Immortalized Pig Fibroblasts: Dual Platform

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Abstract:

In life sciences, cell culture has become a cornerstone for drug screening, toxicity testing, and disease modeling. While primary cells rapidly senesce in vitro, immortalized cell lines overcome proliferation limitations and offer homogeneity for biomedical research. Bama Xiang pig, with unique biological stability and utility in disease modeling, has gained prominence in biomedical studies. Its embryonic fibroblasts (BmPEFs) serve as ideal targets for transgenic research and viral propagation due to low differentiation and high proliferative capacity.

This study established BmPEF lines using 38-day embryos through optimized enzymatic digestion (collagenase IV yielding 7.2×10^{7}~1.29×10^{8} cells/embryo) and contamination screening. Culture conditions were optimized to DMEM with 10% FBS and 2 ng/mL bFGF. Spontaneous immortalization achieved 17-50 passages, while telomerase overexpression enabled stable proliferation beyond 50 passages. Characterized BmPEFs exhibited fibroblast morphology (vimentin+/CKpan-), stable growth curves with transient proliferation decline at P26-39, and normal cell cycle profiles. Species-specific cytochrome B sequencing confirmed porcine origin without plasmid residues.

Preliminary applications demonstrated BmPEFs' susceptibility to PEDV (showing distinct cytopathic effects vs Vero cells) and CRISPR/Cas9-mediated USH2A knockout efficiency. These immortalized BmPEFs provide valuable resources for vaccine development (viral propagation) and genetic disease modeling (gene editing), offering a standardized cellular platform for biomedical research.