Redox-Reactive Field-Effect Transistor Nanodevices for the Direct Monitoring of Small Metabolites in Biofluids toward Implantable Nanosensors Arrays

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ABSTRACT: Chemically modified field-effect transistor (FET) nanodevices were shown to be a selective and extremely sensitive detection platform. In FET-based sensors, signal amplification and transduction is based on electrostatic gating of the nanometric semiconductor channel by analyte–receptor interactions, which measurably affect the transconductance of the device. However, chemically modified FETs must overcome several fundamental limitations before they can be effectively deployed as real-time sensors for bioevents occurring on their surface in complex biofluids. Here, we demonstrate the development of amperoFET devices for the real-time continuous monitoring of small molecular metabolites in biofluids. The surface of the nanowires is covalently modified with a redox reversible moiety, which is easily oxidized in the presence of H$_2$O$_2$. The reversible redox transformation of the surface-confined molecules is carried out by a hot electron injection mechanism, conducted simply by the modulation of the source–drain current through the nanoFET sensing device. By this approach, electrons may be injected by the nanowire element into the surface-confined redox moiety and thus maintain a whole-electrically actuated redox system in which the oxidation state is completely controlled by the current applied to the amperoFET system. The modulation of the source–drain current allows the control of the reduced versus oxidized redox moieties population on the nanowire surface, and this, in turn, is applied as the main sensing mechanism. At a given constant source–drain and gate voltage, the chemical perturbation exerted by the presence of chemical oxidants in the tested biofluid will lead to a measurable conductance change. Alteration in the concentration of the specific metabolite will chemically regulate the extent of perturbation applied to the redox system, which can be utilized for the quantification of the molecular metabolite of interest. These 'equilibrium'-type sensors are fully electrically operated and can be further used in implantable sensing applications.

KEYWORDS: silicon nanowires, field-effect transistor, redox, biomolecules, metabolites, nanosensors

Chemically modified field-effect transistor (FET) nanodevices were shown to be a selective and extremely sensitive detection platform. In FET-based sensors, signal amplification and transduction is based on electrostatic gating of the nanometric semiconductor channel by analyte–receptor interactions, which measurably affect the transconductance of the device. However, chemically modified FETs must overcome three fundamental limitations before they can be effectively deployed in real-world biosensing applications. First, the electrical double layer in biofluids of high ionic concentration dramatically shields charge carriers, thus limiting the high ionic concentration dramatically shields charge carriers, thus limiting the gating effect as a consequence of biorecognition events. The extent of shielding is characterized by the Debye length, <1 nm in physiological fluids. Second, small uncharged molecular targets exert a minimal gating effect on semiconductor transconductance unless they affect the surface potential through triggering changes in receptors within the Debye length. Third, most detection schemes based on
nanoFETs require washing steps following the biorecognition events, in order for the sensor to be able to perform additional cycles of detection. This in turn strongly limits the use of nanoFET devices in implantable in vivo applications, where real-time equilibrium-based sensors are highly desirable. Thus, a strong need exists for the development of nanoFET devices capable of directly monitoring bioevents occurring on the surface of the sensor in real-time in complex biofluid environments.

Our group recently demonstrated the development of redox-reactive nanowire biosensors for multiplex monitoring of metabolic activity in physiological environments. The surface of the nanowires is covalently modified with a redox reversible system, 9,10-dihydroxyanthracene/9,10-anthraquinone (DHA/AQ). The reversible redox transformation of the surface-confined DHA molecules can occur by alternatively applying chemical reductants and oxidants, such as NaN-dithiylhydroxylamine (DEHA), and hydrogen peroxide, respectively. DHA tends to selectively react with oxygen reactive species (ROS) or with H2O2. Therefore, by introducing a metabolite, glucose, for example, to a specific oxidase enzyme, such as glucose oxidase, the coproduction of H2O2 will occur. ROS or H2O2 will selectively oxidize the DHA on the surface of the nanowire FET, and as a result, the charge density on the nanowire’s surface will deeply change and alter the conductivity of the device. Initially, measurements were obtained using the chemical reductant DEHA, for performing the chemical reduction of the redox-reactive moiety. This type of analysis was shown very useful for non-invasive ex vivo monitoring of excreted extracellular metabolites.

