Supplementary Information

AIMS: Acoustofluidic Interfaces for the Mechanobiological Secretome of MSCs

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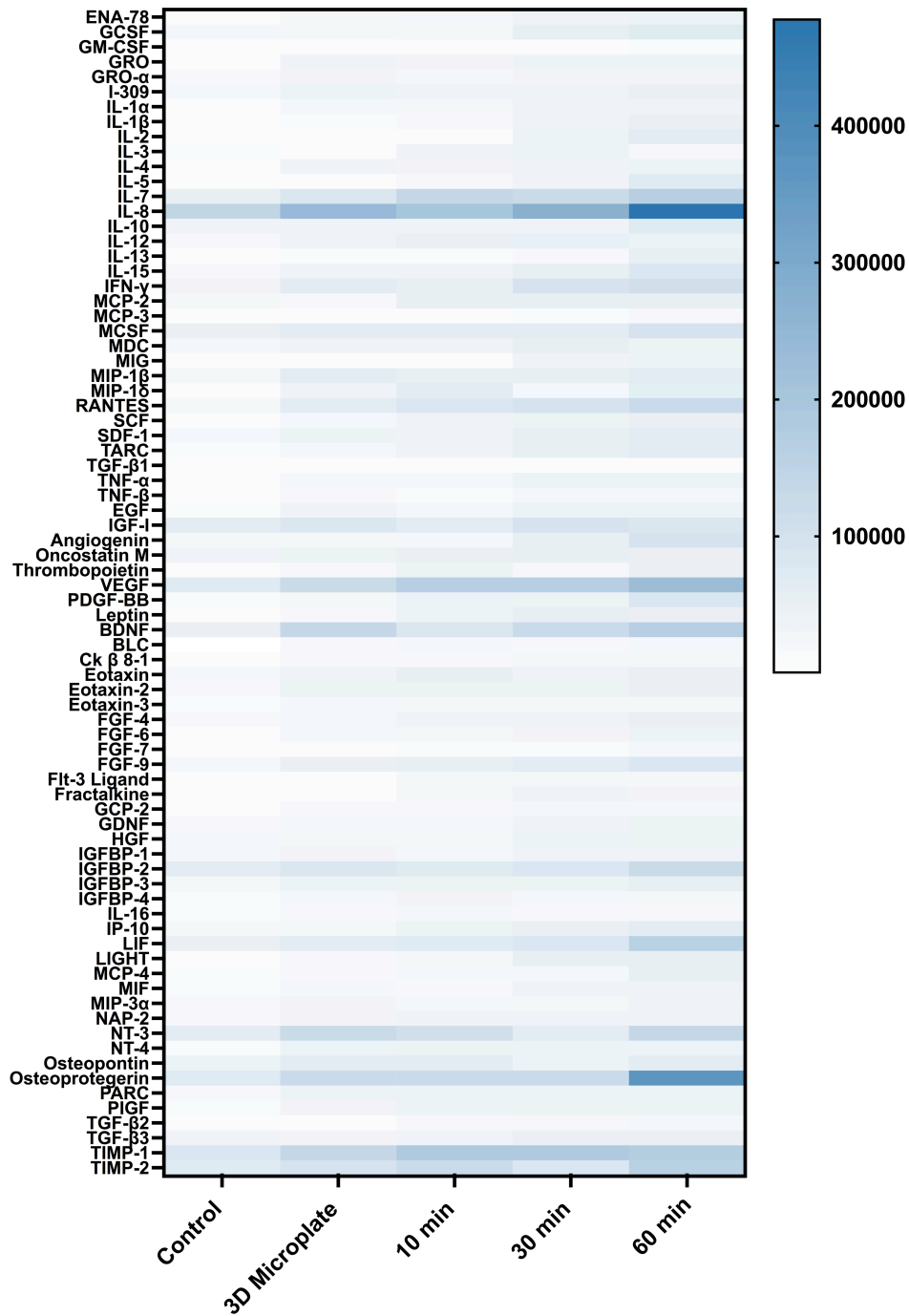
Supplementary Figure 1 | Heatmap presenting the integrated densities of different groups after incubating with the cytokine array for 4 days (without Z-score normalization and clustering process). Source data are provided as a Source Data file.
