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(54) **Title:** ESTABLISHMENT OF A HUMAN EMBRYONIC STEM CELL LINE USING MAMMALIAN CELLS

(57) **Abstract:** Purified preparations of human embryonic stem cells with certain population-specific characteristics are disclosed. This preparation is characterized by the positive expression of the following pluripotent cell surface markers: SSEA-1 (-); SSEA-4 (+); TRA-1-60 (+); TRA-1-81 (+); alkaline phosphatase (+), as well as a set of ES cell markers including Oct-4, Nanog, Rexl, Sox2, Thyl, FGF4, ABCG2, Dppa5, UTFl, Criptol, hTERT, Connexin-43 and Connexin-45. The cells of the preparation are negative for lineage specific markers like Keratin 8, Sox-1, NFH (ectoderm), MyoD, brachyury, cardiac-actin (mesoderm), HNF-3 beta, albumin, and PDXl (endoderm). The cells of the preparation are human embryonic stem cells, have normal karyotypes, exhibit high telomerase activity and continue to proliferate in an undifferentiated state after continuous culture for over 40 passages. The embryonic stem cell line Relicell™ Hes1 also retains the ability, throughout the culture, to differentiate into cell and tissue types derived from all three embryonic germ layers (endoderm, mesoderm and ectoderm). Methods for isolating a human embryonic stem cell line are also disclosed.

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