However, in order to realize implantable and real-time continuous monitoring sensing devices, it is impractical to use the described chemical reduction of the AQ moiety, thus it will be highly desirable and important to be able to electrically reduce/oxidize the surface-confined redox moieties by the nanowire element directly.

In 2002, Professor Lieber’s group presented the first molecular nonvolatile memory constructed on a back-gated Si nanowire FET covered by redox-reactive molecules. Here, the switching between on and off states has been realized via controlling the Si nanowire FET channel conductance through back-gate voltage. This reversible on and off states switching process relied on hot electron injection from the channel into the redox-reactive molecules, which served as a floating gate, through a tunneling native oxide layer of the Si nanowire. Therefore, in an aqueous environment, where H+ is available, and particularly in biofluids, modulating the applied gate electrode voltage, Vg, allows maintaining a constant population of DHA/AQ, or a constant reduced/oxidized ratio, on the nanowire surface, which results in an equilibrium state between the gate voltage and the ‘reduced-to-oxidized’ moieties population on the nanowire surface. By this mechanism, electrons may be injected by the nanowire element into the surface-confined redox moiety and thus maintain a whole-electrically actuated redox system in which the state of oxidation is completely controlled by the gate voltage applied to the amperoFET system.

Here, we demonstrate the development of amperoFET devices for the real-time continuous monitoring of small molecular metabolites in biofluids. The modulation of the gate voltage allows the control of the reduced versus oxidized redox moieties population on the nanowire surface, and this, in turn, is applied as the main sensing mechanism. At a given constant gate voltage, the chemical perturbation exerted by the presence of chemical oxidants in the tested biofluid will lead to a measurable conductance change. Change in the concentration of the specific metabolite will chemically regulate the extent of perturbation applied to the redox system and eventually used for the quantification of the molecular metabolite of interest. These ‘equilibrium’-type sensors are fully electrically operated and can be further used in implantable sensing applications.

RESULTS

Electrical Reduction of Redox Species on Surface-Modified Silicon Nanowire FETs. The effect of different gate voltages applied to the biosensing system, while maintaining a constant source–drain voltage (Vsd), was investigated. SiNW devices were converted to their fully oxidized state, specifically, the AQ population was increased to its maximum on the surface. This was achieved by introducing a 1 mM H2O2 solution in phosphate buffer saline (PBS) to the SiNW device that receives electrons from DHA and consequently oxidizes it. Then, the source–drain voltage was set to a constant (Vsd = 0.3), and a series of negative voltages were applied at the gate electrode. Consequently, after the gate voltage returned back to zero, a decrease in the current signal from the SiNW devices was received, namely a change of potential on the surface of the SiNWs FET occurred, resulting from the reduction of the redox-reactive moieties. For comparison, we performed a ‘chemical’ reduction experiment, in which the SiNW devices were first converted to their fully oxidized state, using 1 mM H2O2 solution in PBS, in a similar manner. In this case, the gate voltage (Vg) was turned off (Vg = 0 V) for the entire experiment, while the source–drain voltage was set to a constant for the time frame of obtaining the signals and turned off for the reductant injection (Table 1).

Table 1. Summary of the Source–Drain and Gate Voltages as a Function of the Redox-Reactive Nanosensors Operation Modes

| Operation mode | Vsd (V) | Vg (V) |
|----------------|--------|--------|
| Chemical reduction | 0.3    | 0.0    |
| AmperoFET – detection limit | 0.3    | −0.9   |
| AmperoFET – continuous sensing in biofluids | 0.3    | −0.5   |

nanowire device was fully reduced by 1 vol % DEHA (the reductant) in PBS, as we have shown in previous work by XPS analysis and the device response to H2O2. In Figure 1, it is clearly shown that a SiNW surface reduction achieved by applying a voltage on the gate is as effective as the chemical reduction.

