invitrogen

Conquer cell counting

Cell

Countess II Automated Cell Counters

Fast | Accurate | Affordable

ThermoFisher SCIENTIFIC

Countess II Automated Cell Counters

Advanced technology at an affordable price

Accurate counts in as little as 10 seconds

We offer two high-performance automated cell counters designed to meet the needs of any lab. The Invitrogen™ Countess™ II and Countess™ II FL Automated Cell Counters contain advanced autofocusing and counting algorithms to allow you to quickly and accurately count cells, while avoiding user variation associated with counting.

Both automated cell counters offer the following features:

- **Accuracy**—autofocus and auto-lighting minimize user-to-user variability
- **Speed**—results in as little as 10 seconds
- **Convenience**—built-in dilution calculator and ability to save up to 10 user profiles

Feature	Countess II counter	Countess II FL counter
Counting mode	Brightfield	Brightfield channel plus two optional, user-changeable fluorescence channels
Slides	Disposable slides	Reusable and disposable slides
Counting time	As little as 10 seconds	
Focus	Autofocus with manual focus option	
User profiles	Customize and save up to 10 user profiles	
User interface	Intuitive touch screen with ability to use a mouse, if desired	

"It outperformed all other suppliers' models that we tried."

—Michael DaCosta, The Jackson Laboratory

Features of the Countess II instruments

Reduced variability improves accuracy

The autofocus feature of Countess II instruments works by analyzing more than 30 focal planes, then selecting the plane with the best focal quality. This helps ensure that any variance from sample to sample, user to user, and slide to slide is minimized. Results are based on the optimal focal plane to enable highly accurate cell and viability counts. The Results view shown in Figure 1 allows quick visual confirmation of cells as being live or dead.

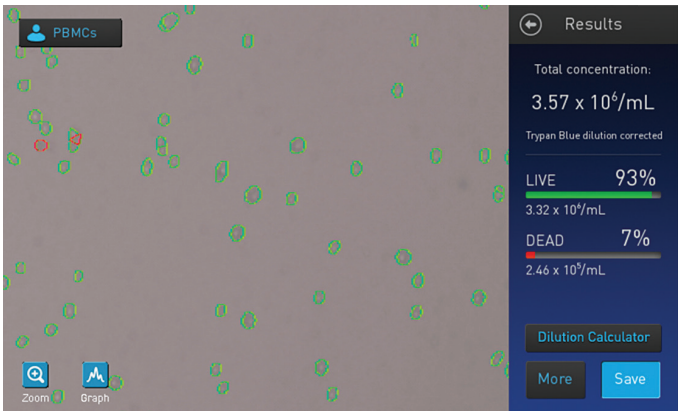


Figure 1. Peripheral blood mononuclear cells (PBMCs) counted using the Countess II Automated Cell Counter. Counting and viability measurement is done using trypan blue staining.

Reusable or disposable slides available

The Countess II FL Automated Cell Counter was designed to work with a reusable glass slide to help significantly reduce the long-term consumable costs associated with automated counting. Convenient disposable slides are also available.

Countess II FL counter | Reusable slide | An end to manual counting

The ability of the Countess II cell counters to gate cells based on cell size, brightness, and circularity using quantitative measurements rather than operator judgment also helps to reduce subjectivity, and allows increased repeatability between samples and users (Figure 2).

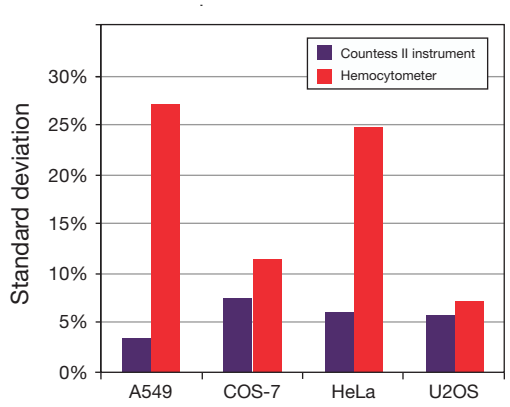


Figure 2. Count variability using a hemocytometer compared to using a Countess II instrument. Identical samples of A549, COS-7, HeLa, and U2OS cells were counted by three different operators using a Countess II cell counter and then manually with a hemocytometer and microscope. The user-to-user variability for the hemocytometer is much higher than for the Countess II instrument.

Time savings

The additional time it takes to manually count cells compared to counting with the Countess II Automated Cell Counters is often overlooked as an added cost. An individual counting five slides per day (two samples per slide) can save ~10 hours per month by switching to an automated counter with a reusable slide (Figure 3). This additional time can be applied to other activities in the lab, resulting in a significant advantage when switching from manual to automated cell counting.

