A preliminary study for cancer therapy based on amino acid-combined nanobubbles: an approach to lung cancer cells (A549)

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Nanobubbles are extremely ultrafine bubbles (less than 500 nm in size) and have attracted much attention in a wide range of advanced science and technology, including medical, agricultural, and food. In recent years, nanobubble particularly have been applied in a variety of ways in the medical and clinical areas. For example, nanobubbles are intensively used as ultrasonic contrast medium and therapeutic delivery[1]. Besides therapeutic delivery, oxidative nanobubbles have several advantages; in particular, it has the advantage of destroying or inactivating pathogens such as bacteria and viruses. In this study, we report that amino acid-combined oxygen nanobubbles (O_2NBs) were actively able to inactivate cancer cells compared to normal O_2NBS. For this purpose, an experiment using lung cancer cells (A549) with the highest mortality rate was conducted. In addition, lysine, one of the basic amino acids, was used in this study. Since the extracellular pH of cancer cells is slightly acidic, less than 6.7, basic amino acids can assist in the introduction of nanobubbles into the cell. Lysine solutions at 0.5, 1, and 3 mM were used to find the optimal lysin concentration. The experimental set-up and cell viability under different conditions are shown in Figure 1.

(a) (b)

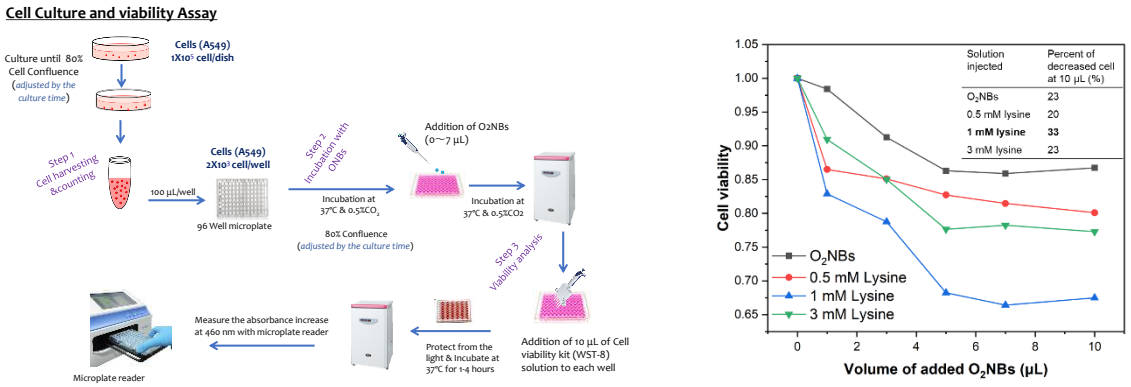

Figure 1. (a) Experimental set-up for cell culture and viability assay and (b) cell viability under different conditions.

O_2NBs were generated by mixing oxygen into pure water through a generator with air gap structure-, consequently showing a concentration of ca. 2×10^8 particles/mL. $2x10^3$ of A549 cells were cultured on a 96-well microplate for two days, and then nanobubbles, or lysin-including O_2NBs, were added. One day after, cell viability was measured using a cell counting kit-8. As shown in Figure 1b, the cancer cells tended to decrease with O_2NBs; however, it is more effective when mixed with lysine. On the other hand, higher lysin concentrations over 3 mM was inefficient and the 1 mM lysine is an appropriate and more effective concentration in this case. The current results suggests the synergy effect of lysine and nanobubbles and we are working toward further optimization of experimental conditions to investigate the biological mechanism of O_2NBs in the lung cancer and normal cell lines.

References

[1] Hansen, H.H.W.B; Cha, H; Ouyang, L; Zhang, J; Jin, B;Stratton, H; Nguyen, N. T; An, H. Nanobubble technologies: Applications in therapy from molecular to cellular level. Biotechnology Advances 2023, 63, 108091. https://doi.org/10.1016/j.biotechadv.2022.108091.