

Healthy Cells Broad Capabilities Better Science

Gentle, customizable benchtop microfluidic cell sorting.





High cell integrity without compromise •••

The WOLF was originally created by a team of scientists and engineers who wanted to solve a classic challenge in biological research: how to sort cells effectively, easily, and of high quality. Now, as science continues to move forward into increasingly complex realms, the WOLF is moving forward with it. The WOLF G2 instrument has significantly expanded the capabilities of gentle benchtop microfluidic cell sorting with two lasers and up to nine colors, while maintaining simple workflows for either bulk sorting or singlecell dispensing. Single-cell sorting can be completed in 96- or 384-well plates when using the WOLF G2 in conjunction with the N1 Single-Cell Dispenser. This flexibility in performance, along with the additional abilities of the second laser, makes it ideal for use in many different research fields and application areas like single-cell genomics, cell line development, gene editing, antibody discovery, immunology, infectious



Contaminant- and Biohazard-Free

Disposable, aerosol-free microfluidic cartridge allows for sterile sorting that protects the sample from the environment and scientist from the sample.



Compact

At under 2 cubic feet, NanoCellect's benchmark for access and performance allows every lab for the flexibility to do analysis and sorting into tubes or 96- and 384-well plates.



Simple and at your Bench

Intuitive software, fixed optics, no fluidics cart and less than one minute clean-up time.



Expanding the WOLF's Capabilities

With two lasers and up to nine fluorescent channels, the WOLF G2 aligns with a broad set of research applications and experiments. Three different laser configurations allows options specific to your needs.

Applications •••

Gene Editing



- With a sorting pressure of < 2 psi, gentle microfluidic cell sorting with the WOLF, along with single cell deposition using the N1 single-cell dispenser, eliminate the challenges that pluripotent stem cells face.
- · Without decompression shock and shear stress, single-cell deposition results in high monoclonal outgrowth.
- This improved workflow will allow for higher throughput pluripotent stem cell research, which is valuable for both basic and clinical research.

Infectious Disease

Viral Particles or Antigens

infectious disease research.

Viral Culture

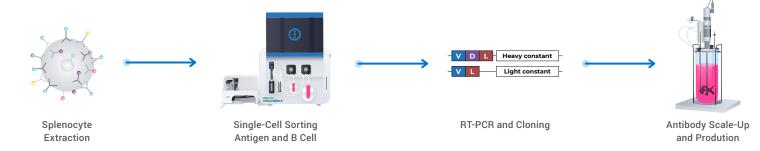


Therapeutic **Antibody Production**

· The WOLF G2 microfluidic cell sorter is aerosol-free and compact • The WOLF G2's disposable cartridges also avoid cross-contamination enough to fit into standard biosafety hoods, making it ideal for and create a safe and effective workflow for the kind of speed and efficiency required to combat infectious disease threats.

and B cell Sorting

Immunology and Antibody Discovery



- The WOLF G2 Cell Sorter provides a simple cell sorting solution that gently sorts high antibody-producing clones and dispenses single cells for optimal clone outgrowth.
- With two lasers and up to nine fluorescent channels, the WOLF G2 provides the most comprehensive benchtop cell sorting solution.

Cell Line Development



- The WOLF G2 Cell Sorter protects cell viability after sorting and improves sorting efficiency by increasing the number of viable clones growing per plate.
- · Contaminant- and biohazard-free, the WOLF cell sorter's rapidly exchangeable cartridge and tubing set eliminates carryover between samples and allows for guick and easy cleanup.
- When paired with the N1 Single-Cell Dispenser, the WOLF can select viable single cells and plate them into 96- or 384-well plates.
- · Low pressure microfluidics greatly improves cell integrity over conventional droplet sorters, resulting in high-outgrowth and high-titer monoclonal lines.

Genomics



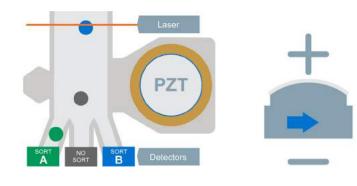
- The WOLF G2's gentle microfluidic system eliminates unintentional sequencing and analysis of dying or off-target cells or debris while maintaining cell integrity and avoiding stressing cells and affecting downstream data.
- Researchers can sort cells with a wide range of genome sizes, including human, plant, and animal cells or microorganisms like yeast and bacteria, providing insight on population genetics or disease.
- The WOLF G2 also allows labs to be completely flexible when it comes to sequencing research and eliminates carryover with a 100% disposable fluidic cartridge.

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Microfluidic Cartridge Technology •••

The WOLF Cell Sorter uses patented, microfluidic-based sorting with robust laser-excitation and sensitive PMT detectors to isolate mammalian cells, microbes, plant cells and more. A gentle and precise piezoelectric actuator directs cells into collection channels and allows analysis and sorting in a disposable format. This eliminates sample-to-sample contamination and biohazard exposure or cleanup.

