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Short communication

Inkjet printed and "doctor blade" TiO_2 photodetectors for DNA biosensors

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ABSTRACT

A dye sensitized TiO_2 photodetector has been integrated with a DNA detection method based on non-cross-linking hybridization of DNA-functionalized gold nanoparticles, resulting in a disposable colorimetric biosensor. We present a new approach for the fabrication of dye sensitized TiO_2 photodetectors by an inkjet printing technique—a non-contact digital, additive, no mask and no vacuum patterning method, ideal for cost efficient mass production. The developed biosensor was compared against a dye sensitized photodetector fabricated by the traditional "doctor blade" method. Detection of gold nanoparticle aggregation was possible for concentrations as low as 1.0 nM for the "doctor blade" system, and 1.5 nM for the inkjet printed photodetector. The demonstrated sensitivity limits of developed biosensors are comparable to those of spectrophotometric techniques (1.0 nM). Our results show that a difference higher than 17% by traditional photodetector and 6% by inkjet printed in the photoresponses for the complementary and non-complementary gold nanoprobe assays could be attained for a specific DNA sequence from *Mycobacterium tuberculosis*, the etiologic agent of human tuberculosis. The decrease of costs associated with molecular diagnostic provided by a platform such as the one presented here may prove of paramount importance in developing countries.

1. Introduction

There is an increasing need to develop simple, rapid, and inexpensive detection platforms for diagnostic applications, monitoring food and water contamination by pathogens, environmental analysis, among others (Pejcic et al., 2006). Currently, the majority of available assays is performed under laboratory conditions, frequently with the assistance of expensive instruments and trained personnel. To circumvent these issues, we have combined three emerging technologies—dye sensitized TiO_2 photodetection (O'Regan and Grätzel, 1991), inkjet printing technology (Le, 1998) and colorimetric DNA detection method based on DNA-functionalized gold nanoparticles (Storhoff et al., 1998). This combination is of utmost relevance for the fabrication of a biosensing platform that leads to significant cost and time reduction in detection assays.

The increasing control over the unique size-dependent properties of nanoscale materials has paved the way for integration of such materials into many and/or improved technologies. One such technology has been a range of colorimetric assays for DNA detection based on the distance-dependant optical properties of oligonucleotide functionalized gold nanoparticles. These nanoparticle based systems have been extensively used for the labeling of DNA/protein such that the specific nanoparticle tag allows for the detection of target molecules based on the properties of the material (Baptista et al., 2008). The surface plasmon resonance (SPR) of gold nanoparticles is responsible for their intense colors. In solution, monodisperse gold nanoparticles (AuNPs) appear red and exhibit a relatively narrow SPR band centered around 520 nm in the UV/vis spectrum. In contrast, a solution containing aggregated AuNPs appears blue in color, corresponding to a characteristic red shift of the SPR. DNA or protein can be used as a linking molecule to aggregate the AuNPs and thus taking advantage of the optical properties of disperse versus aggregated gold particles for biodetection assays (Mirkin et al., 1996). Recently, a non-cross-linking hybridization method has been presented, where aggregation of the DNA-functionalized gold nanoparticles (Au-nanoprobes) is induced by an increasing ionic strength of the solution—the presence of a complementary target to the Au-nanoprobe prevents aggregation and the solution remains red; whereas the presence of non-complementary/mismatched targets does not prevent aggregation, resulting in a visible color change from red to blue (Baptista

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et al., 2006; Doria et al., 2007). We have previously integrated this system with an optoelectronic platform, using a high intensity light source and a color sensitive amorphous/nanocrystalline silicon photodetector, so as to create a high efficiency biosensor that can be used for the detection of specific DNA sequences (Fortunato et al., 2006; Martins et al., 2007; Silva et al., 2008).

Here, we report a new approach for the fabrication of dye sensitized nanocrystalline TiO_2 photodetectors (known also as Grätzel cell (O'Regan and Grätzel, 1991)) by an inkjet printing technique. This inkjet printed photodetector was optimized for use in conjunction with the Au-nanoprobe assay towards utilization as biosensor. This low cost and user-friendly biosensor may lead to significant cost reduction in more than one order of magnitude by minimizing the costs related to the high technology associated with fabrication of silicon-based photodetectors without losing the required sensitivity.

