Supplementary Information

Photosensor characterization

The calibration and optoelectronic characterization of the a-Si:H PIN photosensor chip[1] response to the incident flux of photons is shown in Fig. S1a for two wavelengths (405 and 605 nm). These wavelengths were chosen because they correspond respectively to the excitation and emission wavelengths of the selected QDs to fluorescently label the target HIV-1 Vif protein. As shown in Fig. S1a, the photodiode chip responds linearly to the flux of photons at 605 nm (the emission wavelength of the QDs), in the range of 10^8 to 10^{15} cm^{-2} s^{-1}. On the other hand, the current density from the photodiode for a photon flux of ~ 10^{14} cm^{-2} s^{-1} at 405 nm was five orders of magnitude lower than at 605 nm (Fig. S1a), as expected considering that the photodiode chip was designed in order to have very low sensitivity at this wavelength. The measured current density-voltage (*J-V*) characteristics of the photodiode under light irradiation at 405 nm is comparable to the dark current (photoresponse in the dark) and significantly lower that the photoresponse obtained when irradiating light at 605 nm (Fig. S1b). A maximum ratio of ~ 10^5 is observed between the photocurrent measured under 605 nm and 405 nm light irradiation, obtained at a voltage bias of 0 V. Collectively, the data presented in Fig. S1 clearly reveals the efficiency of the a-SiC:H fluorescence filter to suppress the 405 nm light which will be used for the fluorescent excitation of the QDs. The current density obtained from the photodiode when irradiated at 405 nm at 0V bias was considered the device noise signal.

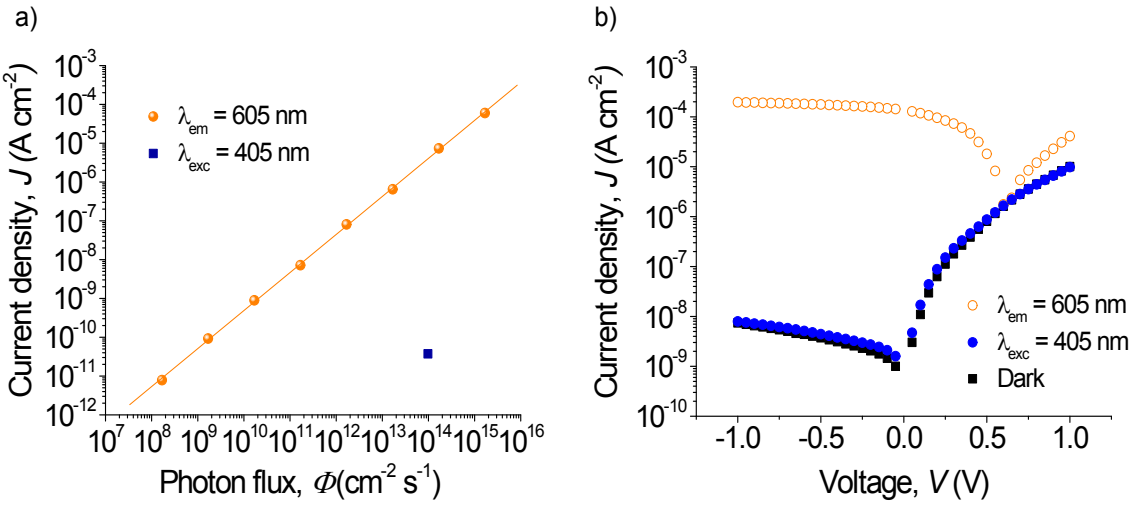

Fig. S1 a) Photodiode response to 605 emission light as a function of the incident photon flux. The line represents the linear fit to the experimental results. Device response is also shown at

the excitation wavelength. b) Experimental *J-V* characteristics of the a-Si:H PIN photodiode measured in the dark and under illumination.

