Supporting Information

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SI Materials and Methods

Scheme for the Synthesis of the Cyclodextrin Dimer. Reagents and chemicals were purchased from commercial sources. Diisobutylaluminium hydride (DIBAL-H) was purchased from Aldrich (1.5 M in toluene), and palladium were purchased from Aldrich. The solvents were purified by distillation before use or dried by standard methods: Toluene was distilled over sodium, Dichloromethane was distilled over phosphorous pentoxide, dimethyl formamide (DMF) was dried and kept on molecular sieve 4 Å. The hydrogenations catalyzed by palladium were carried out at 1 bar of pressure. Reactions were monitored by TLC on a precoated plate of silica gel $^{60}F_{254}$ (layer thickness 0.2 mm; E. Merck) and detection by UV (254 nm) or charring with sulfuric acid. Flash column chromatography was performed on silica gel 60 (230–400 mesh, E. Merck). NMR spectra were recorded at room temperature with a 300 MHz Bruker AVANCE 300, a 400 MHz Bruker AVANCE 400, or a 600 MHz Bruker AVANCE 600 spectrometer.

Monol CD2. To a solution of perbenzylated cyclodextrin (CD; Fig. S4, compound **1**) (13.6 g, 4.5 mmol, 1 eq.) in toluene (20 mL) under nitrogen atmosphere, DIBAL-H (1.5 M in toluene, 60 mL, 90 mmol, 20 eq.) was added dropwise at room temperature. The reaction mixture was heated at 60 °C for 30 min and poured on ice. HCl solution (150 mL, 1 M in water) and EtOAc (200 mL) were added and the solution was stirred during 1 h. The aqueous layer was extracted with EtOAc (3 × 100 mL). The combined organic layers were dried over $MgSO_4$, filtered and concentrated under vacuum. After purification by silica gel chromatography (cyclohexane/EtOAc 6:1), monol CD **2** (Fig. S4, compound 2) (7.2 g, 62%) (1) was obtained as a white foam. R_f 0.66 (cyclohexane/EtOAc 2:1).

Mesyl CD 3. To a solution of monol CD **2** (3.6 g, 1.22 mmol, 1 eq.) in anhydrous CH_2Cl_2 (15 mL), were added triethylamine (340 µL, 2.45 mmol, 2 eq.) and mesyl chloride (190 µL, 2.45 mmol, 2 eq.) at 0 °C. The reaction mixture was stirred at room temperature for 2 h under nitrogen; then, water (10 mL) was added slowly, the layers were separated, and the aqueous layer was extracted with CH_2Cl_2 (3 × 10 mL). The combined organic layers were dried over $MgSO_4$, filtered and concentrated. Silica gel flash column chromatography of the residue (cyclohexane/EtOAc 5:1) afforded the mesylated β-cyclodextrin **3** (Fig. S4, compound 3) (3.68 g, quant.) as a white foam.

Azido CD 4. To a solution of mesylated cyclodextrin **3** (3.5 g, 1.16 mmol, 1 eq.) in anhydrous DMF (20 mL), was added sodium azide (302 mg, 4.64 mmol, 4 eq.) at room temperature under nitrogen. The reaction mixture was heated at 80 °C during 12 h and then cooled down to room temperature, and the solvent was evaporated. The residue was dissolved in ethyl acetate (20 mL) and washed with water (20 mL). The aqueous layer was extracted with ethyl acetate (3 × 20 mL). The combined organic layers were dried over $MgSO_4$, filtered, and concentrated under vacuum. Silica gel flash chromatography of the residue (cyclohexane/EtOAc 5:1) gave monazido-β-cyclodextrin **4** (Fig. S4, compound 4) (3.37 g, 98%) as a white foam.

Protected Dimer 5. diisopropyl ethyl amine (95 µL, 0.54 mmol, 8 eq.), tris(benzyltriazolylmethyl)amine (29 mg, 0.054 mmol, 0.8 eq.), and $Cu(CH_3CN)_4PF_6$ (20 mg, 0.054 mmol, 0.8 eq.) were added to a solution of azido β-CD **4** (400 mg, 0.135 mmol, 2 eq.) and 1,4-diethynylbenzene (9 mg, 0.067 mmol, 1 eq.) in anhydrous DMF (8 mL) at room temperature under nitrogen in a microwave vial. The reaction mixture was heated at 150 °C under microwaves for 1 h. The solvent was evaporated and the residue was purified by silica gel chromatography (cyclohexane/EtOAc 3:1) to give the compound **5** in Fig. S4 as a white foam (406 mg, quant.). Derivatization in triazolium to be detected by mass spectrometry: CD (2 mg) was solubilized in CH_3CN (0.5 mL), and iodomethane (10 µL) was added in a microwave vial. The solution was stirred at 100 °C for 30 min under microwaves. Then 1 µL of this solution was injected into mass spectrometer.