Next, control experiments were performed in order to confirm that the reductive abilities of the applied gate voltages are attributed to the specific reduction of the AQ moieties. For this purpose, SiNW devices were modified with the nonredox reactive molecule (3-aminopropyl)-dimethyl-ethoxysilane (APDMES). In both systems, the SiNW devices were initially oxidized using a 1 mM H2O2 solution. Then, the source–drain voltage was set to Vsd = 0.3 V, while the gate voltage was turned off during signal collection. In our previous work with redox-reactive nanowires sensors, this setup of voltages has shown a great sensitivity (down to 100 nM to oxidative spics) in high ionic flux samples. Afterward, each system, three different gate voltages were applied in order to investigate the
Next, the oxidant ROS or H$_2$O$_2$ is introduced to the system reduced DHA population on the nanowire surface is achieved. By applying a given constant gate voltage, the reduced to DHA, and its population is maintained on the nanowire surface by applying a given constant gate voltage. In this way, the equilibrium between the gate voltage and the reduced DHA population on the nanowire surface is achieved. The signals obtained for the detection of H$_2$O$_2$ species from the vicinity of the modified nanodevice, resulting in a conductivity change. Importantly, by removing the ROS or H$_2$O$_2$ species, the equilibrium is perturbed. These species oxidize the electron from the nanowire are injected into the AQ oxidized (AQ) reacted through a decrease in the current signal, similarly to the previously demonstrated electrochemical reduction by the gate voltage (Figure 1). According to these results, we deduce that the electrochemical gate-induced reduction is specific to the AQ redox-reactive moiety.

Figure 3 schematically represents the electrochemical equilibrium sensing mechanism achieved by the redox-reactive moiety modified nanowire device. Initially, the moiety is fully reduced to DHA, and its population is maintained fixed on the nanowire surface by applying a given constant gate voltage. In this way, the equilibrium between the gate voltage and the reduced DHA population on the nanowire surface is achieved. The oxidant ROS or H$_2$O$_2$ is introduced to the system and perturbs this equilibrium. These species oxidize the population of DHA on the nanowire surface, resulting in a conductivity change. Importantly, by removing the ROS or H$_2$O$_2$ species from the vicinity of the modified nanodevice, electrons from the nanowire are injected into the AQ oxidized units, which in turn reduce back to DHA, a process which is also accompanied by a change in the conductivity of the device.

**Metabolites Sensing under Physiological Conditions.** The redox-reactive nanoFET array was first calibrated for sensing of H$_2$O$_2$ and glucose, under physiological conditions (Figures 4 and 5). The signals obtained for the detection of various concentrations of H$_2$O$_2$ (from 100 nM to 1 mM) correlated ($R = 0.961$, Figure 3b) and covered the concentration of H$_2$O$_2$ in plasma ($\sim$1–5 μM). Notably, H$_2$O$_2$ and ROS play a very important role in cellular signaling and are normally produced as a byproduct of metabolism. Nonetheless, increased ROS/H$_2$O$_2$ production may lead to "oxidative stress" and signifies the development of various diseases.

Importantly, the redox-reactive nanoFET is not limited to the detection of ROS/H$_2$O$_2$ and can be forward expanded to the detection of a wide variety of metabolites by coupling the nanodevice with corresponding oxidase enzymes. The calibrated response signal was obtained using eq 1:

$$\text{calibrated response (\%)} = \frac{I_0 - I_t}{I_0 - I_{\text{max,ox}}}$$

where $I_0$ is the current at time $= 0$, $I_t$ is the current at time $= t$, and $I_{\text{max,ox}}$ is the current when the device is maximally oxidized during the measurement, which is the lowest current value.