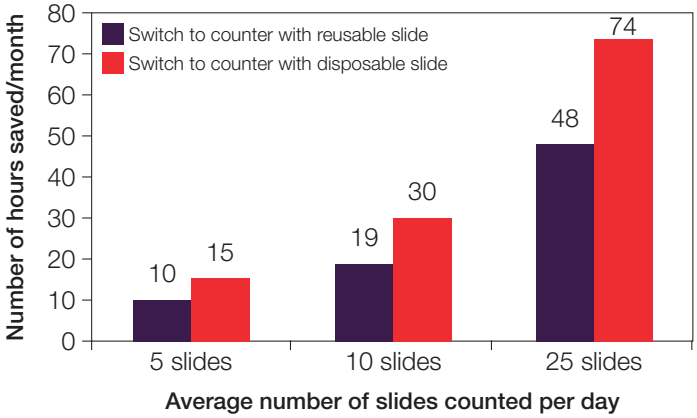


Figure 3. Estimated hours saved per month when switching from manual cell counting to using an automated cell counter.

Countess II FL Automated Cell Counter

Fluorescence detection beyond GFP and RFP

The Countess II FL Automated Cell Counter offers one brightfield and two optional, user-interchangeable fluorescent channels. The fluorescent colors detected are determined by the insertion of individual light cubes. The wide variety of light cubes available provides flexibility for multiple research applications.

Interchangeable LED light cubes

At the heart of the Countess system's fluorescence technology lie the unique Invitrogen™ EVOS™ light cubes. Each cube contains an LED, illuminating optics, and filters. Light cubes are user-interchangeable and auto-detected by the Countess II FL system for plug-and-play capability. There are more than 20 light cubes to choose from.

Table 1 lists some of the most commonly used light cubes.

Table 1. List of commonly used light cubes.

Light cube	Excitation (nm)	Emission (nm)	Common compatible dyes/fluorescent proteins
DAPI	357/44	447/60	DAPI, Hoechst, BFP
CFP	445/45	510/42	ECFP, Lucifer Yellow
GFP	470/22	510/42	GFP, Alexa Fluor™ 488, SYBR™ Green, FITC
YFP	500/24	524/27	EYFP, acridine orange
RFP	531/40	593/40	RFP, Alexa Fluor™ 546, Alexa Fluor™ 555, Cy®3, DsRed, Rhodamine Red, dTomato
Texas Red™	585/29	624/40	Texas Red, Alexa Fluor™ 568, Alexa Fluor™ 594, MitoTracker™ Red, mCherry
Cy®5	628/40	692/40	Cy®5, Alexa Fluor™ 647, Alexa Fluor™ 660, DRAQ5™

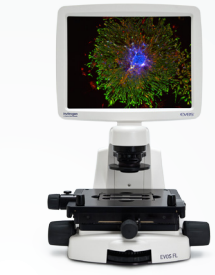
For a complete list of available common and specialty light cubes, go to **thermofisher.com/evoslightcubes**

Customizable instruments for your fluorescence experiments

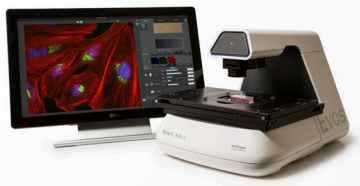
You can get more out of your research with easy-to-use, modular systems that can adjust to fit your experimental needs. We offer imaging systems that can be customized with a variety of LED light cubes, vessel holders, and objectives.

Invitrogen™ EVOS™ FL and EVOS™ FL Auto 2 Imaging Systems are powered by the same light cubes that are used in the Countess II FL system. The light cubes can be easily transferred between systems to increase the experimental capabilities of your lab while saving money.

Learn more about our full line of imaging systems at **thermofisher.com/microscopes**



EVOS FL Imaging System



EVOS FL Auto 2 Imaging System

Applications

Viability

Assessing cell viability is a key step in daily cell manipulation, and is required for accurate and efficient downstream processing. With the Countess II FL instrument, there are many fluorescence options in addition to trypan blue staining to quickly and easily check viability prior to downstream sample analysis. Figure 4 shows results obtained using the Invitrogen™ LIVE/DEAD™ Viability/Cytotoxicity Kit combined with the EVOS GFP and Texas Red light cubes.

Apoptosis

In addition to cell viability, knowing how many cells are dead or dying are key pieces of information that can be determined quickly with the Countess II FL instrument. Cells of interest can be stained with a viability dye such as Invitrogen™ SYTOX™ Red Dead Cell Stain, as well as an apoptosis indicator that measures caspase activation, such as Invitrogen™ CellEvent™ Caspase-3/7 Green Detection Reagent. This staining combination, used together with EVOS GFP and Cy5 light cubes, offers a fast and simple method to obtain apoptosis and viability data (Figure 5).

Transfection

The ability to assess the number of cells successfully transfected or transduced in a cell population is a basic and critical evaluation parameter. Commonly, transfection or transduction of cells of interest results in the expression of a fluorescent protein reporter, such as GFP. The Countess II FL instrument, loaded with your choice of light cubes, offers a quick and simple method to easily obtain transfection efficiency data (Figure 6).