- Unique to NanoCellect are our disposable cartridges that allow for bulk sorting or single-cell sorting.
- The sorting cartridges use a piezoacoustic actuator that gently directs cells into collection channels; an embedded cell sorting verification system gives instant feedback of sorting accuracy.
- This technology allows the WOLF G2 to sort up to 200 cells per second with high accuracy and effective recovery.
- Sort two selected cell populations with bulk sorting while the remainder of cells collects in a third channel.
- Deposit 1 to 100 cells per well in a 96- or 384 well plate using a single-cell sorting cartridge along with the N1 Single-Cell Dispenser accessory.



Magnified Sorting Junction

Actuator

Key Benefits



Sterile

Cartridges are individually wrapped in sterile packaging to avoid cross-contamination.



Disposable

Anything that the sample or sheath fluid touches is disposable.



Gentle Sorting

A gentle sorting mechanism results in improved viability of cells and higher outgrowth.



No aerosols

No dangerous aerosols are produced during sorting of hazardous samples.



Easy clean-up

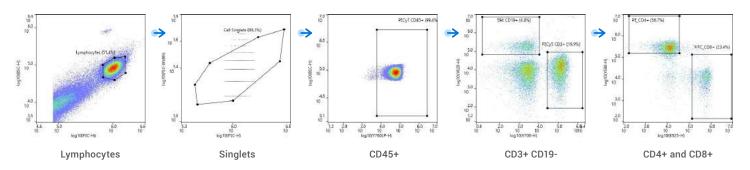
Clean-up in one minute and simply discard the cartridge.



WOLF G2 Cell Sorting •••

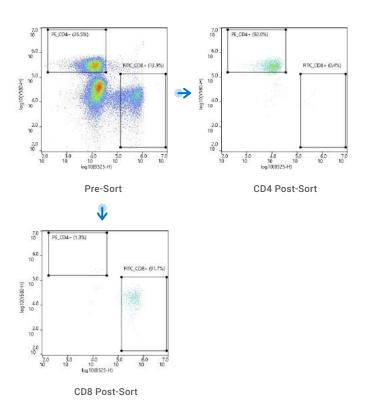
5-Color Immune Cell Sorting:

To verify sorting performance, CD4 and CD8 T Cells were sorted from BioLegend's PBMC Veri-Cells. They were gated from the CD3+CD19-CD45+lymphocyte population.



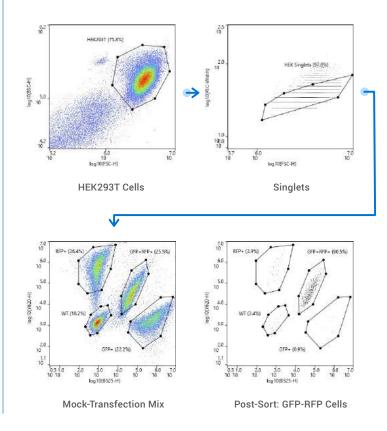
5-Color Sort Performance:

CD4 and CD8 T Cell populations were evaluated based on Cells and Singlets gates. CD4 T cells were enriched to 92.0% from a 26.5% target population. CD8 T cells were enriched to 91.7% from a 13.9% target population.



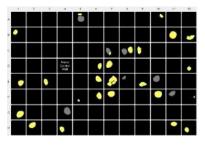
Transfected Cell Lines:

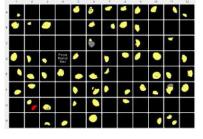
Four HEK293T cell lines were mixed to mimic a GFP+RFP+ dual-expressor transfection. To verify sorting performance, the GFP+RFP+ cells were sorted from the rest to result in 90.5% postsort purity.



Monoclonal HEK Outgrowth

The same 25% GFP+RFP+ dual-expressor cell mix was also sorted for single cells into 96-well plates and incubated for 14 days. As a rigorous limiting dilution comparison, 100% GFP+RFP+ dual-expressor cells were dispensed at 1 cell/well. Even without the advantages of the limiting dilution plate, the G2 plate still yielded more than a 2-fold increase in targeted monoclonal colonies.





Limiting Dilution: 1 cell/well from 100% GFP+RFP+ HEK293T cell mix

G2 Single-cell Dispense: 1 cell/well from 25% GFP+RFP+ HEK293T cell mix



RFP+ only fluorescence

GFP+RFP+ fluorescence

N1 Single Cell Dispenser

Designed to sort and dispense into 96- and 384-well plates, the N1 provides higher rates of singlet detection compared to cell printers or limiting dilution. Users can perform simple, labelfree dispensing or advanced multicolor panel single-cell dispensing.

