Supporting Information: Modulating the oxidation of cucurbit[n]urils

Jade A. McCune,[†] Edina Rosta,[‡] and Oren A. Scherman[*,†]

†*Melville Laboratory for Polymer Synthesis, Department of Chemistry, University of Cambridge, Lensfield Road, Cambridge CB2 1EW, UK.*

‡*Department of Chemistry, King's College London, London SE1 1DB, U.K.*

E-mail: oas23@cam.ac.uk

Materials and methods

All chemicals used were reagent grade or higher and used without further purification unless otherwise stated. $(NH_4)_2S_2O_8$ was purchased from Sigma Aldrich. Milli-Q water was produced by Synergy Ultrapure Water Systems. ITC experiments were carried out on a MicroCal iTC_{200} from GE Healthcare Life Sciences, Microcal Inc. at 25 ℃. CB[n] was synthesised from glycoluril with formaldehyde by a basic procedures published by Day and Kim. Mass spectra were recorded on Thermos LTQ Velos. Samples (250 μL) were introduced into the spectrometer *via* syringe with a flow rate set at 15 μL/min. 0.2 μm syringe filters were purchased from GE Healthcare Life Sciences.

Synthesis of guest compounds G1–G5

Synthesis of bisimidazolium compounds were all carried out according to previously reported procedures. G1 *via* a procedure reported by Zhao *et al.*[1] and G2, G3 and G4 in accordance

with previously reported procedures by Jiao *et al.*[2] Synthesis of G5 was also carried out in accordance with a previously reported procedure bu Ueda and co-workers.[3]

Determination of solution binding constants using ITC

Titration experiments were carried out at 25 ℃ sodium acetate buffer (50 mM, pH = 4.2). Fresh analyte solutions were dissolved by sonication and heating up to 60 ℃. All solutions were degassed prior to titration. The binding equilibria were studied using a cellular solution of CB[n] and guest solution was titrated. 19 consecutive injections of 2 μL each were used, whereby the first injection was chosen to be 0.2 μL in all cases. Thus the first data point was removed from the data set prior to curve fitting. Heats of dilution were checked by titration into the buffer solution and were found to be negligible. The data was analysed with Origin 7.0 software, using the one set of sites model for CB[n] complexes.

ITC curves of G1–G5 to CB[n]

The binding constant of **G1** to CB[6] has been previously reported by Zhao *et al.*[1] but the K_a of **G1** to CB[7] and CB[8] has not been previously reported . The ITC curves for **G1**to CB[7] and CB[8] are shown below in Figure **G2**–**G5** the ITC values had been previously reported.[2,4,5]

Protocol for oxidation of CB[n]

CB[n] (0.01 mmol) and guest (0.01 mmol) were placed in a 25 mL round bottomed flask with 10 mL of H_2O (Millipore, 18.2 MΩ·cm) and fitted with a rubber septum. The solution was heated to 85 ℃ and purged with nitrogen. After 30 min, APS (0.01 mmol) was dissolved in the minimum volume of H_2O (Millipore, 18.2 MΩ·cm) (0.1 mL) added in one portion to flask *via* a syringe.

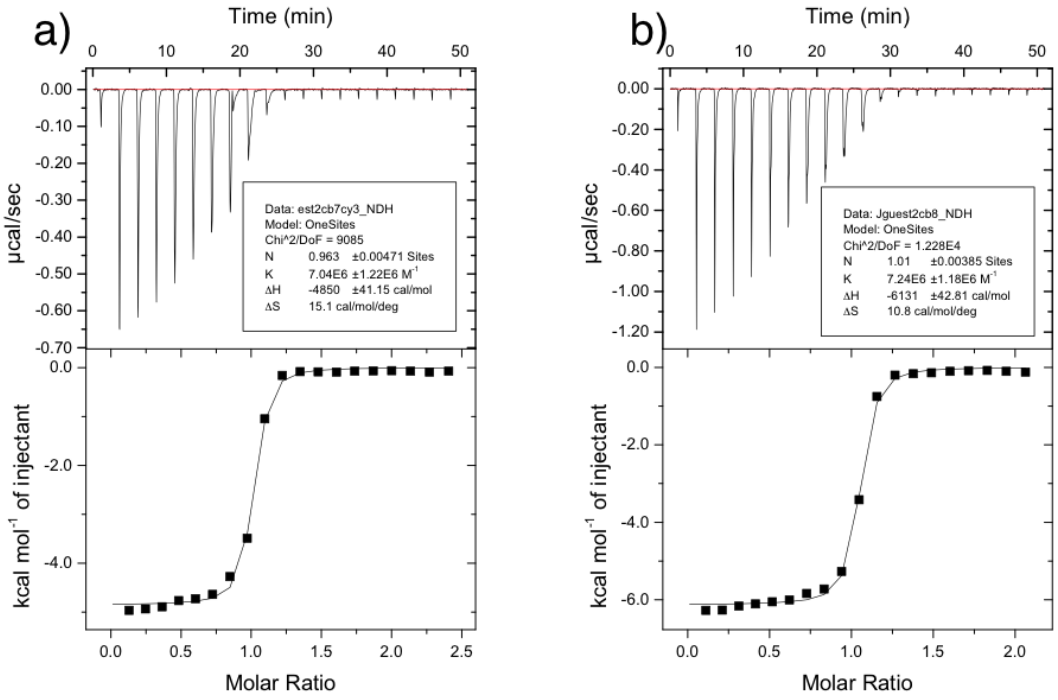

Figure S1: ITC curves showing the binding constants (K_a) of **G1** to (a) CB[7] and (b) CB[8]

Method for sampling oxidation reaction

Samples of the reaction mixture ($\approx$ 0.5 mL) were removed at set time intervals *via* a syringe, placed in a vial and immediately dropped into a bath of liquid N_2. When the sample was completely frozen it was removed from the liquid N_2 bath and allowed to slowly warm to room temperature. Upon reaching room temperature, the sample was filtered through a 0.2 μm syringe filter and the diluted with 1 mL of HPLC grade water before injection into the ESI-MS.

