

# Supporting Information

## **Cover and Uncover: Chiral Switching Exploiting Templating and Layer by Layer Grafting**

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## **Experimental section**

### **Materials**

Ethylene glycol dimethacrylate (EGDMA), methacrylic acid (MAA), triethylamine, and ethyl chloroformate were obtained from Aldrich (Deisenhofen, Germany). EGDMA was purified by washing consecutively with 10% aqueous NaOH, water, brine and finally water. After drying over MgSO<sub>4</sub>, pure, dry EGDMA was obtained by distillation under reduced pressure. MAA was purified by distillation under reduced pressure. Methanol and acetic acid (AcOH) were obtained from Fluka (Deisenhofen, Germany). HPLC grade acetonitrile (MeCN) and methanol were purchased from Merck. The templates L- and D-phenylalanine anilide (L-PA, D-PA) were synthesized following a previously described procedure.<sup>1</sup> Anhydrous solvents, tetrahydrofuran (THF), toluene were purchased from Fluka and used as received. The following silica was used as supports. For the RAFT immobilization macroporous beads (Si500) (Fuji Silysia Japan)

(30 $\mu$ m average particle size) with a surface area (S) of 45m<sup>2</sup>/g; an average pore diameter (D<sub>p</sub>) of 50nm and an pore volume (V<sub>p</sub>) of 0.7 mL/g) was used. The RAFT agent 4-cyanopentanoic acid dithiobenzoate was purchased from Strem chemicals Germany.

### **Apparatus and methods**

*HPLC:* The HPLC measurements were carried out on Hewlett-Packard HP 1050 instruments (Agilent Technologie, Waldbronn, Germany).

*Elemental analysis:* Carbon, nitrogen and sulphur contents were determined by elemental analysis at the Department of Organic Chemistry, Johannes Guttenberg Universität Mainz using a Heraeus CHN-rapid analyser (Hanau, Germany).

*SEM:* The particle morphology, size and size distribution were determined using Hitachi H-S4500 FEG Microscope in secondary electron mode with an acceleration voltage of 1 kV at the Department of Biochemical and Chemical Engineering, TU Dortmund. The samples were deposited on holders with a carbon foil without gold sputtering.

*TGA:* Thermogravimetric analysis was carried out using a TGAQ50 (TA instruments, ESchborn, Germany). The sample (10-15mg) was placed in a platinum pan, which is suspended in a sensitive balance together with the reference pan. The sample was then heated in a furnace with at a heating rate of 20 °C/min, under N<sub>2</sub> atmosphere.

*Nitrogen sorption:* Nitrogen sorption measurements were performed on a Quantachrome 4000e (Quantachrome Corporation, Boynton Beach, FL) automatic adsorption instrument. Prior to measurements, 50-100 mg of the samples were heated at 40-60°C under high vacuum (10<sup>-5</sup> Pa) for at least 12 hours. The specific surface areas (S) were evaluated using the multi point BET

method, the specific pore volumes ( $V_p$ ) following the Gurvitch method and the average pore diameter ( $D_p$ ) using the BJH theory applied to the desorption branch of the isotherm.

### Film thickness estimation

The calculation of the film thickness  $d$  (nm) was performed assuming a homogeneous grafted layer as follows.

*From elemental analysis:*

$$(1) \quad d = \frac{m_c \times M_w}{M_c \times \rho \times S} \times 10^3$$

$$(2) \quad m_c = \frac{\%C}{100 - \left( \frac{\%C \times M_w}{M_c} \right)}$$

where  $m_c$  = weight of carbon of the grafted polymer per gram of bare silica support,  $M_w$  = weighted average molecular weight of the grafted polymer assuming stoichiometric incorporation of reactive monomers,  $M_c$  = weighted average molecular weight of the carbon fraction of the grafted polymer,  $\rho$  = weighted average density of monomers ( $\text{g mL}^{-1}$ ) and  $S$  = specific surface area of the bare silica support ( $\text{m}^2\text{g}^{-1}$ ).

