Focused ultrasound induced inertial cavitation by amino-propyl functionalized ZnO nanoparticles

Andrea Ancona¹, Giancarlo Canavese¹, Adriano Troia², Veronica Vighetto¹, Marco Laurenti¹, Nadia Garino¹, Valentia Cauda¹

¹Department of Applied Science and Technology, Politecnico di Torino
²Istituto Nazionale di Ricerca e Metrologia (INRiM), Torino
Corresponding author: valentina.cauda@polito.it

Introduction

Inertial cavitation (IC) refers to the rapid growth and violent collapse of bubbles after exposure to ultrasound. As ultrasound wave travels through a liquid/tissue, any gas bubbles in the liquid are forced to oscillate in the applied acoustic field. Increasing acoustic pressure, this oscillation becomes unstable and eventually the bubble implodes, generating extremely high temperatures and pressures at the center of the collapsing bubble [1]. IC has been identified as a key mechanism behind ultrasound-related biomedical applications, such as improved delivery of drugs, sonodynamic therapy and tissue ablation. It has been largely demonstrated that the presence of nanoparticles (NPs) in aqueous solutions decreases the cavitation threshold [2]. NPs are indeed able to stabilize nanobubbles on their surface and inside well-defined cavities. These nanobubbles can initiate acoustic inertial cavitation acting as cavitation nuclei. Different approaches have been used in the literature to stabilize gas nanobubbles on the surface of solid nanoparticles [3]. Yildrim et al. exploited hydrophobic functionalization of mesoporous silica nanoparticles to stabilize surface nanobubbles able to generate cavitation under US exposure [4]. Jin et al. discovered that superhydrophobic polytetrafluoroethylene (PTFE) NPs were able to initiate IC and ROS generation [5], while Kwan et al. developed cup-shaped polystyrene NPs that could trap nanobubbles on their surface after the drying process [6]. Here we show that surface functionalization of zinc oxide nanometers particles with amino-propyl groups (ZnO-NH₂ NPs) greatly enhance their ability to incept IC under ultrasonic irradiation by stabilizing gas nanobubbles on their surface, thus making them suitable for therapeutic and imaging applications based on IC activity.

Methods

Zinc oxide nanoparticles were synthesized using a novel microwave-assisted wet-chemical synthetic approach. They were subsequently chemically modified by anchoring amino-propyl group to the ZnO surface. This approach leads to round-shaped nanocrystals with an average size of 15 nm. A homemade focused ultrasonic transducer centered at 985 KHz was used to irradiate the ultrapure water solution containing the ZnO-NH₂ NPs. Ultrasound was applied as single pulses of 100 cycles each with a Peak Rarefaction negative pressure of 1.8 MPa. Acoustic cavitation inception by ZnO-NH₂ nanoparticles was tested by recording the broad band acoustic emissions generated by collapsing bubbles using a passive cavitation detector centered at 5 MHz (Precision Acoustics). The foci of both transducers were adjusted to overlap at the same position. The IC signal was received and amplified by a 25 dB hydrophone booster amplifier (Precision Acoustics). The recorded time-domain signal acquired at a sampling rate of 1 Gsamples/s was first transformed into a frequency domain spectrum A(f) using Fast Fourier transform. Then the integral values of area under the frequency spectrum from 2 to 10 MHz was calculated and termed as cavitation dose (V/Hz), which can be used to assess the IC intensity.

Results

Table 1 shows the cavitation dose generated by water and water containing ZnO NPs (200 μg/mL) or ZnO - NH₂ NPs (200 μg/mL). Amino-propyl functionalization enhanced the ability of ZnO nanoparticles to induce inertial cavitation. Our hypothesis is that the amino-propyl group attached to the
ZnO NP, by increasing the local hydrophobicity, induced the formation of a layer of gas molecules adsorbed to the surface. To test this hypothesis, two different experiments were performed. The first one consisted in degassing the solution containing water and ZnO-NH$_2$ NPs (200 µg/mL) by applying vacuum to the chamber. Table 1 shows that after the process of vacuum degassing, ZnO-NH$_2$ were no longer able to induce inertial cavitation. This result suggests that by vacuum degassing, the gas nanobubbles adsorbed on their surface were removed, thus preventing the induction of inertial cavitation. To further explore this hypothesis, ZnO-NH$_2$ nanoparticles were treated with ethanol (90 v/v %). Ethanol is expected to destroy surface nanobubbles by wetting the surface of the nanoparticles and displace the entrapped gas [4]. As shown in Table 1, ethanol treatment drastically reduced the cavitation dose intensity detected. Together, these data suggest that the acoustic inertial cavitation initiated by ZnO-NH$_2$ nanoparticles under focused ultrasonic irradiation is due to the periodic oscillation and subsequent implosion of the surface nanobubbles trapped at the surface of ZnO-NH$_2$ nanoparticles.

**Conclusions**

We have reported an innovative strategy to increase the ability of zinc oxide nanoparticles to induce inertial cavitation by functionalizing their surface with amino-propyl groups. We validated their acoustic activity under focused ultrasound irradiation by recording the broad band noise emitted by collapsing bubbles. We suggest that a gas layer adsorbed on the surface of the ZnO-NH$_2$ nanoparticles could be responsible for the induction of inertial cavitation, and we validated this hypothesis by showing that vacuum degassing and ethanol addition completely inhibited the ability of ZnO-NH$_2$ NPs to induce inertial cavitation.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Water (V/Hz)</th>
<th>ZnO NPs (V/Hz)</th>
<th>ZnO-NH$_2$ NPs (V/Hz)</th>
</tr>
</thead>
<tbody>
<tr>
<td>None</td>
<td>272 ± 95</td>
<td>1183 ± 89</td>
<td>9785 ± 123</td>
</tr>
<tr>
<td>Vacuum degassing</td>
<td>128 ± 92</td>
<td>234 ± 87</td>
<td>484 ± 88</td>
</tr>
<tr>
<td>Ethanol addition (90 v/v %)</td>
<td>148 ± 98</td>
<td>289 ± 93</td>
<td>382 ± 94</td>
</tr>
</tbody>
</table>

Table 1. Inertial cavitation dose of water solution, Zinc Oxide (ZnO) and Amino-propyl zinc oxide (ZnO-NH$_2$) nanoparticles under different experimental conditions, n=3

**References**


