LAH 405

FOREWORD TO THE THIRD EDITION

This is a light-hearted and very useful book on a complex but very widely used technology. When we had in hand the first working model of a fluorescence-activated cell sorter in 1969, we expected that the major application would be the sorting of live fluorescently stained cells to obtain pure populations of cells that would then be further analyzed off-line. However, implicit in the technology was the on-line analytical capability, so nicely described in this book by Howard Shapiro.

The first two editions from Dr. Shapiro's prolific pen maintained the fiction that working scientists, especially biologists and medically oriented scientists, would build their own flow cytometers. In this Third Edition, Shapiro has bowed to practical reality, and does not predicate this excellent text on flow cytometry and sorting on the "Cytomutt" and improvements as he had in the First Edition, but continues the tradition he started in the Second Edition of presenting the principles of modern multiparameter analysis (importantly, explaining what a parameter is), so that working biomedical scientists can understand how to get the best machines for their money, how to evaluate capabilities of these machines, how and where errors can come in, how to use the instruments most effectively in their important biomedical experiments, and, finally, how some technologically-minded folk are trying to advance the art of flow cytometry and sorting.

This technology has spawned an estimated four hundred million dollars a year in sales of instruments and reagents in 1994. More than 900 participants, mostly machine operators, engineers, technological buffs, staff members of principal investigators, and a relative few of the principal investigators themselves, are expected to attend the International Society for Analytical Cytology's meeting in Lake Placid in the Fall of 1994. Also present will be many members of the large and small companies that hope to provide the material base for this field. Rubbing elbows at this meeting will be immunologists, both basic and clinical, oncologists and cell biologists, as well as molecular biologists, AIDS specialists (and activists), pharmacologists, and too many types of flow cytometrists to name in this Foreword. Nevertheless, all will find information that interests and helps them throughout this book.

Highly capable computers are vitally important components that must be included in modern cell analysis and sorting. Two-parameter analysis is the minimum that any flow cytometer offers. Three, four, and five fluorescence parameters are available from the major producers in 1994. Six, seven, and even ten such parameters are available on some experimental machines being put into practical this same year. Soon thereafter, there may be considerably more than ten measurement parameters on the more advanced instruments.

Consider the data taken at the Stanford Shared FACS Facility in mid-1994 as typical of a heavily used multi-user flow cytometry center. About 125 experiments are analyzed per week, averaging 30 samples per experiment, or 3,750 samples/week. This requires approximately 300 megabytes (30,000 cells, 6 measurements, and 9 bit resolution produce about 200 kilobytes/sample). Thus, we must store 15 gigabytes of new data per year. This creates a need for very

extensive and sophisticated means of data management, retrieval, and analysis.

We soon will have all these many gigabytes of data available on-line, with access to investigator names, dates, experimental parameters, etc.; all of the ancillary information needed for analysis of the accumulated data from current as well as previous experiments will be easily accessible to the investigator.

In order for all this data to be meaningful, excellent standardization, compensation, and stability of measurement will have to be featured in the specifications of all serious machines. This is done now in the Shared FACS Facility at Stanford and should be done everywhere flow cytometry is used.

Shapiro covers many technical and scientific considerations in this excellent book and, as I said, treats them with light-hearted humor. Take, for example, "Flow's Golden Oldies" as a heading on page 35, or aphorisms like "Shapiro's First Law of Flow Cytometry: A 51 μm Particle CLOGS a 50 μm Orifice", on page 16.

I recommend a thorough reading for all who are using and plan to use flow cytometry in analysis and sorting of cells and other biological particles.

Leonard A. Herzenberg
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