All-carbon suspended nanowire sensors as a rapid highly-sensitive label-free chemiresistive biosensing platform

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ABSTRACT

Nanowire sensors offer great potential as highly sensitive electrochemical and electronic biosensors because of their small size, high aspect ratios, and electronic properties. Nevertheless, the available methods to fabricate carbon nanowires in a controlled manner remain limited to expensive techniques. This paper presents a simple fabrication technique for sub-100 nm suspended carbon nanowire sensors by integrating electrospinning and photolithography techniques. Carbon Microelectromechanical Systems (C-MEMS) fabrication techniques allow fabrication of high aspect ratio carbon structures by patterning photoresist polymers into desired shapes and subsequent carbonization of resultant structures by pyrolysis. In our sensor platform, suspended nanowires were deposited by electrospinning while photolithography was used to fabricate support structures. We have achieved suspended carbon nanowires with sub-100 nm diameters in this study. The sensor platform was then integrated with a microfluidic chip to form a lab-on-chip device for label-free chemiresistive biosensing. We have investigated this nanoelectronics label-free biosensor’s performance towards bacterial sensing by functionalization with Salmonella-specific aptamer probes. The device was tested with varying concentrations of Salmonella Typhimurium to evaluate sensitivity and various other bacteria to investigate specificity. The results showed that the sensor is highly specific and sensitive in detection of Salmonella with a detection limit of 10 CFU mL⁻¹. Moreover, this proposed chemiresistive assay has a reduced turnaround time of 5 min and sample volume requirement of 5 µL which are much less than reported in the literature.

1. Introduction

Carbon, the building block life, is now poised to become the building block of next-generation electronics. Carbon nanomaterials such as graphene and carbon nanotubes (CNT) offer superior electrical, thermal and mechanical properties as well as energy efficiency than current silicon-based electronics (Peng et al., 2014; Shulaker et al., 2013). Furthermore, their fast electron transfer kinetics, excellent conductivity, and ease of biofunctionalization give rise to numerous sensor applications (Barsan et al., 2015; Hernández et al., 2014; Qiu et al., 2015). However, these two allotropes of carbon, graphene and CNTs, are heterogeneous and hard to handle (Yang et al., 2010). Moreover, they are difficult to fabricate into any desired shapes, for instance, a suspended nanowire for electronic sensors. Graphene and CNT based bio transistor sensors have been synthesized by chemical vapor deposition or chemical reduction methods which are still hard to integrate with established large-scale manufacturing techniques for electronics. Currently, nanowires in electronic sensors are mainly fabricated by focused ion beam (Fujii et al., 2017) and electron beam lithography (Cui et al., 2001; Juhasz et al., 2005). While these techniques are capable of high-resolution patterns down to tens of nanometers, they are relatively expensive and slow for mass-manufacturing. In this report, we fabricated highly defined suspended carbon nanowires by simple electrospinning.

Keywords:
Suspended carbon nanowire
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process combined with standard Carbon-Microelectromechanical Systems (C-MEMS) techniques for rapid, ultrasensitive, label-free electronic biosensor application.

C-MEMS offers a way to control fabrication of carbon materials in desired shapes from macro to nanoscale dimensions. A typical C-MEMS fabrication involves patterning polymer structures and subsequent carbonization to convert them into carbon structures. Patterning can be performed by standard photolithography techniques on photosensitive polymers with high carbon content. SU-8 photoresist is a typical polymer used in C-MEMS because they are patternable to high aspect ratio and 3-dimensional structures (Martinez-Duarte, 2014). Carbonization is performed by pyrolysis of patterned polymer structures in an inert atmosphere at an elevated temperature ranging from 500°C to 120°C. This process converts polymer patterns to conductive carbon while retaining their shape. Carbon structures obtained from this technique are found to have glossy carbon like properties (Martinez-Duarte, 2014). Glassy carbon is a widely used electrode material for electrochemical analysis because of its inert nature, stability and wide electrochemical windows (Bollella et al., 2017). Using C-MEMS fabrication techniques, researchers have fabricated high aspect ratio carbon micro 3D electrodes for various applications. These include next-generation battery electrodes (Long et al., 2004), electrochemical sensors (Kamath and Madou, 2014), gas sensors (Sharma and Madou, 2012) and glucose sensors (Xi et al., 2013).

