Evaluation of Galai CIS-1 for Measuring Size Distribution of Suspended Primary Particles in the Ocean

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ABSTRACT

CIS-1 is a particle sizer based on time-of-transition principle. It scans particles with a laser beam, and determines their sizes by measuring the time lengths of light obscuration by the particles. Studies have shown that CIS-1 provides good size information for standard reference spherical particles and for homogeneous material. This study examines the accuracy of its size information by using field particles obtained in the ocean. These particles differ from the above materials in that field particles have a wide range of shape and optical properties; they may affect the performance of a particle sizer. The volume-based cumulative size distribution of primary particles in 8 offshore and 4 coastal water samples were obtained by CIS-1 and by microscopy. Their comparison generally showed good agreements. The maximum difference in the median size of the same sample was 1 µm, or 17%, and the standard deviations and the modes also were close. The average relative size deviations between the two methods varied between 2% and 17.3%. The mean of the average relative deviation among all samples was 9.7%. The results of this study also showed that the distributions of primary particles in all samples were highly similar, irrespective whether they were from offshore or coastal waters. There were no primary particles larger than 16 µm, and their median sizes were within the range of 4–6 µm. By averaging the size distributions obtained for all samples, the median size and the mode of the resulting distribution were 4.7 and 5.5 µm, respectively.

1. INTRODUCTION

Suspended particles in the ocean consist of organic matter, skeletal debris and mineral grains. They are the major source of the bottom sediments. In addition they are known to be associated with pollutants in the ocean. The transport of suspended particles offers a pathway of the transport of pollutants. More recently, the sinking of suspended particles has been increasingly recognized as an important process in the transfer of CO₂ from the atmosphere to the ocean bottom. Hence, there is an intensifying interest in understanding the origin and characteristics of suspended particles.

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Particle size distribution is a fundamental characteristic of suspended particles. Several methods have been used to determine size distributions of suspended particles in aquatic environments. Microscopy was used by Honjo et al. (1974) and Yamamoto (1979). Paffenhofer et al. (1980), Kranck (1980) and McCave (1983) studied particle size spectra by using Coulter counter. Photographic and holographic techniques were also adopted by Eisma et al. (1983), Kranck (1984), Asper (1987) and Gibbs et al. (1989) for floc size studies. All these methods have their own limitations. Microscopy has long been recognized as a standard method, but it requires a lengthy operator time. Measurements done on Coulter counter may need considerably less time than microscopy, but aggregated flocs may be broken up during the measurement (Gibbs, 1982). Size range of each technique is also an important consideration. A significant amount of particles in the ocean are smaller than 50 µm, but the photographic method has only a resolution of 50 µm. Although the holographic technique can record particles as small as 5 µm, Gibbs et al. (1989) incorporated microscopy to analyze particles less than 10 µm.

In recent years, due to the progress of laser optics technology, size distribution can now be measured by using laser-based particle sizer. There are several advantages in using laser particle sizer. With this type of instrument, operator time can be greatly reduced (particularly, in comparison to microscopy). Laser sizers have a wide size range, typically between 0.5 µm and about 1800 µm, and their measurements can be conducted on board ship. In addition, since only the laser beam comes into contact with the particles, this type of sizer can be designed such that the disruption of particle structure is minimal. All these advantages make laser sizer particularly suitable for measuring suspended aggregated flocs. Bale & Morris (1987) developed a submersible instrument based on a laser particle sizer which converts Fraunhofer diffraction spectrum to particle size spectrum. It was successfully tested in situ in estuarine waters. However, laser sizers based on Fraunhofer diffraction principle have a limit of particle concentration below which insufficient amount of light is diffracted for meaningful measurements. Since, in general, the particle concentration in the offshore area is below that limit, this limitation precludes the application of this type of instrument in the open ocean.

Galai CIS-1 is also a laser-based particle sizer, but it operates on a different principle, called time-of-transition. It measures particle size by detecting the time of obscuration of laser beam by particles. With this detection principle, there is no lower limit on particle concentration. Thus, CIS-1 appears to be a viable instrument for size determination on suspended particles in the ocean.

