

# Energy-efficient stirred-tank photobioreactors for simultaneous carbon capture and municipal wastewater treatment

K. Mohammed, S. Z. Ahammad, P. J. Sallis and C. R. Mota

## ABSTRACT

Algal based wastewater treatment (WWT) technologies are attracting renewed attention because they couple energy-efficient sustainable treatment with carbon capture, and reduce the carbon footprint of the process. A low-cost energy-efficient mixed microalgal culture-based pilot WWT system, coupled with carbon dioxide (CO<sub>2</sub>) sequestration, was investigated. The 21 L stirred-tank photobioreactors (STPBR) used light-emitting diodes as the light source, resulting in substantially reduced operational costs. The STPBR were operated at average optimal light intensity of 582.7 μmol. s<sup>-1</sup>.m<sup>-2</sup>, treating synthetic municipal wastewater containing approximately 250, 90 and 10 mg.L<sup>-1</sup> of soluble chemical oxygen demand (SCOD), ammonium (NH<sub>4</sub>-N), and phosphate, respectively. The STPBR were maintained for 64 days without oxygen supplementation, but had a supply of CO<sub>2</sub> (25 mL.min<sup>-1</sup>, 25% v/v in N<sub>2</sub>). Relatively high SCOD removal efficiency (>70%) was achieved in all STPBR. Low operational cost was achieved by eliminating the need for mechanical aeration, with microalgal photosynthesis providing all oxygenation. The STPBR achieved an energy saving of up to 95%, compared to the conventional AS system. This study demonstrates that microalgal photobioreactors can provide effective WWT and carbon capture, simultaneously, in a system with potential for scaling-up to municipal WWT plants.

**Key words** | activated sludge, bacterial oxygen requirement, carbon capture, light-emitting diodes, microalgae, synthetic municipal wastewater

## INTRODUCTION

Energy is required to treat municipal wastewater, this being obtained mostly from the combustion of fossil fuels. The energy requirement for conventional wastewater treatment (WWT) processes, such as activated sludge (AS), and their associated carbon footprint, is relatively high (Ahammad *et al.* 2013). Furthermore, undesirable carbon emissions and escalating energy prices are additional drawbacks to current WWT practices. This calls for the development and optimisation of energy-efficient carbon-neutral WWT processes to help mitigate climate change. Novel WWT technologies that utilise microalgae could address these issues by harnessing the ability of microalgae to capture carbon and assimilate nutrients from wastewater through photosynthesis (Negoro *et al.* 1993).

The idea of using microalgal photosynthesis to treat municipal wastewater is not novel (e.g. Ludwig *et al.* 1951;

Oswald *et al.* 1957). However, traditional microalgal and related WWT methods have many limitations, but recently developed semiconductor technologies now permit the development of energy-efficient carbon-neutral treatment technologies. The quality and quantity of light photons is an essential and limiting factor in microalgal growth and productivity (Matthijs *et al.* 1996). As such, optimising this parameter can greatly influence the efficiency of microalgal WWT systems. In addition, exploitation of the dissolved oxygen (DO), produced through microalgal photosynthesis, can serve as a potential investment towards minimising aeration in the aerobic systems, and subsequent reduction in the energy cost of the overall process.

In view of the need to provide high quality photosynthetically active radiation, and to overcome the light limitation and photo-inhibition commonly encountered in microalgal

**K. Mohammed** (corresponding author)

**S. Z. Ahammad**

**P. J. Sallis**

**C. R. Mota**

School of Civil Engineering and Geosciences,  
Newcastle University,  
Newcastle upon Tyne, NE1 7RU,  
United Kingdom  
E-mail: kmohammed.civ@buk.edu.ng

**K. Mohammed**

Department of Civil Engineering,  
Bayero University,  
PMB 3011, Kano,  
Nigeria

**S. Z. Ahammad**

Department of Biochemical Engineering and  
Biotechnology,  
Indian Institute of Technology Delhi,  
Hauz Khas, New Delhi,  
India

