FlowCAM®

An Imaging Particle Analysis System for the Automated Identification and Classification of Particulate Matter

Particle analysis has been practiced since the early days of the microscope, and microscopy still remains as a standard method for particle analysis due to the tremendous amount of information that microscope images can convey. However, microscopy is time consuming, manually intensive and generally can not produce high statistical confidence in results due to the fact that statistically significant numbers of particles take too long to observe in this fashion.

Volumetric-based particle analysis techniques, such as Coulter counters, laser diffraction and light obscuration have the advantage that they can very rapidly and repeatedly measure statistically significant numbers of particles. These systems can measure tens of thousands of particles in a matter of minutes, whereas manual microscopy can typically only measure hundreds of particles in times measured in hours.

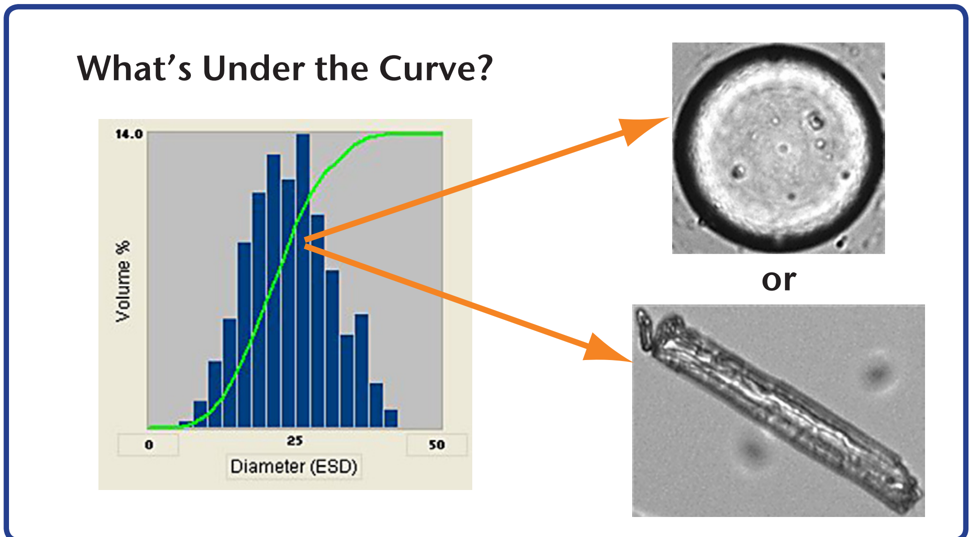


Figure 1: Volumetric-based particle analysis systems can not distinguish between 2 similar sized particles with very different shapes

Figure 2: Benchtop FlowCAM

The primary disadvantage to these volumetric-based systems is that they can only measure Equivalent Spherical Diameter (ESD), operating on a (false) assumption that all particles are spherical in shape. Centuries of microscopic analysis tells us that particles are not all spherical in shape. Even in a homogenous sample, particles can have widely varying shape. In the real world, however, we are most often interested in analyzing samples which are heterogeneous collections of many different particle types. The volumetric-based analysis systems can not even make the

