

Solid-state nanopores for biosensing with submolecular resolution

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Abstract

Biological cell membranes contain various types of ion channels and transmembrane pores in the 1–100 nm range, which are vital for cellular function. Individual channels can be probed electrically, as demonstrated by Neher and Sakmann in 1976 using the patch-clamp technique [Neher and Sakmann (1976) *Nature* **260**, 799–802]. Since the 1990s, this work has inspired the use of protein or solid-state nanopores as inexpensive and ultrafast sensors for the detection of biomolecules, including DNA, RNA and proteins, but with particular focus on DNA sequencing. Solid-state nanopores in particular have the advantage that the pore size can be tailored to the analyte in question and that they can be modified using semi-conductor processing technology. This establishes solid-state nanopores as a new class of single-molecule biosensor devices, in some cases with submolecular resolution. In the present review, we discuss a few of the most important recent developments in this field and how they might be applied to studying protein–protein and protein–DNA interactions or in the context of ultra-fast DNA sequencing.

Introduction

Biological cell membranes contain various types of ion channels and transmembrane pores in the 1–100 nm range, which are vital for cellular function. Individual channels can be probed electrically, as demonstrated by Neher and Sakmann in 1976 [1] using the patch-clamp technique. Since the 1990s, this work has inspired the use of protein or solid-state nanopores as inexpensive and ultrafast sensors for the detection of biomolecules, including DNA, RNA and proteins, but with particular focus on DNA sequencing [2,3]. The basic principle behind this novel biosensor is relatively simple: the nanopore membrane separates an electrolyte-filled cell into two compartments, which are connected through a single or small number of well-defined nanopores. Each compartment contains at least one electrode, typically a silver/silver chloride electrode immersed in a chloride-containing solution. Upon application of a bias voltage, an ion current starts to flow due to the conversion of silver and silver chloride at the anode and cathode respectively. The steady-state current depends on the dimensions of the nanopore, the conductivity of the electrode and the bias voltage; surface effects become important for small nanopores. According to Ohm's law, at constant applied bias voltage, the potential fall in the cell is largest at the

nanopore, since the nanopore resistance R_{pore} is typically large compared with the solution resistances R_s . With a membrane thickness of the order 100 nm, this means that the local electric field 1 V bias is approximately 10^7 V/m. This field is the driving force for the transport of charged species through the pore ('translocation'). In a simple volume-exclusion model, a translocating biomolecule blocks part of the pore volume and thus reduces the number of ions transported in a given time. The ion current is decreased as long as the biopolymer resides inside the pore and returns to the 'open pore' value immediately after it leaves [4–6] (Figure 1a). More accurate models take into account the charge of the translocating biomolecule, relative to the electrolyte, and conformational effects, as well as surface interactions. Interestingly, translocation can also lead to an increase in the pore current, if the biomolecule is highly charged and the electrolyte concentration is low [7]. If the nanopore is small enough, a biopolymer such as DNA is forced into a linear configuration, which may be exploited for single-biopolymer analysis with potentially submolecular resolution [7] (see below).

Detection of DNA translocation through solid-state nanopores was first demonstrated by Li et al. [8] for 500-bp DNA fragments. The nanopores were fabricated by ion beam sculpting into Si_3N_4 membranes. Further studies showed that information about DNA length, structure and conformation could be extracted from the associated current blockades [5,9,10].

In addition, biological and solid-state nanopores allow discrimination of different nucleic acids, ss (single-stranded) DNA and ds (double-stranded) DNA [11–14], unzipping dsDNA and hairpins [15,16], as well as determining the folding state of dsDNA [5,9,10,14].

Key words: biosensing, DNA sequencing, graphene, nanopore, protein–DNA interaction, protein–protein interaction.

