Machine vision for the quality assessment of emulsions in pharmaceutical processing

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Abstract— **Emulsion quality evaluation using machine vision techniques depends on the efficiency of the image segmentation algorithms. Two different machine vision techniques are investigated to determine their competency in detecting droplets from in-process microscopic images of a cream emulsion. Histogram-based segmentation shows promising potential compared to edge and symmetry detection. A statistical study of the droplet characteristics was conducted. The results demonstrate that the histogram-based approach is more proficient in the progressive analysis of droplet evolution during emulsification. A real-time integration of the technique is proposed, as a soft sensor, to predict the optimum process time and to increase manufacturing efficiency in chemical industries.**

Keywords—automated image analysis, droplet characteristics, emulsion, machine vision, quality evaluation, sustainable manufacturing

I. INTRODUCTION

Product quality assessment, during an emulsification process, has been identified as a challenging task in chemical industries [1]. Emulsions are multi-phase colloidal dispersions obtained by mixing insoluble liquids. A two-phase oil in water emulsion (o/w) is formed when oil gets dispersed as tiny droplets in water (continuous phase). Emulsification is the process of breaking big oil globules into a homogenous distribution of microscopic droplets. The quality of the final emulsion product is highly dependent on the droplet size distribution, which is influenced by the operating conditions and process parameters [2]. Inline droplet size monitoring has been recommended as a route to identify the process parameters that provides the optimal target product specification [3]. Machine vision techniques have been previously applied for the quantitative real time measurement of droplet size in various multiphase systems [4-6]. These studies have found that automated image analysis for droplet counting and characterisation is approximately fifty times faster and less erroneous compared to manual assessment.

There have been several machine vision techniques reported in the literature for the segmentation and analysis of optical microscopic images of emulsions [7-11]. Zhou *et al.* [12] have conducted a critical review of various image processing approaches in real-time crystal size measurements. ImageJ [13] and Matlab are the most popular and widely used machine vision software for automated droplet detection in emulsions according to previous research [4, 7, 9]. Maaß *et al.* [4] has developed a machine vision tool in Matlab for automated counting and measurement of particles/droplets in multiphase systems including emulsions. However, the previous methods are limited to circular particles and Maaß *et al.* [4] have proposed a future extension of the method to additionally detect irregular shapes as well. The droplet detection techniques previously referenced have used machine vision software mainly to identify the droplet border on the phase transition between oil and water using edge detection [7]. Such edge detection techniques have proved unsuccessful in the image analysis of highly concentrated emulsions with dispersed phase fraction greater than 10 to 15 % [4-6, 14]. In addition to that, there has been no previous research published so far on a progressive analysis of droplet characteristics other than droplet diameter during emulsification.

This paper presents an investigation of two different image segmentation techniques that were performed to extract various droplet characteristics, from optical micrographs, during an emulsification process. The main objective of the study was to develop and assess suitable machine vision techniques capable of determining industrial process equilibrium (point at which the emulsification process is completed) through the detection of droplets and their corresponding characteristics at various stages of emulsification.

II. IMAGE SEGMENTATION AND ANALYSIS

Microscopic images (micrographs), of an oil in water (o/w) cream emulsion, were acquired at 5 minute intervals from the start to the end of a 30 minutes emulsification process. A Zeiss Microscope Axio imager A2m was employed to obtain bright field micrographs of 40x magnification under standard illumination settings. Two different automated image segmentation techniques were developed in Fiji version 1.51h to identify the region of interest, which is the oil droplet, in the

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micrographs. Subsequently, the droplet characteristics were extracted using both the techniques. Fiji is an extended version of ImageJ [15, 16].

A. Edge and Symmetry Detection

A macro, user-defined program to execute specific tasks, was programmed in Fiji to execute the edge and symmetry based image segmentation steps dynamically in the following order.

