

Construction, expression and characterisation of a single chain variable fragment using phage display technology to recognize MCF-7 breast cancer cells in the *E. coli* periplasm

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A functional single chain variable fragment (scFv) recognising the MCF-7 breast cancer carcinoma cell line was constructed from the C3A8 hybridoma using phage display technology. The scFv coding sequence was cloned in frame with the pIII phage coat protein. The signal sequence included in the C terminus directed the expression of the scFv in the *E. coli* periplasm. Following several rounds of biopanning, colonies that expressed a scFv that recognized MCF-7 cells in Western blots and ELISAs were isolated. A 750-bp scFv gene was successfully isolated. Cloning and two rounds of biopanning isolated the candidate with the highest activity (clone B7), as screened by ELISA. Following SDS-PAGE of the purified product, a 32-kDa band was observed. A similar-sized band was observed following Western blot analysis with an E tag-specific antibody. The indirect ELISA established that the affinity of the best representative clones of the single-chain antibodies. The recombinant antibody technology used in this study is a rapid and effective approach that will aid in the development of the next generation of immunodiagnostic reagents.

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