Journal of Civil Engineering and Environmental Technology
p-ISSN: 2349-8404; e-ISSN: 2349-879X; Volume 6, Issue 4; April-June, 2019, pp. 184-189
© Krishi Sanskriti Publications
http://www.krishisanskriti.org/Publication.html

Formation of Aerobic Granules in Lab Scale Sequential Batch Reactor for Waste Water Treatment

Avneet Kaur[1] and Puneet Pal Singh Cheema[2]

*[1]PG Student, Department of Civil Engineering,
Guru Nanak Dev Engineering College,
Ludhiana, Punjab, India
[2]Assistant Professor, Department of Civil Engineering,
Guru Nanak Dev Engineering College,
Ludhiana, Punjab, India
E-mail: [1]avneet.khera@gmail.cm, [2]ppsc390@gmail.com*

Abstract—*Wastewater treatment contributes to an effective solids-liquid parting and the biological treatment method involves microorganisms which pose the problem and require a large footprint. An effective biotechnological process of AGS represents an inventive cell immobilization strategy cultivated in SBRs. The several advantages like intense and condensed microbial structure, high biomass retention and greater settleability are the key factors possessed by the AG technology. The number of conditions have to be fulfilled acting as operating parameters like the organic loading rate (OLR), Hydrodynamic shear force generated by aeration and measured as SUAV, Settling time (ST) and volume exchange ratio (VER). The values for each parameter were (OLR – 2.5 kg COD m^{-3} d^{-1}, SUAV – 2.78 cm/sec., ST - 10min and VER – 50 %) were attempted in three trials. The other operating conditions were kept constant throughout the study to evaluate the performance of the reactor. The influence of the parameters like sludge volume index (SVI), time taken for the appearance of aerobic granules and COD, nitrates, total phosphates removal efficiencies had been examined frequently. The optimized values of the operational parameters studied which showed the excellent performance in terms of aerobic granule formation, settleability of sludge, COD removal (91.78 %), nitrates removal (51.51%) and total phosphorus removal (79.89%). Under the proscribed operating conditions, the present study has established the suitability of AG technology. The Prioritization of major influencing factors is necessary for the design of pilot plants and for scaling-up of SBRs and the optimized values of operational parameters were proved successfully for the treatment of a real wastewater.*

1. INTRODUCTION

The wastewater generation increases due to the increase in the urbanization, at the same time earth's dwindling resources use had also increased. Recovering the water, nutrients and other materials enclosed in wastewater is a key opportunity to be seized. The untreated wastewater discharge in water bodies does not only lead to eutrophication and human health risks, it also generates gases like nitrous oxide and methane which contributes significantly to Greenhouse Gas (GHG) emissions. The objectives like safeguarding human health and environmental protection are of principal importance. Decisive and large-scale actions are needed to increase wastewater treatment methods and fecal sludge treatment. The decouple economic growth and development from the consumption of exhaustible resources has emerged as the concept of circular economy in response to the drawbacks of the conventional 'take-make-consume and dispose' model of growth. The water which is used, regarded as one of the most under- exploited resources we have. The resource recovery from wastewater including bio-gas, fertilizer and perhaps most importantly, it is a source of 'new' water. As a whole the aim of the biological treatment for the wastewater discharge from industries and domestic area are Oxidizing the particulate and dissolved biodegradable constituents into stable and secure end products. Collect and subsume non settleable and suspended colloidal solids into the biofilm for suspended or attached growth process. Removal of nutrients like nitrogen, phosphorus and many others as some of the constituents are also toxic to the microorganisms. Therefore, activated sludge process (ASP) derives its name from the biological mass that formed when air is continuously infused into the wastewater. In this, the microorganisms are mixed thoroughly with organic compounds that are contained in wastewater under conditions which stimulate their growth with use of the organic compounds as substrate. The microorganisms grow as they mixed by the air and the individual organisms flocculate to form an active microbial mass which is called as an activated sludge (AS). The conformist ASP systems are built as:(1) An aeration tank where biological reactions like organic carbon removal and nitrification occurs.(2) A settling tank where the AS is separated from the water treated by the flocculation method. In aeration tank low biomass concentrations further increases the requirement of recycling of the activated sludge from settling tank to aeration tank. Large floor area require for ASP treatment with high energy for aeration to generate recycling flows. The poor settling ability of AS (i.e. sludge bulking) can further lead to drop of effluent quality due to the extreme loss of sludge in the effluent, unrestrained sludge ages and loss of treatment effectiveness. The AG are considered to be a special case of a bio film compose of the self-immobilized cells. In the last 30 years, intensively deep research in the field of biological wastewater treatment and other applications had demonstrated that the bio films were often more proficient for wastewater purification than suspended ASP. To date, the applications of the AGS are regarded as one of the promising biotechnologies in wastewater treatment. In addition, the AG were known to exhibit following attributes as Regular, even and nearly encircling in structure, Tremendous settleability, Impenetrable and physically powerful microbial structures, High biomass withholding capacity, Capability to withstand at high organic loadings, Good acceptance to toxicity (Adav et al., 2008). The creation of stable aerobic granular sludge (AGS) depends upon more than one parameter which selects the slow growing microorganisms and wash-out the fast growing microorganisms. Early studies showed that high SUAV are essential for the formation of AGS and it's stability. It is also now evident that formation of AGS is possible even at low SUAV telling other parameters are of equally significant in AGS formation. The extracellular polymeric substances (EPS) production and trigger cell-cell interactions are induced by high HSF and compaction force helps in shaping the granules. The settling time (ST) was considered important since the invention of AGS as short ST will select fast settling aggregates while slow settling flocs will get washed out. Adav et al., (2008) adopted ST as 2-10 min, Wang et al. (2006) reported formation of AG with higher VER by diverse from 20 to 80 %. Liu et al. (2005) show three parameters as mathematically unified into the single united selection pressure called minimum settling velocity (Vs_{min}).

