

# Morphological Analysis of Activated Sludge Flocs and Filaments

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**Abstract**—Purification of waste water is commonly done using the activated sludge process. The ratio of the activated sludge flocs and filamentous bacteria play a key role in the purification process of waste water. The sludge bulking or filamentous bulking is a common problem in activated sludge plants that prevents flocs to settle down. Digital imaging techniques can play an important role in monitoring activated sludge flocs and filaments in waste water treatment plants (WWTPs). In this paper, an algorithm to segment the flocs and the filaments of the microscopic sludge images captured at 4 times magnification in brightfield microscopy has been proposed. Morphological parameters, like, compactness, roundness, convexity, equivalent diameter are analyzed. Comparison with laser particle size analysis method has been done for the interpretation of the imaging results.

**Keywords**— *Image Processing; Image Analysis; Segmentation; Morphology; Waste Water Treatment*

## I. INTRODUCTION

The activated sludge process is one of the most frequently employed wastewater purification process worldwide. The biological mass of microbial aggregates known as sludge flocs are formed in the activated sludge process and plays an important role in determining the performance of the waste water purification system. When the common problem of filamentous bulking occurs, the sludge flocs are prevented from being formed properly, affecting their proper sedimentation in the settling phase of the process and causing the effluent quality of the plant to deteriorate. To maintain the plants' performances, the effluent and the sludge needs to be observed frequently and the operating conditions in the tank are adjusted accordingly [1, 2]. The traditional methods of monitoring WWTPs use chemical-physical methods. These methods measure the sludge volume index (SVI), total suspended solids (TSS) etc. These methods are somewhat limited in their ability to remedy the filamentous bulking and other abnormal conditions before the condition becomes severe due to delay in information gathering [3]. Digital image analysis involves the observation of microscopic images of activated sludge flocs and filament samples. These images are

stored on the computer in digital form and are analyzed to gather information about activated sludge on the microscopic level. Digital image analysis can help to identify the changes in the morphology of the activated sludge flocs (shape, size, compactness, roundness), a phenomenon that cannot be readily observed in the chemical-physical measurements. Recently digital image analysis has been incorporated to detect and analyze the flocs and the filaments in activated sludge [1, 4-9].

Hence digital image analysis can help to identify the onset of abnormal conditions in the WWTPs well beforehand as compared to the traditional chemical-physical measurements. The identification, quantification and morphological analysis of activated sludge flocs and filaments using digital image analysis play a crucial role in avoiding abnormal conditions in activated sludge WWTPs. Therefore in this study emphasis has been given on the segmentation and morphological analysis of activated sludge flocs and filaments.

The rest of the paper is organized as follows. In section 2 we will discuss the method for data acquisition. The proposed algorithm for the segmentation of flocs and filaments will be discussed in section 3, followed by discussion on segmentation results in section 4. The morphological analysis will be presented in section 5. Comparison of results is done with the results from laser particle analyzer in section 6. Finally the paper is concluded in section 7 followed by references.

## II. IMAGE ACQUISITION

The samples of waste water are acquired from the activated sludge tanks of local WWTP that deals with municipal effluents. For image acquisition a Zeiss Primostar microscope equipped with a CCD camera (Zeiss AxioCam ERC 5s) connected to a personal computer is used to capture the digital images. The sample is prepared simply by placing 80 $\mu$ l of activated sludge sample on microscopic slides using micro pipette. The sample is observed and 30 images are captured under the objective magnification of x4 using brightfield microscopy. The simulation platform is a laptop computer with Intel Centrino Duo Intel 1.66GHz CPU. MATLAB software is used for programming. All images are saved in JPEG file

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format. The image resolution is 1920 x 2560 pixels. A sample image is shown in Fig. 1.

