

Brown Flow Cytometry Facility

Cell Sorting Guidelines

Cell Sorting:

- Sorting is done by prior appointment only. Due to set up time required, please explain your needs thoroughly when scheduling an appointment. A completed Cell Sort Request Form must be submitted before your appointment will be confirmed. Scheduling generally should be done about one week in advance.
- Sorting hours are from 10:00 A.M. to 4:30 P.M. Monday through Friday. Extended hours (early or late time) can be arranged at the time of initial scheduling.
- Any cancellations with less than a 24 hour notice before scheduled start time will be charged 1 hour. No-shows will be charged for time reserved.
- All users must provide an account for billing at time of scheduling. Users will be billed monthly.
- Scheduling is on a first come, first served basis.

SORT SAMPLE PREPARATION GUIDELINES:

- Do not fill sample tubes over 2/3 full.
- Suspend cells in PBS at 5×10^6 per ml - 1×10^7 per ml.
- Sample cell concentration affects sort speed.
 - Use the highest concentration possible while maintaining a single cell suspension.
 - We can dilute samples at sorting facility but cannot concentrate them.
- **Control Samples:** For all sort operations a negative or unstained control and a separate single stain control sample tube for each stain to be used is required for instrument pre-sort calibration. *(ie: if you are using FITC, PE, PEcy7, APC, a control sample of each is required for calibration and compensation of instrument. In this case, 5 control samples total including unstained cell sample)*
- **GFP sort** requires only a negative or not-transfected (not-transduced) control sample.
- Control samples should be prepared the same as the sort sample from the

same type of cells as experiment samples.

- Each control sample should contain a minimum of around 200,000 cells.

COLLECTION TUBE PREPARATION:

- Pre-coating tubes with serum or media/buffer is recommended. Collection media/buffer can also be added to the tubes (no more than 1 ml).
- Populations can be simultaneously be sorted into: 12 X 75mm tubes or 17 X 120mm tubes or 15 ml tubes

SINGLE CELL SORTING:

- Single cells can be sorted at 1 – 50,000 cells into 6 to 96 well micro titer plates or onto a slide.
- Plates must be pre-filled (100 microliters of complete medium in 96 well plate).

For any further questions regarding sorting, please contact us:

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