BD Accuri™ C6 and BD Stemflow™ Human Definitive and Pancreatic Endoderm Analysis Kit:

Monitoring human embryonic stem cell differentiation to definitive endoderm

This experimental data demonstrates the characterization of human embryonic stem cells (hESCs) differentiated to definitive endoderm using the BD Accuri™ C6 personal flow cytometer.

Experiment 1

H9 hESCs were differentiated to definitive endoderm in the presence of low serum and Activin A for three days. Cells were dissociated and analyzed for expression of key pluripotency (Nanog) and definitive endoderm (Sox17 and FoxA2) differentiation markers.

Data was collected and analyzed on a BD Accuri C6 personal flow cytometer. All data was gated based on the light scatter properties of H9 hESC derivatives.

Material

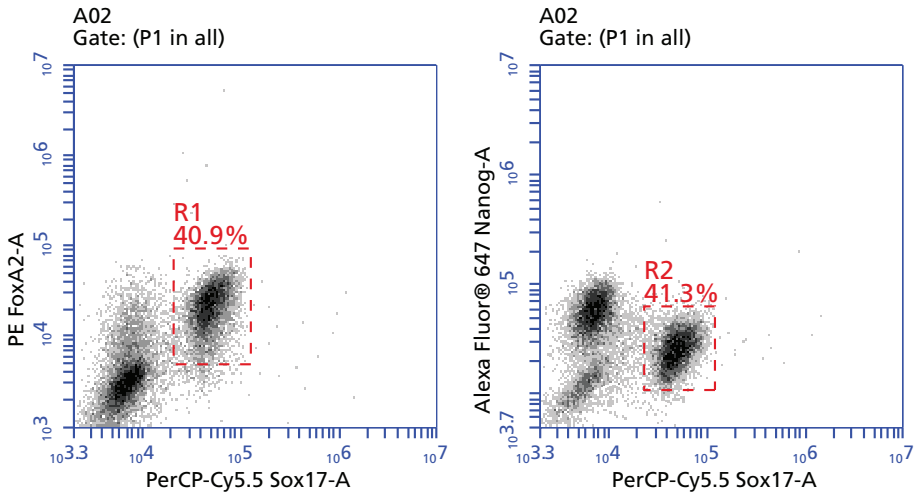
Reagents

BD Stemflow™ Human Definitive and Pancreatic Endoderm Analysis Kit including Pax-6, Sox2, Sox17, FoxA2, PDX-1, and Nanog markers (Cat. No. 562496)

Solution

BD™ Accutase Cell Detachment Solution (Cat. No. 561527)

Data



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Experiment 2

H9 hESCs were differentiated to definitive endoderm in the presence of low serum and Activin A for three days. Cells were then dissociated and analyzed for assessment of their apoptotic status (cleaved PARP–positive cells), DNA damage (pH2AX-positive cells), and proliferation status (BrdU-positive staining). Data was collected and analyzed on a BD Accuri C6 personal flow cytometer. All data was gated based on the light scatter properties of H9 hESC derivatives.

Material

Reagents

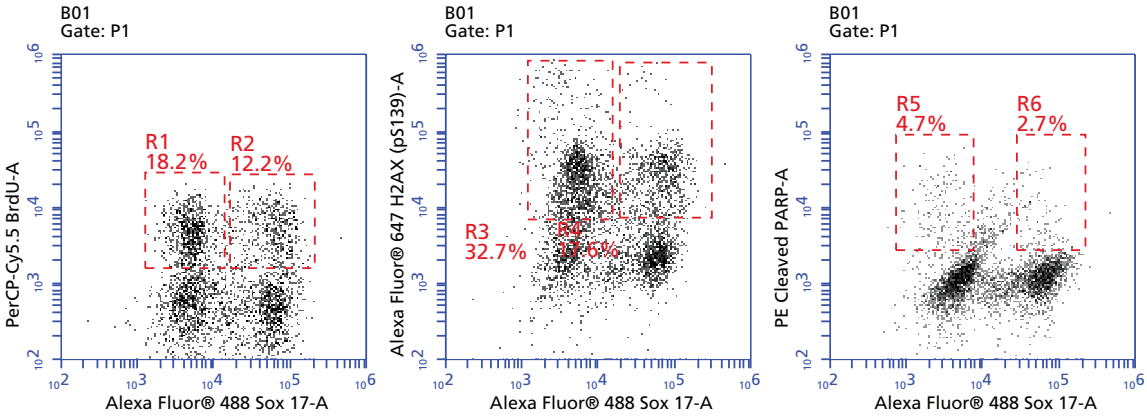
BD Pharmingen™ Apoptosis, DNA Damage, and Cell Proliferation Kit including BrdU, anti-BrdU, anti-cleaved PARP, and anti-phosphorylated H2AX (pS139) antibodies (Cat. No. 562253)

BD Pharmingen™ Alexa Fluor® 488 anti-Sox17 antibody (Cat. No. 562205)

Solution

BD Accutase Cell Detachment Solution (Cat. No. 561527)

Data



Discussion

The data demonstrates that rapid and multiparametric flow cytometry using the BD Accuri C6 personal flow cytometer and BD Stemflow kits is effective for characterization of stem cell differentiation to pancreatic and definitive endoderm.

Reference

D'Amour KA, Agulnick AD, Eliazer S, Kelly OG, Kroon E, Baetge EE. Efficient differentiation of human embryonic stem cells to definitive endoderm. *Nat Biotechnol.* 2005;23:1534-1541.

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