The photocurrent generation of these dye sensitized TiO_2 photodetectors depends on the incident photon energy and the highest spectral response occurs around the 530 nm region (the SPR peak of monodisperse Au-nanoprobes), thus enhancing the already high potential of this biosensing platform. Moreover, the sensitivity of this device can easily be tuned and adjusted by changing the dyes used in the sensitization (O'Regan and Grätzel, 1991). The main limitation of dye sensitized photodetectors over their silicon-based counterparts is the fact that the liquid electrolyte causes significant problems associated with the device sealing and stability that affect the commercialization of these devices. This problem can be overcome by employing solid state dye sensitized photodetectors that contain no volatile electrolytes (Senadeera et al., 2002). The other disadvantage of these photodetectors, when comparing with their Si counterparts, is the lower light to electricity conversion efficiency. Nevertheless, for application of photodetectors in a colorimetric biosensor, conversion efficiency does not play a very important role.

The main development is the introduction of adequate and low cost processing technique for the fabrication of dye sensitized photodetectors—the inkjet printing technology. The use of inkjet printing technique, not only simplifies a patterning process, but also reduces the consumption of materials and energy, allowing the scaling-up from lab prototype device to industrial production. It is a non-contact, no mask and no vacuum patterning method, ideal for cost efficient mass production (Calvert, 2001), being a very attractive and powerful technology for the development of biosensors (Hart et al., 1996) and DNA arrays (Schena et al., 1998). The decrease of costs associated with molecular diagnostic provided by a platform such as the one presented here may prove of paramount importance in developing countries. The use of inkjet printing in mass production of dye sensitized photodetectors reduces strongly their prices and opens up the possibility of easy fabrication of disposable photodetectors to be used in these biosensors, thus effective for broad application without compromising the device's reliability.

2. Experimental

2.1. Fabrication of photodetectors

In the fabrication of the photodetectors by inkjet printing technique, viscosity and surface tension controlled aqueous TiO_2 dispersion (ink) were prepared using appropriate amounts of TiO_2 (Solaronix SA, Aubonne, Switzerland) and Triton X-100 (JT Baker Chemical, Phillipsburg, USA). The dispersion was filtered through a 1.2 μm pore size filter (Chromofil PET-120/25; Macherey-Nagel GmbH & Co. KG, Düren, Germany) and ultrasonically mixed with the appropriate amount of ethanol. A modified commercially available Canon PIXMA IP4500 desktop printer (Canon Virginia, Inc., Newport News, USA) with a resolution of 9600 × 2400 dpi was used as the "device printer". With this printing unit we can achieve the deposition of the material as a digital printing process, where the ink/material is ejected directly onto a glass substrate from 1536 nozzles driven by an electronic signal. The fabrication of the active layer of TiO_2 with a thickness of ~4–5 μm by inkjet printing was performed at room temperature and atmosphere pressure on pre-cleaned, indium doped tin oxide conducting glass substrates (ITO, 10–12 $\Omega\,sq^{-1}$; Nippon Sheet Glass, Tokyo, Japan) and then sintered at 450 °C for 30 min in oven (Nabertherm, Lilienthal/Bremen, Germany). Improved electrical contacts from the ITO substrates were then printed with a commercial solvent-based silver nanoparticle formulation (SunTronic Jettable Silver-U5741; Sun Chemical Corporation, Parsippany, USA) using the same printer and afterwards sintered at 120 °C for 30 min. Substrates were then stained in an absolute ethanol solution of Ruthenium dye (N3, cisdi(thiocyanato)bis(2,2-bipyridyl-4,4-dicarboxylate) ruthenium(ll); Solaronix SA, Aubonne, Switzerland) for 24 h. Photodetectors were then fabricated by sandwiching these photoelectrodes with a Pt-coated fluorine doped conducting glass (FTO; Nippon Sheet Glass, Tokyo, Japan). The redox electrolyte containing lithium iodide (0.5 M; Sigma–Aldrich, Steinheim, Germany) and iodine (0.05 M; Sigma–Aldrich, Steinheim, Germany) in a 6:4 (v/v) mixture of acetonitrile (Sigma–Aldrich, Steinheim, Germany) and propylene carbonate (Sigma–Aldrich, Steinheim, Germany) was introduced between the dye coated TiO_2 electrode and the Pt-counter electrode. Photodetectors with a TiO_2 thickness ~7 μm were fabricated using the same TiO_2 paste by "doctor blade" method, as described by Senadeera et al. (2005).

