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## CLUMPING DIGESTION with DNase FOR CELL SORTING

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Sort purity can be reduced when sorted target cells are attached to non-target cells, as well as poor recovery when coincident aborts exclude all clumped cells. DNA from lysed cells in the medium can cause cells to clump.

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### Procedure

1. Treat cells for 15 to 30 minutes in a solution of 100 µg/mL DNase and 5 mM MgCl<sub>2</sub> in HBSS at room temperature.
2. Wash the cells with 5 mM MgCl<sub>2</sub> in HBSS.
3. Suspend the cells with Staining Buffer (PBS + 1%BSA) containing MgCl<sub>2</sub> and 25-50 µg/mL DNase as a maintenance dose during the sort.
4. Filter sample through standard FCM tube with cell-strainer cap.

### Reagents

- DNase (Sigma D-4513) 100 µg/ml in Hank's Balanced Salt solution (HBSS)
- Magnesium chloride hexahydrate (Sigma M-2670) MW=203.3 (203 mg/ml = 1000 mM or 200X )

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### References

Crissman, HA, Mullaney, PF, and Steinkamp, JA. *Methods and applications of flow systems for analysis and sorting of mammalian cells. Meth. Cell Biol.* 9:175 (1975).

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