*From % mass loss (TGA)*

$$d = \frac{D_p}{2} \left[ 1 - \sqrt{1 - \left( \frac{\%wt. loss}{(100 - \%wt. loss)\rho \cdot V_p} \right)} \right]$$

Where,  $D_p$  = pore diameter of the composite (nm),  $V_p$  = pore volume of the composite ( $\text{mL/g}$ ),

$\rho$  = weighted average density of monomers ( $\text{g mL}^{-1}$ )

(Note: Formula does not fit if mass loss is higher than 40%. For third layer film thickness calculation we took maximum mass loss)

### **Immobilization of RAFT agent (Table S1)**

In a three-necked round bottom flask (500 mL), equipped with a dropping funnel, an overhead stirrer and an ethanol thermometer, was introduced 250 mL dry THF and the flask purged with nitrogen. 795.2mg (2.85 mmol) 4-cyanopentanoic acid dithiobenzoate, 274 $\mu\text{L}$  (309.3mg, 2.85 mmol) ethylchloroformate and 397 $\mu\text{L}$  (288.4mg, 2.85 mmol) triethylamine were consecutively added. The mixture was then cooled at  $-78^\circ\text{C}$  using a liquid-nitrogen-ethanol bath. After stirring for 30 min, 25g of aminommodified silica was added to the mixture and the suspension was stirred for 3h at  $-78^\circ\text{C}$  and then for 4 h at  $-10^\circ\text{C}$ . The product was then filtered, washed with THF and MeOH and dried under vacuum at room temperature. The surface density of RAFT agent calculated based on % mass loss by Thermogravimetry (TGA) was  $1.54 \mu\text{mol}/\text{m}^2$ .

### **Layer by layer grafting of MIPs via RAFT-modified silica (Table S2)**

*Layer 1 (SiP<sub>L</sub>):* Si500-RAFT particles (1g) were suspended in a prepolymerization mixture containing L-PA (7.7mg, 0.032mmol), MAA (22 $\mu\text{L}$ , 0.26mmol) and EDMA (244  $\mu\text{L}$ , 1.3 mmol) dissolved in 20 mL of dry toluene. The polymerization mixture was subjected to three freeze-thaw cycles under nitrogen where after the initiator ABDV (8.7mg), was added. This corresponds to a ratio of RAFT/initiator of 2. Polymerization was initiated at  $50^\circ\text{C}$  for 24h. After polymerization the particles were filtered through a sintered glass funnel and washed with methanol/formic acid/water, 80:15:5(v/v/v) and pure methanol and then the polymer was dried under vacuum at  $40^\circ\text{C}$  overnight.

*Layer 2 (SiP<sub>L</sub>P<sub>D</sub>):* 600mg of layer1 (SiPL) composites were suspended in a prepolymerization mixture containing D-PA (4.6mg), MAA (13.2μL, 0.15mmol) and EDMA (146μL, 0.77mmol) in 12 mL of dry toluene. The polymerization mixture was subjected to three freeze-thaw cycles under nitrogen where after the initiator ABDV was added. This corresponds to a ratio of RAFT/initiator of 2. Polymerization was initiated at 50°C for 24h. After polymerization the particles were filtered through a sintered glass funnel and washed with methanol/formic acid/water, 80:15:5(v/v/v) and pure methanol and then the polymer was dried under vacuum at 40°C overnight.

*Layer 3 (SiP<sub>L</sub>P<sub>D</sub>P<sub>N</sub>):* This was prepared as layer 1 and 2 but in absence of template using a prepolymerization mixture containing MAA (20μL, 0.24 mmol) and EDMA (229μL, 1.21mmol) and 400mg of layer 2 (SiP<sub>L</sub>P<sub>D</sub>) composite. The RAFT/initiator ratio was adjusted to 2.

### **HPLC measurements and evaluation**

The composite materials were typically slurry packed into stainless steel columns (33x4mm or 20x2mm), using MeOH/H<sub>2</sub>O 80:20 (v/v) as pushing solvent, and evaluated chromatographically. The flow rate was adjusted between 0.2mL/min to 0.5mL/min in order to achieve a similar linear velocity for all columns. 10μL aliquots of 1 mM solutions of pure enantiomers or racemate were injected unless otherwise mentioned. The elution was monitored at 260nm. The retention factors ( $k_L$  and  $k_D$ ) and the separation factor ( $\alpha$ ) were calculated using the following formulae:  $k_L=(t_L-t_0)/t_0$ ;  $k_D=(t_D-t_0)/t_0$ ;  $\alpha=k_L/k_D$  where  $t_L$  is the retention time of the L-enantiomer,  $t_D$  is the retention time of the D-enantiomer and  $t_0$  is the retention time of the void marker, acetone.