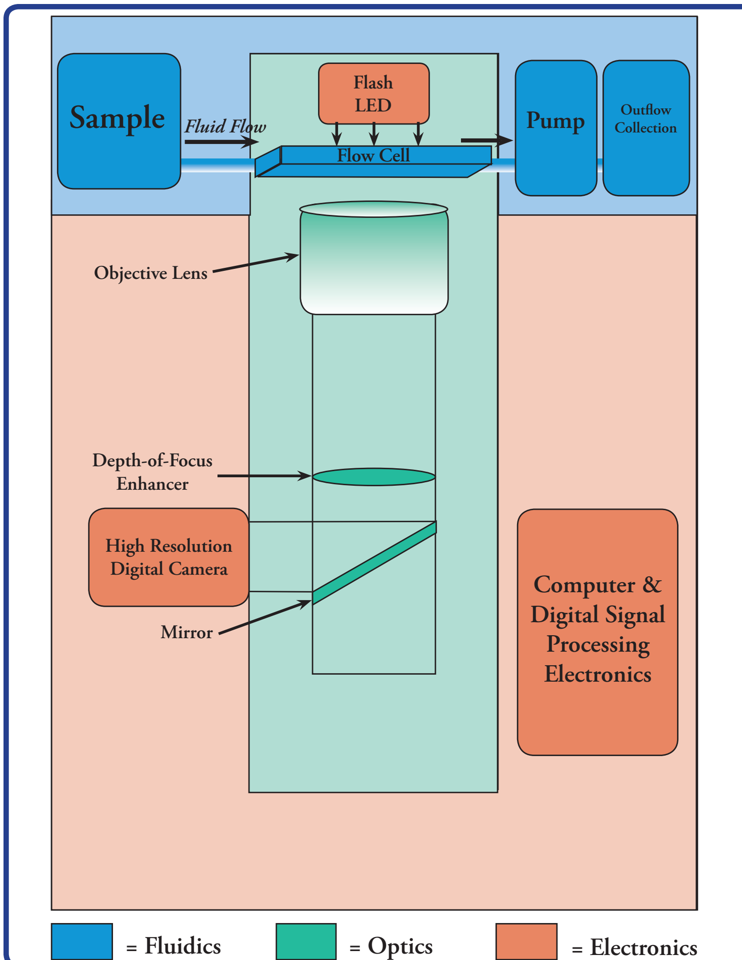


Figure 3: FlowCAM Architecture Block Diagram

most basic differentiation between a rod-shaped and a spherical shaped particle, as seen in Figure 1.

FlowCAM® is an imaging-based particle analyzer that enables microscopic particle measurements to be made rapidly enough to produce statistically significant amounts of particle data similar to the volumetric-based systems (see Figures 2 and 3). The enormous advantage of FlowCAM is that, unlike the volumetric based systems, it collects many different measurements (both spatial and spectral) from each particle. The multitude of measurements obtained allows for software post-processing which can automatically differentiate different particle types in a heterogeneous sample based upon particle morphology (shape) and spectral (gray-scale and color) measurements, in addition to ESD.

Example 1

The first example of how FlowCAM can use the additional information gained from imaging involves quality control of silica gels. Silica gels have many uses, but one of the most critical is as the stationary phase in column chromatography. While the diameter of the particles is important (and the material is usually characterized by diameter), the shape is also extremely critical, as it affects how the particles fit together when packed into the column, and therefore will directly affect the column efficiency.

In this example, two different lots of silica having roughly the same diameter distributions were analyzed in the FlowCAM. Figure 4 shows the results of these runs. Both samples had a mean size (ESD) around 16μm, but as can be seen from the images, very different shape characteristics. Sample A (on left in Figure 4) contains the desired characteristic of being roughly uniform (basically spherical) in shape, which means they will pack very consistently in the column. Sample B (on right in Figure 4) contains non-uniform (erose, non-spherical) shaped particles, which will not pack uniformly in the column, creating all sorts of varying pressure (and hence efficiency). The lot containing Sample B should be rejected by the column packer.

While this is easy to see qualitatively from the images, the beauty of imaging particle analysis is that measurements made from the particle images will yield quantitative proof to back up what is easily seen. In this instance, the parameter "Aspect Ratio" (width/length) is sufficient to easily distinguish the two samples: Sample A, Aspect Ratio = 0.91, standard deviation = 0.05, whereas Sample B, Aspect Ratio = 0.80 , standard deviation = 0.09. However, if the difference between the two samples were even more subtle, a more discriminatory measurement, called "Roughness", collected by the FlowCAM could have been used. This measurement distinguishes very subtle differences in the irregularity of the particle's surface by comparing the ratio of the perimeter of the particle to its convex perimeter.

