Characterizing Shape of Effluent Particles by Image Analysis

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Abstract: Particle shape of wastewater effluents were investigated by implementing a dynamic particle image analyzer which provided data on the particle count, size, shape and circularity concurrently with images. The goals of this study were to characterize removal efficiency based on size and shape parameters for an in-line filtration process and to study correlation between size and shape parameters. Results showed that filter removal efficiency can be determined by various size and shape parameters, where different information is obtained for each analysis. This method has potential in evaluating treatment process efficiencies by coupling size and shape parameters into a meaningful tool.

Keywords: Particle shape, circularity, filtration, image analysis, feret diameter, effluent

INTRODUCTION

Israel is situated in a semi-arid zone with insufficient water sources, thus wastewater effluent reuse for irrigation provides a major alternative for fresh water shortage. The Israeli national water company, Mekorot, has initiated a series of research projects for producing tertiary effluents for
unrestricted irrigation (1). One of the projects includes optimization of UV and chlorine disinfection for flocculated filtered effluents. Previous studies showed that particles and wastewater flocs interfere with chemical and physical disinfection (2–5). However these studies did not directly investigate the impact of particle shape on filtration and disinfection efficacy.

Filtration is an advanced tertiary wastewater treatment used to reduce the particulate content of effluents. It is especially important to remove particles from effluents as particles can clog drip irrigation systems and enhance membrane fouling (6). Particles in the effluent are generally evaluated through analysis of turbidity. Turbidity is the most common parameter used for monitoring particles, however, it is insensitive to particles in the size of a few microns (7), and does not provide information on the size, and concentration of the individual particles in the water. Moreover, turbidity may not be the best true measure of particle removal processes. Adin (8) concluded that optimum removal of submicron particles upon alum coagulation was represented by turbidity measurement while removal of particles larger than 1 μm was measurable best through particle size analyzers. Thus, to obtain more sensitive data on particles it is necessary to employ particle counting methods that provide information on particle size distribution.

The size of irregular particles may be simplified to one parameter by defining particles as spheres and isolating the “equivalent spherical diameter” (ECD). Natural particles actually acquire a variety of complex shapes as spheres, ellipsoids, rods, plates, or combination of these. Therefore the assumption of particles as spheres is not satisfactory with particles from water and wastewater effluents as they are not spherical in nature and are frequently in aggregates.

There are a variety of methods to measure particle size such as microscopy methods including imaging techniques, light interaction methods, electrical property methods, sedimentation methods, sorting and classification methods. Comparison of particle sizing obtained by different methods is difficult. In most applications of particle distribution analysis usually the size of the particle is defined and not the shape. Microscopy and image analysis are common methods to characterize shape of particles. However, analysis of particles by microscopy is limited to a small number of images thus data is not statistically representative, data analysis is laborious and results can be subjective. The use of dynamic digital image analysis of particles is increasingly being recognized as a sensitive and rapid technique to characterize particle size, shape, count and transparency which can provide insights to particle density and composition (9, 10). This technology generates data by capturing direct images of each particle in flowing liquids through automated sample introduction, image acquisition and analysis.

The shape of particles may be correlated to the process wastewater effluents undergo. For example, activated sludge treatment normally yields particles of a well defined, typically oval shape, while wastewater reservoir effluents contain a variety of particles typically characterized by gelatinous
particle shape (8). The specific objectives of this study are to characterize particles through particle size and shape parameters (such as circularity, feret max diameter, equivalent spherical diameter and transparency) for various water samples such as effluents from an in-line filtration process with or without use of a coagulant and effluents from secondary treatment after UV and chlorine exposure. Over the last decades, depth filtration has been studied extensively with respect to particle removal (11); however, removal was usually based on size and not on other parameters such as shape. Knowledge of the number, size, and shape distribution of the particles present in water, before and after filtration, can provide designers and operators means for improving filtration efficiency. Although the shape has been stated as significant for various processes, the relationship between particle shape and size on process removal efficiency has not been widely investigated in wastewater treatment plants (WWTP) and this type of analysis may provide a new look into particles.

