# **Cellometer®** K2 Fluorescent Viability Cell Counter





## Live/Dead Nucleated Cell Counts using Dual-fluorescence

### Why Dual-fluorescence?

Because brightfield cell counting does not differentiate nucleated from non-nucleated cells and trypan blue staining is not as consistent as fluorescent staining, dual-color fluorescence is strongly recommended for accurate viability analysis for primary cells. The K2 is equipped with standard assays for dual-fluorescence analysis of a variety of cells stained with Acridine Orange and Propidium Iodide (AO/PI).

## The AO/PI Method

Acridine Orange (AO) is a nuclear staining (nucleic acid binding) dye permeable to both live and dead cells. It stains all nucleated cells to generate green fluorescence. Propidium Iodide (PI) can only enter dead cells with compromised membranes. It stains all dead nucleated cells to generate red fluorescence. Cells stained with both AO and PI fluoresce red due to quenching, so all live nucleated cells fluoresce green and all dead nucleated cells fluoresce red. Staining with AO/PI also doesn't require incubation, saving time compared to trypan blue.



#### Brightfield

AO/PI

The brightfield image on the left shows the combination of nucleated cells, red blood cells, and platelets present in the sample. The red blood cells are not visible in the fluorescent image on the right, only the live (green) and dead (red) nucleated cells are counted.

## No Interference from Red Blood Cells, Platelets, or Debris

The dual-fluorescence AO/PI method utilizes nuclear staining dyes that bind to nucleic acids in the cell nucleus. Because most mature mammalian red blood cells do not contain nuclei, only live and dead mononuclear cells produce a fluorescent signal. There is no need to lyse red blood cells, saving time and eliminating an extra sample preparation step. Red blood cells, platelets, and debris are not counted in the fluorescent channels.

## Simple and Fast Cell Counting Workflow with Results in Seconds



1. Pipette 20 µl



2. Insert slide and count



3. Get images and data

"The Cellometer K2, coupled with ViaStain AOPI stain, allows users to easily stain nucleated cells in samples containing red blood cells. No lysis buffer is required, which makes getting results very quick."

- Susan Hamilla, Sorrento Therapeutics



## Advanced Fluorescence Cell Counter

The Cellometer K2, powered by Matrix software, utilizes brightfield imaging and dual-fluorescence imaging to quickly and accurately identify and count individual cells. Cell count, concentration, diameter, and % viability are automatically calculated and reported.

#### The Cellometer K2 has the following advantages:

- **Dual-fluorescence and brightfield imaging –** stain only nucleated cells for the most accurate count and viability information
- Fast results count, size, concentration, and viability calculations in <60 seconds
- Analyze complex samples designed for analysis of complex and messy samples including whole blood, peripheral blood, cord blood, and bone marrow
- **Multiple fields of view –** increased accuracy with the ability to capture one, four, or eight images per sample
- Built-in predefined assays quickly analyze viability, apoptosis, and transfection efficiency
- Built-in cell types includes saved parameters for over 400 cell types
- Small sample volume only 10 µl of cell sample required
- **Customizable reports –** includes predefined reports with the ability to create new ones with graphs, images, charts, and tables
- Multi-language support over 7,000 languages available
- **21 CFR Part 11 ready** optional add-on that includes an audit trail, user access control, and digital signature

"We love our Cellometer K2 and every lab should have one! Gone are the days of manual cell counting and we can now reliably and quickly count thousands of cells in a few seconds."

- Nav Masani, AstraZeneca









## **Results Display**



## Accuracy from Cell Lines to Primary Samples

The Cellometer K2 can be customized to handle a variety of cell types, including primary cells, tumor digest, insect cells, cell lines, fragile cells, and more at low or high concentrations.



## Predefined Assays and Cell Types

Take the guesswork out of setting up your cell quantification experiments. The Cellometer K2 comes with frequently used assays and cell types with predefined settings to ensure consistent results from sample to sample. Easily build custom assays and cell types to fit your experimental needs.

Not sure what settings to use? Our customer success team and field application specialists are here to help you develop fit-for-purpose assays and protocols for your specific research and development requirements.

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## **Capture Multiple Fields of View**

Capture one, four, or eight images per sample. The instrument default is set to four images, which is the equivalent of six quadrants on a hemocytometer. Eight images are equivalent to twelve quadrants on a hemocytometer. The ability to capture multiple images improves the cell counting dynamic range and accuracy of results.