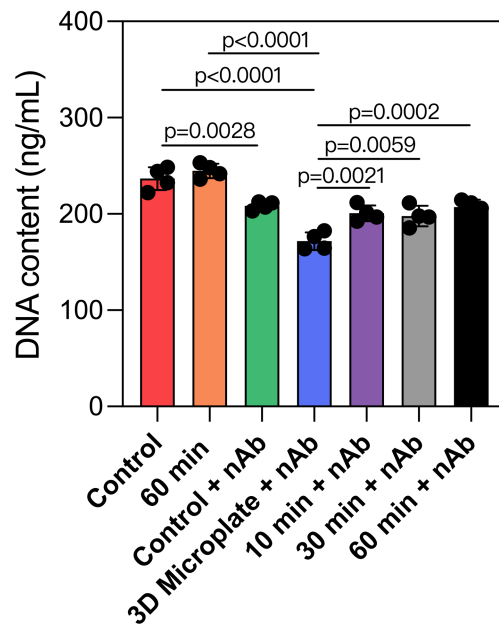
Supplementary Figure 2 | CyQUANT™ Cell Proliferation Assay measuring the DNA contents after functional blocking of N-cadherin of MSCs and incubating for 4 days. Data are graphed as the mean ± SD (n=4, biological repeats). The statistical analysis was performed using one-way ANOVA with Tukey's post-hoc test by GraphPad Prism 9.0. Source data are provided as a Source Data file.
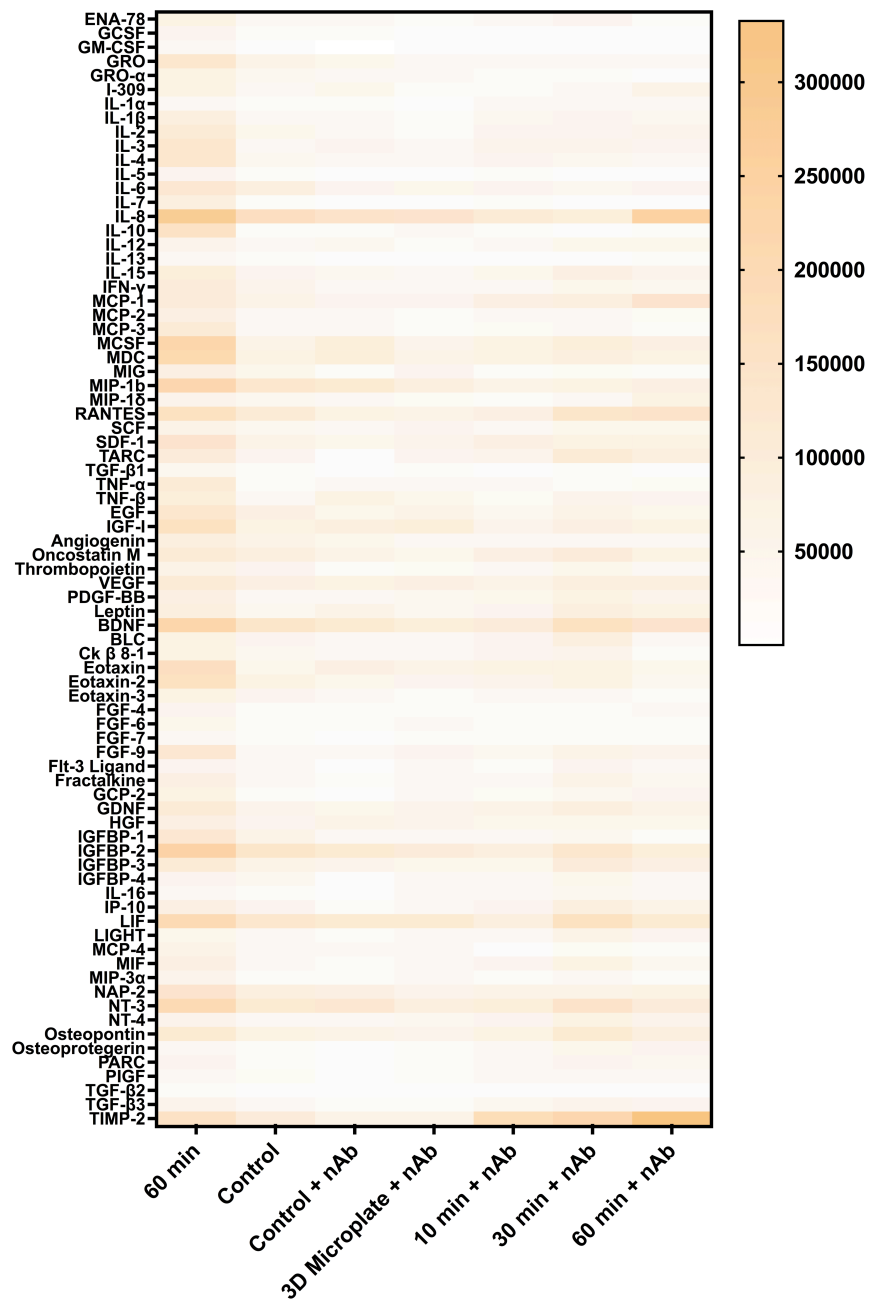
