ISO 21501 – A Standard Methodology to Optical Particle Counter Calibration and What It Means to Cleanroom Owners

Tony Harrison and Bob Latimer

INTRODUCTION

ISO 21501 is a new family of standards describing the instruments and calibration requirements for determining particle size distribution using light interaction methods. It represents the culmination of work by instrumentation manufacturers and industry users and comes at a critical time for the life science industry with the increasing trend for real-time air particle monitoring in cleanrooms using light scattering air particle counters.

Air Particle Counters and ISO 21501

In comparison to liquid particle counters, the calibration of air particle counters presents greater challenges due to the need to generate air samples containing sub-microscopic particles of homogenous size and distribution. Although the technology of air particle counting is well understood, the ability to calibrate any two air particle counters so that they produce the same results when sampling the same air sample has proven to be challenging, bringing into question the accuracy of these instruments. ISO 21501 now delivers a calibration method that can significantly improve the repeatability and reproducibility of air particle counters.

Liquid Particle Counters and ISO 21501

ISO 21501 also applies to liquid particle counters used for determination of particulate contamination in infusions and injections. Until recently, the calibration requirements [known as “IST” methods] for liquid particle counters used to test infusions and injections were described in detail in the United States Pharmacopoeia (USP) chapter <788>. However, in the interest of international harmonization of the pharmacopoeias, the details of these IST calibration methods have been removed in order to simplify the text of USP <788>. ISO 21501 now offers an alternative to these IST tests and establishes calibration methods to ensure accurate and repeatable performance of liquid particle counters.

BACKGROUND

Optical instrumentation has been used to determine particle contamination in air and liquids in the life science industry for many years. In addition, the correlation between airborne particles and final product quality has long been recognized in the semiconductor, flat panel display and hard disk storage manufacturing industries, where improvement of air quality (reduction of particulate contamination) has led to increases in final product yield.

Differing techniques are used to determine the number and size of particles depending on the size of particles that are of interest (see Figure 1).

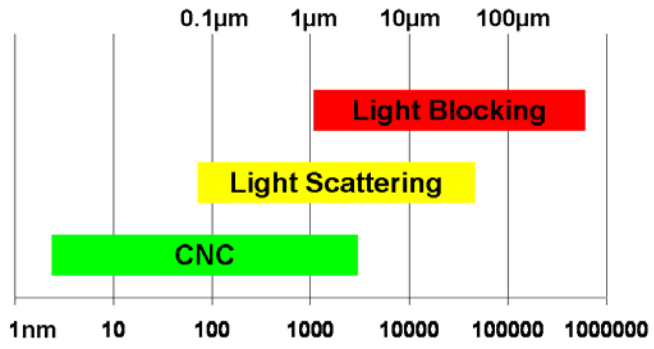

Figure 1 - Particle size range and counting techniques

In liquid particle counting for infusions and injections, the sizes of interest are =>10µm and =>25µm, whereas for the life science industry, the sizes of interest for cleanroom air particle cleanliness are =>0.5µm and =>5µm. Higher sensitivities are required for semiconductor manufacturing plants where cleanroom and

mini-environment air is routinely monitored at 0.1 µm and lower. Hard disk manufacturers typically monitor to around 0.2 µm to 0.3 µm and flat panel display manufacturing environments monitor to 0.3 µm and 1.0 µm.

USP <788>, EU 2.9.19 and JP 6.07 recognize that light obscuration is suitable for liquid particle counting in infusions and injections, whereas ISO 14644-1 recognizes that light scattering particle counters are appropriate for determining airborne contamination in cleanrooms.

