# Automation in Image Analysis for Particle Size and Shape Measurement

**Fully automated particle characterisation using imaging and data analysis techniques can deliver statistically significant size and shape information in a single measurement, and with minimal user intervention. This article examines how the latest instrumentation is addressing industry's need for increasingly detailed particle characterisation.**

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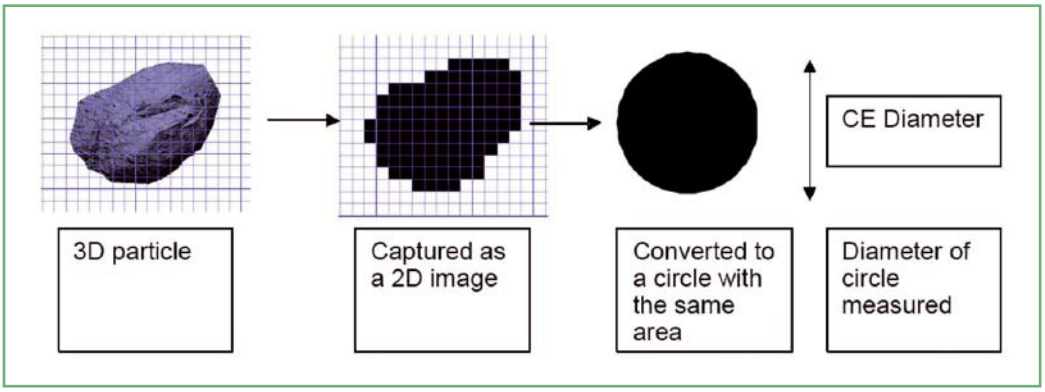



**Fig. 1: Calculation of CE diameter**

A knowledge and understanding of particle shape as well as size is essential in the development and control of many of today's industrial processes. Particle characterisation using image analysis-based systems can provide real insight into the nature of particles in a process. It gives a better understanding of their behaviour, from initial development through to production and final QC.

In the pharmaceutical industry for example, measuring both the particle size and shape distribution of many raw materials is important in quality control. From an intellectual property perspective this information can be used to tighten the specification on a particular product and make it difficult to replicate. This type of detailed analysis is possible thanks to a new generation of automated analysers, the best of which provide size and morphological information through imaging particles and generating a variety of quantitative shape data. Importantly, their full automation enables both high throughput sample preparation and the rapid analysis of extremely large numbers of particles.

## In the Beginning…

Particle size is a long-established parameter in the development and production of a wide range of materials and is measured and controlled at many different stages of research, development, manufacture and quality control. Particle shape is now assuming a similar importance, driven by the more recent advent of technology that enables its rapid and reliable measurement.

For many years manual microscopy was the only practical method of determining particle shape. However, it suffered enormously

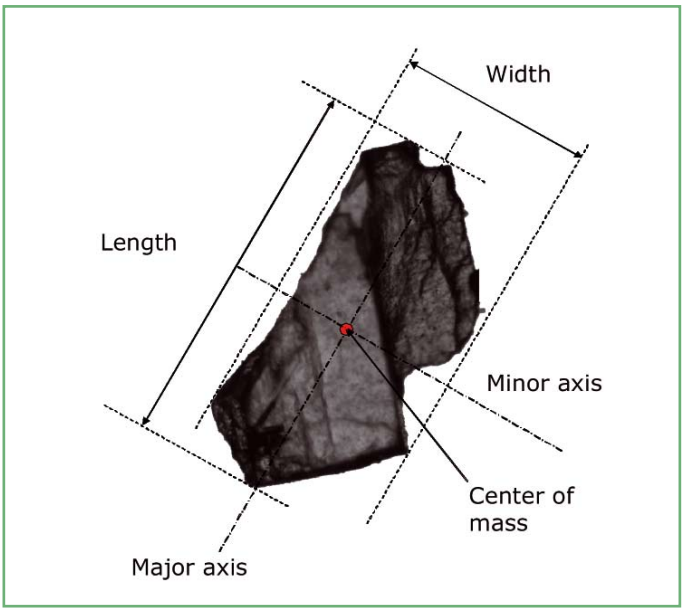



**Fig. 2: Key dimensions**

from being both highly labour-intensive and subject to operator bias: and measuring a statistically significant number of particles is almost impossible. Now, modern image analysis-based alternatives overcome these problems. They combine state-of-the-art hardware with sophisticated data analysis, for high-level automation, delivering information on both particle size and shape.

## Why Particle Shape is Important

Even very small differences in particle size or shape can have a significant effect on the properties and performance of materials. In pharmaceuticals, for example, bioavailability, flowability, stability, blending or tabletting efficiency may all be affected. Measuring particle size alone may not be sensitive enough to identify the important but subtle differences between samples. Particles having very different shapes but the same area may be characterised as identical.

Applying image analysis techniques is a highly effective way of measuring both particle size and shape. A strong element of "seeing is believing" brings the comfort of being able to view every single particle. Furthermore, particle characterisation by image analysis is complementary to many of the more established particle sizing methods such as laser diffraction. So, not only does it provide additional information, it also offers a means of validating other techniques.