Protocol for oxidation of CB[7] under guest free conditions

CB[7] (0.01 mmol) was placed in a 25 mL round bottomed flask with 10 mL of H_2O (Millipore, 18.2 MΩ·cm) and fitted with a rubber septum. The solution was heated to 85 ℃ and purged with nitrogen. After 30 min, APS (0.01 mmol) was dissolved in the minimum volume of H_2O (Millipore, 18.2 MΩ·cm) (0.1 mL) added in one portion to flask *via* a syringe. A stock

solution of guest was made in the HPLC grade water such that the 1 mL needed for dilution prior to mass spec analysis contained 1 equivalent of **G1**.

Protocol for optimised CB[7] oxidation

CB[7] (0.01 mmol) and guest (G1) (0.01 mmol) were placed in a 25 mL round bottomed flask with 10 mL of H_2O (Millipore, 18.2 MΩ·cm) and fitted with a rubber septum. The solution was heated to 85 ℃ and purged with nitrogen. After 30 min, APS (0.033 mmol) was dissolved in the minimum volume of H_2O (Millipore, 18.2 MΩ·cm) (0.1 mL) added in one portion to flask *via* a syringe. Following 35 minutes the reaction was quenched by submerging in liquid nitrogen.

Method for ESI-MS analysis

All samples were injected into the ESI-MS at a flow rate of 15 μL/min. The ESI-MS was flushed extensively between uses until all peaks corresponding to the previously run sample were at a relative intensity less than 1. In all cases, the m/z range studied was 400 – 1000. Spectra were recorded and an average obtained over acquisition times of at least 2 min after the signal following injection had stabilised.

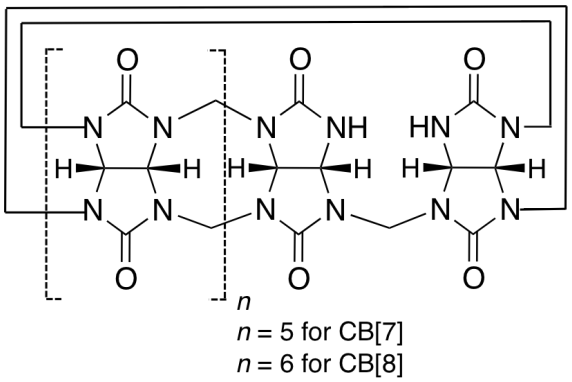

Figure S2: Proposed structure of product with m/z 6 less than CB[n] *deg*-CB[n], caused by the breakdown of one methylene bridge between adjacent glycoluril units.

