



Standard Guide for Subvisible Particle Measurement in Biopharmaceutical Manufacturing Using Dynamic (Flow) Imaging Microscopy¹

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1. Scope

1.1 Biotherapeutic drugs and vaccines are susceptible to inherent protein aggregate formation which may change over the product shelf life. Intrinsic particles, including excipients, silicone oil, and other particles from the process, container/closures, equipment or delivery devices, and extrinsic particles which originate from sources outside of the contained process, may also be present. Monitoring and identifying the source of the subvisible particles throughout the product life cycle (from initial characterization and formulation through finished product expiry) can optimize product development, process design, improve process control, improve the manufacturing process, and ensure lot-to-lot consistency.

1.2 Understanding the nature of particles and their source is a key to the ability to take actions to adjust the manufacturing process to ensure final product quality. Dynamic imaging microscopy is a useful technique for particle analysis and characterization (proteinaceous and other types) during product development, in-process and commercial release with a sensitive detection and characterization of subvisible particles at ≥ 2 and ≤ 100 micrometers (although smaller and larger particles may also be reported if data are available). In this technique brightfield illumination is used to capture images either directly in a process stream, or as a continuous sample stream passes through a flow cell positioned in the field of view of an imaging system. An algorithm performs a particle detection routine. This process is a key step during dynamic imaging. The digital particle images in the sample are processed by image morphology analysis software that quantifies the particles in size, count, and other morphological parameters. Dynamic imaging particle analyzers can produce direct determinations of the particle count per unit volume (that is, particle concentration), as a function of particle size by dividing the particle count by the volume of imaged fluid (see [Appendix X1](#)).

1.3 This guide will describe best practices and considerations in applying dynamic imaging to identification of poten-

tial sources and causes of particles during biomanufacturing. These results can be used to monitor these particles and where possible, to adjust the manufacturing process to avoid their formation. This guide will also address the fundamental principles of dynamic imaging analysis including image analysis methods, sample preparation, instrument calibration and verification and data reporting.

1.4 The values stated in SI units are to be regarded as standard. No other units of measurement are included in this standard.

1.5 *This standard does not purport to address all of the safety concerns, if any, associated with its use. It is the responsibility of the user of this standard to establish appropriate safety and health practices and determine the applicability of regulatory limitations prior to use.*

2. Referenced Documents

2.1 ASTM Standards:²

[E2589 Terminology Relating to Nonsieving Methods of Powder Characterization](#)

2.2 ISO Standards:³

[ISO 2859 Sampling Procedures for Inspection by Attributes](#)
[ISO 8871 Elastomeric Parts for Parenterals and for Devices for Pharmaceutical Use](#)

[ISO 9276-6 Representation of Results of Particle Size Analysis Part 6: Descriptive and Quantitative Representation of Particle Shape and Morphology](#)

2.3 Other Standards:

[ANSI/ASQ Z1.4-2003 Sampling Procedures and Tables for Inspection by Attributes](#)³

[ASME BPE-2014 Bioprocessing Equipment](#)⁴

² For referenced ASTM standards, visit the ASTM website, www.astm.org, or contact ASTM Customer Service at service@astm.org. For *Annual Book of ASTM Standards* volume information, refer to the standard's Document Summary page on the ASTM website.

³ Available from American National Standards Institute (ANSI), 25 W. 43rd St., 4th Floor, New York, NY 10036, <http://www.ansi.org>.

⁴ Available from American Society of Mechanical Engineers (ASME), ASME International Headquarters, Two Park Ave., New York, NY 10016-5990, <http://www.asme.org>.

¹ This guide is under the jurisdiction of ASTM Committee E55 on Manufacture of Pharmaceutical and Biopharmaceutical Products and is the direct responsibility of Subcommittee E55.03 on General Pharmaceutical Standards.

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