1. The micrographs were calibrated to a micrometer scale such as 4 pixels/µm.
2. The images were then converted to 8-bit in order to facilitate further processing.
3. Edge and Symmetry filter (ESF) was applied to detect the oil droplets in the images.
4. The filtered images were auto-thresholded through the red channel to enhance the detected droplets.
5. The images were then converted into binary and applied 'watershed segmentation' (separating the droplets that touch/overlap each other).

ESF is a multistage algorithm which operates in two major steps. These are edge detection followed by radial symmetry detection. A sample detection of a circular object using the ESF algorithm is shown in Fig 1. The edge detection stage includes identification of the edges of all the objects in the image, centralising the detected edge points by preserving the local maxima and also minimising the false edge (noise) detection. The radial symmetry phase makes use of a radius parameter to draw the symmetry of the objects in the image.

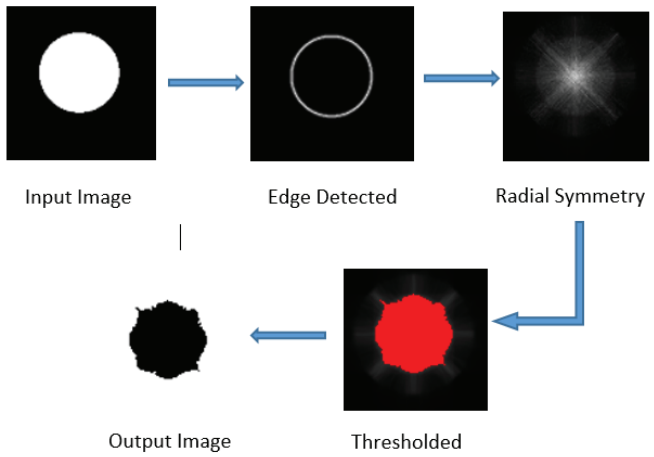

Fig. 1. Detection of a circular object using edge and symmetry based image segmentation.

B. Histogram-based Detection

A second macro was developed in Fiji to perform an alternative image segmentation procedure based on the histogram of the distribution of gray values (pixel intensity) in the region of interest.

Calibration of the micrographs and conversion to 8-bit (similar to section A discussed above) were the initial steps of this macro. This was followed by histogram-based image segmentation, which was executed as follows:

- A histogram was computed from all of the pixels in the images.
- The mean pixel intensity of the histogram was calculated and stored in a variable.
- The images were then thresholded using the calculated intensity value.
- Converted to binary and applied watershed segmentation.

Histogram-based detection of a circular object is shown in Fig 2.

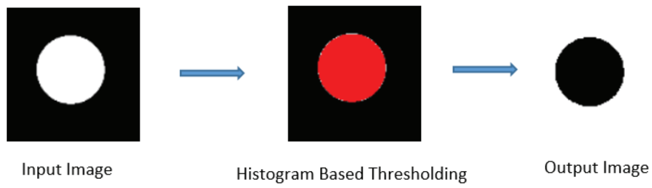

Fig. 2. Detection of a circular object using histogram-based image segmentation.

C. Extraction of Droplet Characteristics

The droplets detected using the two automated methods of image segmentation were analysed using the 'Analyze Particles' functionality in Fiji. This was the final step of both macros. The purpose of this image analysis was to extract the characteristics of the detected droplets for a user-specified range of droplet area (μm^2) and circularity. A diverse set of characteristics was extracted for each droplet such as size, shape, orientation, centroid, solidity *etc*.

An example of the droplet detection from an emulsion micrograph using both segmentation methods is shown in Fig. 3.

