Evaluation of novel high sensitivity fluorescent reporters in multiparameter flow cytometry

Brilliant Violet™ Reporters

Brilliant Violet™ Reporters are a unique class of fluorescent dyes for biological detection. These dyes are characterized by their high brightness and spectral properties, which make them exceptionally useful in flow cytometry.

Methods

Brilliant Violet™ Reporters are uniformly conjugated to monoclonal antibodies using standard cromatochemistry. Resulting antibodies were purified by size exclusion chromatography and ion exchange chromatography. Geometric nanosizes were validated by titration at different concentrations for antigen receptor expression in order to determine optimal parameters for flow cytometry.

Initial experiments were conducted to assess the performance of Brilliant Violet™ when using a standard method. The highly sensitive marker of CD4 was selected and the commonly used fluorochrome, Pacific Blue™, was chosen as a performance control (Figure 1). Figure 1 illustrates that non-specific binding with Brilliant Violet 421™ RPA-T4 is markedly lower than with Pacific Blue™. Staining with Brilliant Violet™ showed significantly higher MFI and stain indices than Pacific Blue™. Discrimination between positive and negative populations was also markedly clearer. Once again Brilliant Violet™ had a significantly higher brightness than Pacific Blue™.

Brilliant Violet 421™ is well matched to the optics of commercially available infrared lasers. We have developed an exceptionally bright novel polymeric fluorescent dye, Brilliant Violet™, which was designed to be used in conjunction with high sensitivity detectors for flow cytometry. These will substantially increase the opportunities for their use in flow cytometry. These will substantially increase the opportunities for their use in flow cytometry.

Results

Initial experiments were conducted to assess the performance of Brilliant Violet™ 613™ when using a standard method. The highly sensitive marker of CD4 was selected and the commonly used fluorochrome, Pacific Blue™, was chosen as a performance control (Figure 2). Figure 2 illustrates that non-specific binding with Brilliant Violet 613™ RPA-T4 is markedly lower than with Pacific Blue™. Staining with Brilliant Violet™ showed significantly higher MFI and stain indices than Pacific Blue™. Discrimination between positive and negative populations was also markedly clearer. Once again Brilliant Violet™ had a significantly higher brightness than Pacific Blue™.

Brilliant Violet 613™ is well matched to the optics of commercially available infrared lasers. We have developed an exceptionally bright novel polymeric fluorescent dye, Brilliant Violet™, which was designed to be used in conjunction with high sensitivity detectors for flow cytometry. These will substantially increase the opportunities for their use in flow cytometry. These will substantially increase the opportunities for their use in flow cytometry.