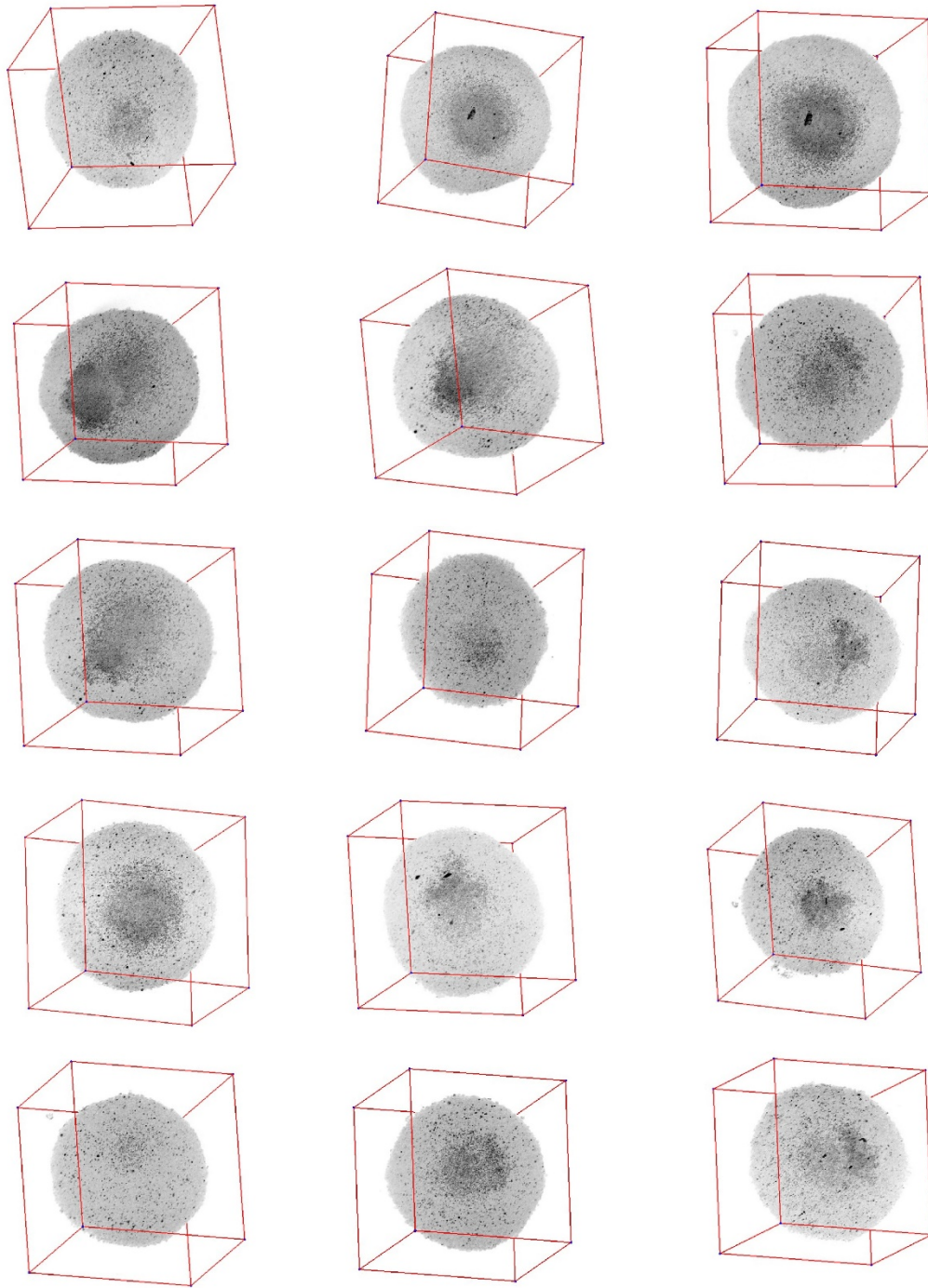**Electronic Supplementary Material**

1. Python code for automatic organoid detection.

2. Movie showing automatic sample detection in flow.

3. Image stacks of glioblastoma organoids mounted fixed in agarose vs. in flow recorded on the digitally scanned light-sheet microscope.

4. Image stack of a glioblastoma organoid in flow recorded on the Ultramicroscope II.

5. Image stack of a microglia-populated organoid recorded in flow on the digitally scanned light-sheet microscope.

Supplementary figure 1 | 3D renders of glioblastoma spheroids with nuclear counterstain on the Leica SP8 DLS