SPHERICAL TIP SELECTION GUIDELINES

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Choosing an appropriate tip size is related to the structure of the sample and the kind of experiment one wishes to perform. When measuring the global elasticity of a sample, a larger tip would be more suitable, since the applied and recorded force of each indentation would effectively average over a larger area. For measuring small spatial features, a smaller tip could be more suitable. Tip size should be carefully considered when working with porous materials: to determine the overall structural properties of a porous material, select a tip size several times larger than the largest pore. For examining local features within one pore, select a tip size that is several times smaller than the pore diameter.

There are a few other considerations regarding the tip radius:

1. The maximum indentation depth shouldn't be more than 16% of the tip radius. This is the Hertz model assumption of the parabolic indenter used to calculate the contact area during indentation: $a = \sqrt{h * R}$, a - contact radius, h - indentation depth, R - tip radius.
2. The maximum indentation depth shouldn't be more than 5-10% of the sample thickness to not sense the substrate underneath the sample. Furthermore, the substrate should be much stiffer than the sample itself.
3. The sample is also assumed to be an infinite half-space, thus, tip size should be chosen so that this assumption is true.
4. When performing indentation mapping, the step size should be at least two times the contact radius *a*, to avoid oversampling.

Regarding the tip size, you should balance between the spatial resolution of the mapping which depends on the heterogeneity of the sample, and the scale of the measurement in terms of depth. If you only want to sense surface properties, a smaller indentation depth is desirable. If you want to sense more bulk/averaged properties, you should consider measurements at larger depths with a larger radius. However, when using larger spheres, step size needs to be higher and, thus, you lose spatial resolution during indentation mapping.

General guideline:

TYPE OF SAMPLE	RADIUS	INDENTATION-DEPTH (DMAX)
Single cells, 2D monolayers	3, 10 µm	0.5, 1.6 µm
Hydrogels	25, 50, 100, 250 µm	4, 8, 16, 40 µm
Tissues	25, 50, 100 µm	4, 8, 16 µm
Spheroids, Organoids, Oocytes	10, 25 µm	1.6, 4 µm

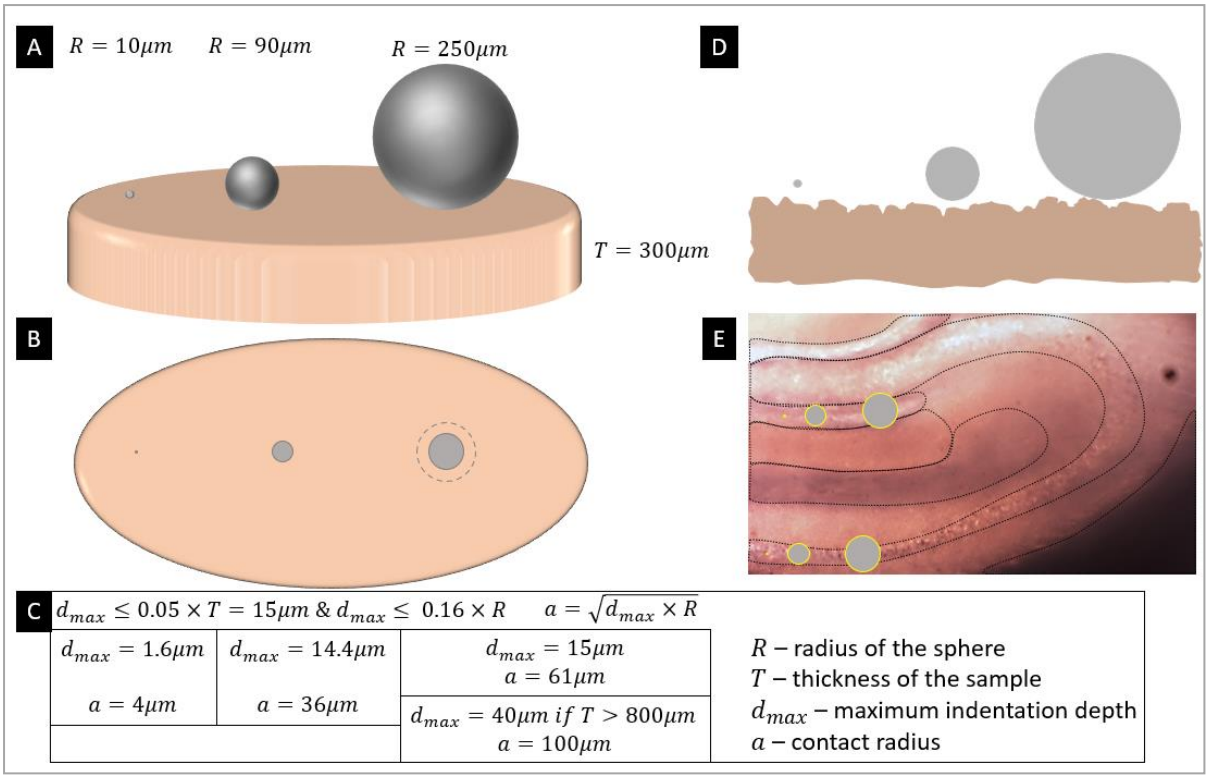

*Figure 1: Example of maximum indentation depth, $\mathbf{d_{max}}$, and B) contact radius, **a**, for three tip radius, **R**, concerning the A) sample thickness, **T**, D) surface roughness and E) regional heterogeneity (hippocampus).*