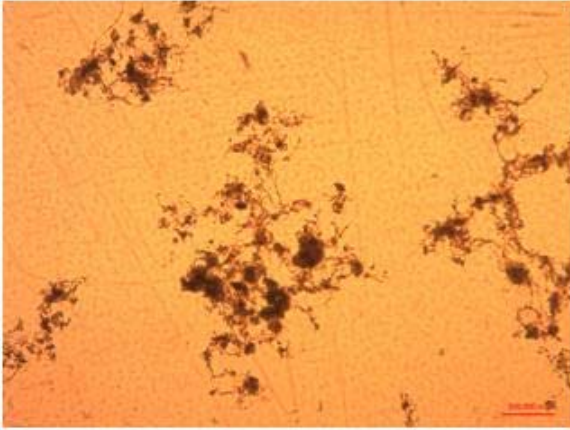


Fig1. Original image of activated sludge

### III. IMAGE SEGMENTATION

All images are captured using the brightfield microscopy technique where flocs and filaments show dark intensity values against a brighter background. It is observed that at x4 objective magnification, the filaments are very thin and threadlike. Hence in the proposed segmentation algorithm the filament pixels are replaced with the surrounding bright intensity values by means of median filtering. The resultant image can be called ‘floc image’. Adding or subtracting operations involving the floc image and a gray scale original image gives us the replaced pixels by the previous median filtering. Most of the replaced pixels represent the filament objects. This is the basis of segmentation used in the proposed algorithm.

In the proposed algorithm H-minima Transform [14] is used to segment flocs and filaments. The grayscale image obtained from the original image (hereby onwards referred to as the original image), is obtained by using the H-minima transform. Regional minima in the images that have a intensity/depth, less than a given value (50 in our experiments) are eliminated by the operation. Next, median filtering with a moderately large window size (25 x 25) is applied to the resultant image to eliminate the filament objects. Median filtering is followed by 10th order morphological erosion using a disk type structuring element to bulk up the remaining objects. Complementing this image produces the ‘floc’ image.

The addition of the ‘floc’ image and the ‘floc and filament’ image produces the ‘filament’ image. Binary conversion is then done using the Otsu thresholding method [14]. At this point, the morphological parameter of roundness as shown in (1) is used to eliminate the segmented binary regions that are not sufficiently elongated. Roundness is defined as the ratio of an object’s area to a circle/disc’s area such that the circle/disc is equal in length to the object. Roundness can be defined as follows:

$$Roundness = (4 / \pi)(area / length^2) \quad (1)$$

The roundness of an object varies from a value of 1 for a circle to values closer to zero for increasingly elongated objects. For our purposes here, ‘length’ is defined as the major axis length, which is the length (in pixels) of the major axis of the ellipse that has the same normalized second central moments as the object. The variable of ‘area’ is similarly measured in pixels as the ‘length’. Only objects with roundness values greater than 0.3 remain after the operation. The remaining regions after the operation give the segmented filaments.

The overall flow of the filament and floc detection and segmentation algorithm is shown in Fig 2.

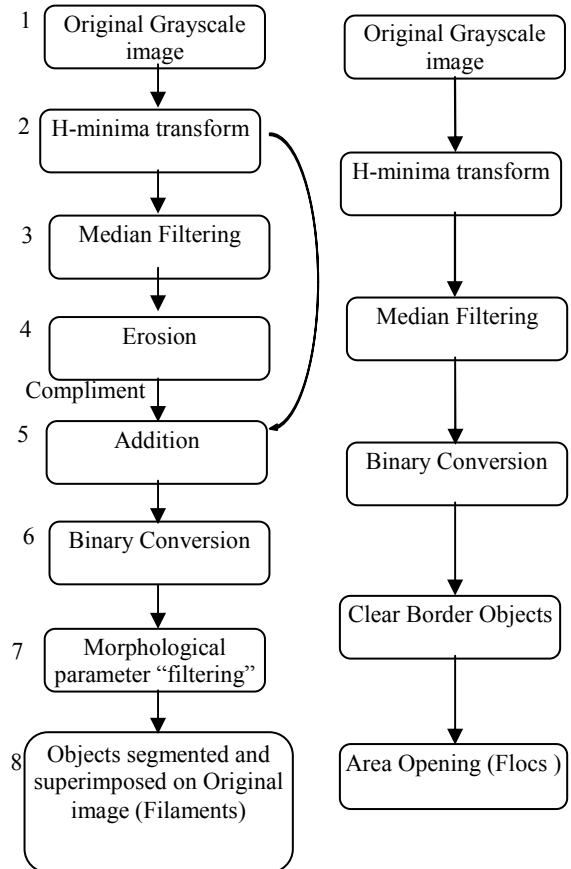


Fig. 2 Filament and Floc segmentation flowchart

### IV. SEGMENTATION RESULTS

The proposed algorithm is able to detect the flocs and filaments quite well from the microscopic images. The shape of the flocs show a good match as shown in Fig 3 (a) and (c), whereas quite a good number of filaments are detected and segmented, as seen in Fig. 3 (d).