## Silica removal

Portions (400mg) of the composite materials were suspended in 4 mL of 3M NH<sub>4</sub>HF<sub>2</sub> (aq.) in Teflon flasks. The suspensions were shaken at room temperature for 2 days and then filtered through glass funnel. The resulting polymer was washed with water to remove unreacted NH<sub>4</sub>HF<sub>2</sub> and dried in vacuum oven at 40 °C for 24h.

**Table S1. Characterization of the RAFT modified silica supports used for grafting**

Si-500 support <sup>a</sup>	%wt loss (100-900 <sup>o</sup> C)	Elemental comp.			Area density (Ds) <sup>b</sup> μmol/m <sup>2</sup>	Coverage <sup>c</sup> %
		%C	%N	%S		
Si500APS	2	0.55	0.28		1.72 (2.53)	22 (32)
Si500RAFT	3.9	1.18	0.29	0.35	0.91 (1.54)	11 (19)

a) The immobilizations were performed by ethyl chloroformate catalyzed coupling of 4-cynopentanoic acid dithiobenzoate to amino modified silica beads. The modified supports analyzed by elemental analysis and TGA.

b) The area density (Ds) was calculated from the increase in carbon or average of the increase in sulfur and nitrogen content after the corresponding coupling as:  $D = m_x / (M_x S)$ , where  $m_x = X\% / (100 - X\% M_w / M_x)$ ,  $M_w$  = molecular weight of immobilized silane (Step 1) or 4-cynopentanoic acid dithiobenzoate (Step2),  $M_x$  = weight of carbon (X=C), nitrogen (X=N) or sulfur (X=S) per mole of immobilized species and S = surface area of the silica support. A value in bracket was calculated from the increase in percentage mass loss content after coupling as reported elsewhere<sup>2</sup>

$$Ds (\mu\text{molm}^{-2}) = \frac{\left[ \frac{\%W_{100-900}}{100 - \%W_{100-900}} \right] \times 100 - \%W_{\text{silica}}}{S \times M_w \times 100} \times 10^6$$

Where

$\%W_{100-900}$  = % mass loss of modified silica between 100 °C and 900 °C

$\% W_{\text{silica}} = \% \text{ mass loss of starting silica}$

Mw = Molecular weight of coupled ligand

c) The coverage (C) was calculated as:  $C=100xD/8$ , assuming a maximum silanol group density of  $8\mu\text{mol}/\text{m}^2$ .

**Table S2. Polymer feed composition with respect to nominal film thickness.**

composite Si-500- RAFT	Nominal film thickness value indicated in bracket (nm)			Silica Support	Template (LPA/DPA) mg	Monomer (MAA) $\mu\text{L}$	Crosslinker (EGDMA) $\mu\text{L}$	RAFT/ABDV	Toluene mL
	Layer1	Layer2	Layer3						
SiP <sub>L</sub>	L-PA (5)	--	--	1g	7.7	22	244	2	20
SiP <sub>L</sub> P <sub>D</sub>	--	D-PA (5)	--	600mg	4.6	13	146	2	12
SiP <sub>L</sub> P <sub>D</sub> P <sub>N</sub>	--	--	NIP (15)	400mg	--	20	229	2	8