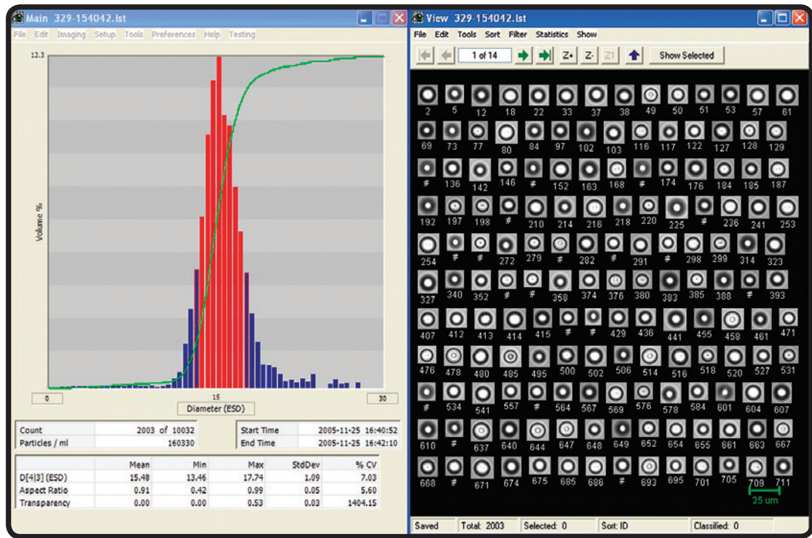


Figure 4: Results of FlowCAM® runs for two Silica samples: Sample A or the two samples are quite different as evidenced by the

Example 2

The first example above shows a relatively simple situation where a relatively uniform (homogeneous) sample was analyzed, with shape measurements used to determine whether a product could be accepted or not in a quality control environment. Where the FlowCAM really shines, however, is when looking at samples

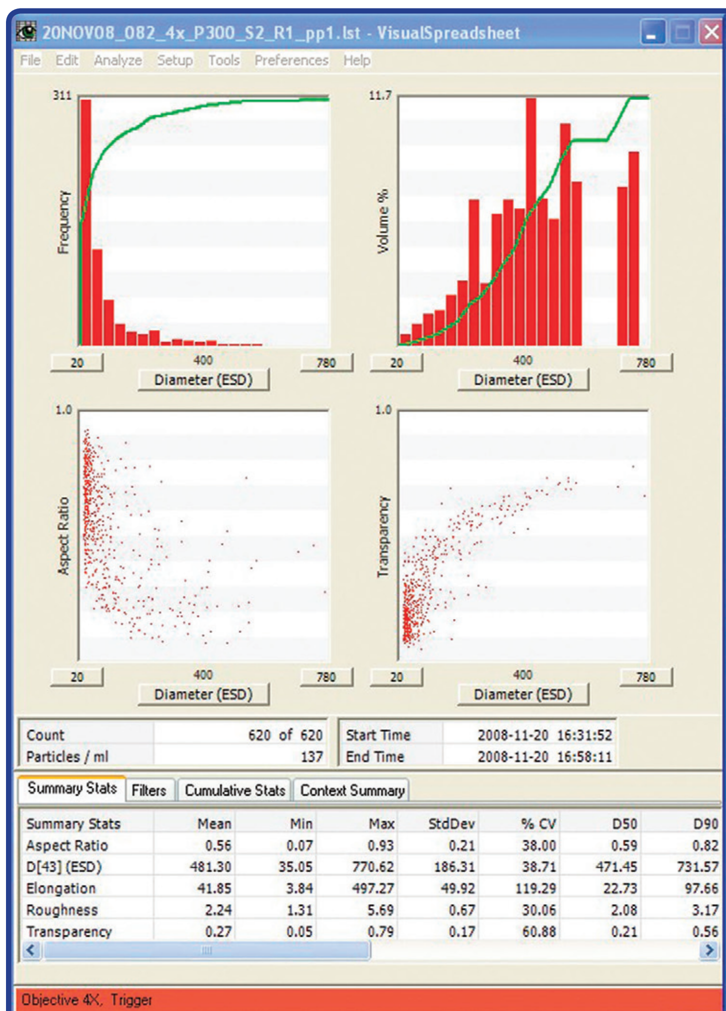
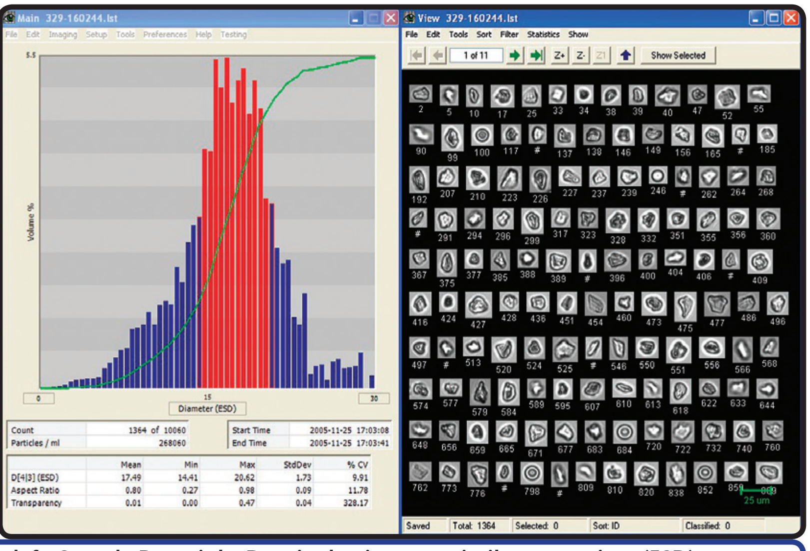


