

Development of anti-FZD7 scFv antibody and evaluation of its apoptotic effect on breast cancer cells

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Frizzled family receptor 7 (FZD7) is one of the 10 members of Frizzled family of receptors. It interacts with Wnt ligands to activate canonical Wnt signaling, which function to turn on different downstream transcription factors essential for modulating cellular proliferation, polarity, and differentiation. Altered expression of FZD7 receptor is associated with development and progression breast cancer. An effective targeted therapy for breast cancer through modulating ligand-receptor interaction may involve using antibodies to antagonize FZD7. ScFvs (single-chain fragment variable) have provided an alternative to full-length monoclonal antibodies (mAbs) in diagnostic and therapeutic applications. A phage antibody library of scFv was used and selection of specific scFvs were performed by 4 rounds of panning process against an immunodominant epitope of FZD7, followed by PCR and fingerprinting of the selected clones. ELISA was used to confirm the specificity of the clones. Apoptotic effects of the selected scFv on MDA-MB-231 cell line were assessed by annexin V/PI assay after 24 h and 48 h. A specific scFv with the frequency of 35% was isolated which produced positive ELISA with the corresponding epitope. After 24h treatment with the selected scFv, MDA-MB-231 cells showed 48.7% apoptotic cell death (Annexin V+/PI−). However, this amount increased to 81.6% following 48h treatment with scFv. Due to unique apoptotic properties of selected scFv including human origin, high affinity and specificity, this agent has been applied in cancer immunotherapy. The specific anti-FZD7 scFv selected in this study has the potential to be used for inhibiting Wnt signaling pathway in breast cancer cells.

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