Furthermore, blood glucose concentration in a healthy body varies from 4.4 to 6.1 mM, while impaired glucose tolerance and diabetes set the blood glucose concentration range to limits as high as 10 mM. First, we performed a set of sensing experiments detecting glucose blood concentration, covering hyperglycemic (>7.8 mM) and hypoglycemic (<3.9 mM) ranges, under physiological conditions (PBS). These measurements (Figure 4) were realized using glucose oxidase, which produces H$_2$O$_2$ from glucose oxidation, as aforementioned. Each time a new sample with different concentrations of glucose is injected into sensing channel, the amperoFET’s nanodevices were turned off ($V_{sd} = 0$ V), in order to prevent electronic noise caused by pressure changes inside the sensing channel during the sample injection. Then, the nanodevices were turned on in amperoFET mode in which the $V_{sd} = 0.3$ V and $V_{g} = -0.4$ V for glucose concentration sensing. These lower electrical voltages better suit future in vivo sensing applications, although less sensitivity compared to the conditions in Figure 4. The obtained calibrated response from the nanodevices for the detection of different concentrations of glucose (from 0.1 mM to 10 mM) was perfectly correlated (correlation coefficients = 0.99 and 1.00, Figure 5b) and covered the glucose concentration range in blood. Moreover, the sensitivity that was achieved with glucose and H$_2$O$_2$ sensing suggests that other important metabolites, like lactate, can be easily monitored.

**Continuous Glucose Monitoring under Physiological Conditions.** Finally, for simulating the most reliable conditions of physiological glucose concentration alterations, including sudden spikes after food consumption, we performed glucose sensing in unprocessed blood samples, spiked with varying glucose concentrations. The blood was continuously injected into a sensing channel through tubing using a syringe pump. The amperoFET nanodevices inside the sensing channel were connected to a printed circuit board (PCB) using a wire bonder in order to connect the sensing chip to the multiplex direct current input/output system (Figure 6d and Figure S7). The nanodevice measured calibrated response correlated ($R = 0.993$) with increasing concentrations of glucose (Figure 6a,b). Next, we performed continuous glucose monitoring in a simulated interstitial fluid, containing 25% bovine serum and 75% PBS, while applying concentration changes to the injected sample, achieved by constant dilution/
concentration of the sample at a known rate. Previous studies demonstrated the correlation between glucose concentrations in blood and in interstitial fluid. The signals acquired from two modified nanowire devices were concentration dependent and correlated, as presented in Figure 6c. Through all the experiments, the source−drain voltage was set to $V_{sd} = 0.3$ V, and $V_g$ was set to 0 V, except times that the gate was used for hot electron injection. (a) Redox reactive devices modified with redox reversible moiety. When applying a sufficient negative gate voltage (higher than $V_g = -0.3$ V and $V_g = -0.4$ at time 240 s and $V_g = -0.5$ at time 420 s) for ~3 min, a decline in the current of the nanodevice is shown, after the $V_g$ was back to 0 V, which indicates a reduction process. (b) Nonredox reactive devices, modified with APDMES only, demonstrate no visible changes in the current after applying a negative gate voltage ($V_g = -0.3$, $V_g = -0.4$, and $V_g = -0.5$) for ~3 min and then turning off $V_g$ voltage ($V_g = 0$).

Figure 2. A comparison between nonredox reactive and redox-reactive moiety modified nanoFET devices. Before each experiment, the AQ and APDMES modified nanoFETs were introduced to 1 mM H$_2$O$_2$ (at time = 0 s). All the measurements were performed in PBS solution (155 mM pH = 7.45). During all the measurements, the $V_{sd}$ was 0.3 V, and $V_g$ was set to 0 V, except times that the gate was used for hot electron injection.

Figure 3. Metabolite sensing mechanism of redox-reactive nanoFET based on nanowire-to-redox moiety electron injection. The left panel of the scheme illustrates the hot electrons ($e^−$) injection, from the conductive band throughout the native oxide layer into the AQ molecules, which receive protons (H$^+$) from the aqueous surroundings, in order to be fully reduced to DHA. In the right panel, the DHA population on the nanoFET surface is maintained fixed by applying a given constant $V_g$ and $V_{sd}$, maintaining a constant conductivity of the device. The presence of H$_2$O$_2$ from oxidase enzyme activity, or other oxidant species, perturbs the DHA population on the nanoFET surface, causing a change in the conductivity of the nanodevice.