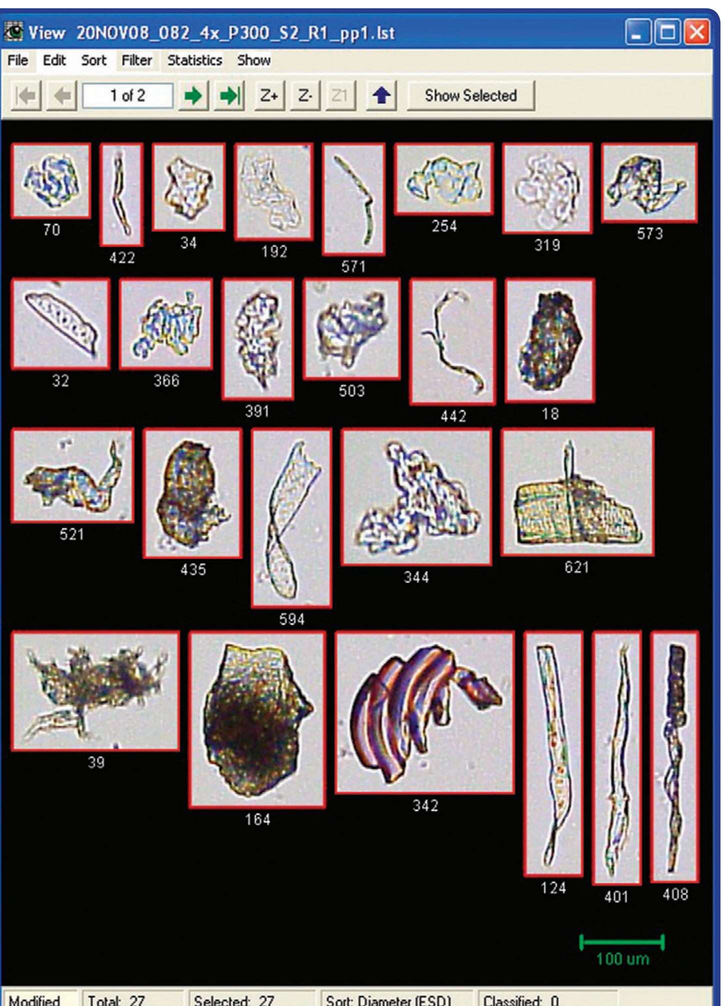
Figure 5: Results of wash water sample ru are shown on left window, with e



n left, Sample B on right.Despite having very similar mean sizes (ESD), images. Sample B is unacceptable, and will be rejected.

that are *heterogeneous* in nature, where multiple particle types are present. In these situations, using imaging particle analysis and pattern recognition techniques can provide an automated method for characterizing the types and quantities of particles present.

In this second example, we show how this works using a “wash water” example. In this case, the sample analyzed is wash water from a manufacuring process for electronic devices. After manufacturing, these devices are washed to remove traces of fibers, metals and plastics that could remain from the manufacturing process. It is important that the wash water contains less than a certain number of each of these particle types, as too many leftover particles could cause failures. In addition, the types and numbers of particles washed off can serve as an indicator for any problems arising in the actual production process itself.



n through FlowCAM. Summary statistics example particle images on right.

Figure 5 shows the results of running the wash water sample through the FlowCAM, with graphs and statistics shown in the left hand window, and sample images shown in the right window. Note the diversity of the particle types found: the long, skinny particles are fibers, the semi-transparent particles are metal shavings, and the more opaque particles are plastics.