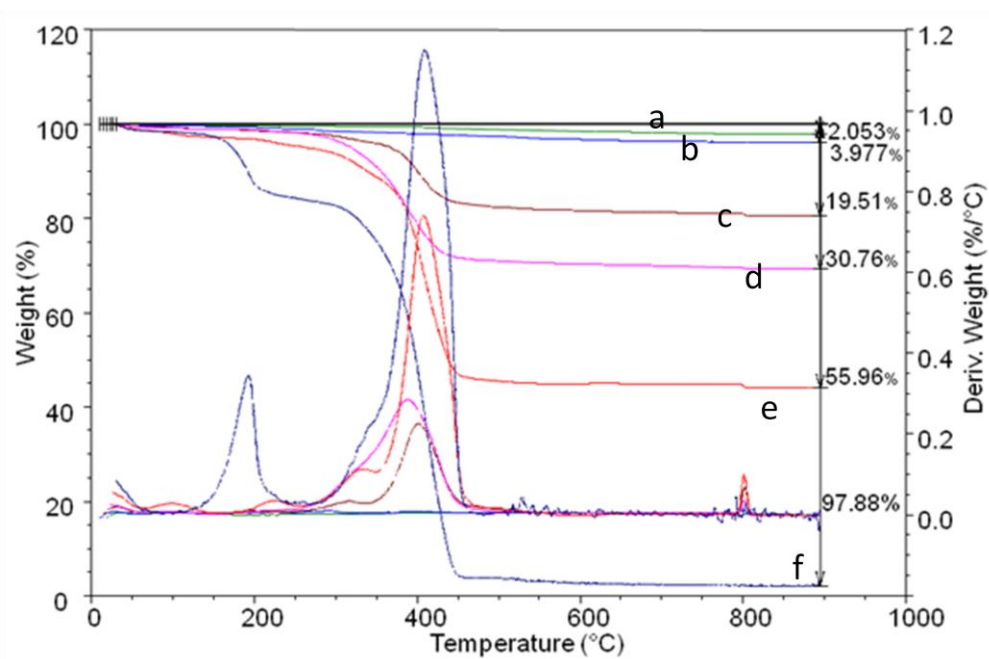


Figure S1. TGA curves of (a) aminosilica (b)RAFT modified silica (c) SiP<sub>L</sub>(Layer1), (d) SiP<sub>L</sub>P<sub>D</sub> (Layer2), (e) SiP<sub>L</sub>P<sub>D</sub>P<sub>N</sub> (Layer3), (f) P<sub>L</sub>P<sub>D</sub>P<sub>N</sub>, after removal of silica by etching.

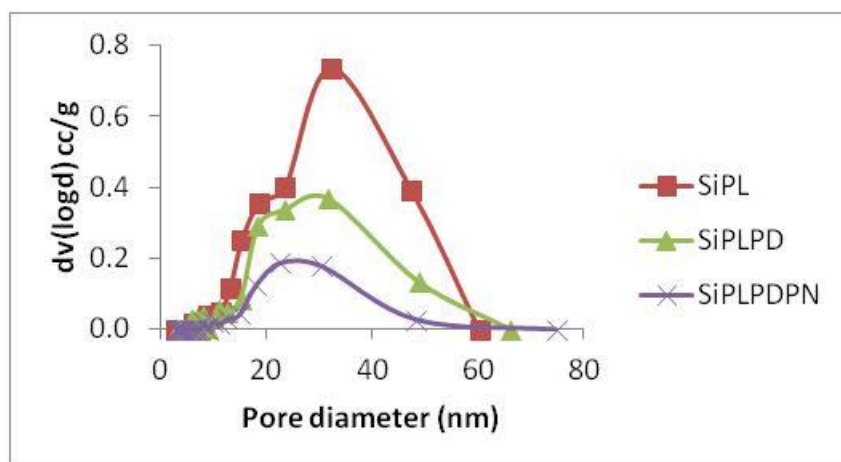


Figure S2. Typical pore size distribution of consecutive grafted layer.