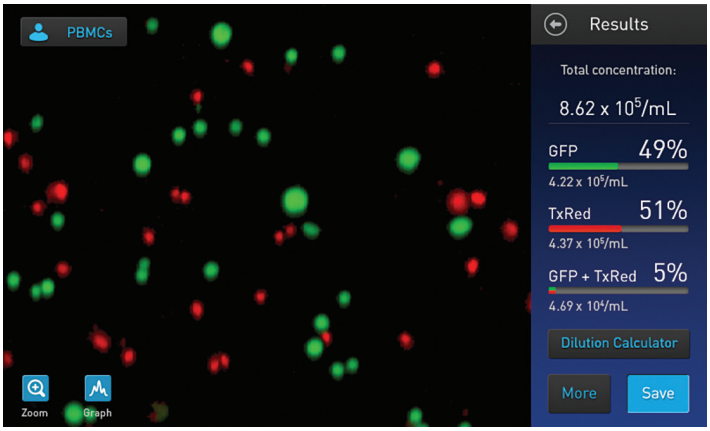


Figure 4. Viability assay using the LIVE/DEAD Viability/Cytotoxicity Kit.

Figure 5. Apoptotic and dead cells counted on a Countess II FL Automated Cell Counter.

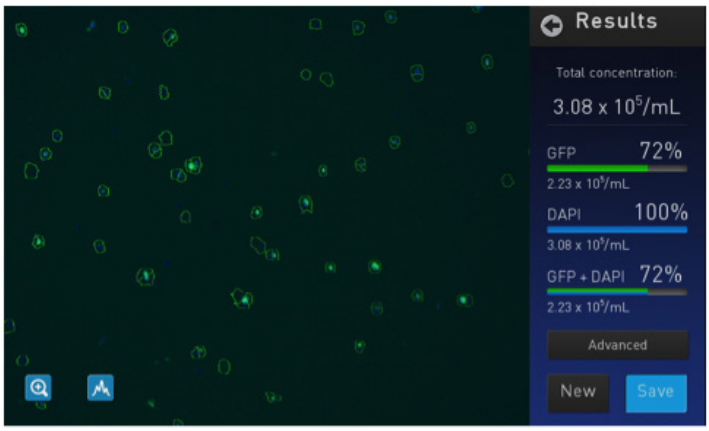
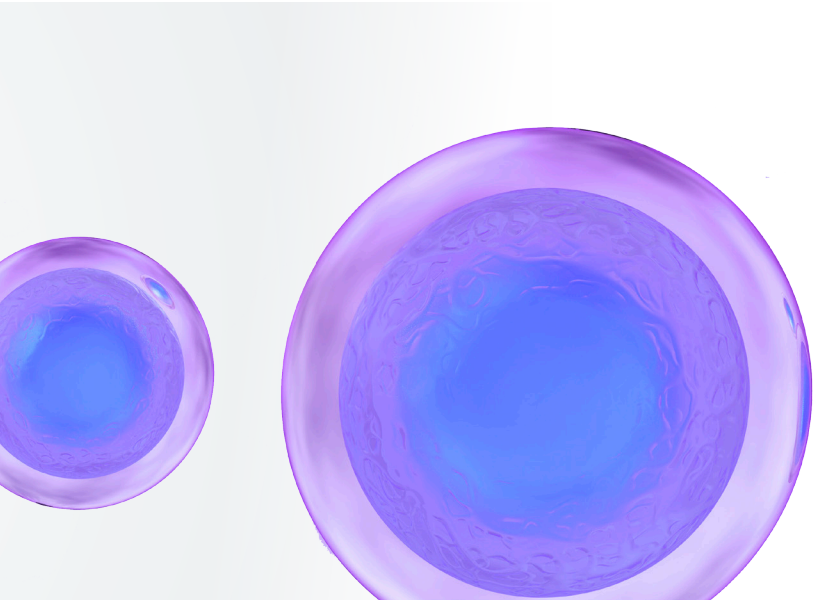


Figure 6. Determination of transduction efficiency in HeLa cells.



Easily gate cells and view histograms

Cells can be easily gated based on cell size, brightness, and circularity to fine-tune precisely what is included or excluded for specific applications. View the histogram by selecting the graph icon, and watch the changes in real time as you use the gating features. The average cell size is also displayed on the histogram (Figure 7).

