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SOLUBILISATION OF WASTE ACTIVATED SLUDGE USING GARBAGE ENZYME PRODUCED FROM DIFFERENT PRE-CONSUMER ORGANIC WASTE.

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Abstract

The conversion of pre- consumer solid waste into value added product and utilisation of this for the treatment of activated sludge into reusable form without creating toxic effect to the environment is much focussed in the present days. In the present work, different types of garbage enzymes were produced from pre-consumer waste (pineapple, cauliflower, orange tomato, and mango dregs) and the characteristic of each garbage enzyme produced were investigated. The sludge solubilisation was performed with different types of garbage enzyme at different pH and time. When the treatment time increased from 48- 60 hours, a higher reduction of VSS (Volatile Suspended solids), TSS (Total Suspended solids) and also higher increase of solubility of COD (Chemical oxygen demand), TKN (Total Kjeldhal Nitrogen) and TP (Total phosphorus) were obtained for all types of garbage enzyme with pH 7. The pineapple and orange garbage enzyme showed higher reduction % of VSS and TSS nearly 20-25% and also increased % solubilisation of COD, TKN and TP nearly 20-25 %, 15-20%, 9-11% respectively in treated WAS (Waste activated sludge) compared with other garbage enzyme. This significant result showed that garbage enzyme solution has the capability to solubilize the complex (insoluble organic) compounds to soluble organic compounds which can be subsequently treated by anaerobic microbes to produce methane or hydrogen.

Key words: Pre-consumer waste; Garbage enzyme; Solubilisation; Waste activated sludge.

1. Introduction

In recent decades the developments of food processing industries are in the increasing trend in the developing countries. These types of industries are producing pre-consumer vegetable and fruit organic waste. On one hand improper disposal of these organic wastes along with other municipal solid waste in open dumps, generates unpleasant odour and increases the disease-causing organisms affecting the human health[1]. On the other hand organic waste disposal by landfill methods produce greenhouse gases and leachate affecting the atmosphere and the water environment in a larger extent[2]. The organic waste and sludge on landfill will ultimately degrade to produce carbon dioxide and methane thereby recirculating carbon back to the atmosphere causing global warming[3]. The discharge of greenhouse gases (GHGs) into the atmosphere is expected to have significant impact on the environment, human health and the economy. Subsequently an environment-friendly and sustainable technology at low cost is needed for the management and reuse of pre-consumer organic wastes[4].The pre-consumer organic waste can be used to produce garbage enzyme by fermentation. Garbage enzyme can be used as fertilizer, plant growth hormone, pesticides, insecticides, waste water treatment and antimicrobial agent[5].

The wastewater treatment plants, for industries and domestic (Municipal) wastewater, increasing day by day, to achieve the permissible limit for discharge of wastewater stipulated by environmental conservation and protection organisations like WHO (World Health Organization), pollution control boards etc. Due to increase of wastewater treatment plants, the generation sludge from them also increased significantly. Sludge produced is usually rich in poorly stabilised organic matter, affecting air, water and soil environment during storage and land spreading. The management of high sludge generated has become one of the challenging tasks for wastewater treatment plants[6]. The incineration and landfilling are the most common methods used to dispose sludge from waste water treatment plants. Recent

legislation in the developing countries is forcing the industries to reduce the amount of sludge entering landfills and adopting alternate methods to increase the recycling of sludge. Anaerobic digestion and composting are the suitable technology to treat the solid waste and it has been considered as a waste to wealth technology[7,8]. The operating cost of treatment of high-organic industrial wastewater is less by anaerobic digestion than by aerobic composting[9]. The production of biogas through anaerobic digestion offers the most environment friendly and energy-efficient technology for bioenergy production. The anaerobic digestion process has four essential stages namely hydrolysis, acidogenesis, acetogenesis and methanogens. Among these stages, the hydrolysis stage is a rate limiting step[10] as it involves depolymerisation of complex organic matter (insoluble state). This problem can be overcome by solubilizing the insoluble complex organic matter before entering anaerobic digestion because when the organic matter in soluble state, the microorganisms can digest the organic matter at a faster rate without further breakdown. Various physical[11, 12], chemical[13, 14, 15], and biological methods[16, 17, 18, 19] are available to solubilizes the complex organic matter but the biological (microbial or enzyme) methods are preferred due to eco-friendly and low operating cost[20, 21]. In addition, these methods are preferred to improve the solubility of sludge for further utilization or disposal. In the enzymatic hydrolysis, enzyme acts on WAS and releases nutrient into soluble form with reduction of solids[22]. Guo and Xu[23] reported that mostly in the biological treatment, the hydrolysis and degradation of complex biodegradable organic matters depended on the presence of hydrolytic enzymes. Nagina et al[24] reported the alkaline protease; a hydrolytic enzyme showed a beneficial effect in pathogen reduction, solids reduction and also improved dewatering of sewage sludge. Roman et al[25] investigated the combined effect of commercially available enzymes (Cellulase and pronase E) in solubilising the organic municipal waste activated sludge . All above cited investigation were based on hydrolysis of municipal sludge

treatment with commercial enzyme. Fazna and meera[26] studied the treatment of grey water using 5% and 10% of garbage enzyme and confirms that 10% garbage enzyme has the ability to reduce BOD, COD TDS up to 70, 50, and 39 %.respectively. Tang and Tong[27] reported that 9% solution of garbage enzyme in wastewater was found to be most cost-effective in removing ammonia nitrogen and phosphorus, and also neutralizing the domestic wastewater. Till now no attempt has been made to solubilise industrial waste activated sludge using garbage enzymes. Also the garbage enzyme production cost is cheaper as it produced from organic solid waste and hence one can get the advantage of both solid waste treatment of preconsumer organic waste and activated sludge solubilisation.