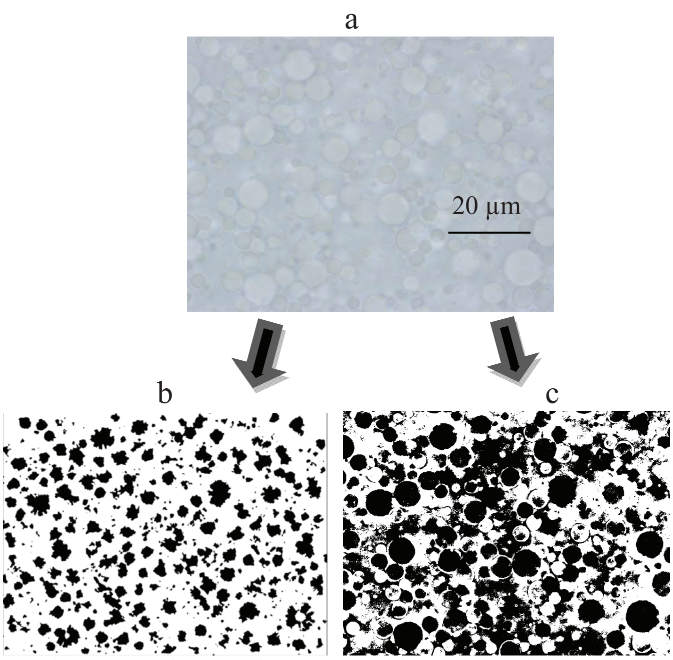

Fig. 3. Droplet Detection. (a) emulsion micrograph, (b) Edge and symmetry method and (c) Histogram-based method.

D. *Statistical Analysis*

A statistical analysis of the droplet characteristics was performed in RStudio, version 1.1.383 for all the micrographs. The evolution of average droplet size in terms of area (µm^2) and Feret diameter (µm) and the evolution of droplet count at each 5 minutes processing interval were studied in detail. The oil concentration, *i.e.*, the % of area occupied by the droplets, was also determined for each micrograph. The statistical study of the remaining droplet characteristics, obtained through the image analysis, are not within the scope of this paper.

III. Results and Discussion

A random sample of ten micrographs were selected at each 5 minutes interval (from 5 to 30 minutes) of the emulsification process. A comparative study of the droplet characteristics obtained from both image segmentation techniques and their statistical analysis results are discussed in this section.

A. *Method 1 – Edge and Symmetry*

Box plots of average area and average Feret diameter of the droplets obtained using edge & symmetry filter are presented in Fig. 4. Feret diameter is the longest distance between any two points along the droplet boundary. The average droplet area decreases from around 30 to 5 µm^2 in the first 15 minutes of the emulsification process and it appears to be varying only slightly for the rest of the processing period. Similarly, the average Feret diameter falls from around 7 to 3 µm in the initial 15 minutes and doesn't appear to vary much after that.

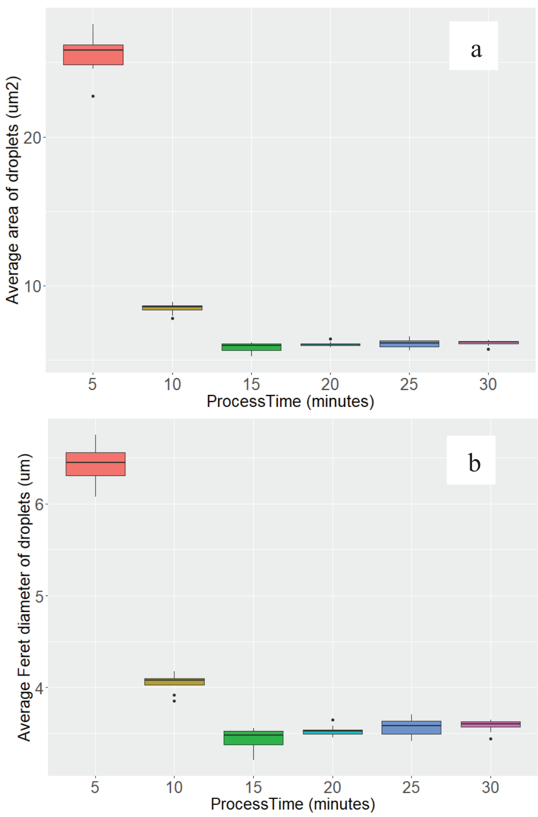

Fig. 4. Box plots of (a) average droplet area and (b) average Feret diameter obtained from edge and symmetry detection.

