Scheme S1 The process of making the microfluidic chip.
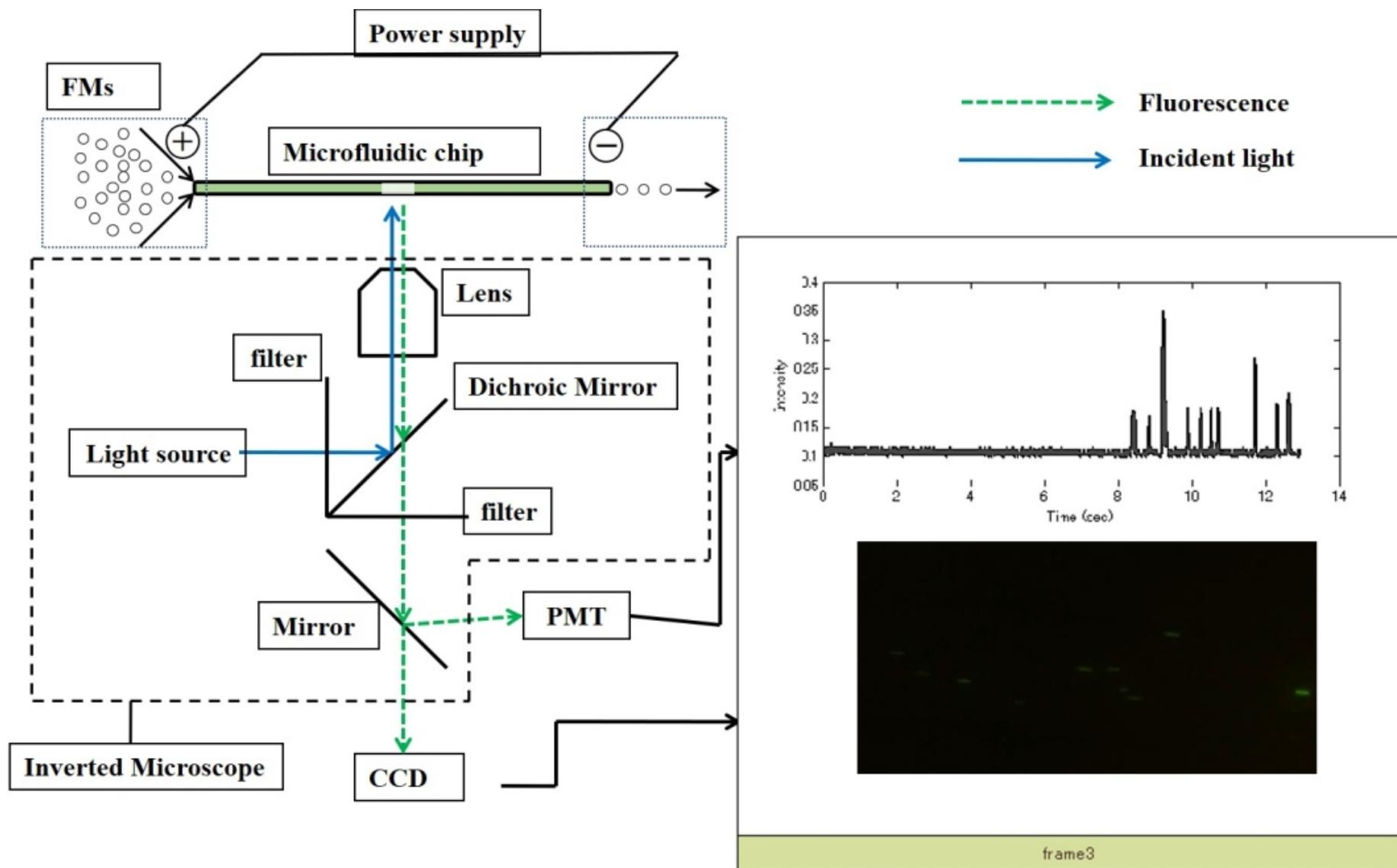
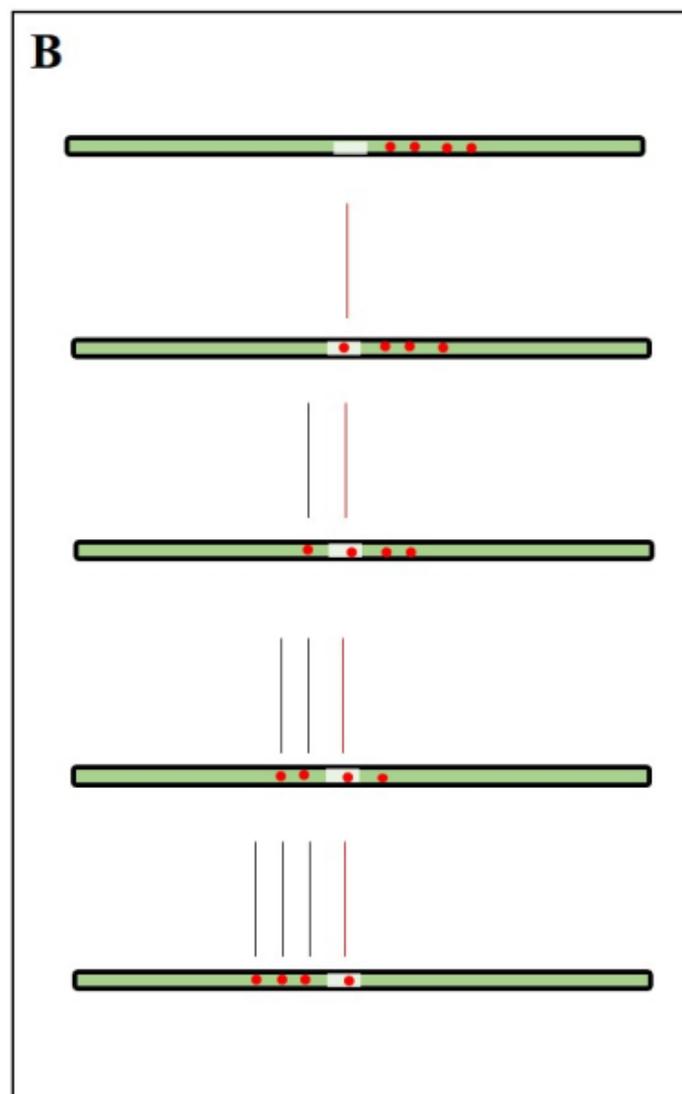