Supplementary information for

Tracking single-particle dynamics via combined optical and electrical sensing

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The Supplementary Information includes:

1. Analytical derivation of ionic current through microchannels

2. Supplementary Figures (Fig. S1-S11)
Figure S1. Scanning electron micrographs of a bow-tie shaped microchannel.  

a, False colored image of the fluidic channel fabricated on a SiO$_2$-coated cover glass substrate. Yellow-colored region denote Pt/Au microelectrodes used as external markers for EB-lithography drawing.  
b, Magnified image of the fluidic channel.  $w_4$ = 25 μm and $w_3$ = 5 μm are the width of the channel and the spacing between squared micropillars, respectively.  
c, Close view of the bow-tie shaped microchannel. $w_1$ = 1.2 μm and $w_2$ = 2.6 μm are, respectively, the widths of the narrowest constriction and the orifice of microchannel, while $L$ = 4.0 μm denote the channel length. The channel height is uniformly 1.2 μm.
Figure S2. Fabrication procedure of the microchannel structure. a, (i) A 1.5 μm thick SiO₂ layer is grown on a cover glass substrate. (ii) After that, a micro-electrode pattern is delineated on the substrate using a standard photo-lithography method, which is followed by (iii) deposition of Pt/Au (2nm/30nm) layer by a radio-frequency (RF) magnetron sputtering process. (iv) The substrate is then immersed in N,N-dimethylformamide for half a day and sonicated for lift-off. (v) After fabrication of microelectrodes, a 100 nm thick Cr layer is deposited using RF sputtering technique. (vi) Subsequently, a microchannel is rendered using a standard electron-beam (EB) lithography method. (vii) The exposed Cr layer was then removed by a wet etching process. (viii) The microchannel is sculpted in the SiO₂ using a reactive ion etching (RIE) method. (ix) The residual Cr is removed by a wet etching process. b, Layout of the sample setup. PDMS block with a fluidic channel formed on one side of the surface was used for sealing of the microchannel. Six holes are punched in the PDMS. Two pairs of holes served as inlet and outlet for a liquid flow into the microchannel and the other two are used to insert Ag/AgCl electrodes for the ionic current measurements.
Figure S3. Liquid flow through fluidic channels. a, Microchannel device fabricated on a SiO₂-coated cover glass. Four holes in the PDMS block are utilized for injecting a particle solution into the microchannel. b, SEM image of the channel formed on one side of the PDMS block. c, Top view of the microchannel sealed with the PDMS block. Liquid is introduced via the four conduit prepared on the PDMS surface. The distance between the pair of channels $L_{\text{ext}}$ is 175 μm as indicated in the figure.
Figure S4. Structure of a straight type microchannel.  a, SEM micrograph showing a fluidic channel consisting of two conduits with embedded micropillars connected via a straight microchannel.  b, Magnified image of (a).  $L = 4.0 \, \mu m$ and $w = 1.4 \, \mu m$ are the length and the width of the channel, respectively.
Figure S5. Optical identifications of ionic current spikes.  **a-c,** Temporal changes of the ionic current through a straight-type microchannel with structure shown in Fig. S4. Resistive pulses of height about 1 nA marked as S are attributed to single-particle translocation through the channel as confirmed by fluorescence observations. On the other hand, we observe no change in the microchannel by the optical imaging at the moment when the signals with asterisks are detected. These anomalous current spikes are perhaps mere external noise or charging/discharging phenomena occurring at the channel (Fig. S1). Whereas the extraordinarily large pulses (a-b) can be readily judged as atypical events and be excluded from the data for single-particle analyses in the conventional Coulter counter method, the right two unidentified signals of size 1 nA in (c) cannot be
discriminated from the resistive pulses of particle translocation origins without fluorescent imaging.
Figure S6. Fluorescent imaging of single-particle translocation through a straight-type microchannel. **a-c,** Consecutive fluorescence images used to assess the
flow speed of polystyrene single-particles for three different pathways: along the wall of the micropillar $pw$ (a), in the middle of the channel $mc$ (b), and in vicinity of the side surface of the channel $cw$ (c). Particles are voltage-driven to move from the left to the right side. Digits denote the frame number. As evident in the images, particles are passing through the microchannel much faster than the exposure time of 5 ms.
Figure S7. Velocity of 1 μm-sized polystyrene single-particles $v_{\text{Pst}}$ flowing through a fluidic channel shown in Fig. 3 deduced from fluorescence images.  

a-i, $v_{\text{Pst}}$ vs $t$ plots for $pw$ (a-c), $mc$ (d-f), and $cw$ events (g-i) that corresponds to a particle moving in the middle ($mc$) or along the side walls of a channel ($cw$) or a pillar ($pw$). The average of these data is used in Fig. 4c.
Figure S8. Electric-field-driven acceleration of 1 μm-sized polystyrene single-particles $a_{Pst}$ flowing through a fluidic channel shown in Fig. 3 obtained from the data in Fig. S5. a-i, $v_{Pst}$-t plots for $pw$ (a-c), $mc$ (d-f), and $cw$ events (g-i) that corresponds to a particle moving in the middle ($mc$) or along the channel ($cw$) or pillar walls ($pw$). The average of these data is shown in Fig. 4d.
Figure S9. Resistive pulse suggestive of two-particle-cluster translocations.  