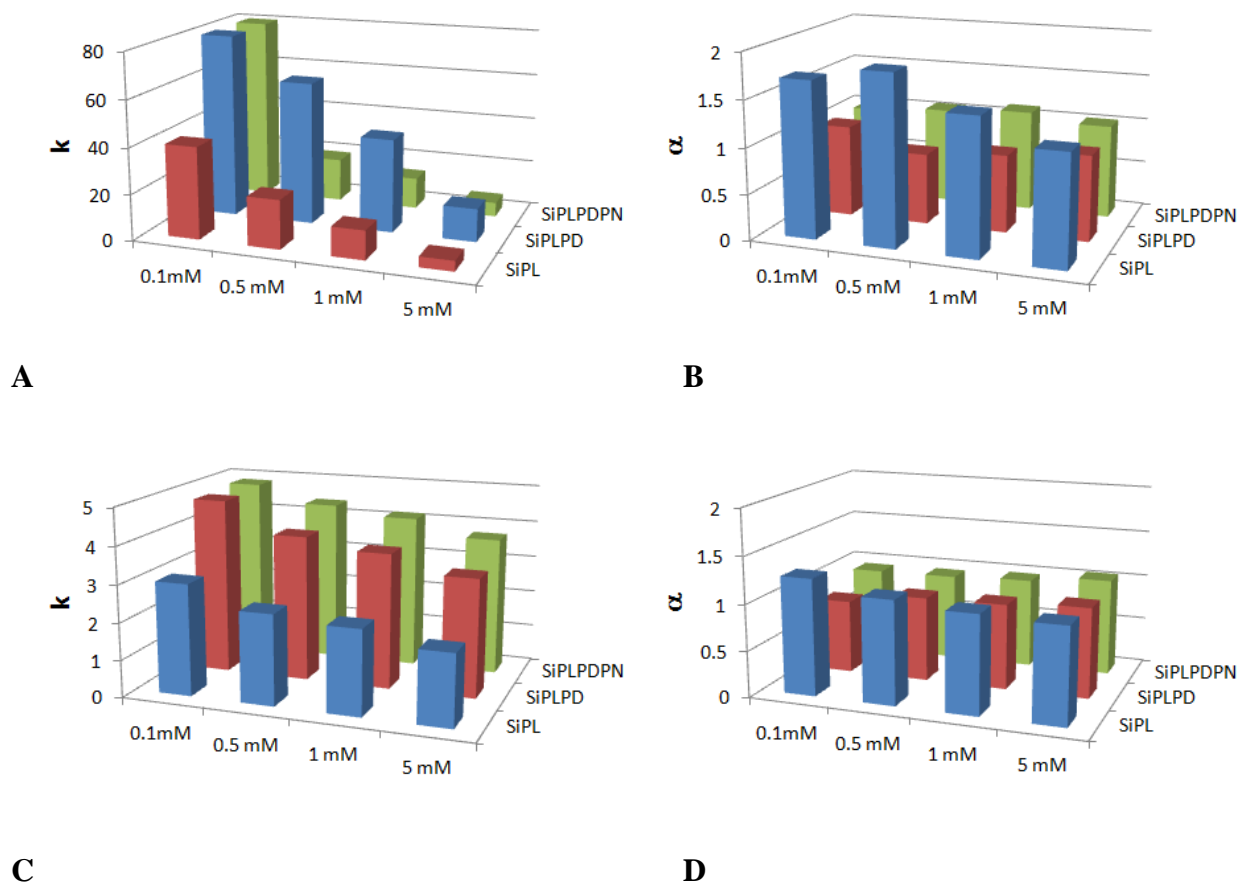


Figure S3: Retention factors ( $k_L$ ) for L-PA (A,C) and enantiomer separation factors ( $\alpha$ ) (B,D) obtained in the chromatographic mode after separate injection of stock solutions of D- and L-PA at four different sample loads for corresponding imprinted composites beads prepared by consecutive grafting from RAFT modified support. Mobile phase: (A, B) acetonitrile, (C,D) MeCN/Sodium acetate buffer pH 4.8(70/30,v/v)

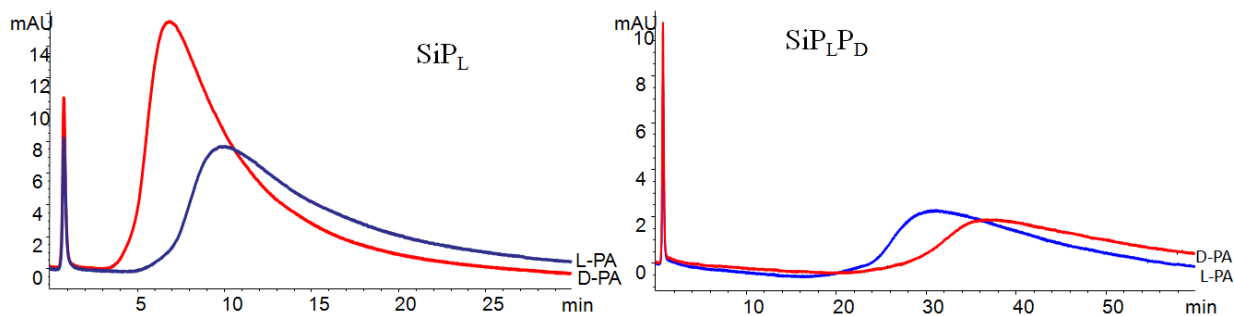


Figure S4. Elution profiles of D (red trace), L (blue trace) phenylalanine anilide (PA) injected (10 $\mu$ L of 1mM solution) separately on columns (35mm x 4.6 mm) packed with the material indicated. Mobile phase: Acetonitrile, flow rate 0.5mL/min, DAD=260 nm.

1. C. Sulitzky, B. Ruckert, A. J. Hall, F. Lanza, K. Unger and B. Sellergren, *Macromolecules*, 2002, **35**, 79-91.
2. C. Bartholome, E. Beyou, E. Bourgeat-Lami, P. Chaumont and N. Zydowicz, *Polymer*, 2005, **46**, 8502-8510.