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Nitrogen Removal in a Redox-Stratified Constructed Wetland System

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ABSTRACT

A redox-stratified multi-stage vertical flow constructed wetland system was designed to enhance the removal of nitrogen from synthetic domestic wastewater with high nitrogen concentration. The overall aim was to further our mechanistic understanding of nitrogen removal in such systems and in constructed wetlands in general, and to determine the impact of design and operational variables on the overall removal efficiency. A tidal flow operational strategy was used to enhance aeration and thus promote nitrogen removal in the system. The system was designed and operated as a multi-stage system with a hydraulic loading rate and retention time of 0.76 $m^3/m^2 \cdot d$ and 8 hours, respectively. Results obtained over a 7 month period showed that the multi-stage system was able to efficiently remove the contaminants with average removal efficiencies of 97% for chemical oxygen demand, 97% for ammonium-nitrogen and 75% for total nitrogen; however, only 12% of TP was removed in the system. Results further reveal that the oxidation redox potential and dissolved oxygen are the significant operational variables impacting upon nitrogen removal in the system.

Keywords: Nitrogen; redox-stratified; synthetic domestic wastewater; tidal flow; vertical constructed wetlands

1. INTRODUCTION

Constructed wetlands (CWs) system is considered as a cost effective and environmentally friendly system for wastewater treatment; in particular, for the removal of nitrogen and organic matter from wastewater. Nitrogen and phosphorus are nutrients which in high concentration can lead to eutrophication. Eutrophication is an excessive richness of nutrients in a lake or other body of water, frequently due to run-off from the land, which causes a dense growth of plant life on the water course that consumes oxygen and enhances the algae growing and therefore depletes the oxygen in the water and affects aquatic life.

Numerous research studies have been conducted on the effectiveness of CWs in removing nitrogen from wastewater; and it is generally acknowledged that CWs are poor in removing nitrogen from ammonium-rich wastewater due to the limiting nitrification process in the saturated sediment (De Boer et al., 1990; Hunt and Poach, 2001; Vymazal and Kröpfelová, 2015). In recent years, vertical flow constructed wetlands (VFCWs) have been developed to achieve nitrification and remove organic matter and suspended solids. From the tidal flow operational strategy, tidal

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flow means fill (saturated), drain (unsaturated) and rest. Thus, the saturated time refers to the period of loading with the wastewater into the CW; the unsaturated period refers to unsaturated time (draining the load out of CW). Tidal flow allows more oxygen to enter the wetland media and promotes nitrification. Table 1 shows a review of different studies on tidal flow and their main findings.

Table 1 A summary review of different studies on tidal flow strategy on CW

References	Main findings
Wu et al. (2015)	Limited buffering capacity of tidal-operated CWs to influent ammonium pulse loadings because of the short contact time. High buffering capacity of tidal-operated CWs to suddenly changed organic matter loadings.
Li et al. (2015b)	Quantitative responses suggested that, under the FT constraints, the anammox 16S rRNA was the key factor of the NH_4^+–N transformation rates, whereas, (*napA*+*narG*) was the key factor regulating the NO_3^-–N transformation rate.
Li et al. (2015a)	Higher carbon supply (C:N) to enhance TN removal as well as the removal of phosphorus was strongly influenced by organic loadings.
Liu et al. (2014)	The zeolite-based tidal flow CW (TFCW) outperformed the quartz sand, ceramsite, and volcanic-based TFCWs in removing NH_4^+-N and TN under the same operational conditions.
Zhi and Ji (2014)	Molecular biological analyses confirmed the existence of simultaneous nitrification, anammox, and denitrification (SNAD) processes in the system, which could potentially help increase nitrogen removal under different C/N ratios, and the role of microbes in nitrogen transformation processes.
Ju et al. (2014a)	A novel electrolysis-integrated TFCW was developed to investigate how it intensifies nutrient removal. Current intensity plays a very important role in the dynamics of nitrogen transformation.
Ju et al. (2014b)	The current intensity of electrolysis plays an important role in nitrogen transformations. Phosphorus removal was significantly enhanced, in the electrolysis-integrated CW system.
Hu et al. (2014a)	High inorganic carbon (IC) could promote both partial nitrification and anammox activities but had no substantial effect on nitrite oxidization, which provided a strong selective method to maintain the Canon pathway.
Hu et al. (2014b)	The contact/rest scheme should be optimized according to the adsorption/nitrification kinetics. The retained OM and biomass were major contributors to the overall NH_4^+-N adsorption capacity. SND could be intensive during the bed resting periods.
Cui et al. (2012)	The recirculation hybrid tidal flow CW has better performance in removal of nutrients than that of the traditional vertical flow CW.
This study	With a shorter saturated time and longer unsaturated time in the tidal flow, the CWs systems were more efficient in the removal of various pollutants due to enhanced oxygen supply into the CW.