Supplementary Figure 3 | Heatmap presenting the integrated densities of different groups after functional blocking N-cadherin of MSCs and incubating with the cytokine array for 4 days (without Z-score normalization and clustering process). Source data are provided as a Source Data file.
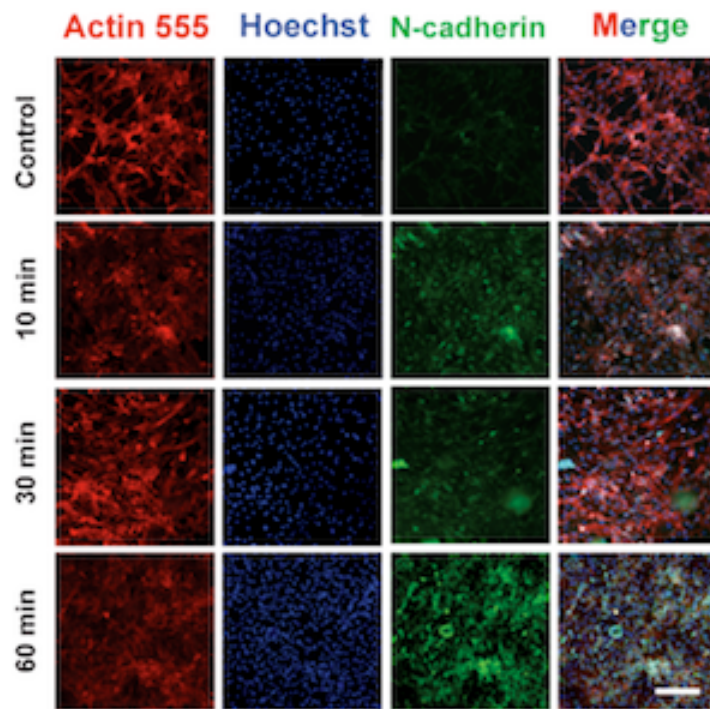
Supplementary Figure 4 | Representative immunofluorescence images of different groups after incubating for 3 days. The expression of N-cadherin is shown in green. The cytoskeleton is shown in red. The cell nucleus is shown in blue. n=3 tests. Scale bar: 50 µm.