**C. R. Mota**

Departamento de Engenharia Sanitaria e  
Ambiental,  
Universidade Federal de Minas Gerais,  
Belo Horizonte,  
Minas Gerais CEP 31270-901,  
Brasil

cultivation systems (Curtis *et al.* 1994; Heaven *et al.* 2005), this study explored the use of red light-emitting diodes (LED) as the sole source of microalgal illumination in stirred-tank photobioreactors (STPBR) treating synthetic municipal wastewater. Red photons have a property of being weakly absorbed by water molecules (Blankenship 2002), allowing most of the irradiance to reach microalgal cells, thus reducing light limitations. Red light also enhances photosynthetic efficiency due to its characteristic relatively long wavelength (Curtis *et al.* 1994), which overlaps with the chlorophyll absorption band (Hall & Rao 1999). It also has a low tendency to exert photoinhibition, due to its relatively low energy (Matthijs *et al.* 1996; Blankenship 2002). In addition, the STPBR used in this study were enriched with industrial-grade carbon dioxide (CO<sub>2</sub>) to avoid carbon limitation, and to promote photosynthetic carbon capture. The resulting system potentially serves as a green technology that can simultaneously capture carbon and treat municipal wastewater.

## MATERIALS AND METHODS

### Experimental set-up

Pilot-scale (21 L), internally-illuminated, transparent Plexiglas STPBR were used to treat modified synthetic municipal wastewater (MSMW) in the laboratory. Red LED were used to illuminate three STPBR under optimal irradiance, determined previously by Mohammed *et al.*

(2013). A mixture of aerobic AS seed culture collected from Spennymoor municipal WWT plant (UK), and mixed microalgal culture was used to inoculate the STPBR. Three different mixed liquor volatile suspended solids (MLVSS) concentrations of 50, 300 and 600 mg.L<sup>-1</sup> were used as the control parameter for STPBR operation. The STPBR were operated at 4-d hydraulic retention time (HRT), and 16:8 light-dark cycles, in continuous mode, for 64 days. Peristaltic pumps (Watson-Marlow, UK) were used to supply wastewater continuously from a feed tank. Settlers were used to return settled biomass into the STPBR, maintaining the desired MLVSS concentrations. Optical density (OD) measurements and gravimetric analyses were used to estimate MLVSS concentration. Mixing was provided by a single rectangular impeller, 150 × 80 mm, rotating at 100 ± 1 rpm, driven by an overhead stirrer (IKA, UK).

### Wastewater

The MSMW used for STPBR was adapted from Bracklow *et al.* (2007), the composition and characteristics being shown in Table 1.

A concentrate of the wastewater (Table 1) was prepared and autoclaved (Rodwell Scientific Equipment, UK) at 120 °C for 15 minutes, and stored at 4 °C for the duration of the experiments. The algal culture growth medium (ACGM) was prepared from a mixture of Modified Bold's Basal Media (MBBM), a portion of the concentrate, and distilled water; this was fed to the STPBR, as set out in Table 2.

**Table 1** | Composition and characteristics of the MSMW

Composition of concentrate		Characteristics of the ACGM feedstock	
Constituents	Concentration (g.L <sup>-1</sup> )	Parameter	Concentration (mg.L <sup>-1</sup> ) <sup>a</sup>
Peptone	1.740	SCOD	254 (1.73)
Yeast extract	5.220	NH <sub>4</sub> -N	88.1 (1.19)
Glucose	6.100	Total carbon	391.4 (5.92)
NH <sub>4</sub> -acetate	31.76	Inorganic carbon	n/d
KH <sub>2</sub> PO <sub>4</sub>	0.585	PO <sub>4</sub> <sup>3-</sup>	12.4 (0.081)
MgNH <sub>4</sub> PO <sub>4</sub>	0.725	NO <sub>2</sub> -N	n/d
K <sub>2</sub> HPO <sub>4</sub>	0.525	NO <sub>3</sub> -N	0.015 (0.001)
Urea	9.170	DO	3.50 (0.006)
NH <sub>4</sub> Cl	1.280	pH	5.70 (0.00)
FeSO <sub>4</sub> ·7H <sub>2</sub> O	0.580	Temperature (°C)	22.1 (0.10)

<sup>a</sup>Unit not applicable to pH and temperature.

n/d, not detected.

