra. Comparison of experimental and simulated CD spectra suggests that both *E*- and *Z*-glycomacrocycles adopt preferentially *P*-helicity for the azobenzene moiety. As example, the optimized geometry of *E*- and *Z*-(*P*)-**4** is shown in Figure 1d. The *E*→*Z* photoisomerization led to a large conformational change of the macrocycles (Figure 1d), while the sugar ⁴C₁ conformation is conserved in both *E*- and *Z*-isomers, as confirmed by ¹H NMR studies.^[13,14]

Furthermore, we have accidentally observed the gelation ability of the 4,6-*O*-bridged *manno* macrolactone **1** in cyclohexane or in EtOH, with a reversible solution↔gel transition through repetitive heating and cooling procedure (Figure 2). Light irradiation at 365 nm induced an irreversible gel→solution transition in cyclohexane, leading to an opaque solution due to the reduced solubility of the more polar *Z*-macrocycle; while the gel→solution transition can be partially switched back to the gel state in EtOH after illumination at 435 nm. Very interestingly, the cyclohexane gel showed a positive Cotton effect at 460 nm (Figure 3, red line, at 20 °C) which is inverted compared to its solution state (Figure 3, blue line, at 70 °C), as a consequence of a chiral supramolecular assembly in the gel state.

Table 1. Comparison of the photochromic parameters, CD and $[\alpha]_D$ data of glycoazobenzene derivatives in MeCN before and after illumination.

Compound	1	2	3	4	5	6
λ_{\max} of <i>E</i> -isomer (nm)	454	451	451	458	440	440
λ_{\max} at PSS ₃₆₅ (nm)	420	407	420	430	430	430
<i>E</i> : <i>Z</i> ratio at PSS ₃₆₅	5:95	10:90	20:80	15:80	- ^a	- ^a
<i>E</i> : <i>Z</i> ratio at PSS ₄₃₅	53:47	70:30	67:33	70:30	- ^a	- ^a
$\tau_{1/2}$ of <i>Z</i> -isomer	51 d	72 d	37 d	56 d	5.8 d	10.5 d
CD λ_{\max} of <i>E</i> -isomer (nm)	454	454	- ^b	454	- ^b	- ^b
$\Delta\epsilon$ (M ⁻¹ cm ⁻¹)	-1.43	-1.39		-2.92		
CD λ_{\max} at PSS ₃₆₅ (nm)	413	410	407	415	- ^b	- ^b
$\Delta\epsilon$ (M ⁻¹ cm ⁻¹)	-1.47	-2.10	-1.13	-3.07		
$[\alpha]_D$ of <i>E</i> -isomer (°)	-406	-60.7	+128.4	-904	+52.7	+51.4
$[\alpha]_D$ at PSS ₃₆₅ (°)	+76	+50	-127.3	-596.1	-25.3	-20.3

^aNot determined; ^bNo significant Cotton effect was observed.

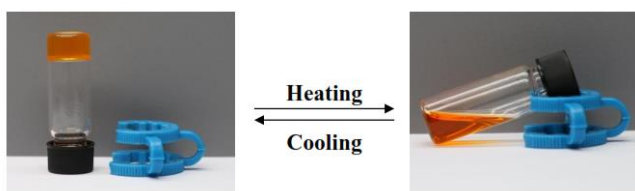


Figure 2. Reversible gel \leftrightarrow solution transition of the organogel **1** in cyclohexane (3 mM).