2. MATERIALS AND METHODS

Granulation process is influenced by number of operational parameters like organic loading rate (OLR), settling time (ST),hydraulic shear force in terms of superficial upflow air velocity (SUAV) and volume exchange ratio (VER). To investigate the influence of all the above key operational parameters in a laboratory set-up of SBR is the main objective of this study.

2.1 Experimental setup

Feeding is done by pump and speed was accustomed according to desired dosing rate. The Solenoid valve with operating pressure was used for the present study. An aerator (air compressor) having flow of oxygen varying from minimum 0 L/min. to 10L/min by controlling with the flow regulator on the pump. The air is evenly distributed in the reactor with the aid of stone diffusers placed at the underside of the reactor surface. The reactor of glass with working volume 12L designed in the rectangular shape**.** The timers used for the set up to control automatically. The influent was fed after primary treatment. The drum was placed for the influent wastewater collection and treated wastewater collection with the capacity. The cycle of operation was 6 hrs (4 cycles/day) following feeding 1 min., non-aeration 24 min., aeration 300 min., settling 10 min. and decanting25 min. The laboratory set-up is shown in figure-1.

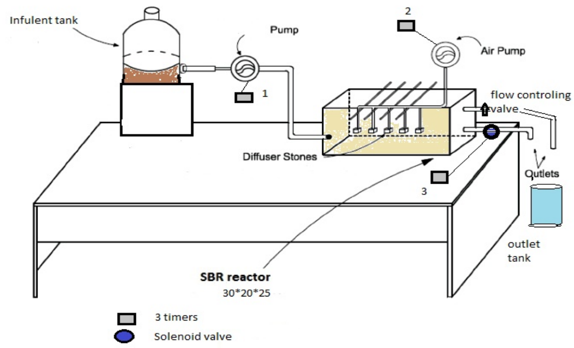


Figure-1: Laboratory setup

2.2 Seed sludge

The sample of seed sludge was used during the start-up, from the AS processing unit of a STP Bhattian, Ludhiana. The severe washout and bulking of sludge was observed but afterwards decreased and satisfactory performance was shown. The seed sludge has grey-blackish color with MLSS value of 5406 mg/L, MLVSS as 3068 mg/L and SVI as 178 mg/L.

2.3Feed composition

Kitchen wastewater is one of the main sectors that play an important role in domestic wastewater as it contains high organic content in itself. The wastewater with high organic content during the treatment and disposal creates problem where availability of land is a constraint and treatment by aerobic granulation is a viable option. Hence wastewater from the kitchen was selected as a real wastewater for the present study. The wastewater of the mess/kitchen consists of high COD strength which is a mean parameter for the formation of the granules and glucose as the sole carbon source was used for the study. The composition of the feeding wastewater was in rage as: pH 4-9, temp. 19-28^{0}C, DO 0.60-2 mg/L, TSS 250-450 mg/L, alkalinity 280-700 mg/L, BOD 500-800 mg/L, COD 900-1700 mg/L, ammonia 1.5-4 mg/L, nitrate 20-55 mg/L and total phosphorus 40-90 mg/L.