While photolithography in C-MEMS offers a way to fabricate high aspect ratio microstructures, electrospinning provides a simple and cost-effective way to fabricate nanostructures. In electrospinning, a high voltage (5-30kV) is applied between syringe tip and collector. The electric field stretches polymer droplet to nanoscale as it ejects from syringe tip towards collector and deposits as a nanofiber mat. Electrospinning has been widely explored in a variety of applications like filters filtration (Gopalan et al., 2006), tissue engineering scaffolds (Li et al., 2002) to polymer field effect transistors (FET) (Babel et al., 2005; Manuelli et al., 2014). However, a major obstacle of integrating electrospinning into electronics and sensor fabrication is that electrospinning inherently is a random process, improving control of electrospinning process, with the focus on positioning single nanowires and integrating with support structures, is an active research area (Pramanick et al., 2016). Recently, Prakash et al. electrospin multi-walled carbon nanotubes (MWNT) embedded SU-8 polymer fibers on micro copper poles for biosensing (Prakash et al., 2016). The authors reported controlling electrospinning time for deposition of one single fiber on the support structure. This way of controlling, however, tends to require mostly manual observation of resultant fibers (Sharma et al., 2012). In a more recent study, Paul et al. (2017) have reported deposition of MWNTs-zinc oxide nanofibers as stretchable biosensors deposited between gold electrodes. Although fibers were fabricated by electrospinning, fiber mats were collected by dissolving in a solvent and subsequent drop-casting of the solution onto support structures. In this report, we describe on-site suspended carbon nanowire fabrication by integration of electrospinning and photolithography. By electrospinning photoresist polymer, the resulting mat can be photopatterned and crosslinked selectively in the desired area. Thus, this can be integrated with standard photolithography methods for suspended nanowire sensor fabrication. By converting the resulting electrospun photoresist nanowires to carbon by the C-MEMS fabrication, we can achieve photo-patterning fabrication of carbon nanowires while pushing C-MEMS forward into nanodomain as Carbon Nanoelectromechanical Systems (C-NEMS).

On-chip electrochemical biosensors technologies utilize varieties of analytical methods to probe molecular bindings at the electrode surface to liquid interface. These include cyclic voltammetry (Zhao and Liu, 2016), chronoamperometry (Lin et al., 2017) and electrode impedance spectrometry (Jiang et al., 2014). Polymeric and metal nanowire electrochemical biosensors which use above sensing methods can be fabricated by electrochemical deposition of nanowires on a planar substrate (Lin et al., 2012; Spain et al., 2015; Zhuang et al., 2016). These methods employ current sensing in electrodes through a liquid medium and analyze changes in charge transfer characteristics. On the other hand, in chemiresistive biosensors, the conductivity of sensor material changes in response to surface binding. As the dimension of sensor components in chemiresistive sensors are comparable to Debye length, a slight change in surface properties can vary the material’s electronic properties significantly (Prakash et al., 2016). Here, sensing took place in the sensor substrate and the current doesn’t need to flow through the liquid medium. Hence, unlike other electrochemical biosensing platforms, chemiresistors do not require conductive medium and redox species to perform analysis, resulting in shorter time, simpler assay protocol and reduced reagents. Besides, chemiresistive sensors require simpler instrumentation for analysis as only the conductivity of substrate need to be measured (Janata, 2009) making them more suitable for point-of-care diagnosis. Hence, they have been widely applied in biosensing applications such as DNA sensing, protein sensing and bacteria sensing (Paul et al., 2017; Prakash et al., 2016; Rajesh et al., 2013). Our on-site electrospun deposition enables suspension of nanowire sensors to the height of tens of micrometers, which has additional advantages of having better contact with the analyte as target molecules can bind from all sides and reducing electronic interface from the substrate surface (Pramanick et al., 2016).