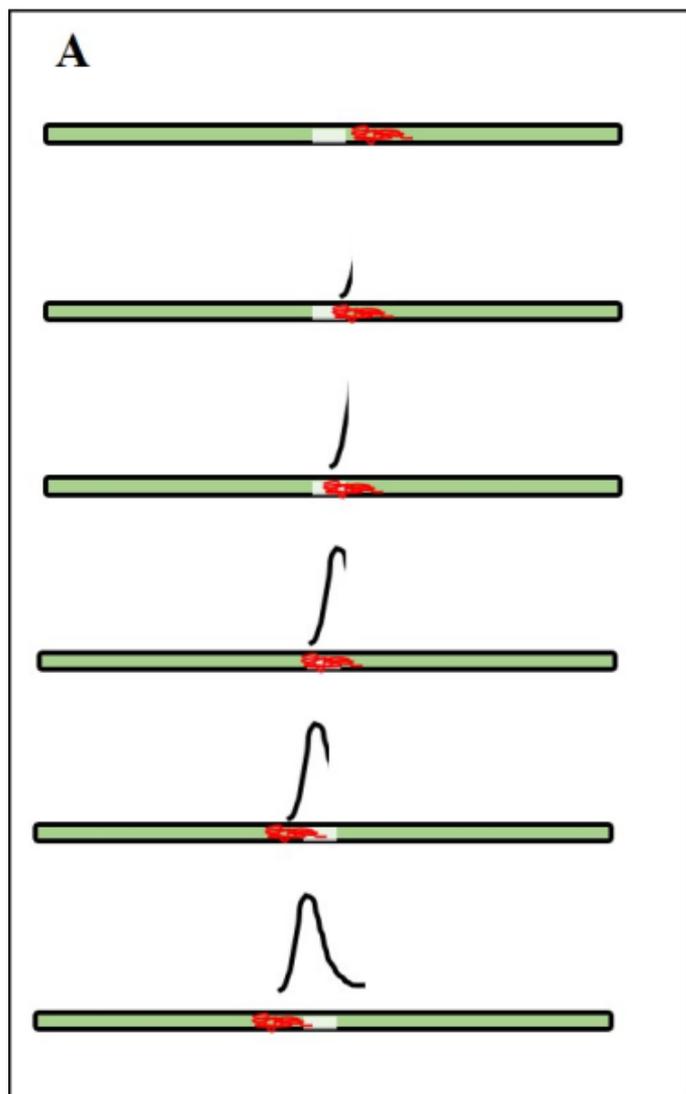**Fig.S1** The working principle of the CE system

