Standardizing Application Setup Across Multiple Flow Cytometers Using BD FACSDiva™ Version 6 Software

Ellen Meinelt, Mervi Reunanen, Mark Edinger, Maria Jaimes, Alan Stall, Dennis Sasaki, Joe Trotter

**Optical Configuration**

<table>
<thead>
<tr>
<th>Laser</th>
<th>PMT</th>
<th>LP Mirror</th>
<th>BP filter</th>
<th>fluorochromes</th>
</tr>
</thead>
<tbody>
<tr>
<td>355 nm</td>
<td>A</td>
<td>770</td>
<td>820/60</td>
<td>UV 780/60</td>
</tr>
<tr>
<td>B</td>
<td>690</td>
<td>740/35</td>
<td>UV 740/35</td>
<td>BV737</td>
</tr>
<tr>
<td>C</td>
<td>630</td>
<td>670/25</td>
<td>UV 670/25</td>
<td>BV661</td>
</tr>
<tr>
<td>D</td>
<td>660</td>
<td>615/44</td>
<td>UV 615/24</td>
<td>BV595</td>
</tr>
<tr>
<td>E</td>
<td>550</td>
<td>580/20</td>
<td>UV 580/20</td>
<td>BV563</td>
</tr>
<tr>
<td>F</td>
<td>450</td>
<td>515/30</td>
<td>UV 515/30</td>
<td>BV496</td>
</tr>
<tr>
<td>G</td>
<td>410</td>
<td>450/50</td>
<td>UV 450/50</td>
<td>DAPE, V550</td>
</tr>
<tr>
<td>H</td>
<td>370</td>
<td>379/28</td>
<td>UV 379/28</td>
<td>BV395</td>
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</table>

**405 nm**

<table>
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<th>BP filter</th>
<th>fluorochromes</th>
</tr>
</thead>
<tbody>
<tr>
<td>violet</td>
<td>A</td>
<td>770</td>
<td>800/30</td>
<td>Vis 800/30</td>
</tr>
<tr>
<td>B</td>
<td>735</td>
<td>750/35</td>
<td>Vis 750/35</td>
<td>BV750</td>
</tr>
<tr>
<td>C</td>
<td>685</td>
<td>710/20</td>
<td>Vis 710/20</td>
<td>BV711</td>
</tr>
<tr>
<td>D</td>
<td>640</td>
<td>670/25</td>
<td>Vis 670/25</td>
<td>BV660</td>
</tr>
<tr>
<td>E</td>
<td>590</td>
<td>590/20</td>
<td>Vis 590/20</td>
<td>BV580</td>
</tr>
<tr>
<td>F</td>
<td>550</td>
<td>585/15</td>
<td>Vis 585/15</td>
<td>BV570</td>
</tr>
<tr>
<td>G</td>
<td>505</td>
<td>525/30</td>
<td>Vis 525/30</td>
<td>BV510 or BV480</td>
</tr>
<tr>
<td>H</td>
<td>410</td>
<td>431/28</td>
<td>Vis 431/28</td>
<td>BV422, e450, CFP</td>
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</tbody>
</table>

**488 nm**

<table>
<thead>
<tr>
<th>Laser</th>
<th>PMT</th>
<th>LP Mirror</th>
<th>BP filter</th>
<th>fluorochromes</th>
</tr>
</thead>
<tbody>
<tr>
<td>blue</td>
<td>A</td>
<td>770</td>
<td>820/60</td>
<td>Blue 820/60</td>
</tr>
<tr>
<td>B</td>
<td>735</td>
<td>750/35</td>
<td>Blue 750/35</td>
<td>BV735</td>
</tr>
<tr>
<td>C</td>
<td>685</td>
<td>710/20</td>
<td>Blue 710/20</td>
<td>BV711</td>
</tr>
<tr>
<td>D</td>
<td>640</td>
<td>670/25</td>
<td>Blue 670/25</td>
<td>BV660</td>
</tr>
<tr>
<td>E</td>
<td>590</td>
<td>590/20</td>
<td>Blue 590/20</td>
<td>BV580</td>
</tr>
<tr>
<td>F</td>
<td>505</td>
<td>530/15</td>
<td>Blue 530/15</td>
<td>BV515, FITC, AF488, GFP</td>
</tr>
<tr>
<td>G</td>
<td>-</td>
<td>488/10</td>
<td>555</td>
<td>SSC</td>
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</table>

**561 nm**

<table>
<thead>
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<th>PMT</th>
<th>LP Mirror</th>
<th>BP filter</th>
<th>fluorochromes</th>
</tr>
</thead>
<tbody>
<tr>
<td>yellow/green</td>
<td>A</td>
<td>750</td>
<td>780/60</td>
<td>YG 780/60</td>
</tr>
<tr>
<td>B</td>
<td>635</td>
<td>670/30</td>
<td>YG 670/30</td>
<td>PE-Cy5, 7-AAD, BV6790</td>
</tr>
<tr>
<td>C</td>
<td>600</td>
<td>610/30</td>
<td>YG 610/30</td>
<td>PE TexasRed, PE-Cy594, PI, mCherry, RFP</td>
</tr>
<tr>
<td>D</td>
<td>570</td>
<td>585/15</td>
<td>YG 585/15</td>
<td>PE, DilRed, Cy5, BV5668</td>
</tr>
</tbody>
</table>

**637 nm**

<table>
<thead>
<tr>
<th>Laser</th>
<th>PMT</th>
<th>LP Mirror</th>
<th>BP filter</th>
<th>fluorochromes</th>
</tr>
</thead>
<tbody>
<tr>
<td>red</td>
<td>A</td>
<td>750</td>
<td>780/60</td>
<td>Red 780/60</td>
</tr>
<tr>
<td>B</td>
<td>690</td>
<td>730/45</td>
<td>Red 730/45</td>
<td>APC-R700, AF700</td>
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<tr>
<td>C</td>
<td>655</td>
<td>670/30</td>
<td>Red 670/30</td>
<td>APC, AF647, e760</td>
</tr>
</tbody>
</table>

**Optimum PMT Voltage**

Optimal settings based on PBMCs using BD CS&T bright bead target values for each fluorescence detector, to obtain consistent and reproducible results over time.

**Compensation table**

**Spread table**

Reference: Standardizing Application Setup Across Multiple Flow Cytometers Using BD FACSDiva™ Version 6 Software

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