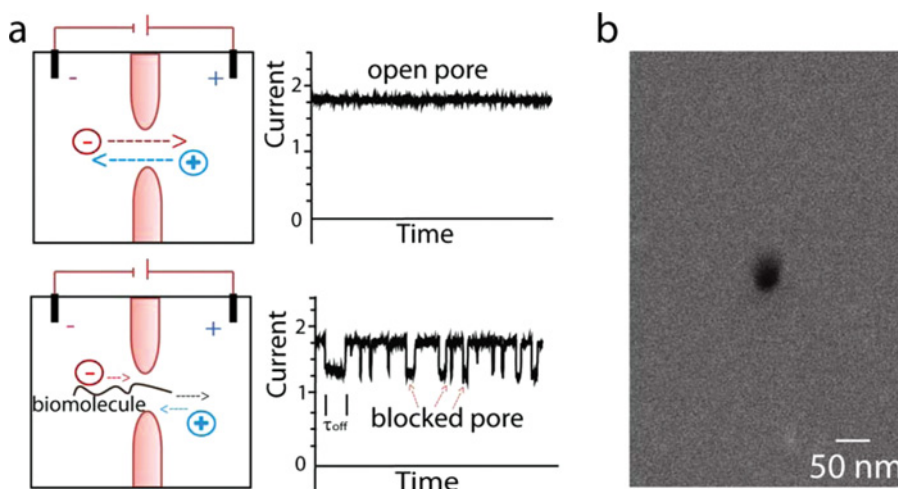
Abbreviations used: ds, double-stranded; β -hCG, β -human chorionic gonadotropin; PNA, peptide nucleic acid; ss, single-stranded; STM, scanning tunnelling microscopy.

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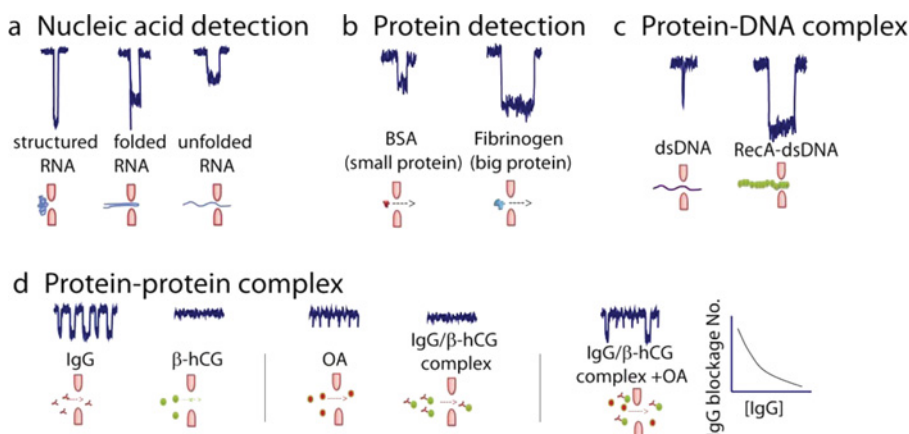
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Figure 1 | Principle of nanopore sensors for biomolecule analysis

(a) Biomolecule translocation through a solid-state nanopore. The membrane (in red) separates two chambers filled with electrolyte solution (e.g. 0.1 M KCl). A bias voltage applied between the two electrodes results in ion transport according to the potential gradient in the cell. The strong local electric field at the pore captures a (charged) biomolecule in the vicinity of the pore, pulls it through and into the opposite fluid compartment. Biopolymer translocation leads to a transient current modulation, which can be related to the properties of the biomolecule. See the main text for further details. (b) Example of a solid-state nanopore, fabricated in an Si_3N_4 membrane. The pore was both milled and imaged using a focused ion beam (FIB)/scanning electron microscope (SEM) instrument (Carl Zeiss XB 1540 Cross-Beam; ion acceleration voltage at 30 kV; milling current at 1 pA).

**Figure 2 | Solid-state nanopores for sensing biomolecular interactions**

(a) Nucleic acid detection; folded and unfolded RNA have caused difference ionic current blockages, which could be detected when translocating through 5 nm Si_3N_4 pores [14]. (b) Protein detection; different sizes of proteins (BSA and fibrinogen) could be estimated on the basis of ionic current blockages when they translocate through 16 nm Si_3N_4 pores [17]. (c) Protein-DNA complex; free dsDNA and RecA protein coating on dsDNA could be detected with 20–30 Si_3N_4 pores [39]. (d) Protein-protein complex: IgG and ovalbumin (OA) translocation through the nanopore caused high and low ionic current blockage levels respectively, whereas β -hCG could not be detected when translocating through the same nanopore. It was suggested that translocation of the IgG- β -hCG complex is too fast to be detected because of an increased electrophoretic mobility of the bound antibody at low pH. Addition of β -hCG to the solution containing the mixture of IgG and ovalbumin decreased the translocation frequency of IgG, which indicates binding of IgG and β -hCG [38].

