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Co-culture and transduction of murine thymocytes on Delta-like 4-expressing stromal cells: A method to study oncogenes in T-cell leukemia

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Protocol status: Working
We use this protocol and it's working

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Keywords: transducing primary thymocyte, transduction of murine thymocyte, cultivating hematopoietic progenitor cell, generating transgenic mice, dl4 stromal cell, dl4 cell, hematopoietic progenitor cell, murine thymocyte, transgenic mice, thymocyte, retroviral transduction, primary thymocyte, immature thymocyte, expressing stromal cell, leukemogenesi, expressing bone marrow, transduced mouse, dividing cell, stromal cell, different types of cell, cell leukemia, cell, specific gene, bone marrow

Abstract

Transduced mouse
immature thymocytes can be differentiated into T cells *in vitro* using
the delta-like-4-expressing bone marrow
stromal cell line co-culture system (OP9-DL4). As retroviral transduction requires dividing cells for transgene integration, OP9-DL4 provides a suitable *in vitro* environment for
cultivating hematopoietic progenitor cells. This is particularly advantageous
when studying the effects of expression of a specific gene during normal T-cell
development and leukemogenesis, as it allows researchers to circumvent the time-consuming
process of generating transgenic mice. To achieve successful outcomes, a
series of coordinated steps involving the simultaneous manipulation of
different types of cells must be carefully performed. Although these are very well-established procedures, the lack of a common source in the literature often demands a series of
optimizations, which can be time-consuming. This protocol has been shown to be
efficient in transducing primary thymocytes followed by differentiation on
OP9-DL4 cells. Detailed here are the steps that can serve as a quick and
optimized guide for the co-culture of retrovirally transduced thymocytes on
OP9-DL4 stromal cells.

Attachments

DOCX

Protocol_Co-culture ...

22.6MB

Troubleshooting