Plate sorting specifications

Time to plate (96 wells): 3 - 8 minutes Time to plate (384 wells): 32 minutes

Droplet volume: 7-10 µL

Sample plate options: 96 or 384 wells (flat bottom, V-bottom, U-bottom, PCR)



WOLFViewer Software

The WOLFViewer software has an intuitive workflow menu that walks users through their experimental process, and is designed for both novice and expert users. New users can be performing their first sorts in about 20 minutes and can be on their own on day one.

Plate sorting specifications

System power up: 3 minutes

System setup and calibration: 20 minutes

System shutdown: 3 minutes

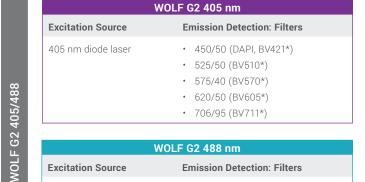
Additional features: Auto-alignment, intuitive compensation, advanced coloring and gating options,

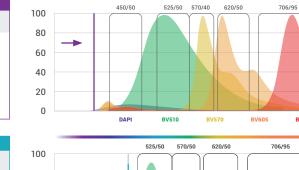
FSC files compatible with FlowJo



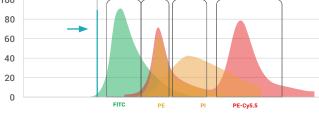
WOLF G2 Configuration Guide •••

The WOLF G2's three possible laser configurations significantly enhance the number of fluorescent markers that may be utilized.



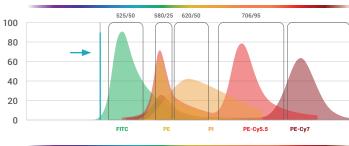


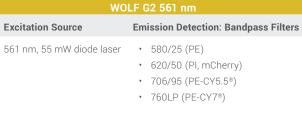
WOLF G2 488 nm			
Excitation Source	Emission Detection: Filters		
488 nm, 55 mW diode laser	 525/50 (FITC, GFP) 575/40 (PE) 620/50 (PI) 706/95 (PE-CY5.5°) 		

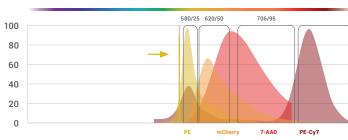


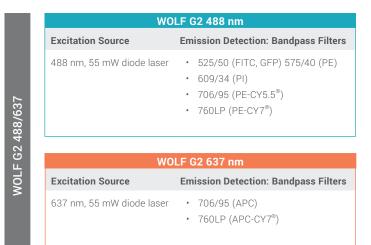
WOLF G2 488 nm	
Emission Detection: Filters	
 525/50 (FITC, GFP) 580/25 (PE) 620/50 (PI) 706/95 (PE-CY5.5*) 760LP (PE-CY7*) 	

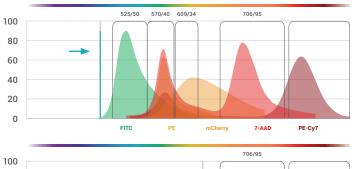
WOLF G2 488/561

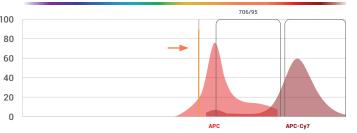












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Fluorophore Guide •••

Instrument	Excitation Laser	Emission Filter	Fluorescent Dyes	Fluorescent Proteins
/488		450/50	Alexa Fluor® 405, DAPI, Brilliant Violet™ 421, BD Horizon™ V450	eBFP, Cerulean
		525/50	Pacific Green, Qdot® 525, Brilliant Violet™ 510	AmCyan, CFP
	405 nm	575/40	Pacific Orange, Brilliant Violet™ 570, Qdot® 565, Qdot® 585	
		620/50	Brilliant Violet™ 605, Qdot® 605, Qdot® 625	
;2 40E	2 405	706/95	Qdot® 705, Brilliant Violet™650, Brilliant Violet™711	
WOLF G2 405/488		525/50	Alexa Fluor® 488, FITC, SYTOX® Green, Brilliant Violet™ 515	eGFP, eYFP
	488 nm	575/40	PE, PE-Texas Red®, ECD	eYFP, mCitrine
		620/50	PE-Cy5.5®, PerCP	
		706/95	PE-Cy7®, Qdot® 800, PE-Vio770	
//561	488 nm	525/50	Alexa Fluor® 488, FITC, Brilliant Violet™ 515	eGFP, eYFP, Emerald
		580/25	PE	mKate, mBeRFP
		620/50	PI, Texas Red®, PE-Texas Red®, PE-Alexa Fluor® 594, ECD	DsRED
		706/95	PerCP, PE-Cy5.5®, PE-Cy5®, PerCP-Cy5®	
2 488	WOLF G2 488/561	760LP	PE-Cy7®, PE-Vio®770	
LF G		580/25	PE	DsRED
WO	561 nm	620/50	Texas Red®, PE-Texas Red®, Alexa Fluor® 594, PE-Alexa Fluor® 594, ECD	DsRED, mCherry, mStrawberry
		706/95	PE-Cy5.5®, PerCP, 7-AAD, DRAQ5	
		760LP	PE-Cy7®, DRAQ5™, DRAQ7™	
WOLF G2 488/637	488 nm	525/50	Alexa Fluor® 488, FITC, Brilliant Violet™ 515	eGFP, eYFP
		575/40	PE, PE-610	eYFP, mCitrine
		609/34	PI	
		706/95	PerCP, PE-Cy5®, PE-Cy5.5®	
OLF		760LP	PE-Cy7®	
	627	706/95	APC, Alexa Fluor® 633	
	637 nm	760LP	APC-Cy7®, APC-Horizon™7	