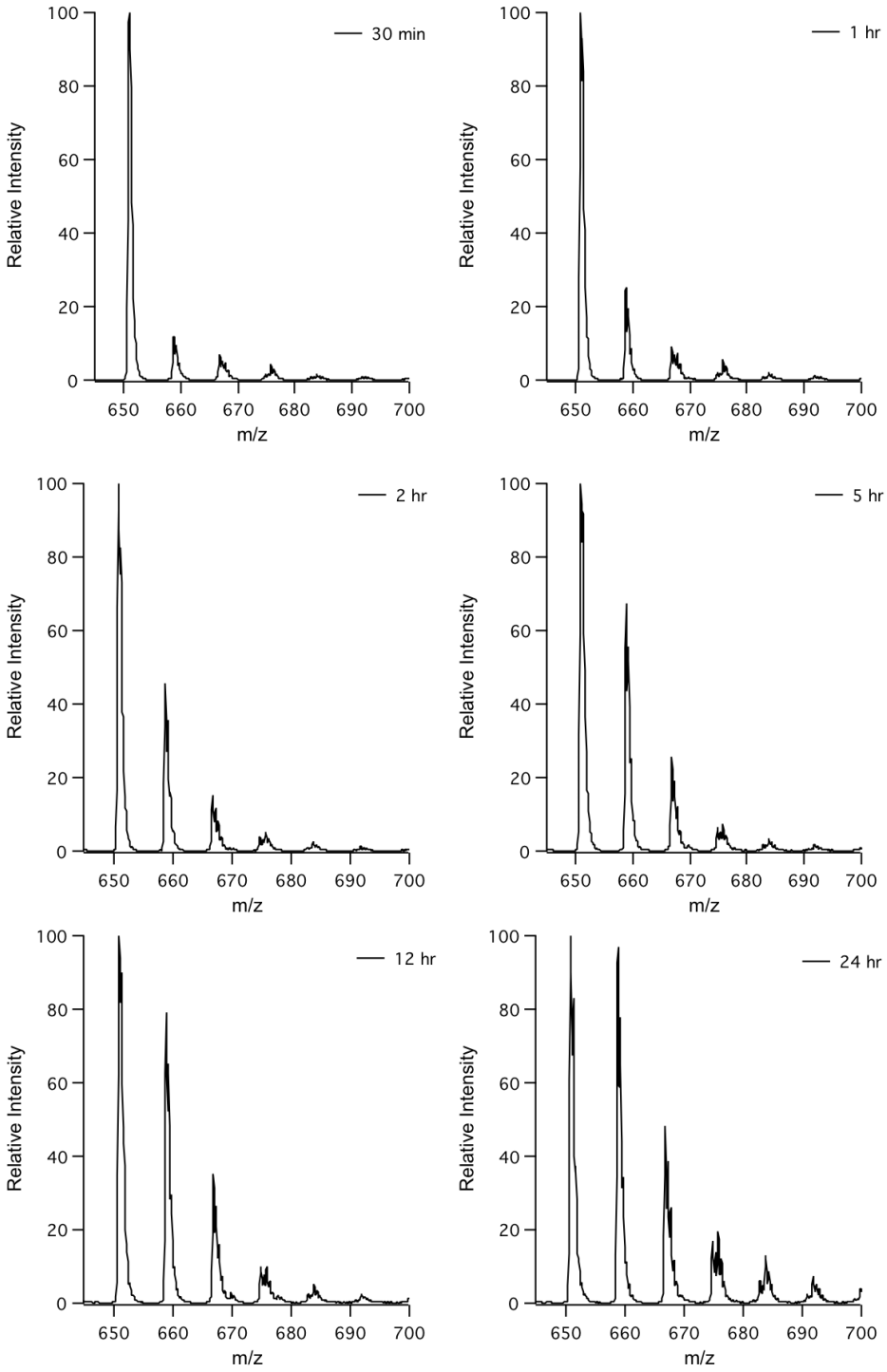

Figure S3: Representative ESI-MS spectra of CB[6]·**G1** and functionalised derivatives following oxidation at selected time intervals.

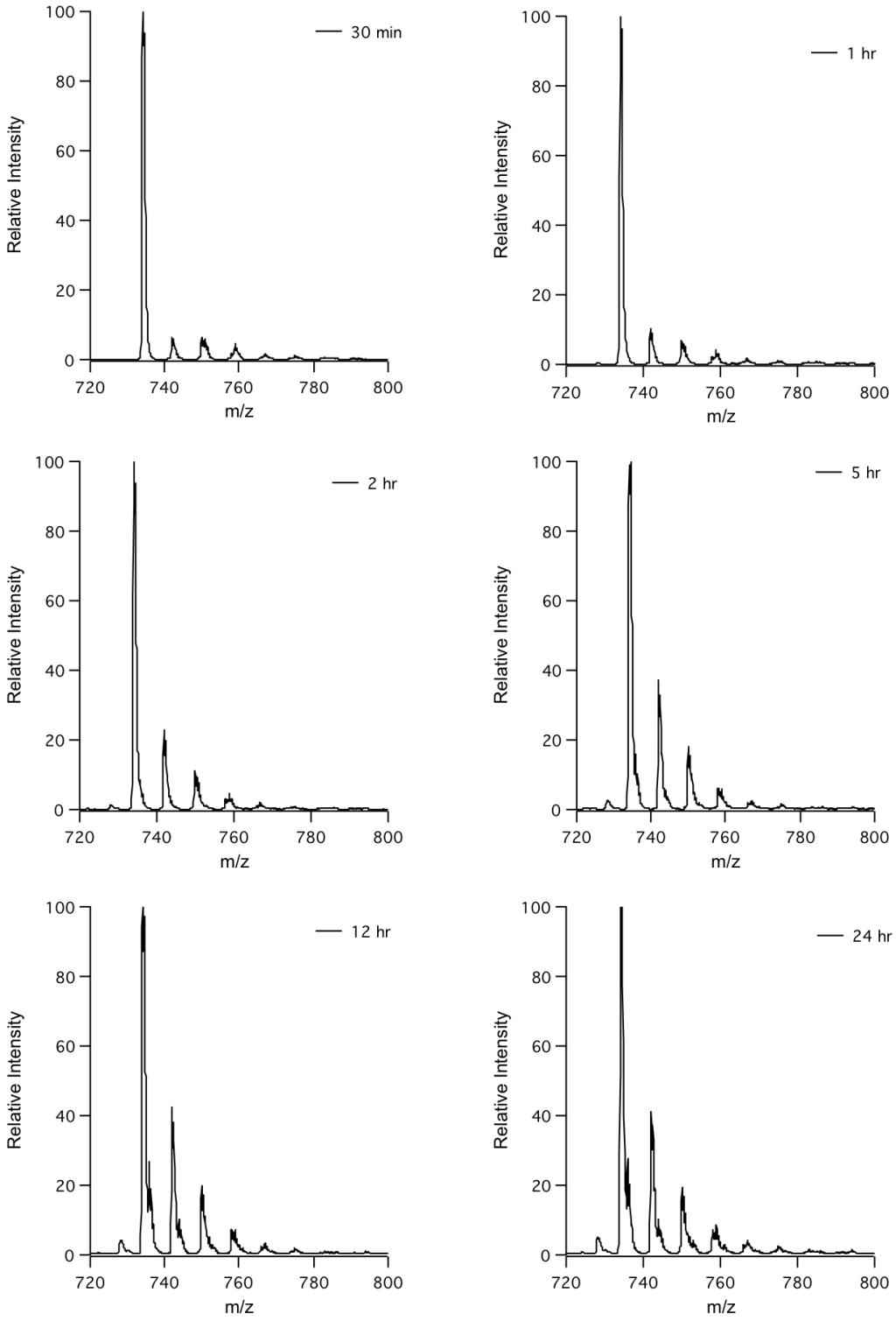

Figure S4: Representative ESI-MS spectra of CB[7]·**G1** and functionalised derivatives following oxidation at selected time intervals.