Dimeric CD 6. The dimer β-CD **5** (680 mg, 0.11 mmol, 1 eq.) was dissolved in THF/MeOH/H_2O (10:4:1, 102 mL) under nitrogen. $Pd(OH)_2$ was added (1.36 g, 2 eq. in mass). The reaction mixture was purged with H_2, then stirred under H_2 at room temperature for 12 h. Palladium was filtered through a Celite pad and the residue was washed with MeOH/H_2O (100 mL). The solvents were evaporated and the residue was dissolved in water and was freeze dried. The product **6** in Fig. S4 (275 mg, quant.) (2) was obtained as a white amorphous powder.

1. Lecourt T, Herault A, Pearce AJ, Sollogoub M, Sinaÿ P (2004) Triisobutylaluminium and diisobutylaluminium hydride as molecular scalpels: The regioselective stripping of perbenzylated sugars and cyclodextrins. *Chemistry* 10(12):2960–2971.

2. Mourer M, Hapiot F, Tilloy S, Monflier E, Menuel S (2008) Easily accessible mono- and polytopic β-cyclodextrin hosts by click chemistry. *Eur J Org Chem* 2008:5723–5730.

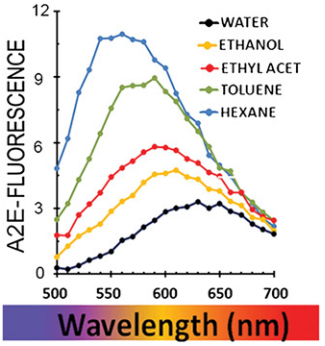

Fig. S1. Sensitivity of the fluorescence spectrum of A2E to the polarity of its environment. Emission spectra of 5 µM A2E in Water (black line), Ethanol (yellow line), Ethyl Acetate (red line), Toluene (green line), and Hexane (blue line).

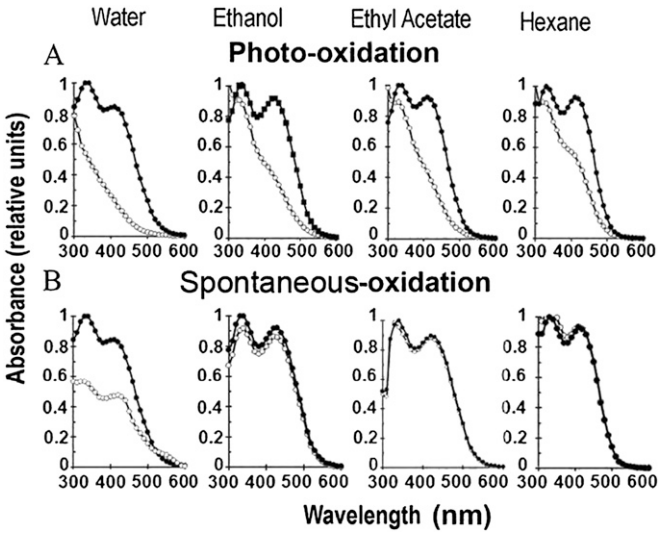

Fig. S2. Protection of A2E against oxidation by organic solvents. Protection was monitored by changes in the UV-visible absorption spectra of 5 μM A2E solutions. (*A*) A2E oxidative status before (●) and after (○) blue-light irradiation in the presence of indicated solvents. (*B*) A2E oxidative status at time 0 (●) and 1 d (○) after incubation at room temperature in the dark in the presence of organic solvents. The different protective capacities of nonpolar solvents and CDs are consistent with the notion that light-dependent and -independent oxidative reactions proceed through different pathways.

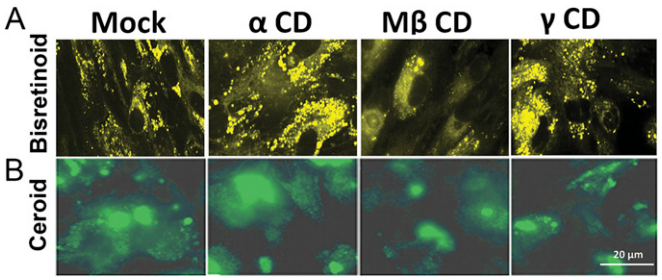

Fig. S3. Methyl-β-CD (but neither α- nor γ-CDs) reduces levels of bisretinoids without significantly affecting ceroid-lipofuscin deposits. Live autofluorescence images of cells plated on glass bottom dishes, loaded with bisretinoids (*A*) or ceroid- (*B*) lipofuscin, and exposed 48 h to mock, α-, β-, Methyl-β-, and γ-CD treatments.

