

## New and Notable

### Cell Mechanics: Combining Speed with Precision

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The mechanical response of cells is a powerful biophysical marker for cell state. Information on a cell's elasticity can, for instance, be used to distinguish between different cell phenotypes, or between healthy and diseased cells.

Depending on the application, either the accuracy or the speed at which cell properties can be accessed is more important. However, it is still difficult, as of this writing, to measure the mechanical properties of cells both at high speed and with high precision. In this issue of *Biophysical Journal*, Mietke et al. (1) present a combined theoretical and experimental study that might just bring us an important step closer to this goal of combining speed with precision in cell mechanics.

The methods that are available as of this writing for measuring the mechanical response of cells can be roughly divided into static methods, where mechanical properties are measured for one cell at a time, and continuous flow methods, where the properties of many cells are probed in a continuous flow, using microfluidic devices. One of the earliest static methods to be developed was the so-called cell poking device (shown schematically in Fig. 1 a) (2), where a carefully calibrated thin and flexible glass rod is used to directly press on a single cell, enabling the controlled application of forces in the piconewton regime. Atomic force microscopy (3) (Fig. 1 b) has also been used in a

similar fashion, yielding precise force displacement curves associated with the indentation of a cell by the atomic force microscopy tip. To yield information on the full elastic response of a cell, both these direct poking methods would also require imaging a cell's shape during deformation. Another popular static technique is micropipette aspiration (4) (Fig. 1 c), which can often be performed using equipment that is readily available in many biological laboratories. In this technique, a micromanipulator is used to bring a micropipette into contact with a cell surface and suction is applied. The resultant local deformation of the cell is indicative of the cell's mechanical properties; if the cytoskeletal elasticity is negligible, it is a highly accurate probe of the cortical tension of the cell membrane. In a related method, capillary micromechanics (shown in Fig. 1 d) (5), the experimental setup is essentially inverted; here cells are flown inside a tapered glass capillary by applying pressure to the inlet. Because the tip of the capillary is smaller than the cells, a single cell is trapped near the tip, blocking further flow. By analyzing the shape of the deformed cell as a function of the applied pressure, both the bulk (compressive) elastic modulus  $K$  as well as the shear elastic modulus  $G$  of the cell can be extracted. Such information on the response of a cell to different modes of deformation can be valuable and nontrivial, as illustrated by a recent study where monocytic cells were shown to become more deformable (lower  $G$ ), but less compressible (higher  $K$ ) upon activation (6).

Continuous flow methods inherently enable a higher measurement speed, as cells do not have to be loaded or located individually for each measurement, but instead are continuously flown through the channels of a microfluidic device. One strategy is to flow cells into a constriction of width  $W$  smaller than the cell diameter  $D$ , as shown schematically in (Fig. 1 e),

which predefines a minimum level of deformation. Now the velocity of a cell passing through the constriction at fixed pressure difference can be used as a measure for mechanical phenotyping (7). However, as the cell walls are generally in contact with the channel walls, this velocity can be determined to a large degree by the friction and interactions between the cell and the channel surfaces. It is therefore often difficult to extract quantitative mechanical properties from such measurements. These problems can be circumvented by performing measurements in constrictions that are wider than the size of the cells,  $W > D$ , as schematically shown in (Fig. 1 f). In this case, no direct contact between the cells and the surfaces of the channels occur; cell deformation thus occurs solely because of hydrodynamic forces induced by the shear flows in the constriction. While for stiffer types of cells it is often difficult to induce shear rates that result in deformations large enough to be accurately probed, for softer cells such as red blood cells, devices with  $W > D$  have been successfully used for mechanical measurements. Higher levels of cell deformation at still modest flow rates can be achieved in devices that employ an extensional flow, as realized in a cross-slot type device with two inlet and two outlet channels, resulting in a strong extensional flow, as shown schematically in (Fig. 1 g). This leads to significant shear and inertial forces driving cell deformation, but only if the incoming cells are centered well enough in the middle of the incoming channels. This crucial alignment problem has recently been overcome in an elegant setup where inertial focusing is employed to align cells at the middle of the incoming channels (8); the technique is referred to as “deformability cytometry” and enables deformability fingerprinting at rates of up to 2000 cells/s.