Values in parentheses are standard deviations.

**Table 2** | Operating conditions for the STPBR run at 4-d HRT

Bioreactor conditions				ACGM feedstock composition (%)			
Bioreactor	V (L)	Q (L.day <sup>-1</sup> )	MLVSS (mg.L <sup>-1</sup> )	MBBM	MSMW	Distilled water	Total
STPBR1	14.92	3.73	50	1.4	1.0	97.6	100
STPBR2	14.92	3.73	300	1.4	1.0	97.6	100
STPBR3	15.44	3.86	600	1.4	1.0	97.6	100

## Inoculum

The STPBR were inoculated with a mixed culture of microalgae and AS at an estimated initial microalgae-bacteria ratio of about 90:10 (data not shown). The microalgal culture was obtained from an algal harvesting tank maintained in the laboratory. The micro algal culture was centrifuged at 1,000 g for 20 minutes at room temperature,  $22 \pm 2$  °C, (APHA 2005), and maintained in a 1 L Pyrex beaker under red LED illumination and agitated with a magnetic stirrer (Hanna Instruments, UK) prior to the experiments. The AS was obtained from Tudhoe Mill Sewerage Works, Spennymoor, in the north east of England. The sludge was allowed to settle under quiescent conditions. The settled portion of the AS was then mixed with the microalgal culture, and maintained under ambient conditions at  $582.7 \mu\text{mol.m}^{-2}.\text{s}^{-1}$  red LED irradiance and 16:8 light-dark cycles, in one of the STPBR, for 3 days, prior to the experiments. Mixed liquor was collected from the STPBR, fixed according to Eland *et al.* (2012) and analysed using flow cytometry to estimate the aforementioned microalgae-bacteria ratio.

## Illumination

LEDs emitting red light at a 660 nm characteristic wavelength and electrical power consumption of about 0.044 watt (i.e., typical current of 20 mA at a forward voltage of 2.2 V; Maplin Electronics, UK), were used to illuminate the STPBR internally at an average apparent optimum irradiance of  $582.7 \mu\text{mol.m}^{-2}.\text{s}^{-1}$ . This optimum level of irradiance was previously determined by Mohammed *et al.* (2013) using the same STPBR but operated under different light regimes. A total of 189 LEDs were used in the study (nine Vero boards of 21 LEDs vertically arranged at the centre of the STPBR in a water-proof chamber).

## Carbon dioxide

Industrial-grade gas comprising 25% CO<sub>2</sub> and 75% N<sub>2</sub> (BOC, UK) was bubbled into the STPBR via a 0.5 mm-pore gas sparger

(SUPA Aquatic Supplies Ltd, UK) at  $25 \text{ mL.min}^{-1}$  using rotameters (Key Instruments, Trevoze, USA). The premixed gas was supplied concomitant with illumination. CO<sub>2</sub> was added to the STPBR with a view to overcoming any inorganic carbon (IC) limitation (rather than relying on atmospheric CO<sub>2</sub> supply), and this possibly enhanced microalgal activity.