and the gate voltage to −0.5 V. These results demonstrate the capability of our system to quantitatively and selectively monitor in real-time different small molecular metabolites directly from complex biofluids, such as blood and interstitial fluid, based on the application of our fully electrically controlled amperoFET devices. In addition, our nanodevice concentration of the sample at a known rate. Previous studies demonstrated the correlation between glucose concentrations in blood and in interstitial fluid. The signals acquired from two modified nanowire devices were concentration dependent and correlated, as presented in Figure 6c. Through all the experiments, the source−drain voltage was set to $V_{sd} = 0.3$ V,
in amperoFET mode can perform continuous metabolic monitoring for at least 2 h (Figure 6c) and can operate properly as a redox reactive nanosensor for a period of several weeks.\textsuperscript{13,48}

CONCLUSIONS

We hereby present the development of a SiNW amperoFET device, modified with redox-reactive moieties, able to perform multiplex, real-time, and continuous monitoring of metabolic activity under real-world physiological conditions. Moreover, our method is calibrated for sensing in physiological environments, without preprocessing of the sample. Electrochemical reduction of the surface-confined redox moieties by direct application of different gate voltages (from $-0.4$ V to $-0.9$ V) was demonstrated. Furthermore, the reduction selectivity for the specific redox-reactive moiety was verified. Using our SiNW amperoFET device, calibration curves for glucose (from 0.1 mM to 10 mM) and H$_2$O$_2$ (from 0.1 $\mu$M to 1 mM) were obtained, covering the entire physiological range. Next, glucose monitoring in unprocessed blood, and continuous glucose monitoring in simulated interstitial fluid, including glucose concentration changes, was successfully performed. For future applications, self-calibration of the sensing platform could be achieved using a combined array of redox-reactive and nonredox reactive devices. All of these findings highlight the potential of our sensing devices for continuous glucose and other metabolites monitoring, as an implantable wearable patch, without the need for additional blood withdrawal for system calibration.
EXPERIMENTAL SECTION

Materials. Gold nanoparticles 20 nm (Ted Pella), poly-L-lysine (Ted Pella), silicon wafer covered with 600 nm thermal oxide layer (<0.005 Ω/cm, SSP prime grade, Silicon Quest International), LOR5A (Microchem) and 500 nm S1805 (Shipley), sodium 9,10-anthraquinone-2-sulfonate (743038, Sigma-Aldrich), oxalyl chloride (O880, Sigma-Aldrich), N,N-dimethylformamide (227056, Sigma-Aldrich), (244511, Sigma-Aldrich), acetone (9005-68, J.T. Baker), isopropanol (9079-05, J. T. Baker), deionized water (18 MΩ·cm), glovebox (150B-G, Mbraun), (3-aminopropyl)-dimethyl-ethoxysilane (SIA0603.0, Gelest), anhydrous toluene (244511, Sigma-Aldrich), anhydrous pyridine (270970, Sigma-Aldrich), plasma-enhanced chemical vapor deposition (Benchmark 800 ICP, Axic), atomic layer deposition (Savannah 200 system, Cambridge Nanotech), Rapid Thermal Processor system (AnnealSys, AS-Micro), probe station include DAQ card (PCI-6030E, National Instrument) and current preamplifier (DL Instruments, model 1211), mass spectroscopy (Autospec M250Q, Waters Corp. USA), X-ray photoelectron spectroscopy (Multi-Technique System 5600, PHI), wire-bonder (Model 8850, West Bond), current recording system (FES-SM32P), syringe pump (Fusion 200, Chemxy).

Silicon Nanowires P-Type Synthesis. The synthesis of silicon nanowires (Si NWs) by chemical vapor deposition (CVD) was performed as previously described (see Supporting Information Section 1).

Silicon Nanowires Field-Effect Transistor Array Fabrication. The SiNW-FET array was fabricated by photolithography as previously described, with minor changes (see Supporting Information Section 2 and Figure S1).

Electrical Characterization of Silicon Nanowire Devices. As an initial quality control, prior to the completion of the SiNW FET devices, the electrical properties of the SiNW devices on the sensor chip were characterized in deionized water by using a probe station.

