

## ABSTRACT

### *Characterization of Activated Sludge Flocs By Confocal Laser Scanning Microscopy and Image Analysis*

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The purpose of this study was to characterize microbial floc structure and properties under phosphorus (P) limiting and non-limiting regimes. The P-limitation applied to the biomass did not significantly impact on reactor performance in terms of COD removal and MLSS. The composition of EPS was affected by the P-limitation with significantly increased accumulation of carbohydrates, uronic acids and proteins. Under both P-conditions, CLSM and glycoconjugate mapping revealed bacterial cells enmeshed in heterogeneous EPS known to contain glucose, mannose, *N*-acetylglucosamine, and galactose. The relative abundance of  $\alpha$  and  $\beta$ -*N*-acetylgalactosaminyl/galactopyranosyl and *N*-acetylglucosaminyl residues was affected by P-limitation, suggesting changes in microbial populations within the floc structure, which in turn could cause the compositional changes of EPS. The image analysis performed on CLSM images indicated structural changes in area1 parameters of flocs under P-limited conditions. Under non-limiting conditions the cell clumps within the floc were significantly smaller (mean average diffusion distance of 1.16  $\mu\text{m}$ ) as compared to P-limiting conditions (mean average diffusion distance of 1.36  $\mu\text{m}$ ). The fractal dimension and porosity measurements for non-limiting conditions were 1.57 and 0.80 respectively. The fractal dimension and porosity under limiting conditions were either significantly higher or lower than under P-rich conditions.