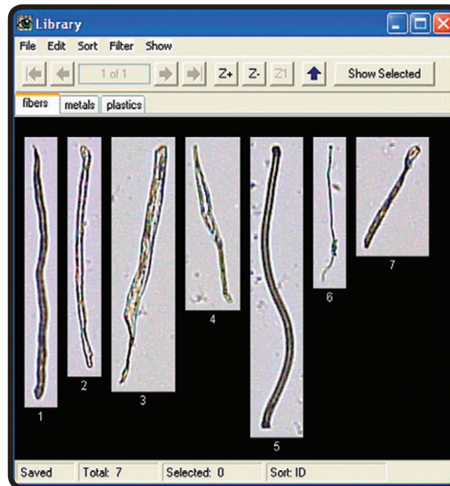


Figure 6: “Fibers” library images

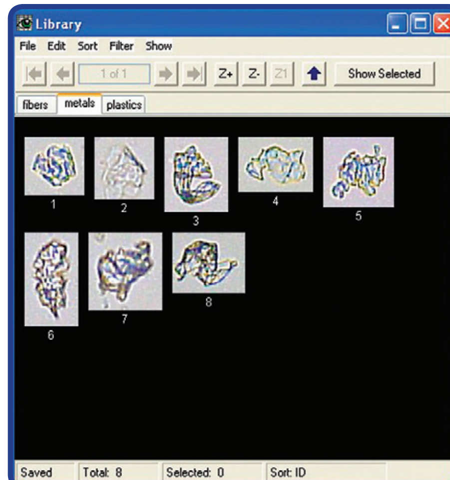


Figure 7: “Metals” library images

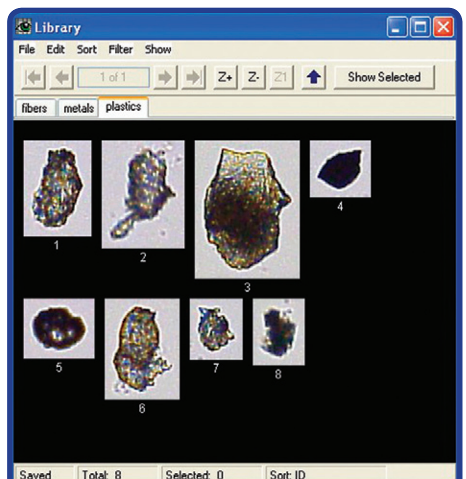


Figure 8: “Plastics” library images

The determination of what each particle type looks like is based upon knowledge supplied by the user, who has examined these particles closely before using a microscope, and therefore knows what the images represent. This “expert” user begins the particle classification process by defining “libraries” of particle images that represent each particle type that it is desired to quantify. Figures 6, 7, and 8 show the libraries built for each particle type interactively using the VisualSpreadsheet™ software. These libraries can be saved and used on every run so that the same statistical comparison is being used for every analysis.