MATERIAL AND METHODS

Pilot Plant

A filtration pilot plant was constructed at a WWTP designed to produce effluents that follow guidelines for Israeli unrestricted irrigation (12). The pilot plant is a unit located near the actual wastewater reuse facility (Shafdan, Tel Aviv, Israel). The secondary effluent from the activated sludge basin flows into the tertiary pilot plant which is based on in-line filtration process that integrates the coagulants during the processes to reduce effluent particulate load.

The pilot plant units are high-rate deep bed filters with filtration velocities of 10–15 m/sec and a flow of 5 m$^3$/hr each one. The filters are 1.4 m high and are composed of two layers with 70 cm anthracite and 70 cm sand with porosity of 0.27 and 0.43 mm respectively. The coagulant poly aluminum chloride (PAC) at concentration of 2.3 mg/L was added directly on the filter media to improve filtration in a process termed in-line filtration while the other filter is identical but without addition of the coagulant. Samples for analysis include secondary effluent prior to filtration, samples after filtration and backwash samples. Samples after filtration will be termed “PAC” or “NPAC” for filtration with or without PAC respectively. The goal is to determine removal efficiency through particle size and shape parameters. A control system is responsible for measurement of the pressure before and after the filter that controls the backwash automatically at a backwash flow of 11 m$^3$/hr and velocity of 35 m/hr. The filtration process run is 6 to 8 hours or 300 cm of head loss.

Additional samples after secondary treatment in another WWTP (Ramat HaSharon, Israel) after UV or chlorine disinfection are held in a reservoir and examined after two weeks to study the correlation between size and shape.
parameters. These samples will be termed “Chlorine” or “UV”. Another inorganic sample taken from the soil and separated to the silt fraction was analyzed and termed “Silt”. Soil samples were collected from the North Negev region, Israel, which is characterized by high silt content. The separation method for the silt fraction was based on the piped method (13).

**Image Analysis**

Particles suspended in liquid are analyzed by the “Micro Flow Imaging” (MFI) technology (DPA 4100, Brightwell Technologies Inc, Ottawa Ontario, Canada; www.brightwelltech.com). This apparatus employs a digital camera with an illumination and magnification system to capture in-situ images of suspended particles in a flowing sample. Basically a sample fluid is drawn through a flow cell and a sections of the fluid are illuminated with LED light source at $\lambda = 475 \text{ nm}$, magnified and imaged onto a digital camera. These captured images are automatically analyzed to determine various size and shape parameters that represent the two dimensional projection of the particles.

Analysis was conducted with low magnification set point with particle analysis between 2.25–400 $\mu$m. The pixel density is 1.3 Mega pixels, and the size of each pixel is $6.7 \times 6.7 \mu m$, with a field of view of $1760 \times 1400 \mu m$ with a resolution of $0.25 \mu m$. The threshold value which separates the background from the particles is predefined; however, there is a possibility for manual threshold and background is subtracted by optimizing illumination. During the run images of particles in binary and grey scale files are obtained, with a predefined number of images captured for each run. System calibration is conducted with polymer microspheres with mean diameters of $5 \mu m \pm 0.05 \mu m$ and $10 \mu m \pm 0.1 \mu m$ with narrow size distributions and certified by manufacturer (Duke Scientific, USA).

The geometric characteristics of samples were expressed in the following parameters:

1. **ECD ($\mu m$)** is the equivalent circular diameter.
2. **Circularity** is a dimensionless number between 0–1, where circularity $= 1$ for a spherical particle and circularity $= 0$ non spherical particle. It is defined as $(\text{circumference of an equivalent area circle})/(\text{perimeter of the particle}).$
3. **Average intensity** is defined as $(\text{sum of all pixel intensities in object})/ (\text{Number of pixels})$. A pixel with intensity greater than the predefined threshold will be considered by the system to be carrier fluid, while intensity levels below the threshold will be considered to be particles. High intensity means transparent particles.
4. **Max Feret’s (or Feret’s) Diameter** is defined as the max diameter between two points on the perimeter between which a line can be drawn within the perimeter.
5. Area is defined as the total numbers of pixels covering the object.
6. Perimeter is defined as the total number of perimeter pixels.