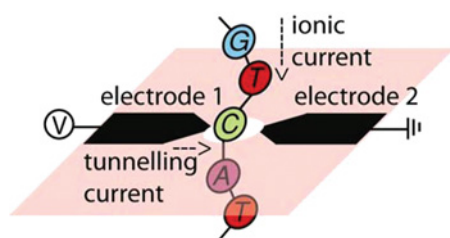


In 2007, Li and co-workers were the first to demonstrate protein translocation through solid-state nanopores using negatively charged BSA [17] (Figure 2b). They confirmed that the relative charge and size of protein molecules could be estimated from the amplitude of

the current blockage, the duration of the translocation event and the charge transferred. Subsequent work also showed that protein denaturation in the presence of guanidium chloride could be detected using Si_3N_4 nanopores [18–20].

Figure 3 | Translocation of a DNA strand in a tunnelling current detector integrated into a nanopore device

Tunnelling electrodes (black) are deposited on the membrane material (pink) in the vicinity of the nanopore [32].



Metallic nanopores have been used for DNA translocation experiments recently, which holds interesting prospects for externally modulating the charge on the pore surface (by applying an external potential) and surface modification (by self-assembled monolayers) [21,22]. Both aspects may be exploited for controlling the translocation process or for detecting stochastic binding to the nanopore surface [23].

However, apart from difficulties in controlling the dynamics of the translocation process (e.g. speed or molecular configuration), the lack of spatial resolution and molecular specificity has proven to be a significant shortcoming, hampering the application of nanopore sensors in gene profiling or DNA sequencing, for example. To this end, several approaches are currently under development aiming to overcome these issues, which is the focus of the present review. The interested reader is referred to the review by Howorka and Siwy [24] for a more comprehensive overview of nanopore sensing as a field.

Solid-state nanopores as sensors with submolecular resolution

DNA sequencing-by-tunnelling in a nanopore

One way to improve both the spatial resolution and specificity of nanopore sensors may be to replace the ion current as a probe by the tunnelling current, for example in the configuration shown in Figure 3. A pair of electrodes with a gap smaller than 5 nm and aligned with the pore opening then serves as the 'read head', while DNA passes through the pore. Tunnelling is a quantum-mechanical effect, which rests on subtle electronic orbital interactions between two closely spaced electrodes, mediated by the molecular medium in between. The latter provides some degree of molecular specificity. In the so-called coherent tunnelling regime, the tunnelling current has a characteristic decay length of ~ 1 nm. This ultimately results in high spatial resolution [cf. STM (scanning tunnelling microscopy)], because even closely spaced features may be resolved with subnanometre resolution. The combination of both features, i.e. high spatial resolution and electronic specificity, may thus help to address the two issues mentioned above.

The notion of 'sequencing-by-tunnelling' is supported by theoretical work by Zwolak and Di Ventra [25], and experimental results from the Lindsay and Kawai groups using STM and mechanical break-junctions [26–29], demonstrating that: (i) individual DNA bases can be distinguished on the basis of their tunnelling conductance [26,30], provided that there is a well-defined base-electrode interaction (recognition tunnelling); (ii) the bases remain electronically distinct, even when part of an oligonucleotide [31]; (iii) individual bases within short DNA oligomers can be identified, which is another important step towards DNA sequencing-by-tunnelling [27,28]; and (iv) differentiation of methylated and non-methylated cytosine has been shown, opening up perspectives of epigenetic profiling [28]. Integration into a high-throughput platform (such as nanopore sensors) is an important pre-requisite for commercial applications in sequencing or diagnostics. This is not trivial from the device engineering point of view, e.g. it would require close alignment between the nanopore and the electrode junction with nanometre precision. However, this has recently been achieved by Ivanov et al. [32], who were the first to demonstrate concurrent detection of dsDNA by ion and tunnelling current.