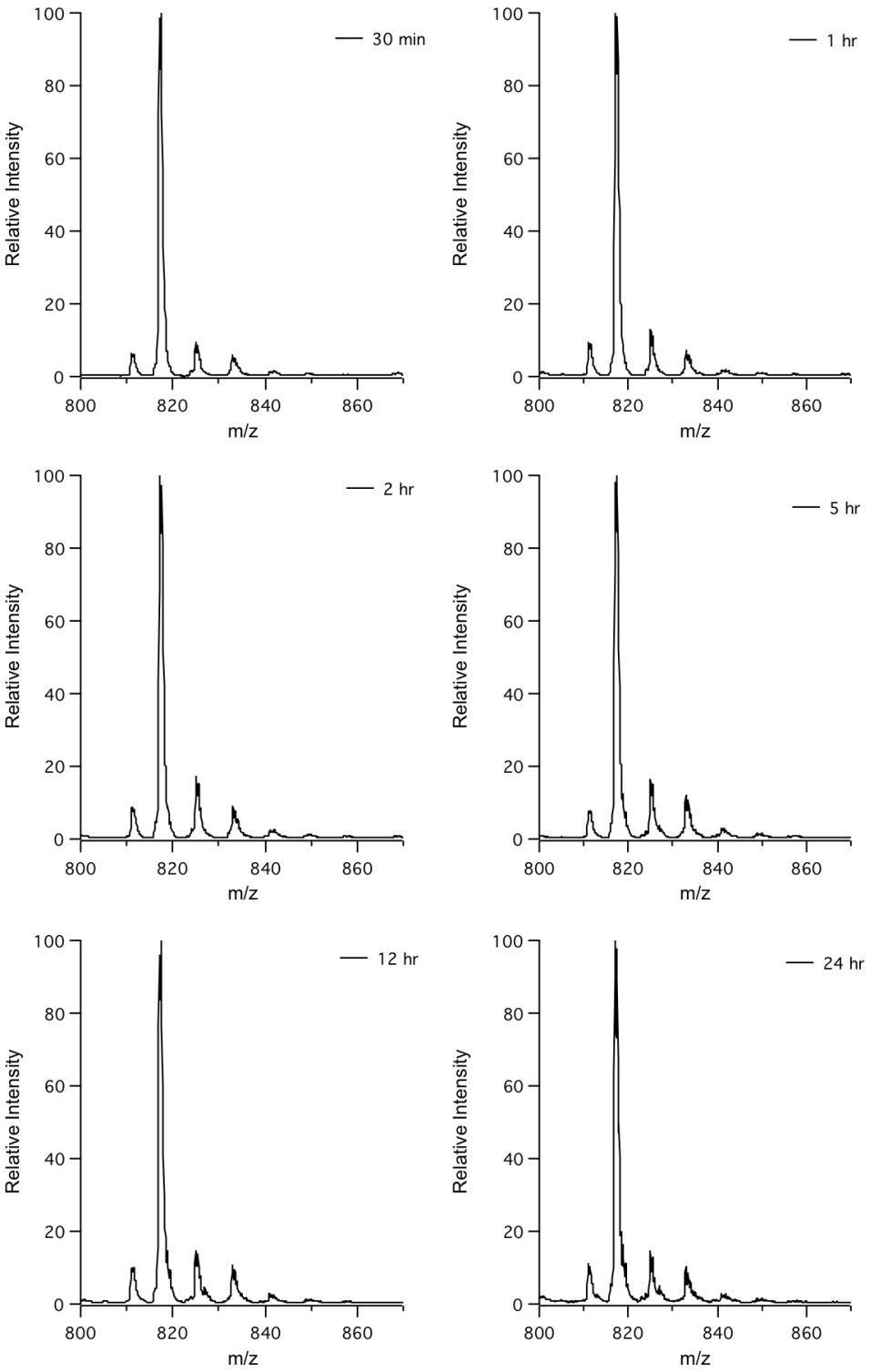

Figure S5: Representative ESI-MS spectra of CB[8]·**G1** and functionalised derivatives following oxidation at selected time intervals.