Among biorecognition elements for sensors, aptamers have become recent alternatives for the traditional usage of antibodies as biorecognition element. Although antibodies were used as conventional probes in biosensing due to their high affinity and high specificity to a broad range of analytes, some limitations and drawbacks still hinder their development as biosensors (Zhou et al., 2014). For instance, antibodies are susceptible to chemical modification and high temperatures (Han et al., 2010). The production of antibodies is also an expensive and complex process as compared to in-vitro selection and amplification process of the aptamer (Han et al., 2010). Aptamers are single-stranded nucleic acid (DNA or RNA) that interacts and binds the target analyte with high affinity, selectivity, and sensitivity (Hamaguchi et al., 2001). Aptamers can be easily tailored and chemically modified to bind with various target analytes such as antibodies, nucleic acids, cells, and microorganisms with dissociation constants of aptamer-target complexes ranging from picomolar to micromolar (Hamula et al., 2006). Owing to these advantages, extensive research and effort have been channeled to the application of aptasensor for foodborne pathogen detection. Besides offering high-stability to chemical modifications, aptamers are also attractive candidates for novel, label-free and direct detection of whole-cell bacteria using electronic biosensing platform. Aptamers can interact and undergo conformational changes with outer membrane proteins of a bacterial cell to form a unique 3D-structure, and thus enable them to discriminate between protein isoforms of another bacterial cell (Davydova et al., 2016; Joshi et al., 2009; Muniai et al., 2017). Another major advantage of aptamer to antibodies as biorecognition probes is that aptamers are much smaller than antibodies and hence, aptamer probes reduce distance between the electrode surface and bio-analyte. This results in higher sensitivity in FET sensors and chemiresistive sensors by reducing Debye screening effect (Maehashi et al., 2007; Zhang et al., 2016).

In this work, we report the application of suspended carbon nanowire based aptasensor as an innovative platform for chemiresistive biosensing. The suspended carbon nanowires were fabricated by integrating electrospinning and photolithography with C-MEMS techniques. The performance of the sensor platform was evaluated for the label-free detection of Salmonella Typhimurium (S. Typhimurium), a common food-borne pathogen, using a specific aptamer probe. Amine-ended aptamers were immobilized on suspended nanowire surface by
means of carbodiimide crosslinker chemistry with carboxylic groups on the carbon nanowire. This nanoelectronics biosensor platform enables rapid, real-time, highly-sensitive and specific label-free detection. By functionalization with other suitable aptamer probes, this platform can also be used for label-free detection of various analytes such as DNA, antibodies and cancer cells.

2. Methods and materials

2.1. Fabrication of suspended carbon nanowire electrodes

Suspended carbon nanowire sensors were fabricated by C-MEMS process of SU-8 photolithography followed by carbonization. First, SU-8 2015 photoresist (MicroChem, USA) was spin-coated to 15 μm thickness onto a silicon wafer coated with a silicon dioxide insulation layer. Supporting structures were then patterned using standard photolithography techniques which includes soft bake, UV exposure under a designed mask, post-exposure bake followed by resist development in SU-8 developer (MicroChem, USA). The baking temperature and exposure were primarily followed from photoresist datasheet albeit a slight variation of longer baking time to optimize for pyrolysis. After development, wafers were dried in nitrogen gas flow.

SU-8 nanofibers were then electrospun on the patterned silicon wafer. Electrospun parameters were optimized to achieve sub-100 nm fibers after pyrolysis. Second step photolithography under a different mask was then performed to selectively crosslink the suspended nanowires between support structures. The wafer was then developed again in resist developer to dissolve and wash non-crosslinked fibers away.

