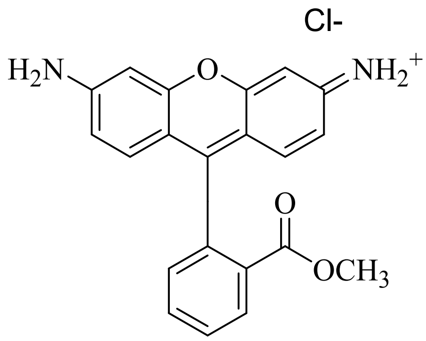
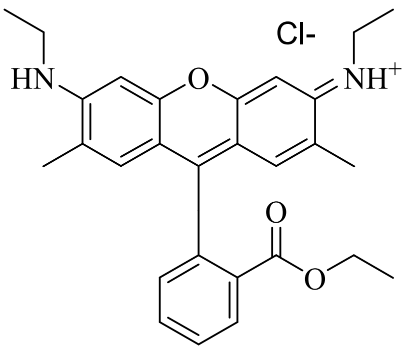
Electronic Supplementary Material (ESI) for Nanoscale.

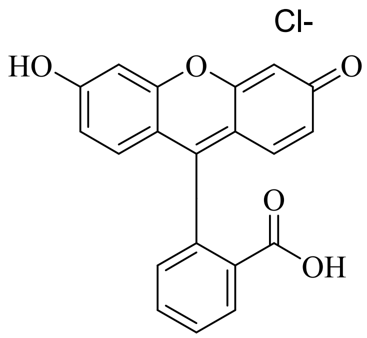
SUPPORTING INFORMATION



Rhodamine 123 (R123)



Rhodamine 6G (R6G)



Fluorescein (FLU)

Figure SI.1. Chemical structure and acronyms of the different fluorescent dyes.

Optimization of polymer composition

Imprinted polymers were prepared by bulk polymerization keeping constant the template/functional monomer molar ratio (0.5:4) and varying the concentration of cross-linker (T/FM/CL, 0.5:4:2.5–16) (Table SI.1). After monolith grinding, the polymers (20 mg) were shaken in 25 mL of ethanol for 2 h for template removal. The use of acidic alcohol solutions, that favor MAA protonation and thus template release from the polymer network, were avoided to prevent degradation of the metallic layer. The amount of extracted template was monitored by HPLC (see LC-UV analysis section) and the recoveries (%) are summarized in Table SI.1. When the amount of cross-linker (%) with respect to the total amount of monomers involved in the polymerization was lower than 40%, bulk polymers were not obtained. However, for concentrations higher than 50%, template recovery decreased down to 63% for 80% CL.

Table SI.1. Template recovery after polymerization. Washing conditions: 2 h shaking in ethanol. T: template. FM: functional monomer. CL: cross-linker (n = 3).

T/FM/CL mole ratio	CL (%)	Template recovery (%)	RSD (%)
0.5:4:2.5	38	No polymerization	---
0.5:4:4	50	91	4.3
0.5:4:5	55	83	5.5
0.5:4:10	71	72	4.7
0.5:4:12	75	69	4.4
0.5:4:16	80	63	5.3

In agreement with these results, an increase in the CL concentration in the three dimensional polymer network leads to a more efficient entrapment of R123. In this way the extracting solvent has less accessibility to the binding cavities in the polymer decreasing the extraction recoveries. A molar ratio 0.5:4:4, T/FM/CL, was selected for the synthesis of the MIP nanostructures.

Determination of the binding capacity by equilibrium rebinding analysis

The experimental data were fitted to the Freundlich (FI) isotherm model [Yan, M.; Ramström, O., Molecularly Imprinted Materials: Science and Technology. Marcel Dekker, New York, 2005] (Eq. 1):

$$B = aF^m \qquad \text{(Eq. 1)}$$

where *B* is the concentration of bound ligand, *F* the concentration of free ligand and *a* is a Freundlich parameter related with the binding affinity and *m* represents the heterogeneity index of the polymer and can take values from 1 to 0, increasing with decreasing heterogeneity of the material.

The affinity distribution (AD) [Rampey, A. M.; Umpleby, R. J.; Rushton, G. T.; Iseman, J. C.; Shah, R. N.; Shimizu, K. D. *Anal. Chem.* **2004,** 76, 1123-1133] was calculated using Eq. (1) and the experimentally derived FI fitting parameters (*a* and *m*):

The average affinity constant, *K*, and the average number of binding sites, *N*, was calculated using Eqs 2 and 3:

$$K = \left(\frac{m}{m-1}\right)\frac{K_1^{1-m} - K_2^{1-m}}{K_1^{-m} - K_2^{-m}} \qquad \text{(Eq. 2)}$$

$$N = a\left(1 - m^2\right)\left(K_1^{-m} - K_2^{-m}\right) \qquad \text{(Eq. 3)}$$

Calculation of the propagation length of surface plasmon polariton (SPP) at the aluminum/pre-polymerization mixture interface at 532 nm wavelength

The SPP propagation length (δ_{SP}) at a metal/dielectric interface can be found from the following equation [Raether, H. Surface Plasmons (ed. Hohler, G.) (Springer, Berlin, 1988)]:

$$\delta_{SP} = \frac{\lambda}{2\pi}\left(\frac{\varepsilon_m^r + \varepsilon_d}{\varepsilon_m^r \varepsilon_d}\right)^{\frac{3}{2}} \frac{\left(\varepsilon_m^r\right)^2}{\varepsilon_m^i} \qquad \text{(Eq. 4)}$$

where λ is the free-space wavelength, ε_m^r and ε_m^i are the real and imaginary parts of the dielectric function the metal and ε_d is the dielectric function of the dielectric material.

For Al, ε_m^r (λ=532 nm) = -37.7 and ε_m^i (λ=532 nm) = 11.2 [from A. D. Rakić, A. B. Djurišic, J. M. Elazar, and M. L. Majewski. Optical properties of metallic films for vertical-cavity optoelectronic devices, *Appl. Opt.* **37**, 5271-5283 (1998)]; for the pre-polymerization mixture, ε_d (λ=532 nm) = 1.4. Therefore, from Eq. 4: $\delta_{SP} = 3.6$ μm.