Supporting information

Paper-Based Strips for the Electrochemical Detection of Single and Double Stranded DNA

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Abstract

The Supporting Information file shows the following information: photogtaphs related to the synthesis of gold nanoparticles (AuNPs) starting from precursors and the final aspect that AuNPs should have (Figure S1), the UV-Spectra related to the plasmon resonance band of the synthesized AuNPs (Figure S2), the schematic representation of the single- and double-stranded DNA targets that have been analyzed during work (Figure S3), the square wave voltammograms obtained in presence of different concentrations of hairpin target (Figure S4), the evaluation of the repeatability of the paper-based device obtained by testing 8 replicates (Figure S5), the effect of bivalent cations in triplex formation (Figure S6), and the study of the selectivity (Figure S7). In addition, all the analytical information regarding the fit equation, the correlation coefficient, the (apparent) binding constants and the calculated detection limits for each platform have been reported in Table S1.

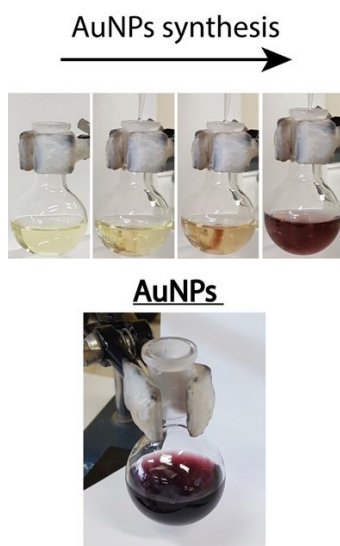### AuNPs: synthesis and characterization

Figure S1. Color variation during the reduction of gold precursor by means of sodium borohydride addition in presence of citrate ions as the capping agents. The synthesized AuNPs display their characteristic color.
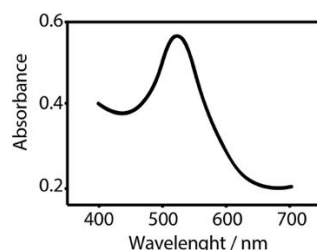
Figure S2. UV-Vis spectra for 10-fold diluted gold nanoparticles in aqueous solution.

Due to the high concentration of gold nanoparticles, the dispersion has been diluted 10-fold, in order to stay in the linear range of the Lambert-Beer law. The spectrum is consistent with the presence of the characteristic plasmonic surface resonance band of the gold nanoparticles centered at 523 nm.

Single and double stranded targets

Figure S3. Single stranded (ssDNA) and double stranded (dsDNA, hairpin) DNA utilized as targets during detection.

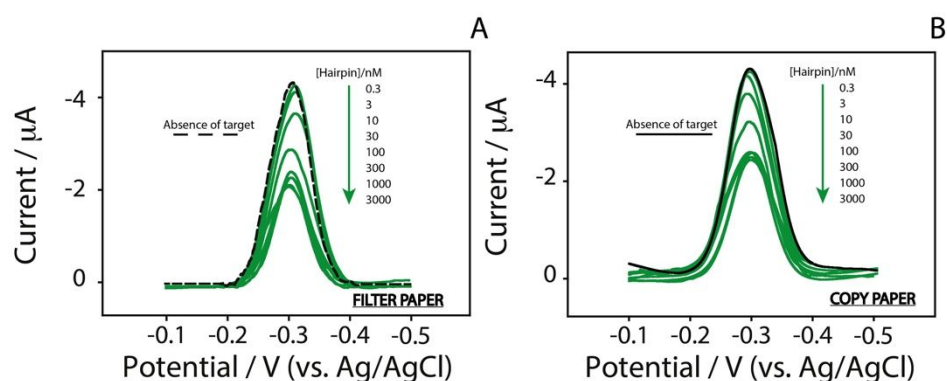## Square Wave Voltammograms in presence of hairpin

Figure S4. Square Wave Voltammograms obtained by using by using electrodes screen-printed onto A) filter paper and B) copy paper. Hairpin has been chosen as the target in the 0.3-3000 nM range of concentration.
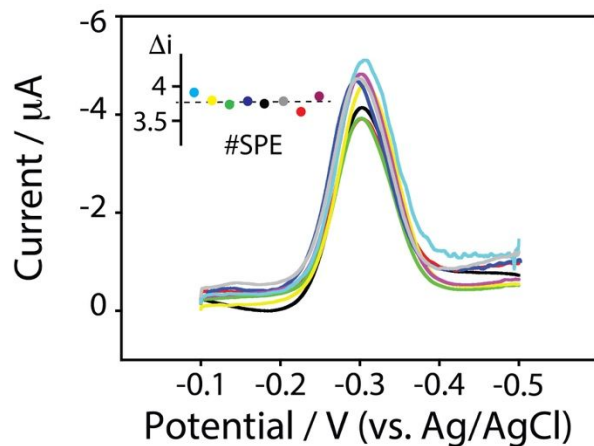
Repeatability of the paper-based platform

Figure S5. Repeatability study by evaluating the initial response of 8 different paper-based SPEs in buffer solution.