The resultant SU-8 structures were converted to carbon electrode by pyrolyzing in a tube furnace (Nabertherm, Turkey) under nitrogen gas flow at a flow rate of 2000 sccm. The pyrolysis step involved annealing at 300 °C for 30 min and then increasing temperature to 900 °C with a ramp rate of 6 °C/min. The furnace temperature was then held at 900 °C for 1 h before cooling to room temperature under nitrogen flow.

2.2. Functionalization of carbon nanowires

Carbon electrodes fabricated using C-MEMS process are found to be inert with little to no functional groups on the surface. Hence, surface functionalization is necessary to prepare nanowire electrodes for biofunctionalization. Activation of carboxylic (−COOH) groups on pyrolyzed carbon surface was performed by plasma treatment as described in previous studies (Hirabayashi et al., 2013; Thiha et al., 2016).

2.3. Fabrication of microfluidic chip

The base microfluidic structures on sensor chip were fabricated using photolithography of SU-8 photoresist as shown in Fig. 1. This step has the dual purpose of insulating non-active electrode area and making a microfluidic channel on the chip exposing only nanowire sensing element. The process involved spin-coating carbonized nanowire electrodes with SU-8 photoresist to a thickness of 3 μm. SU-8 microfluidic pattern was obtained after UV exposure under a designed mask and resist development. The resultant sensor chips were diced and incorporated into a microfluidic housing made of Poly(methyl methacrylate)

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**Fig. 1.** Schematic illustration for fabrication steps of carbon nanowire biosensor. Insets show the magnification of suspended nanowire area.
(PMMA) fabricated using Computer Numerical Control (CNC) machining. Binding between the SU-8 microfluidic layer on the sensor chip and PMMA structures were achieved by a pressure sensitive adhesive layer which was patterned using a cutting plotter (GCC PUMA II, USA).

2.4. Immobilization of DNA aptamer probe on carbon biosensor

First, Salmonella-specific DNA aptamer probe (Joshi et al., 2009) with sequences of (5’-TATGGCGGCTACCCGACGGAGCTTGACAT-TATGACAG-3’) was modified with amine (-NH$_2$) group at the 5’ end. Aptamers were dissolved in deionized water to a concentration of 5 µM. The amine-functionalized aptamer was covalently attached to the carboxylic groups with the assistance of sulfo-N-hydroxysuccinimide (sulfo-NHS) and N-(3-dimethylaminopropyl)-N-ethylcarbodiimide hydrochloride (EDC) (sulfo-NHS 0.1 M, EDC 0.4 M) mixed in 0.1 M MES buffer (pH 5.5–5.9) for 30 min. The electrodes were then washed with 0.1 M MES buffer to remove the unbound residues and were dried. Then, 5 µL of 5 µM aptamer solution was drop-casted on the sensor chip and incubated for another 1 h. Current-voltage (I-V) characterization was used to investigate conductivity of nanowire in each step of functionalization.

2.5. Preparation of bacteria and bacterial sensing

To evaluate the biosensing platform, 5 µL volume of bacterial suspensions of different concentrations were introduced to microfluidic chips and incubated for 5 min at room temperature. Serially diluted S. Typhimurium cell suspensions with concentrations ranging from 10$^5$–10$^7$ CFU mL$^{-1}$ were used in the experiment. Dilutions of bacterial cultures were confirmed by viable plate count. To test the selectivity of the sensing platform, different types of bacteria namely, Escherichia coli, Shigella dysenteriae, Vibrio cholerae and Klebsiella pneumoniae suspensions were prepared. After incubating with the bacterial suspensions for 5 min, the I-V characterizations were carried out for the individual sensor chips. Bacterial cultures were obtained from the culture collection of the Biomedical Science Laboratory, University of Malaya, Malaysia. The developed biosensor platform was further tested with real food samples. 10 g of beef samples were collected from 6 different markets. The samples were homogenized in 225 mL of Buffer Peptone Water (BPW) for 4 h at room temperature. To compare electronic detection results with plate count, meat homogenate was transferred into Rapport Vassiliadis broth followed by 24 h of incubation at 42°C. This enriched broth was then serially diluted to obtain a bacterial suspension in a range of 10$^3$–10$^7$ CFU mL$^{-1}$. Bacterial cell density in diluted suspensions were evaluated by plating onto Brilliance Salmonella agar plates followed by incubating at 37°C. Parallelly, diluted bacterial suspensions were introduced to carbon nanowire electrodes and incubated for 5 min followed by the electrical detection. The results were cross-checked with viable plate count method.