Nevertheless, although many elements of a sequencing-by-tunnelling technology have been implemented, significant challenges still lay ahead. These include controlling the speed of the translocation process and the interaction of the biopolymer with the electrode junction, developing sufficiently fast and sensitive electronics to resolve each base, and improving the statistical significance of the detection events, thus reducing the error rate. Resolving these is not trivial, but the prospects are very attractive, in that the approach is label-free and the same device platform could also be used to detect other biologically relevant molecules, such as RNA or protein fragments.

Nanopore sensing with ultra-thin membranes: graphene

Graphene is a semi-metallic material with atomic scale thickness, just 0.6 nm. With excellent mechanical properties, high electrical conductivity in the graphene plane, but surprisingly insulating to ion transport across, graphene appears to be an attractive material for nanopore-based sensing of biomolecules and potentially even label-free DNA sequencing [33–35]. DNA translocation through single- or multi-layer graphene nanopores has been reported recently and independently by several research groups [33,35,36]. The graphene sheet is mounted across a larger, typically micrometre-sized, opening in an Si_3N_4 template; the pore is then drilled in the conventional way using a focused electron (or ion) beam. Given the atomic-scale thickness of the graphene membrane of approximately 0.6 nm, which is shorter than two neighbouring nucleotides in a DNA strand, it is conceivable that a graphene-based device may be able to distinguish individual bases by ion current modulation. Alternatively, the in-plane conductivity of the graphene itself may be exploited as a probe, if translocating analytes

modulate the potential distribution at the circumference of the nanopore, say in a narrow strip of graphene [34,37]. However, so far, DNA translocation experiments through graphene pores have failed to provide sufficient resolution for sequencing applications and the high translocation speed makes electronic readout difficult. Note that in the case of graphene, the pore resistance is limited by the so-called access resistance at the pore, not by the pore channel itself (which is extremely short). Merchant et al. [33] point out that the capacitive noise of unmodified graphene membranes is in fact worse than for other solid-state membranes, thus limiting the bandwidth/resolution of the device. However, they demonstrate that depositing TiO₂ on to the graphene layer can reduce this noise significantly [33].

Sensing protein–DNA interactions

Exploiting protein–DNA or protein–protein interactions are a third way of resolving submolecular detail, for example with a view to detecting particular sequences or base modifications along the DNA strand with antibodies or PNA (peptide nucleic acid) labels. It may also shed light on mechanistic details of biological processes (see below). The requirements on the spatial resolution of the nanopore sensor are less stringent and perhaps an order of magnitude lower than in the case of DNA sequencing. Hence conventional nanopore sensors may be used, with two strategies being most common. The first technique is based on the modification of the pore walls themselves, e.g. with a ‘bait’ protein. During translocation, DNA interacts with the bait protein, modulating the current signal in a characteristic way. The second relies on modifying the target DNA already in solution; the protein–DNA complex forms before translocation, but is then transported as a whole across the pore [38–40].

Timp and co-workers reported for the first time on the capability of synthetic nanopores for sensing protein–DNA complexes [41,42]. They detected a restriction enzyme bound to a specific sequence of dsDNA using 2.5 nm Si₃N₄ nanopores and observed different voltage thresholds compared with mutated dsDNA [41,42]. Dekker and co-workers later examined the translocation of coated dsDNA with a DNA-repair protein (RecA). They showed local detection of protein along DNA with a spatial resolution of 8 nm [39] (Figure 2c). In 2010, Meller and co-workers demonstrated the synchronous optical and electrical detection of protein–DNA complexes through a 4 nm synthetic solid-state nanopore [43]. This method involves a customized TIRF (total internal reflection fluorescence) and labelled avidin protein bound to biotinylated dsDNA with an ss overhang.

The same group also demonstrated the detection of specific DNA sequences by hybridizing dsDNA with PNA through sub-5 nm Si₃N₄ pores. This technique differs from above as the sequence detection is only performed by changes in blockade ionic current upon translocation of individual DNA and PNA–DNA molecules [44].

Han et al. [38] used 20–30 nm Si₃N₄ nanopores in an attempt to detect an interaction of IgG and β -hCG (β -human chorionic gonadotropin) proteins. Failing to observe distinct translocation events directly, they suggested that the complex is too fast to be detected (KCl solution, pH 6.0) owing to the high electrophoretic mobility of the complex under these conditions [38]. However, they could still probe the interaction of IgG and β -hCG by performing a dose–response experiment and monitoring the translocation frequency of IgG: by adding β -hCG to a mixture of IgG and ovalbumin, they observed a decrease in translocation frequency of IgG as a function of β -hCG concentration, indicating that IgG indeed binds to β -hCG under the conditions used.