A box plot showing the evolution of droplet count is given in Fig 5. The droplet count seems to increase by 4000 in the first 20 minutes of the emulsification process. Then it varies slightly in the next 5 minutes followed by a steady state.

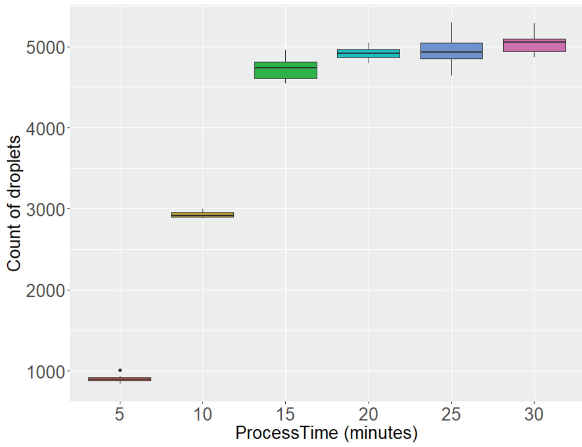

Fig. 5. Evolution of droplet count (edge and symmetry detection) from 5 to 30 minutes of emulsification.

The edge and symmetry detection technique highly depends on well-defined droplet borders and also on the radius parameter of the algorithm specified by the user. As the droplet size decreases with emulsification, it is difficult for the user to identify an accurate value for the droplet radius. This is a disadvantage of this technique. Multiple detections will be required for a single micrograph, using a set range of radii, to detect droplets from a varying size distribution, which is not feasible in terms of time and accuracy.

B. *Method 2 – Histogram-based*

Box plots of average area and Feret diameter of the droplets detected at each 5 minutes interval of the emulsification process using histogram-based detection are presented in Fig. 6. In the initial 10 minutes, the droplet area decreases dramatically from around 30 to 10 µm^2 while the Feret diameter also drops from 7 to 5 µm. During the following 20 minutes, the droplet size appears to decrease gradually and attains a steady state towards the end of the emulsification process. The evolution of droplet count is presented in Fig. 7, which shows a very smooth increase from around 2000 to 8000 droplets during the total 30 minutes of the emulsification process.

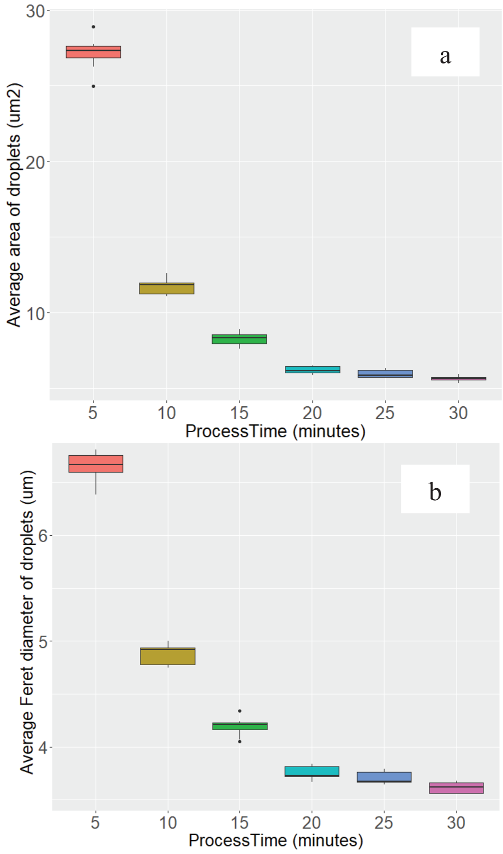

Fig. 6. Box plots of (a) average droplet area and (b) average feret diameter obtained from histogram-based detection.