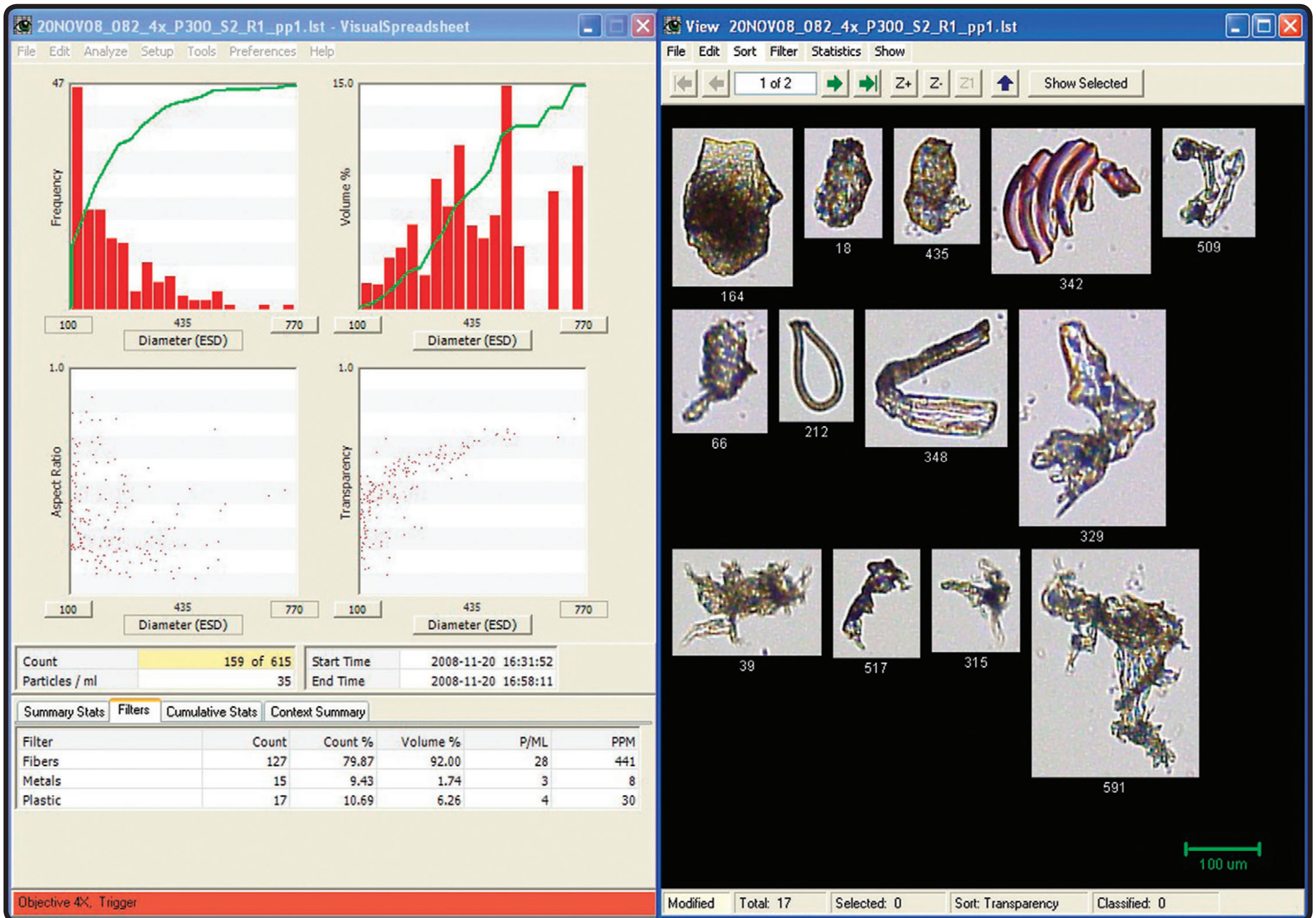


Figure 9: Results after filters applied to wash water sample run through FlowCAM. Filter results showing count and concentration can be seen at the bottom of the left hand window, and the images for the "plastics" particles are seen on the right.

Once the libraries are saved, they can be used to "filter" the sample results to ***automatically*** characterize the particles into the different types desired. Figure 9 shows the results after the filters have been applied (note that in this application only particles >100µm in ESD were required to be measured, so the graph's x-axes were adjusted to only display those particles). On the lower left hand window, you can see that out of 159 total particles, 127 were classified as fibers, 15 as metals and 17 as plastics, and the corresponding volume %, particles/ml and PPM have also been calcualted.

Conclusion

FlowCAM's imaging based particle analysis approach overcomes the limitations imposed by traditional volumetric based systems. Not only does it yield real, image-based measurements for far more information, but the use of powerful statistical- based pattern recognition tools in the VisualSpreadsheet software enables *automatic* particle type characterization in heterogeneous samples. We invite you to contact us, send a sample for analysis, and let us prove how FlowCAM can be a valuable tool in your laboratory!

FlowCAM Basic Specifications

Parameter	Value (Range)
Size Range (ESD)	1µm - 2,000µm (Count) 3µm - 2,000µm (Shape)
Basic Shape Measurements	Equivalent Spherical Diameter (ESD), Area Based Diameter (ABD), length, width, aspect ratio, area, volume
Advanced Morphology Measurements	Circularity, Elongation, Compactness, Circle Fit, Perimeter, Convex Perimeter, Edge Gradient
Gray-Scale and Color (optional) Measurements	Intensity, Average Intensity, Sigma Intensity,Transparency, Average Red, Green, Blue, R/G Ratio, R/B Ratio, G/B Ratio
Power Requirements	Benchtop: 100-250 VAC, 50/60 Hz Portable: 100-250 VAC, 50/60 Hz or 12V DC
Dimensions	Benchtop: 21" width X 16" deep X 12" high Portable: 18" width X 15" deep X 26" high (lid open, 14" lid closed)
Shipping Weight (crated)	Benchtop: 95lbs (60lbs unit only) Portable: 85lbs (40lbs unit only)