"This unit works fast to get accurate and precise numbers. It takes a fraction of the time trypan blue or other manual methods for cell counting. It allows for greater throughput of samples and simplifies our workload. It is simple to use and miles ahead of any of the competitors. "

- Matthew Wilgo, New England Cord Blood Bank

## **Dual-fluorescent Staining for Clumpy Cells**

The fluorescent image (far right) shows bright green AO-positive hepatocytes declustered by the Cellometer K2 algorithm. Red circled hepatocytes are PI-positive (dead) while free nuclei are not counted.





Live AO-positive

Free nuclei are not counted Dead, PI-positive

Declustered clumpy cells are accurately counted

## Predefined and Customizable Reports

Use default or customized reports based on the data and images you want to be included for your specific experimental needs. Automatically export images and data reports including CSV, Excel, Word, or PDF files. Perform statistical analysis for a wide range of parameters such as average, variance, min/ max, and standard deviation of cell size.



## 21 CFR Part 11 Ready

An optional module can be purchased to meet requirements for 21 CFR Part 11 ready software. The additional module comes with the following features:

- User login with passwords
- User assigned permissions
- Audit trail
- Error log files
- Electronic signatures



## Consumables that Work for You

We offer two different disposable counting slides, one with protective coverings on both sides and a ready-to-use option packed in microscope slide boxes. Several key advantages include:



- No clogging
- Time savings no washing
- No risk of cross-contamination
- Reduce biohazard risk to users

## Performance of the Cellometer K2





Table of results for cell concentration dynamic range

#### **Concentration Dynamic Range**

The dynamic range for cell concentration measurements on Cellometer K2. This data set was taken on a concentration series of a cultured Jurkat cell line.

Samples from  $1 \times 10^5 - 1 \times 10^7$  cells/ml can be counted without further dilution.

The %CV at each concentration was below 10%.

#### **Viability Dynamic Range**

The viability dynamic range is 0 - 100% for Cellometer K2 using dual-fluorescence AO/PI stain.

Sample	N Value	Average Live Cell Concentration	% Viability	CV of Concentration	CV of Viability
Jurkat	24	3.61E+06	92.2%	8.9%	1.0%
Human PBMC	10	5.94E+06	96.0%	4.7%	0.5%
Mouse Splenocyte	10	1.86E+07	88.6%	5.6%	0.7%

Table of results for cell concentration and viability using AO/PI

The results indicate the accuracy of the Cellometer K2 instrument in assessing the viability of Jurkat cells using AO/PI for cell viability. Jurkat, human PBMC, and mouse splenocytes were tested at 24, 10, and 10 sample replications, respectively. The viability average was calculated and plotted. The results show the reliability and accuracy of the Cellometer K2 in measuring cell concentration and viability of mammalian cells.

Low Sample Volume, Complete Counts

Cell samples can be precious. K2 requires only 10 µl for accurate counts.

## Applications for Cellometer K2 Fluorescent Cell Counter



**Peripheral Blood Mononuclear Cells (PBMC)** Measure live cell concentration and viability without lysing red blood cells for consistent results from patient samples.



#### Measure GFP Transfection

Rapidly identify fluorescence-positive cells from a sample, calculate cell concentration, size, and determine the GFP transfection percentage automatically.



## WBCs in Whole Blood

Measure nucleated cell concentration without lysing red blood cells using nuclear staining dyes (AO), for human and mouse blood.



#### Immunology Research

Quantify cell viability and concentration for a variety of immunologically relevant samples such as: bone marrow, cord blood, splenocytes, lymphocytes, isolated mononuclear cells, tumor digests, murine samples, and others.



### Insect Cells

Measure live cell concentration and viability for baculovirus infected insect cells.



#### NCI-60 Cancer Cell Lines

Measure live cell concentration and viability of cancer cell lines used in oncology research and biology research.



#### Single Cell Sequencing

Scientists in single cell genomics choose the Cellometer K2 because accurate cell counts are critical in sample preparation. Viability and clumps also need to be assessed to minimize double rate.



#### **Adoptive Cell Transfer Therapy**

Perform cell based assays and measure cell size, viability, and concentration of cell lines and primary samples used in adoptive cell therapy research.



#### **Apoptosis and Necrosis**

Detect and analyze apoptotic and necrotic cells with Annexin V and PI.



#### **Primary Hepatocytes**

Measure live hepatocyte concentration and viability from fresh and cryo preserved samples using dual-fluorescent nuclear stains for human, rat, mouse, and horse.

## Need higher-throughput?

High-throughput Automated Cell Counter





Count 24 samples in less than 3 minutes *Learn more: www.nexcelom.com/cellaca* 

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