There is a requirement to follow the guidelines in EU GMP and cGMP for cleanroom users that aseptically manufacture pharmaceutical products for the European and American markets. Both documents define the airborne particulate count limits for different cleanroom operations, but neither defines the methods required to determine these count limits, nor do they define the instrument to be used and how it should be calibrated. However, EU GMP states that ISO 14644-1 should be used for methodology to determine cleanroom air particle cleanliness classification and that ISO 14644-2 should be used for methodology for demonstrating continued compliance. The introduction in ISO 21501-4 states, *"Monitoring particle contamination levels is required in various fields, e.g. in the electronic industry, in the pharmaceutical industry, in the manufacturing of precision machines and in medical operations. Particle counters are useful instruments for monitoring particle contamination in air. The purpose of this part of ISO 21501 is to provide a calibration procedure and verification method for particle counters, so as to minimize the inaccuracy in the measurement result by a counter, as well as the differences in the results measured by different instruments."* The scope of ISO 21501-4 states, *"Instruments that conform to this part of ISO 21501 are used for the classification of air cleanliness in cleanrooms and associated controlled environments in accordance with ISO 14644-1"*. So the importance of ISO 21501 to cleanroom users looking to follow the guidance in GMP is evident.

Equally the scope of ISO 21501-2 states, *"Instruments that meet this standard are used for the evaluation of cleanliness of pharmaceutical products (injections, water for injections, infusions), as well as the measurements of number/size distribution of particles in various liquids."* So the importance of ISO 21501 to those in the pharmaceutical industry manufacturing injections, water for injections or infusions is also evident.

WHAT STANDARDS EXIST? WHAT IS ISO 21501 REPLACING?

ISO 14644 is a widely used standard for cleanroom classification using optical particle counters. Despite the existence of ISO 14644, prior to the ratification and introduction of ISO 21501 at the beginning of 2007, there were no ISO standards dealing with calibration and performance of the optical particle counters (OPC) used to classify cleanrooms to ISO 14644. Comprehensive non-ISO standards and calibration methods guidelines did exist however and have been employed by most major particle counter manufacturers. In summary, these standards are:

- ASTM F 328-98(2003) "*Standard Practice for Calibration of an Airborne Particle Counter Using Monodisperse Spherical Particles"* (withdrawn May 2007).
- IEST-RP-CC014.1 "*Calibration and Characterization of Optical Airborne Particle Counters"* (providing actual methods to perform the calibration).
- JIS B 9921:1997 "*Light scattering automatic particle counter*", a Japanese standard which comprehensively deals with OPC design performance, most notably in the area of counting efficiency.

The counting efficiency parameter has presented the most significant variable when it came to the actual count accuracy of individual OPC's, especially air particle counters.

Counting Efficiency

OPC's typically feature a number of size channels into which particle counts are binned, each channel being calibrated to count particles greater than a specific particle size. Particle sizes are typically expressed in micrometers (µm). The term *counting efficiency* primarily refers to the ability of the OPC instrument to count particles at a specified size. Typically, calibration involves passing a continuous stream of standard, mono-

sized particles through the OPC's sensor, which results in a stream of electrical pulses, each pulse being proportional to the size of each particle. The mono-sized standard particles produce a distribution of pulse heights, the median of which is typically regarded as the appropriate channel calibration threshold for that size. Therefore, in the real world a particle exactly the same size as a given channel would have a 50% probability of being counted (see Figure 2a). As a result, OPC's calibrated in this manner are said to have a counting efficiency of 50%. Note however that this does not mean that the OPC will only count half of the particles in the real world.

ISO 21501 makes use of the specification for counting efficiency accepted in the JIS B 9921 standard. This states that the counting efficiency should be 50% +/-20% (i.e. between 30% → 70%) in the first channel (Figure 2a). Additionally, particles of between 1.5 X to 2.0 X the channel 1 particle size should be counted with an efficiency of 100% +/-10% (i.e. between 90% → 110%) in the first channel (Figure 2b.)