Therefore in the present work, an attempt was made to produce different types of garbage enzymes from pre-consumer waste (pineapple, cauliflower, orange tomato, and mango dregs separately) and the characteristic of each garbage enzyme produced were investigated. Also, the experiments were performed for the solubilisation of dairy waste activated sludge using different crude garbage enzymes. The parameters like VSS, TSS, Soluble COD, Soluble total Kjeldhal nitrogen, and soluble total phosphorus before and after treatment were studied to find out the effect of treatment time and pH on solubilisation of WAS.

2. Materials and methods

2.1 Production of garbage enzyme from different types of pre-consumer organic waste

In this study pre-consumer organic waste like pineapple, orange, tomato, cauliflower and mango peels and dregs were collected from vegetable markets and fruit shop in Tiruchirappalli and stored in refrigerator at 4^{o}C for the production of garbage enzyme. Five 2-liter airtight containers were taken and named as PGE (Pineapple garbage enzyme), OGE (Orange garbage enzyme), TGE (Tomato garbage enzyme), CGE (cauliflower garbage enzyme), and MGE (Mango garbage enzyme). To each container 500 ml of water and 50 grams of molasses were added with sufficient mixing. 150 grams of pineapple peels were

added and well mixed in PGE container and this procedure was repeated for remaining four containers with respective pre-consumer waste. These air tight containers were placed in a cool, dry and well-ventilated area for three months of fermentation.

2.2 Characterisation of different types of garbage enzyme

After three months of fermentation, the solution from each container was filtered and centrifuged at 3000 rpm for 30 minutes and the purified solution were stored separately in refrigerator at 4^{o}C. The parameters like pH, TS (Total solids), TDS (Total dissolved solids), BOD (Biological oxygen demand), COD and MPN (Most probable number) of different types of garbage enzyme were analysed according to the Standard methods [28] ,Citric acid concentration using HPLC method were determined and presented in the Table 1. From this Table 1 it is observed that all the above analysed parameters are more or less equal in all the enzyme solution and these values are taken into account while determining the environmental parameters of treated WAS with garbage enzyme solution.

-Table. 1-

Cell-free enzyme activities in the garbage enzyme were determined by centrifuging 10ml of solution at 3000 rpm for 10min. The supernatant was collected and used for the measurement of cell-free enzyme activities. Amylase activity was measured using the method of Bernfeld[29]. The assay solution containing 0.5ml of 1.0% soluble starch solution, 0.5ml of enzyme solution was incubated at 25^{o}C for 10min and 1 ml of dinitrosalicylic acid colour reagent was added. Then the mixture solution was incubated in boiling water bath for 5 minutes and cooled to room temperature. The absorbance of the mixture was read at 540nm. The reducing groups namely maltose released from starch were measured by the reduction of 3,5-dinitrosalicylic acid.

1 ml of garbage enzyme solution was mixed with 1ml 2% of casein, the resulting solution is prewarmed for 10 min to allow the reaction to proceed, the reaction was then terminated by the addition of 2ml of trichloroacetic acid solution and then incubated in a water bath at 35 ^{o}C for 10min. After the centrifugation of this mixture at 3000 rpm, 1 ml of supernatant was taken to it 5ml of Na_2CO_3 and 1ml folin phenol reagent were added[30]. The absorbance of the mixture was read at 660nm. The activity of protease was expressed as the amount of enzyme that releases 1mg of tyrosine equivalent per minutes.

Lipase activity was determined spectrophotometrically using the procedure of Pandey[31] et.al. The reaction mixture contains 50 µl of enzyme solution and 950 µl of substrate solution(1 part of 3.0 mM p-NPPin 2 propanol with 9parts of 0.4% Triton X100 and -0.1% gum Arabic).The reaction mixture was incubated at 37 ^{o}C for 20min and the absorbance of the mixture was read at 410nm. The activity of lipase was expressed as the amount of enzyme that releases 1 µmole of p- nitrophenol per minute of tyrosine equivalent per minutes.

2.3 Sampling and characterization of WAS sludge

The waste activated sludge (WAS) collected from a dairy industry at Trichy in Tamil Nadu (India) and stored in refrigerator at 4^{o}C. The characteristics of the raw sludge namely pH, TS, VSS, TSS, BOD, TCOD(Total chemical oxygen demand), SCOD(Soluble chemical oxygen demand), TKN(Total Kjeldhal nitrogen), STKN(Soluble Total Kjeldhal nitrogen), TP(Total phosphorus), STP(soluble total phosphorus) were analysed according to APHA methods [28]. Total proteins in the sludge was analysed with help of Lowry's method and carbohydrates by phenol sulphuric acid method and results are presented in Table 2.

-Table. 2-

2.4 Treatment of sludge using different types of garbage enzyme

20 ml of the concentrated PGE, OGE, TGE, CGE, MGE enzyme solution were diluted with 200 ml of ultra-pure water. The pH of garbage enzyme was adjusted to 3.5 and 7 with help of sodium citrate and phosphate buffer solution. These diluted garbage enzyme solution with pH adjusted were used for the treatment to improve the soluble COD, TKN and TP in WAS. Five numbers of 250 ml conical flasks were taken and 20 grams of WAS was added in all the flasks. After this 50 ml of diluted PGE, OGE, TGE, CGE and MGE enzyme were added separately in all the flasks, labelled respectively. Another 250ml conical flask labelled as control was taken and 20 grams of WAS only added with respective buffer solution. All the conical flask are kept in incubator shaker at 100 rpm and sludge treatment experiments were conducted for 60 hours by maintaining temperature at 35^{o}C. The solubility of sludge was evaluated by determining the COD solubilisation, VSS and TSS reduction and nutrient (nitrogen and phosphorus) solubilisation after treatment. At regular time interval the above parameters were estimated and the experiments were repeated twice to determine the consistency in the result obtained. The increase in COD Solubilisation %, STKN % and STP % were calculated by the following equation 1, 2, 3 respectively.