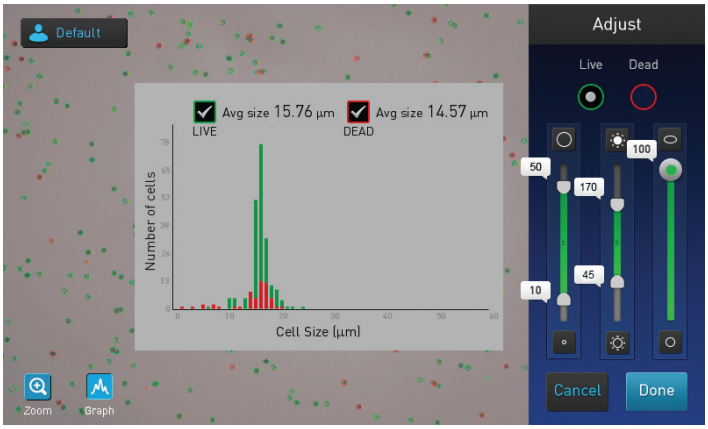


Figure 7. Histogram is updated to reflect gating changes. Multiple gating options can be selected separately for live cells and dead cells, allowing easy inclusion or exclusion of debris or specific populations.

Gating based on fluorescence intensity

The ability to use up to two customizable fluorescence colors with the Countess II FL Automated Cell Counter allows for more accurate counts when dealing with complex samples that contain significant amounts of platelets, RBCs, or debris. In addition, the ability to gate based on fluorescence intensity to determine how many cells are dim vs. bright, or are expressing a fluorescent protein, can save time and effort prior to downstream analysis with techniques such as flow cytometry or high-content analysis (Figure 8).

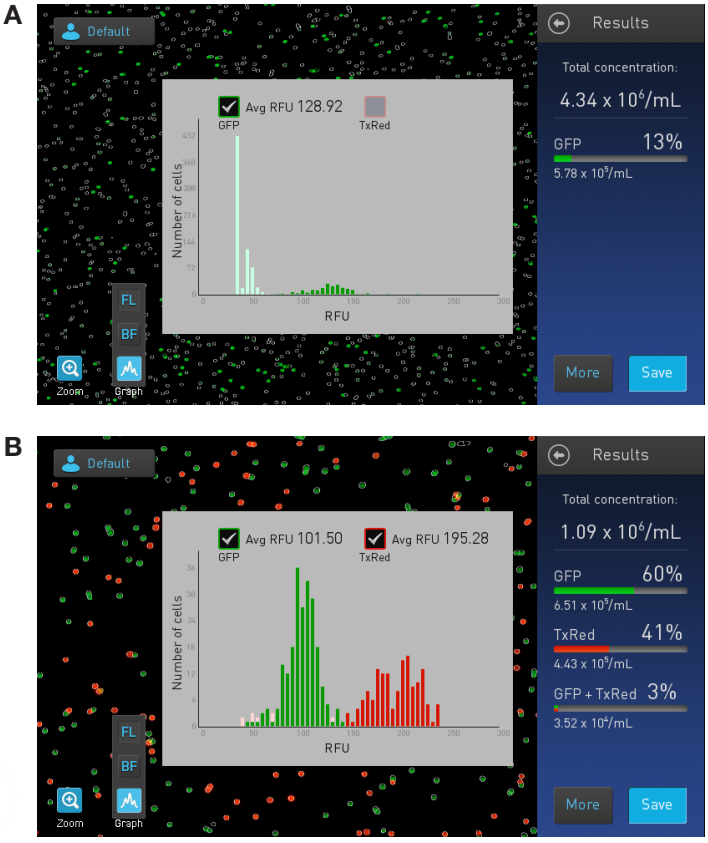
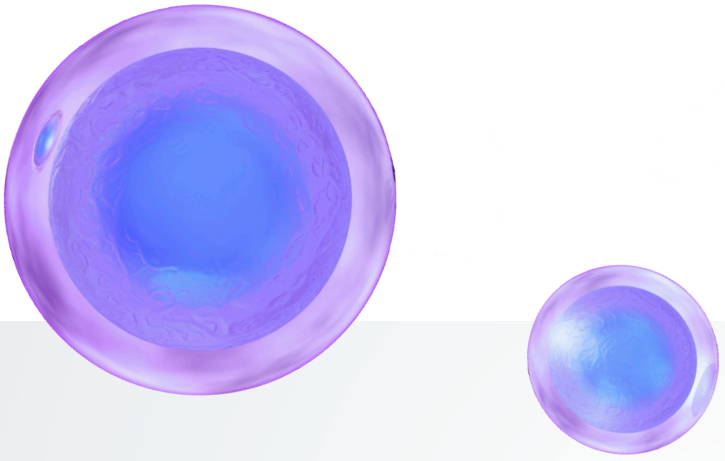
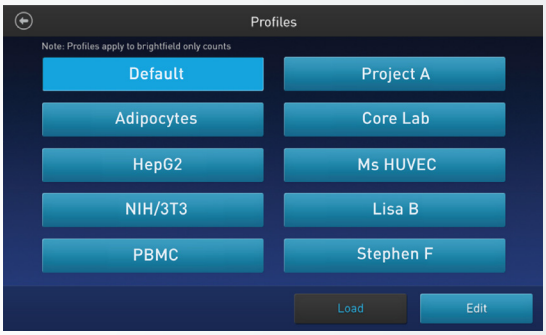


Figure 8. Gating based on fluorescence intensity. (A) The dim cells (around 50 RFU) have been excluded from the GFP-positive count. **(B)** Two fluorescence colors are present, each of which can be gated by size, brightness, circularity, and fluorescence intensity.



