FEATURES

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Pyrosequencing for Accurate Imprinted Allele Expression Analysis

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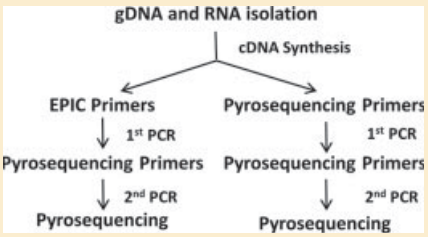
Genomic imprinting is an epigenetic mechanism that restricts gene expression to one inherited allele. Improper maintenance of imprinting has been implicated in a number of human diseases and developmental syndromes. Assays are needed that can quantify the contribution of each paternal allele to a gene expression profile. The authors have developed a rapid, sensitive quantitative assay for the measurement of individual allelic ratios termed Pyrosequencing for Imprinted Expression (PIE). Advantages of PIE over other approaches include shorter experimental time, decreased labor, avoiding the need for restriction endonuclease enzymes at polymorphic sites, and prevent heteroduplex formation which is problematic in quantitative PCR-based methods. The authors demonstrate the improved sensitivity of PIE including the ability to detect differences in allelic expression down to 1%. The assay is capable of measuring genomic heterozygosity as well as imprinting in a single run. PIE is applied to determine the status of *Insulin-like Growth Factor-2* (IGF2) imprinting in human and mouse tissues.



Development of Hematopoietic Stem and Progenitor Cells From Human Pluripotent Stem Cells

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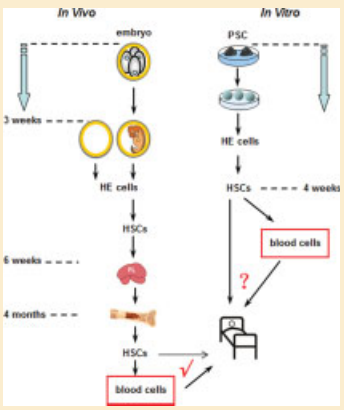
Human pluripotent stem cells (hPSCs), including human embryonic stem cells (hESCs) and human induced pluripotent stem cells (hiPSCs), provide a new cell source for regenerative medicine, disease modeling, drug discovery, and preclinical toxicity screening. Understanding of the onset and the sequential process of hematopoietic cells from differentiated hPSCs will enable the achievement of personalized medicine and provide an in vitro platform for studying of human hematopoietic development and disease. During embryogenesis, hemogenic endothelial cells, a specified subset of endothelial cells in embryonic endothelium, are the primary source of multipotent hematopoietic stem cells. The authors discuss current status in the generation of multipotent hematopoietic stem and progenitor cells from hPSCs via hemogenic endothelial cells. In addition, review the achievements in direct reprogramming from non-hematopoietic cells to hematopoietic stem and progenitor cells. Further characterization of hematopoietic differentiation in hPSCs will improve understanding of blood development and expedite the development of hPSC-derived blood products for therapeutic purpose.



Journal of Cellular Biochemistry

Flavonol Regulation in Tumor Cells

Michael A. Lea

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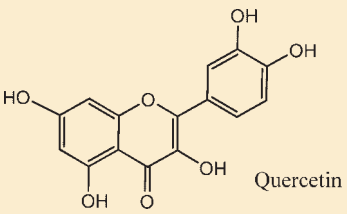

Flavonols comprise a group of flavonoid molecules that are widely distributes in fruits and vegetables. There is epidemiological data to suggest that consumption of flavonols can be accompanied by decreased cancer incidence. The anti-oxidant activity of flavonols may have an important role in preventing carcinogenesis. Therapeutic potential of flavonols is indicated by growth inhibitory action accompanied by a decrease in several hallmarks of cancer such as resistance to apoptosis. Multiple mechanisms of action have been reported for the action of flavonols on cancer cells. Particular emphasis has been directed to inhibitory effects on several protein kinases and on the potential for prooxidant effects. The diversity of actions presents a problem in trying to elucidate primary and secondary effects but it may be a strength of the therapeutic potential of flavonols that it renders development of resistance more difficult for cancer cells. Cancer chemotherapy is usually characterized by the use of drug combinations. Some additive or synergistic combinations have been identified for flavonols and is an area of ongoing investigation. As with other polyphenolic molecules there have been questions of cellular uptake and bioavailability. Several investigations have been and are being conducted to modify the structures of flavonols with the goal of increasing bioavailability. At present many investigators are sufficiently encouraged by past observations that they are responding to the challenge to optimize the dietary and therapeutic use of flavonols in cancer prevention and treatment.

Osteoporosis-Associated Alteration in the Signalling Status of BMP-2 in Human MSCs Under Adipogenic Conditions

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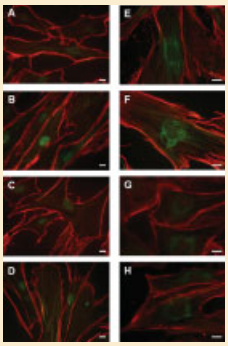

The authors studied bone morphogenetic protein-2 (BMP-2) signaling in mesenchymal stem cells (MSCs) obtained from bone marrow of control or osteoporotic volunteer postmenopausal women. MSCs were cultured under basal, adipogenic (AD) or AD plus BMP-2 conditions. The protein content of PPARγ, p-PPARγ, Runx2, bone morphogenetic receptor IA (BMPR IA), phosphorylated Smad-1/5/8 (p-Smad) and Smad 4 were determined by specific western blots. mRNA level for BMPRs was determined by PCR and cell localization of p-Smad-1/5/8 were detected by immunocytochemistry. Control MSCs showed a differential response to both AD and AD plus BMP-2 treatments: BMP-2 exerted an anti-adipogenic effect increasing both transcription factors analyzed. Moreover, p-Smads-1/5/8 were detected in nuclei after short term BMP-2 treatment. Osteoporotic MSCs showed no response to exogenous added BMP-2, as shown by p-PPARγ/PPARγ ratio and Runx2 levels, although BMPR-IA level was significantly higher in osteoporotic than in control MSCs. In addition, staining for p-Smad-1/5/8 in o-MSCs was observed around nuclei at all experimental conditions. Taken together results demonstrate failure of BMP-2 signaling in osteoporotic MSCs.