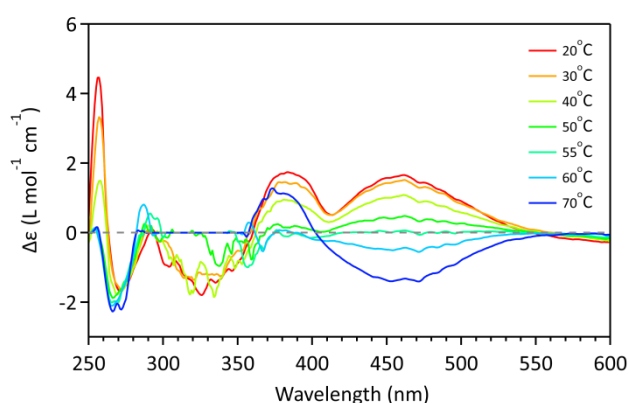
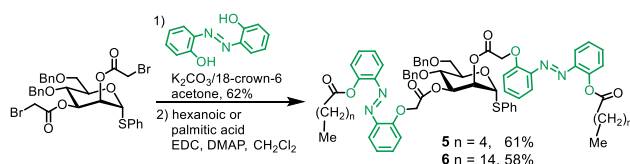


Figure 3. Temperature-dependent CD spectra of **1** gel in cyclohexane.

3. AZOBENZENE-FUNCTIONALIZED GLYCOLIPIDS

As part of the natural glycoconjugate family, glycolipids are involved in various structural, physiological and biological functions. The ability of optically controlling the glycolipids structure and function would therefore be very useful. Promising photoswitchable properties of glycolipids mimics in Langmuir films have been recently reported.^[15] From the previously described phenyl di-*O*-bromoacetyl thio- α -D-mannopyranoside, we have synthesized two photoswitchable glycolipids **5** and **6** (Scheme 2).



Scheme 2. Synthesis of photoswitchable glycolipids **5** and **6**.

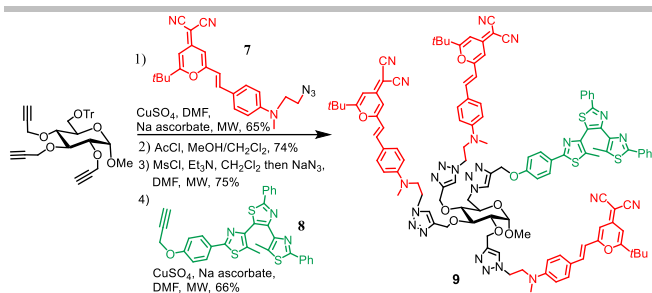
Similar to the azobenzene-based glycomacrolactones, the glycolipids **5** and **6** can

be reversibly photoisomerized after illumination at 365 and 435 nm (Figure 1c), with good photochromic properties and high fatigue resistance.^[16] The half-lives of *Z*-**5,6** are shorter than the glycomacrolactones, with a longer half-life for the *Z*-**6** bearing a longer lipid chain (5.8 and 10.5 days for *Z*-**5** and *Z*-**6** respectively, Table 1). However, due to the absence of structural constraints, no significant chiroptical property has been observed. Compared to cyclic glycoazobenzenes **1-4**, change in specific rotation between *E* and *Z* isomers of **5** and **6** is smaller too (Table 1).

4. FLUORESCENT AND PHOTOCROMIC SUGAR

The development of “intelligent” molecular systems, *i.e.* sensitive to external stimuli and able to operate complex functions is nowadays a very attractive field of fundamental and applied research. The chemical functions at the molecular level yield properties, which are translated in terms of specific electronic or optical functions to the materials and device level. One of the challenges is to synthesize organic nanomaterials with a high degree of structural order and well-defined properties yielding high-performance functions. Carbohydrates are polyfunctional molecules with well-defined stereogenic centers which constitute excellent “platforms” to link several chromophores in one molecular unit with controllable orientations. Among various applications, the development of photochemical devices based on sugar derivatives is particularly appealing for the advances in supramolecular systems *e.g.* for optical data storage media.

As a continuous interest in the development of fluorescent and/or photoswitchable carbohydrates,^[17-23] we have designed and synthesized, through copper-catalysed azide-alkyne cycloaddition reaction, a first multichromophoric monosaccharide **9** bearing three dicyanomethylene-4*H*-pyran (DCM) fluorophores (**7**) and one diarylethene (DAE) photochrome (**8**) from 2,3,4-tri-*O*-propargyl glucoside (Scheme 3).^[24]



Scheme 3. Synthesis of multichromophoric glycoside **9**.