2.6. Electrical characterization

The fabricated suspended carbon nanowires were electrically characterized before and after functionalization and after incubation with bacterial suspensions at different concentrations. The I-V characterization was carried out from -0.1 V to 0.1 V at a scan rate of 10 mV/s in Autolab electrochemical workstation. Changes in resistance as a percentage were calculated and plotted.

3. Results and discussion

3.1. Suspended nanowire electrode fabrication

First, electrospinning parameters were optimized to obtain SU-8 nanofibers with thin and uniform diameters. SU-8 2015 solution was electrospun with a 22-gauge metal syringe at varying distances, voltages and flow rates. Electrospun fibers are deposited on a silicon substrate and observed under optical microscopy and field emission scanning electron microscopy (FESEM). The optimization process was carried out by first eliminating the parameters that produced beads, short ends, beaded fibers or fibers with diameters more than 1 µm. Then, the voltage and distance from needle-to-collector were fine-tuned while keeping flow rate constant as shown in Fig. 2. Needle-to-collector distance less than 10 cm resulted in occasional large polymer droplet de-

![Fig. 2. Optimization of electrospinning parameters of voltage and distance between needle and collector at the flow rate of 10 µL/min.](image-url)
posing on the silicon substrate while voltage greater than 12 kV produced spindle short ends instead of cylindrical wires. The finest and most uniform nanofibers were deposited at a voltage of 12 kV with a needle-to-collector distance of 100 mm at a flow rate of 10 μL/min, resulting in average fiber diameter of 275 nm with a standard deviation of 96 nm. These conditions were used to synthesize nanofibers in subsequent fabrication steps.

Step-by-step evolution of polymer nanowire to carbon nanowire sensor chip is illustrated in Fig. 3. Confined UV exposure at supporting structures crosslinks SU-8 polymer fibers between them. After development, non-crosslinked fibers are washed away while retaining crosslink polymer nanowires as shown in Fig. 3(b). Upon pyrolysis, the supporting structures shrank at the top while the base retained the original size. This shrinkage of support structure mechanically stretched the nanowire during pyrolysis making it longer, thinner and more graphitic by rearranging carbon layers under strain (Sharma et al., 2012). We have also observed that the suspended nanowires shrank 50-70% during pyrolysis. The gap between the top of the support structures widens from 20 μm to 44 μm as can be observed from micrographs, extending nanowire to an average 100% of the original length. As fabricated, carbon nanowires were found to have an average diameter of (84 ± 29) nm with the height of suspension at 10 μm as measured from micrograph. This suspension enabled the carbon nanowire to have better contact with bacteria suspended inside the solution while eliminating electronic interferences from the silicon surface. During fabrication process, we used a 100 nm diameter silicon wafer which can hold 24 electrode patterns. When electrospinning was performed on the whole silicon wafer, suspended wires were obtained in 19–20 electrodes with patterns on the edge of wafer sometimes failed to receive nanowire deposition. This translates to yield of approximately 80% of suspended nanowire deposition in 5 repetitions of our wafer fabrication under optimized parameters. Among which, approximately 20% results in single nanowire suspension while two to a maximum of three nanowires were deposited in the rest. Micrographs of multiple suspended nanowire depositions can be found in Supplementary materials. Fig. 3(e) shows the sensor chip after insulating with the SU-8 layer. Microfluidic channel exposed the nanowire only while covering supporting carbon structures. This is to avoid unwanted immobilization and chemical reactions at supports which could affect the analysis. Fig. 3(b) shows the as-fabricated sensor integrated with PMMA microfluidic chip. Electrode contact pads were coated with silver paste for better contact with mini-crocodile clip and prevent scratching of the carbon layer during the experiments. Silver paste layer also reduced the contact resistance between the clip and sensor giving a more reliable reading of sensor value.

