



# CERTIFIED REFERENCE MATERIAL BCR<sup>®</sup> – 166

## CERTIFICATE OF ANALYSIS

LATEX SPHERES IN AQUEOUS SUSPENSION		
	Certified value <sup>1)</sup> [μm]	Uncertainty <sup>2)</sup> [μm]
Average particle diameter	4.821	0.019
1) This value is the unweighted mean of the means of 5 accepted sets of results. These sets of results were independently obtained by different laboratories. The value is traceable to the International System of Units (SI).		
2) The uncertainty is taken as the 95 % confidence interval of the certified value.		

This certificate is valid for one year after purchase.

Sales date:

The minimum amount of sample to be used is 200 μL.

### DESCRIPTION OF THE SAMPLE

The material is available as BCR-166A. Each vial contains 2 mL of an aqueous suspension of latex spheres, at a mass concentration of about 2 g/L. About 0.5 % of the particles are agglomerated doublets. Additional information on the material is given in the report.

### NOTE

This material has been certified by BCR (Community Bureau of Reference, the former reference materials programme of the European Commission). The certificate has been revised under the responsibility of IRMM.

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Signed: \_\_\_\_\_

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## **ANALYTICAL METHOD USED FOR CERTIFICATION**

Array sizing by optical microscopy

## **PARTICIPANTS**

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## **SAFETY INFORMATION**

The usual laboratory safety precautions for working with particles in the  $\mu\text{m}$  range apply. Drying of the material should be avoided.

## **INSTRUCTIONS FOR USE**

- Array sizing was adopted as a procedure of measurements for the certification of particle diameter.
- The method involved selecting straight alignments of particles in two-dimensional arrays without any visible imperfections, or selecting alignments of similar particles in a capillary.
- This preselection of particles resulted in fact in investigating the particles with the highest frequency in the narrow size distribution of latex suspension. The diameter which is certified corresponds actually to the mode of the size distribution.
- The certified reference material is therefore well suited for use in instruments which measure a large number of particles and provide a histogram or distribution curve.
- An aliquot containing 5 000 particles can be considered as a representative sub sample exhibiting the same characteristics as the parent particle population.
- When using the latex suspensions for flow-through counting (e.g. electrical sensing zone instruments) it is recommended to employ the diluent specified by the instrument manufacturer.
- The latex particles are particularly suited to verify the instrument functioning, to assess its sensitivity, its resolution power and to test its stability. The reference material can only be used for calibration when the particles under investigation have physical properties close to those of latex spheres (including size, shape, deformability, conductivity). Further information relevant to the use of latex particles in cytology and haematology can be found in chapter 13 of the report.
- The spheres can also be used with other types of instruments e.g. with microscopes as particle size standards. Here the user should examine a sufficiently large proportion of particles to select those representative of the central part of the size distribution. If only a few particles are considered and used as particle size standards, a careful evaluation of the uncertainties must be made. In addition to the uncertainty related to size distribution estimated  $\pm 2\%$ , the user shall assess other possible contributions to the uncertainty due to the observation e.g. ambiguity of edge location.
- Measurements with an electrical sensing zone counter equipped with hydrodynamic focusing show that between 98 and 99 % of the particles have a distribution close to a Gaussian distribution. 1 to 2 % are irregular particles. 99 % of the particles which are normally distributed have a size which does not differ by more than  $\pm 2\%$  from the mean size. Detailed information on the parameters characterizing the particle size distribution can be found in chapter 10 of the report.
- The particles are stable in aqueous solution (with surfactant) but their stability is not demonstrated after drying. Stability of latex particles in electron microscopy is not guaranteed in all operation conditions.
- In all applications for electron microscopy the aliquots taken from the vial shall be washed with distilled water, in order to remove excess of surfactant.
- The following procedure can be employed:
  1. Place 200  $\mu\text{L}$  of latex particle suspension in a container.
  2. Centrifuge in order to collect the solids in the bottom and not onto the walls of the container.
  3. Carefully withdraw the supernatant from the solids. The remaining volume of sediment would be less than 5  $\mu\text{L}$ .
  4. Resuspend the particles with 200  $\mu\text{L}$  of distilled water.This washing operation should be repeated three times.

## STORAGE

Upon receipt samples must be kept at room temperature and in the dark. A stabilizer (surfactant) has been added to prevent flocculation of the particles. When opened, it is recommended to protect the suspension remaining in the vial against microbial growth by an adequate bactericide-fungicide. However, the European Commission cannot be held responsible for changes that happen during storage of the material at the customer's premises, especially of opened samples.

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## NOTE

A technical report on the production of BCR-166 is available on the internet (<http://www.irmm.jrc.be>). A paper copy can be obtained from IRMM on request.