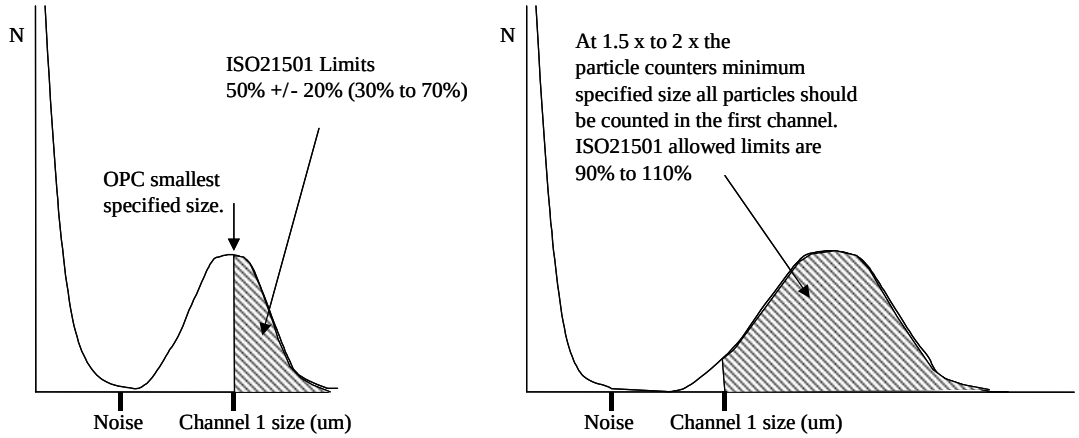

Figure 2a: The 50% calibration point **2b: Verifying 100% efficiency at a higher size**

WHY WAS A NEW STANDARD REQUIRED?

Prior to ISO 21501, it was not required that counting efficiency be checked at each calibration interval. There are many things that can impact counting efficiency during the lifetime of an OPC; for example a slight opto-mechanical misalignment of the illumination source can go undetected. Therefore, the situation exists where even though a given OPC may correctly size particles, it may be undercounting - in effect missing some of the particles. The new ISO 21501 standard requires (among other elements) that the all-critical counting efficiency element be checked during calibration. To check counting efficiency, it is necessary that once calibrated for sizing characteristics using traceable size standards, the OPC under test must be run and compared to either an Electrostatic Classifier or an OPC instrument with higher sensitivity than the OPC under test. This OPC is considered to be a "secondary standard", having been formally compared to an Electrostatic Classifier and verified as having 100% counting efficiency at the size of interest, i.e. the channel 1 size of the OPC to be certified.

The full list of elements that ISO 21501 requires to be tested in addition to the basic size calibration are as follows:

- Counting efficiency
- Sizing resolution
- False count rate
- Concentration limit
- Sampling flow rate
- Sampling time
- Sampling volume

WHAT IS ISO 21501 AND WHAT IMPROVEMENTS WILL IT BRING?

To quote from the ISO 21501 standard:

"The purpose of this part of ISO 21501 is to provide a calibration procedure and verification method for particle counters, so as to minimize the inaccuracy in the measurement result by a counter, as well as the differences in the results measured by different instruments."

Simply put, the ISO 21501 standard will ensure that OPC instruments will size and count particles correctly, using a traceable reference instrument. Different OPC models from different manufacturers will therefore closely correlate in terms of actual particle counts recorded. This presents a significant step forward in providing traceable, accurate OPC tools to classify and validate cleanrooms to ISO 14644.

WHO SHOULD ADOPT ISO 21501?

Manufacturers of products requiring processing or assembly all goods and materials within a cleanroom environment classified under ISO 14644 should require that OPC instruments be calibrated to the ISO 21501 standard. This is particularly applicable to the pharmaceutical manufacturing facilities employing sterile processing or filling lines.

Users of cleanrooms OPC's with questions or concerns regarding the transition to ISO 21501 should contact their particle counter supplier or cleanroom certifier.

ADDITIONAL INFORMATION

The ISO 21501 family of standards extends beyond air particle counters to include both scattering and extinction type liquid particle counters. The standard is split into four parts and all are available from ISO at http://www.iso.org.

ISO 21501 Determination of particle size distribution – Single particle light interaction methods -

- Part 2: Light scattering liquidborne particle counter
- Part 3: Light extinction liquidborne particle counter
- Part 4: Light scattering airborne particle counter for clean spaces

Hach Ultra manufactures a range of ISO 21501 compliant particle counters and is currently in the process of deploying an ISO 21501 field service capability for the calibration of existing products.

Particle counter owners and users with specific questions or concerns regarding IS0 21501 are invited to email the Hach Ultra ISO 21501 support team at iso21501@hachultra.com. Through this email address, one can access a panel of experts regarding ISO 21501 and receive prompt and accurate answers to questions.

CONTACT

Tony Harrison - Life Sciences Key Account Manager
Bob Latimer - Market Manager, Electronics
iso21501@hachultra.com
www.hachultra.com