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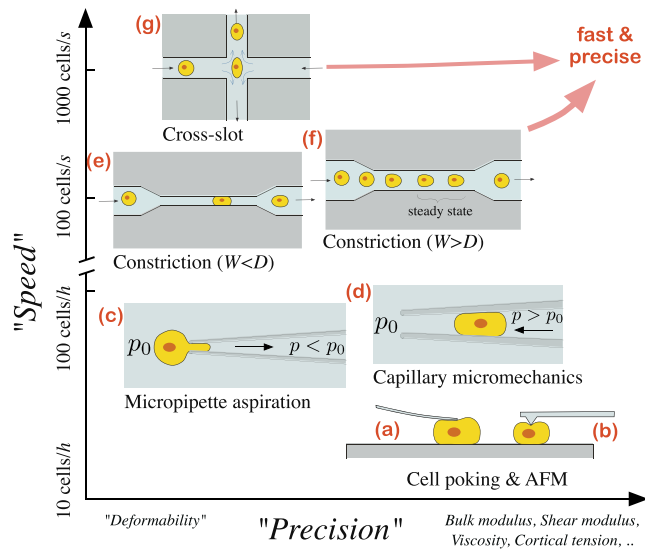


FIGURE 1 Examples of methods for probing cell mechanics. Static methods: (a) cell poking (2), (b) atomic force microscopy (3), (c) micropipette aspiration (4), and (d) capillary micromechanics (5). Continuous flow methods: (e) flow through constriction with  $W > D$  (7), (f) flow through constriction with  $W < D$  (10), and (g) extensional flow in a cross-slot device (8). To see this figure in color, go online.

Thus, as illustrated in Fig. 1, static methods are typically more accurate and can yield precise mechanical information such as the cortical tension of the cell membrane or the bulk and shear elastic moduli of the cell as a whole, while the main advantage of continuous flow methods lies in the speed at which measurements can be performed, which makes them suitable for high-throughput biophysical fingerprinting of large cell populations. However, the degree of quantitative information that can currently be extracted from these continuous flow methods is still limited, as it is not straightforward to separate the effects of cell elasticity, viscosity, membrane tension, surface friction, etc., on the observed cell deformations. Thus, even when deformability can be measured accurately, these measurements do generally not provide precise information on mechanical properties. In order to combine this precision of static methods with the speed of continuous flow methods, advances in the interpretation of the latter are clearly required.

The work of Mietke et al. (1) makes an important contribution to the analysis of cell deformation data, enabling a better link to mechanical properties. The same group of authors has recently developed an improved experimental setup for deformability measurements employing constrictions with  $D < W$  (Fig. 1f), which enables cell deformations to be analyzed in real-time; they refer to this as “real-time deformability cytometry” (9). In this issue, Mietke et al. (1) present a detailed study that combines analytical theory with simulations and experiments to link the observed deformability to mechanical properties. The basis is their derivation of an analytical solution for the flow around a sphere within a channel flow. By calculating the corresponding surface stresses, the authors predict the expected shape deformation of such as a sphere as a function of its elastic modulus. While this approach is strictly valid only in the regime of small deformations, the theoretical predictions are in good agreement with experiments on soft spheres of known elasticity. The authors are able

to disentangle the effects of size and elasticity for the soft objects or cells being probed, and demonstrate that elastic properties can be measured at high speeds.

These developments, as well as expected upcoming improvement of alternative continuous flow approaches, contribute to the rapidly developing ability to use mechanical cues as biophysical markers in cell studies and open the door to high-throughput screening and sorting of large cell populations based on the precise mechanical properties of each individual cell.

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