

PROPOSED GATING STANDARD FOR FLOW CYTOMETRY

Josef Spidlen¹, Robert Gentleman², Perry Haaland³, Michael F. Ochs⁴, Charles Schmitt⁵, Clayton Smith⁶, Adam S. Treister⁷, Ryan R. Brinkman⁸

¹British Columbia Cancer Research Centre, Terry Fox Laboratory, Vancouver, British Columbia, Canada;

²Fred Hutchinson Cancer Reserch Center, Seattle, Washington;

³BD ViaSante, Carrboro, North Carolina;

⁴Fox Chase Cancer Center, Philadelphia, Pennsylvania;

⁵BD Technologies, Research Triangle Park, North Carolina;

⁶Terry Fox Laboratory, BC Cancer Research Centre, Vancouver, British Columbia, Canada;

⁷Tree Star, Inc., Redwood City, California;

⁸British Columbia Cancer Research Centre, Vancouver, British Columbia, Canada.

Abstract.

Gating in flow cytometry is a well known and highly important process for selecting populations of interests by defining the characteristics of particles for further data acquisition or analysis. It may also be used for sorting purposes, e.g., for distinguishing among multiple heterogonous populations in a single sample.

Although flow cytometry has a successful data format standard (FCS files), there is no shared representation of gates. This prevents a variety of collaborative opportunities to recreate experimental methods and results.

Several partners from academy as well as from industry joined within the project NIH R01 EB-5034 to collaborate on the development of data standards in flow cytometry.

We have developed a proposal on how to form XML-based gate definitions that can facilitate the interchange and validation of data between different software packages. It currently consists of four parts as follows:

1. A detailed description of the gating specification.
2. A W3C schema document defining the syntax of XML documents describing gates. The schema can also be used to validate them.
3. A comprehensive user documentation presenting the schema in a lucid manner.
4. A set of examples of gating XML files.

The specification currently supports rectangular gates in n dimensions (i.e., from 1D range gates up to n -dimensional hyper-rectangular regions), polygon gates in 2 (and more) dimensions, ellipsoid gates, decision tree structures, and Boolean collections of the any of the types of gates. We also provide mechanisms to link the gating file to a particular FCS file, to express transformations of dimensions, or to express fuzzy gates.

Moreover, we are preparing a platform independent open source software tool that is capable of reading gate definition files, applying them to specified FCS data files, and providing common descriptive statistics for selected parameters. We plan to release this tool along with the gating specification in order to provide a freely available reference implementation for independent software developers.

The draft of the gating standard has progressed through several iterations among the project collaborators and is now being presented to the broader ISAC community with a kindly request for comments. The most up-to-date version can be downloaded from the project web site at www.flowcyt.org.