2.4 Analytical methods

The influent absorption of COD was not fixed at every time but the average COD value is around 1250 mg/Las per the feed composition given in table. Each stock of the feed coming was checked for total COD to maintain accuracy. The effluent sample was collected thrice a week and analyzed for COD, BOD, nitrate, total phosphate, DO and SS. While the reaction phase (aeration) grab samples of reactor stuffing were also drawn thrice a week and analyzed for mixed liquor suspended solids (MLSS), mixed liquor volatile suspended solid (MLVSS) and SVI. Sampling of the reactor content was done during the peak of aeration period in the operation cycle, at same time every day DO of the reactor contents were measured and monitored. Wastewater from mess/kitchen contains nitrogen and phosphate along with high amount of organic matter. The appearance and development of granules were monitored regularly. Analysis of morphological characteristics like specific gravity and settling velocity is done as and when required. The COD was analyzed by closed reflux method (Section 5220 B).Temperature of the influents and effluents were checked regularly with the help of temperature sensor. The pH of the feed and the effluent was measured using an Oakton pH meter ion 6+ meter kit. The DO meter (HACH Instruments, HQ30d) respectively as per standard methods each SVI_{30} determination was done by pouring the mixed liquor in 100ml cylinder and allowed to settle for 30 minutes then calculated with the help of MLSS concentration. The BOD_5 was measured for each inlet and the outlet sample along the other parameters using standard methods of examination of water and wastewater 5210B. The nitrate was measured using UV-VIS spectrophotometer as per standard method (4500-NO3-B). The Phosphorus content in the inlet and outlet sample was estimated using UV-VIS spectrophotometer as per standard (4500-P C) at the absorbance of 470nm.

3. RESULTS AND DISCUSSION

The four major parameters (organic loading rate, hydraulic shear force, settling time and volume exchange ratio) which may critically influence the process were selected as the main objectives of the present study. The experiment was conducted in the laboratory. The results are reported and discussed provides relative grading or ranking of the influencing factors, for the optimum performance of the reactor. So, while designing plants this will provide necessary help for the successful design and working of any treatment system. The wastewater of the mess/kitchen consists of high COD strength which is a key parameter for the formation of the granules and glucose as the sole carbon source was used for the study, using the optimal values obtained from the experiment.

The reactor was operated with high strength average COD 1250 mg/L for 40 days. Volume exchange ratio for the reactor was kept at 50%. The operational parameters chosen for the investigation i.e. temperature, COD, BOD, alkalinity, pH, Nitrates, TSS, DO and total phosphorus which were examined twice a week at both inlet and outlet to learn the efficiency of the reactor in the terms of pollutant removal. The MLSS, MLVSS and SVI of the reactor during the process was examined and maintained frequently to obtain accurate F/M ratio. The reactor was examined regularly to find the impact of feeds and substrate on the granule development and formation. Distinctive investigation with OLR 2.5 kg COD $m^{-3}d^{-1}$ was made on the reactor. The reactor was worked in progressive SBR cycles for 40 days of operation for 24×7. The Superficial up flow air velocity (hydrodynamic shear forces) was 2.78 cm/sec and HRT of the operation was 24 hr. All four parameters of the third objective were analyzed during each trail to learn their influence on the aerobic granule formation.