Streamline your workflow with custom profiles

Custom profiles can be saved and easily accessed from the capture and results screens. Settings for cell size, brightness, and circularity as well as fluorescence thresholds can be customized and saved for easy retrieval.



Dilution calculator

Easily determine the amount of cell sample and buffer needed for your experiment with the built-in dilution calculator. The cell count results are automatically used in the calculation; simply input the desired concentration and the volume needed (Figure 9).

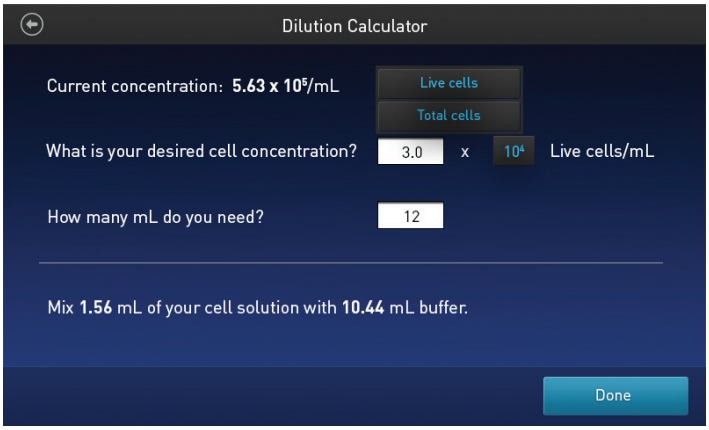


Figure 9. A dilution calculator quickly determines the amount of cell sample and buffer needed.

Saving files

Using the USB port on the Countess II FL Automated Cell Counter, you can save your cell count results and images, and then transfer them to your own computer. Image files can be saved as TIF, PNG, or JPG files, and the results are saved as a CSV file. A printable PDF report with results, images, and settings used can also be saved (Figure 10).

