Electronic Supplementary Material (ESI) for Analyst.

Supplementary Information

Label-free detection of nosocomial bacteria using a nanophotonic interferometric biosensor

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1. XPS analysis

The XPS experiments were performed using a Phoibos 150 analyzer (SPECS GmbH, Berlin, Germany) in ultra-high vacuum condition (base pressure 1×10^{-10} mbar). A monochromatic $Al_{K\alpha}$ X-ray source (1486.74 eV) operating at 400 W was employed. Survey spectra were collected from 0 to 1380 eV with a pass energy of 50 eV, and high-resolution spectra were collected for each element (e.g. C, N and O) with a pass energy of 20 eV. Survey and high-resolution spectra were collected at 0º take off angle, defined as the angle spanned by the electron path to the analyser and the sample surface. The spectra were obtained at room temperature.

XPS survey spectra illustrated significant changes in carbon, silicon, oxygen and nitrogen peaks, due to the addition of the different molecular layers (see Figure S2).[1] The spectra with the antibody immobilized shows a significant increase of the carbon and decrease of the nitrogen and the silicon, both being contributions from the Si_3N_4 substrate and the SiO_2 oxidation layer. This demonstrates how Si_3N_4 is not exposed but coated with silane-PEG-COOH and antibodies. The elemental analysis indicates a remarkable increase of carbon content after antibody immobilization. Carbon increased from 6.2% (Si_3N_4) to 20.7% on the antibody coated surface while nitrogen levels decreased on the silanized surface and was even lower with the antibody attached (see Table S1). The high-resolution C 1s spectra (see Figure S3) showed that the C 1s photoelectron peaks are much broader after the immobilization with antibodies. The C 1s peaks were deconvoluted according to binding energies of carbons in antibody. The first peak was centred at 284.7 (Figure S3a) and related to aliphatic carbon (C-C). The second peak at 286.5 eV corresponds to carbon bonded to oxygen group (C-O) (Figure S3b) which evidences the PEG-silane-COOH presence. The antibody attachment by the peak around 287.5 eV, corresponds to –C (-O)-NH_2 peptide carbon (Figure S3c).[2, 3] The binding energies of N 1s detected on the samples (close to 400 eV) are typical for organic materials and are related to C-N bonds, [4, 5] at 398.2

eV, the spectre corresponding to Si_3N_4 (see Figure S4a). At the same time, the silane-PEG-COOH (Figure S4.b) and the antibody onto Si_3N_4 (Figure S4c) are indicative of complete coverage of the silicon nitride surface. The component around 402 eV peak was assigned to imide-N, found on the amino acid proteins (see Figure S4.c). [6]

