426k Blue Fluorescent Protein Variants with Enhanced Brightness from a Computationally Designed Library

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The use of both computational design and library screening for obtaining proteins with new or improved function has increased dramatically in recent years. Computational design was applied to guide the construction of a library of mutants of the blue fluorescent protein (BFP). This library was constructed using gene assembly and screened using fluorescence activated cell sorting (FACS). Twenty variants with improved whole-cell brightness relative to the commonly-used Y66H, Y154F mutant were isolated. The brightest clones were further characterized to correct for differences in expression, and in some cases, showed improved fluorescence relative to the parental BFP. This study highlights the importance of coupling library design with screening approaches to improve protein properties.