RESULTS AND DISCUSSION

Particle Size Distribution

Figure 1 illustrates the particle size distribution (PSD) as a function of ECD along the filtration run for PAC and NPAC samples. The retention time for the specific filter velocity and media porosity studied was approximately 2.5 minutes; therefore 2.5 minutes were subtracted from the actual time effluent samples were taken to obtain a corrected time. PAC filter reached 300 cm of head loss after 240 minutes and therefore filtration run was terminated at that time for PAC and NPAC filters and backwash followed. Results indicated that a large number of particles or flocs (approximately 60%) of the filter influent are compacted in the lower size range between 2.5–3 μm. Usually the PSD in wastewater effluents is not normally distributed (14) as observed in this study. According to the PSD and the integrated total count, a higher concentration of particles in the effluent was observed for NPAC filter compared to PAC filter.

Figure 1. Particle size distribution (PSD) as a function of ECD filtered effluent with and without PAC. NP represents effluent of NPAC filter (dotted lines) while P represents effluent of PAC filter (full lines); Numbers in legend represent corrected time sample collected from filter effluent.
Average turbidity values of the filter influent is 3.15 ± 0.25 NTU. For PAC filter, the average effluent turbidity reduced from 1.7 NTU (at 2.5 min start time) to a value of 0.6 ± 0.04 NTU (after 12.5 min till the end of the filtration run at 240 min). For NPAC filter the average effluent turbidity value remained constant with a value of 1.8 ± 0.17 NTU (from 2.5 min till the end of the filtration run at 240 min). In general, filter ripening takes place when a significant reduction in the filtrate turbidity occurs and beyond that value filtrate turbidity remains at steady values. This shows that with PAC on turbidity basis it takes approximately 12.5 minutes to reach ripening while with NPAC samples effluent quality based on turbidity does not improve over time, thus filter ripening is not attained.

Similarly, total particle count from 2–100 μm shows that with PAC after 12.5 minutes total count reduced from approximately 46,000 particles/ml to 5000 particles/ml. For NPAC sample the total count after 2.5 minutes up to 60 min remains constant with an average of 37,000 particle/ml and after that count decreases to about 30,000 particle per ml and remains constant at that value. Thus, based on PSD, with PAC sample it took 12.5 minutes for the filter to ripen, while for the NPAC filter, the filter did not ripen. However, turbidity values were not sensitive as particle count to the small change after 60 min in total count of the NPAC samples (data not shown).

Particles transport to the filter media grain by different mechanisms which depend on the particle size. Small particles (<1 μm) are transported by diffusion while larger particles are transported by gravity, interception, and straining. PSD analysis can allow obtaining more accurate filtration models and improve our understanding of filtration (15). The larger particles in the inflow raw suspension are probably aggregates of biomass that were not removed in the secondary sedimentation prior to filtration. Yao et al., (16) showed that filter effluent particles in the size range of 1–2 μm in size are minimally removed by the filter. In this study, particles less than 2.5 μm are not measured and probably represent dispersed particles or microorganisms. The high concentration of particles in size range of 3.5–5.0 μm for NPAC samples may imply minimum transport for particles with a dimension close to minimum transport efficiency such as Cryptosporidium particles (11) or it may demonstrate the impact of filter influent PSD shape on effluent PSD shape. Effluent samples after filtration for NPAC show the same distribution shape as raw sample but with lower counts while with PAC particles larger than 3.5 μm are totally removed (Fig. 1). The influent PSD samples at different times showed similar pattern and concentration numbers except for influent at 120 min that had an approximately 50% lower particle count, with a similar distribution pattern (data not shown).

Filter Removal Efficiency

Percent relative removal efficiency (RRE) is calculated by Eq. (1) where \( C_i \) is influent concentration of raw sample and \( C_e \) is concentration at the effluent
with PAC (PAC) or without PAC (NPAC).

\[
\% \text{RRE} = \left( \frac{C_i - C_e}{C_i} \right) \cdot 100
\]  

Influent particles from secondary treatment may show variability in their concentration and size distribution therefore $C_i$ was sampled simultaneously with $C_e$ to determine removal. Percent RRE of particles as a function of ECD is illustrated in Fig. 2. During depth filtration the suspended particles are removed by attachment to the filter media or to particles that were previously retained and serve as additional collector sites that result in improved removal efficiency (17). Removal efficiency (RRE) is higher along samples with PAC as compared to samples that pass the filtration without inserting PAC. Application of flocculants highly improved removal efficiency and the difference is noticed even at the beginning of the filtration run. Without PAC, the removal efficiency of the filtration process based on size is low for the small particles and higher for the larger particles, while with PAC removal efficiency is constant at all particle size range.