$$COD\ Solubilisation\ \% = \frac{SCOD\ after\ treatment}{TCOD\ after\ treatment} * 100 \qquad 1$$

$$TKN\ Solubilisation\ \% = \frac{STKN\ \ after\ treatment}{TKN\ after\ treatment} * 100 \qquad 2$$

$$TP\ Solubilisation\ \% = \frac{STP\ \ after\ treatment}{TP\ after\ treatment} * 100 \qquad 3$$

3. Result and discussion

3.1 Hydrolytic enzyme activity in garbage enzyme solution

In the present study cell free hydrolytic enzyme activities in garbage enzyme solution produced from different pre consumer organic waste were determined and results are

198 presented in Fig.1a and Fig.1b. From these figures, it is observed that all types of garbage
199 enzyme at pH 3.5 and pH 7 have amylase, protease and lipase activity. Hydrolytic enzyme
200 activity is higher for garbage enzyme solution with pH 7 when compared to garbage enzyme
201 solution with pH 3.5. Among them the amylase activity is higher for tomato garbage enzyme
202 solution and lower for mango garbage enzyme. Similarly protease activity is higher for
203 pineapple garbage enzyme solution and lower for tomato garbage enzyme solution. Lipase
204 activity is higher for pineapple garbage enzyme and all other garbage enzyme solution
205 contains comparable lipase activity. Thus this experiment confirms the presence of hydrolytic
206 enzyme activity in all types of garbage enzyme solution at pH 7 is higher when compared
207 with pH 3.

208 -Fig. 1a-

209 -Fig. 1b-

210

211 **3.2 VSS and TSS reduction**

212 Stability and effectiveness of sludge treatment process can be determined using VSS and TSS
213 reduction[32]. The removal percentage of volatile solids and suspended solids from sludge after
214 treatment with different types of garbage enzymes (pH 3.5 and 7) are presented in Figs. 2a,
215 2b and Figs. 3a, 3b respectively. From these figures it is observed that the removal percentage
216 of VSS and TSS increased for all types of garbage enzyme, when the treatment time
217 increased from 12- 60 hours at both the pH. But the significant reduction in VSS and TSS is
218 higher for the sludge treated with garbage enzyme at a pH 7 when compared with garbage
219 enzyme at a pH 3.5. The reason for higher reduction of VSS and TSS at pH 7 is due to
220 enhanced activity of hydrolytic enzyme at pH 7 whereas enzyme activity got suppressed at

pH 3.5 due to acidic condition. Similarly Qi Yanga et al., [22]demonstrated municipal secondary sludge treatment with protease, amylase, mixed-enzyme treatment and concluded that the solid reduction was found to be 42%, 56.32% and 68.43% of respectively.

-Fig. 2a-

-Fig. 2b-

It is also observed that WAS treated with PGE and OGE showed increase in VSS and TSS reduction from 21 -25 %. The reason for higher VSS and TSS reduction by PGE and OGE enzyme treated sludge is explained as follows,

OGE contains organic acids mainly citric acid as it was produced from fermentation of citrus fruit peels. Citric acid has the power to disturb the extracellular polymeric substances (EPS) and releases hydrolytic enzyme [12, 33]. In addition to garbage enzyme these released hydrolytic enzyme also has an impact on sludge solubilisation. Thus citric acid has a property to enhance the sludge matrix breakage, which in turn resulted in higher % of VSS and TSS reduction, when sludge treated with OGE. MGE has lower citric acid concentration when compared to other garbage enzyme thus it shows lower removal % of solids (Table 1).

The PGE enzyme solution is produced by fermentation from the peels of pineapple along with water and molasses. During the production of this enzyme, at acidic condition protease from the peels of pineapple released into garbage solution. This extracellular Proteolytic enzyme has higher activity at pH 7, which activates the hydrolysis of protein present in dairy waste activated sludge. Because of this reason the VSS and TSS reduction % is increased, when sludge treated with PGE.

-Fig. 3a-

-Fig. 3b-

3.3 COD Solubilisation

Treatment process of sludge aims to improve the biodegradability and bioavailability of sludge organic matter in soluble form. The increase in biodegradability is directly proportional to the solubilized COD[34, 35.] Since SCOD calculation is considered as a main parameter for the evaluation of the maximum level of sludge solubilisation[32]. Figs 4a and 4b, present the effect of different garbage enzyme on COD solubilisation of WAS at pH 3.5 and 7 respectively. From Figs 4a and 4b, it is observed that the COD solubilisation of WAS at both the pH (3.5 and 7) starts increasing for all the types of garbage enzyme (PGE, OGE, TGE, CGE, MGE) when compared to control (WAS with respective buffer solution) while the treatment time increased from 12 -60 hours. Also, the sludge treated with garbage enzyme at a pH 7 showed significant increase of COD solubilisation, compared with garbage enzyme at a pH 3.5. The reason for higher COD solubilisation rate at pH 7 is due to the enhanced activity of hydrolytic enzyme at that pH whereas its activity got suppressed at pH 3.5(acidic), due to loss in enzyme stability. The increase in SCOD level in treated sludge indicates that the sludge containing large amount of soluble substances. When organic particles are solubilised and it can be readily degraded by microorganism during anaerobic digestion process to produce biogas. Similarly Roman et al[25] investigated the combined effect of commercially available enzymes (Cellulase and pronase E) in solubilising the organic municipal waste activated sludge (MWAS) and reported the increases in SCOD level in MWAS after treatment with the enzymes.