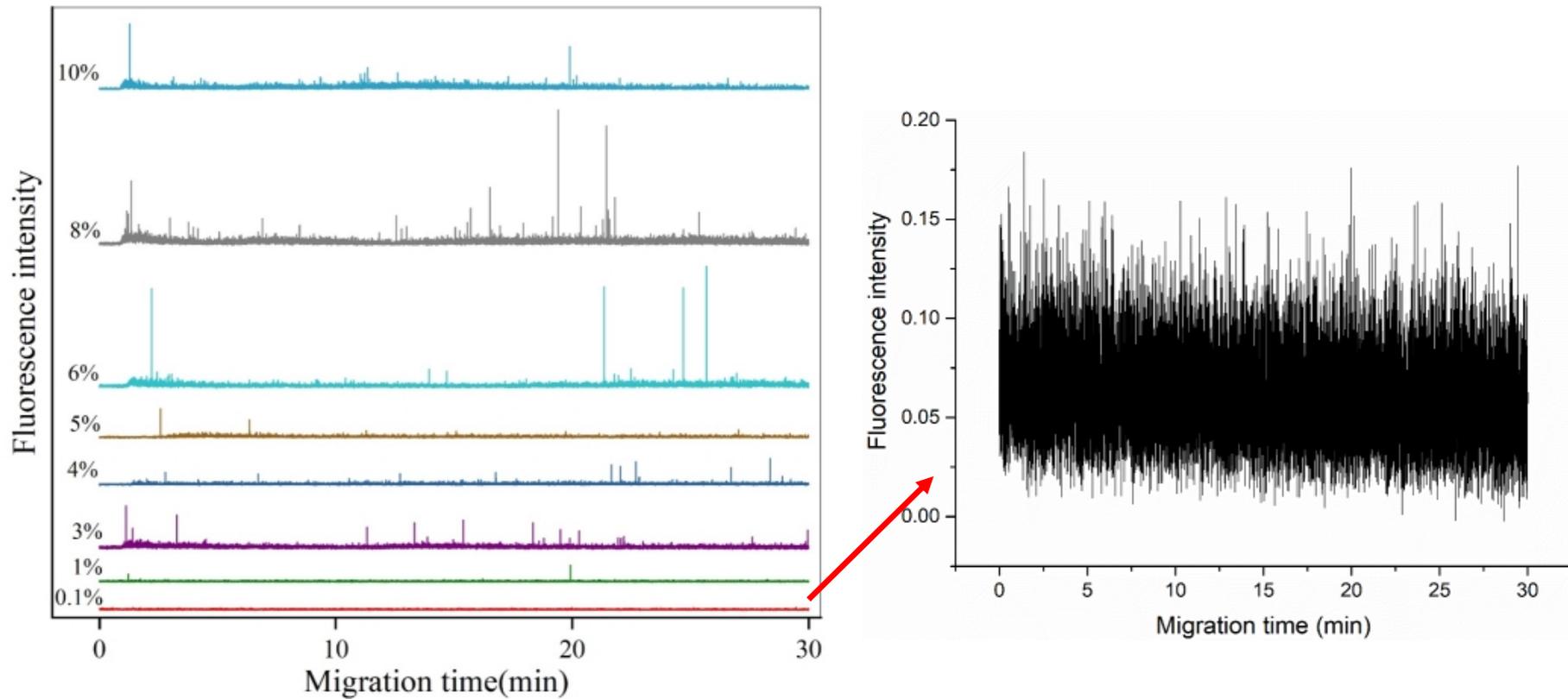**Fig.S2** The peak of the FMs in electropherogram: (A) FMs aggregate (B) dispersed FMs.