3.1 Parameters variations

The MLSSS/MLVSS ratio was 0.77. The sole carbon source, glucose was used in the reactor at every 48 hours. The MLSS and MLVSS values keep on increasing till the end with the ratio of 0.82 as MLSS/MLVSS. The SVI of the reactor was measured as 182 ml/g during the start-up, the washout of the sludge took place in the initial period but the stability increases in the reactor leading to drop in the SVI value at 36 ml/g at the end of the operation. As SVI at 80 ml/g or less indicates sludge with dense and rapid settling characteristics so it was the indication of the successful formation of the aerobic granules with good settling properties. The Chemical oxygen demand was measured regularly to keep the food to the microorganisms ratio accurate to achieve stability. The efficiency of the reactor for COD removal during was 91.78% (maximum) and 82.52% (minimum). Hence the results showed that the proficient removal. The biochemical oxygen demand is related to the chemical oxygen demand and the organic matter in the wastewater contains both biological as well as chemical oxygen requirements. The average value of the BOD in the wastewater was 685 mg/L as discussed above during the whole operation. The BOD efficiency during operation was at maximum value 90.51% and minimum at 85.38 %. The BOD result reveals the size of the wastewater treatment facilities. The alkalinity of the wastewater ranged between 300 – 700 mg/L as $CaCO_3$ and the maximum removal efficiency achieved was 67.70% and minimum as 65.97%. The total suspended solids removal efficiency was maximum at 84.72% and minimum at 79.01%. The pH of the mess/kitchen was not fixed as it has huge variations due to the salts used while cooking and the use of soap while cleaning. The pH must range in 6-9 for the growth of the biological life, as its extreme concentration cause difficulty in the biological treatment. During the treatment the pH value was adjusted by the use of HCL or NaOH by dilutions. The temperature was the room temperature ranged as 20–24^{0}C.The dissolved oxygen to achieve strong hydrodynamic shear forces, which results in AG formation acts as a key parameter. The air flow rate was 10 min/L resulting in SUAV 2.78 cm/sec. The influent had DO ≤ 1.5 while the treated wastewater had the DO≥7 mg/L which shows that the DO was much higher in the effluent as compared to the influents. The nitrates removal was 51.51% as maximum and 26.19% as minimum. The T.P. efficiency was as 76.92% at maximum and 73.33% at minimum.

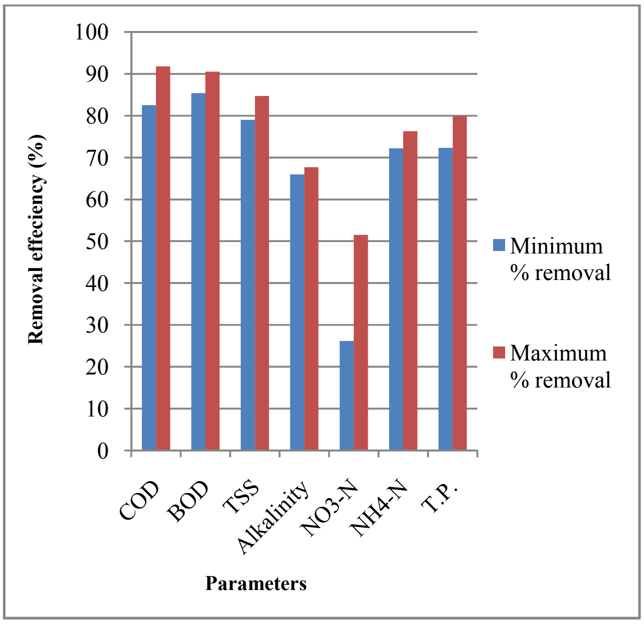


Figure-2: The minimum and maximum removal efficiency of the various parameters present in wastewater during the successful formation of aerobic granules.

3.2 FTIR (Fourier-change infrared spectroscopy) SPECTRA OF AG

The FTIR analysis of the sludge obtained at the end of the operation day 40 (mature granular sludge) was carried out to clarify the roles of functional groups and protein structure. The strong fluid or gas and high ghostly determination information in the aerobic granulation sludge is determined using Fourier-change infrared spectroscopy (FTIR). FTIR spectra were gone up against a Infrared and Raman spectrometer to distinguish the particle gatherings of crisp and treated granular slop. The sample (EPS extracted) of aerobic granules was directly used for spectrometry review.

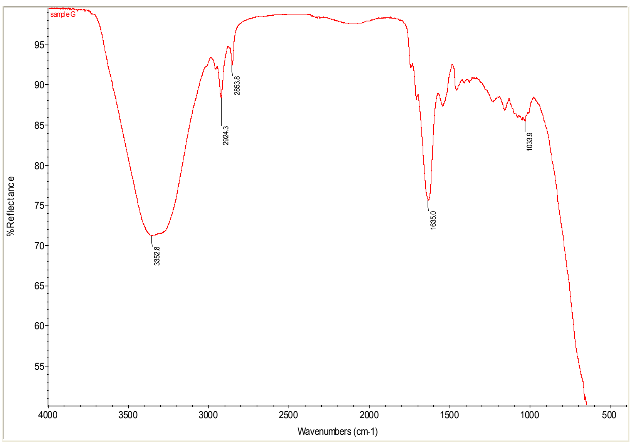
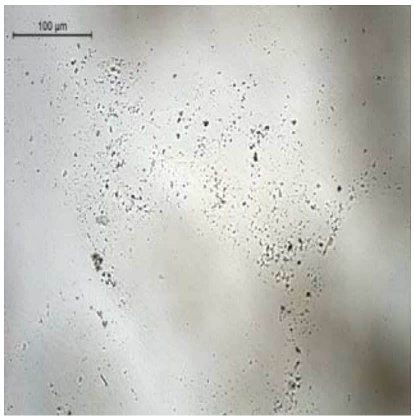