In the first step of the algorithm, a moderately large scalar value was manually chosen for the H-minima transform, and the operation is seen to eliminate most of the regional minima in the flocs and some of the unwanted noise objects in the images which are lighter than the flocs. This operation can be

interchanged with a ‘morphological reconstruction’ operation that utilizes the original image and a darkened version of itself, and both produce very similar results.

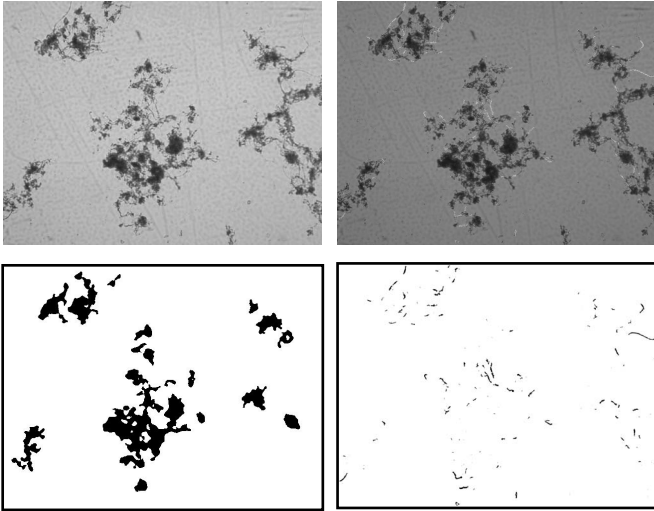


Fig. 3 Left to right, top to bottom, (a) Original Grayscale Image (b) Superimposed Filament Segmentations (c) Floc Segmentation (d) Filament Segmentation

There exists difference in background lighting conditions across images. However, the algorithm is robust across such differences. Although efforts can be made to keep illumination conditions similar or constant throughout image acquisition, it can be difficult to recreate the exact lighting condition after it has been altered, for example after switching to a different objective lens (which requires setting a new illumination) and then switching back, a common occurrence. Hence, the illumination-invariant feature of the algorithm is useful and allows for greater freedom in image acquisition.

Filaments are different from flocs in their shape as these are thin and elongated. After binary conversion, roundness is chosen as a morphological parameter to eliminate the less-elongated pixels in the binary image. By taking into account the object’s length and the occupied area, the roundness is a parameter that is sensitive to the elongation of the objects. In the binary image, the pixels that represent the filaments are seen to be well connected, and together they make long and thin regions. Therefore, only short, blob-like noise objects are deleted.

## V. MORPHOLOGICAL ANALYSIS

A very simple morphological analysis can be done on the basis of shape and size of the flocs. The sludge flocs can be divided into 3 different classes based on their sizes [12]. These are small flocs with diameter less than 100  $\mu\text{m}$ , medium flocs with diameter between 100 and 500  $\mu\text{m}$ , and large flocs with diameter more than 500  $\mu\text{m}$ . From Fig. 4, it can be seen that the flocs analyzed can be categorized into small and medium sized

flocs. Small size flocs make up approximately 70% of the total while medium size flocs make up the remaining 30%.

The Pearson’s linear correlation coefficient for various morphological parameters is calculated for the segmented floc objects utilizing four morphological parameters; these are equivalent diameter, roundness, compactness and convexity. This is shown in Table 1. The values of Pearson’s coefficient range between -1 and +1; where -1 corresponds to a perfect negative correlation and +1 corresponds to perfect positive correlation.

The equivalent diameter is defined as the diameter of a circle with the same area as the area of a 2D captured floc. Roundness is defined by Eq (1). Compactness or Form Factor is defined by Eq (2); and is calculated as the ratio of an object’s area to the area of a circle that has the same parameter/border as the object. For any constant parameter/border length, more compact objects possess larger area values and hence larger compactness values. Convexity is given by Eq (3) and is calculated as the ratio of an object’s area to its convex hull’s area. Convex hull refers to the minimum-size shape that makes an object convex.

$$Compactness = (4\pi * area) / perimeter^2 \quad (2)$$

$$Convexity = area / (convex\_hull\_area) \quad (3)$$

All morphological parameters above except for the equivalent diameter range from 0 to 1.