Fig.S3 The electropherogram of fluorescent microspheres with concentration from 0.1% to 10%.
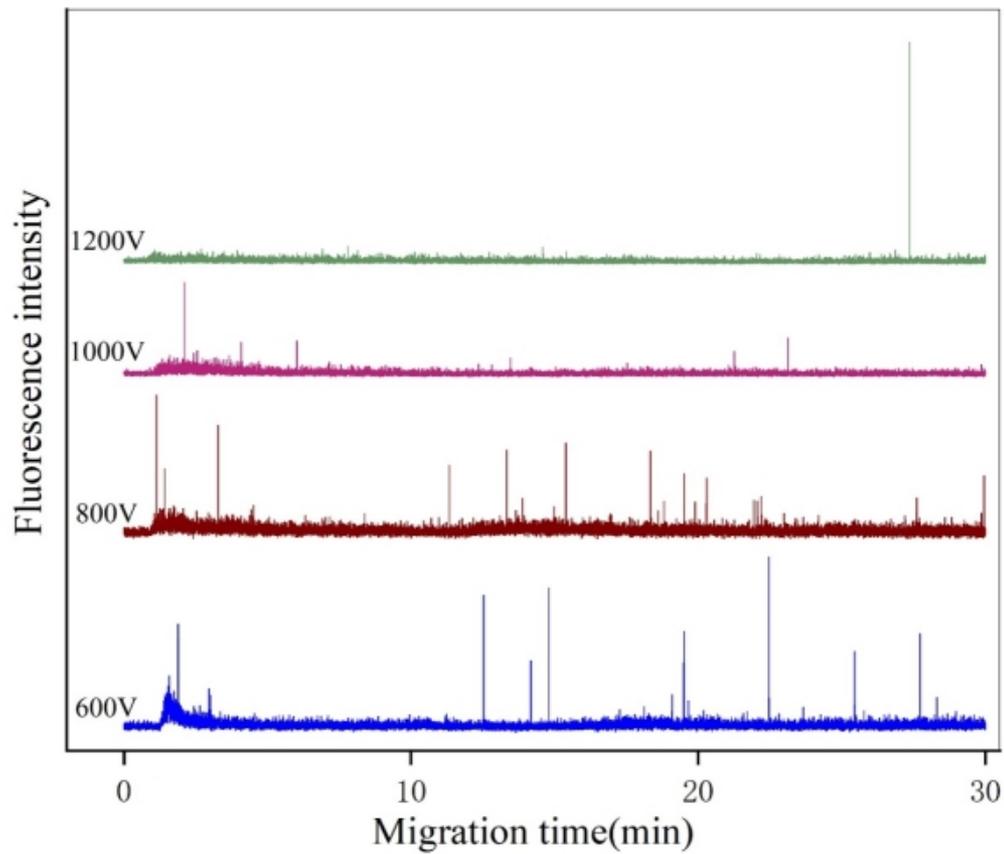
Fig.S4 The electropherogram of fluorescent microspheres migrating in the microchannel at voltages from 600 V to 1200 V.