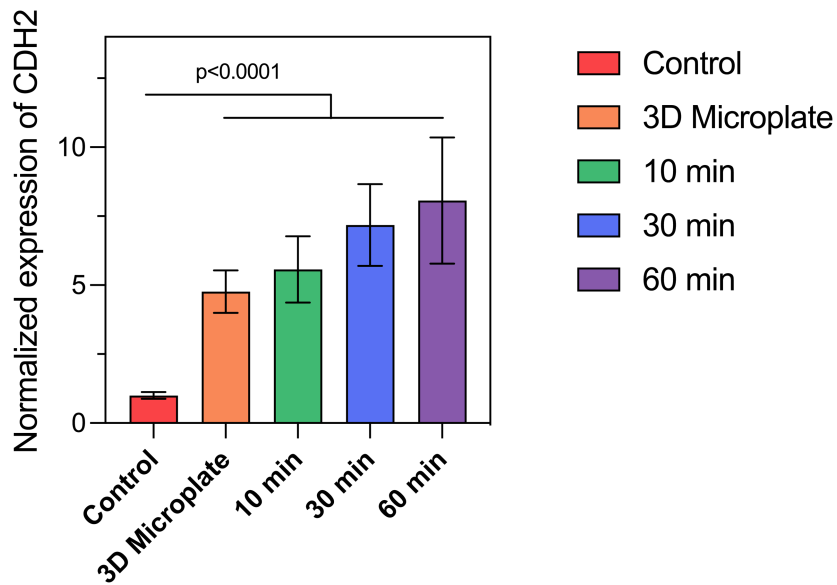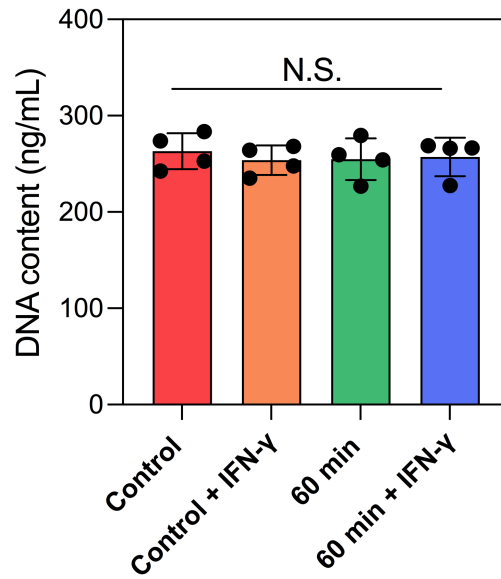
Supplementary Figure 5 | QPCR measuring the relative expression of CDH2 gene (N-cadherin) of different groups after incubating for 3 days. Data are graphed as the mean ± SD (n=3, biological repeats). The statistical analysis was performed using one-way ANOVA with Tukey's post-hoc test by GraphPad Prism 9.0. Source data are provided as a Source Data file.
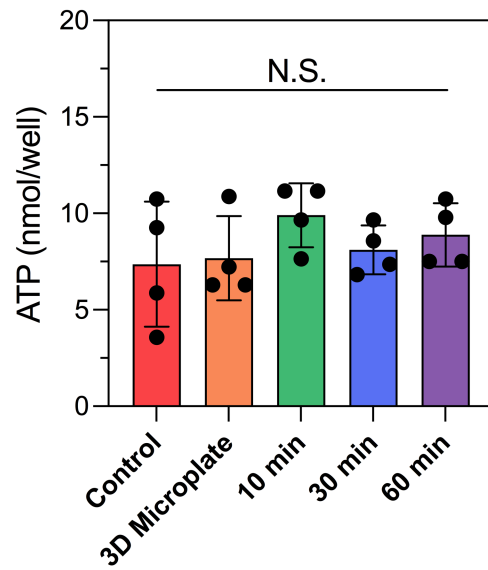
Supplementary Figure 6 | The analysis of ATP concentrations of different groups after incubating for 4 days. Data are graphed as the mean ± SD (n=4, biological repeats). The statistical analysis was performed using one-way ANOVA with Tukey's post-hoc test by GraphPad Prism 9.0. Source data are provided as a Source Data file.
