Nemours | Alfred I. duPont Hospital for Children
Nemours Children's Clinic

BCL

Biomolecular Core Facility

AI Dupont Hospital for Children, Rockland Center One, Room 214
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310 - DNA Fragment Analysis Submission Protocol

Currently used for quantative analysis, mutation detection and genotyping for linkage analysis. Researchers are asked to provide a fluorescently labeled diluted PCR reaction along with a submission sheet to the core for processing.

Sample sign-up

Contact the Core (302-651-6712) the day before you would like your samples to be run. For capillary electrophoresis, each sample takes approximately 30 minutes to run. If the instrument is full for that day, your samples will be placed in a queue for overnight or next day processing. Advance notification is required to ensure availability of the machine. This is a first-come, first-serve service.

DNA Prep

Routinely, each researcher performs the PCRs using fluorescently labeled primers. Before submitting to the core, the PCR products are diluted to an appropriate concentration determined by the user. A good start is to prepare a 1:20 dilution in a clean eppendorf tube. The core will typically perform electrophoresis on 2 ul of your diluted sample. If performing multiplex reactions: mix FAM, HEX, or NED labeled PCR products in a 1FAM : 2HEX : 2NED ratio. The internal control dye for FAM, HEX and TET will be TAMRA. The internal control dye for FAM, HEX and NED will be ROX.

Sample Submission and Turnaround Time

Diluted samples should be protected from light and placed in the -20^{o} Core freezer in G25 A/R building by 1:30 PM or room 214 Rockland Center One by 2:30 PM. Place your submission sheet in the CE inbox. Our normal fragment analysis processing is overnight. We will notify you via phone or email the next morning when your run is complete.

Same day processing is available by advance arrangement only and staff and time permitting: A maximum of 8 samples can be run during the day. Samples should be submitted by 10:00 AM. It is the responsibility of the researcher to deliver the samples to the Core lab. Your results will be ready for you by 4:00 PM that afternoon. Any re-runs may delay your results until the next business day.

Please note: Any samples arriving after the submission deadline will be processed as time/work load permitting and may be subjected to a delay (see Avoiding delays below). The Core may grant exceptions with advance notification only.
Avoiding delays: Since others may have samples to run after yours, we can only guarantee a spot for the exact number of samples that you originally reserved (i.e. if you

reserved 10 samples for Monday, we can only guarantee that 10 will be run on Monday). All additional samples will be accommodated as room permits. In addition, if you reserved space on Monday and your samples don't arrive until Tuesday, they will be placed at the bottom of the order on Tuesday. This is meant to provide fairness and consideration to others who have already signed up for this service.

Data

You will receive a copy of the electropherogram for each of your samples. In addition, a Genotyper file will be created and saved in your folder on the core Mac or in your shared folder on the medsci network. The Genotyper program is available on several Core Mac computers. All customers are welcome to use them. In addition, we can make Genotyper available for labs to download to one of their own Mac computers. There are system requirements that need to be met. Please contact the Core Assistant Director, Jennifer Holbrook (x6712) for assistance.

The DNA Fragment Analysis Core offers several levels of services depending on the needs of the researchers. Please consult with the core director, Dr. Katia Sol-Church for the best fit for your experimental design. High throughput genotyping may require the need for robotics set-up. We also offer genotyping software training to anyone interested.

Fee

The cost per injection is $5.00 and will be billed to your Lawson activity number on a quarterly basis. For high volume discount, please contact the Director, Dr. Katia Sol-Church. All non-AIDHC affiliated customers will be charged a 10% surcharge.