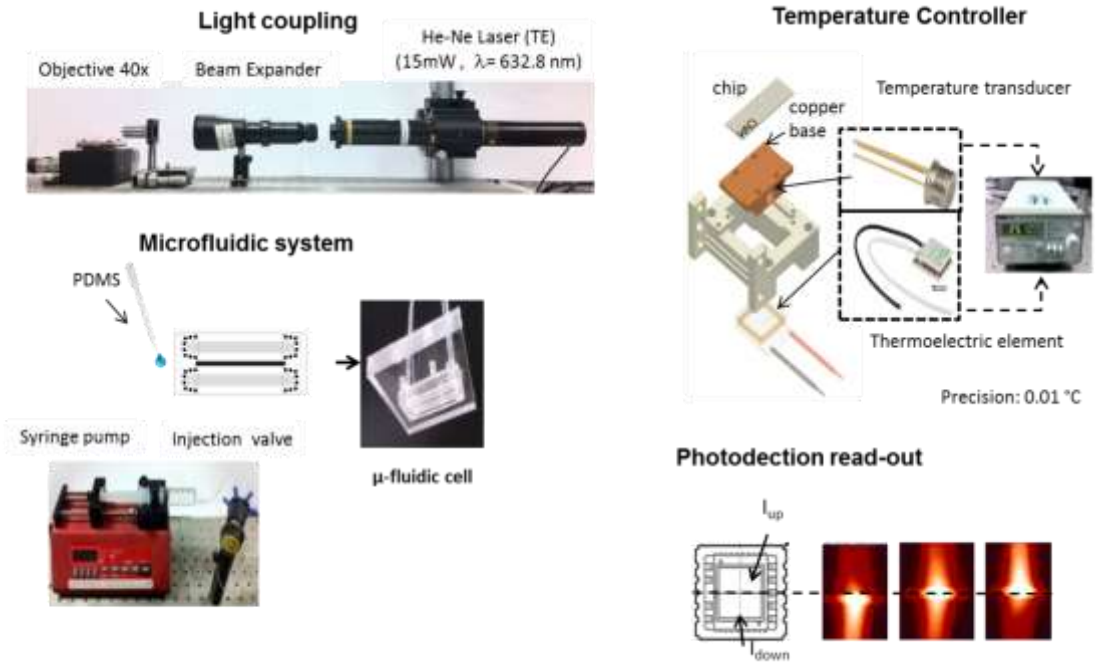

Figure S1. Schematic representation of the experimental set-up for the evaluation of the BiMW biosensors system. The set-up comprised three mainly components: a flow cell and flow delivery system, optical system for in coupling of the light and the read-out and data acquisition system.

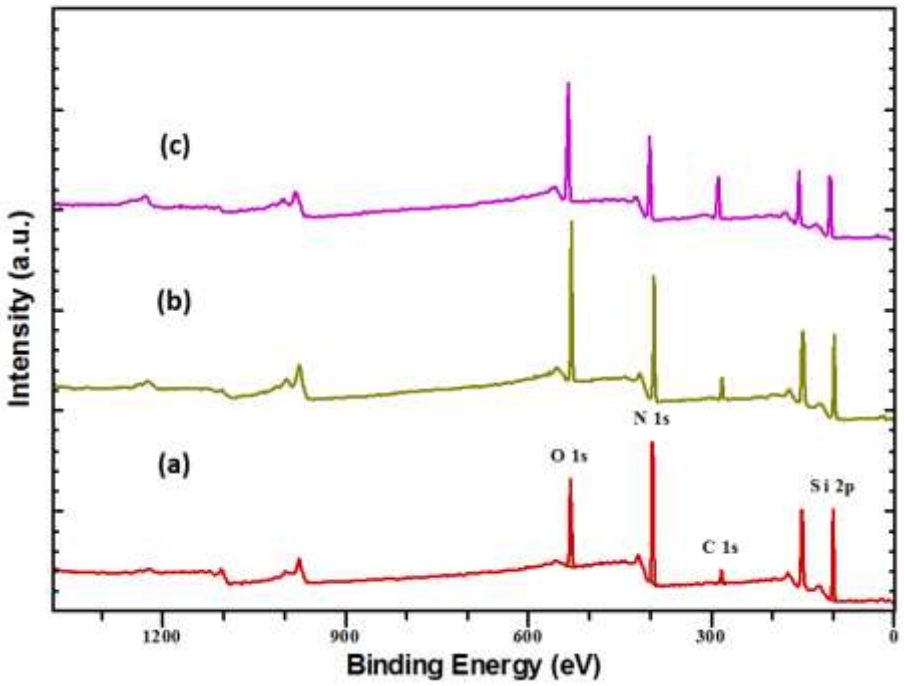

Figure S2. XPS survey spectra of: a) Si_3N_4, b) Si_3N_4 coated with silane-PEG-COOH and c) silanized surface with antibody covalently attached to the COOH groups.