Fig. 1 shows a schematic of the production of both photodetectors. The inset shows a scanning electron micrograph of the surface morphology of the TiO_2 film prepared by inkjet printing technology after sintering at 450 °C. No cracks on the surface are observed which indicates a very high inter-particle connectivity. The average diameter of TiO_2 particles is 25 nm.

2.2. Characterization of photodetectors

The effectiveness of the photosensor to convert light of various wavelengths into electrical current was measured as the incident photon to current conversion efficiency (*IPCE*). *IPCE* (%) is defined as the number of electrons generated by light per number of photons incident on the photodetector, formulated by the simplified equation:

$$IPCE = \frac{1240 J_{sc}}{\lambda W_i} \quad (1)$$

where J_{sc} is the short-circuit current density ($\mu A\,cm^{-2}$), λ the excitation wavelength (nm) and W_i is the photon flux ($W\,m^{-2}$). The *IPCE* was measured using a homemade setup coupled to a Keithley Multimeter 238 (Keithley Instruments, Inc., Cleveland, USA) via a computer.

Thickness of TiO_2 layers was determined by a profilometer Ambios Technology XP-200 (Ambios Technology, Inc., Santa Cruz, USA). Scanning electron micrograph of inkjet printed TiO_2 layer was done using Hitachi S4100 (Hitachi, Tokyo, Japan). Absorption spectrum of ethanolic solution of the Ruthenium dye was measured using a UV/vis/NIR scanning spectrophotometer UV-3101PC (Shimadzu, Kyoto, Japan).

2.3. Preparation of DNA-functionalized gold nanoparticles

All oligonucleotides were purchased from STAB Vida, Oeiras, Portugal. The Au-nanoprobes were synthesized by derivatizing an aqueous solution of AuNPs with a thiolated oligonu-