Adin and Elimelech (18), found that the removal efficiency of the secondary effluent filtered through a sand bed showed linear removal up to 10 $\mu$m followed by constant 80% removal for larger particles. Specifically, after 60 min without PAC (NPAC), removal was low (removal 40–60%) for the smaller particles below 10 $\mu$m and higher for the larger particles (removal 60–80%). With NPAC filter after 240 min, removal was higher than at 60 min filtration time especially for the smaller particles below 10 $\mu$m (removal 70–80%) and similar to 60 min filtration for the larger

![Figure 2](image-url)

**Figure 2.** Percent removal of particles as function of ECD for PAC and NPAC samples after 60 and 240 min of filtration.
particles. Improved removal of the small particles was not correlated to the effluent NPAC turbidity measurements. At 60 and 240 min for the PAC filter, removal efficiency is constant at all particle size range from 2.5–30 μm with values between 85–100%. This means that PAC captures also small particles thus results in an effluent with a narrower distribution.

Removal of particles by granular filtration is a simultaneous process of particle attachment to the collector, detachment and reattachment of those detached particles in deeper layers of the filter, or detachment towards the effluent as observed herein as spikes in Fig. 2. The spikes in the graph that relate to sudden lower removal efficiencies in particle removal imply that larger particles may be leaving the filter compared to particles that entered the filter, suggesting particle detachment. This occurs since the structure of accumulated deposits on the filter are not equally strong, and under the hydrodynamic forces caused by the flow this structure may be partially destroyed and may disintegrate previously fixed flocs (19). The spikes in the graph are more obvious after 60 min for NPAC sample. After 240 min for NPAC filter, there is less detachment and a higher efficiency removal for the smaller particles, since particles that were previously removed may act as an additional collectors (11). Detachment or floc break off of the captured particles might occur and impact particle effluent morphology over time. Hydrodynamic mechanism for particle transport to the filter is a result of particle rotation across streamlines and is related to particle shape (20). Thus, particle shape can influence net removal and will be further investigated.

Shape determination of particles based on perimeter is mostly useful when particles analyzed are for particles with a narrow size range to reduce the influence of shape changing with size (21). Tysmans et al., (22) showed that since the perimeter of the particle projection is resolution dependent, it may result in different circularity values for circular particles depending on the particle size. They concluded that larger particles and finer resolution result in more accurate measurements. For example a 100 μm particle had a circularity of 0.99, particles in the size range between 15–90 μm had a circularity of 0.98 but for a 6 μm particle the circularity was 0.88. Since removal of particles in filter effluent as a function of circularity may depend on the size, data was analyzed in size bins. Narrowing down the particle range may strengthen the conclusion that particles with a lower circularity are more easily captured by the filter compared to particles with higher circularity, especially evident for the smaller particles with different circularities.

Generally the shape of particles in wastewater is very complex as natural particles are not spherical when observing the 3-dimensional (3D) images of particles. Particle shape can be described by the circularity which is the ratio of the perimeter of a circle with the same area as the particle divided by the perimeter of the actual particle image. Circularity has values in the range 0–1, with a perfect circle a circularity of 1 while an irregular object has circularity closer to zero. Figure 3 illustrates the percent removal of particles as a function of circularity for PAC and NPAC samples at various filtration times.
Removal based on circularity shows that with PAC high removal efficiency occurs at all particles with all circularity values. With NPAC samples it is evident that removal improves over time (120 min compared to 240 min) and improved removal occurs with less circular particles. However the improved removal is not entirely correct, as particles with a variety of circularity values even for the small particles are minimally removed (Fig. 2), while large particles whether circular or not are highly removed. Therefore it would be more accurate to study removal based on circularity by dividing particles to size bands and observing removal as a function of circularity for narrow size bands as suggested previously by Hentschel and Page (21).