One of the challenges of nanopore-based protein sensing is clogging of protein inside the pore and absorption of protein on the nanopore wall. Mayer and colleagues addressed this problem by introducing the ‘fluid-wall technology’, originally inspired by olfactory processes in insects [45]. In their work, solid-state nanopores were coated with a fluid lipid bilayer which remains mobile on the solid substrate. First of all, this allows modification of surface chemistry and tuning of pore diameter by changing the bilayer headgroups and length. Secondly, it permits incorporation of specific molecular receptors into the bilayer, providing specificity with regard to species in solution. Dilute protein samples can thus be pre-concentrated on the surface and translocated while surface-bound [45].

Conclusions

Nanopore sensing has seen considerable progress in detection of single molecules in recent years. The present review outlines some important developments and applications of solid-state nanopore sensing with particular focus on detecting submolecular features. Many applications are still at an early stage, but have significant potential for biochemical assays, DNA sequence analysis, epigenetics, gene profiling and molecular barcoding. The relative ease of integrating solid-state nanopore sensors into lab-on-a-chip platforms and straightforward wafer-scale fabrication of these devices adds prospects of their use as a sensing technology outside research laboratory in the future.

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References

- 1 Neher, E. and Sakmann, B. (1976) Single-channel currents recorded from membrane of denervated frog muscle fibres. *Nature* **260**, 799–802
- 2 Bezrukov, S.M. and Vodyanoy, I. (1993) Probing alamethicin channels with water-soluble polymers: effects on conductance of channel states. *Biophys. J.* **64**, 16–25