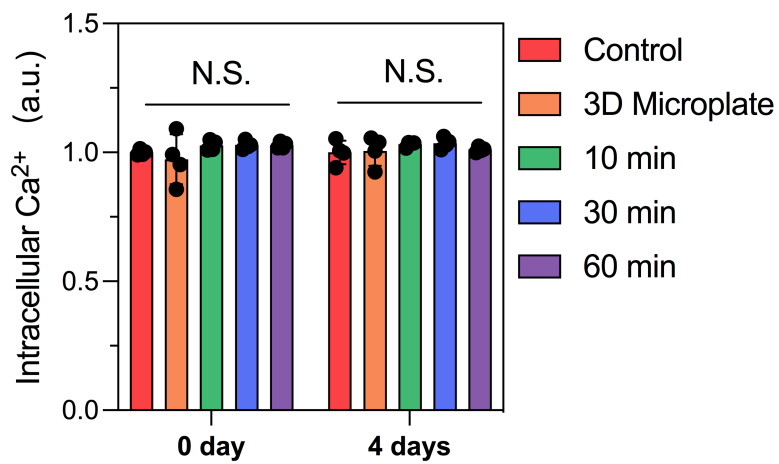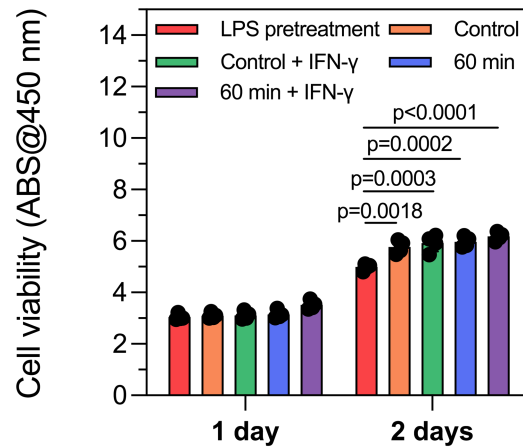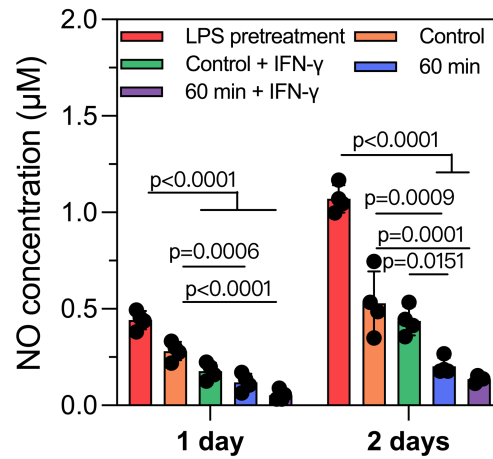
Supplementary Figure 7 | The analysis of intracellular Ca^{2+} concentrations of different groups after incubating for 4 days. Data are graphed as the mean ± SD (n=4, biological repeats). The statistical analysis was performed using one-way ANOVA with Tukey's post-hoc test by GraphPad Prism 9.0. N.S.: not significant. Source data are provided as a Source Data file.

Supplementary Figure 8 | CyQUANT™ Cell Proliferation Assay measuring the DNA contents of IFN-γ primed MSCs from different groups and incubating for 4 days. Data are graphed as the mean ± SD (n=4, biological repeats). The statistical analysis was performed using one-way ANOVA with Tukey's post-hoc test by GraphPad Prism 9.0. N.S.: not significant. Source data are provided as a Source Data file.