Figure 4 illustrates removal based on circularity after 120 min of filtration for particles between 2.5–5 μm, 5–10 μm and above 10 μm. Particles with lower circularity are removed more efficiently compared to the highly circular particles. These results also suggest that the spikes in the size based removal may be due to the more circular particles for similar size range that are removed less efficiently and not necessarily due to detachment of previously retained particles as suggested by other researchers. For a particle with circularity below 0.6 at all size range removal was near 100%. Between circularity of 0.6 to 0.9, there is a trend in decrease in removal efficiency with an increase in circularity for particles up to 10 μm. In a few cases negative removal based on circularity was observed, which repeated itself in all samples for the higher circularity values. Negative values can be obtained in the case of having more particles in the effluent than the influent for a specific circularity, which suggest that larger flocs may breakdown and form smaller more circular particles or that particles in the effluent are more circular than influent particles. Removal of NPAC filter at 120 min based on feret diameter shows that longer more elongated particles are retained by the filter almost completely (above 95%) for particles with feret max above 12 μm. Removal on ECD basis for NPAC sample is gradual (Fig. 2) and

Figure 3. Percent removal of particles as function of circularity for PAC (left image) and NPAC (right image) samples after 120, 180, and 240 min of filtration.
there is not a cut off value of ECD that results in complete removal as with feret max number. For PAC samples at 120 min, above feret number of 6 μm removal is complete along the filter run time. This point may be very important in evaluation of the minimum feret max value that results in complete particle removal. To conclude, using a statistical test for coupling

Figure 4. Removal based on circularity after 120 min of filtration for NPAC samples between 2.5–5 μm, 5–10 μm and above 10 μm.
size and shape “coupled distribution” for each individual sample has an advantage when comparing samples (22), and is suggested as future work.

**Correlation between Shape Parameters: Circularity and ECD**

The goal of this experiment was to test the impact of ECD on circularity with irregular shaped particles. The increase in particle ECD was obtained by intentionally increasing sample concentration of latex beads at a size of $90.7 \pm 17.7 \, \mu m$ overtime to induce particles to contact closely other particles and form various particle dimensions. Figure 5 illustrates circularity versus ECD for latex bead particles, while the insert represent the PSD of the latex beads. It is obvious that there is a decrease in circularity with increase in ECD. This experiment shows the difficulty in interpreting particle shape parameters for irregular shaped effluent particles that are secondary particles composed of primary particles with a variety of shapes.

**Correlation between Shape Parameters: Feret Diameter, ECD, and Count**

Many floc morphological characteristics are important when modeling floc behavior, such as the longest dimension, width, and shape factors (23). To understand the floc characteristics, samples that are uniform and spherical were first analyzed. Two types of polystyrene microspheres were analyzed in this study, where an ECD of $5 \, \mu m$ correlated to an average feret diameter of $5.55 \, \mu m$ and an average ECD of $10 \, \mu m$ correlated to and average feret diameter of $10.4 \, \mu m$. Thus, with dispersed spherical particles,

*Figure 5.* Circularity versus ECD for latex bead particles. The insert represent the PSD of the latex beads.
the ECD and feret diameters match. The latex bead used in this study formed various shapes such as doublets, triplets and a variety of irregular forms, when their concentration in the sample was intentionally increased. Table 1 shows selected images of latex beads taken from the image analyzer, with its corresponding ECD and feret max diameter. A linear relationship is noticeable between feret diameter and the number of particles that join each other in a row. For example, a feret of 88 µm is obtained for one latex bead particle, 162 µm for two particles in a row, 254 µm for three particles in a row, and 355 µm for four particles in a row. When particles join each other in other more compact configurations (not in a line), the feret max is less than that obtained if the same number of particles will be theoretically stretched in a continuous row. It is difficult to understand results obtained even for particles that are originally relatively spherical with narrow PSD in their disperse state. However, working with mean diameters (ECD) can lose information while the feret number is not a mean parameter and it relates to the actual raw particle information.

Samples after secondary treatment in a WWTP (Ramat HaSharon, Israel), were disinfected either with UV or chlorine (termed “UV” or “chlorine”), held in a reservoir and examined after two weeks for particle analysis. Figure 6 illustrates the PSD of the chlorine sample. A bimodal distribution was |Table 1. Selected images of latex beads, with its corresponding ECD and feret max diameter|

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<tr>
<th>Latex bead image</th>
<th>ECD (µm)</th>
<th>Feret diameter max (µm)</th>
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<tr>
<td>83.13</td>
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obtained where most of the particles are near the 4–5 μm and other mode occurs between 14–19 μm. Representation by image analysis can provide indication of the type of particle being analyzed, as the modes can be correlated with the image of the particles.