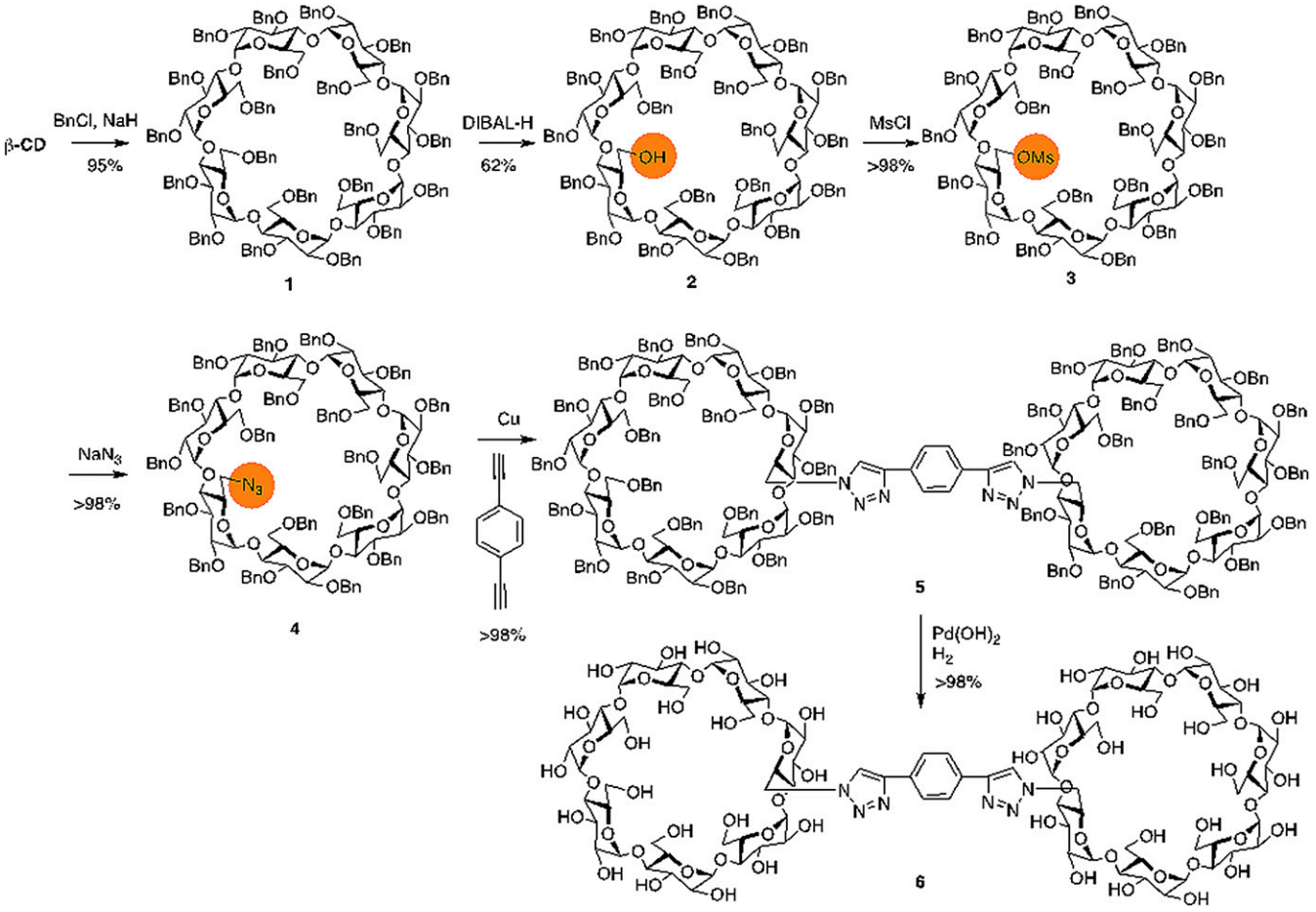

Fig. S4. Synthetic scheme for β-CD dimer.