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## **Electronic Supplementary Information**

## Site-specific immobilization of DNA on silicon surfaces by thiol-yne reaction

Jorge Escorihuela, María-José Bañuls, Rosa Puchades and Ángel Maquieira\*

Centro de Reconocimiento Molecular y Desarrollo Tecnológico, Departamento de Química, Universitat Politècnica de València, Camino de Vera s/n, 46022 Valencia, Spain. \*To whom correspondence should be addressed. Tel: +34 963879349; Fax: +34 963879349; E-mail: amaquieira@qim.upv.es



**Fig. S1.** WCA evolution for the functionalization of the epoxy-terminated surface with propargylamine.



**Fig. S2.** Fluorescence intensity *vs* probe concentration for different irradiation times, after irradiation of Cy5-labeled, thiol-modified oligonucleotide (Probe A) immobilized on the alkyne-functionalized silicon surface.



**Fig. S3.** Standard calibration curve for oligonucleotide immobilization density calculation for (A) Probe A and (B) Probe B.

Fluore





**Fig. S4.** Images of static water contact angles on epoxy (A), alkyne (B) and DNA (C) immobilized on silicon.



**Fig. S5.** Comparison of Probe A droplets (20 nL) on alkyne (A), alkene (B) and epoxy (C) terminated silicon surfaces.



Fig. S6. XPS spectra of raw (black), epoxy (red), alkyne (blue) and DNA (green) modified silicon surfaces.

Sample	Si (%)	O (%)	C (%)	N (%)	S (%)
Bare SiO <sub>2</sub>	29.5	54.3	16.2		
Epoxy-modified	28.6	50.5	20.9		
Alkyne-modified	22.4	39.2	33.3	5.1	
ssDNA attached	22.0	33.9	38.4	4.4	1.3
dsDNA attached	20.7	30.0	42.6	5.1	1.6

Table S1. Quantitative XPS analysis of atomic composition for bare, epoxy, alkyne and DNA functionalized (1  $\mu$ M) Si substrates.



**Fig. S7.** XPS high-resolution C 1s spectrum of epoxy (A), alkyne (B) ssDNA (C) and dsDNA (D) modified silicon surfaces.



Fig. S8. Partial IRRAS spectrum of alkyne functionalized silicon surface.



**Fig. S9.** AFM images (2  $\mu$ m × 2  $\mu$ m; z-range 100 nm) recorded in air for: epoxy-functionalized (A), alkyne-terminated (B), ssDNA-functionalized (C) and dsDNA-functionalized (D) silicon surfaces.

Sampla	<b>RMS roughness</b>	Average roughness	Peak to valley	
Sample	(nm)	(nm)	(nm)	
Bare SiO <sub>2</sub>	$0.28 \pm 0.08$	$0.25\pm0.07$	1.16	
Epoxy-modified	$0.21\pm0.02$	$0.17\pm0.04$	0.62	
Alkyne-modified	$2.10\pm0.03$	$2.29\pm0.08$	1.34	
ssDNA attached	$3.24\pm0.09$	$3.09\pm0.07$	3.94	
dsDNA attached	$3.47\pm0.08$	$3.27\pm0.09$	4.33	

Table S2. RMS roughness measured by AFM on  $SiO_2$  surfaces.



**Fig. S10.** Optimization of hybridization temperature of Cy5-labeled target. Hybridization time was kept at 1h.



## Norma

**Fig. S11.** Relative fluorescence intensity of Cy5-labeled target at different hybridization times. Hybridization temperature was kept at 37 °C.



Fig. S12. Standard calibration curve for oligonucleotide hybridization density calculation.



**Fig. S13.** Hybridization results for different Target A concentrations (1000-1 nM) at several Probe A spotted concentrations.



**Fig. S14.** Signal intensity profile for immobilized microarrays stored at 4 °C for 1 to 8 weeks before performing the hybridization assay.



**Fig. S15.** Variation of thickness (nm) and mass (ng/mm<sup>2</sup>) after DNA hybridization monitored by DPI.



Fig. S16. Effect of formamide content in the detection of SNPs using different hybridization conditions: (A)  $3 \times SSC$  and (B)  $0.1 \times SSC$ 



Fig. S17. Detection of bacterial Escherichia coli.