

## SUPPORTING INFORMATIONS

### SERS-active metal-dielectric nanostructures integrated in microfluidic devices for label-free quantitative detection of miRNA

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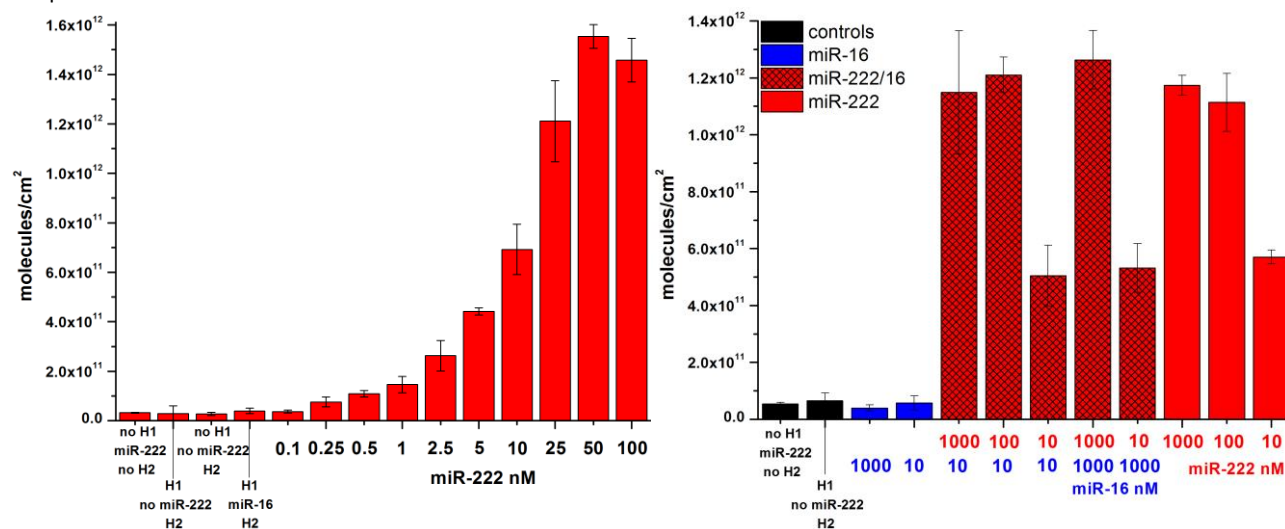
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**ELISA-like detection of different concentration of miR-222 by the two-step hybridization protocol.** The two-step hybridization protocol was applied to the detection of different concentrations of miR-222 by ELISA-like assay. The results of this calibration are reported in Fig. S1a. This test clearly demonstrated that the optimized two-step hybridization protocol allows to specifically detect the target miRNA in a dynamic range of concentrations of 0.25-50 nM. Moreover, in comparison with the one-step protocol previously performed on commercial 96-well plates, or on the properly functionalized PSD substrates<sup>1</sup>, the two-step assay revealed a similar dynamic range and sensitivity.

In order to confirm the specificity of this two-step bioassay, a test with various mixtures of miR-222 and miR-16 at different concentrations was performed. In Fig. S1b the results of this test are illustrated. This plot clearly denote that there is no influence of other interfering miRNA sequences on the specific detection of the miR-222 by using this two-step hybridization assay. In fact, the signal recorded for the target miR-222 alone at different concentrations is not significantly different from the one recorded in the presence of the miR-16. Indeed, even the presence of 1  $\mu$ M miR-16 did not impair the detection of the miR-222 at concentration as low as 10 nM. This result is fundamental for a real application of this bioassay to the miRNA profiling into cellular samples.



**Figure S1.** a) Detection of different concentration of miR-222 by the two-step hybridization assay; b) ELISA-like assay of the PSD substrates incubated with different concentrations of the target miR-222 incubated alone, or mixed with various concentration of miR-16.

### References

- 1 A. Chiadò, C. Novara, A. Lamberti, F. Geobaldo, F. Giorgis and P. Rivolo, *Anal. Chem.*, 2016, **88**, 9554–9563.