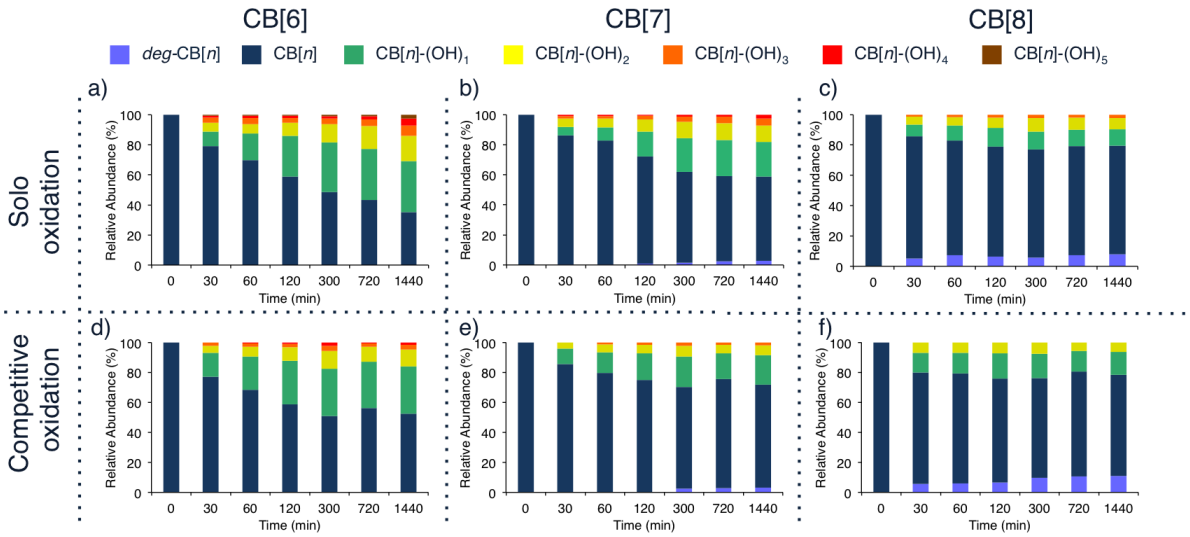

Figure S6: Stacked bar chart plots showing the relative abundance of CB[n] and f-CB[n] derivatives at time points taken throughout the reaction. Graphs (a – c) highlight the amount of each CB[n]-$(OH)_x$ derivative present when the oxidation is carried out on a single homologue, *i.e.* only one parent CB[n] homologue is present. Graphs (d – f) depict the modulated reactivity when the parent CB[n] is present in a complex mixture of equimolar CB[n] (n = 6, 7 & 8.)

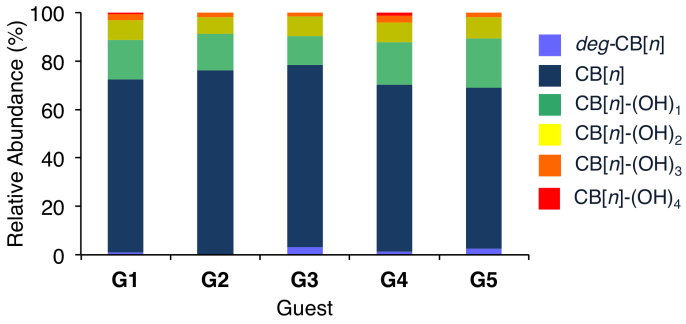

Figure S7: Stacked bar chart plots showing the relative abundance of CB[n] and each f-CB[n] derivatives following oxidation in the presence of different guest moieties **G1**–**G5** following 120 min reaction time.

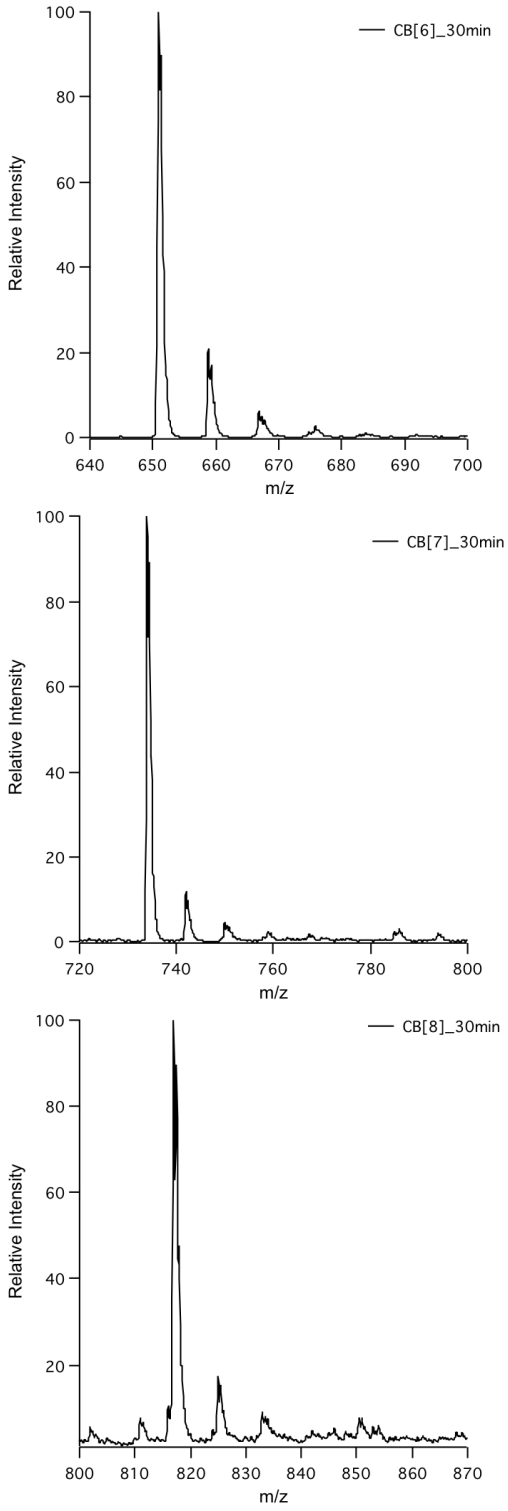

Figure S8: Representative ESI-MS spectra of CB[6]·**G1**, CB[7]·**G1**, CB[8]·**G1** and their respective functionalised derivatives. Spectra are representative of the abundance of each CB[n] homologue and their functionalised derivatives present the reaction mixture containing equimolar quantities of each CB[n] homologue following 30 min under oxidative reactive conditions.