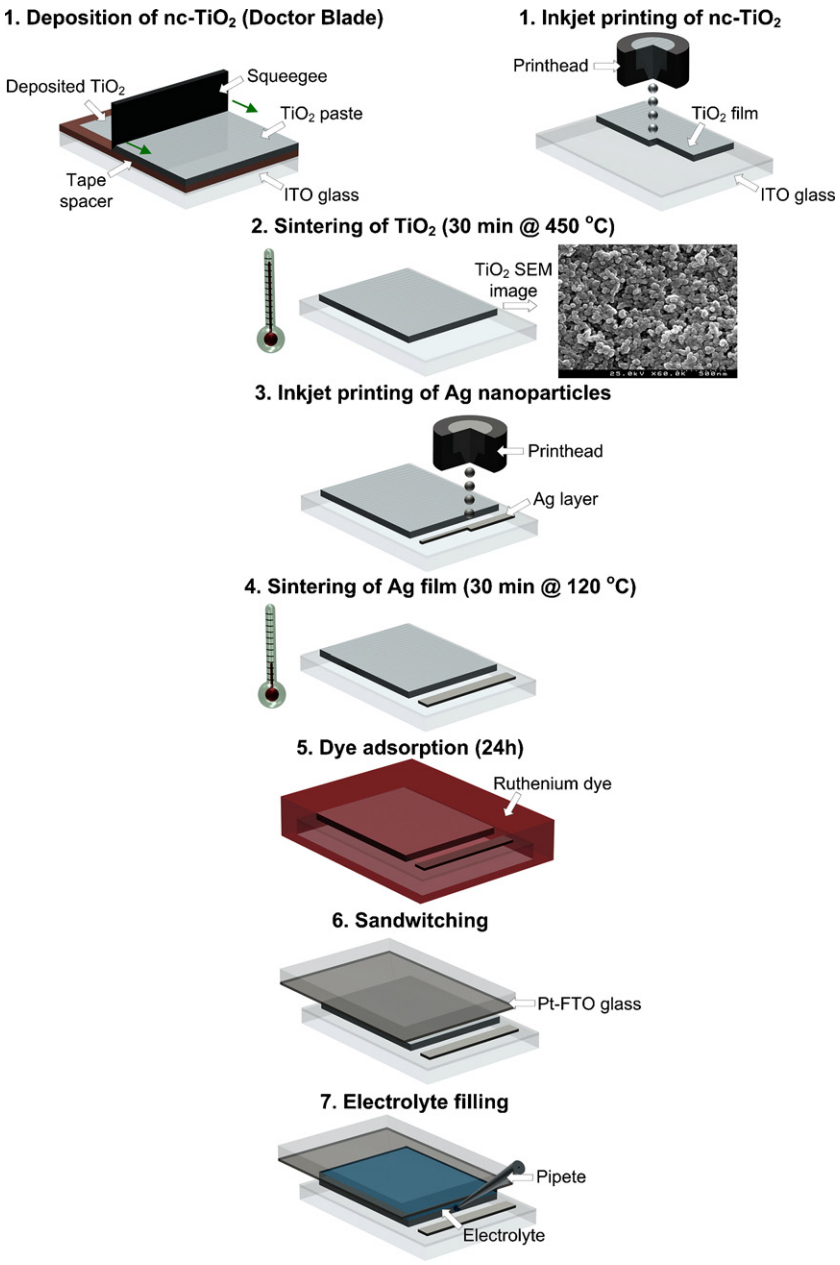

Fig. 1. Schematic of the TiO_2 photodetector fabrication by "doctor blade" and inkjet printing technique. The scanning electron micrograph presented the surface morphology of the TiO_2 film prepared by inkjet printing technology after sintering at 450 °C. The film is about 4 μm thick, no cracks on the surface are observed which indicates a very high inter-particle connectivity. The average diameter of TiO_2 particles is 25 nm.

cleotide (5′-thiol-GGACGTGGAGGCGATC-3′) recognizing a specific sequence within the rpo-beta gene of *Mycobacterium tuberculosis* (GenBank accession no. BX842574) and used as described by Baptista et al. (2006), with either a complementary DNA oligonucleotide harboring part of the rpo-beta gene of *M. tuberculosis* (5′-GATCGCCTCCACGTCCGTTGGTATCAAGGTT-3′) or a non-complementary DNA oligonucleotide (5′-GGTCGTCAGACTGTCG-ATGAAGCC-3′), both at a final concentration of 1.33 μM.

2.4. Detection procedure

In the prototype biosensor, plastic cuvettes (10 mm path length; Plastibrand, Wertheim, Germany), filled with 75 μl of AuNPs or Au-nanoprobes solution, were placed between the light source (a high power LED, 530 nm light, 35 nm spectral half-width, Luxeon® K2 Emitter; Quadica Developments Inc., Brantford, Canada) and the photodetector. The entire platform is placed within a black environment, in order to eliminate ambient light interference. Photocurrents generated by photodetectors were measured by a digital electrometer (Keithley 238; Keithley Instruments, Inc., Cleveland, USA) connected to a computer via USB/GPIB interface card (Agilent® 82357A; Agilent Technologies, Santa Clara, USA) with developed software (MatLab®).

Measurements were performed after 15 min of salt addition to AuNPs or Au-nanoprobes solution (0.02 M NaCl or $MgCl_2$, respectively). The measurements were repeated 6 times for each sample with a time interval of 150 s. Ultra pure water (Millipore, Billerica, USA) and 10 mM phosphate buffer (pH 8), 0.1 M NaCl were used as reference solutions for AuNPs and Au-nanoprobes, respectively. For validation of the prototype, UV-3101PC and UVmini-1240 UV/vis/NIR scanning spectrophotometers (Shimadzu, Kyoto, Japan) were used for AuNPs and Au-nanoprobes measurements, respec-

tively. DNA detection reactions were carried out with the appropriate target and Au-nanoprobes at a final concentration of 2.5 nM, in 10 mM phosphate buffer (pH 8). After 10 min of denaturation at 95 °C, the mixtures were allowed to slowly cool down to 20 °C until salt addition.

The reported expanded uncertainties are based on a standard uncertainty multiplied by a coverage factor $k = 2.65$, providing a coverage probability of approximately 95%. The uncertainty evaluation has been carried out in accordance with International Organization for Standardization requirements.