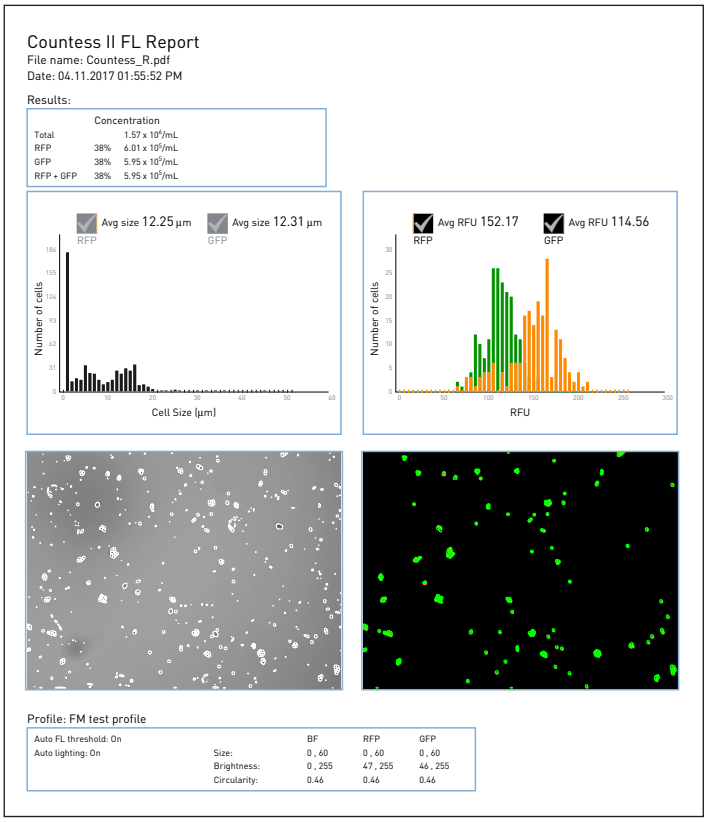
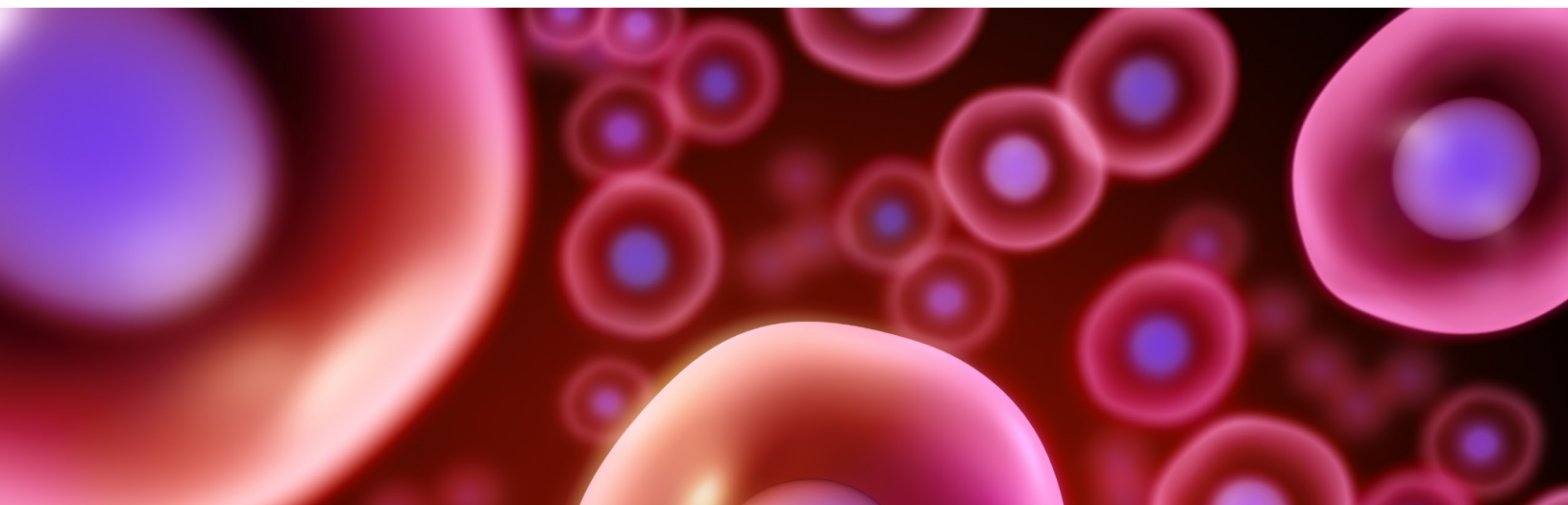
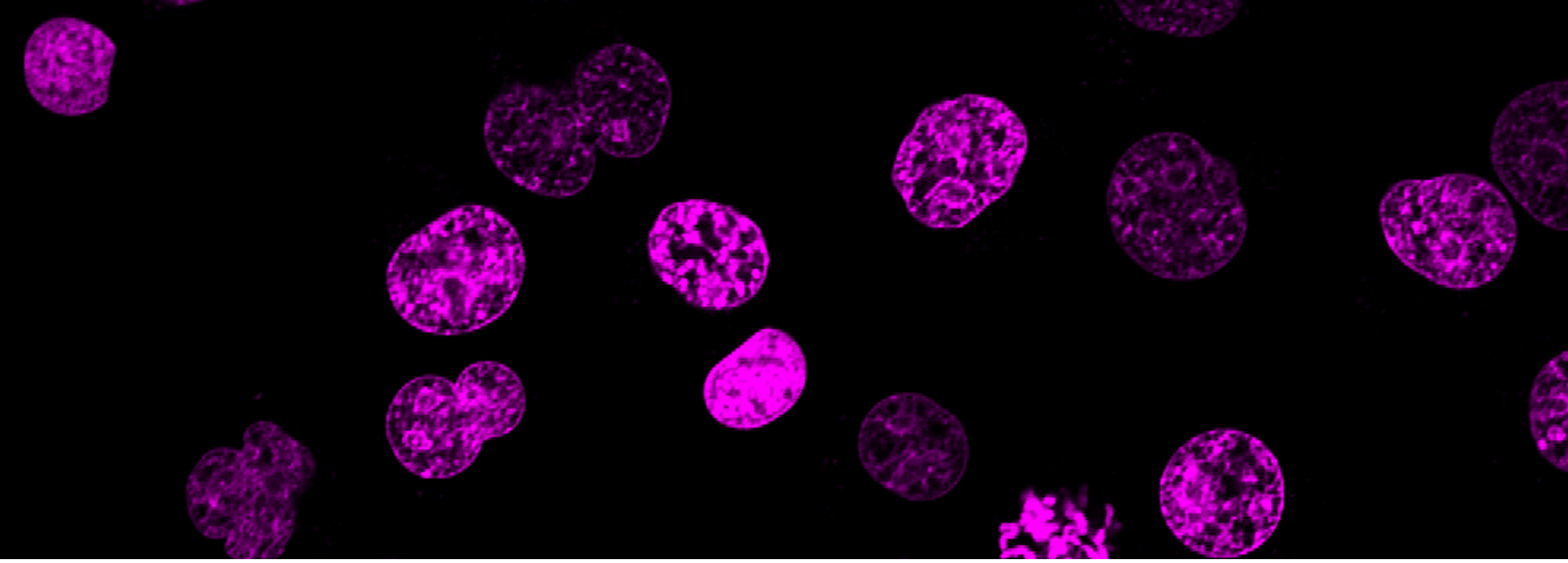


Figure 10. PDF report with results and images.