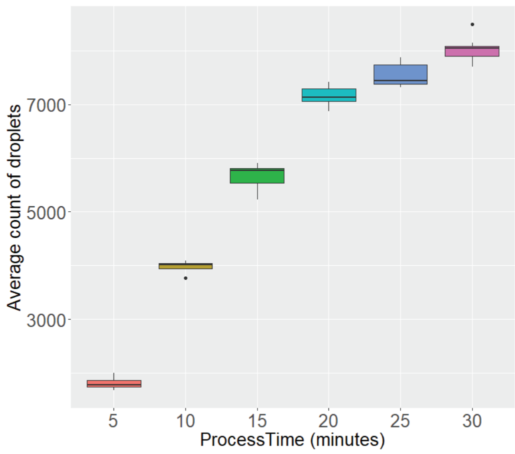

Fig. 7. Evolution of droplet count (histogram-based detection) from 5 to 30 minutes of emulsification

The histogram-based technique is capable of detecting both bigger and smaller droplets thus providing a smooth evolution of droplet size and count. The droplet size decreases significantly during the last minutes of processing and they appear as texture rather than discrete droplets as seen in Fig. 3. The histogram-based approach demonstrates proficiency in detecting those droplets.

C. *Comparison of both techniques*

The machine vision techniques, methods 1 & 2, were compared by analysing the oil concentration obtained using the image analysis. Box plots were generated for the concentration of oil obtained using the micrographs from 5 to 30 minutes of emulsification process as shown in Fig. 8. The box plot shown in Fig. 8(a), obtained using edge & symmetry, shows inconsistency in the oil concentration throughout the process and the values do not agree with the oil to water ratio of the product. On the other hand, the oil concentration results obtained using the histogram-based method (Fig. 8(b)) was in close agreement with the emulsion product and showed consistency throughout the process.

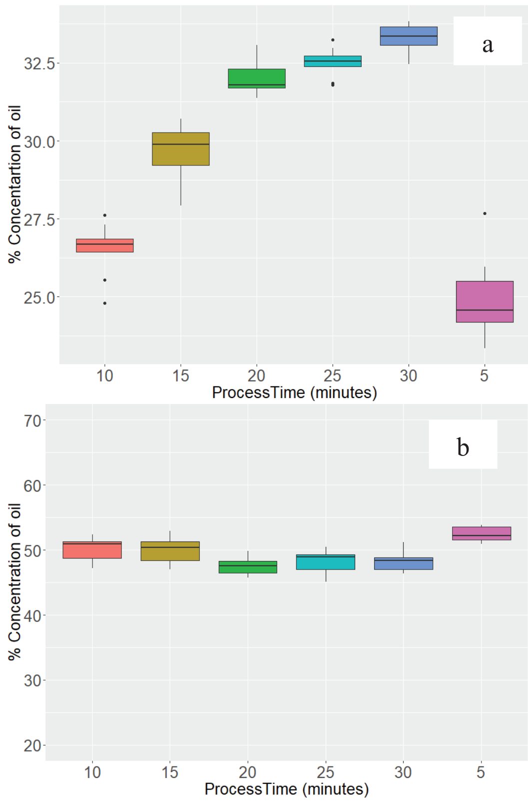

Fig. 8. Box plot showing the oil concentration from 5 to 30 minutes of emulsification. (a) Edge & symmetry detection, (b) histogram-based detection

The histogram-based machine vision technique demonstrates novel potential in assessing the evolution of droplet characteristics during the emulsification process until the desired characteristics are achieved. Therefore, the technique can be implemented as an automated tool in chemical industries to predict and identify the optimum process time. This can contribute to eliminating over-processing and associated resources that can be regarded as surplus to production requirements. A flowchart, derived from this research, is shown in Fig. 9. This scheme can entirely automate, in real-time, the quality assessment of the emulsion. Future work is planned to implement this scheme by integrating the machine vision technique with real-time imaging (an endoscope coupled with a CCD camera) using the experimental set-up applied by Maaß *et al.*[4]. The bias in the microscopical analysis performed by human examiners can also be entirely rectified through automated quality evaluation as suggested by Boxall *et al*. [17]. The proposed approach can be applied as a soft sensor, for in-situ process monitoring, to provide real-time feedback on emulsion quality.