3. Results and discussion

We developed a disposable biosensor integrating a dye sensitized TiO_2 photodetector obtained by standard "doctor blade" method (from now on referred as "doctor blade" photodetector, DBP; Smestad, 1998) and by inkjet printing technology (described as inkjet printed photodetector, IPP) to be used in conjunction with the Au-nanoprobe based method. In the case of fabrication of the IPP, through an improved formulation of specific water-based inks (Section 2), we attained a TiO_2 photodetector by thermal inkjet printing technology. The inks must meet strict physicochemical properties (viscosity, surface tension, etc.) to achieve optimal performance of printed device. The optimum composition of TiO_2 water-based ink was 20 wt% of ethanol and 0.12 wt% of TiO_2. Such composition resulted in a viscosity that was acceptable for conventional office printers (1–10 cP). Additionally, Triton X-100 (5%) was added to the ink, which resulted in more effective jetting while maintaining a well defined and uniform film layer. The dispersion of TiO_2 does not undergo a sedimentation process (particles with diameter smaller than 1.2 μm). Long term viscosity measurements showed that this dispersion is also free of aggregation and demonstrated a greater chemical stability of crystal over solution in comparison with sol–gel ink. Highly conductive contact tracks from inkjet printed silver nanoparticle further helped in the stability and performance of this photodetector. This inkjet printing technique may circumvent the existing time-consuming and expensive microfabrication of conductive tracks by lithography or electrolysis.

Fig. 2 depicts the incident photon to current conversion efficiencies (*IPCE*) of the traditional DBP and novel IPP. The inset of the figure shows the absorption spectrum of the ethanolic solution of Ruthenium (N3) dye. While the absorption spectrum of the N3 in ethanol showed an intense absorption at ~539 nm, the *IPCE* peaks of DBP and IPP were at 537 and 525 nm, respectively. As it is generally observed with the dye sensitized photodevices, the close relationship and shifts of the *IPCE* peaks with the optical absorption spectra indicate that the photoelectrochemical conversion is achieved through photosensitization, namely that N3 molecules absorbed photons and generated excited electrons, and the excited electrons were subsequently transferred to the conduction band of the TiO_2 electrode. These shifts mainly depend on the strength of the chelation of the dye molecules to the semiconducting surface and the surface morphology of the semiconductor. For both photodetectors, the maximum *IPCE* was observed around 530 nm, which is in the vicinity of the SPR peak of the monodisperse Au-nanoprobes, confirming the compatibility of the detector and Au-nanoprobes integration in a biosensor. The IPP showed lower *IPCE* than the DBP, probably due to the higher film thickness of the active layer of the DBP, which absorbs more dye molecules, hence creates higher photocurrent density and the *IPCE* (Junghanel and Tributsch, 2005; Kang et al., 2004).

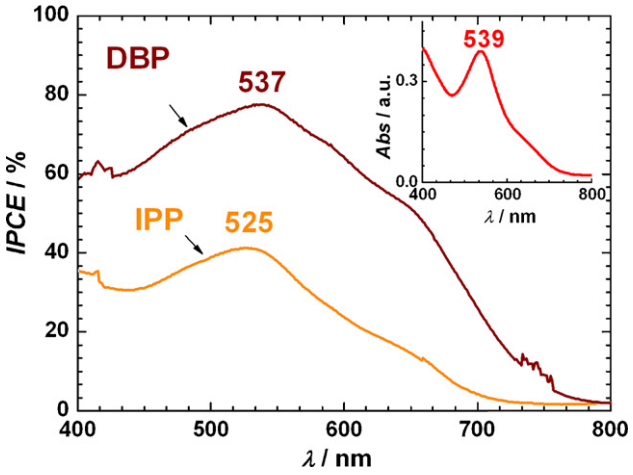

Fig. 2. Incident photon to current conversion efficiency (*IPCE*) of the photodetector obtained by "doctor blade" method (DBP) and by inkjet printing (IPP). The TiO_2 layer of DBP is about 1.75 times thicker than in the case of the IPP, which may explain the observed difference at the *IPCE*. *Inset*: Absorption spectrum of the ethanolic solution of Ruthenium N3 dye.