(a) Resistive pulse with anomalously large blockage current of 1.6 nA. Consecutive fluorescence images taken simultaneously with the ionic current trace are also shown.  

(b) Current spike and fluorescence images signifying single-particle translocation are displayed for comparison. The fluorescence micrographs clearly show higher intensity of the particle in the microchannel shown in (a) than that in (b). If the blockage current $I_p$ of 1.6 nA is attributed to a single-particle translocation, the diameter of the particle $d_{Pst}$ is estimated to be $d_{Pst} = 1.2 \, \mu m$ by back calculation, which is out of the nominal size distribution of TransFluoSpheres® beads: $d_{Pst} = 1.10 \, \mu m \pm 55 \, nm$. Nonetheless, this would yield 19 % increase in the number of fluorophores on the particle surface. On the other hand, when a two-particle-cluster of 1.06 μm beads passes through the channel, it is analytically anticipated to cause a temporal blockage of the ionic current by 1.7 nA, which is close to that in (a). For the cluster, the number of fluorophores increases by 200%. Therefore, as both cases would lead to an enhanced fluorescence intensity in the images, the large resistive pulse in (a) can be assigned to either translocation of a large particle or a dimer. However, considering that blockage current larger than 1.4 nA is seldom observed in the measurements (2 out of 233 events as shown in Fig. 4h), and that the two data with large $I_p$ is very close to each other, it is more probable that the event in (a) corresponds to
translocation of a dimer particle than imagining fortuitous observations of two passing-by events of particles of a peculiar size that exceeds the nominal size distribution.
Figure S10. In-channel velocity profiles of a polystyrene particle along a straight-type microchannel. a-b, Analytically deduced ionic current $I$ through a straight-type microchannel of structure shown in Fig. S4 with a 1.1 μm particle entering (a) and exiting (b) it. The position of center of the particle $x$ is taken along the channel. $x$ is set to zero when the edge of the particle reaches the channel entrance.

c-e, Resistive pulses for particles moving along a side surface of the micropillar $pw$ (c), in the middle of the channel $mc$ (b), and on the channel wall $cw$ (e).

f-k, The velocity of the particles $v_{\text{p}}$ entering (I) or exiting (II) the channel for $pw$ (f,i), $mc$ (g,j), and $cw$ (h,k) estimated using the theoretical $I - x$ in (a-b).
Figure S11. Current measurements through a standard 100 MΩ resistor at different sampling rates.  

- **a-b**, Current through the resistor I sampled under a DC voltage of 6 V at 1 MHz (a) and 100 kHz (b).  

- **c-d**, $I-t$ curves obtained under a square wave voltage of $V_{p-p} = 0.5$ V amplitude at 1 MHz (c) and 100 kHz (d). While noise increases more than a factor of two, there is no difference in the mean current levels digitized at 1 MHz and 100 kHz.
1. Analytical derivation of ionic current through microchannels

Ionic current blockage during a polymeric bead translocation through a microchannel is estimated by analytical calculations as described below. The resistance in the bow-tie shaped microchannel $R_{micro}$ is described as

\[
R_{micro} = \int_0^{L/2} \rho \frac{dl}{S_{micro}}
\]

\[
S_{micro} = \left\{ w_1 + \frac{2(w_2 - w_1)l}{L} \right\} h
\]

, where $\rho = 10 \, \Omega m$ is the resistivity of TE buffer, $S$ is the cross-sectional area of the channel, $w_1 = 1.4 \, \mu m$ and $w_2 = 2.6 \, \mu m$ are the widths while $L = 4 \, \mu m$ and $h = 1.2 \, \mu m$ are the length and the height of the bow-tie channel, and $l$ is a coordinate along the channel length (see Fig. S1 (c) for the configuration of the bow-tie type channel). From the above expression, we obtain

\[
R_{micro} = 2 \int_0^{L/2} \rho \frac{dl}{a + bl} , \quad a = hw_1 , \quad b = \frac{2h(w_2 - w_1)}{L}
\]

, which leads to

\[
R_{micro} = 2 \int_0^{L/2} \frac{\rho}{b} \log(a + bl) dl
\]

As a result, we obtain $R_{micro} = 17.2 \, M\Omega$. In addition to $R_{micro}$, the conduit extends for about $L_1 = 175 \, \mu m$ from the microchannel (Fig. S3 (c)) at both sides largely contributes to the measured ionic current. This resistance $R_{ext}$ can be roughly written as $R_{ext} = \rho L_1/3w_3h + \rho L_1/w_4h = 126 \, M\Omega$, where $w_3 = 5 \, \mu m$ and $w_4 = 25 \, \mu m$ are the spacing between the micropillars and the width of the large channel (see Fig. S1 (a,b)). The open channel current $I_{open}$ is then calculated as $I_{open} = V_0/(R_{micro} + R_{ext}) = 69.6 \, nA$, which fairly agrees with the measured value, $I_{open} = 69 \, nA$.

