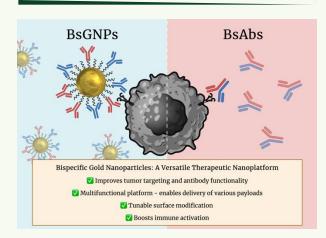
Enhancing Therapeutic Efficacy and Immune Response with Bispecific Gold Nanoparticles in Cancer

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Introduction

Bispecific antibodies (bsAbs) are emerging as promising new cancer therapies, redirecting immune cells to tumors and blocking tumor survival pathways. However, their clinical use faces various challenges, including complex design, poor pharmacokinetics, limited stability, and limited penetration into solid tumors [1]. This study presents a gold nanoparticle (GNP)-based nanoplatform that harnesses the inherent biocompatibility and tunable surface chemistry of GNPs to enhance bsAb therapies. The resulting bispecific gold nanoparticles (BsGNPs) are designed to overcome current therapeutic limitations and improve overall treatment efficacy.



Methods

GNPs were functionalized with trastuzumab and pertuzumab via a PEG linker. The resulting BsGNPs were assessed in breast cancer cell lines for cytotoxic ability (CyQuant analysis) and immune activation (via co-culture with human NK cells and flow cytometry). In a mouse model of breast cancer, BsGNPs were evaluated for tumor accumulation computed tomography (CT) imaging, for anti-tumor efficacy by tumor size measurements.

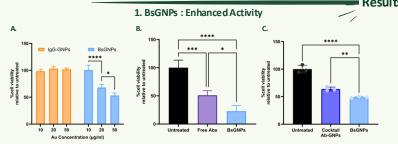


Fig. 1. Tumor cell killing abilities of BsGNPs. SKBR3 breast cancer cell line was treated with BsGNPs or controls and efficacy was evaluated. (A) Dose-dependent cyto toxicity of BsGNPs (solid bars) vs control IgG-GNPs (dotted bars), using 10, 20, and 50 μg/mL gold. (B) %Cell viability after treatment with either free antibodies or BsGNPs (an equal antibod y quantity was present in both free and conjugated groups). Results show that conjugating antibodies to GNPs enhances their efficacy. (C) %Cell viability after treatment either with BsGNPs or with a cocktail of GNPs conjugated to either trastuzumab or pertuzumab. Results demonstrate superior tumor cell killing effect of BsGNPs. Data are presented as mean ± 50. *Student's t-test. *p < 0.01.



Fig. 2. BsGNPs induce NK cell-mediated antibody dependent cellular cytotoxicity (ADCC). (A) %Cell viability of SKBR-3 cells either alone or in co-culture with human NK cells, and treated with BsGNPs. BsGNPs enhanced cancer cell kiling in NK co-culture. Results presented as mean ± SBM; ***p<0.001 (Student's t-test). (B) SKBR-3 breast cancer cells were co-cultured with NK cells and treated with either BsGNPs, IgG-conjugated GNPs, or free antibodies. CD107a was analyzed by flow cytometry analysis. Results show enhanced degranulation and activation of NK cells after treatment with BsGNPs. Thus, BsGNPs likely induce NK cell-mediated ADCC.

3. BsGNPs Enhance Targeting and Therapeutic Impact

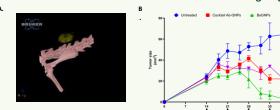


Figure 3. Targeting and therapeutic efficacy of BsGNPs in a mouse model of breast cancer. (A) Representative 3D micro-CT image showing accumulation of BsGNPs within the tumor. Mice were scanned 24hrs after IV injection of BsGNPs. (B) Tumor size over time following IV administration of either BsGNPs, free antibodies, or a cocktail of trastuzmab/pertuzumab-conjugated GNPs. BsGNPs significantly decreased tumor size as compared to controls. Arrows indicate administration days. Data are shown as mean ± SEM (n = 3/group).

Conclusion

This study demonstrates the successful development of BsGNPs, highlighting the potential of this approach for dual-antibody cancer therapy. Our results show enhanced in vitro and in vivo efficacy, along with precise tumor targeting. BsGNPs offer a significant advancement in nanoparticle-based therapy by combining the benefits of gold nanoparticle delivery with synergistic, bispecific antibody targeting.

^[1] Nejadmoghaddam, M., Minai-Tehrani, A., and Ghahremanzadeh, R., Avicenna J Med Biotechnol. 2019:11(1):3-23