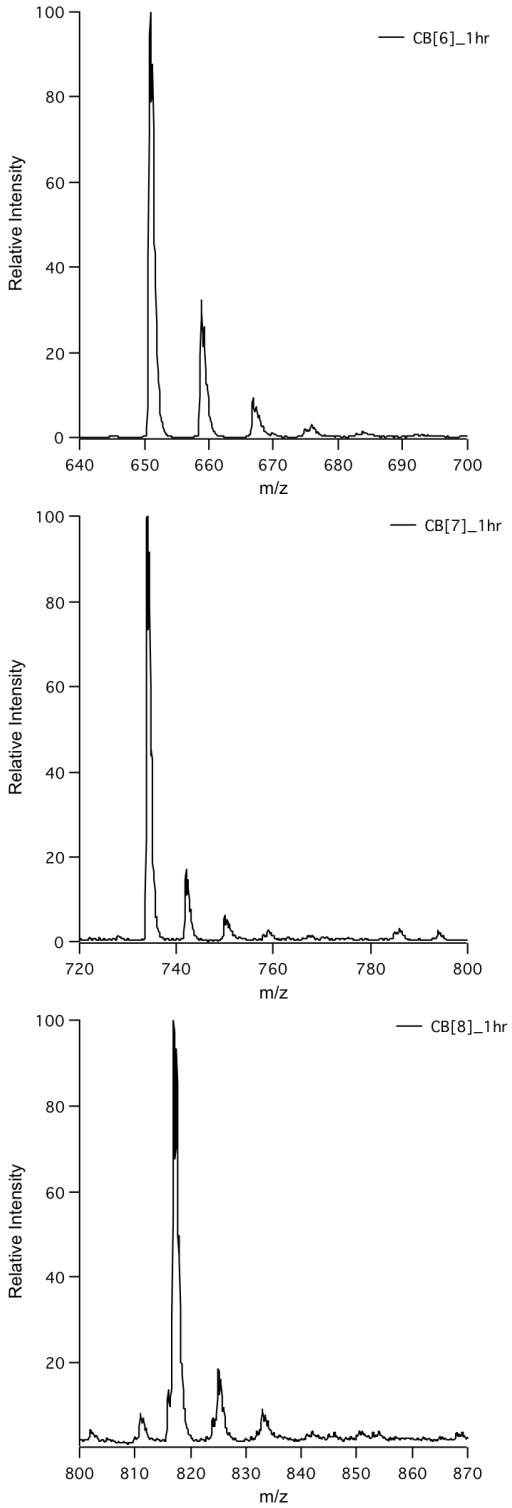

Figure S9: Representative ESI-MS spectra of CB[6]·**G1**, CB[7]·**G1**, CB[8]·**G1** and their respective functionalised derivatives. Spectra are representative of the abundance of each CB[n] homologue and their functionalised derivatives present the reaction mixture containing equimolar quantities of each CB[n] homologue following 1 hr under oxidative reactive conditions.

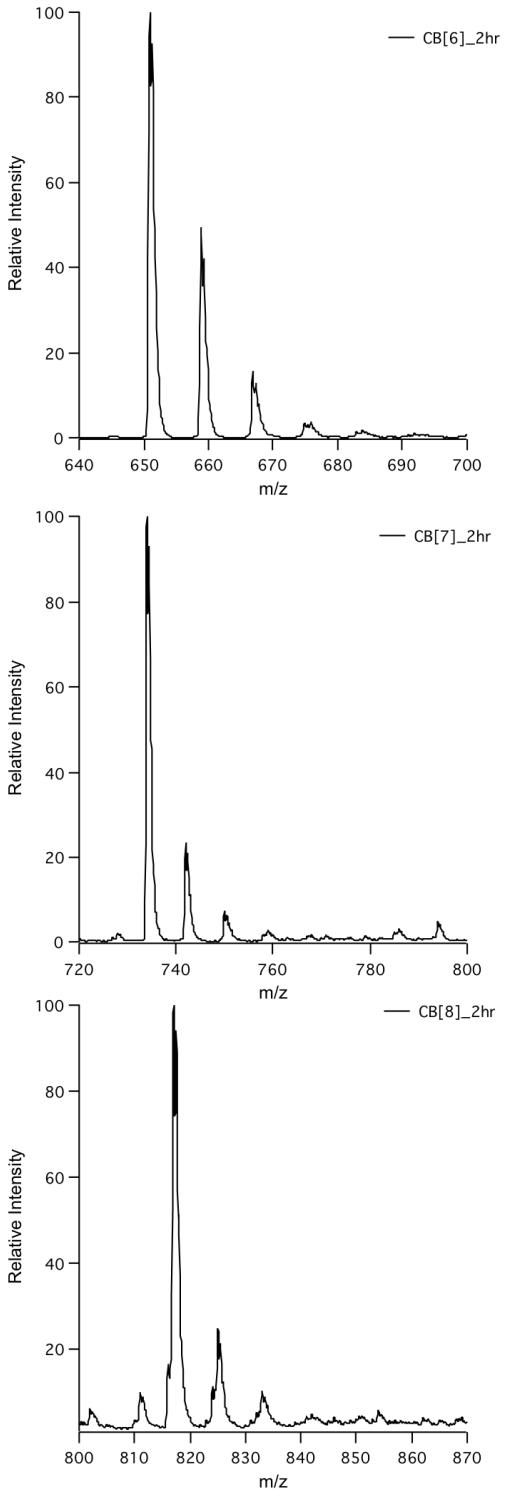

Figure S10: Representative ESI-MS spectra of CB[6]·**G1**, CB[7]·**G1**, CB[8]·**G1** and their respective functionalised derivatives. Spectra are representative of the abundance of each CB[n] homologue and their functionalised derivatives present the reaction mixture containing equimolar quantities of each CB[n] homologue following 2 hr under oxidative reactive conditions.