Figure-3: FTIR spectra of granule sludge

Amide, carboxylic, and carbohydrate functional groups as the major bands located in region 1800–900 cm^{-1}. The peak at 1635 cm^{-1} associated with the C=O stretching vibration of 3-turn helix and β-sheets in secondary protein structure, which favored bio-flocculation. The shoulder peak at 3352.8 cm^{-1} was associated with N-H asymmetric stretching of secondary amides of proteins, indicating that tyrosine protein might be a structural component of mature granular sludge (Zhu ey al.2012,Dong et al.,2017). Previous studies indicate that bands in 2921–2853 cm^{-1} were associated with the vibration (symmetric and asymmetric) stretching of CH_2 and CH_3 of proteins and lipids. The two most common functional groups in any carbohydrate, differences in the position and frequency of occurrence, C–O bonds which have been widely used to differentiate and quantify different isomers and forms of food science carbohydrates in the FTIR spectra .The 984 and 991 cm^{-1} peak shows weak vibrations indicating squat amounts of extracellular nucleic acids , which is a marker for sludge quality in terms of the health of the microbial population and biomass, lesser extracellular nucleic acid content showed elevated biological activity in mature granular.

3.3 Optical results

With the help of image analysis morphological characteristics like size, shape and regularity of the seed sludge was evaluated for granular sludge and seed sludge. The IA of the granule sludge system is done with the help of Optical microscope LEICA DM5000 B. The automated microscope LEICA DM5000 B is used for live cell analysis and cell morphology studies features.



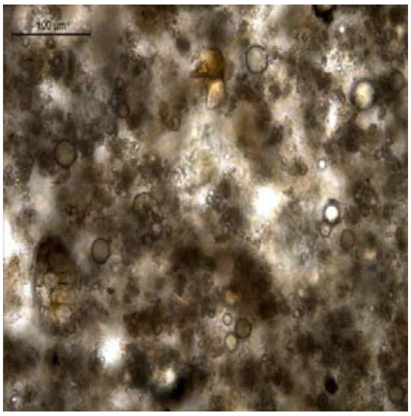


Figure-4: Optical image of the seed sludge at 100µm (a) and granule sludge at 100µm (b)

The optical results expose that the seed sludge optical structure was very loose hence, its settling property is poor but the optical structure of the AG is compact having good settling properties. So AGS is proven as the most promising technology for the effective treatment.

4. CONCLUSION

The OLR, SUAV, ST and VER are decisive parameters in the aerobic granule formation. The weak selection pressures such as low shear force, long settling time and low VER are not positive key to aerobic granulation formation technology. An important role of OLR had been studied on aerobic granules developed with OLR 2.5 kg COD m^{-3} d^{-1}, resulted in a high settling velocity, low SVI (36 ml/g) and COD removal of 91.78%. Further, shear force enriched the biomass growth by reducing the SVI and improvement in the sludge settling. So, most favorable shear force has to be chosen based on the performance and financial viable. The shorting of settling time can retain denser and more compact granules, whereas long settling time can't yield a fully developed granular sludge with good settling character. Even though, the high MLVSS/MLSS ratio (0.82) shown up a healthy biological treatment system.

5. ACKNOWLEDGEMENTS

This research work was supported by civil department of Guru Nanak Dev Engineering College, Ludhiana.

REFERENCES

[1] Adav, S.S., Lee, D.J., Show, K.Y., Tay, J.H., (2008), Aerobic granular sludge: Recent advances. *Biotechnology Advances*, 26, 411–423.

[2] Barr, Andrew E. Cook and Phillip L. Bond, (2010), Granule Formation Mechanisms within an Aerobic Wastewater System for Phosphorus Removal. *Applied and Environmental Microbiology*,2010, 76(22):7588

[3] Chen, J Ming, Brandon A. Yoza, J. Liang, X. Li, H. Guo, Z. Liu, J. Deng, Q. Wang, (2019), Characterization of aerobic granular sludge used for the treatment of petroleum wastewater. *Bioresource Technology*, 271, 353–359

[4] Franca, H. M. Pinheiro, Mark C.M, N. D. Lourenço., (2018), Stability of aerobic granules during long-term bioreactor operation. *Biotechnology Advances*, 0734-9750

[5] Guldhe, S. Kumari, L. Ramanna, P. Ram., P. S., I. Rawat, F. Bux ,(2017), Prospects, recent advancements and challenges of different wastewater streams for microalgal cultivation. *Journal of Environmental Management*, 203, 299e315

[6] Guo, Y., Zhang, B., Zhang, Z., Shi, W., Zhang, R., Cheng, J., Li, W., Cui, F., (2018), Enhanced aerobic granulation by applying the low-intensity direct current electric field via reactive iron anode. *Water Research*,10.095.