Figure 6. Glucose sensing in unprocessed blood samples and continuous glucose monitoring in simulated interstitial fluid samples. During all the measurements, the $V_{sd} = 0.3$ V and $V_{g} = −0.5$ V. (a) Calibrated response signals acquired from the redox-reactive nanoFET device after adding different concentrations of glucose to the untreated blood sample (from 2 mM to 5 mM). Prior to the injection of a new sample, the device was switched off ($V_{sd} = 0$ V). (b) Concentration-dependent ($R = 0.993$) calibration curve of the calibrated response versus glucose concentration in the blood sample. The current values of the last 50 s of each measurement were used for the extraction of the calibration curve. See Experimental Section for detailed error analysis. (c) Continuous glucose monitoring in a simulated interstitial fluid sample. The sample was introduced to the device at a flow rate of 5 μL/min (for the times 0–2500 s and 3250–end s) and 15 μL/min (for the times 2500–3250 s), through a fluid delivery system. The simulated interstitial fluid contained 25% bovine serum and 75% phosphate-buffered saline. (d) The entire composition of our sensing system, including the sensing chip with a PDMS microfluidic channel, tubing for fluidic delivery, and electrical contacts. See Supporting Information Sections 8–9 for the detailed system assembly.

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The best performing devices were selected and mapped for future sensing applications (for more details see Supporting Information Section 3 and Figure S2).

**Scanning Electron Microscope Analysis.** The synthesized p-type SiNWs and the SiNW devices were analyzed by Quanta 200 FEG environmental scanning electron microscope (ESEM). The images indicate the good quality of the synthesized nanowires (Figure S3s) and their successful assembly to FET devices (Figure S3b).

**Silicon Nanowire Devices Surface Modification.** Following the fabrication of the SiNW FET array, the chip was chemically modified with 9,10-anthraquinone-2-sulphochloride to enable the sensing of cellular metabolites. The preparation and analysis of 9,10-anthraquinone-2-sulphochloride and surface modification of the nanodevices are described in Supporting Information Sections 5, 6, and 8.

**Fabrication of Fluid Delivery System.** The fabrication of the fluid delivery device from flexible polydimethylsiloxane (PDMS) elastomer was performed as previously described, with minor changes (see Supporting Information Section 8).

**Metabolic Sensing Electrical Measurements.** The selected devices were wire-bonded and integrated into sensing array device chip using the PDMS microfluidic channels which connected to the syringe pump (Figure S7) as previously described. Current versus time signals were recorded at 1 s intervals by the ‘current recording’ system. A more detailed description can be found in Supporting Information Section 9.

**Error Analysis.** To estimate the error in the reported calibrated response values, the standard deviation (SD) of the calibrated response values for 50 s was taken before the devices were switched off (Vd = 0). This method was used for the estimation of the error in Y-axis in Figure 4b and Figure 5b.

**REFERENCES**

(1) Perez-Soroka, H.; Pezvzer, A.; Davidi, G.; Naddaka, V.; Tirosh, R.; Flaxer, E.; Patolsky, F. Optically-Gated Self-Calibrating Nanosensors: Monitoring pH and Metabolic Activity of Living Cells. *Nano Lett.* 2013, 13, 3157–3168.

(2) Kwiat, M.; Elnathan, R.; Kwak, M.; de Vries, J. W.; Pezvzer, A.; Engel, Y.; Burstein, L.; Khatchtourints, A.; Lichtenstein, A.; Flaxer, E.; Herrmann, A.; Patolsky, F. Non-Covalent Monolayer-Fiercing Anchoring of Lipophilic Nucleic Acids: Preparation, Characterization, and Sensing Applications. *J. Am. Chem. Soc.* 2012, 134, 280–292.

(3) Pezvzer, A.; Engel, Y.; Elnathan, R.; Tsukernik, A.; Barkay, Z.; Patolsky, F. Confinement-Guided Shaping of Semiconductor Nanowires and Nanoribbons: Writing with Nanowires. *Nano Lett.* 2012, 12, 7–12.