-Fig. 4a-

-Fig. 4b-

3.4 TKN and TP solubilisation

WAS contains a large amount of nitrogenous compounds in the form of organic nitrogen, ammonia, and ammonium and among them most of them are in insoluble complex form namely amino acids, amino sugars and proteins[36]. By observing the characteristic of WAS before treatment with garbage enzyme solution (Table 2) it is seen that less than 20-25% of nitrogenous compounds are in soluble form and remaining 75- 80% are in insoluble in nature. Therefore solubilisation process of such waste activated sludge is required to increase the soluble nitrogen components, which in turn minimizes the rate limiting hydrolysis stage during biological treatment of sludge. Hence, the sludge was treated with different garbage enzyme solution and STKN in WAS after treatment with respect to treatment time is presented in Figs 5a and 5b. From Figs 5a and 5b, it is observed that soluble TKN increases when compared to control while the treatment time increases from 12 to 60 hours. The reason for the increasing soluble TKN % is due to the presence of organic acids (carbon source) in garbage enzyme solution.

-Fig. 5a-

-Fig. 5b-

By observing the characteristics of WAS before treatment with garbage enzyme solution (Table 2) it is seen that less than 9% of phosphorus are in soluble form and remaining 91 % are insoluble form. The phosphorus content of waste activated sludge includes orthophosphate, polyphosphate and organic phosphate. Polyphosphate (insoluble) in sludge should be converted to orthophosphate (soluble) by the process of hydrolysis[36] .Therefore, WAS was treated with different garbage enzyme solution and STP in WAS after treatment with respect to treatment time is presented in Figs 6a and 6b. From the Figs 6a and 6b, it is observed that the increase of soluble phosphorus in WAS, when compared to control while the treatment time increases from 12 to 60 hours for all types of garbage enzymes. The

maximum increase of STKN (15 -20 %) and STP (9-11%) were found, when the sludge was treated with PGE and OGE.

The reason for increase in solubilisation of TKN and TP in treated sludge is due the presence of organic acid (carbon source) and hydrolytic enzyme in the garbage enzyme solution, which helped in breakdown of insoluble form of minerals to soluble from. Ely Nahas[37] reported the similar observation, when investigating the microbial solubilisation of phosphorus, carbon and nitrogen in soil.

-Fig. 6a-

-Fig. 6b-

4. Conclusion

The cell free hydrolytic enzyme activities in garbage enzyme solution produced from different pre consumer organic waste were determined. Thus this experiment confirms the presence of hydrolytic enzyme activity in all types of garbage enzyme solution at pH 7. The WAS treatment was performed with different types of garbage enzyme at pH 3.5 and 7 and treatment time (12, 24, 36, 48 and 60 hours). The pineapple and orange garbage enzyme showed slightly higher reduction % of VS and SS nearly 20-25% and also increased % solubilisation of COD, TKN and TP nearly 20-25 %, 15-20%, 9-11% respectively were obtained in treated WAS. The above significant results showed that garbage enzyme solution have the capability to solubilize the complex (i.e.) insoluble organic compounds to soluble organic compounds which can be subsequently treated by anaerobic microbes to produce methane or hydrogen.

References

[1]Dan iel Hoornweg ., Perinaz Bhada ., What a waste- A Global Review of Solid Waste Management, Urban Development Series, 2012.World bank, 15, Washington, USA.

[2]Daniel Pleissner., Carol Sze Ki Lin., Sustain Chem Processes., 2013, 1, 21.

[3]Levlin E., International scientific seminar, Research and application of new technologies in wastewater treatment and municipal solid waste diposal in Ukraine, Sweden and Poland 23-25 September 2009 Stockholm, Polish-Swedish, TRITALWR report, 2010, 3026, 95-104.

[4]Luque, R., Clark, J.H., Sustain Chem Processes, 2013,1,10.

[5]Bhavani prakash., 2011, http://www.ecowalkthetalk.com Accessed 17 December 2013.

[6]Neyens, E.,Baeyens, J., J.Hazard.Mater, 2003, 98,51-67.

[7]Sonakya, V., Raizada, N., Kalia, V.C., Biotechnol. Lett. 2001, 23,1463–1466.

[8]Garg, V.K., Renuka Gupta., Ecotox Environ Safe. 2011, 74, 19–24.

[9]Clisso,M.,(2010), http://water.me.vccs.edu/courses/env108/anaerobic.htm, Accessed 17 November 2013.

[10] Bougrier, C., Delgenès, J.P., Carrère, H., Biochem.Eng. J., 2008, 34, 20–27.

[11] Jianbo Xu., Haiping Yuan., Jinxian Lin., Wenxiang Yuan., J. Taiwan Institute of Chem Engineers. 2014, 45, 2531–2536.

[12] Gayathri, T., Kavitha, S., Adish Kumar, S., Kaliappan, S., Ick Tae Yeom, Rajesh Banu, J., Ultrason Sonochem. 2015, 22 , 333–340.

[13] Tae-Hun Kim., Tak-Hyun Kim., Seungho Yu., Youn Ku Nam., Dong-Kyu Choi., Sang Ryul Lee., Myun-Joo Lee., J. Ind. Eng. Chem., 2007. 13, 7, 1149-1153.

[14] Heo, N.H., Park, S,C,, Lee ,J.S., Kang H., Water Sci Technol. 2003, 48(8), 211-9

[15] Xu Zhou, Guangming Jiang, Qilin Wang and Zhiguo Yuan, RSC Adv., 2014, 4, 50644.

