Original Paper

Isolation and culture of interfollicular epidermal stem cells from newborn mouse skin without feeder layer

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Abstract

Background and Objective: Epidermis is the outer layer of skin, regenerating continuously. Epidermal stem cells play important roles in tissue regeneration, scar regeneration and neoplasm formation.This study was displayed for the isolation and culture of interfollicular epidermal stem cells from newborn mouse skin without feeder layer.

Materials and Methods: This experimental study was displayed on 0-3 old-day newborn NMRI mouse skin 60-70 gr weight. The epidermal keratinocytes were separated mechanically and enzymatically from 0-3 old day newborn mice skin (NMRI strain) and seeded on fibronectin-collagen culture substrates. Putative epidermal stem cells were selected by rapid adherence for 10 minutes on this composite matrix of type 1 collagen and fibronectin and the unattached cells were discarded and attached cells were cultured in essential minimal eagle medium (EMEM) (ca+2-free culture medium containing 0.05 mM Ca+2, 9% FBS, 50% conditioned medium, EGF (epidermal growth factor) and Cholera Toxin. The immunocytochemistry of β1-integrin analysis used to indicate their stemness nature.

Results: The results indicated that rapid adherence yields 50% purity. By using this method, the stem cells have been subcultured continuously without any change in the cell properties. The isolated interfollicular epidermal stem cells, expressed epidermal stem cells special marker (β1-integrin) in high levels, which indicates stem cell nature.

Conclusion: This new method yields pure viable epidermal stem cells that can be used in regenerative medicine and cell therapy.

Keywords: Interfollicular epidermal stem cells, Feeder layer, β1-integrin, Keratinocyte

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