

428p Guiding Human Embryonic Stem Cell Fate Choice to the Keratinocyte Lineage

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Human embryonic stem cells (hESCs) hold tremendous potential in the future of tissue engineering, offering promise as a source of virtually unlimited amounts of desired cell and tissue types. We have identified soluble chemical and extracellular matrix factors that permit isolation of epidermal keratinocytes from hESCs. Culturing embryoid bodies (EBs) formed from hESCs in a defined serum free keratinocyte growth medium on a gelatin matrix generates keratin 14 (K14) expressing cells with an epithelial morphology. K14 is a well-established marker for proliferative basal keratinocytes. These K14 expressing cells derived from hESCs can be subcultured in medium supplemented with hydrocortisone and induced to stratify and terminally differentiate by addition of calcium ions. Optimum times for obtaining K14 expressing cells exist for EB formation and for differentiation and growth of cultures after EB plating. EB formation is not necessary to generate keratinocytes; direct transfer of hESC colonies to keratinocyte growth medium permits keratinocyte differentiation. This culture system also leads to formation of melanocytes, another epidermal cell type. With further studies to optimize generation and purification of hESC-derived keratinocytes, these cells could provide a source of epidermal cells for skin tissue engineering applications, *in vitro* or *in vivo*.