342 [16] Veera Lakshmi, M., Merrylin, J., Kavitha , S., Adish Kumar, S., Rajesh Banu, J.,
343 Ick-Tae Yeom., Environ Sci Pollut Res., 2014, 21,2733–2743.
344 [17] Kavitha, S., Jayashree, C., Adish Kumar, S., Yeom, I. T., Rajesh Banu, J.,
345 Bioresour. Technol., 2014, 168, 159–166.
346 [18] Merrylin, J., Adish Kumar, S., Kaliappan, S., Ick-Tae Yeom ., Rajesh Banu J.,
347 Environ. Technol., 2013, 34, 2113–2123.
348 [19] S. Kavitha, T. Saranya, S. Kaliappan, S. A. Kumar, I. Tae and J. R. Banu, 2015, 175,
349 396–405.
350 [20] Tsuyoshi Imai., Yuyu Liu., Masao Ukita., Yung-Tse Hung., Environmental
351 Bioengineering,Handbook of Environmental Engineering, 2010, 11,75-122.
352 [21] Parawira, W., Crit. Rev. Biotechnol. 2012, 32, 172–186
353 [22] Qi Yanga., Kun Luo., Xiao-ming Li., Dong-bo Wang., Wei Zheng., Guang-ming
354 Zeng., Jing-jin Liu., Bioresource Technol., 2010, 101, 2924–2930.
355 [23] Guo, J.S., Xu, Y,F., Review of Enzymatic Sludge Hydrolysis. J Bioremed Biodegrad.
356 2011, 2,130.
357 [24] Nagina parmar., Ajay Singh., Owen, P. ward., World J Microb Biotec., 2001, 17,169-
358 172.
359 [25] Roman, H.J., Burgess, J.E., Pletschke, B., Afr J Biotechnol., 2006, 5 (10), 963-967.
360 [26] Fazna Nazim., Meera, V., (2013). Bonfring International Journal of Industrial
361 Engineering and Management Science. 3(4), 111- 117.
362 [27] Fu E. Tang., Chung W. Tong., World Academy of Science, Engineering and
363 Technology 60 2011
364 [28] American public Health Association (APHA)., Standard Methods for examination of
365 water and waste water, 21 st ed., 2005,Washington, DC.

366 [29] Bernfeld P., 1951, Enzymes of starch degradation and synthesis. In: Nord FF (Ed),
367 Advances in Enzymology, New York, Interscience Publication,. pp. 379
368 [30] Lowry O H, Rosebrough N J, Farr A L and Randall R J, J. Biol.Chem, 1951, 193: 265
369 [31] Pandey, A., Benjamin, S., Nigam, P., Soccol C.R., Krieger N., Biotechnol Appl.
370 1999, 29: 119-131.
371 [32] Uma Rani, R., Adish Kumar, S., Kaliappan, S., IckTae Yeom., Rajesh Banu, J., Waste
372 Management, 2013, 33, 1119–1127.
373 [33] Wawrzyńczyk, J., Recktenwald, M., Norrlöw, O., Dey Szwajcer, E., Afr J
374 Biotechnol., 2007, 4 (17), 1994-1999.
375 [34] Dwyer J., Starrenburg D., Tait S., Barr K., Batstone D. J., Lant P. Water Research,
376 2008, 42, 4699–4709
377 [35] Kavitha, S., Kumar, S. A., Kaliappan, S., Tae I., Banu, J. R., Bioresour. Technol.,
378 2014, 169, 700–706.
379 [36] Met calf., Eddy., Wastewater Engineering: Treatment and Reuse, Fourth Ed.,
380 McGraw-Hill, 2002, New York.
381 [37] Ely Nahas., First International Meeting on Microbial Phosphate Solubilisation,
382 Developments in Plant and Soil Sciences. 2007,102, 111-115
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391 **Figure captions**

392 Fig.1a. Determination of Hydrolytic enzyme activity in different garbage enzyme solution
393 with pH 3.5.

394 Fig.1b. Determination of Hydrolytic enzyme activity in different garbage enzyme solution
395 with pH 7.

396 Fig.2a. Effect of garbage enzyme with pH 3.5 on VSS reduction in WAS with respect to
397 treatment time.

398 Fig.2b. Effect of garbage enzyme with pH 7 on VSS reduction in WAS with respect to
399 treatment time.

400 Fig.3a. Effect of garbage enzyme with pH 3.5 on TSS reduction in treated WAS with respect
401 to treatment time.

402 Fig.3b. Effect of garbage enzyme with pH 7 on TSS reduction in treated WAS with respect to
403 treatment time.

404 Fig.4a. Effect of garbage enzyme with pH 3.5 on SCOD increase in treated WAS with respect
405 to treatment time.

406 Fig.4b. Effect of garbage enzyme with pH 7 on SCOD increase in treated WAS with respect
407 to treatment time.

408 Fig.5b. Effect of garbage enzyme with pH 3.5 on STKN increase in treated WAS with respect
409 to treatment time.

410 Fig.5b. Effect of garbage enzyme with pH 7 on STKN increase in treated WAS with respect
411 to treatment time.

412 Fig.6a. Effect of garbage enzyme with pH 3.5 on STP increase in treated WAS with respect
413 to treatment time.

414 Fig.6b. Effect of garbage enzyme with pH 7 on STP increase in treated WAS with respect to
415 treatment time.

416

417

418 Table 1 Characteristic of different types of garbage enzymes

Parameters	PGE	OGE	TGE	CGE	MGE
			Range		
pH	3.4-3.7	3.2.-3.3	3.1-3.4	3.4-3.6	3.5-3.7
TDS (mg/l)	997-1006	995-1008	1013-1019	1006-1020	1009-1027
BOD (mg/l)	70-79	65-74	69-81	67-79	71-78
COD (mg/l)	150-157	152-160	151-158	154-160	151-154
MPN(C.F.U/ml)	<3	<3	<3	<3	<3
Citric acid (mg/ml)	2.367	4.402	1.483	1.075	0.5734

419

420 Table 2 Characteristic of dairy waste activated sludge

Parameters	Value
pH	6.7-7.2
Total Solids	9038mg/l
Volatile Suspended solids	4971 mg/l
Total Suspended Solids	5034 mg/l
Total COD	24094 mg/l
Soluble COD	853 mg/l
TKN	1209 mg/l
STKN	283 mg/l
TP	326 mg/l
STP	25mg/l
Total protein	814 mg/l
Carbohydrates	366 mg/l
MPN(C.F.U/100ml)	$9.7*10^7$