## Size and Shape Definitions

Describing a 3-dimensional particle can be much more complex than it first appears. While it can be convenient to use a single number, if the particle is not a perfect sphere there are many different ways in which its size could be described. Image analysis captures a 2-dimensional image of a 3-D particle, from which it calculates various size and shape parameters. Principal among these is circle equivalent (CE) diameter, which can be used to calculate particle size. The captured image is converted to a circle of equivalent area (fig. 1) and while differently shaped particles will influence the CE figure, it has the virtue of being a single number that becomes larger or smaller as the particle does so. Importantly, it is both objective and repeatable.

Particle shape is an even more complex challenge and many parameters may be used to build a complete picture. Figure 2 and table 1 together show just some of the size and shape parameters that can be calculated. The calculation of multiple shape parameters for every particle, and the generation of distributions for each, allows the identification and quantification of even subtle differences.

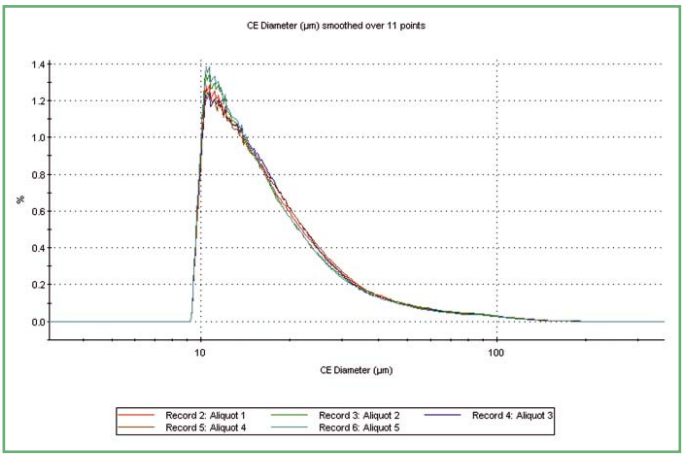
## Sample Preparation Challenges

A significant challenge in making particle characterisation by image analysis a routine operation is the need for fast and consistent sample preparation. Sample preparation must be reproducible and sample must be homogeneously distributed, usually on a glass slide or plate which is then placed on an x-y stage for imaging. Reproducible sample preparation will ensure that results are repeatable while homogeneity allows the option of analysing a subsection, knowing that it is representative of the whole. This translates into shorter measurement times and higher throughput, making image analysis suitable for routine use.

Dry powders pose a particular challenge, requiring strict control of dispersion conditions. Reducing sample preparation times and eliminating user bias demands a high level of automation. One highly successful solution is the integration of a sample dispersion unit (SDU) within the particle characterisation system itself. Figure 3 shows overlays of (a) CE diameter size distributions and (b) shape distributions for five separate aliquots of lactose, illustrating the repeatability of sample preparation using such a unit. (Samples were prepared and analysed on the Malvern Instruments Morphologi G3, with integrated SDU.)

**Table 1: Size and shape parameters (Morphologi G3)**

| Parameter | Example value | Definition |
|---|---|---|
| CE diameter (µm) | 904.14 | The diameter of a circle with the same area as the particle |
| Length (µm) | 1306.14 | All possible lines from one point of the perimeter to another point on the perimeter are projected on the minor axis (axis of maximum rotational energy). The maximum length of these projections is the length of the object. |
| Width (µm) | 678.54 | All possible lines from one point on the perimeter to another point on the perimeter are projected across the minor axis. The maximum length of the projections is the width of the object. |
| Max. Distance (µm) | 1318.07 | Largest distance between any two pixels in a particle. |
| Perimeter (µm) | 3619.42 | Actual perimeter of particle. |
| Major axis$^0$ | 105.52 | Axis of minimum rotational energy. |
| Area (µm$^2$) | 3761550.78 | Actual area of particle in sq. microns. |
| Area (pixels) | 215018 | Number of pixels in a particle. |
| Circularity | 0.785 | Circumference of equivalent area circle divided by the actual perimeter of the particle $=2\sqrt{(\pi \text{ Area})}/\text{Perimeter}$ |
| HS Circularity | 0.616 | High sensitivity circularity (circularity squared) $= 4\pi \text{ Area}/\text{Perimeter}^2$ |
| Convexity | 0.919 | Convex hull perimeter divided by actual particle perimeter |
| Solidity | 0.905 | Actual particle area divided by convex hull area. |
| Aspect ratio | 0.519 | Width divided by length |
| Elongation | 0.461 | 1 – aspect ratio |
| Intensity mean | 61.310 | Average of all the greyscale values of every pixel in the particle |
| Intensity standard deviation | 29.841 | Standard deviation of all the grey scale values of every pixel in the particle |
| Center x position (µm) | 6898271.5 | x co-ordinate of center of mass of particle |
| Center y position (µm) | 1797186.3 | y co-ordinate of center of mass of particle |



