## **Supporting Information**

# A photoresponsive glycosidase mimic

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#### Instrument and reagents:

All reactants and reagents (including 4-nitrophenyl- $\beta$ -D-glucopyranoside) were commercially available and were used without further purification unless otherwise indicated. Solvents used were purified and dried by standard methods. The structures of the compounds were determined by nuclear magnetic resonance spectroscopy and other spectroscopic techniques. <sup>1</sup>H and <sup>13</sup>C NMR spectra were recorded with 500 MHz Jeol instruments. Chemical shifts are reported in  $\delta$  values relative to the solvent peak. The solvents used for the spectroscopy experiments were of the spectroscopic grades and were free from any fluorescent impurities. Double distilled water was used for the spectroscopy experiments. UV spectra were recorded with a Hitachi U-4100 UV-vis spectrophotometer. pH data were recorded with a Sartorius Basic Meter PB-11 calibrated at pH 4, 7 and 10.

#### **Experimental section**

#### Synthesis of azobenzene-3,3'-dicarboxylic acid<sup>1</sup>

Azobenzene-3,3'-dicarboxylic acid was synthesized according to the literature procedure.<sup>1</sup> M.p. 338 °C (reported 340 °C); <sup>1</sup>H NMR (500 MHz, DMSO-*d*<sub>6</sub>): δ 8.39 (s, 2H, ArH), 8.12-8.07 (m, 4H, ArH), 7.67 (t, *J*=7.6 Hz, 2H, ArH); <sup>13</sup>C NMR (500 MHz, DMSO-*d*<sub>6</sub>) δ 167.57, 151.79, 135.32, 132.18, 129.58, 126.59, 122.26; IR (KBr cm-1) 1683, 1590, 2890, 3045, 3440; ESI MS *m*/*z* Calculated for C<sub>14</sub>H<sub>10</sub>N<sub>2</sub>O<sub>4</sub> 270; Obtained 270; UV-vis: λ (nm), ε (M<sup>-1</sup> cm<sup>-1</sup>) For *(E)*-isomer 317 (4.6 × 10<sup>4</sup>), 427 (2.7×10<sup>3</sup>); For *(Z)*-isomer: 317 (2.4 × 10<sup>4</sup>), 427 (3.1 × 10<sup>3</sup>).

#### **Photoisomerization studies**

Photoisomerization experiments were performed with azobenzene-3,3'-dicarboxylic acid (1  $\times 10^{-3}$  M) in DMSO-*d*<sub>6</sub>. The *E* to *Z* photoisomerisation of azobenzene 3,3'-dicarboxylic acid was observed by irradiating sample by 366 nm UV light (8 W, sample placed at a distance of 12 cm from the source) under ice-cold conditions (0 °C). The reverse isomerisation (*Z* to *E*) was achieved by exposing the sample with visible light (200 W, 14 cm) at 0 °C. The isomerization was monitored by studying the change in the UV/Vis absorption spectra and also by <sup>1</sup>H NMR analysis.



**Figure S1**: Changes in electronic absorption spectrum for *E* to *Z* isomerisation of azobenzene-3,3'-dicarboxylic acid on exposure of 366 nm UV light in DMSO solution (10  $\mu$ M).



**Figure S2**: Changes in electronic absorption spectrum for (*Z*) to (*E*) isomerisation of azobenzene-3,3'-dicarboxylic acid on exposure to visible light in DMSO solution (10  $\mu$ M).



**Figure S3**: (A) <sup>1</sup>H NMR spectra of azobenzene-3,3'-dicarboxylic acid on exposure of 366 nm UV light at t = 0 and 300 min at 0 °C. The spectrum recorded after 300 min of UV irradiation corresponds to the PSS; (B) The NMR spectra of the conversion of the PSS to the *E*-isomer on exposure to >490 nm visible light for 30 min.

#### General procedure for the hydrolysis

Each assay was performed on 3 mL of samples prepared from 1.5 mL 1 mM of aqueous KCl solutions of the 4-nitrophenyl- $\beta$ -D-glucopyranoside at various concentrations (0–7.5 mM) mixed with 1.5 mL of acetate (acetic acid/sodium acetate), phosphate (Na<sub>2</sub>HPO<sub>4</sub>/acetic acid), borate (borax/HCl or NaOH) or CHES buffers (CHES/NaOH) containing either the azobenzene derivative (11  $\mu$ M) or nothing (as control) and the final pH values were measured with a pH-meter. The reactions were performed in the darkness inside a box with its walls painted with black. The photoiraadiated (*Z*)-form of the catalyst was exposed to the 366 nm light periodically for ensuring its retention in the (*Z*)-form. The

progress of the hydrolysis was monitored by quenching the aliquots using pH 12 phosphate buffer (0.2 M) and recording the absorbance.