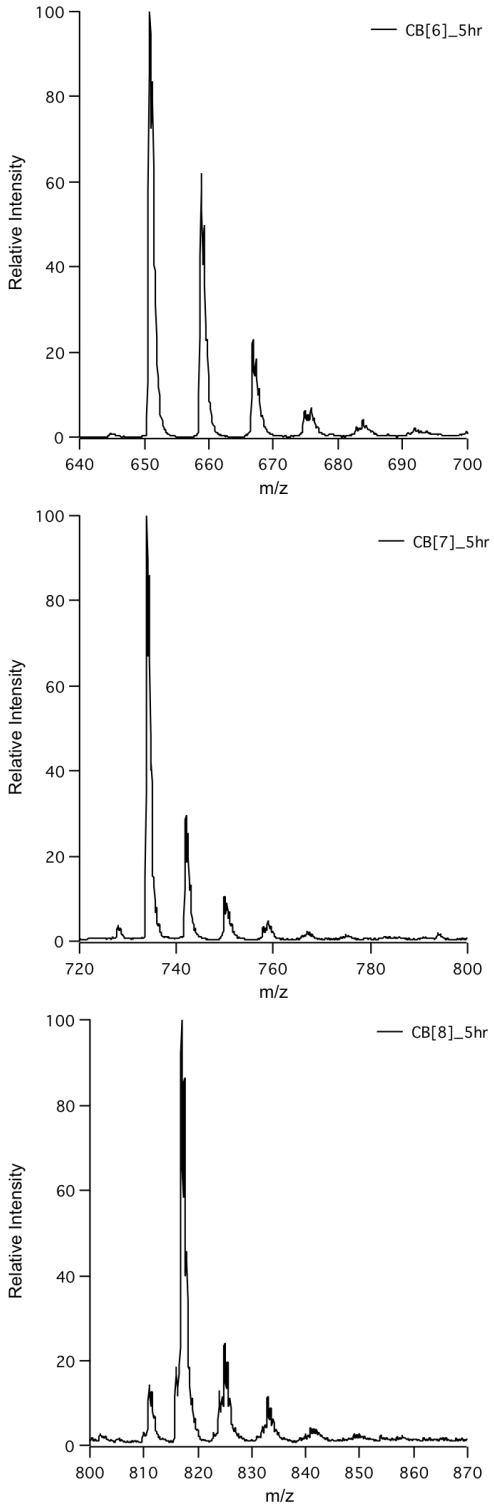

Figure S11: Representative ESI-MS spectra of CB[6]·**G1**, CB[7]·**G1**, CB[8]·**G1** and their respective functionalised derivatives. Spectra are representative of the abundance of each CB[n] homologue and their functionalised derivatives present the reaction mixture containing equimolar quantities of each CB[n] homologue following 5 hr under oxidative reactive conditions.