However, before using CIS-1 for this application, one needs to know the degree of confidence on the measurements of this instrument. Accuracy of any particle sizer can be easily demonstrated, if measurement is conducted using standard spherical particles. Krasikov et al. (1988) have shown that measurements of standard polystyrene divinylbenzene spheres (Duke Scientific, California) and BCR (Community Bureau of Reference) quartz powders by CIS-1 correspond well with the reference diameters and
the certified size distribution for the quartz. Moreover, Krasikov et al. (1987) compared the size of one chalk powder sample measured by the CIS-1 laser sizer and by microscopy. They found that the laser size distribution corresponds well with that obtained by microscopy. Although these reports provided some insight to the accuracy question, their results were obtained by testing homogeneous material albeit with various sizes. Further clarification on the accuracy question is needed for suspended particles in the ocean, since they consist of numerous kinds of material with different compositions and optical properties. This study investigated the accuracy question with specific emphasis on suspended particles in the ocean by comparing size distributions obtained by the CIS-1 with that obtained by microscopy. In effect, this study represents the first effort to measure sizes of in situ suspended particles in the ocean by utilizing this instrument.

Since suspended particles often exist in the water as aggregated flocs of primary particles, particles may be in different aggregated states as they are analyzed by the two different methods. To prevent this, particles were disaggregated to primary particles before analysis. After size distributions were measured, several statistical parameters obtained from the distributions were compared.

2. DESCRIPTION OF THE CIS-1

Galai CIS-1 (Computer inspection system) employs dual techniques for particle inspection: one is laser-based and the other is video microscopic. The laser-based technique, called time-of-transition, measures the time of interaction between the particles and the laser beam as it scans across the particles. In the laser-based system, the optical set-up consists of a 2 mW He-Ne laser, a rotating wedge prism, a focusing lens and a photodiode detector. The laser beam passes through the wedge prism and then is focused by the lens onto the measuring area in the sample cell. Directly behind the sample cell is the photodiode for light intensity detection. This system provides a fine laser beam scanning the measuring area, where the spot size of the laser is 1.2 \( \mu m \) and the scanning diameter 600 \( \mu m \). As the particles in the measuring area interact with the laser spot, the detector generates pulse signals. Since the spot scans at a known speed, the particle size can be computed from the pulse signal (Aharonson et al., 1986; Karasikov et al., 1988).

Figure 1 shows the pulse signals for the cases of particles larger and smaller than the laser spot. As the scanning beam starts to interact with the particle, the scanning beam is increasingly blocked by the particle and the pulse signal decreases with time. Then the signal becomes horizontal at its minimal value when the whole particle blocks the laser beam. Finally, the signal increases with time towards its full strength as the scanning beam leaves the particle. The intervals indicate on the time axis are the times for the scanning beam to travel the distance of one diameter of the particle it is interacting with. Naturally, they are the time intervals corresponding to the sizes of the particles. When the particle is larger than the laser spot, the light can be totally blocked, while the particle is
smaller than the spot the decrease of light intensity is proportional to the diameter of the particle. For the particles much larger than the beam spot, the pulse becomes near rectangular and its width is a measure for the particle. The upper range of the particle size that this system can measure is limited by the diameter of the scanning circle. It must be much larger than the particles so that only a limited arc intersects the particle. With a scanning diameter of 600 µm, the measuring range is 0.5 to 150 µm. By changing the focusing lens and/or wedge prism, the measuring range can be changed to 2–300 µm, 5–600 µm or 10–1200 µm.

Although the time of interaction can directly provide information on particles sizes, two situations may produce incorrect size information. The width of the laser beam varies along its path, and the beam width is smallest at the focusing area. For the particles which are not located at the focusing area, the interaction signal will be longer than that when they are. Secondly, the measured size will be smaller than the actual value, if the laser beam does not intersect the center of the particle. In order to avoid these problems, this system employs algorithms to selectively exclude signals produced by off-focus and off-center particles.

The video microscopic system comprises a microscopic lens, a CCD camera and a monitor. The microscopic image observed by the camera is picked up by a frame grabber and passed to an image monitor. Since the video camera is mounted in perpendicular to the path of laser light, it can be used to observe the particles suspended in the sample cell while the particles were analyzed by the laser-based system. Two dimensional shape analysis can be performed on the video image to obtain various shape and size information by using image analysis computer programs.

