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Nanostructured devices have been able to foster the technology for cell membrane poration. With the size smaller than the cell, nanostructure allows efficient poration on the cell membrane. Emerging nanostructures with various physical transduction have been demonstrated to accommodate effective intracellular delivery. Aside from improving poration and intracellular delivery performance, the nanostructured device also allows the discovery of novel physiochemical phenomenon and biological response of the cell. This article provides a brief introduction to the principles of the nanostructured device for cell poration and outlines the intracellular delivery capability of the technology. In the future, we envision more

thus, are not homogeneous and may not allow the specific substance to be delivered [10].

Aside from chemical agent and electrical field, many other physical methods have been demonstrated to break cell membrane efficiently. [11]. Ultrasound acoustic cavitation shock wave has been able to accommodate DNA transfection into mammalian cells [12 14]. Moreover, shear stress from the oscillating bubble produced by the acoustic wave is able to compromise the cell membrane integrity. Beside acoustic energy, mechanical force to break the cell membrane can be introduced by the fluid flow. A high-speed jet flow of solution with cargo can penetrate the cell seamlessly in vivo [15]. Likewise, shear stress from micromechanical structure also allows a high transfection rate [16]. Similar to the acoustic shock wave, the laser can generate highly localized energy which wounds the cell membrane [17,18]. Although the throughput is far less than the ultrasonic device, the precision of the laser allows a specific cell to be targeted with high efficiency [19,20].

The emerge of nanotechnology has shed light on new cell membrane poration technology. With the size of up to a few hundred nanometers, nanotechnology has the perfect scale to manipulate the cell and its organelles whose dimensions are in the order of few microns. By using a submicron structure, the cytosol is accessible for interrogation with minimal disruption to the cell. In drug delivery study, nanotechnology gains a huge fame especially due to its role in enhancing the efficacy and specific targeting of the drug into the target site [21]. Additionally, the power of nanotechnology is capable to alleviate specific limitation of former cell poration technology by providing precision and accuracy, reducing the adverse

extraction yield of the nanostructured devices compared to chemical lysis.

For cell lysis, the mode of operation is usually by flowing the cell through the structure. Oftentimes, this type of mode of operation integrates the nanostructure in a microfluidic channel where the cell is flown. However, for the cell poration, the mode of operation needs to ensure it is minimally invasive to the cell and maintain the cell viability. Figure 1 represents the mode of operation for the cell poration using mechanical nanostructure. Devices harnessing mechanical energy for cell poration are usually operated by seeding the cell right on the nanostructured materials or by controlling the nanostructure itself. High aspect ratio nanostructures, such as vertically-aligned nanowires, are capable of direct penetration to the cell membrane as the cell spread over the substrate [30]. Whereas, a single nanostructure like nanopipette and nanoneedle, fabricated by modifying AFM tip, can be spatially manipulated to puncture the cell membrane [31,32].

In the first mode of operation of the cell poration device, i.e. seeding the cell on the substrate depicted in Figure 1(a), the penetration of nanostructure is mainly driven by the properties and the types of the cell itself. Puncturing the plasma membrane of the stiffer cell requires less strain and tension compared to the softer cell. Experimental observation shows that lipid membrane could fail when the tension is 1-10 mN/m and the rupture strain is 1-5% [33 35]. Using mechanical model, Xie and coworkers calculated critical tension of membrane failure of 5.6 mN/m and varying rupture strain: 0.7% for a stiff cell, 2% for a regular cell, and 6% for a soft cell [30]. The critical tension value does not assure membrane penetration; it only indicates a threshold value above which penetration of nanowire will start occurring. The mechanisms of how vertical nanowire breaks the cell membrane are described in two ways: impaling and adhesion.
Tf BDC BT1 0 0 1 45.36 575.83 Tm [(a)]1(n)-5(d)-203(F2 2 nle duker1
The impaling mechanism specifies that the penetration occurs when the rounded cell in
134.9nsi EMC /P <</MCd vs8ta when thEMC /P <</(h)6( )9(p)-5(u)6(n)6(d)-5(ed)2(6)-5(BDC BT1 0 0 1 137.78

size-fits-all technology. The effectiveness of the nanostructure

It is important that future research investigate how living cell interacts with nanostructures as limited studies have been done in this area [107,121,122]. A serendipitous response of the cell when interacting with the structure may or may not be desired for the overall performance for cell poration. Culturing the cell in the nanostructured environment can induce various

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11.	Nanostraw electroporation [53] Diameter = 250 nm	Electrical	CHO HEK293	PI DNA	%D: PI < 95% %D: DNA < 81% %D: 2 DNAs < 74% %V < 95%	Dosage control and co-delivery of multiple genes