Carbon nanowires show a reduction in resistance after functionalization with aptamer probe as shown in Fig. 4. However, there is no significant change in resistance of the nanowire in the presence of distilled water or phosphate-buffered saline (PBS) solution which agrees with MW-CNT chemiresistors in a previous study (Fu et al., 2017). We have also observed there is no difference in resistance between wet and dry electrodes after functionalization step. Hence, bacterial sensing experiments were performed in the wet state of the sensor. This is important as, in some existing chemiresistive sensing reports, electrical measurements were performed only after drying of the sensors which is not practical for a rapid lab-on-chip diagnostic device (Fu et al., 2017; Paul et al., 2017).

3.2. Chemiresistive sensing of bacterial cells

In chemiresistive bacterial sensing, binding of bacteria to nanowire can be determined by measuring changes in conductivity of sensor. I-V characterization of electrodes was carried out before and after introduction and incubation with the analyte. After inserting 5 μL of prepared bacteria-containing solution droplet onto the microfluidic chip, the sensor was incubated for 5 min to allow for binding before recording the second I-V characterization. Resistance is obtained from the inverse of I-V curve slope. The percentage changes in resistance after incubation was calculated as ΔR/R0 where ΔR is difference between resistance after incubation with bacteria (R) and original resistance (R0).

From the calibration curve correlating bacteria concentration and percentage change from Fig. 5(a), we can observe that an increase in S. Typhimurium concentration results in a reduction in nanowire resistance with correlation coefficient R2 of 0.95. This increase in conductivity can be explained by the fact that Salmonella is a Gram-negative bacterium which has lipopolysaccharides on its cell wall contributing strong negative charges. Hence, upon binding with the aptamer, negative charges induce electron holes in nanowire, making the carbon nanowire more conductive (Hepel, 2012; Kaisti, 2017). This finding agrees with previous reports of E. coli sensing on graphene (Huang et al., 2011; Mohanty and Berry, 2008).

The specificity of the nanowire sensor to S. Typhimurium was also investigated by incubating with different bacteria (E. coli, Sh. dysenteriae, V. cholerae and K. pneumoniae) with 10^2 CFU mL^-1 concentration and I-V analysis was carried out after 5 min of incubation. The sensors showed no significant changes of conductivity for non-Salmonella bacteria as shown in Fig. 5(b). The results demonstrate the fabricated carbon nanowire chemiresistive sensors are highly selective and specific for S. Typhimurium detection. The sensor was also tested on real food samples collected from the markets (n = 6). Our sensor showed detection of S. Typhimurium in all samples which were also confirmed by

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![Fig. 3. Fabrication of carbon nanowire sensor chip. (a) Electrospin SU-8 nanowire mat on support structures. (b) Single suspended SU-8 nanowire after photolithography and development. (c) Suspended carbon nanowire after pyrolysis. (d) Titled side view of suspended carbon nanowire showing suspension. (e) Nanowire sensor in the SU-8 microfluidic channel. (f) Magnification of SU-8 nanowire from (b). (g) Carbon nanowire after pyrolysis. (h) Suspended carbon nanowire chip inside microfluidic platform.](image-url)
plate count. Quantitative estimation of *S. Typhimurium* was carried out by comparing resistance change with calibration curve equation in Fig. 5(a). The results were compared with viable plate count obtained after overnight incubation in Brilliance *Salmonella* agar (Table 1). Estimation of bacteria cell density from the plate was calculated as number of colonies on the plate multiplied by reciprocal of the dilution factor of samples. The results indicated that the sensor can predict value within an order of magnitude in food samples.