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Brilliant Violet™ is a trademark of Sirigen Group Ltd.

 ${\sf BD\; Horizon^{\tiny TM}}$ is a trademark of Becton, Dickinson and Company.

Vio® is a registered trademark of Miltenyi Biotec GmbH.

For more information on compatible fluorophores, visit nanocellect.com or email info@nanocellect.com

WOLF G2 Specifications •••

F	luidics
Sample input	1.5 and 5.0 mL tubes
Sheath input	50 mL conical tubes
Sheath fluid	PBS or buffer of choice
Sheath fluid usage	7.5 mL/hour
Sample flow rate	24 μL/minute
Sheath flow rate	160 μL/minute
Dead volume	50 μL
Minimum sample volume	150 μL
Tubing diameter (inner)	200 to 500 μm
Flow cell	200 x 70 μm
Smaller channel diameter	50 μm
Sample pressure	< 2 psi
Sample output (bulk sorting)	1.5 mL or 5mL tubes
Sample output (single cell)	96- or 384-well plates (flat/U/V bottom or PCR)

N1 Plate sorting specifications		
Time to plate (96 wells)	3 - 8 minutes	
Time to plate (384 wells)	32 minutes	
Droplet volume	7-10 μL	
Sample plate options	96 or 384 well (flat bottom, V-bottom, U-bottom, PCR)	
	Optics	

Optics		
Laser Profile	25 x 75 μm	
25 x 75 μm	Forward (0 degrees, +/- 15)	
Scatter detection	Back (180 degress, +/- 15)	
Excitation & emission detection	See WOLF G2 Configuration Guide	
Optical alignment	Fixed alignment, no maintenance required	

Performance		
Scatter sensitivity	< 1.5 µm by FSC or BSC	
Scatter resolution	Resolves lymphocytes, monocytes, and granulocytes	
Fluorescence sensitivity	< 200 MESF FITC (using 488 nm laser) < 250 MESF PE (using 561 nm laser)	
Fluorescence resolution	8 peak separation with SPHERO™ Rainbow Calibration Particles	
Analysis speed	> 5,000 events/second	
Sorting	1- and 2-way	
Back-to-back sorting speed	200 events/second	
Absolute counts	Yes	
Volumetric counts	Yes	
Warm-up time	Less than 1 minute	
Sorting purity	Up to 99% purity	

Instrument specifications		
WOLF Dimensions	14.8H x 18.0W x 13.6D inch (37.6H x 45.8W x 34.5D cm)	
WOLF Weight	54 lbs / 24.5 kg	
WOLF Electrical	AC Input: 100-240V, 50-60Hz, 3-1.5A	
N1 Dimensions	8.42W x 6.5H x 8.34D in (21.4 x 16.5 x 20.9)	
N1 Weight	5.5 lbs / 2.5 kg	
N1 Electrical	DC Input: 24V, 1A	

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NanoCellect Technical Support •••



Onboarding

NanoCellect is committed to provide you with the highest level of support and expertise for your cell sorting workflow. We are dedicated to delivering the very best solutions and assistance to help you improve the quality of your research.



Training

Our Sales and FAS teams will assist you during initial instrument installation and for additional training options. Online training videos and other material are always available and being updated on our website's dedicated Knowledge Center.



Technical Support

Our expert technical support, including dedicated Field Application Scientists and Customer Success teams respond quickly and are available for you when you need them. Additional support options include remote TeamViewer sessions, inperson training and repairs, and online educational resources.



Service Contract

Our service offerings have expanded to include two options, allowing you to pick the level that best fits the needs for your budget and lab. In addition to standard customer support, one-on-one TeamViewer sessions, enhanced software updates for greater analysis capability, and preventative instrument maintenance (once per year) and repairs are covered under both plans.

For more information, visit nanocellect.com or email info@nanocellect.com





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