The principle of fluorescence photoswitching is based on absorption and emission spectral features of the chromophoric units to induce efficient Förster resonance energy transfer (FRET) processes. Under light illumination, diarylethenes can undergo reversible photocyclization between their flexible and unconjugated ring open-form (DAE-OF) and conjugated closed-ring form (DAE-CF) (Figure 4a), associated with large spectral absorption changes, as exemplified with DAE **8** in Figure 4b (green and cyan lines for DAE-OF, and DAE-CF at PSS₃₃₅ respectively). To induce fluorescence photoswitching, we have chosen the DCM fluorophore **7** for its emission spectrum (red dashed line, Figure 4b) that overlaps the absorption band of the closed-form of DAE (cyan line, Figure 4b). Illumination of **9** at 335 nm isomerizes the DAE moiety from its open-form to its ring-closed form (PSS₃₃₅), with the concomitant fluorescence quenching of the DCM units by 50% (dashed green lines, Figure 4c). The fluorescence can be fully recovered after irradiation at 575 nm to induce the cycloreversion of DAE-CF to DAE-OF (Figure 4a). Through other photophysical studies, we have demonstrated that the DCM fluorescence quenching is in proportion with the ring open to ring closed-form conversion extent of the DAE moiety.

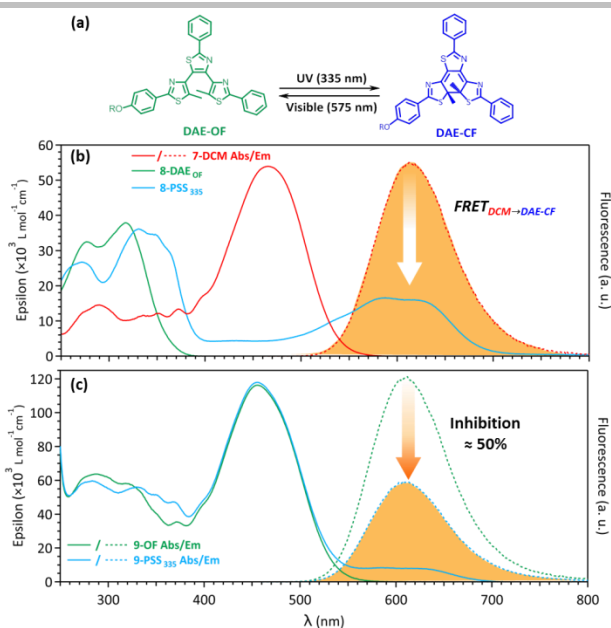


Figure 4. Photoisomerization reaction of DAE (a); absorption and fluorescence spectra of **7** (b), **8** (b), and **9** (c) before and upon UV-Vis illumination.

5. CONCLUSIONS

We have successfully synthesized azobenzene-embedded glycomacrolactones which displayed remarkable photochromic properties and bistability. Although only small numbers of photoswitchable glycomacrocycles have been reported to date,^[13,14,25,26] these ‘smart’ glycomacrocycles have already shown very interesting properties like chirality transfer and supramolecular chirality, photocontrolled molecular shape or solubility. The azobenzene functionalized glycolipids also displayed good photoswitchable functionality. The excellent photoswitching property and bistability of the synthesized glycoazobenenes open a new opportunity for the development of ‘stimuli-responsive’ carbohydrate derivatives. There is plenty of room in this field, by the ‘wedding’ of carbohydrates with different photochromic molecules like DAEs, azobenenes, spiropyran, indigos, and so on.

We have also employed carbohydrates as platform to develop reversible and photostable multichromophoric system for fluorescence photoswitching which use the FRET phenomenon between the DCM and DAE chromophoric units. We are currently working on more complex carbohydrate-based systems for optical applications.

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