The lower limit of detection of the bacterial sensor was determined to be 10 CFU mL\(^{-1}\). This is lower than conventional detection methods of *Salmonella* such as polymeric chain reaction (PCR) and loop-mediated isothermal amplification (LAMP). Comparison of carbon nanowire sensor with different reported techniques is summarized in Table 2. Our detection limit is equal to potentiometric aptasensor recently reported (Munisamy et al., 2017). However, the latter is not a point-of-care microdevice. Among point-of-care lab-on-chip systems, our suspended carbon nanowire sensor has much lower detection limit than recently reported lab-on-Disk microdevice for the foodborne pathogen detection which has the detection limit of 2.7×10\(^4\) CFU mL\(^{-1}\) (Sayad et al., 2018). Moreover, we have achieved the *Salmonella* detection using just 5μl of sample volume. This is much less than recent lab-on-chip platforms which require 12.5–25μl for *Salmonella* detection (Oh et al., 2016; Sayad et al., 2018). PCR and LAMP assays employ DNA extraction, amplification, and florescence detection which require a total of 1–3h for a test. They also usually require centralized facilities

Table 1
Comparison of quantitative *Salmonella* detection in fresh beef samples (n = 6) between plate count and nanowire platforms.

<table>
<thead>
<tr>
<th>Food Samples</th>
<th><em>Salmonella</em> concentration from plate count (CFU mL(^{-1}))</th>
<th>Measured concentration by sensor platform (CFU mL(^{-1}))</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>1.2 × 10(^2)</td>
<td>2.0 × 10(^2)</td>
</tr>
<tr>
<td>2</td>
<td>4.0 × 10(^2)</td>
<td>2.2 × 10(^2)</td>
</tr>
<tr>
<td>3</td>
<td>6.0 × 10(^3)</td>
<td>3.9 × 10(^3)</td>
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<tr>
<td>4</td>
<td>1.6 × 10(^4)</td>
<td>1.6 × 10(^3)</td>
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<tr>
<td>5</td>
<td>2.7 × 10(^3)</td>
<td>4.3 × 10(^3)</td>
</tr>
<tr>
<td>6</td>
<td>8.5 × 10(^3)</td>
<td>1.2 × 10(^4)</td>
</tr>
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and skilled technicians, even though this has started to change with miniaturized point-of-care LAMP system recently introduced (Sayad et al., 2018). However, assay time has not improved much. On the other hand, the graphene oxide based sensor from Muniandy et al. (2017) which used differential pulse voltammetry analysis in a three-electrode electrochemical cell, has reduced assay time to 10 min.

In our proposed chemiresistive assay, the total assay time was estimated to be 5 min, i.e., 5 min of incubation and a few seconds for conductivity measurement. Our assay time is much shorter than conventional methods and even state-of-the-art electrochemical detection methods for S. Typhimurium. In addition, our chemiresistive sensing technique provides a simple, real-time one-step measurement without the need for additional electrochemical buffers. Compared to PCR and LAMP techniques, this carbon nanowire based aptasensor theoretically does not require culturing bacteria and DNA extraction steps, which can add days to total analysis time.

4. Conclusions

We have demonstrated a facile method of fabricating suspended carbon nanowire electrodes and their use as chemiresistive biosensor platform. Sub-100 nm carbon nanowires suspended at 10 μm height between two carbon support structures have been fabricated in this study. This suspended carbon nanowires functionalized with Salmonella-specific aptamer showed rapid detection of S. Typhimurium, foodborne pathogenic bacteria, in 5 min assay time with high sensitivity and specificity while requiring 5 μL of total sample volume for analysis. Hence, our nanoelectronics sensor was able to provide simple, rapid and one-step detection of whole cell bacteria without the need for DNA extraction or additional analytical reagents required in other detection methods. With the on-chip integrated microfluidic system, our lab-on-chip bacteria testing can be applied as a point-of-care detection device for foodborne pathogens. This sensor platform can also be used to detect various biomolecules by functionalization with other appropriate aptamers.

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Conflict of interest

All authors declare no conflict of interests.

Appendix A. Supplementary material

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.bios.2018.02.024.

References