From the values in table 1, it is observed that a very strong correlation exists between the parameter compactness and convexity for the flocs identified. This is expected as shapes that are compact are expected to be convex and vice versa. The very high value however, suggests the absence of highly elongated elliptic-shaped flocs which are convex but not compact. On the other hand, it is seen that roundness and equivalent diameter have very a low correlation coefficient value with each other. This means that floc elongation cannot be inferred from its size and vice versa. The coefficient values between roundness; and compactness and convexity are also relatively high, suggesting that the flocs are considerably well shaped and are not fragmented. Finally the coefficient values between equivalent diameter; and both compactness and convexity shows us that small sized flocs are more compact and convex compared to larger ones. On the basis of these values we can conclude that the plant is in good condition as the flocs obtained are compact. Compact flocs have a higher tendency to settle down, which matches with the state of the plant that is said to be in stable condition.

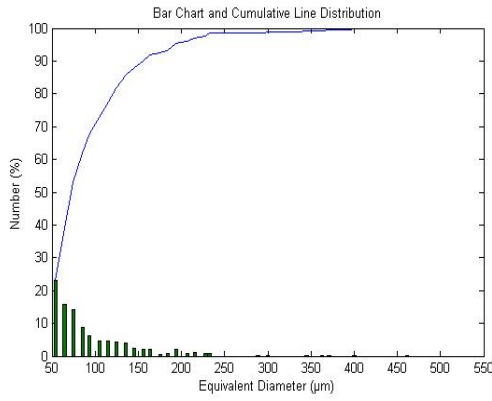


Fig 4. Equivalent diameter versus the number of flocs, bar chart and cumulative distribution

TABLE I. PEARSON'S CORRELATION COEFFICIENTS FOR MORPHOLOGICAL PARAMETERS

	Eq Diameter	Roundness	Compactness	Convexity
Eq Diameter	1.0	-0.156	-0.598	-0.543
Roundness	-0.156	1.0	0.658	0.609
Compactness	-0.598	0.658	1.0	0.949
Convexity	-0.543	0.609	0.949	1.0

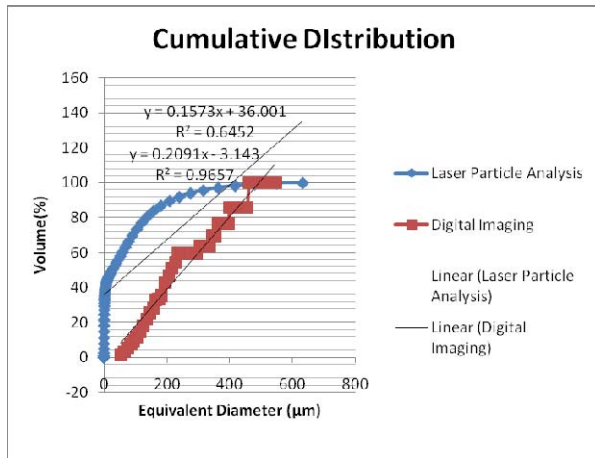


Fig 5. Cumulative volume distribution of floc size using laser particle analyzer

## VI. COMPARISON WITH LASER PARTICLE ANALYZER

Laser particle size analysis is a method that uses laser diffraction to measure particle size distribution by measuring the angular variations in intensities of light scattered as a laser beam passes through a sample. Volume distribution graphs are

produced by this method with other distributions derived from the basic volume graphs. The Malvern Mastersizer MS2000 laser particle size analyzer is used in this work.