421

422

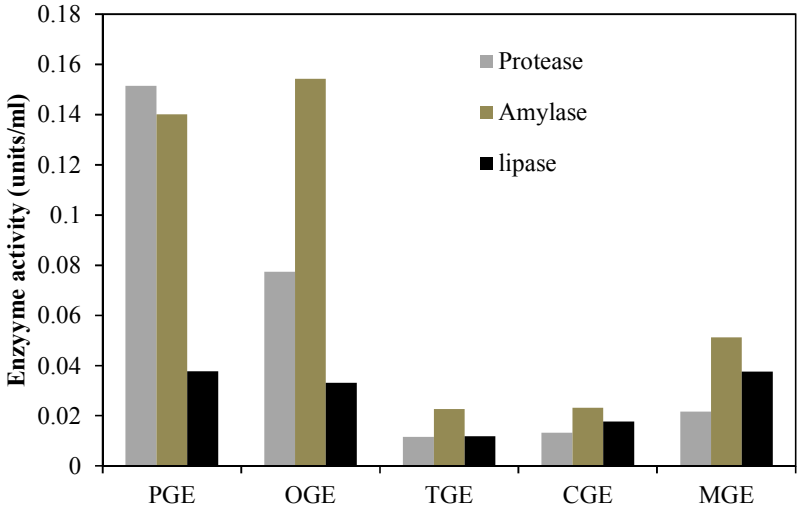

Fig.1a.

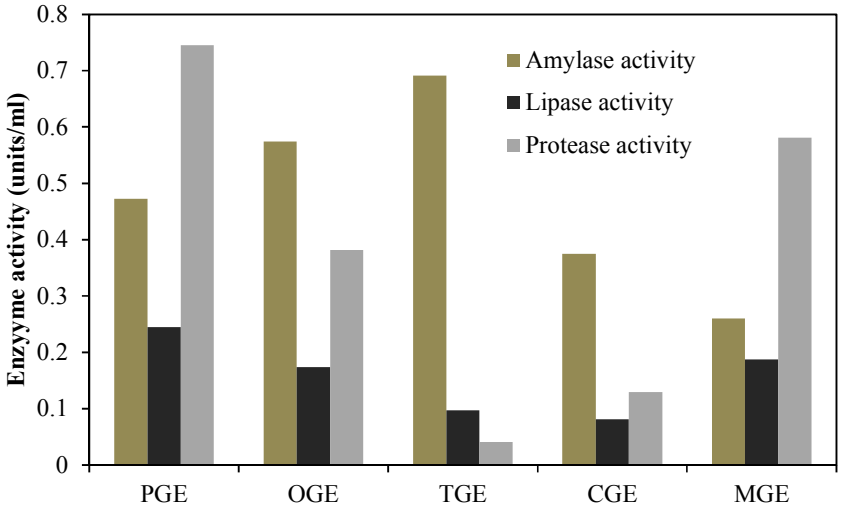

Fig.1b.

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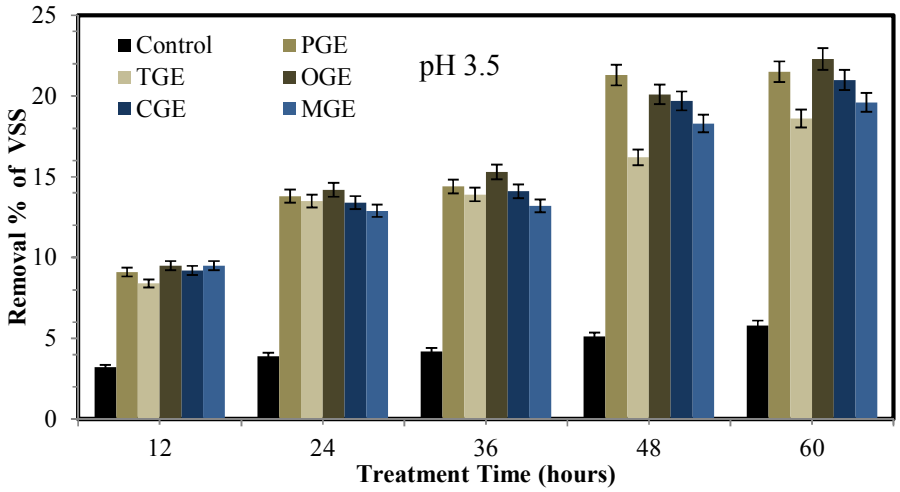

Fig.2a.

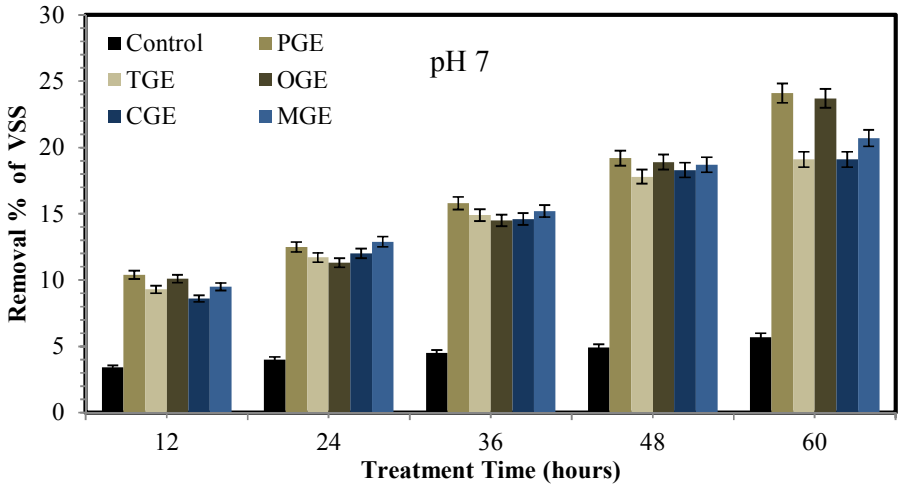

Fig.2b.

