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When “How Big Are My Particles?” Isn’t a Simple Question

How particle counting rules change size distribution measurements

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If you work with particulate materials, you have probably needed a particle size distribution at some point, whether for quality control, regulatory compliance, or to understand why a batch did not perform. The answer depends on how the particles are counted as well as on the instrument.

The counting problem

A 2022 paper by researchers at BAM (the German Federal Institute for Materials Research and Testing) and the European Commission’s Joint Research Centre took one electron microscopy image of zinc oxide particles and applied four legitimate counting approaches. The counts ranged from 2 to 104 particles. The mean particle size ranged from 43 nm to 255 nm.

Why it matters

For regulatory purposes, the 100 nm threshold often decides whether a material is classified as a nanomaterial, which triggers additional testing, labeling, and compliance requirements. Depending on which counting method applies, the same material can fall on either side of that line. The choice also affects anyone trying to understand a material:

- Manufacturing engineers troubleshooting batch-to-batch variation.
- R&D teams correlating particle characteristics with performance.
- Quality control labs setting meaningful specifications.

If you compare results across suppliers, labs, or time and do not know whether the same counting method was used, the comparison may not be valid.

The four counting rules

The paper identifies four counting rules used across standards and regulatory frameworks:

1. CR1 counts only individual, separated particles and ignores anything clumped together.
2. CR2 counts each clump as a single particle, regardless of size.
3. CR3 resolves agglomerates into their components but treats fused aggregates as single units.
4. CR4 resolves everything possible and counts every identifiable particle within agglomerates and aggregates.

ISO standards tend to use CR3. EU regulatory definitions for nanomaterial classification require CR4. Each answers a different question.

Effect of the counting rule

Table 1 shows results for the same TEM image of zinc oxide particles under each rule. The mean size falls above or below 100 nm depending on the rule, which directly affects nanomaterial classification.

Counting rule	Particles counted	Mean minimum Feret diameter (nm)
CR1 (individual only)	2	43
CR2 (clumps as one)	7	255
CR3 (resolve agglomerates)	41	113
CR4 (resolve all)	104	73

Table 1. *Counts and mean minimum Feret diameters from one TEM image of zinc oxide particles under four counting rules (Bresch et al., 2022).*

What this means for your analysis

A request for particle size distribution data should start with the decision the data will support, not with the choice of XRD or SEM imaging.

- For regulatory classification, you need to know which jurisdiction's rules apply.
- For process troubleshooting, you need consistency with your historical data.
- For supplier qualification, you need agreement on the method before comparing results.

Cubic Labs offers image segmentation and particle size distribution analysis from SEM images collected at Purdue University. I start by asking what decision the numbers will inform and agree on the counting rule before measuring, so that you know what the numbers mean when they arrive.

Reference

Bresch, H.; Hodoroaba, V.-D.; Schmidt, A.; Rasmussen, K.; Rauscher, H. Counting Small Particles in Electron Microscopy Images: Proposal for Rules and Their Application in Practice. *Nanomaterials* 2022, 12, 2238. DOI: 10.3390/nano12132238 (open access).

Harald Bresch and Vasile-Dan Hodoroaba work at BAM (Bundesanstalt für Materialforschung und -prüfung) in Berlin, where they develop reference materials and measurement methods for nanoparticle characterization. Kirsten Rasmussen and Hubert Rauscher work at the European Commission's Joint Research Centre on the science behind EU nanomaterial definitions and policy.

About Cubic Labs

Cubic Labs LLC analyzes materials by powder XRD, XRF, and SEM-EDX at Purdue University and helium pycnometry in its own lab. Client details are never shared. If you work with particulate materials and have a characterization question, send it to info@cubiclabsllc.com or call (701) 330-1911. Results stay confidential to you.