3. METHODS

Eight offshore water and four coastal water samples were used in the analysis. The offshore samples were obtained from the area northwest of Keelung at various depths on
CHENG-HAN TSAI AND SU-REN RAU

boards R/V Ocean Researcher I and coastal samples from the sea surface along the north coast of Taiwan (Figure 2). Sites for coastal samples 9, 10 and 11 were of rocky coasts and the 12th site was a beach. Among all samples only the beach sample contained few grains of beach sand. Since their number was low, beach sands could not be uniformly distributed into all sub-samples. Hence, these sand grains were deliberately left out of the analysis.

A portion of each water sample was examined by CIS-1 for direct particle size measurement. The water was first ultrasonically and mechanically agitated to break up any aggregated flocs. Sub-samples, 2 ml each, were then taken from the same agitated sample and placed in sample cells. Magnetic spin bar was placed in the cell, to prevent particles from settling. To ensure good data quality, a confidence level of 95% for measurements was selected. The chosen confidence is the probability that the measured mean size (volume based in this study) of the sample particles is within ±2.5% of the true mean. Hence, the instrument continued scanning the sample until this confidence level was reached.

Fig. 2. Locations of sampling sites.
Microscopy was carried out using 4.5 µm filtered particles. To ensure that the particles filtered were disaggregated primary ones, the water samples were also treated with ultrasonication and mechanical agitation. Twenty to fifty ml of agitated water were filtered. This amount of water was found to produce the best images for microscopic examination. Too much or too little water would either produce too many overlapping particles or too few particles for observation. After filtration the filters were thoroughly rinsed with double filtered distilled water to remove sea salt. Then they were promptly dried and kept in petri dishes to keep airborne particles from settling on the filters. A section of each filter was cut out, placed on a microscope slide and rendered transparent by immersion oil for microscopy. A separate microscope with x40 stage lens was used for examination. Microscopic images were then fed electronically to the video system of the CIS-1 for analysis. It was found that the smallest particle that can be detected by this set-up was about 0.5 µm.

In order to carry out video image analysis, the microscopic image was first converted to black and white by properly adjusting the acceptable gray level on the converted picture. Then the Feret’s diameter, the average of distances between pairs of parallel tangents to the projected outline of each particle, was measured. In the present study, Feret’s diameter was determined as the mean of four line lengths with angles of 0, 45, 90 and 135 degrees, respectively (Figure 3). During the image analysis, a few aggregated flocs were found on some filters. Apparently, they were the ones not broken up by the agitation; they were not counted in this analysis.

4. RESULTS

An assessment of the measurement techniques employed in this study was conducted by analyzing standard polystyrene divinylbenzene spheres with a nominal diameter of 10 µm. The resultant volume-based size distributions obtained by CIS-1 and microscopic procedure were shown in Figure 4. As can be seen, the analysis by microscopy was extremely accurate; about 98% of the volume was contained in the size range of 10.2 µm to 10.6 µm, and the median diameter was 10.4 µm. The median diameter measured by the CIS-1 was 10.9 µm, which was also quite close to the nominal diameter. However, the CIS-1’s distribution spread over a wider size range. The coefficient of variation calculated from this distribution was 15.5%, which was close to that obtained by Karasikov et al. (1988). In their study similar standard spheres with nominal diameters of 200 µm and 646 µm were analyzed, and the coefficients of variations were 14.4% and 20.3%, respectively.

Shown in Figure 5 are the cumulative distributions measured by CIS-1 for 6 subsamples from the sample 7. This figure demonstrated that the distributions exhibited large variations among them. Allen & Davies (1989) compared the performance of most commercially available particle size analyzers. They also found in most cases CIS-1 had the
Fig. 3. Feret's diameters at four different angles (Sela, 1991).