## Analytical tests

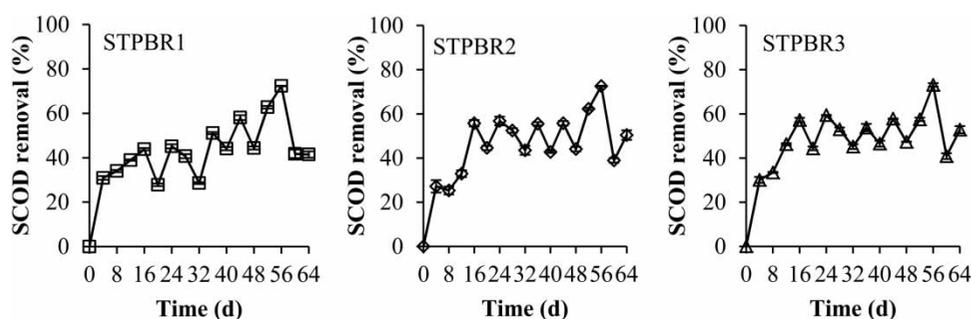
Samples were collected at every HRT cycle (i.e., once every 4 d) from the STPBR, and analysed for soluble chemical oxygen demand (SCOD); IC; other water quality parameters (data not shown); and MLVSS, all according to *Standard Methods* (APHA 2005) to evaluate STPBR performance and WWT efficiency. SCOD was measured using commercial test kits (Merck, Germany) and a spectrophotometer (Spectroquant; Merck, Germany) based on the manufacturer's instructions whereas IC was determined using an automated TOC analyser (Shimadzu, Japan).

All samples for chemical analysis were filtered through 0.2  $\mu\text{m}$  syringe filters (Sartorius, UK). DO and pH were monitored both offline (data not shown) and in real time, at steady-state, using DO and pH probes (Broadley James, UK), respectively. Temperature was monitored using temperature probes (RS Components Ltd, UK). The probes were connected to current-voltage-resistance converters, which were connected to a data logger (both procured from Pico Technology Ltd, UK) and finally to a desktop computer. Furthermore, MLVSS and OD were measured using gravimetry and a UV-1700 spectrophotometer (Shimadzu, Japan), respectively, while the microalgae-bacteria ratio was estimated using a flow cytometer (5 Laser LSRII; BD Biosciences, USA).

## RESULTS AND DISCUSSION

### SCOD removal

Maximum SCOD removal efficiency greater than 70% was achieved in the STPBR (Figure 1).



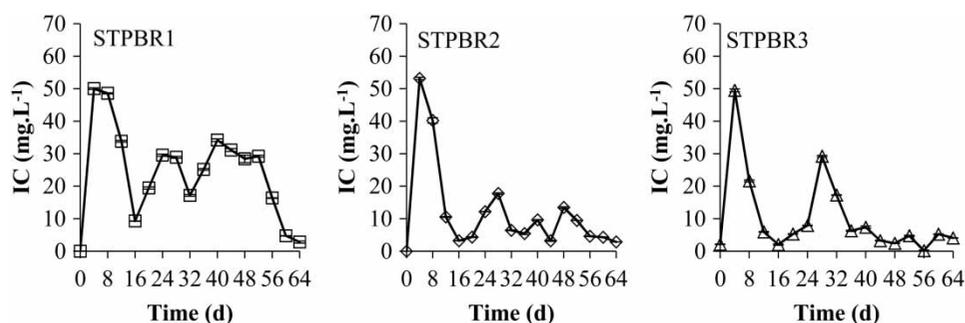
**Figure 1** | Time courses of SCOD removal efficiency in the STPBR operated at same level of red LED irradiance.

The overall SCOD removal efficiencies in the STPBR were not significantly different ( $p = 0.495$ ; one-way analysis of variance, ANOVA). This trend is similar to a previous unpublished laboratory study by the same authors. The maximum SCOD removal efficiencies ranged from 72% in STPBR1 to 73% in both STPBR2 and STPBR3, with average SCOD removal efficiencies ranging from 46% in STPBR1 to 53% in STPBR3 (i.e., averages taken from day 16 to day 64 to minimise the effect of early changes in STPBR performance; Figure 1). However, the overall highest SCOD removal efficiency for the STPBR is at the lower end of the range for conventional AS systems treating municipal wastewater (Tandukar et al. 2007). One possible reason for the relatively low SCOD removal in the current study could be due to acidic conditions in the STPBR, as reflected by pH values lower than 7 (Figure 3(b)). This suggests lower  $\text{CO}_2$  uptake in STPBR1 as reflected by higher IC concentration compared to other STPBR, and low algal activity resulting from possible light attenuation due to higher MLVSS and/or low IC concentration (Figure 2) in STPBR2 and STPBR3 (from day 28 onwards), leading to probable carbon limitation.