Frequently asked questions (FAQs)

Q. What sizes of cells are the Countess II Automated Cell Counter and Countess II FL Automated Cell Counter able to count?
A. Objects of ~4–60 µm can be counted. Cells of ~7–60 µm can be counted and assessed for viability.

Q. How long do the instruments take to count?
A. ~10 seconds per sample.

Q. What is the counting range?
A. The Countess II instruments are designed to read samples with concentrations in the range of 1 x 10^4 cells/mL to 1 x 10^7 cells/mL.

Q. What count-to-count precision should be expected?
A. The count-to-count variability with the Countess II instruments is less than 10%.

Q. Can I buy the Countess II FL instrument without light cubes?
A. Yes, the Countess II FL instrument without light cubes installed will function exactly like the Countess II instrument—as a brightfield cell counter. Customers can easily add light cubes as application needs evolve.

Q. Can I use the reusable slide only on the Countess II FL instrument, or can I use it on the Countess II instrument as well?
A. The reusable slide is only compatible with the Countess II FL instrument.

Q. Can the images be saved?
A. Yes, the results and image files can be saved after each count. Images can be saved as JPG, PNG, TIFF, and BMP files. A report that includes the image, histogram, results, and instrument settings can also be saved as a PDF.

Q. How do I clean and maintain the instrument?
A. Clean the surface of the instrument with a damp cloth. Wipe the screen dry immediately. The Countess II instruments have no moving parts to maintain, no tubes to clean, and no buffers to replace.

Q. How often do I have to calibrate the Countess II instruments?
A. The Countess II instruments come precalibrated. You do not have to calibrate the instruments.

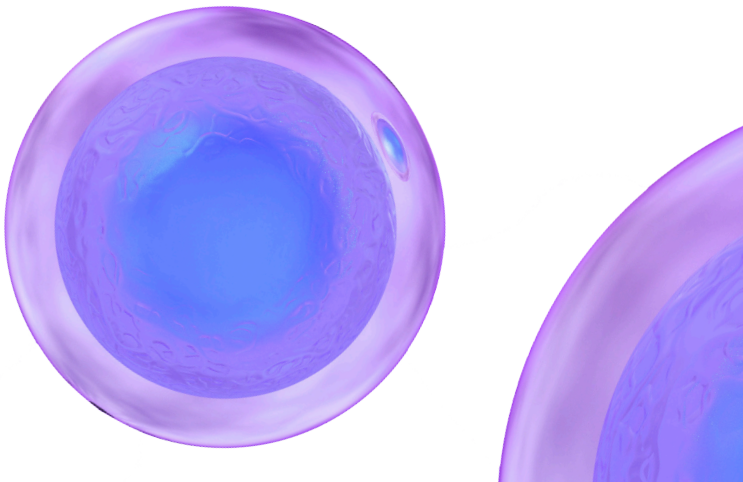
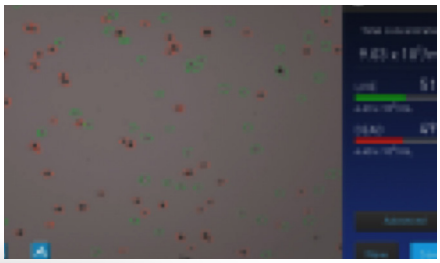


Table 2. Commonly used dyes and light cubes used with the Countess II FL cell counter for viability and apoptosis determination.

Dye	Light cube	Cat. No.
Viability dyes		
ReadyProbes Cell Viability Imaging Kit, Blue/Green	DAPI and GFP	R37609
ReadyProbes Cell Viability Imaging Kit, Blue/Red	DAPI and RFP or Texas Red	R37610
LIVE/DEAD Viability/Cytotoxicity Kit	GFP and RFP or Texas Red	L3224
Propidium Iodide ReadyProbes Reagent	RFP	R37108
SYTOX Green Nucleic Acid Stain	GFP	S7020
SYTOX Red Dead Cell Stain	Cy5	S34859
7-Aminoactinomycin D (7-AAD)	Texas Red or Cy5	A1310
Apoptosis dyes		
CellEvent Caspase-3/7 Green Detection Reagent	GFP	C10423
SYTOX Red Dead Cell Stain	Cy5	S34859

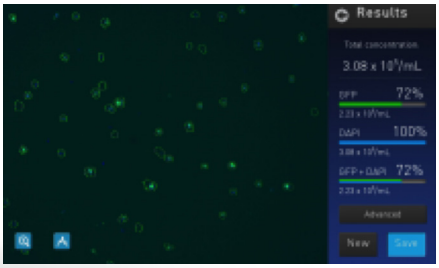
Table 3. Cell lines validated on the Countess II Automated Cell Counters.

