



## **THE USE OF EMIT TECHNOLOGY IN THE GENERATION OF FUNCTIONAL HEPATOCYTES FROM hES CELLS**

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To understand how a stem cell differentiates into a specific tissue-cell requires not only knowledge concerning the molecular pathways of differentiation, but also the identification of the combination of signals the stem cell requires to become a specific type of a differentiated cell tissue. StemCell Services' Emit® (Emission Identification Technology) enables direct noninvasive monitoring of the development process of human stem cells as they differentiate.

Emit® technology depends on the creation of a range of fluorescent proteins controlled by a set of specific promoters able to identify those chemical signals most relevant for the conversion of a stem cell into a mature, fully functional cell. As the stem cell progresses through each stage, it expresses these specific sets of proteins, and in the process switches on one of our fluorescent detector proteins. By following the sequential switching on of individual Emit® detectors in hES cells destined to become hepatocytes we were able to precisely track hepatocyte development. We were then able to look for these triggers in our specially constructed focused ligand collections and growth factor panels.

A variety of markers expressed in hepatocytes, can then be used to characterize the hepatic-like cells - transcription factor 1 & 2 (TCF1 & 2), FOXA1, FOXA2, HNF4, and HNF6, the carrier proteins alpha-fetoprotein (AFP) and albumin and enzymes such as TDO, TAT, CYP 450 enzymes, clotting factors, complement, transporter proteins, bile, lipids and lipoproteins. Using electron microscopy, the morphological features of the cells can be tested and periodic acid shift staining can be used to trace glycogen granules. Hepatocyte function is indicated by the production of urea & albumin, phenobarbital-induced cytochrome P450 expression or the presence of certain metabolic and detoxifying enzymes. The morphological characteristic of the cells can be confirmed using microscopy.

Human hepatocytes are extremely useful in the evaluation of the toxicity of new and existing pharmaceuticals and are highly sought after since donation cannot meet current demand. Emit® has been successfully used to develop a rapid and efficient renewable source of functional hepatocytes from hES cell lines.