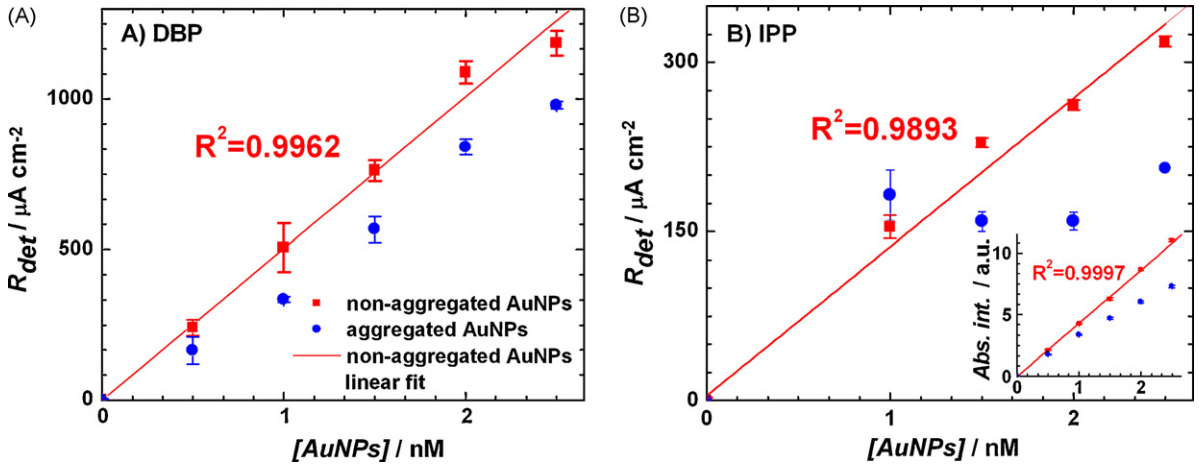

Fig. 3. Responses of the sensor with (A) "doctor blade" photodetector and (B) inkjet printed photodetector for different AuNP concentrations. Non-aggregated (red squares) and aggregated by salt addition (blue circles). The detection response (R_{det})—difference in the photocurrent densities generated by photodetector when illuminating through the reference solution and through the sample solution. Measurements using a high power LED with 530 and 35 nm spectral half-width. *Inset*: Corresponding spectrophotometric measurements: absorption peak area integral from 520 to 540 nm as a function of AuNPs concentration. Standard deviation bars were determined from 6 independent measurements. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

The fabricated photodetectors were individually mounted on a platform inside a black box to cut-off incident background light. Plastic cuvettes filled with each solution were then placed between the light source (green LED) and the photodetector. The photocurrent generated by the photodetector due to the non-absorbed light coming out from the cuvette was measured by an electrometer and relayed to the computer.

Several non-aggregated AuNPs solutions (red) of known concentrations were used to assess the response and the sensitivity of the sensors, and compared with data attained by conventional UV/vis spectrophotometry. Fig. 3A and B shows the results obtained using the novel platforms with either DBP or IPP, respectively. The inset shows the corresponding spectrophotometry results. The detection responses (R_{det}) were calculated by measuring the differences in the photocurrent densities obtained for the reference (ultra pure water and phosphate buffer for AuNPs and Au-nanoprobes, respectively) and the sample solution. Again, the sensor with the IPP showed lower photoresponses probably due to the lower thickness of the active layer compared to the device with DBP. To compare the responses of the developed sensors with those of the spectrophotometer, the integral of the absorption peak over the wavelength range between 520 and 540 nm (spanning the emission spectra of the LED) was used. The output signals of both the sensors and the spectrophotometer show a linear variation for the non-aggregated particles (red color solution) with high accuracy ($R^2 > 0.98$). In the case of the aggregated particles (blue color solution), a non-linear variation is observed for both methodologies. This can be explained by the several different events occurring (e.g. aggregation, flocculation, precipitation and possible adsorption to the plastic surface of the cuvette), which lead to light scattering and decrease the amount of light being transmitted through the solution. Nevertheless, in terms of application as biosensor what is relevant is the capability of discrimination between the aggregated and non-aggregated forms of the AuNPs, and not the linearity of these responses. Very clear differences in the responses of the sensors were observed for the AuNPs solution concentrations routinely used in the Au-nanoprobe based DNA detection method (from 1.5 to 2.5 nM; Baptista et al., 2006; Doria et al., 2007). For low concentrations, below 1.0 nM for DBP and 1.5 nM for IPP, the generated photocurrent densities were nearly the same for both solutions indicating the sensitivity limit of the sensor. The detection limit of the spectrophotometer is superior to 0.5 nM. The measurements were consistent and reproducible, and comparable to those attained for spectrophotometry, showing that the developed sensors may be a low cost and effective alternative for use with the AuNPs based assay. We observe larger fluctuations of the acquired data in the comparison with spectrophotometry, because we are using a LED, which does not have a single wavelength emission but an emission profile of 30 nm half-width peak. This together with the considerable noise introduced by the system, e.g. photodetector, cables, power supply and electrometer is responsible for the larger uncertainty bars. All these factors causing fluctuations during data acquisition need to be dealt upon at a later stage of prototype optimization.

