

Determining the plasticity of CD34 expression

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Rationale:

Previously, we established that OCI-AML-20 requires stroma to maintain CD34 expression. In this experiment, we wanted to see whether those cells grown in suspension are able to regain CD34 expression when cells are reintroduced to stroma.

Methods:

OCI-AML-20 cells that were used in previous experiment (<https://zenodo.org/record/1187083#.Wr5-1C7wbIV>) were collected. Cells previously grown in suspension were plated at a density of 125K/well of a 12-well plate with either MS5, OP9 or no stroma (suspension).

After 1 week and two weeks, cells were collected, stained and analyzed by flow cytometry as previously indicated (link above).

Results and conclusions:

Cells that were grown in suspension for 3 weeks continue to show very low number of CD34 positive cells (Figure 1). OCI-AML-20 cells that had previously grown in suspension were able to regain some CD34 expression in the presence of stroma but not to the levels seen for cells grown continuously on stroma (Figure 1).

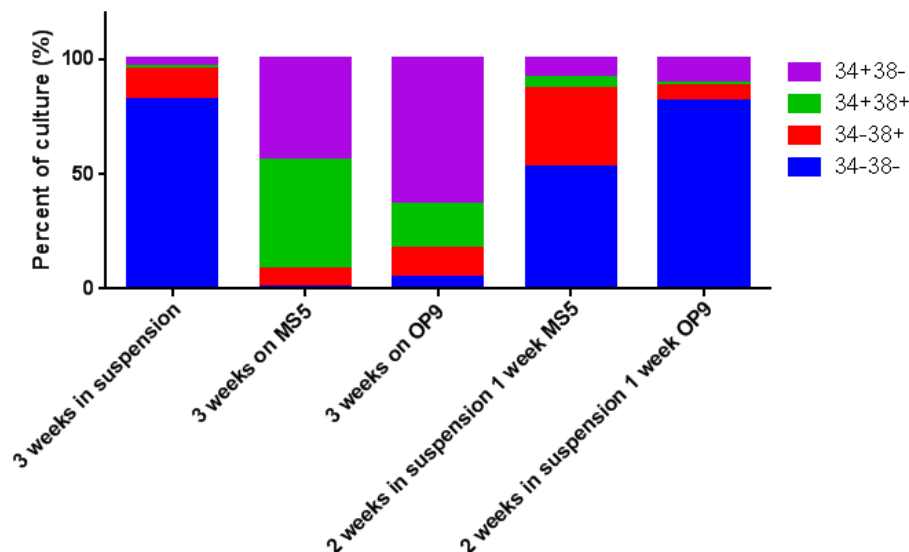


Figure 1: Summary of flow cytometry analysis of cells at 3 weeks. Cells that were transferred from suspension to stroma regain CD34 expression after 1 week, but not to the level seen with cells continuously grown on stroma.

Cells were followed for an additional week. At 4 weeks, there were very low numbers of viable suspension cells to perform flow cytometry and therefore, they were not included in the analysis. OCI-AML-20 cells that were previously grown in suspension and transferred to stroma, had regained even more CD34 expression after 2 weeks on stroma (Figure 2). The levels of CD34⁺CD38⁻ were very similar to cells grown on stroma for the whole 4 weeks at this point (Figure 2).

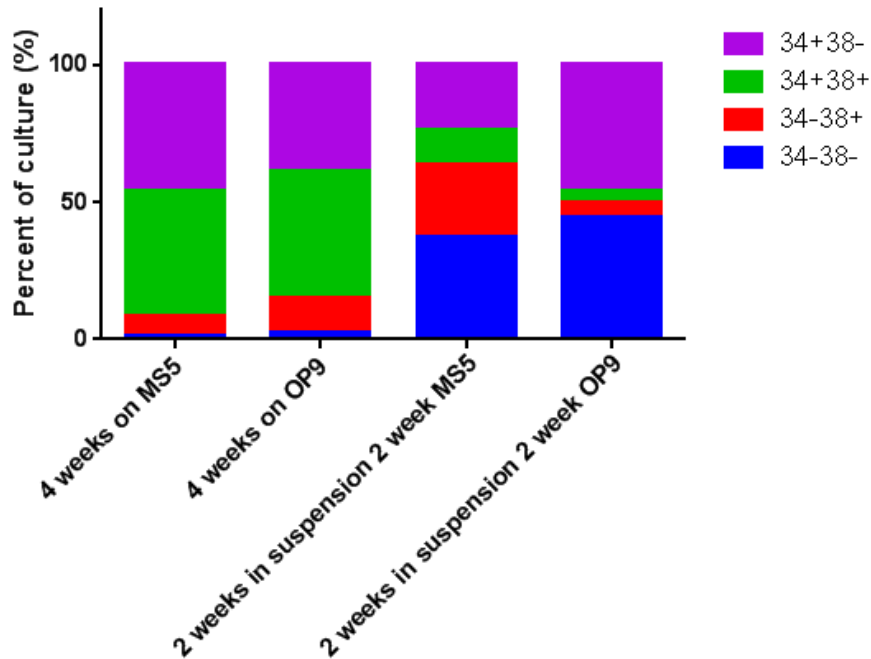


Figure 2: Cells were followed for an additional week (there were too few viable suspension cells at 4 weeks to do flow cytometry analysis)

In conclusion, OCI-AML-20 cells display plasticity in CD34 expression.