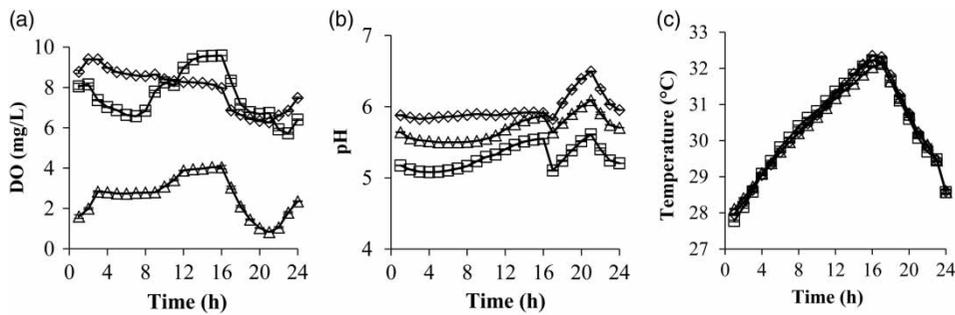
Nevertheless, the resulting DO concentration in the STPBR did not show a lack of oxygenation (Figure 3(a)), and since the headspace was enclosed, with minimal gas exchange between headspace and atmosphere, it was

considered unnecessary to reduce the stirrer speed to eliminate the small vortex around the mixing shaft. Photosynthetic DO produced by algae can achieve locally higher concentrations than oxygen derived from air (21% oxygen), and may be preferentially used in bacterial degradation of organic matter (Shilton & Harrison 2003). Moreover, a slightly acidic pH (Figure 3(b)) might have created conditions unfavourable for microbial growth. In addition, the  $\text{CO}_2$  concentration of the added gas (i.e.,  $25 \text{ mL} \cdot \text{min}^{-1}$ , 25% v/v in  $\text{N}_2$ ) might have inhibited microbial activity. Inhibition of microalgae at  $\text{CO}_2$  concentrations greater than 20% has been reported in the literature (Yun et al. 1996). Nevertheless, the bacterial oxygen requirement was apparently satisfied through photosynthetic oxygenation as reflected by DO concentration higher than  $2 \text{ mg} \cdot \text{L}^{-1}$ , measured in all the STPBR.

Considering the low IC concentration (Figure 2) and the slightly acidic pH values (Figure 3(b)), it may be suggested that the amount of  $\text{CO}_2$  added to the STPBR was too high to control pH within the optimum range for algal growth of pH 7–8.5 (e.g. Park & Craggs 2010). Another possibility could be that the system used in this study performed differently compared to conventional treatment systems, due to the use of red LED light and its manually controlled supply of  $\text{CO}_2$ , unlike, for instance, in the case of Park & Craggs (2010), which involved feedback supply of  $\text{CO}_2$  supply



**Figure 2** | Time courses of IC concentrations in the STPBR operated at same level of red LED irradiance.



**Figure 3** | Real-time data of the average hourly variation of DO, pH and temperature in the STPBR operated at same level of irradiance ( $\square$  STPBR1,  $\diamond$  STPBR2 and  $\triangle$  STPBR3; first 16 h with illumination, and subsequent 8 h in the dark).

based on the microalgal culture pH dynamics. Nevertheless, an in-depth evaluation of the above possibilities regarding the use of  $\text{CO}_2$  addition to control pH as applied to this treatment system (which involved the use of monochromatic LED as the light source) is imperative, and requires further investigation. This may help in better understanding the full potential of this newly introduced hybrid system.

