Supporting information

Spotting of the array was carried out using a spotting robot (SpotBot, Telechem, Sunnyvale, CA) equipped with Megasonic Wash Station and a 50 μm pin (SMP2, Telechem). Standard printing procedure was used. Appropriate samples were spotted onto gold substrates, which were prepared by thermal e-beam deposition of chromium (100 Å) followed by gold (1000 Å, 99.99% purity) onto a silicon wafer. To ensure proper humidity, the substrates were placed on top of a moist filter paper inside a Petri dish. The Petri dish was then wrapped with Parafilm and incubated for 48h at 5°C. Detailed spotting parameters are listed below.

Spot spacing (center to center): 250 μm
Pre-print spots per sample: 10
Sample loading time: 3 s
Pre-print time: 0 s
Print time: 0.2 s
Number of wash/dry cycles: 6
Wash/dry duration: 0.5 s
Last cycle wash duration: 2 s
Last cycle dry duration: 10 s

SECM experiments were carried out with a CHi-900b scanning electrochemical microscope (CH Instruments, Austin, TX). A standard three-electrode cell was used with the 25 μm diameter Pt tip electrode (RG = 2, see supporting information for details), a platinum wire as the counter electrode, and a Ag/AgCl (CH Instruments) as the reference electrode. All measurements were performed in 20 mM Tris-ClO_4 (pH 8.6) containing 1 mM $K_4[Fe(CN)_6]$, 50 mM NaClO4, proceeded by rinsing a sample with copious amounts of 20 mM tris-ClO_4 (pH 8.6). The tip potential was held constant at ET = 0.5 V to oxidize $[Fe(CN)_6]^{4-}$ at a diffusion controlled rate.

Approaching the substrate. Prior to SECM imaging experiments the tip electrode was approached towards an unmodified section of the gold substrate. As the tip-to-sample separation distance, d, decreased instantaneous tip current, I_T, increased due to the recycling of $[Fe(CN)_6]^{3-/4-}$ redox couple between the tip and substrate. The approach was stopped when a normalized tip current $I_T/ I_{T,\infty} = 1.3$ was obtained ($I_{T,\infty}$ - tip current observed in the bulk solution). Positive feedback SECM theory was employed to estimate d (see supporting information). For $I_T/ I_{T,\infty} = 1.3$ separation distance was found to be about 15 μm. After initial positioning and approach, the tip was scanned above the substrate and the recorded tip current was normalized by $I_{T,\infty}$ before plotting. Fig. S1 shows experimental data recorded on approach towards clean gold surface (×) with 25 μm dia. Pt tip. The solid line represents calculated approach curve which was obtained using positive feedback SECM theory.[1] The fit between experiment and theory is very good, therefore, it was used to determine tip-to-sample separation distance and tip electrode RG factor (RG is defined as ratio of the insulator radius to the disk electrode radius). The best fit was obtained for theoretical curve calculated for RG = 2. Dashed line indicates tip-sample separation distance, d = 15 μm ($I_T/I_{T,\infty} = 1.3$), which was used as scanning distance in all SECM imaging experiments.

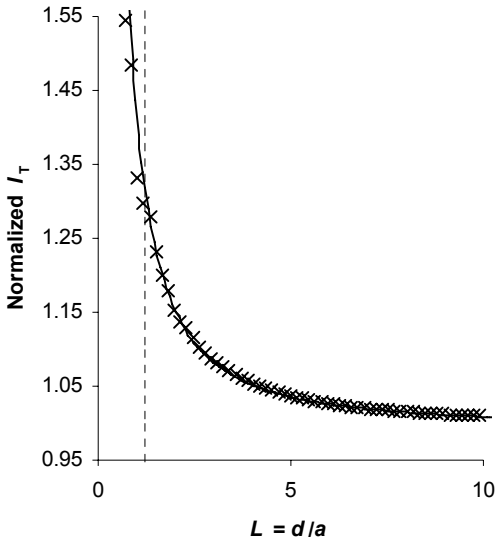

Fig. S1 Normalized approach curve recorded above Au substrate (x). Solid line represents simulated approach curve for RG = 2. Data obtained in 1 mM $K_4Fe(CN)_6$, 50 mM $NaClO_4$, 20 mM tris-ClO_4 (pH 8.6), E_T = 0.5 V.

Diffusion coefficient. Diffusion coefficient for $[Fe(CN)_6]^{3-}$/ $[Fe(CN)_6]^{4-}$ $D = 3.09 \times 10^{-6}$ cm^2s^{-1} was determined from steady-state limiting current as given by:
$i = 4nFDCr$
where i is the measured current, n is the number of electrons, F is the Faraday constant, D is the diffusion coefficient, C is the concentration of the redox mediator and r is the radius of the electrode.

References

[1] A. J. Bard, M. V. Mirkin, *Scanning electrochemical microscopy*, Marcel Dekker, New York, **2001**