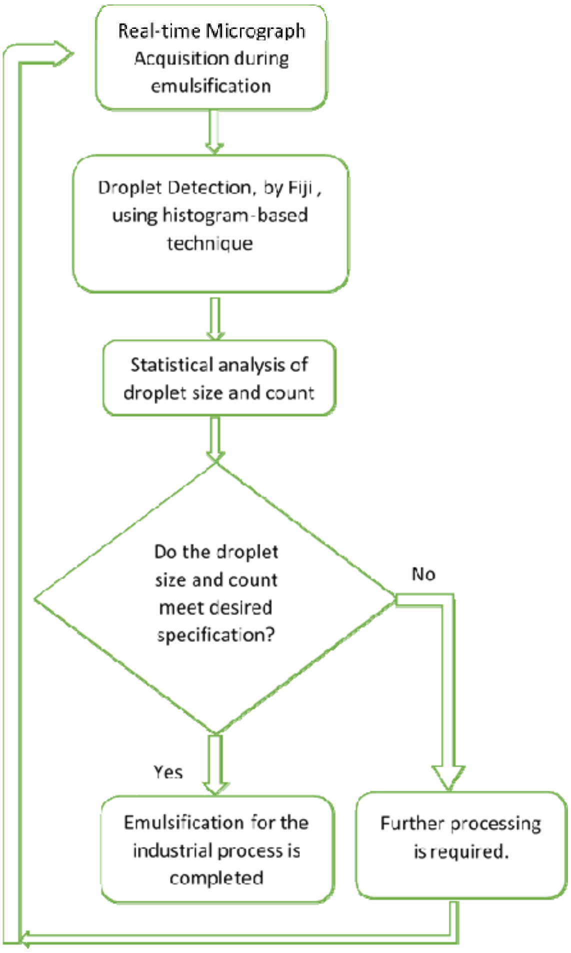

Fig. 9. Flowchart for real-time emulsion quality evaluation.

IV. Conclusion

This study has investigated the competence of edge & symmetry and histogram-based machine vision techniques, applied on optical micrographs, in the evaluation of droplet characteristics during emulsification. Droplet area, Feret diameter and count are the characteristics identified as the product quality indicators. The edge & symmetry technique has a distinct disadvantage, as it cannot be fully automated. This is due to the need to recalibrate the radius parameter as droplet sizes change. On the other hand, the histogram-based approach is fully automated. The histogram-based image segmentation has demonstrated significant potential in the detection of droplets and their corresponding characteristics. The technique has provided a progressive evolution of decreasing droplet size and increasing droplet count. The oil concentration results obtained using the histogram-based approach is in good agreement with the studied emulsion. The technique proved capable of identifying the equilibrium point at the end of the industrial process. A procedural flowchart is developed based on the results obtained. The proposed approach has demonstrated promising potential in avoiding the over-processing of emulsions, leading to smart and sustainable manufacturing practices.

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References

[1] O. Ruscitti, R. Franke, H. Hahn, F. Babick, T. Richter, and M. Stintz, "Application of particle measurement technology in process intensification," *Chemical Engineering & Technology,* vol. 31, no. 2, pp. 270-277, Feb 2008.

[2] J. Zeaiter, J. A. Romagnoli, and V. G. Gomes, "Online control of molar mass and particle-size distributions in emulsion polymerization," *Aiche Journal,* vol. 52, no. 5, pp. 1770-1779, May 2006.