#### Variation of MLVSS with solid retention times (SRT)

Operating the STPBR at controlled MLVSS resulted in different SRT, ranging from a minimum value of 3.9 days in STPBR1 to a maximum value of about 6.2 days in STPBR3. A median SRT of 4.62 was achieved in STPBR2, which was operated at controlled MLVSS of  $300 \text{ mg.L}^{-1}$ . The SRT increased with increasing MLVSS across the STPBR (STPBR3 > STPBR2 > STPBR1). This finding suggests the influence of light attenuation (Heaven *et al.* 2005) in relation to increasing biomass concentration across the STPBR. Interestingly, the microalgae in STPBR1 appeared to grow at a higher rate than in the other two STPBR, possibly due to minimal light attenuation. This observation is supported by the absence of biomass recycling in STPBR1 throughout the experimental period, due to MLVSS production greater than the required  $50 \text{ mg.L}^{-1}$ . This necessitated the use of two clarifiers downstream from STPBR1 (compared to the other STPBR which had only one clarifier each) to aid further settling of the biomass and to facilitate more wastage in order to maintain the MLVSS at the desired level.

#### DO, pH, temperature and algal activity

Real-time data for average hourly variation of DO, pH and temperature for all the STPBR are presented in Figure 3. These parameters were monitored for 24 hours at steady

state. Algal activity appears to be higher in STPBR2 than in the other two STPBR, as reflected by the predominantly highest DO concentrations in this photobioreactor compared to STPBR1 and STPBR3 (Figure 3(a)). The attainment of highest pH values in STPBR2 (Figure 3(b)) supports this observation. This suggests that the biomass concentration in this photobioreactor (i.e.  $300 \text{ mg.L}^{-1}$ ) was the relative optimum for operating the STPBR under the experimental light regime. Therefore, it can be concluded that neither too high nor too low biomass concentration is required to treat municipal wastewater in photobioreactors in order to avoid possible light attenuation. This observation will also apply to pilot- and full-scale hybrid micro algae-activated sludge (HMAS) systems incorporating internal illumination.

It can be seen from Figure 3(b) that all the hourly pH values recorded in the STPBR were below 7, suggesting acidic conditions. The acidic pH values in the STPBR may be connected with the relatively higher  $\text{PO}_4\text{-P}$  concentration observed in the STPBR (data not shown), probably due to the absence of phosphate-accumulating organisms (Saito *et al.* 2004; Oehmen *et al.* 2007). Interestingly, the continuous operation of the STPBR eliminated an apparent light-dependent nitrite accumulation that was observed in a previous study (Mohammed *et al.* 2013) due to the absence of high nitrite concentration in the current study. Despite the apparent luxury uptake (Carberry & Tenney 1973; Powell *et al.* 2009) of P of about 5.4, 6.6, and  $7.3 \text{ mg.PO}_4\text{-P.L}^{-1}$ , corresponding to a removal efficiency of 44, 52 and 53% in STPBR1, STPBR2 and STPBR3, respectively, in the first HRT cycle (i.e., days 1 to 4), this initial removal efficiency was only exceeded (about 55%) in STPBR1, on day 48 (data not shown).

Furthermore, there was a similar pattern of temperature variation across the STPBR (Figure 3(c)). Temperature increased linearly from about  $28^\circ\text{C}$  at the start of

illumination to a maximum of about 32 °C at the end of the light cycle. It then declined to about 29 °C at the end of the dark cycle. Interestingly, the range of temperature encountered in this study may be beneficial when the STPBR are operated outdoors and/or during winter. Possibly, the proximity of the STPBR to anaerobic bioreactors operated at upper mesophilic temperatures in the laboratory might have created a thermal gradient leading to minimal heat loss from or heat gain to the STPBR. Importantly, the thermodynamic effect of temperature on the solubility of pollutants and microbial activities, with its influence on process parameters, such as DO and pH, affects the biochemical reactions taking place during WWT (Paterson & Curtis 2005). As such, the relatively high temperature reported in the current study might have influenced the STPBR performance.

### Energy requirement

Therefore, the efficiency of the current HMAS was evaluated based on the electrical energy required by the STPBR in removing a given amount of SCOD at the operating HRT and photoperiod. The amount of energy required to remove 1 kg of SCOD, based on the maximum treatment efficiency achieved in the STPBR, is given in Table 3. The SCOD removed included that due to the addition of CO<sub>2</sub> into the STPBR, assuming an 80% net uptake of the added CO<sub>2</sub> by microalgal photosynthesis.