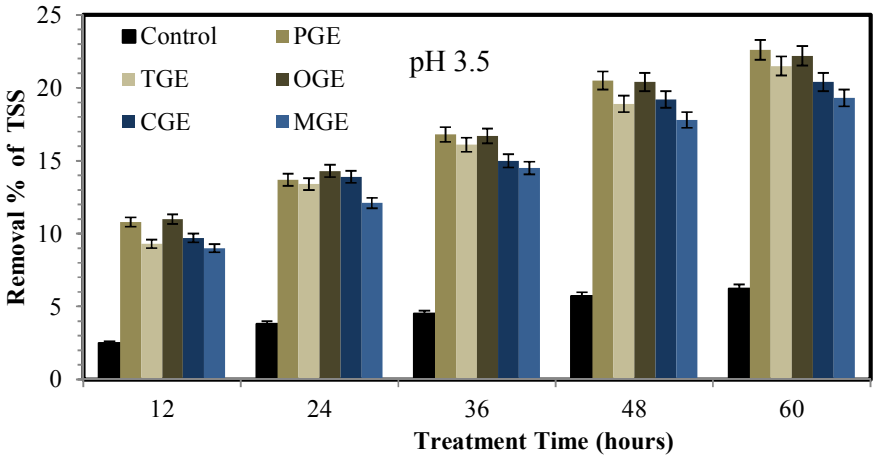

Fig.3a.

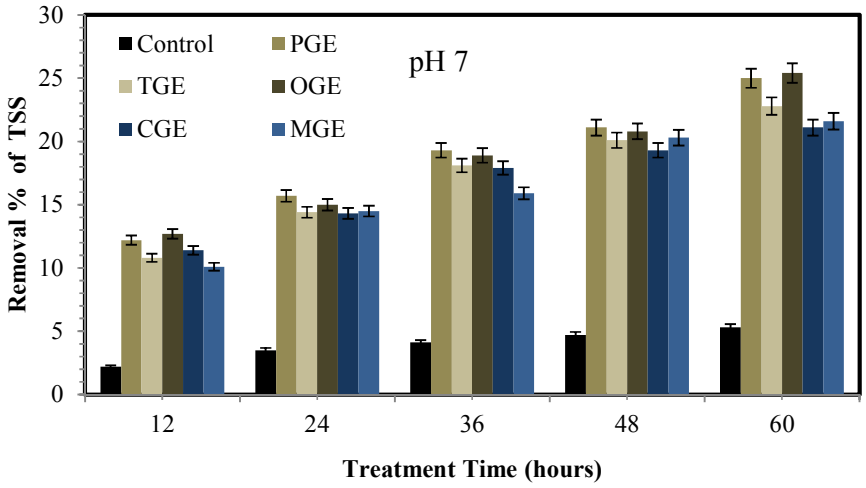

Fig.3b.

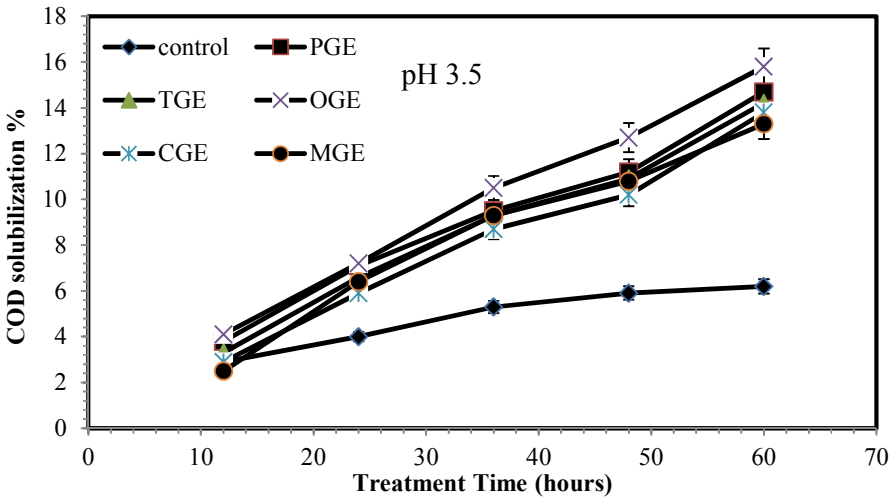

Fig.4a.

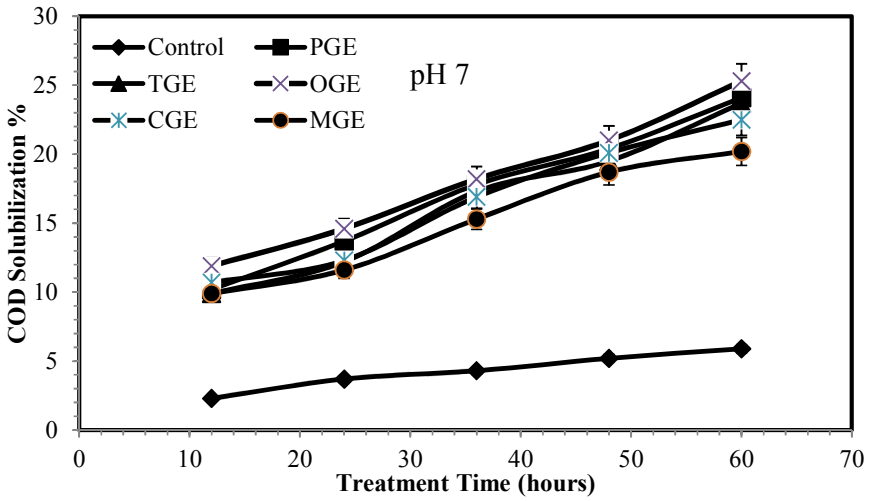

Fig.4b.