[3] G. Crawley and A. Malcolmson, "Online particle sizing as a route to process optimization," (in English), *Chemical Engineering,* Article vol. 111, no. 9, pp. 37-41, SEP 2004 2004.

[4] S. Maaß, J. Rojahn, R. Haensch, and M. Kraume, "Automated drop detection using image analysis for online particle size monitoring in multiphase systems," (in English), *Computers & Chemical Engineering,* Article vol. 45, pp. 27-37, Oct 2012.

[5] A. Khalil, F. Puel, Y. Chevalier, J. M. Galvan, A. Rivoire, and J. P. Klein, "Study of droplet size distribution during an emulsification process using in situ video probe coupled with an automatic image analysis," *Chemical Engineering Journal,* vol. 165, no. 3, pp. 946-957, Dec 2010.

[6] I. Scherze, R. Knofel, and G. Muschiolik, "Automated image analysis as a control tool for multiple emulsions," *Food Hydrocolloids,* vol. 19, no. 3, pp. 617-624, May 2005.

[7] A. Hosseini, S. Jafari, H. Mirzaei, A. Asghari, and S. Akhavan, "Application of image processing to assess emulsion stability and emulsification properties of

Arabic gum," (in English), *Carbohydrate Polymers,* Article vol. 126, pp. 1-8, AUG 1 2015 2015.

[8] K. A. Silva, M. H. Rocha-Leão, and M. A. Z. Coelho, "Evaluation of aging mechanisms of olive oil–lemon juice emulsion through digital image analysis," *Journal of Food Engineering,* vol. 97, no. 3, pp. 335-340, 4// 2010.

[9] S. Schuster *et al.*, "Analysis of W1/O/W2 double emulsions with CLSM: Statistical image processing for droplet size distribution," *Chemical Engineering Science,* vol. 81, no. Supplement C, pp. 84-90, 2012/10/22/ 2012.

[10] R. Tonon, C. Grosso, and M. Hubinger, "Influence of emulsion composition and inlet air temperature on the microencapsulation of flaxseed oil by spray drying," (in English), *Food Research International,* Article vol. 44, no. 1, pp. 282-289, JAN 2011 2011.

[11] N. Meijer, H. Abbes, and W. G. Hansen, "Particle size distribution and dispersion of oil-in-water emulsions: An application of light microscopy," *American Laboratory,* vol. 33, no. 8, pp. 28-+, Apr 2001.

[12] Y. Zhou, R. Srinivasan, and S. Lakshminarayanan, "Critical evaluation of image processing approaches for real-time crystal size measurements," *Computers & Chemical Engineering,* vol. 33, no. 5, pp. 1022-1035, May 2009.

[13] C. A. Schneider, W. S. Rasband, and K. W. Eliceiri, "NIH Image to ImageJ: 25 years of image analysis," *Nature Methods,* vol. 9, p. 671, 06/28/online 2012.

[14] L. M. R. Brás, E. F. Gomes, M. M. M. Ribeiro, and M. M. L. Guimarães, "Drop distribution determination in a liquid-liquid dispersion by image processing " *International Journal of Chemical Engineering,* 2009, Art. no. 746439.

[15] J. Schindelin, "Fiji Is Just ImageJ (batteries included)," in *ImageJ User and Developer Conference* 2008.

[16] J. Schindelin *et al.*, "Fiji: an open-source platform for biological-image analysis," *Nat Meth,* 10.1038/nmeth.2019 vol. 9, no. 7, pp. 676-682, 07//print 2012.

[17] J. A. Boxall, C. A. Koh, E. D. Sloan, A. K. Sum, and D. T. Wu, "Measurement and Calibration of Droplet Size Distributions in Water-in-Oil Emulsions by Particle Video Microscope and a Focused Beam Reflectance Method," (in English), *Industrial & Engineering Chemistry Research,* Article vol. 49, no. 3, pp. 1412-1418, Feb 2010.