- 3 Deamer, D.W. and Akeson, M. (2000) Nanopores and nucleic acids: prospects for ultrarapid sequencing. *Trends Biotechnol.* **18**, 147–151
- 4 Kasianowicz, J.J., Brandin, E., Branton, D. and Deamer, D.W. (1996) Characterization of individual polynucleotide molecules using a membrane channel. *Proc. Natl. Acad. Sci. U.S.A.* **93**, 13770–13773
- 5 Li, J., Gershow, M., Stein, D., Brandin, E. and Golovchenko, J.A. (2003) DNA molecules and configurations in a solid-state nanopore microscope. *Nat. Mater.* **2**, 611–615
- 6 Schoch, R.B., Han, J. and Renaud, P. (2008) Transport phenomena in nanofluidics. *Rev. Mod. Phys.* **80**, 839–883
- 7 Smeets, R.M.M., Keyser, U.F., Krapf, D., Wu, M.Y., Dekker, N.H. and Dekker, C. (2006) Salt dependence of ion transport and DNA translocation through solid-state nanopores. *Nano Lett.* **6**, 89–95
- 8 Li, J., Stein, D., McMullan, C., Branton, D., Aziz, M.J. and Golovchenko, J.A. (2001) Ion-beam sculpting at nanometre length scales. *Nature* **412**, 166–169
- 9 Storm, A.J., Chen, J.H., Zandbergen, H.W. and Dekker, C. (2005) Translocation of double-strand DNA through a silicon oxide nanopore. *Phys. Rev. E: Stat., Nonlinear, Soft Matter Phys.* **71**, 051903
- 10 Fologea, D., Brandin, E., Uplinger, J., Branton, D. and Li, J. (2007) DNA conformation and base number simultaneously determined in a nanopore. *Electrophoresis* **28**, 3186–3192
- 11 Akeson, M., Branton, D., Kasianowicz, J.J., Brandin, E. and Deamer, D.W. (1999) Microsecond time-scale discrimination among polycytidylic acid, polyadenylic acid, and polyuridylic acid as homopolymers or as segments within single RNA molecules. *Biophys. J.* **77**, 3227–3233
- 12 Meller, A., Nivon, L., Brandin, E., Golovchenko, J. and Branton, D. (2000) Rapid nanopore discrimination between single polynucleotide molecules. *Proc. Natl. Acad. Sci. U.S.A.* **97**, 1079–1084
- 13 Kowalczyk, S.W., Tuijtel, M.W., Donkers, S.P. and Dekker, C. (2010) Unraveling single-stranded DNA in a solid-state nanopore. *Nano Lett.* **10**, 1414–1420
- 14 van den Hout, M., Skinner, G.M., Klijnhout, S., Krudde, V. and Dekker, N.H. (2011) The passage of homopolymeric RNA through small solid-state nanopores. *Small* **7**, 2217–2224
- 15 Zhao, Q., Comer, J., Dimitrov, V., Yemenicioglu, S., Aksimentiev, A. and Timp, G. (2008) Stretching and unzipping nucleic acid hairpins using a synthetic nanopore. *Nucleic Acids Res.* **36**, 1532–1541
- 16 McNally, B., Singer, A., Yu, Z.L., Sun, Y.J., Weng, Z.P. and Meller, A. (2010) Optical recognition of converted DNA nucleotides for single-molecule DNA sequencing using nanopore arrays. *Nano Lett.* **10**, 2237–2244
- 17 Fologea, D., Ledden, B., McNabb, D.S. and Li, J. (2007) Electrical characterization of protein molecules by a solid-state nanopore. *Appl. Phys. Lett.* **91**, 053901
- 18 Talaga, D.S. and Li, J. (2009) Single-molecule protein unfolding in solid state nanopores. *J. Am. Chem. Soc.* **131**, 9287–9297
- 19 Oukhaled, A., Cressiot, B., Bacri, L., Pastoriza-Gallego, M., Betton, J.-M., Bourhis, E., Jede, R., Gierak, J., Auvray, L. and Pelta, J. (2011) Dynamics of completely unfolded and native proteins through solid-state nanopores as a function of electric driving force. *ACS Nano* **5**, 3628–3638
- 20 Freedman, K.J., Juürgens, M., Prabhu, A., Ahn, C.W., Jemth, P., Edel, J.B. and Kim, M.J. (2011) Chemical, thermal, and electric field induced unfolding of single protein molecules studied using nanopores. *Anal. Chem.* **83**, 5137–5144
- 21 Ayub, M., Ivanov, A., Hong, J., Kuhn, P., Instuli, E., Edel, J.B. and Albrecht, T. (2010) Precise electrochemical fabrication of sub-20 nm solid-state nanopores for single-molecule biosensing. *J. Phys.: Condensed Matter* **22**, 454128
- 22 Ayub, M., Ivanov, A., Instuli, E., Cecchini, M., Chansin, G., McGilvery, C., Hong, J.G., Baldwin, G., McComb, D., Edel, J.B. and Albrecht, T. (2010) Nanopore/electrode structures for single-molecule biosensing. *Electrochim. Acta* **55**, 8237–8243
- 23 Wei, R., Gatterdam, V., Wieneke, R., Tampé, R. and Rant, U. (2012) Stochastic sensing of proteins with receptor-modified solid-state nanopores. *Nat. Nanotechnol.* **7**, 257–263