Fig 4. Size distributions obtained by microscopy (dashed line) and by CIS-1 (solid line) for standard spheres with a nominal diameter of 10 $\mu$m.
lowest reproducibility. As a matter of fact, low reproducibility is inherent in this type of instrument. Since for any particles to be measured, they have to circulate through a tiny scanning zone which has a diameter of only 600 µm. As a result, when the particle concentration is low, particles in the sample cell are difficult to have even chances of being scanned. Variations between measurements can be reduced, however, by increasing particle concentration. As this was not possible for this test, the average of size distributions for multiple sub-samples had to be used. More distributions that were averaged means that more particles were taken into account, and the averaged distributions should be more stable. Hence, the averages of 6, 8, 10, 12 and 15 distributions were compared (Figure 6). It can be seen that they all collapsed nicely to a single distribution, indicating that it was possible to obtain a stable distribution. Consequently, based on this result, the averages of 6 sub-samples was taken as the representative distribution for all samples.

A similar evaluation procedure was carried out for microscopy to determine the minimal number of particles to be analyzed. The resulting distribution of various particle counts for sample 12 are plotted in Figure 7. The results showed that the distribution approached a stable form as the particle count reached over 1000. It appeared that the predominant effects of increasing particle count over 250 were to smooth out some zigzags of the distribution and to increase the percentages of smaller particles. Although
Fig. 6. Mean distributions over various number of distributions of the sample 7 obtained by CIS-1.

Fig. 7. Size distributions of sample 12 obtained by microscopy using various numbers of particle counts.
the distribution of 250 counts did not differ greatly from that of 1500 count, the microscopic distributions reported here were generated by analyzing about 1200 particles.

Shown in Figure 8 are the distributions obtained by CIS-1 (solid line) and by microscopy (short-dashed line) for all 12 samples. By comparing the distributions between measuring techniques, one can see that, except for the sample 6, CIS-1 detected articles larger than 16 µm in all samples. In contrast, microscopy’s distributions did not extend beyond 16 µm; this was consistent with the exclusion of aggregated flocs in their analytical procedure. This also suggested that some flocs were present in CIS-1’s sample, and they should be excluded for the sake of comparison. Hence, the CIS-1’s distributions were truncated at the size of the largest particles measured by microscopy for each sample, and they were also plotted in Figure 8 (long-dashed line). For most samples, the truncated distributions had higher percentages of smaller particles than microscopic distributions had, and the former ones were smoother than the latter ones. Moreover, it can be seen that the distributions obtained by the two different methods generally followed each other in shape quite closely.

For a more detailed comparison, the following statistical parameters were examined: the median (M), the standard deviation (SD), and the mode (MD) of each distribution and the average deviation between 10th and 90th percentiles (V) between two distributions. The median and the mode are measures of locations. The median expresses the central tendency of the sizes. Although, arithmetic mean is also a measure of location, median is a better expression for the average size, since the mean size can be greatly influenced by the rare occurring extreme values. The standard deviation is a measure of the size dispersion (or sorting). Its value was calculated according to the following equation:

$$SD = \left[ \sum_i \left( X_i^2 \frac{P_i}{100} \right) - \left( \sum_i X_i \frac{P_i}{100} \right)^2 \right]^{1/2} \quad (1)$$

where, $X_i$ is the size of the ith size interval, $P_i$ the volume percentage of the ith size interval and $\Sigma$ is carried out from $i=1$ to 20 (i.e. 20 size intervals).

The average deviation, V, is defined as (Allen & Davies, 1989; Bikker & Konert, 1992):

$$V = \frac{1}{9} \sum_i \left| \frac{X_c(i) - X_p(i)}{X_p(i)} \right| \times 100 \quad (2)$$

where $X_c(i)$ and $X_p(i)$ are the percentile sizes of i=10%, 20%, ..., 90% for the truncated distributions of CIS-1 and that of microscopy, respectively. Thus, V is an average relative deviation of the sizes measured by CIS-1 from that by microscopy irrespective of whether the deviation is positive or negative.
Fig. 8. A comparison of size distributions obtained by the two techniques. (solid line, CIS-1; short-dashed line, microscopy; long-dashed line, truncated CIS-1).
These statistical parameters of all samples are summarized in Table 1. The median size obtained by microscopy varied from 4.1 µm to 6 µm, while that obtained by CIS-1 ranged from 4.2 µm to 5.4 µm. In most cases, the median sizes of the microscopy were larger than that of CIS-1. Sample 8 had the largest difference of 1 µm, or 17%, and samples 3, 4, 11 and 12 had the smallest differences, 0.1 µm or 0 µm. Comparing the standard deviations between the two methods, it can be seen that there was no definite trends to suggest which method produced larger dispersion. Overall, there were good agreements for this parameter. The larger differences in standard deviations for samples 1, 3 and 10 were attributed to the differences of distributions in larger sizes. The agreements between the modes of the distributions were rather good. Only the modes of the sample 8 differed significantly, 8.5 µm from microscopy and 5.5 µm from the CIS-1.