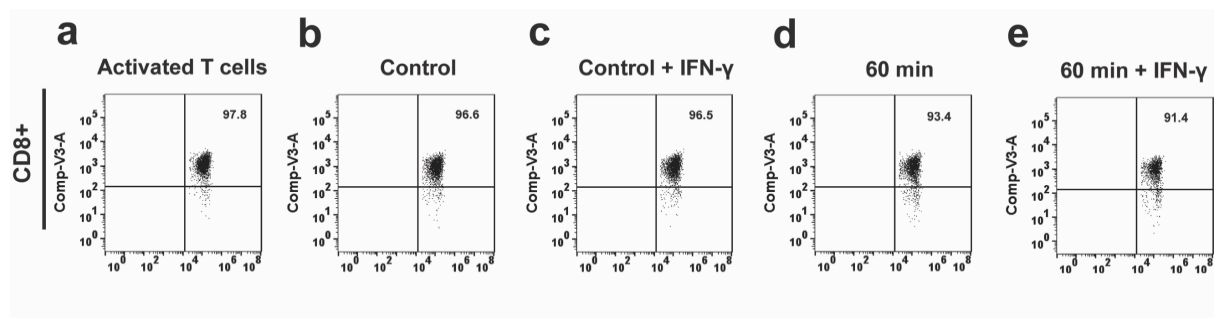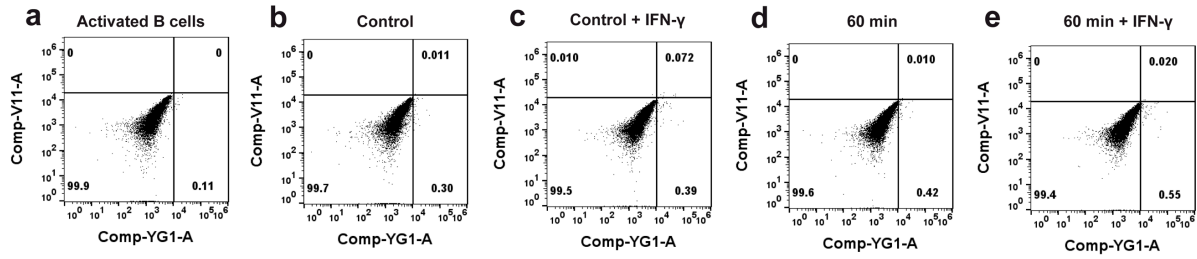
Supplementary Figure 9 | CCK-8 assay measuring the viability of Human THP-1 cells from different groups after incubating for 1 and 2 days. Data are graphed as the mean ± SD (n=4, biological repeats). The statistical analysis was performed using one-way ANOVA with Tukey's post-hoc test by GraphPad Prism 9.0. Source data are provided as a Source Data file.

Supplementary Figure 10 | The analysis of nitric oxide (NO) production of different groups after incubating for 1 and 2 days. Data are graphed as the mean ± SD (n=4, biological repeats). The statistical analysis was performed using one-way ANOVA with Tukey's post-hoc test by GraphPad Prism 9.0. Source data are provided as a Source Data file.

Supplementary Figure 11 | Flow cytometry analysis measuring the percentage of CD8 positive T cells of each group after incubating with the conditional medium from MSCs for 3 days. a: Activated T cells, b: Control, c: Control + IFN-γ, d: 60 min, e: 60 min + IFN-γ. Source data are provided as a Source Data file.

Supplementary Figure 12 | Flow cytometry analysis measuring the B cell apoptosis of each group after incubating with the conditional medium from MSCs for 7 days. a: Activated T cells, b: Control, c: Control + IFN-γ, d: 60 min, e: 60 min + IFN-γ. Source data are provided as a Source Data file.