Importantly, the energy requirement in Table 3 was based solely on the power required to illuminate the LED in the STPBR operating at 4 d HRT and the corresponding photoperiod of 64 h, for a photoperiod of 16 h. It is well known that artificial aeration, with its associated energy requirements, constitutes about 80% of the overall operational costs of aerobic WWT processes, e.g. the AS system, (Driessen & Vereijken 2003). Since about 1 kWh of electrical power is required to remove 1 kg of COD in conventional aerobic WWT systems (Ahammad et al.

2013), this implies that about 0.8 kWh of energy is associated with the aeration process for the removal of 1 kg of COD.

Interestingly, the STPBR exhibits greater potential for energy savings, as well as reduction in the overall operational cost of municipal WWT, as reflected by its excellent energy efficiency, achieving 95% energy saving compared to the conventional AS system (Table 3).

### CONCLUSION

SCOD removal efficiency of greater than 70% was achieved in the STPBR, with energy savings from the elimination of artificial aeration requirements, since bacterial oxygen requirements were satisfied through DO levels that were provided by photosynthetic oxygenation. In addition, using MLVSS as a control parameter, and clarifiers to facilitate biomass settling, can serve as a potential measure to control light attenuation in hybrid microalgal photobioreactors. MLVSS of 300 mg.L<sup>-1</sup> appeared to be the optimum value for the STPBR operating at the given irradiance. The STPBR achieved up to 95% savings in the energy needed for artificial aeration. This investigation demonstrates the potential of using microalgae to couple carbon capture with municipal WWT in a hybrid system combining the characteristics of both AS and advanced high-rate algal ponds. It also demonstrates the potential for scaling up the process to treat municipal wastewater on a larger scale.

### ACKNOWLEDGEMENTS

The authors are grateful to the Petroleum Technology Development Fund, Abuja, Nigeria for awarding a PhD Scholarship to K. Mohammed; Mr Rob Hunter and Mr David Dick (both technical staff at the School of Civil Engineering and Geosciences, Newcastle University, UK) for connecting the LED and fabricating the bioreactor vessels; and Miss L. E. Eland (a research colleague, at the same School) for conducting flow cytometry and helping with its data analysis.

### REFERENCES

- Ahammad, S. Z., Zealand, A., Dolfing, J., Mota, C., Armstrong, D. V. & Graham, D. W. 2013 Low-energy treatment of colourant wastes using sponge biofilters for personal care product industry. *Bioresource Technology* **129**, 634–638.

**Table 3** | Energy required for SCOD removal in the STPBR

Bioreactor	SCOD removed (kg)	Energy (kWh(kg.SCOD) <sup>-1</sup> )
STPBR1	0.0671	0.56 <sup>a</sup>
STPBR2	0.0672	0.10 <sup>a</sup>
STPBR3	0.0673	0.05 <sup>a</sup>
AS	–	1.00 <sup>b</sup>

<sup>a</sup>Denotes values found in current study.

<sup>b</sup>Obtained from Ahammad et al. (2013).