- 24 Howorka, S. and Siwy, Z. (2009) Nanopore analytics: sensing of single molecules. *Chem. Soc. Rev.* **38**, 2360–2384
- 25 Zvolak, M. and Di Ventra, M. (2005) Electronic signature of DNA nucleotides via transverse transport. *Nano Lett.* **5**, 421–424
- 26 Tsutsui, M., Taniguchi, M., Yokota, K. and Kawai, T. (2010) Identifying single nucleotides by tunnelling current. *Nat. Nanotechnol.* **5**, 286–290
- 27 Tsutsui, M., Rahong, S., Iizumi, Y., Okazaki, T., Taniguchi, M. and Kawai, T. (2011) Single-molecule sensing electrode embedded in-plane nanopore. *Sci. Rep.* **1**, 46
- 28 Huang, S., He, J., Chang, S., Zhang, P., Liang, F., Li, S., Tuchband, M., Fuhrmann, A., Ros, R. and Lindsay, S. (2010) Identifying single bases in a DNA oligomer with electron tunnelling. *Nat. Nanotechnol.* **5**, 868–873
- 29 Chang, S., Huang, S., He, J., Liang, F. and Zhang, P. (2010) Electronic signatures of all four DNA nucleosides in a tunneling gap. *Nano Lett.* **10**, 1070–1075
- 30 He, J., Lin, L., Zhang, P. and Lindsay, S. (2007) Identification of DNA basepairing via tunnel-current decay. *Nano Lett.* **7**, 3854–3858
- 31 Nojima, Y., Tanaka, H., Yoshida, Y. and Kawai, T. (2004) High-resolution scanning tunneling microscopy and spectroscopy studies of deoxyribonucleic acid and fluorescein isothiocyanate. *Jpn. J. Appl. Phys., Part 1* **43**, 5526–5527
- 32 Ivanov, A.P., Instuli, E., McGilvery, C.M., Baldwin, G., McComb, D.W., Albrecht, T. and Edel, J.B. (2011) DNA tunneling detector embedded in a nanopore. *Nano Lett.* **11**, 279–285
- 33 Merchant, C.A., Healy, K., Wanunu, M., Ray, V., Peterman, N., Bartel, J., Fischbein, M.D., Venta, K., Luo, Z.T., Johnson, A.T.C. and Drndić, M. (2010) DNA translocation through graphene nanopores. *Nano Lett.* **10**, 2915–2921
- 34 Postma, H.W.C. (2010) Rapid sequencing of individual DNA molecules in graphene nanogaps. *Nano Lett.* **10**, 420–425
- 35 Garaj, S., Hubbard, W., Reina, A., Kong, J., Branton, D. and Golovchenko, J.A. (2010) Graphene as a subnanometre trans-electrode membrane. *Nature* **467**, 190–193
- 36 Schneider, G.F., Kowalczyk, S.W., Calado, V.E., Pandraud, G., Zandbergen, H.W., Vandersypen, L.M.K. and Dekker, C. (2010) DNA translocation through graphene nanopores. *Nano Lett.* **10**, 3163–3167
- 37 Saha, K.K., Drndić, M. and Nikolić, B.K. (2012) DNA base-specific modulation of microampere transverse edge currents through a metallic graphene nanoribbon with a nanopore. *Nano Lett.* **12**, 50–55
- 38 Han, A., Creus, M., Schuurmann, G., Linder, V., Ward, T.R., de Rooij, N.F. and Stauffer, U. (2008) Label-free detection of single protein molecules and protein-protein interactions using synthetic nanopores. *Anal. Chem.* **80**, 4651–4658
- 39 Kowalczyk, S.W., Hall, A.R. and Dekker, C. (2009) Detection of local protein structures along DNA using solid-state nanopores. *Nano Lett.* **10**, 324–328
- 40 Hall, A.R., van Dorp, S., Lemay, S.G. and Dekker, C. (2009) Electrophoretic force on a protein-coated DNA molecule in a solid-state nanopore. *Nano Lett.* **9**, 4441–4445
- 41 Zhao, Q., Sigalov, G., Dimitrov, V., Dorvel, B., Mirsaidov, U., Sligar, S., Aksimentiev, A. and Timp, G. (2007) Detecting SNPs using a synthetic nanopore. *Nano Lett.* **7**, 1680–1685
- 42 Dorvel, B., Sigalov, G., Zhao, Q., Comer, J., Dimitrov, V., Mirsaidov, U., Aksimentiev, A. and Timp, G. (2009) Analyzing the forces binding a restriction endonuclease to DNA using a synthetic nanopore. *Nucleic Acids Res.* **37**, 4170–4179
- 43 Soni, G.V., Singer, A., Yu, Z.L., Sun, Y.J., McNally, B. and Meller, A. (2010) Synchronous optical and electrical detection of biomolecules traversing through solid-state nanopores. *Rev. Sci. Instrum.* **81**, 014301
- 44 Singer, A., Wanunu, M., Morrison, W., Kuhn, H., Frank-Kamenetskii, M. and Meller, A. (2010) Nanopore based sequence specific detection of duplex DNA for genomic profiling. *Nano Lett.* **10**, 738–742
- 45 Yusko, E.C., Johnson, J.M., Majd, S., Prangio, P., Rollings, R.C., Li, J., Yang, J. and Mayer, M. (2011) Controlling protein translocation through nanopores with bio-inspired fluid walls. *Nat. Nanotechnol.* **6**, 253–260

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