

COPAS Vision™

SYSTEM SPECIFICATIONS

INTRODUCTION

The COPAS Vision instrument is a large particle flow cytometer designed to image, analyze, sort and dispense biological materials and other objects ranging in size from 2 to 1500 microns in diameter. The system is designed to handle objects which are too large or too fragile for traditional flow cytometers. Examples include small multicellular organisms, delicate large cells, cell clusters, small seeds, and beads used as micro-carriers or for combinatorial libraries.

CONFIGURATIONS

The COPAS Vision system can be configured to accommodate four ranges of object diameters. Each system features a square cross-section quartz flow cell optimized for maximum sensitivity for a corresponding sample size range.

| | Flow Cell Channel | Object Size Range | Recommended Object Size* | Minimum Sorted Drop Size* |
|-------------------|-------------------|-------------------|--------------------------|---------------------------|
| COPAS Vision 250 | 250 µm | 2-200 µm | 10-175 µm | 1 µl |
| COPAS Vision 500 | 500 µm | 6-400 µm | 30-350 µm | 2 µl |
| COPAS Vision 1000 | 1000 µm | 15-850 µm | 30-750 µm | 7 µl |
| COPAS Vision 2000 | 2000 µm | 40-1500 µm | 100-1400 µm | 15 µl |

* Note: these are general guidelines. Please talk to one of our Applications Scientists about your specific project and sample requirements.

Parameters Measured

This continuous flow system is capable of quickly analyzing small and large quantities of objects using 6 or 12 optical detectors. Objects are passed axially, one by one, through the focus of a laser beam. The resulting signals are then detected and recorded by extinction, scatter and fluorescence detectors. Relative size is measured by the extinction detector which records the length of time that the signal from the blocked light remains above a pre-set threshold level; this parameter corresponds to the time of flight (TOF). The optical density of the object is determined by the total integrated signal of the blocked light; this parameter is the object's extinction (EXT). The forward-scatter (FS), side-scatter (SS), and fluorescence intensity (FLU) can be simultaneously detected at different wavelengths as determined by the excitation and emission filters in the system. TOF is related to an object's axial length, EXT is related to how opaque or transparent an object is, and FLU is related to the amount of fluorescence from a fluorescent tag or labeled antibody that is present.

Sorting

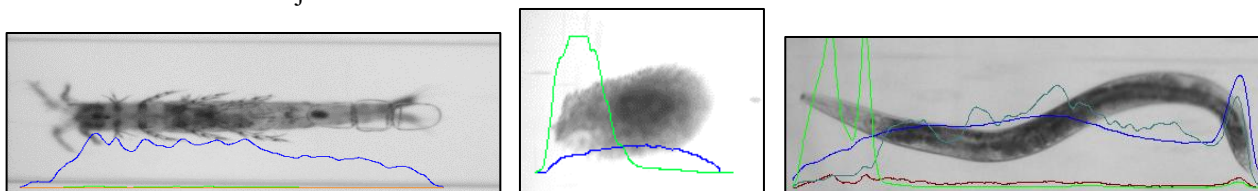
Sorting and dispensing decisions are based on user-selectable values for up to 61 fundamental parameters (31 parameters when using the 6-detector configuration). Additional calculated and derived parameters can also be entered through our FlowPilot-Pro™ software. Up to 32 different regions of data can be monitored and used for setting sorting criteria. Objects can be dispensed into multi-well plates, tubes or bulk receptacles. Using our patented air diverter sorting mechanism, the COPAS Vision instrument is gentle enough to sort and dispense fragile cells and live organisms without affecting viability.

Enclosed Sort Chamber

The sorting chamber is enclosed providing aerosol containment and UV sterilization to enhance biosafety and sterility. Those objects not meeting the sort criteria are diverted by a puff of air to a sample recovery container where they may be recaptured, unharmed and viable.

Profiler

Profiler digitizes the optical signal level into successive point measurements made while the object passes through the flow cell. An object containing a small bright fluorescent spot will produce a fluorescence signal with a corresponding data plot with a narrow peak; more uniform fluorescence will produce a data plot with a plateau. Profiler can be used to select and sort objects with specific localized morphology, as characterized by extinction, scatter, or fluorescence variations internal to the object.