Figure 7 illustrates a 3D plot of ECD as a function of feret number for chlorine sample above. For a particle with a certain ECD, the feret diameter spans over a wide range from circular particles that feret equals to ECD, to

![Figure 6. PSD of chlorine samples (the insert represents an image taken by the image analyzer).](image)

![Figure 7. 3D plot of ECD as a function of feret max diameter for chlorine samples.](image)
elongated particles where feret max is larger than ECD. This data shows that for most particles feret is equal or larger than ECD, thus most of the natural particles in this effluent are elongated even for the small particles. Most of the particles are in the size range up to 3.5 μm, while the small particles are more circular as the feret span in the small particles is narrower compared to larger particles (on ECD basis).

Correlation between Shape Parameters: Intensity and ECD

Intensity is defined as the sum of all pixel intensities in object divided to the total number of pixels. Particles with higher intensity are more transparent and intensity may provide information about particle makeup. Figure 8 shows the intensity of samples as a function of ECD. An inorganic sample taken from the soil was separated to the silt fraction was analyzed and termed “Silt,” the other particles are as previously determined. Data for the natural particles in the effluent of chlorine, UV, and raw filter effluent up to approximately 10 μm fall on top of each other, while the silt particles and the larger mode in the chlorine sample fall on other areas in the graph. Since algae particles can develop in the reservoir of secondary effluents (8), and chlorophyll was detected in both UV and chlorine samples, it is possible that algae particles are dominant in the chlorine samples PSD mode (Fig. 6). Backwash of PAC particles contains a mixture of mostly organic biological and detritus particles. Various intensity levels were detected with decrease in intensity as ECD increase (data not shown). For example, 2–5 μm particles had

![Figure 8. Intensity as a function of ECD for chlorine, silt, and UV samples.](image-url)
intensity of 190–200, 5–10 μm particles with intensity of 170–200 and 10–50 μm particles with intensity range of 160–190 μm. Since flocs are mixed particles it may be difficult to relate transparency to chemical characteristics of specific particles.

The optical absorption of inorganic particles such as silt and clay has very high to very low absorbance depending on the elements; organic detritus has generally high absorbance and biological organisms have usually low absorbance (24). The organic detritus particles range from submicron to 100s of microns, while the inorganic particles range up to a few microns. Assuming absorbance is related inversely to particle transparency, may result in higher transparency for biological organisms and lower transparency for organic non-biological matter in effluents.

Automated image analysis provides also shape and transparency parameters besides particle size and can possibly be used to target specific microorganisms or to discriminate between different size populations. Coupling intensity measurements with other size and shape parameters and the actual images may further be used as a foot print to identify particle makeup however with natural particles this may be complicated as they are a blend of various particle types. Chavez et al., (25) have suggested that particle size distribution as measured by the electrical sensing technology (Coulter counter), can be used as a tool to monitor the microbial quality of wastewater. Clancy et al., (26) showed that dynamic image analysis technology can be used to identify and enumerate biological particles in raw and treated waters such as Cryptosporidium oocysts and algae, and complement manual microscopy.

**Issues to Consider**

Figure 9 illustrates images of effluent samples. The right image represents the image itself, while the left image represents the perimeter of the particle as analyzed for shape parameters. It is clear that there are two different particles in the floc imaged; one of biological organic origin and the other is a floc of organic origin. In the image it is apparent that a microorganism

![Figure 9](image-url)
is associated with an organic effluent particle that may be protected from disinfection either by diffusion of chlorine or penetration of photons with UV. This image shows the importance of observing the images beyond obtaining particle size distribution for the large particles (flocs) prior to disinfection. This technology may be possibly useful to detect particles that are by size smaller than the pore size of membranes; however, due to their elongated shape (feret and circularity) they may be retained on the membrane.

CONCLUSIONS

1. Filter removal efficiency can be determined by various size and shape parameters, where different information is obtained for each analysis.
2. Results suggest that the particle size distribution is not the sole important characteristic in evaluating water effluents as particles are highly heterogeneous with respect to their size and shape.
3. This method has potential in evaluating efficiency of particle separation processes however more robust statistical analysis need to be used to allow coupling all the size and shape parameters into a meaningful tool to evaluate for example the optimum coagulant dose in contact filtration processes.

REFERENCES