Finally, as a proof-of-concept, the sensors were applied in conjunction with the Au-nanoprobe assay to detect a specific DNA sequence (Fig. 4) from *M. tuberculosis* (Baptista et al., 2006), the etiologic agent of human tuberculosis. Mixtures containing the Au-nanoprobe and complementary oligonucleotide target (positive, POS—red) or non-complementary target (negative, NEG—blue), were screened by both the developed sensors and the spectrophotometer. The Au-nanoprobe aggregation (non-complementary assay) induced by salt addition resulted in the shift of the SPR maximum of 26 nm compared to the complementary assay (Fig. 4, inset). A clear difference in the detection response of the sensors between complementary (POS) and non-complementary (NEG) assays is observable in the μA range. The percentage difference between detection response for complementary and non-complementary assays was 17.5% for the DBP and 6.7% for the IPP (Fig. 4B). Even though the inkjet printed photodetectors gave lower difference of detection responses than photodetectors fabricated by the traditional method, it should be noted that the inkjet printing is employed for the first time in the dye sensitized TiO_2 technology, and the properties of IPPs still need to be fully optimized. Nevertheless, the IPP based platform was able to clearly detect the presence or the absence of a specific DNA target.

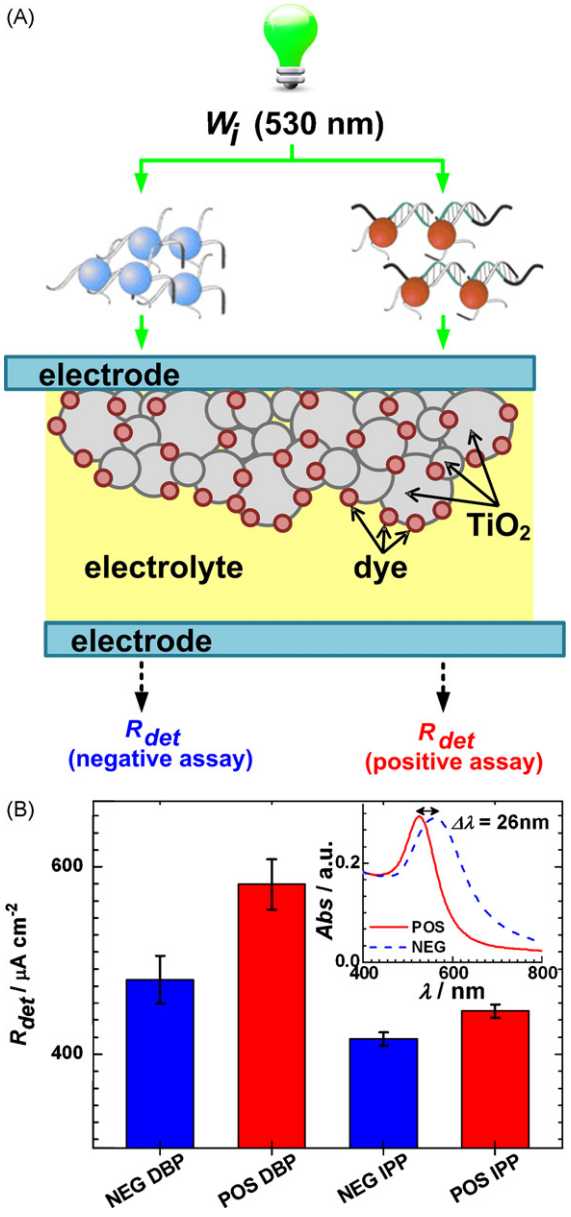

Fig. 4. (A) Schematic representation of DNA detection of the developed biosensor (W_i—the photon flux and R_{det}—detection responses). (B) The Au-nanoprobe based assays for *M. tuberculosis* DNA with the novel sensors with DBP "and IPP: POS—complementary DNA target; NEG—non-complementary DNA target; R_{det}—difference in the photocurrent densities generated by photodetector when illuminating through the reference solution and through the sample solution. *Inset*: Absorption spectra of DNA solutions (in arbitrary units). The absorption spectrum of NEG was shifted for the same absorbance value at λ = 400 nm as POS (shift = −0.0067 a.u.) so as to allow better color change visualization. DNA detection was carried out with the appropriate oligonucleotide target at a final concentration of 1.33 μM and Au-nanoprobes at a final concentration of 2.5 nM. Standard deviation bars were determined from 6 independent measurements.