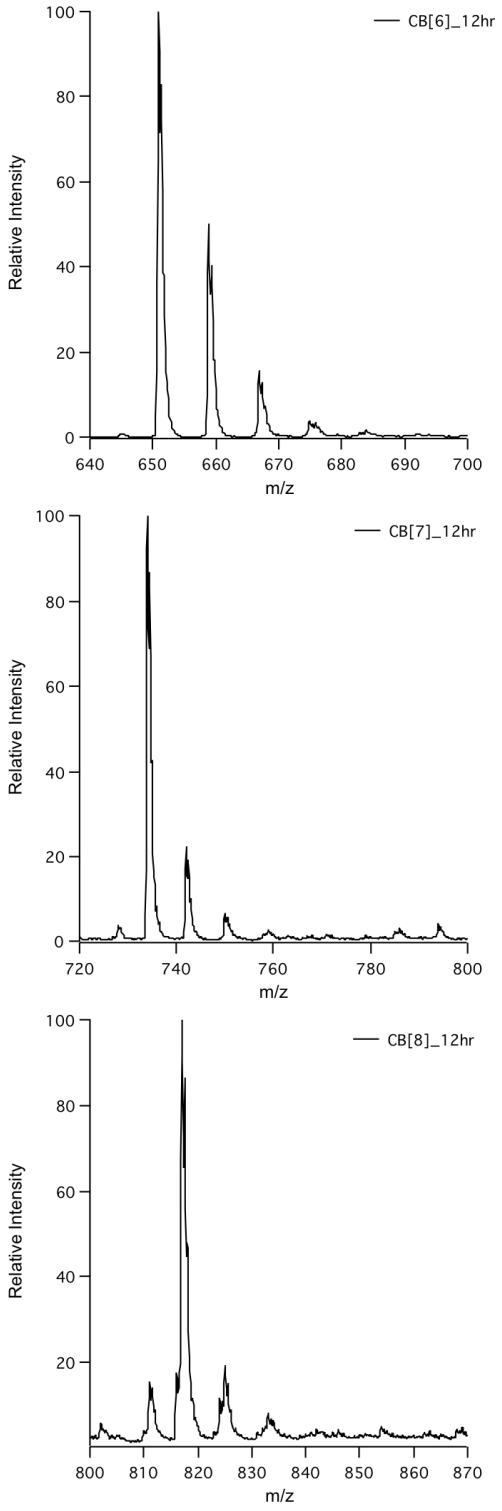

Figure S12: Representative ESI-MS spectra of CB[6]·**G1**, CB[7]·**G1**, CB[8]·**G1** and their respective functionalised derivatives. Spectra are representative of the abundance of each CB[n] homologue and their functionalised derivatives present the reaction mixture containing equimolar quantities of each CB[n] homologue following 12 hr under oxidative reactive conditions.

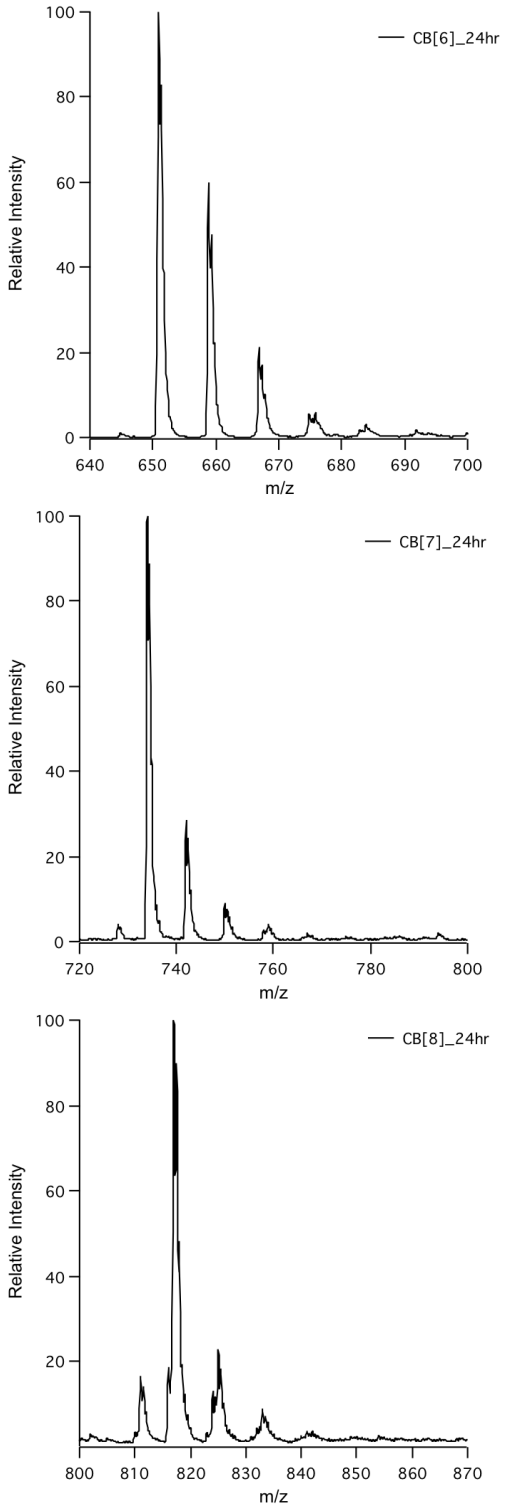

Figure S13: Representative ESI-MS spectra of CB[6]·**G1**, CB[7]·**G1**, CB[8]·**G1** and their respective functionalised derivatives. Spectra are representative of the abundance of each CB[n] homologue and their functionalised derivatives present the reaction mixture containing equimolar quantities of each CB[n] homologue following 24 hr under oxidative reactive conditions.

Table S1: Energies of various CB[7]-$(OH)_2$ structures with second OH group in various positions. [a] Relative energies (kcal/mol) of geometry optimised structures are calculated at the DFT B3LYP-D3 6-31G* level using continuum solvent model. [b] Relative probabilities compared to position 1 were estimated assuming thermodynamically controlled reactions at 298 K temperature.

Position of OH substitution	**Energy**	**Relative probability**
1	0.000	1
2	0.734	0.29
3	0.735	0.29
4	0.758	0.28
5	0.633	0.35
6	0.638	0.34
7	0.563	0.39
8	0.584	0.38
9	0.608	0.36
10	0.640	0.34
11	0.723	0.30
12	0.662	0.33
13	0.581	0.38

References

(1) Zhao, N.; Lloyd, G. O.; Scherman, O. A. *Chem. Commun.* **2012**, *48*, 3070–3072.

(2) Jiao, D.; Biedermann, F.; Scherman, O. A. *Org. Lett.* **2011**, *13*, 3044–3047.

(3) Seino, H.; Mochizuki, A.; Ueda, M. *J. Polym. Sci. Part A: Polym. Chem.* **1999**, *37*, 3584–3590.

(4) Liu, S.; Ruspic, C.; Mukhopadhyay, P.; Chakrabarti, S.; Zavalij, P. Y.; Isaacs, L. *J. Am. Chem. Soc.* **2005**, *127*, 15959–15967.

(5) Wang, R.; Yuan, L.; Macartney, D. H. *Chem. Commun.* **2006**, 2908–2910.