Similarly, the resistance in the bow-tie shaped fluidic channel with a 1 \, \mu m Pst particle located at its narrowest constriction, $R_{Pst}$, is described as

\[
R_{Pst} = 2 \int_0^{L/2} \rho \frac{dl}{S_{micro} - S_{Pst}} + 2 \int_r^{L/2} \rho \frac{dl}{S_{micro}}
\]

\[
S_{micro} = \left\{ w_1 + \frac{2(w_2 - w_1)l}{L} \right\} h
\]

\[
S_{Pst} = \pi(r^2 - l^2)
\]

, where $S_{Pst}$ is the cross-sectional area of the Pst particle of diameter $2r = 1.1 \, \mu m$. The above equation is calculated as

\[
R_{Pst} = 2 \int_0^{L/2} \rho \frac{dl}{\pi (r^2 - r^2 + d)} + 2 \int_r^{L/2} \frac{\rho}{b} \log(a + bl) dl , \quad c = \frac{b}{a} , \quad d = \frac{a}{\pi} - r^2
\]
\[
= \frac{2\rho}{\pi} \int_{0}^{r} \frac{dt}{(t^2 + \frac{b^2}{4})^{3/2}} + 2\rho \left[ \frac{1}{b} \log(a + bl) \right]_{r}
\]
\[
= \frac{2\rho}{\pi} \left[ \frac{1}{\sqrt{d - c^2}} \tan^{-1} \frac{t + \frac{c}{2}}{\sqrt{d - c^2}} \right]_{0}^{r} + 2\rho \left[ \frac{1}{b} \log(a + bl) \right]_{r}
\]

using
\[
\int \frac{dx}{x^2 + a^2} = \frac{1}{a} \tan^{-1} \frac{x}{a} + C
\]

From this, we obtain \( R_{\text{Pst}} = 21.1 \text{ M\Omega} \). The ionic current flowing through the channel with a trapped particle \( I_{\text{block}} \) is then estimated as \( I_{\text{block}} = V_b / (R_{\text{Pst}} + R_{\text{ext}}) = 67.8 \text{ nA} \). Consequently, the current blockage \( I_p \) is deduced as \( I_p = I_{\text{open}} - I_{\text{block}} = 1.83 \text{ nA} \), which is in accordance with \( I_p = 1.9 \text{ nA} \) of the resistive pulses displayed in Fig. 3 (a,b).

Blockade ionic current for 1.1 \( \mu \text{m} \) diameter Pst particles passing through a straight-type microchannel can also be estimated in a similar way. The resistance of a microchannel with a Pst bead residing at the center is written as,

\[
R_{\text{Pst}} = 2\rho \int_{0}^{r} \frac{dt}{hw - \pi r^2} + 2\rho \int_{0}^{r} \frac{dt}{hw \pi r^2 - t^2}
\]
\[
S_{\text{Pst}} = \pi (r^2 - t^2)
\]

, where \( h = 1.4 \text{ \mu m} \), \( L = 4\mu m \), and \( w = 1.4 \text{ \mu m} \) are the height, the length, and the width of the microchannel (see Fig. S4 for the channel structure). The above equation can be mathematically solved as:

\[
R_{\text{Pst}} = 2\rho \int_{0}^{r} \frac{dt}{hw - \pi (r^2 - t^2)} + 2\rho \int_{0}^{r} \frac{dt}{hw \pi r^2 - t^2}
\]
\[
= \frac{2\rho}{\pi} \int_{0}^{r} \frac{dt}{(t^2 + g^2)^{3/2}}
\]
\[
= \frac{2\rho}{\pi} \left[ \frac{1}{g} \tan^{-1} \frac{t}{g} \right]_{0}^{r} + 2\rho \left[ \frac{1}{hw \pi r^2} \right]_{0}^{r}
\]

This yields \( I_p = 1.01 \text{ nA} \) at \( V_b = 5 \text{ V} \) with \( R_{\text{ext}} = \rho L_1/3w_s h + \rho L_1/w_d h = 126 \text{ M\Omega} \) where \( L_1 = 175 \text{ \mu m} \) is the distance between the PDMS channels (see Fig. S3 (c)), which is close to the experimental value of \( I_p = 0.98 \text{ nA} \) (Fig. 3). The above expression is also used to simulate the change in the ionic current through the straight-type microchannel during the entering and the exiting processes as shown in Fig. S8.