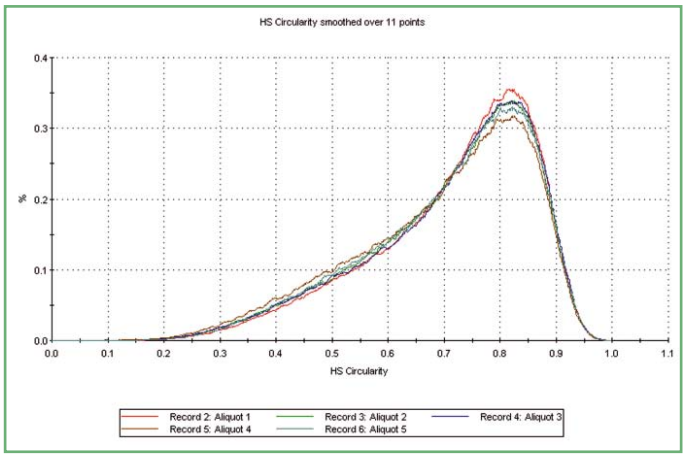




**Fig. 3: Reproducible sample preparation using an integrated SDU**
**a) Overlay of CE diameter size distribution for five lactose aliquots**

**b) Overlay of HS circularity distribution for the same aliquots**

## Sophisticated Analysis

The enormous amount of data produced on a very large number of individual particles makes possible the quick and easy identification of the most important morphological parameters for differentiating samples, and the objective characterisation of changes in a product or process. Translating this data into usable information requires sophisticated analysis, presentation and reporting software. The ability to compare and cluster in order to find differences or similarities between multiple measurements is invaluable. Clear visualisation of measurement data, the facility to plot scattergrams using any size and shape measurement, to filter on any parameter and to group and classify information, are tools offered in the latest systems.

## Not Only Dry Powders…

The type of extended particle characterisation described above is important in a diverse range of industrial applications. While many samples will be presented as dry powders, the technique is by no means confined only to this sample type. This is illustrated in the examples below, which are drawn from quite different industries.

### Pharmaceutical Ointments

Another application for the use of image analysis is the monitoring of needle-shaped particles in commercially available paraffin-based ointments. This product contains an active pharmaceutical ingredient (API) known to form long, thin needle-shaped crystals in equilibrium. Microscopy-based quality control specifications for the product require that a 3 mg sample should contain no particles longer than 250 µm in length and no more than 100 particles over 100 µm. Manual microscopy takes in excess of two hours per sample, but is reduced to just 15 minutes using automated image analysis. The needle-shaped crystals are identified, measured, counted and classified without operator subjectivity. In addition, a permanent record of all measured particles is generated whereas with manual microscopy it is possible to photograph only a small number of representative fields.

### Solder Particles

Used extensively in the manufacture of printed circuit boards, solder paste is a suspension of solder particles in a flux-containing vehicle. When screen printed on to circuit boards the particles must fuse to form a single mass. As well as the composition of the metal, the size and shape of the particles are important. In general highly spherical solder particles are needed, both to ensure optimum distribution of chemical components (low surface area to volume ratio) and to avoid blockages in the screen or stencil through which the paste is printed.

Solder particles are normally classified for size by sieving through meshes. Particle characterisation using image analysis allows improved size and shape monitoring for quality control purposes. Both parameters can be determined in a single measurement and, using filter and classification functions in the instrument software, the proportion of misshapen or oversized particles can be identified. Figure 4 compares the proportion of spherical particles to misshapen or fused particles for three different solder samples. Individual images for every particle allow further investigation if required.

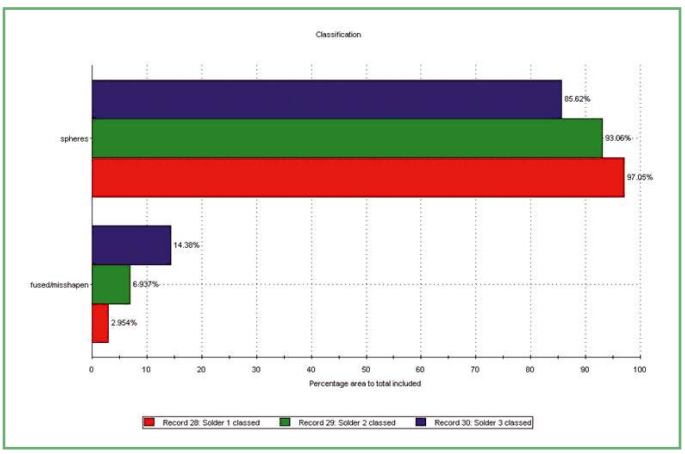



**Fig. 4: Classification result comparing the proportion of spherical particles to misshapen/fused particles within each of the solder samples**

## Conclusion

Knowledge and understanding of particle shape as well as size is now essential for many applications. The advent of automated image analysis-based particle characterisation systems, with powerful measurement and data analysis capabilities, is allowing the more widespread use of this technique for production and process management and for quality control. Automated sample preparation, integrated as part of the measurement process, is proving to be one of the major keys to even higher throughput, especially for dry powders, and is helping to establish the more widespread and routine use of this technique.

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