- APHA 2005 *Standard Methods for the Examination of Water and Wastewater*. 21st edn, APHA, Washington, DC, USA.
- Blankenship, R. E. 2002 *Molecular Mechanisms of Photosynthesis*. Blackwell Science Ltd, UK.
- Bracklow, U., Drews, A., Vocks, M. & Kraume, M. 2007 Comparison of nutrients degradation in small scale membrane bioreactors fed with synthetic/domestic wastewater. *Journal of Hazardous Materials* **144** (3), 620–626.
- Carberry, J. B. & Tenney, M. W. 1973 Luxury uptake of phosphate by activated sludge. *Water Pollution Control Federation* **45** (12), 2444–2462.
- Curtis, T. P., Mara, D. D., Dixo, N. G. H. & Silva, S. A. 1994 Light penetration in waste stabilization ponds. *Water Research* **28** (5), 1031–1038.
- Driessen, W. & Vereijken, T. 2003 *Recent Developments in Biological Treatment of Brewery Effluent*. The Institute and Guild of Brewing Convention, Livingstone, Zambia, 2–7 March.
- Eland, L. E., Davenport, R. & Mota, C. R. 2012 Evaluation of DNA extraction methods for freshwater eukaryotic microalgae. *Water Research* **46** (16), 5355–5364.
- Hall, D. O. & Rao, K. K. 1999 *Photosynthesis*, 6th edn. Cambridge University Press, Cambridge, UK.
- Heaven, S., Banks, C. J. & Zotova, E. A. 2005 Light attenuation parameters for waste stabilisation ponds. *Water Science and Technology* **51** (12), 143–152.
- Ludwig, H. F., Oswald, W. J., Gotaas, H. B. & Lynch, V. 1951 Algae symbiosis in oxidation ponds I: growth characteristics of *Euglena gracilis* cultured in sewage. *Sewage and Industrial Wastes* **23** (11), 1337–1355.
- Matthijs, H. C. P., Balke, H., Hes, U. M. V., Kroon, B. M. A., Mur, L. R. & Binot, R. A. 1996 Application of light-emitting diodes in bioreactors: flashing light effects and energy economy in algal culture (*Chlorella pyrenoidosa*). *Biotechnology and Bioengineering* **50** (1), 98–107.
- Mohammed, K., Ahammad, S. Z., Sallis, P. J. & Mota, C. R. 2013 Optimisation of red light-emitting diodes irradiance for illuminating mixed microalgal culture to treat municipal wastewater. In: *Seventh International Conference on Sustainable Water Resources Management*, 21–23 May 2013, New Forest, UK.
- Negoro, M., Hamasaki, A., Ikuta, Y., Makita, T., Hirayama, K. & Suzuki, S. 1993 Carbon dioxide fixation by microalgae photosynthesis using actual flue gas discharged from a boiler. *Applied Biochemistry and Biotechnology* **39/40** (1), 643–653.
- Oehmen, A., Lemos, P. C., Carvalho, G., Yuan, Z., Keller, J., Blackall, L. L. & Reis, M. A. M. 2007 Advances in enhanced biological phosphorus removal: from micro to macro scale. *Water Research* **41** (11), 2271–2300.
- Oswald, W. J., Gotaas, H. B., Golueke, C. G. & Kellen, W. R. 1957 Algae in waste treatment. *Sewage and Industrial Wastes* **29** (4), 437–457.
- Park, J. B. K. & Craggs, R. J. 2010 Wastewater treatment and algal production in high rate algal ponds with carbon dioxide addition. *Water Science and Technology* **61** (3), 633–639.
- Paterson, C. & Curtis, T. 2005 Physical and chemical environments. In: *Pond Treatment Technology* (A. Shilton, ed.). IWA Publishing, London, pp. 49–65.
- Powell, N., Shilton, A., Chisti, Y. & Pratt, S. 2009 Towards a luxury uptake process via microalgae – Defining the polyphosphate dynamics. *Water Research* **43** (17), 4207–4213.
- Saito, T., Brdjanovic, D. & Loosdrecht, M. C. M. v. 2004 Effect of nitrite on phosphate accumulating organisms. *Water Research* **38** (17), 3760–3768.
- Shilton, A. & Harrison, J. 2003 *Guidelines for the Hydraulic Design of Waste Stabilisation Ponds*. Massey University, Palmerston North, New Zealand.
- Tandukar, M., Ohashi, A. & Harada, H. 2007 Performance comparison of a pilot-scale UASB and DHS system and activated sludge process for the treatment of municipal wastewater. *Water Research* **41** (12), 2697–2705.
- Yun, Y.-S., Park, J. M. & Yang, J.-W. 1996 Enhancement of CO<sub>2</sub> tolerance of *Chlorella vulgaris* by gradual increase of CO<sub>2</sub> concentration. *Biotechnology Techniques* **10** (9), 713–716.

First received 6 September 2013; accepted in revised form 24 February 2014. Available online 10 March 2014