The average deviation, V, unlike the deviation in the median size alone, takes into account the differences at various percentiles. As listed in the table, the V value varies from 2.0% to 17.3%, with an average of 9.7% over all samples. There were 7 samples whose V values were larger than 10% and only 3 samples less than 5% (samples 3, 4 and 11). From Figure 8, one can also see that the distributions for the latter three samples have the best comparison.

It is possible to get a comparative sense for the V values obtained here. Allen & Davies (1989) analyzed the size distribution by CIS-1 using BCR standard quartz powders. They reported that the average deviation relative to the standard sizes of BCR 70 (0.5 µm to 12 µm), BCR 67 (3 µm to 20 µm) and BCR 69 (12 µm to 90 µm) were 28.4%, 14.7% and 2.2%, respectively. Their average deviations were generally close to those reported here. However, the V value for BCR 70, which was in the same size range

Table 1. Comparisons of various parameters derived from volume based size distributions of suspended primary particles measured by CIS-1 and microscopy. (D=median size, SD=standard deviation, MD=mode, V=average relative deviation; subscripts: 'p'=microscopy, 'c'=truncated CIS-1)

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<th>(D_c) (µm)</th>
<th>(SD_p) (µm)</th>
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as that of particles in this study, was much higher than that shown in Table 1. This suggests that the overall comparison of size measurements between the two methods described here is good.

The size distributions obtained by either method showed a high degree of similarities between samples (Figure 8). They all bore a near S shape, and no particles were larger than 16 µm. Thus, it is clear that one single distribution can represent that of primary particles sampled in this study. The averaged cumulative size distribution for offshore waters (short-dashed line) and for coastal waters (long-dashed line) are shown in Figure 9. The median sizes were 4.9 µm and 4.4 µm for offshore and coastal waters, respectively. That is, the former had less percentage in small particles than the latter. However, the difference was not significant as these two averaged distributions were remarkably similar. Therefore their averaged distribution (cumulative one and histogram) was also plotted in the figure (solid line). As can be seen, the volume percentage in the histogram had a plateau between the sizes of 1.2 µm to 2.5 µm. After that it increased sharply towards the mode at 5.5 µm. Then the percentage decreased quickly and reached zero at 16 µm. The median size was found to be 4.7 µm. This finding is comparable to that obtained by Bale & Morris (1987) for primary particles in the Tamar Estuary. They found that no particles

![Graph showing averaged size distributions of primary particles](image)

*Fig. 9. Averaged size distributions of primary particles. (long-dashed line, coastal waters; short-dashed line, offshore waters; solid line, all samples)*
were larger than 20 \( \mu m \); the largest 30th percentile size was 1.9 \( \mu m \); and the median sizes were in the range of 6-10 \( \mu m \). More importantly, they also found that their primary particle distributions were highly uniform among samples throughout the estuary.

5. DISCUSSION AND SUMMARY

Techniques and types of instruments for analyzing particle sizes are numerous, but not all of them are suitable for measuring suspended particles in the ocean. In order to do so they should be able to measure particles with wide ranging sizes (0.5 \( \mu m \) to several \( mm \)) and optical properties and with low concentration. They should not significantly cause the breakage or further aggregation of particles to be measured and their measurements should be conducted \textit{in situ} or on board ship. Galai CIS-1, which is based on the time-of-transition principle, is a particle size analyzer which is the only commercially available and laser-based instrument that satisfies most of the above requirements, except for the size range. CIS-1’s measuring range is between 0.5 and 1200 \( \mu m \). For particles larger than 1200 \( \mu m \), photographic technique may be used. This study evaluated this instrument’s accuracy for this application by comparing its measurements with that by microscopy using mechanically disaggregated primary particles suspended in the offshore and coastal waters. A preliminary assessment of the measurement techniques utilized by this study was carried out by analyzing standard spheres. It was demonstrated that the microscopy procedure produced extremely accurate results. The size measurement obtained by CIS-1 was also quite good.