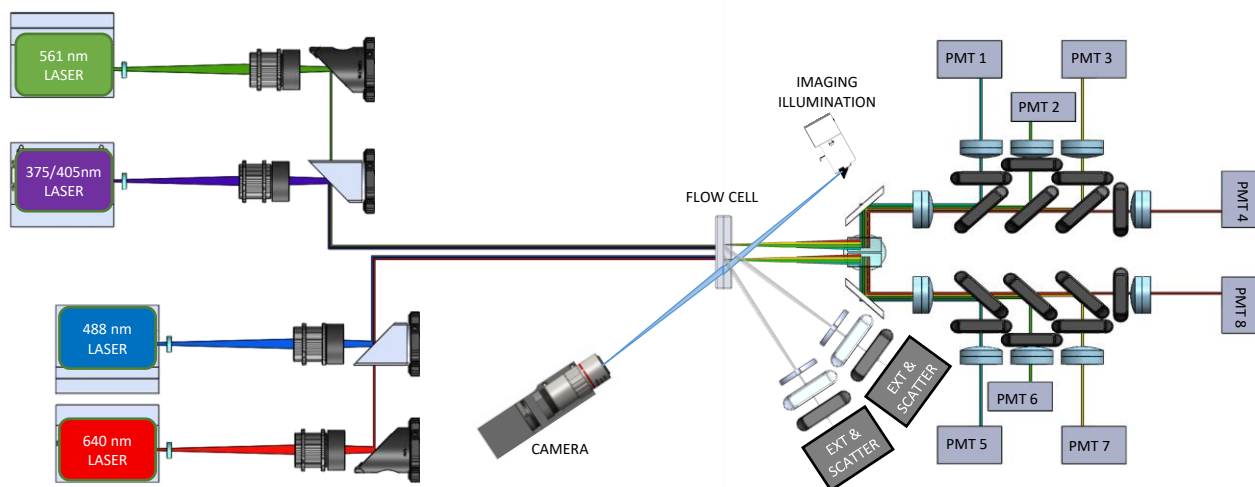
Imaging

Bright field images of each object can be captured as it flows through the flow cell. This image data can be stored and correlated with Profiler data and other parameters. Images can be captured for all objects or just those that meet user-selectable criteria. Up to 300 images per second can be captured.

| | Field of View | Image Resolution |
|-------------------|---------------------------|-------------------|
| COPAS Vision 250 | 250 x 1700 μm | 1.4 μm |
| COPAS Vision 500 | 500 x 1700 μm | 1.4 μm |
| COPAS Vision 1000 | 1000 x 1700 μm | 1.4 μm |
| COPAS Vision 2000 | 2000 x 3600 μm | 3.5 μm |

OPTICAL ASSEMBLY DESIGN SPECIFICATIONS

COPAS Vision instruments are available with multiple excitation lasers and detection optics to accommodate a variety of fluorescence excitation and emission requirements. Various lasers are used as excitation sources for fluorescence. Custom optics and low dispersion lenses allow these instruments to precisely focus the excitation lasers across all laser wavelengths and flow cell sizes. Bright field transillumination is used for capturing images. User changeable optical filters and dichroic mirrors allow precise customizations specific to a sample. The figure below is a common configuration though other configurations are possible.



Laser Excitation Sources (1 to 4 lasers)

The COPAS Vision system can accommodate up to four lasers. Low optical noise, solid-state lasers are used for optimal sensitivity and consistent performance. Lasers can be configured co-linearly or spatially separated depending upon application. Some of the most popular wavelength choices are shown above but other options are available so please inquire if you have a special request.

Extinction Detectors (1 or 2 detectors)

The Extinction detector (EXT) is a PIN Photodiode which detects the amount of light blocked as the object passes in front of the laser beam. The COPAS Vision system has one Extinction detector and can be configured as an optional second detector.

Forward Scatter Detector (1 or 2 detectors)

The Forward Scatter (FS) detector is a PIN Photodiode which detects the amount of light scattered off an object at a shallow angle as it passes in front of the laser beam. The COPAS Vision system has one Forward Scatter detector and can be configured with an optional second detector.

Side Scatter & Fluorescence Detectors (4 or 8 detectors)

High-sensitivity photomultiplier tube (PMT) detectors and collection optics are utilized to maximize detection of fluorescence and side scatter light. The COPAS Vision can be configured for simultaneous detection using either four or eight PMT detectors. Optical filters and dichroic mirrors can select which wavelengths of side scatter and / or fluorescence emissions are collected. These filters and mirrors can be easily configured for different setups. Optional fluorescence detector filters are available in a variety of ranges and can be interchanged with the standard filters by the end user. One example of a filter setup is shown in the table below.