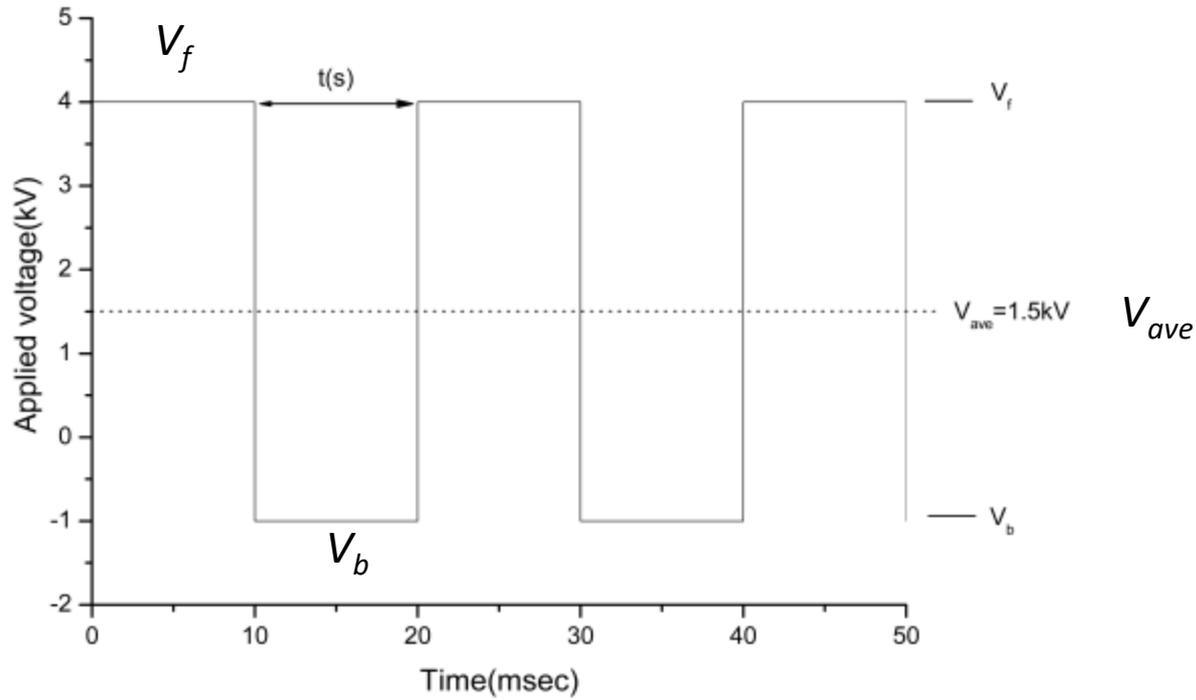
Fig.S5 The definition of pulsed field electric field.

$$V_{ave} = \frac{V_f + V_b}{2}$$

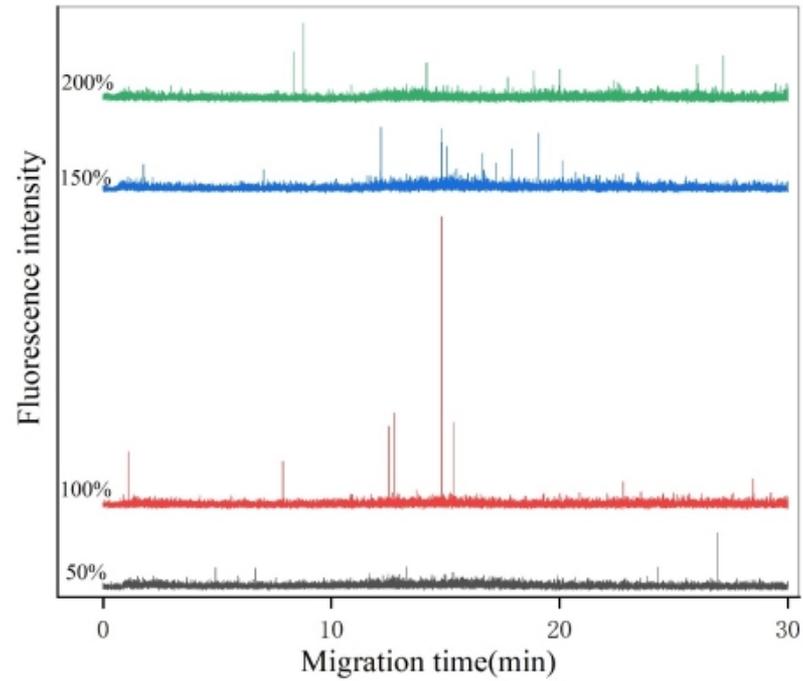$$Modulation : M = \frac{V_f - V_{ave}}{V_{ave}} \times 100(\%)$$

Fig.S6 The electropherogram of fluorescent microspheres migrating in the microchannel at pulsed electric field strength with modulation depth from 50% to 200%.