

**Biophysical Journal**

**Supporting Material**

**Characterizing Cellular Biophysical Responses to Stress by Relating Density, Deformability, and Size**

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## Supporting Material

### Estimating the error contribution from converting buoyant mass to volume with an average cell density using a Monte Carlo simulation

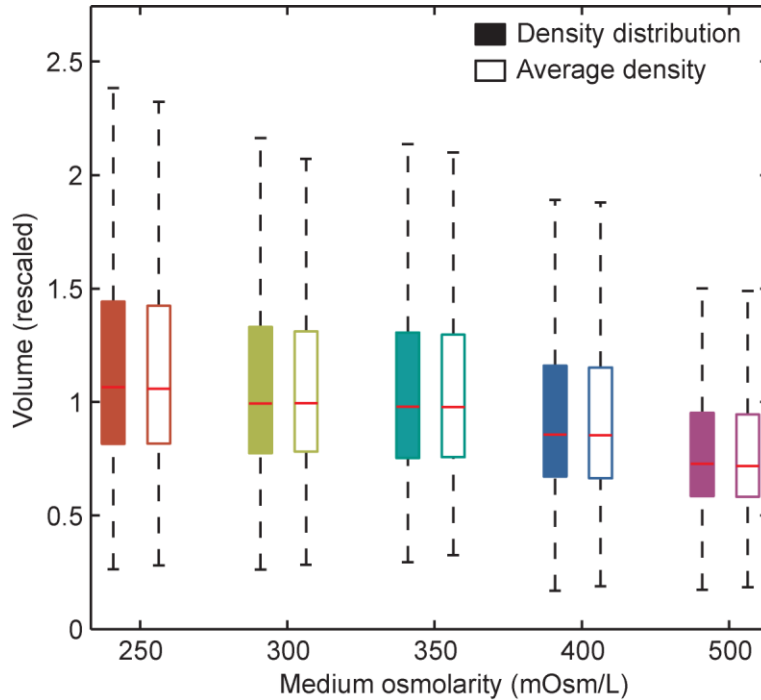


Fig. S1. Converting the buoyant mass from the constriction SMR to a volume using a density distribution (solid color) or average density (outline) value does not significantly increase the width of the subsequent volume distribution. A Monte Carlo simulation ( $n = 10000$ ) was used to calculate a volume distribution (solid color) based on independent, random sampling of values from buoyant mass and density distributions. This distribution was compared to a distribution calculated by converting a buoyant mass distribution to a volume distribution using an average density value (outline). Red lines represent the median for each value, the top and bottom box boundaries represent the 75<sup>th</sup> and 25<sup>th</sup> percentile of the data, respectively, and the whiskers represent the most extreme data points not considered outliers. Representative data is from measurements at 250, 300, 350, 400, and 500 mOsm/L.

### Estimating changes to water content

To determine the changes to water content following osmotic stress, we employed the model described below (1). We start by describing the total mass of the cell ( $m_{tot}$ ) as composed of aqueous ( $m_{water}$ ) and dry ( $m_{dry}$ ) material:

$$m_{tot} = m_{water} + m_{dry} \quad (1)$$

We can rewrite this equation in terms of density and volume, based on the general relationship between mass, density and volume described in equation (2):

$$m = \rho V \quad (2)$$

$$\rho_{tot} V_{tot} = \rho_{water} V_{water} + \rho_{dry} V_{dry} \quad (3)$$

where  $\rho_{tot}$ ,  $\rho_{water}$  and  $\rho_{dry}$  refer to the density of the total cellular, aqueous and dry material, respectively, and  $V_{tot}$ ,  $V_{water}$  and  $V_{dry}$  refer to the volume of the total, aqueous and dry material. We can also describe the volume of the cell in a manner similar to equation (1):

$$V_{tot} = V_{water} + V_{dry} \quad (4)$$

By combining equations (3) and (4), and assuming a water density of 1 g/mL, we can obtain expressions for the amounts of aqueous and dry material, described in terms of volume:

$$V_{dry} = V_{tot} \left( \frac{\rho_{tot} - 1}{\rho_{dry} - 1} \right) \quad (5)$$

$$V_{water} = V_{tot} \left( 1 - \left( \frac{\rho_{tot} - 1}{\rho_{dry} - 1} \right) \right) \quad (6)$$

Table S1. Biological effects and mechanisms of drugs used in Fig. 5.

Drug	Biological effects and mechanisms
Latrunculin B	<i>Inhibition of actin polymerization</i> : deformability decrease, apoptosis (2), cell cycle arrest (3), protein synthesis inhibition (4)
Staurosporine	<i>Inhibition of protein kinases</i> : apoptosis, deformability change (5), cell cycle arrest (6), protein synthesis inhibition (7)
Torin 1	<i>Inhibition of mTOR</i> : cell cycle arrest, modification of actin polymerization (8, 9), apoptosis (10) , protein synthesis inhibition (11)
Rapamycin	
Cycloheximide	<i>Inhibition of translocation</i> : protein synthesis inhibition, deformability change (12), apoptosis (13), cytoprotection (14), cell cycle arrest (15)

## Supporting References

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