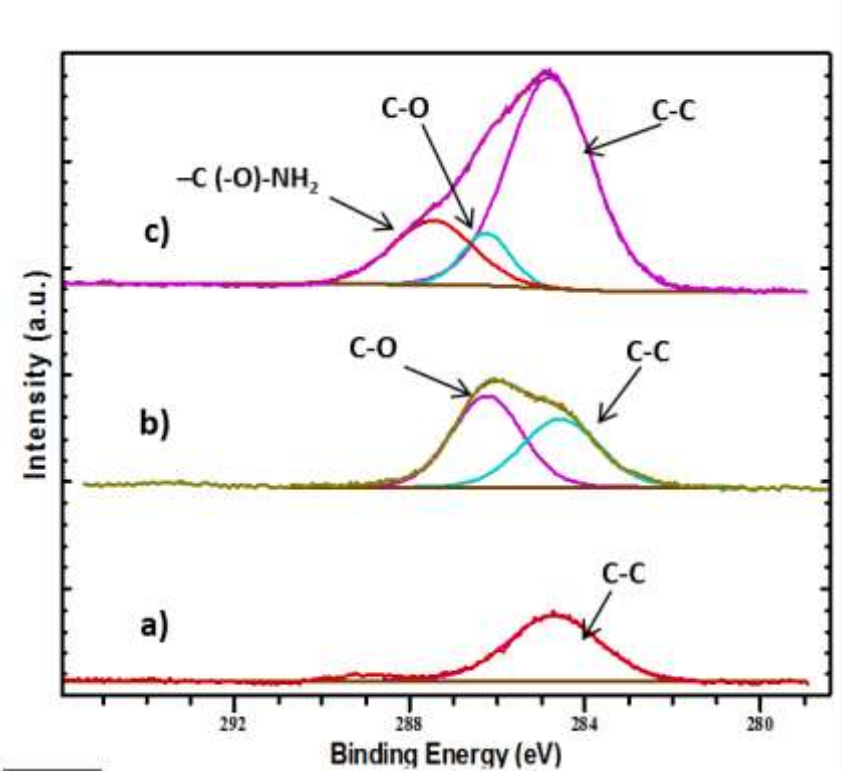

Figure S3. XPS high resolution spectra of C 1s, a) untreated Si_3N_4, b) silane-PEG-COOH and c) antibody immobilized.

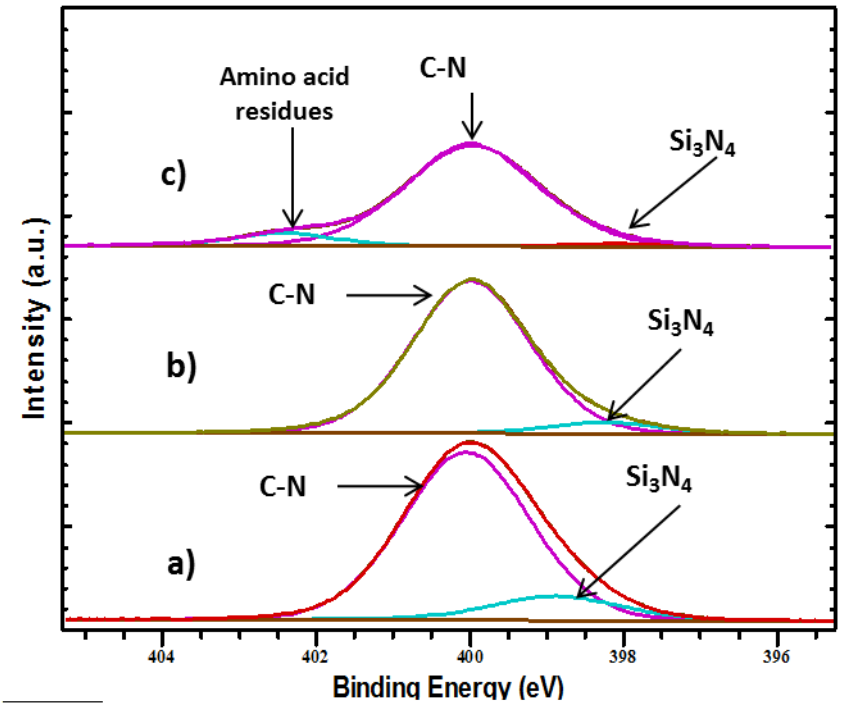

Figure S4. XPS high resolution spectra of N 1s, a) untreated Si_3N_4, b) silane-PEG-COOH and c) antibody immobilized.

