Preparation of lipid microbubbles

- Stock solutions of lipids DSPC and DSPE were prepared in chloroform and mixed in molar ratio 90:10 in glass vial.
- Chloroform was removed by vacuum desiccation, 10-15 min.
- Lipids were hydrated by adding PBS and vortexed for 30 s.
- After 1-2 h Incubation at 60°C with gentle shaking, lipids were cooled down on ice for 5 min.
- Perfluorohexane was added directly in the vial with the syringe.
- Microbubbles were obtained by sonication (3x 1s ON, 10s OFF, 20% amplitude).
- Samples were centrifuged to separate microbubbles from lipid debris (1000 rpm, 3 min).