| | Example | Detected Wavelengths | | | Example | Detected Wavelengths |
|-------|--------------|----------------------|--|-------|---------|----------------------|
| PMT 1 | Side Scatter | 488 nm | | PMT 5 | NADPH | 423 – 468 nm |
| PMT 2 | FITC | 506 – 534 nm | | PMT 6 | GFP | 500 – 525 nm |
| PMT 3 | PE | 576 – 596 nm | | PMT 7 | YFP | 532 – 554 nm |
| PMT 4 | Cy5 | 659 – 701 nm | | PMT 8 | DsRed | 603 – 627 nm |

FLUIDIC SPECIFICATIONS

Sample Introduction

The standard sample introduction uses a 50 ml conical tube with suspended stir bar (40 ml working volume). Our large particle auto-sampler, the LP Sampler™, can be used to introduce samples from multi-well plates. The OSIS, an oscillating sample introduction system for extremely delicate samples, can be used with disposable syringes ranging from 3 to 30 ml, and our 750 ml and 1.5 liter sample cups with suspended stir bars can be added for introducing larger sample volumes. These gentle, low-pressure sample introduction systems are designed to operate at sample pressures of 0.2 - 6 psi [0.01 - 0.4 bar] depending upon the sample and flow cell size.

Sample Collection Cups

Sorted sample can be collected in multiple types of collection tubes: 50 ml, 15 ml, and microcentrifuge tubes. Additionally, sample can be sorted into multi-well plates or bulk dishes. Non-sorted sample is collected in a sample recovery container and can be re-run on the instrument or used again in another application.

Fluid Regulation

Sample Fluid – the sample flow is controlled to +/- 0.5% accuracy with automatic feedback pressure regulation.

Sheath Fluid – a high precision metering pump is used to deliver precise and repeatable sheath flow rates.

All other fluids are controlled by the instrument's software and electronics producing repeatability and flow stability of all fluids.

Fluid Bottle Capacity

10 Liters – Sheath, Waste, and Water

1 Liter – Cleaning, Ethanol, and Hypochlorite (bleach) flushing solutions

TECHNICAL SPECIFICATIONS

| | |
|---------------------|--|
| Workstation: | Windows® based workstation running FlowPilot-Pro™ software with real-time data acquisition and 64 bit processing. |
| Stage: | X-Y stage for dispensing into multi-well plates, 50 ml, 15 ml, and microcentrifuge tubes. |
| Data: | Data resolution using 16 bit A/D converters with 32-bit computation producing 16-bit data granularity (1:65536) at data acquisition speeds up to 10 MHz. Maximum object sampling resolution of better than 0.4 microns per data point. |
| Imaging: | Image resolution of 2 microns using 5X / 0.14 NA Plan Apochromat Objective and a 5 Mega Pixel camera. Brightfield illumination using a 450 nm light source. |

INSTALLATION

| | |
|----------------------------|---|
| Instrument Size: | 22 x 26 x 18 in (56 x 66 x 44 cm) depth x width x height |
| Fluidics Cart Size: | 18 x 23 x 23 in (46 x 59 x 59 cm) depth x width x height |
| Workspace: | Recommend a permanent, open, level, vibration-free work space with the dimensions: 24 x 48 x 30 in (61 x 120 x 76 cm) depth x width x height |
| System Weight: | 120 lbs (55 kg) not including computer or Fluidics Cart |
| Power: | 350 Watts. Place the COPAS Vision instrument within 6 feet (2 meters) of the power outlets. |

For 110-120v Countries: One single phase, 120VAC, 20 amp, 50/60 HZ, separate dedicated line with protective earth ground, using the detachable cords supplied. *If the optional Air Compressor is used, it is recommended that a second separate dedicated line (100-120VAC, 15 amp, 50/60 HZ, single phase with protective earth ground) is provided. Always ensure that local electrical codes are followed.

For 220-240v Countries: One single phase, 220/240VAC, 16 amp, 50/60 HZ, separate dedicated line with protective earth ground using the detachable power cords provided or CE7/VII approved equivalent detachable cords. *If the optional Air Compressor is used, it is recommended that a second separate dedicated line (220/240VAC, 10 amp, 50/60 HZ, with protective earth ground) is provided. Always ensure that local electrical codes are followed.

| | |
|-------------------------|---|
| Pressurized Air: | 40-100 psi (3-7 bar) at 1 CFM, of filtered, water and oil-free air can be supplied by house-air or optional compressor. Actual sample pressure during sorting is typically less than 6 psi depending on flow cell size and sample type. |
| Temperature: | 60°F (15°C) to 85°F (30°C) environmental temperature limits. Temperature should not vary more than +/- 1 degree C from the time of experimental setup through completion. The system generates approximately 3800 BTU/Hr. |
| Humidity: | 0 to 85% non-condensing environmental relative humidity limits. |

