## Culture of MEL murine cell lines

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## Obtaining cell lines:

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Mouse erythroleukemia (MEL), derived from a B-cell lymphoma, are rapidly dividing murine cell lines that are maintained in suspension cultures and grow as loose clumps. Cells should be maintained at $37^{\circ} \mathrm{C}$ with $5 \% \mathrm{CO}_{2}$ at a density between $1 \times 10^{5}$ and $1 \times 10^{6}$ cells $/ \mathrm{mL}$. Cultures should be split $1: 8-10 \sim$ every two days to maintain this concentration.

MEL growth medium: 10\% HI-FBS(heat-inactivated), 1\% Pen-strep(Penicillin and Streptomycin), RPMI 1640 with L-Glutamine;
MEL freezing medium: $90 \% \mathrm{HI}-\mathrm{FBS}, 10 \%$ fresh DMSO, freeze at a concentration of $1 \times 10^{7}$ cells $/ \mathrm{ml}$.

## Starting cultures from frozen stocks:

1. Thaw cell vials at $37^{\circ} \mathrm{C}$.
2. Add to 15 mL tube with 10 ml fresh growth medium, centrifuge $1500 \mathrm{rpm} /$ 3 min ., and discard supernatant.
3. Resuspend cells in 10 ml fresh growth medium, add to small flask and incubate in $5 \% \mathrm{CO}_{2} / 37^{\circ} \mathrm{C}$.
4. Cells can normally be split $\sim 1: 8$ after 2 days and maintained as described above.

DMSO-induction of MEL cells: To induce erythroid differentiation, MEL cells are treated with $2 \%$ DMSO. DMSO-induction of differentiation slows growth so that cells only need to be split 1:2 at day 2 or 3 after induction. Differentiating cells will become noticeably red when pelleted by 2-3 days after induction and color will increase in following days.