Cell type	Animal	Tissue/organ source	Cell size (diameter)
A431	Human	Skin	15.5 µm
Adipocytes	Human	Adipose-derived stem cells	13 µm
Aortic smooth muscle	Human	Smooth muscle	20 µm
Blood, whole lysed	Human	Blood	NA
CHO-M1WT2	Chinese hamster	Ovary	NA
CHSE	Chinook salmon	Embryo	16–17 µm
COLO-205	Human	Colon	NA
COS-7	African monkey	Kidney	NA
HEK293	Human	Kidney	13 µm
HeLa	Human	Cervix	NA
HepG2	Human	Liver	18 µm
HL-60	Human	Blood	NA
J774A.1	Mouse	Blood	13–14 µm
Jurkat	Human	Blood	12 µm
K562	Human	Bone marrow	NA
MCF-7	Human	Breast	20–24 µm
MRC-5	Human	Lung	18 µm
NIH/3T3	Mouse	Embryo	18 µm
PBMC	Human	Blood	7–8 µm
PC-12	Rat	Adrenal gland	12–14 µm
Pulmonary artery endothelial cells	Human	Blood vessel	13 µm
Pulmonary artery smooth muscle	Human	Smooth muscle	20 µm
SF-21	Insect	Ovary	NA
U266	Human	Blood	12–13 µm
U2OS	Human	Bone	NA
Umbilical vein endothelial cells	Human	Blood vessel	17 µm



Countess II Automated Cell Counter (Cat. No. AMQAX1000)

Technical specifications	
Optics	Brightfield only
Slides	Countess™ Cell Counting Chamber Slides (disposable)
Processing time	As little as 10 seconds
Required sample volume	10 µL
Sample concentration range	1 x 10^4–1 x 10^7 cells/mL
Particle and cell diameter range	Particles: ~4–60 µm; cell diameter: ~7–60 µm
Objective	2.5x
Total magnification	~55x
Illumination	LED (50,000 hours)
Camera	5 megapixels
Output	JPG, TIF, PDF, CSV, BMP, ANG
Instrument dimensions (W x D x H)	9 x 5.5 x 9 in.; 23 x 14 x 23 mm
Weight	8 lb
Operating power	100–240 VAC, 0.58 A max.
Frequency	50/60 Hz
Electrical input	12 V DC, 2 A



Countess II FL Automated Cell Counter (Cat. No. AMQAF1000)

Technical specifications	
Optics	3 channels (brightfield and 2 slots for EVOS LED light cubes)
Slides	Countess™ II FL Reusable Slide or Countess Cell Counting Chamber Slides (disposable)
Processing time	As little as 10 seconds
Required sample volume	10 µL
Sample concentration range	1 x 10^4–1 x 10^7 cells/mL
Particle and cell diameter range	Particles: ~4–60 µm; cell diameter ~7–60 µm
Objective	2.5x
Total magnification	~55x
Illumination	LED (50,000 hours)
Camera	5 megapixels
Output	JPG, TIF, PDF, CSV, BMP, ANG
Instrument dimensions (W x D x H)	9 x 5.5 x 9 in.; 23 x 14 x 23 mm
Weight	8 lb
Operating power	100–240 VAC, 0.58 A max.
Frequency	50/60 Hz
Electrical input	12 V DC, 2 A

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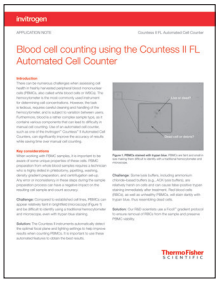


Application notes

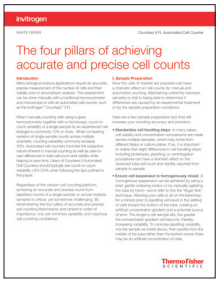
Find application notes with protocols for viability, apoptosis, transfection, and PBMC counting using the Countess II instruments.

thermofisher.com/countess

Convince your lab

We can help address key areas that labs are concerned about when considering purchasing an automated cell counter. Download the prewritten letter, petition, and application note demonstrating the benefits of an automated cell counter.

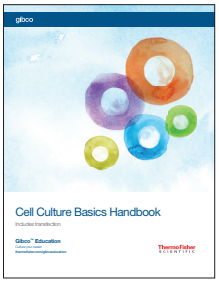
thermofisher.com/stopthemadness



White paper

Improve your cell counting results with this white paper discussing the four pillars of accurate cell counting.

thermofisher.com/accuratecounting



Cell culture learning center

Find the information you need for successful cell culture—including application notes, videos, webinars, and Gibco™ Education through virtual labs.

thermofisher.com/cellculturelearning

Ordering information

Product	Quantity	Cat. No.
Countess II FL Automated Cell Counter	1	AMQAF1000
Countess II Automated Cell Counter	1	AMQAX1000
Countess Cell Counting Chamber Slides	50	C10228
Countess II FL Reusable Slide	1	A25750
Countess™ II FL Reusable Slide Holder	1	AMEP4746

Find out more at **thermofisher.com/countess**

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