

**1999-2000  
RESEARCH CYTOMETRY LABORATORY  
SUMMARY OF ACTIVITIES**

The Research Cytometry Laboratory (RCL) provides a wide range of cell-analysis techniques including three-color immunophenotyping, apoptosis quantitation, GFP detection, cell sorting and cell cloning, immunofluorescence detection of intracellular markers, confocal fluorescence imaging and 3-D reconstruction, DNA (ploidy) analysis, and cellular activation studies such as kinetics analysis of intracellular Ca<sup>++</sup> and pH modulation.

Funding of an NIH Shared Instrument Grant application this year provided funds for the purchase of a new confocal laser scanning microscope, a Leica TCS SP2. Other instrumentation in the Laboratory includes a Becton Dickinson FACSVantage SE flow cytometer, a Photon Technology Incorporated (PTI) M-Series fluorescence detection system, and a Molecular Devices Cytosensor microphysiometer.

<u>Laboratory Utilization</u> (through Aug 28)	<u>1999-2000</u>	<u>1998-1999</u>
FACS (samples)	1931	870
ACAS (hours)	226	267
Cytosensor (experiments)	7	53
PTI (hours)	14	8

Twenty-five investigators representing the following nine departments used the services of the RCL during 1999-2000: Biochemistry and Molecular Biology, Medicine, Microbiology and Immunology, Pathology, Pediatrics, Pharmacology, Physiology, Cell Biology and Neuroscience, and Orthopaedics.

**Director's Evaluation of the Laboratory**

Overall activity was up approximately 25%, compared to last year, based on the dollar amount of invoices for services rendered with this increase due mostly to the use of our new flow cytometer. Likewise, addition of the new Leica confocal laser scanning microscope (October 1, 2000) is expected to increase image analysis revenue and further boost production of the laboratory.

**Future Directions**

New directions will focus mainly on image analysis as a result of the acquisition of the new Leica confocal system. This instrument has the capability of acquiring three colors simultaneously at frame rates of up to 25 fps. The TCS will have three lasers providing investigators with a wide range of possibilities in their selection of fluorochromes. DIC/fluorescence image overlays, full-field fluorescence viewing, and the unique capabilities provided by an upright microscope are all new additions to the Research Cytometry Laboratory.

**Extramural Support****Active**

NIH DRR/BRS Shared Instrumentation Grant  
"Leica TCS SP Spectral Confocal Microscope"  
1 S10 RR13732-01A1

R.B. Hester, P.I.

9/01/2000 to 8/31/2001

\$336,843.

**Publications**

W. Radding, S.E. Jordan, R.B. Hester, and H.C. Blair. Intracellular Calcium Puffs in Osteoclasts. *Exp. Cell Res.* 253:689-696 (1999).