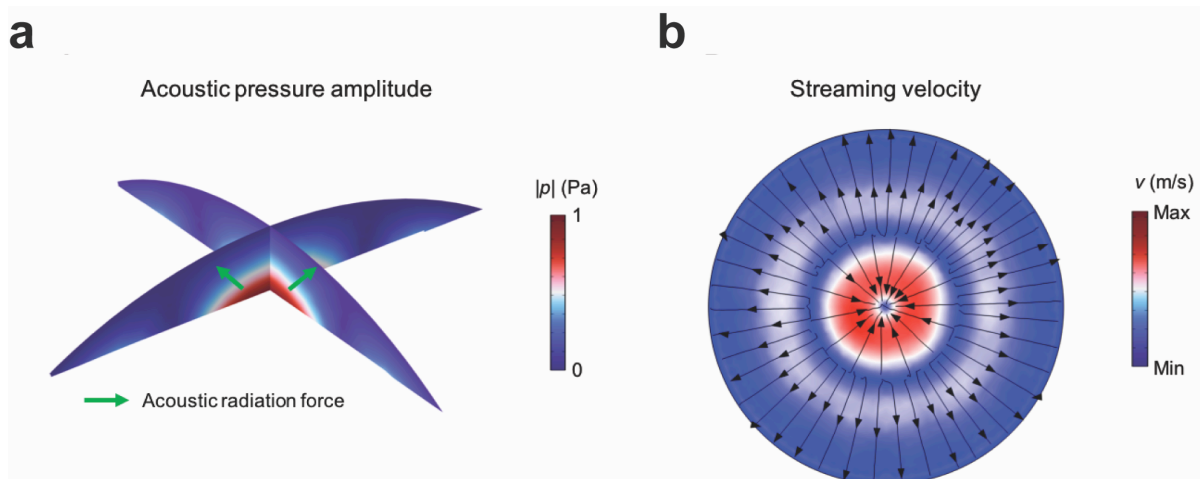
Supplementary Figure 13 | Simulation results depict the acoustic pressure amplitude **(a)** and acoustic streaming velocity **(b)** on a cross-sectional plane near the bottom of the device.
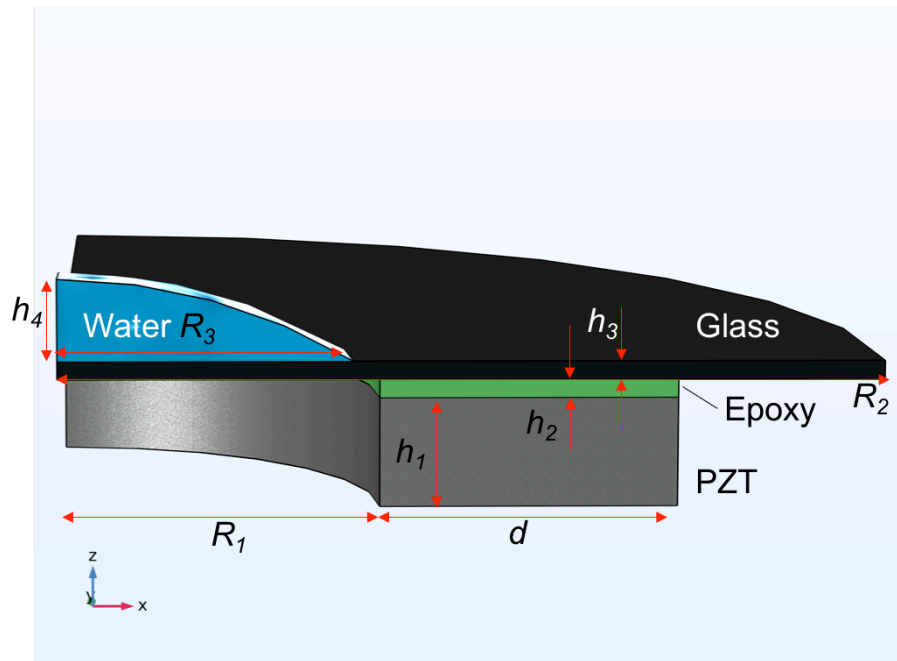
Supplementary Figure 14 | Illustration of the geometry and dimensions of the simulation model. A fully coupled three-dimensional model was developed by coupling the acoustic pressure module and solid mechanics module to calculate the acoustic intensity distribution. The model setup consisted of a ring piezoelectric transducer (PZT) located at the base of a glass substrate, affixed with a thin layer of Epoxy. Positioned atop this glass substrate was a small droplet. The geometric parameters of the model can be find in Table 1 (main text).