4. Conclusions

To our knowledge the use of dye sensitized TiO_2 sensors for DNA detection by means of Au-nanoprobes has been demonstrated for the first time. Moreover, we describe here pioneer results of inkjet printing technology application to the fabrication of the active layers of photodetectors. The developed biosensor including dye sensitized photodetector fabricated by traditional "doctor blade" method detected the aggregation of AuNPs concentrations as low as 1.0 nM, while for the inkjet printed photodetector the limit was set at 1.5 nM. The sensitivity limits of the developed biosensors are comparable to the sensitivity limit of the spectrophotometry based techniques (1.0 nM). Both photodetectors gave clear and distinct differences in the detection responses for the complementary and non-complementary DNA assays. Our results show that a difference of 17.5% by traditional photodetector and 6.7% by inkjet printed in photoresponses for the complementary and non-complementary Au-nanoprobe assays could be attained. The DBP gives nearly as good results as spectrophotometry, while usability and reliability of the established IPP used in DNA sequence identification still require further experimental development, mainly concerning the determination of the most adequate thickness and role of TiO_2 nanoparticles size on the overall device performance. Moreover, the photodetectors' sensitivity can be further improved by enhancing the spectral responses of the detector through the use of different dyes. Once the photodetectors have been optimized, we will apply the same methodology to other relevant DNA target sequences and with increased complexity. Further research is being also carried out to evaluate the possible association within a fully integrated, portable biosensor by means of microfluidic technology. The combination of three emerging technologies: (i) dye sensitized TiO_2 photodetection, (ii) inkjet printing technology and (iii) colorimetric DNA detection based on DNA-functionalized gold nanoparticles may lead to significant cost and time reduction in DNA detection.

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References

Baptista, P., Koziol-Montewka, M., Paluch-Oles, J., Doria, G., Franco, R., 2006. Clin. Chem. 52, 1433–1434.
Baptista, P., Pereira, E., Eaton, P., Doria, G., Miranda, A., Gomes, I., Quaresma, P., Franco, R., 2008. Anal. Bioanal. Chem. 391, 943–950.
Calvert, P., 2001. Chem. Mater. 13, 3299–3305.
Doria, G., Franco, R., Baptista, P., 2007. IET Nanobiotechnol. 1, 53–57.
Fortunato, E., Martins, R., Baptista, P., FCT-UNL, 2006. Patent: 103561-PT.
Hart, A.L., Turner, A.P.F., Hopcroft, D., 1996. Biosens. Bioelectron. 11, 263–270.
Junghanel, M., Tributsch, H., 2005. J. Phys. Chem. B 109, 22876–22883.
Kang, M.G., Ryu, K.S., Chang, S.H., Park, N.G., Hong, J.P., Kim, K.J., 2004. Bull. Korean Chem. Soc. 25, 742–744.
Le, H.P., 1998. J. Imaging Sci. Technol. 42, 49–62.
Martins, R., Baptista, P., Raniero, L., Doria, G., Silva, L., Franco, R., Fortunato, E., 2007. Appl. Phys. Lett., 90, 023903(1–3).
Mirkin, C.A., Letsinger, R.L., Mucic, R.C., Storhoff, J.J., 1996. Nature 382, 607–609.
O'Regan, B., Grätzel, M., 1991. Nature 353, 737–740.
Pejcic, B., De Marco, R., Parkinson, G., 2006. Analyst 131, 1079–1090.
Schena, M., Heller, R.A., Theriault, T.P., Konrad, K., Lachenmeier, E., Davis, R.W., 1998. Trend Biotechnol. 16, 301–306.
Senadeera, G.K.R., Jayaweera, P.V.V., Perera, V.P.S., Tennakone, K., 2002. Sol. Energy Mater. Sol. Cells 73, 103–108.
Senadeera, R., Fukuri, N., Saito, Y., Kitamura, T., Wada, Y., Yanagida, S., 2005. Chem. Commun. 17, 2259–2261.
Silva, L.B., Baptista, P., Raniero, L., Doria, G., Martins, R., Fortunato, E., 2008. Sens. Actuator B: Chem. 132, 508–511.
Smestad, G.P., 1998. Sol. Energy Mater. Sol. Cells 55, 157–178.
Storhoff, J.J., Elghanian, R., Mucic, R.C., Mirkin, C.A., Letsinger, R.L., 1998. J. Am. Chem. Soc. 120, 1959–1964.