[7] Hamza, O. T. Iorhemen, J. H. Tay, (2016), Occurrence, impacts and removal of emerging substances of concern from wastewater. *Environmental Technology & Innovation*, 161–175

[8] Hamza, O. T. Iorhemen, M. S. Zaghloul, J. H. Tay., (2018), Rapid formation and characterization of aerobic granules in pilot-scale sequential batch reactor for high-strength organic wastewater treatment. *Journal of Water Process Engineering,* 22 (2018) 27–33.

[9]He, Q. Song, S. Zhang, Wei , Wang, (2018), Simultaneous nitrification, denitrification and phosphorus removal in an aerobic granular sequencing batch reactor with mixed carbon sources: reactor performance, extracellular polymeric substances and microbial successions. *Chemical Engineering Journal*, 331, 841–849

[10] Kang and Yuan, (2017), Long-term stability and nutrient removal efficiency of aerobic granules at low organic loads, *Bioresource Technology*, 234, 336–342.

[11] Li, Y., Liu, Y., Shen, L., Chen, F., (2008a), DO diffusion profile in aerobic granule and its microbiological implications. Enzyme and Microbial Technology, 43, 349–354.

[12] Liu, Y., Tay, J.H., (2002), The essential role of hydrodynamic shear force in the formation of biofilm and granular sludge. *Water Research*, 36 (7), 1653–1665.

[13] Liu, Y., Wang, Z.W., Tay, J.H., (2005b), A unified theory for upscaling aerobic granular sludge sequencing batch reactors. *Biotechnology Advances*, 23 (5), 335-344.

[14] Liu, Y.Q., Tay, J.H., (2008), Influence of starvation time on formation and stability of aerobic granules in sequencing batch reactors. *Bioresource Technology*, 99, 980-985.

[15] Long, X. Xuan, C. Yang, L. Zhang, Y. Cheng, J. Wang., (2019), Stability of aerobic granular sludge in a pilot scale sequencing batch reactor enhanced by granular particle size control. *Chemosphere* 225, 460-469

[16] Metcalf and Eddy, (2003), Wastewater Engineering Treatment and Reuse. (fourth ed.) *McGraw Hill*

[17] Nancharaiah and Reddy, (2018), Aerobic granular sludge technology: Mechanisms of granulation and biotechnological applications. *Bioresource Technology*, 247, 1128–1143.

[18] Qin, L., Tay, J.H., Liu, Y., (2004a), Selection pressure is a driving force of aerobic granulation in sequencing batch reactors. *Process Biochemistry*, 39, 579–584.

[19] Rollemberg, L. Q. Oliveira, Barros, Melo, Firmino, Santos, (2019), Effects of carbon source on the formation, stability, bioactivity and biodiversity of the aerobic granule sludge. *Bioresource Technology*, 278 (2019) 195–204.

[20] Tay, J.H., Liu, Q.S., Liu, Y., (2001a), Microscopic observation of aerobic granulation in sequential aerobic sludge blanket reactor. *Applied Microbiology*, 91, 168– 75.

[20] Tay, J.H., Liu, Q.S., Liu, Y., (2004a), The effect of upflow air velocity on the structure of aerobic granules cultivated in a sequencing batch reactor. *Water Science and Technology*, 49(11), 35-40.

[21] Wang, F., Yang, F.L., Zhang, X.W., Liu, Y.H., Zhang, H.M., Zhou, J., (2005), Effects of cycle time on properties of aerobic granules in sequencing batch airlift reactors. *World Journal of Microbiology and Biotechnology*, 21, 1379-1384.

[22] Wei Cai, Huang, Z. Lei, Zhang, Lee, Y. Adachi, (2019), Granulation of activated sludge using butyrate and valerate as additional carbon source and granular phosphorus removal capacity during wastewater treatment, *Bioresource Technology* 282, 269–274