The formation of the 4-nitrophenol (4-NP) was monitored by the appearance of peak at 400 nm which was monitored using UV/vis spectroscopy at room temperature. The concentration of the 4-NP species was calculated at individual pH from the molar absorptivity of the species at each of the pH at 400 nm.

The Michaelis-Menten kinetics data were obtained from the initial rates (<15%) of the reactions and its subsequent linearization in the Lineweaver-Burk form as presented in Figure 5(A) and Figure 5(B) of the main manuscript.

#### pH-rate profile

The buffers used in determining the pH - rate profile were acetate buffers, phosphate, borate buffers and CHES. The buffers were freshly prepared before each experiment. The experiments at pH 5.8 used the sodium acetate/acetic acid buffer. The pH of the buffer solutions were measured using an electrode calibrated at pH 4.0, 7.0 and 10.0 at 25 °C. The pH was measured before and after each hydrolytic assay and was found not to vary significantly in the course of the experiment.



Figure S4: <sup>1</sup>H NMR spectrum of azobenzene-3,3'-dicarboxylic acid in DMSO-*d*<sub>6</sub>.



Figure S5: <sup>13</sup>C NMR spectrum of azobenzene-3,3'-dicarboxylic acid in DMSO- $d_6$ .

#### **Docking studies**

The docking studies have been performed on using the ArgusLab 4.0.1 software<sup>2</sup> with the previously minimized structures using ZINDO methods. The docked structure was equilibrated using the program. The structures were explicitly solvated using the SCRF solvent model<sup>3</sup> with water in a  $20 \times 20 \times 20$  Å<sup>3</sup> cube placed around the azobenzene docked sugar substrate. The docking studies were run 10 times to produce the best results.



**Figure S6**: 4-nitrophenyl- $\beta$ -D-glucopyranoside docked to the Z-isomer of the azobenzene-3,3'-dicarboxylic acid (acid-carboxylate form). Only the two water molecules relevant to the catalysis have been shown here explicitly. The other water molecules and some of the Hatoms in the structures have not been shown for clarity. The yellow line has been manually drawn to indicate the attacking sites as shown in Figure 6 in the main paper.



**Figure S7**: Alternate exposure to UV (366 nm, 3h) and visible light (.490 nm, 30 min) in a reaction mixture containing 4-nitrophenyl- $\beta$ -D-glucopyranoside and the (*Z*)-azobenzene-3,3'-dicarboxylic acid at pH 5.8.

#### Thermal stability of the (Z)-isomer of the azobenzene-3,3'-dicarboxylic acid

Thermal stability of the (*Z*)- azobenzene-3,3'-dicarboxylic acid was studied in DMSO using UV-vis spectroscopic methods.<sup>4</sup> The samples were placed in a Peltier thermostat attached to a spectrophotometer and the spectra were recorded at various time intervals. The data were plotted at 317 nm using both zero and first order kinetics – the first order kinetic data offered better fitting of the data with higher regression coefficients. Thus the first order kinetic data have been used for the studies presented here in the subsequent sections.

The error in the absorbance values of the photoisomerization reaction run in triplicate varied by less than 5%.



**Figure S8.** Thermal isomerisation studies of the (*Z*)-isomer of the azobenzene-3,3'dicarboxylic acid at various temperatures. The plots A–F display the time dependence of the mole fraction composition of the (*Z*) isomer with time at various temperatures (45–70 °C with an interval of 5 °C) in DMSO. The error in the temperature reading is  $\pm 1$  °C.

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**Figure S9.** (A) Arrhenius and (B) Eyring plots for determination of the activation parameters for the thermal reversal of the (Z) to the (E) isomer.

**Table S1.** Activation parameters for the (Z) to (E) isomerisation of the azobenzene-3,3'-dicarboxylic acid.

Parameters	ΔE <sub>act</sub> (kcal mol <sup>-1</sup> )	ΔH <sup>‡</sup> (kcal mol <sup>-1</sup> )	$\Delta S^{\ddagger}$ (cal mol <sup>-1</sup> K <sup>-1</sup> )
Values±(error)	23.0±(0.7)	23.6 ±(0.7)	22.3 ±(6.8)

#### Reference

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<sup>1. (</sup>a) S. Ghosh, D. Usharani, A. Paul, S. De, E. D. Jemmis and S. Bhattacharya, *Bioconjugate Chem.*, 2008, **19**, 2332; (b) M. Y. Qiu, J. C. Zhang, Y. Wei, Q. C. Jia and Y. S. Niu, *Asian J. Chem.*, 2012, **24**, 2295; (c) D. H. Marrian, P. B. Russell, B. J. F. Hudson, J. W. Cornforth, R. H. Cornforth, W.J. Dunstand and M. L. Tomilson, *J. Chem. Soc.*, 1946, 753.

<sup>2.</sup> ArgusLab 4.0, M. Thompson, Planaria Software LLC, Seattle, http://www.ArgusLab.com.