(4) Pezvzer, A.; Engel, Y.; Elnathan, R.; Ducobni, T.; Ben-Ishai, M.; Reddy, K.; Shpaisman, N.; Tsukernik, A.; Oksman, M.; Patolsky, F. Knocking Down Highly-Ordered Large-Scale Nanowire Arrays. *Nano Lett.* 2010, 10, 1202–1208.

(5) Elnathan, R.; Kwiat, M.; Pezvzer, A.; Engel, Y.; Burstein, L.; Khatchtourints, A.; Lichtenstein, A.; Kantaev, R.; Patolsky, F. Biorecognition Layer Engineering: Overcoming Screening Limitations of Nanowire-Based FET Devices. *Nano Lett.* 2012, 12, 5245–5254.

(6) Krivitsky, V.; Hsiung, L.-C.; Lichtenstein, A.; Brudnik, B.; Kantaev, R.; Elnathan, R.; Pezvzer, A.; Khatchtourints, A.; Patolsky, F. Si Nanowires Forest-Based On-Chip Biomolecular Filtering, Separation and Preconcentration Devices: Nanowires Do It All. *Nano Lett.* 2012, 12, 4748–4756.

(7) Patolsky, F.; Zheng, G.; Lieber, C. M. Nanowire-Based Biosensors. *Anal. Chem.* 2006, 78, 4260–4269.

(8) Krivitsky, V.; Zverzhinetsky, M.; Patolsky, F. Antigen-Dissociation from Antibody-Modified Nanotransistor Sensor Arrays as a Direct Biomarker Detection Method in Unprocessed Biosamples. *Nano Lett.* 2016, 16, 6272–6281.

(9) Leifer, S.; Vizel, R.; Yeru, E.; Granot, E.; Heiifer, O.; Kwiat, M.; Krivitsky, V.; Weil, M.; Yaish, Y. E.; Patolsky, F. Multicolor Spectral-Specific Silicon Nanodetectors Based on Molecularly Embedded Nanowires. *Nano Lett.* 2018, 18, 190–201.

(10) Patolsky, F.; Krivitsky, V.; Heiifer, O.; Zverzhinetsky, M. Method and System for Subcutaneous Sensing. U.S. Patent US20190223795A1, 2019.

(11) Patolsky, F.; Krivitsky, V.; Zverzhinetsky, M. Method and System for Sensing by Modified Nanostructure. U.S. Patent US20180372678A1, 2018.

(12) Kwiat, M.; Cohen, S.; Pezvzer, A.; Patolsky, F. Large-Scale Ordered 1D-Nanomaterials Arrays: Assembly or Not? *Nano Today* 2013, 8, 677–694.

(13) Krivitsky, V.; Zverzhinetsky, M.; Krivitsky, V.; Hsiung, L.-C.; Naddaka, V.; Gabriel, I.; Leifer, S.; Conroy, J.; Burstein, L.; Patolsky, F. Cellular Metabolomics by a Universal Redox-Reactive Nanosensors Array: From the Cell Level to Tumor-On-A-Chip Analysis. *Nano Lett.* 2019, 19, 2478–2488.

(14) Patolsky, F.; Hsiung, L.-C.; Krivitsky, V.; Naddaka, V. Method and System for Sensing. U.S. Patent US20190234900A1, 2019.

(15) Borberg, E.; Zverzhinetsky, M.; Krivitsky, V.; Kodloff, A.; Heiifer, O.; Degabi, G.; Soroka, H. P.; Fainaro, R. S.; Burstein, L.; Leiferu, S.; Diamant, H.; Krivitsky, V.; Patolsky, F. Light-Controlled Selective Collection-and-Release of Biomolecules by an On-Chip Nanostructured Device. *Nano Lett.* 2019, 19, 5868–5878.

(16) Zheng, G. F.; Patolsky, F.; Cui, Y.; Wang, W. U.; Lieber, C. M. Multiplexed Electrical Detection of Cancer Markers with Nanowire Sensor Arrays. *Nat. Biotechnol.* 2005, 23, 1294–1301.

(17) Jain, K. K. Nanotechnology in Clinical Laboratory Diagnostics. *Clin. Chim. Acta* 2005, 358, 37–54.