Experimental Section

Fabrication and Characterization of the a-Si:H photodiode: 200 μm × 200 μm hydrogenated amorphous silicon (a-Si:H) PIN photodiodes were microfabricated on a glass substrate (Schott AF45) as described.[2,3] The current density-voltage characteristics of the a-Si:H photodiode were measured using a picoammeter (Keithley 237) in the dark and under illumination. The current density as function of the photon flux was measured using a lock-in amplifier (EG&G Princeton Applied Research, model 5209) and a light beam chopper (HMS). The photon flux of light from a tungsten-halogen lamp, passing through a monochromator (McPherson, model 2053) was determined using a calibrated crystalline silicon photodetector (Advanced Photonix, Inc., SD100-11-11-021). The photosensor response was characterized at the emission wavelength of the quantum dot (QD), λ_{em} = 605 nm, and at the laser excitation wavelength, λ_{exc} = 405 nm.

Assembly of the microfluidic chip and preparation of the biosensor: A microchannel with dimensions of 300 μm in width, 20 μm in height and 2 mm in length was fabricated in PDMS by soft lithography. A master mold of the microchannel was fabricated using SU-8 2015 (Microchem) photoresist deposited on a silicon substrate that was patterned with UV light through an aluminum mask on quartz. The PDMS pre-polymer (Sylgard 184, Dow Corning) was prepared by mixing the base solution and the curing agent in a 10:1 weight ratio followed by vacuum degassing. The PDMS mixture was poured into the mold and cured for 1h at 80 ºC. The solidified PDMS was peeled off from molds and bonded to microscopy cover-slip glass chips (No. 1.5; VWR International) after an UV-ozone treatment for 40 minutes. The sealed PDSM-glass chips were further cured at 100ºC for 30 min and silanized by flowing 2.5% MPTS in anhydrous acetone. The MPTS-activated microchannels were further cured at 110 °C for 2 h, incubated with 10 mM dithiothreitol in PBS for 30 min at 37 ºC, and activated with a 33 mM solution of GSH in PBS buffer pH 7.0 for 30 min at room temperature. The scFv 4BL was immobilized by flowing a GST-scFv 4BL solution (100 μg mL^{-1}) at 0.5 μL min^{-1} for 15 min, and the channels were flushed with PBS, 0.1% Tween 20 at 5 μL min^{-1} for 5 min.

Preparation of recombinant proteins and cell extracts: The single chain variable fragment (scFv) 4BL antibody was cloned into the *Escherichia coli* expression vector pGEX-4T. The expression of the encoded glutathione S-transferase (GST)-scFv 4BL fusion protein was induced by 1mM IPTG added to a 2-L *E. coli* BL21 (DE3) culture in a bioreactor. After expression for 48 h the cells were lysed by sonication and the released GST-scFv 4BL protein was purified on a single step affinity chromatography using a glutathione sepharose 4 Fast Flow XK16 column (GE healthcare), dialyzed against PBS buffer and formulated with 10% glycerol.

The HIV-1 virion infectivity factor (Vif) was produced and purified according to previously published procedures,[4,5] and labeled with the QDs previously activated by standard EDC/sulfo-NHS chemistry.[6]

Whole cell extracts were prepared from HEK 293T cells expressing HIV-1 Vif cells by lysis with 50 mM Tris, 150 mM NaCl, pH 7.5, supplemented with anti-proteases (30 min incubation at 4ºC) and clarified by centrifugation.[4]

Detection assay: The solution containing the target (QD-Vif) was flowed in the microchannels at $Q = 0.5$ μL min^{-1} for 15 min. The microchannels were further washed with a PBS flow at 5 μL min^{-1} for 5 min, mounted and aligned with the photodetector under a stereomicroscope and placed inside a dark box. A 405 nm diode laser (Laser Components) with a spot size of approximately 4 mm was used as excitation light source and the sensor photocurrent was measured at zero bias over a 100 s time interval using a picoammeter (Keithley 6487).

References

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