It was found (by Allen \& Davies, 1989 as well) that the CIS-1 can produce measurements with low reproducibility, especially when the particle concentration is as low as that of the samples used in this study, which is less than 12 mg/l. However, by measuring the distribution with a required confidence of 95\% for six times. The average of the six distributions was shown to be highly reproducible. This suggests that for the measurements to be reproducible, number of particle scanned should exceed a certain value. This can be achieved by increasing the measurement time for one sample or increasing the number of sub-samples for repetitive measurements. However, it is always preferable to use multiple sub-samples, since subjecting particles to the stirring actions of the magnetic stirrer for too long may cause them to change their state of aggregation. For samples with particle concentration different from that of this study, it is recommended that the procedure similar to that reported here should be used so that the minimal number of repetitive measurement for obtaining stable results can be determined. Moreover, one should always use the largest possible measuring size range. The larger the measuring range, the larger the scanning zone, and hence more particles are scanned per unit time. This would shorten the required measurement time.

For the samples studied, the median size obtained by the two methods differed at most by 1 \( \mu m \), or 17\%. The modes of the distributions generally agreed well. The average
relative deviation between 10th and 90th percentiles varied between 2% and 17.3% with its mean among all samples of 9.7%. It is well known that measurements of sizes using different principles may yield slightly different results, since different parameters are measured. Size measured by time-of-transition, in effect, is the average of lengths of scanning line on two dimensional projected outline of particles at various angles, while the size measured by microscopy reported here represents the mean Feret’s diameter at four different angles. Despite this difference, it was shown that the size distributions obtained by CIS-1 generally are not very far off from that obtained by microscopy. Hence, one should be confident enough to conclude that the sizes of primary particles analyzed by CIS-1 is generally correct.

As mentioned in the introduction, this study was only the first attempt to use CIS-1 in measuring oceanic particles. The real goal of the size study is to be able to analyze aggregated floes. Further studies on the accuracy of measuring floes in the ocean by CIS-1 and the best method in obtaining reproducible results in an acceptable time period has been underway.

It was also demonstrated that the distributions of primary particles were highly uniform among all samples. Their median sizes and modes were in the ranges of 4–6 $\mu m$ and 4.7–8.5 $\mu m$, respectively. The distribution averaged over all samples had a median and mode of 4.7 $\mu m$ and 5.5 $\mu m$, respectively. This averaged distribution may be regarded as the representative one for the primary particles in the present studying area. The significance of the distribution of primary particles is that these particles are the basic building blocks of aggregated floes.

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使用 Galai CIS-1 測量海水中懸浮基本顆粒粒徑分佈之評估

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摘 要

CIS-1 是一種利用遮蔭時間 (time-of-transition) 原理的粒徑測定儀。它使用一束光束掃描顆粒，藉由顆粒遮蔽光線的時間長短來決定顆粒的大小。現已有些研究，用 CIS-1 測量圓球狀的標準參考顆粒及均質 (homogeneous) 顆粒，他們發現 CIS-1 能夠測得令人相當滿意的數據。本文探討以 CIS-1 來測量與上述物質不同的海水懸浮顆粒大小的準確度。海水中的顆粒由具有不同光學性質及形狀的顆粒所組成，這可能影響粒徑分析儀測量結果的好壞。本研究將 8 個外海及 4 個近岸海水中懸浮基本顆粒粒徑由 CIS-1 及光學顯微鏡之影像分析所得之粒徑分佈 (體積計)作比較，結果顯示兩方法所測得之數據相差不多。粒徑中間值之最大差異為 1 µm 或 17%，而粒徑分佈之標準偏差及眾數也都很相近。至於兩方法所得之粒徑平均相對偏差 (於第十及第九十百分位數間) 則於 2% 及 17.3% 之區間內，12 個樣本之平均值為 9.7%。本研究亦發現基本顆粒之粒徑分佈於各水樣中也彼此很接近。所測得之最大顆粒沒超過 16 µm，其中間粒徑都在 4 µm 至 6 µm 區間內，若將所有水樣之粒徑分佈平均後，所得之分佈中間值及眾數各為 4.7 µm 及 5.5 µm。