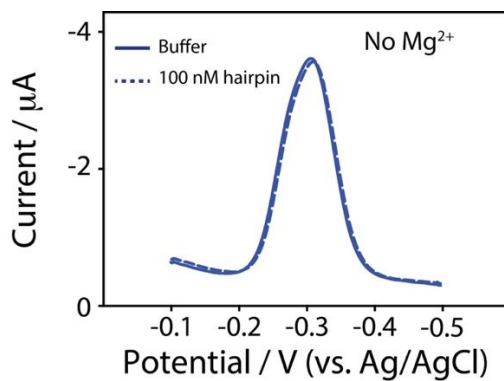## Effect of Mg^{2+} on triplex formation

Figure S6. Analysis of 100 nM hairpin in absence (dashed line) of magnesium ions in the working solution. The solid line corresponds to the blank analysis in absence of target.
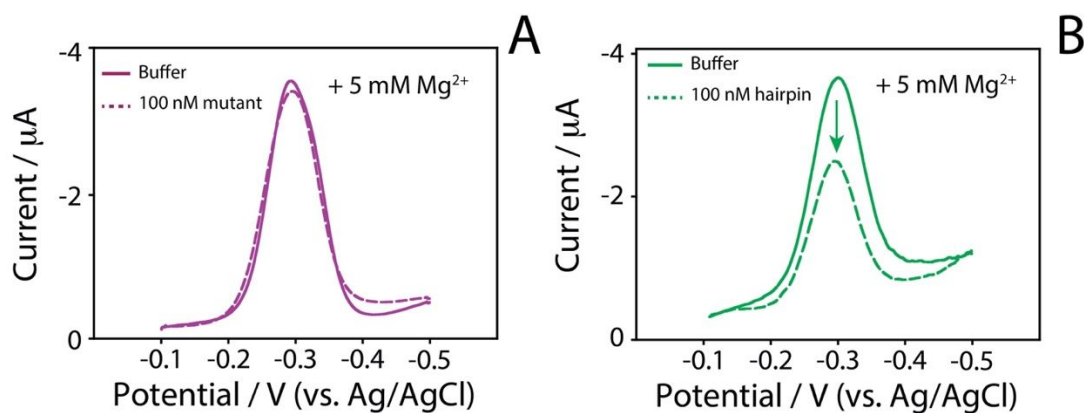
Selectivity study

Figure S7. A) Analysis of 100 nM mutated hairpin (purple dashed line); B) Signal suppression in presence of 100 nM hairpin (green dashed line). All the measurements have been carried out in presence of 5 mM magnesium ions in the working solution, and thensolid lines correspond to the blank analysis in absence of targets.

Table S1. Experimental and analytical parameters for single-stranded and double-stranded DNA detetion at paper-based electrochemical strips.

Target	Paper	Working solution	Fit Equation*	R^2	Binding costants (nM)	Detection limit (nM)
ssDNA	Copy	Buffer	$y = 0.01 + \dfrac{1.04x^{0.97}}{18^{0.97} + x^{0.97}}$	0.998	18±2	3.1±0.3
dsDNA	Copy	Buffer	$y = 0.05 + \dfrac{0.97x^{0.91}}{44^{0.91} + x^{0.91}}$	0.986	45±5	6.8±0.7
Hairpin	Copy	Buffer	$y = 0.04 + \dfrac{1.06x^{1.03}}{46^{1.03} + x^{1.03}}$	0.991	46±6	7.0±0.7
ssDNA	Filter	Buffer	$y = 0.02 + \dfrac{0.97x^{1.50}}{20^{1.50} + x^{1.50}}$	0.994	20±2	3.2±0.3
dsDNA	Filter	Buffer	$y = 0.02 + \dfrac{1.01x^{1.79}}{49^{1.79} + x^{1.79}}$	0.997	49±5	7.1±0.7
Hairpin	Filter	Buffer	$y = 0.01 + \dfrac{0.98x^{1.80}}{53^{1.80} + x^{1.80}}$	0.990	53±6	7.3±0.6
Hairpin	Filter	Human serum	$y = 0.01 + \dfrac{1.01x^{1.92}}{56^{1.92} + x^{1.92}}$	0.992	56±6	7.9±0.8
Synthetic PCR amplified	Filter	Human serum	$y = 0.01 + \dfrac{1.07x^{1.24}}{230^{1.24} + x^{1.24}}$	0.995	230±30	30±4

* y represents the normalized signal change, x represents the target concentration expressed in nM