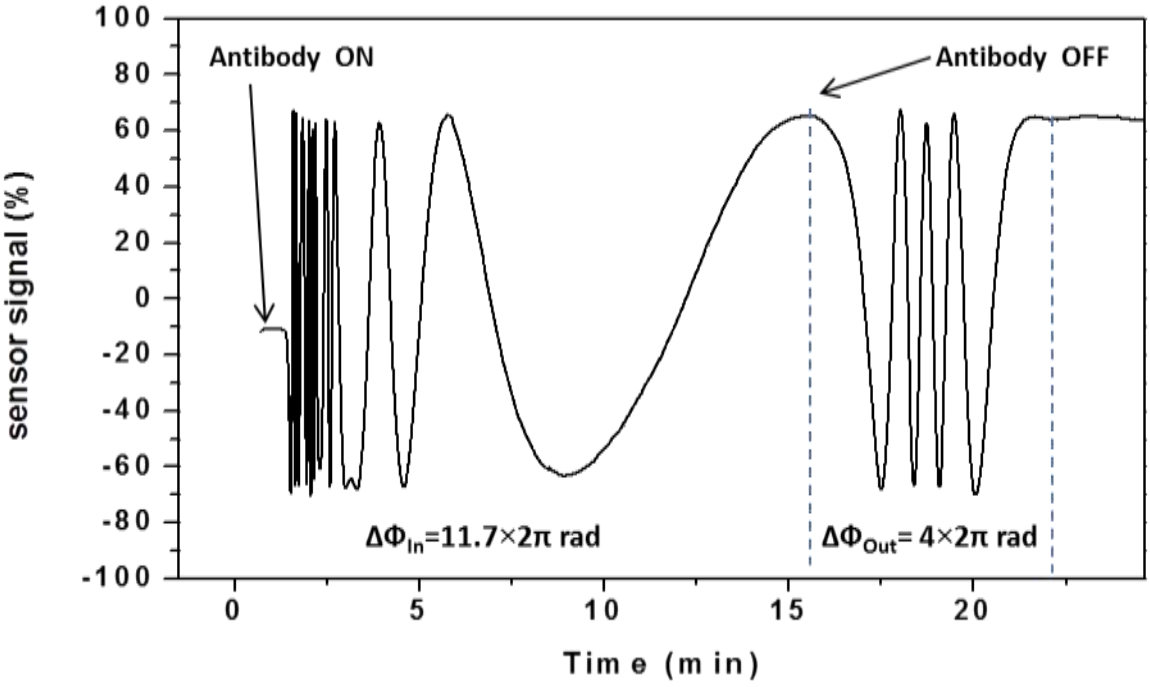

Figure S5. Real-time sensorgram showing the antibody immobilization. Phase variation due to the refractive index bulk change and the immobilization of antibody is 7.7×2π rad ($\Delta\Phi_{In}$-$\Delta\Phi_{Out}$).

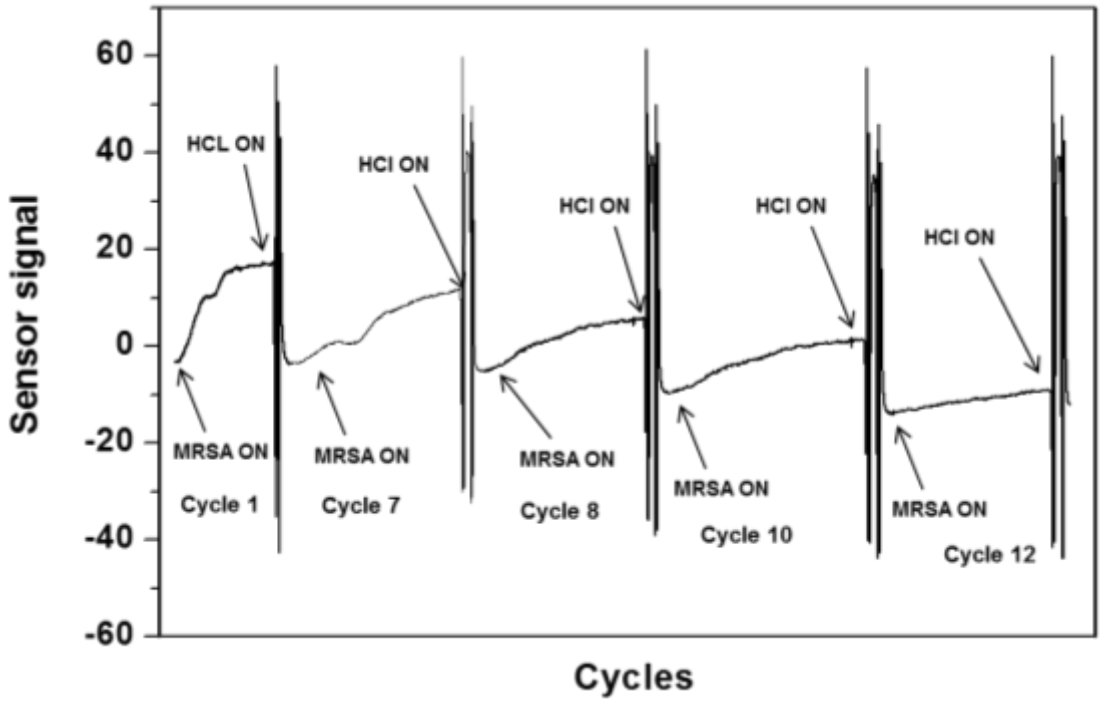

Figure S6. Real-time detection of 1, 7, 8, 10, and 12 measurement-regeneration cycles of MRSA detection regenerated with 25 mM HCl.

Table S1. Elemental composition of Si_3N_4 susbtrates before and after surface modification with silane-PEG-COOH and antibody

Sample	Elemental composition (at %)			
	C	N	O	Si
SI_3N_4	6.26	33.62	14.59	45.53
Silane-PEG	10.15	26.68	21.86	41.31
Antibody	23.32	22.37	20.7	33

References

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