(18) Burg, T. P.; Godin, M.; Knuuttila, S. M.; Shen, W.; Carlson, G.; Foster, J. S.; Babcock, K.; Malinis, S. R. Weighing of Biomolecules, Single Cells and Single Nanoparticles in Fluid. *Nature 2007*, 446, 1066–1069.

(19) Stern, E.; Wagner, R.; Sigworth, F. J.; Breaker, R.; Fahmy, T. M.; Reed, M. A. Importance of the Debye Screening Length on
Transistors and Biosensors Using Chemical Lift-Off Lithography. Feasibility in Physiological Salt Environments. 2012–2015

InP Monitored by Changes in Substrate Electronic Properties. Chem. Phys. 1989

Neuronal Signals with High-Density Nanowire Transistor Arrays. Zheng, G. F.; Lieber, C. M. Detection, Stimulation, and Inhibition of Neuronal Signals with High-Density Nanowire Transistor Arrays. Science 2002, 2, 487–490.

X.; Lieber, C. M. Electrical Detection of Single Viruses. Proc. Natl. Acad. Sci. U. S. A. 2004, 101, 14017.

Fabrication of High-Performance Ultrathin In2O3 Film Field-Effect Transistors for Detection of Biological and Chemical Species. Science 2006, 313, 1100–1104.

Rothe, J.; Stettler, A.; Chen, Y.; Patolsky, F.; Hierlemann, A. Monolithic Integration of a Silicon Nanowire Field-Effect Transistor Array on a Complementary Metal-Oxide Semiconductor Chip for Biochemical Sensor Applications. Anal. Chem. 2015, 87, 9982–9990.

Leibovitch, R.; Yeung, Y.; Flaxer, E.; Lieber, C. M. Nanowire Biosensors for Highly Sensitive and Selective Detection of Biological and Chemical Species. Science 2001, 293, 1289–1292.

Kim, J.; Rim, Y. S.; Chen, H.; Cao, H. H.; Nakatsuka, N.; Hinton, H. L.; Zhao, C.; Andrews, A. M.; Yang, Y.; Weiss, P. S. Fabrication of High-Performance Ultrasound InSb Film Field-Effect Transistors and Biosensors Using Chemical Lift-Off Lithography. ACS Nano 2015, 9, 4572–4582.

Casal, P.; Wen, X.; Gupta, S.; Nicholson, T.; Hill, A.; Rotho, J.; Stettler, A.; Chen, Y.; Patolsky, F.; Hierlemann, A. Monolithic Integration of a Silicon Nanowire Field-Effect Transistors Array on a Complementary Metal-Oxide Semiconductor Chip for Biochemical Sensor Applications. Anal. Chem. 2015, 87, 9982–9990.

Leibovitch, R.; Yeung, Y.; Flaxer, E.; Lieber, C. M. Nanowire Biosensors for Highly Sensitive and Selective Detection of Biological and Chemical Species. Science 2001, 293, 1289–1292.

Kim, J.; Rim, Y. S.; Chen, H.; Cao, H. H.; Nakatsuka, N.; Hinton, H. L.; Zhao, C.; Andrews, A. M.; Yang, Y.; Weiss, P. S. Fabrication of High-Performance Ultrasound InSb Film Field-Effect Transistors and Biosensors Using Chemical Lift-Off Lithography. ACS Nano 2015, 9, 4572–4582.

Casal, P.; Wen, X.; Gupta, S.; Nicholson, T.; Hill, A.; Rotho, J.; Stettler, A.; Chen, Y.; Patolsky, F.; Hierlemann, A. Monolithic Integration of a Silicon Nanowire Field-Effect Transistors Array on a Complementary Metal-Oxide Semiconductor Chip for Biochemical Sensor Applications. Anal. Chem. 2015, 87, 9982–9990.

Leibovitch, R.; Yeung, Y.; Flaxer, E.; Lieber, C. M. Nanowire Biosensors for Highly Sensitive and Selective Detection of Biological and Chemical Species. Science 2001, 293, 1289–1292.