

These recommended voltages are a good starting point. If your single stain control is too bright, the voltage should be lowered. In general, increasing the voltage above the recommended settings has no benefit.

### Fortessa X20

Laser	Laser	Filter	Fluorophore tested	Starting voltage
UV	355	379/28	BUV396	450
UV	355	450/50	DAPI	458
UV	355	515/30	BUV496	600
UV	355	740/35	BUV737	600
UV	355	820/60	BUV805	600
Violet	405	450/50	BV421	450
Violet	405	525/50	BV510	500
Violet	405	610/20	BV605	600
Violet	405	710/50	BV711	600
Violet	405	780/60	BV786	514
Blue	488	530/30	FITC	500
Blue	488	710/50	PerCP-Cy5.5	600
Yellow/Green	561	586/15	PE	500
Yellow/Green	561	610/20	PE-Dazzle595	450
Yellow/Green	561	780/60	PE-Cy7	450
Red	640	670/30	APC	500
Red	640	710/50	APCR700	600
Red	640	780/60	APC-Cy7	600

Fluorophore tested	Separation Index	Brightness Estimation
APC	223	Very Bright
PE	222	Very Bright
PE-Dazzle595	214	Very Bright
BV421	198	Very Bright
PE-Cy7	181	Bright
BV786	178	Bright
BV711	172	Bright
BV605	135	Moderate
PerCP-Cy5.5	113	Moderate
BUV737	104	Moderate
AF700	86	Dim
BV510	85	Dim
AF488	85	Dim
BUV396	71	Dim
APC-Cy7	71	Dim
Pacific Blue	61	Dim
BUV496	33	Very Dim
BUV805	30	Very Dim
DAPI	not tested	