

through the DOE genome center, is designed to run 384 lanes simultaneously with a low-viscosity sieving medium. The entire system would be loaded automatically, run, and set up for the next run at 3-hour intervals. If successful, it should provide a 20- to 40-fold increase in throughput over existing machines.

An LLNL-designed high-precision spotting robot, which should allow a density of 98,304 spots in 96 cm², is now operating. The goal of this effort is to create high-density filters representing a 10× BAC coverage of both human and mouse genomes (30,000 clones = 1× coverage). Thus each filter would provide ~3× coverage, and eight such filters would provide the desired coverage for both genomes. The filters would be hybridized with amplicons from individual or region-specific cDNAs and ESTs; given the density of the BAC libraries, clones that hybridize should represent a binned set of BACs for a region of interest. These BACs could be the initial substrate for a BAC sequencing strategy. Performing hybridizations in parallel in mouse and human DNA facilitates the development of the mouse map (with ORNL involvement), and sequencing

BACs from both species identifies evolutionarily conserved and, perhaps, regulatory regions.

Information generated by sequencing human and mouse DNA in parallel is expected to expand LLNL efforts in functional genomics. Comparative sequence data will be used to develop a high-resolution synteny map of conserved mouse-human domains and incorporate automated northern expression analysis of newly identified genes. Long range, the center hopes to take advantage of a variety of forms of expression analysis, including site-directed mutation analysis in the mouse.

Summary

The Livermore Human Genome Center has undergone a dramatic shift in emphasis toward commitment to large-scale, high-accuracy sequencing of chromosome 19, other chromosomes, and targeted genomic regions in the human and mouse. The center also is committed to exploiting sequence information for functional genomics studies and for other programs, both in house and collaboratively.