Fig. 5 shows the cumulative distribution of floc size using laser particle analyzer and image analysis. It is seen that the digital imaging method has a little tendency to overestimate the size of the objects [13]. This implies that in future more accurate floc segmentation has to be devised so that the results show more correlation. The hundredth percentile is reached by our method at approximately 500µm. From the graph, peaks are observed at around the range of 100-200 µm of particle size from both lines. The linear approximating equations for laser particle analyzer and digital imaging are shown in Eq (4) and Eq (5) respectively and are observed to possess similar gradient values.

$$y_{\text{Laser}} = 0.1573x + 36.001 \quad (4)$$

$$y_{\text{DI}} = 0.2091x - 3.143 \quad (5)$$

## VII. CONCLUSION

In this paper, an algorithm to identify and segment flocs and the filament objects found in the microscopic activated sludge images of the wastewater treatment system under the x4 objective magnification has been proposed using the roundness morphological parameter. The filament segmentation algorithm is robust across different background lighting conditions in the different images tested. The detected flocs have been analyzed with different morphological parameters and compared with the laser particle size analysis method for further processing and comparison purposes. The compactness of flocs show that the plant is in stable condition that has also been verified by the information obtained by the operating parameters of the actual plant. The segmented flocs and filaments can be further utilized for more in-depth diagnosis of the state of the sample and the wastewater treatment plant in future.

## REFERENCES

- [1] D. P. Mesquita, A. L. Amaral, E. C. Ferreira, "Disturbances Detection in Activated Sludge Systems," *Semana de Engenharia* 2010.
- [2] D. P. Mesquita, O. Dias, A. M. A. Dias, A. L. Amaral, E. C. Ferreira, "Correlation between sludge settleability and image analysis information using Partial Least Squares,"— *Proceedings of International Conference on Chemometrics in Analytical Chemistry (CAC2008)*, Montpellier, 2008.
- [3] G. Gins, I. V. Smets, R. Jenne, J. F. Van Impe, "Activated Sludge Image Analysis Data Classification: An LS-SVM Approach," *16th IFAC World Congress*, Prague 2005.
- [4] D. P. Mesquita, O. Dias, A. M. A. Dias, A. L. Amaral, E. C. Ferreira, "Evaluation of Activated Sludge Systems by Image Analysis Procedures"— *Proceedings of the International Symposium on Sanitary and Environmental Engineering: SIDISA 08 [CD-ROM]*, Florence, Italy, 24-27 June, pp: 1-8, 2008.

- [5] W. Heine, I. Sekoulov, H. Burkhardt, L. Begen, J. Behrendt, "Early Warning-System for Operation-Failures in Biological Stages of WWTPs by On-Line Image Analysis"- IWA Conference in Berlin, 15-19 October 2001.
- [6] A. L. Amaral, C. Baptiste, M. N. Pons, A. Nicolau, N. Lima, E. C. Ferreira, M. Mota, H. Vivier (1999) "Semi-Automated Recognition of Protozoa by Image Analysis" *Biotechnol. Tech.* 13: 111-118
- [7] X. Y. Lee, H. Nisar, K. H. Yeap, "An Approach for Segmentation and Quantification of Activated Sludge Floc Blobs, " *Advanced Science Letters*, July 2013.
- [8] H. Nisar, X. Y. Lee, K. H. Yeap, V. V. Yap, C. S. Soh, "Application of Imaging Techniques for Monitoring Flocs in Activated Sludge" *Intl. Conf. on Biomedical Engineering*, Malaysia, Feb 27-28, 2012.
- [9] M. Sikora, B. Smolka, "Feature Analysis of Activated Sludge Based on Microscopic Images" *Canadian Conf. on Electrical and Computer Engineering*, Canada, May 13-16, 2001.
- [10] A.L. Amaral, "Image analysis in biotechnological processes: Applications to wastewater treatment" Ph.D. Thesis, Braga, Portugal, 2003.
- [11] E. Liwarska-Bizukojc, "Application of Image Analysis Techniques in Activated Sludge Wastewater Treatment Processes" *Biotechnology Letters* (2005) 27: 1427-1433, DOI 10.1007/s10529-005-1303-2.
- [12] D.H. Eikelboom, H.J.J. van Buijsen "Handbuch für die mikrobiologische Schlammuntersuchung" 3rd edn. Munich: F. Hirshammer Verlag GmbH, 1992.
- [13] R. Govoreanu, H. Saveyn, P. Van der Meeren, P.A. Vanrolleghem, "Simultaneous determination of activated sludge floc size distribution by different techniques" *Water Science and Technology* Vol 50 No 12 pp 39-46, IWA Publishing 2004.
- [14] Rafael C. Gonzalez, Richard E. Woods, *Digital Image Processing*, Third Edition, pp. 455-456, 2010.