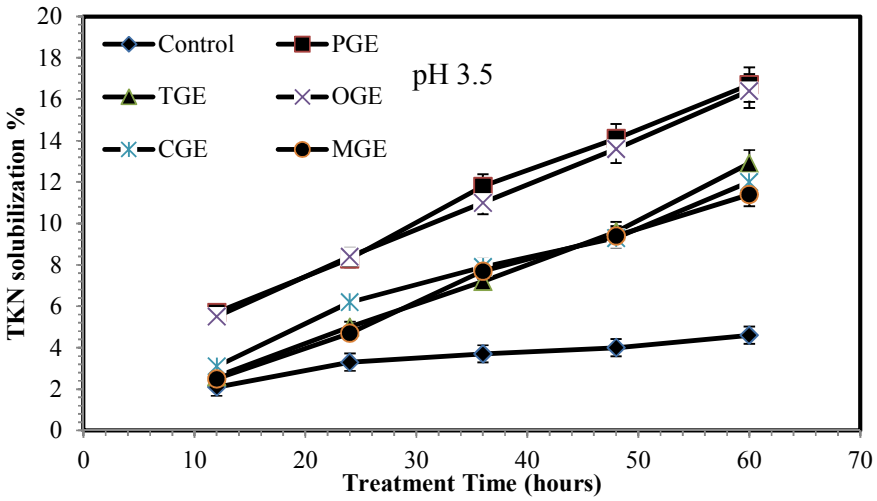

Fig.5a.

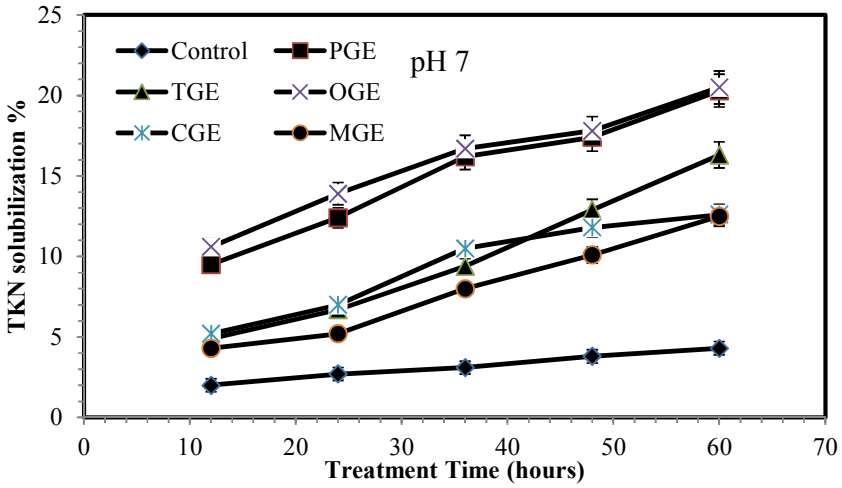

Fig.5b.

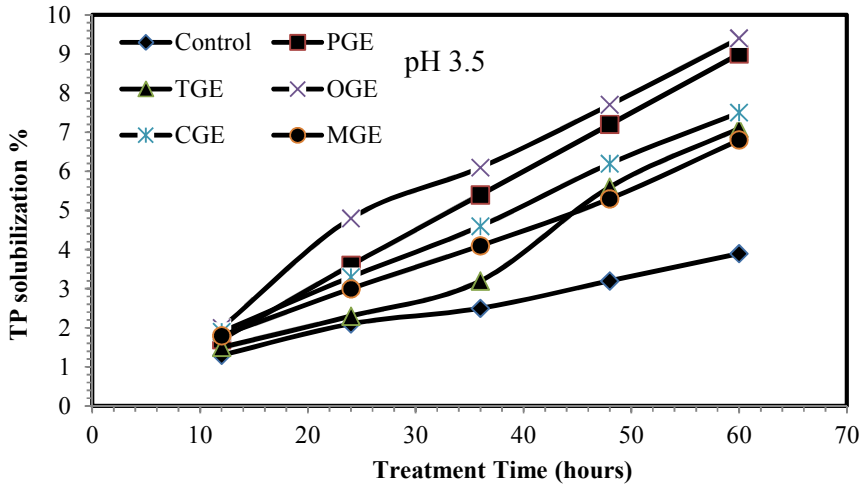

Fig.6a.

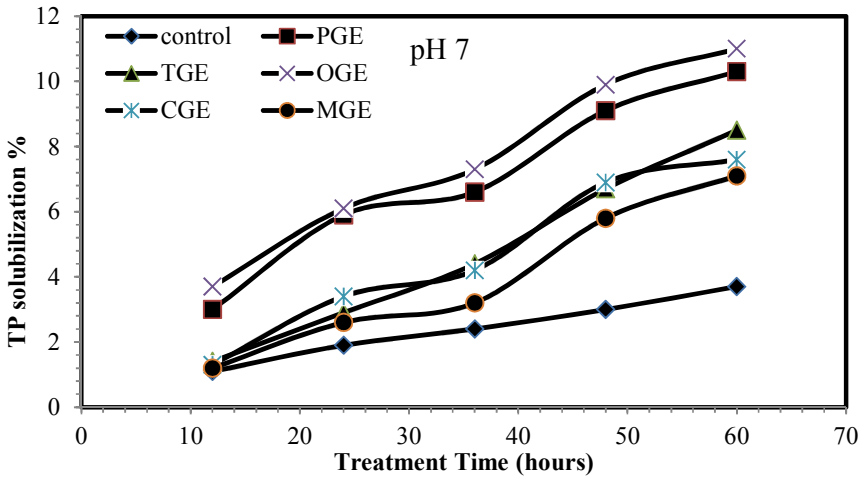

Fig.6b.