However, the present study reports on the performance of VFCW that combines four stages as shown in Fig. 1.This is based on the process design, whereby the first stage is aimed at organic matter removal and initiation of the nitrification step, the second stage and third stage (depending on the load) for nitrification and denitrification and the fourth stage for denitrification. The aim of this study is to examine the engineering aspects (i.e. hydraulic loading rate, hydraulic retention time and pollutant loading), and determine the pathways and mechanisms involved in nitrogen removal in a redox-stratified engineered wetland system. Findings from the study can further our understanding of nitrogen removal and thus promote the efficient removal of nitrogen.

2. MATERIALS AND METHODS

Fig. 1 shows the redox-stratified multi-stage constructed wetland system. The system consists of four identical stages made from PVC with each stage being 100 cm in height and 10 cm in diameter with a total treatment surface area of 0.032 m² (0.008 m² each). A VFCW was designed and constructed to treat synthetic domestic wastewater with high nitrogen content. The experiment was carried out, installed and constructed outdoors. It was therefore exposed to the natural environmental conditions (temperature range between 5 to 21˚C), the weather being almost high humidity (average annual 73-88%) and rainy (average annual rainfall 63-116 mm) (Weather, 2016); the system was exposed also to the sun rays, but due to the cloudy weather there was little exposure. Gravel was used as the main substrate in all the four stages: in each wetland, a 10 cm bottom layer of coarse gravel (20-25 mm) was filled and served as the supporting and drainage layer; the following layer was filled with gravel (5-9 mm) as the main substrate layer with a depth of 50 cm; a 10 cm top layer of gravel (11-19 mm) was added for facilitating the dispersion and the distribution of wastewater and the growth of plants. Each column was planted with *Phragmites australis*, because they are the mostly common used reference plant for CWs in Europe and able to survive in most conditions. Moreover, they have a comparatively high oxygen transfer ability from their leaves through stems and rhizomes, which facilitates aerobic degradation of pollutants (Barbera et al., 2009; Wang et al., 2012).

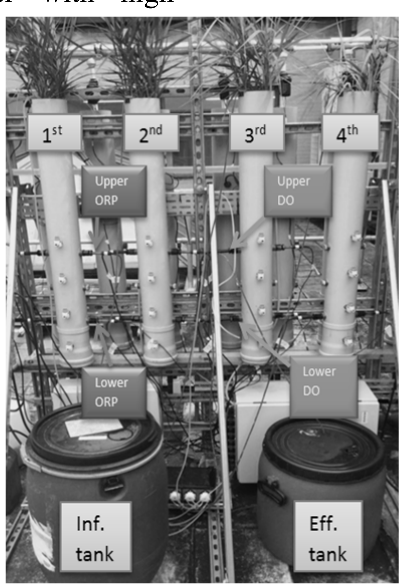

Figure 1 Redox-stratified multi-stage constructed wetland system

The system was fed with synthetic wastewater simulating domestic wastewater. The characteristics of the synthetic wastewater are: 0.95 g/L (CH_3COONa), 0.025 g/L (KH_2PO_4), 0.025 g/L (K_2HPO_4), 0.74 g/L (KCl), 0.58 g/L (NaCl), 0.2181 g/L (NH_4Cl), 0.1 g/L ($MgSO_4\cdot7H_2O$) and 0.1 g/L ($CaCl_2\cdot2H_2O$) with 1 ml/L of the trace elements mixture (containing 0.5 L) consisting of: 0.5 g/L ($FeSO_4\cdot7H_2O$), 0.035 g/L ($ZnCl_2$), 0.05 g/L ($MnCl_2\cdot4H_2O$), 0.003 g/L (H_3BO_3), 0.065 g/L ($CaCl_2\cdot6H_2O$), 0.001 g/L ($CuCl_2\cdot2H_2O$), 0.012 g/L ($NiCl_2\cdot6H_2O$), 0.018 g/L ($Na_2MoO_4\cdot2H_2O$) and 0.119 g/L ($CoCl_2\cdot6H_2O$). The synthetic wastewater simulates typical domestic wastewater with a high concentration of nitrogen and organic carbon source to obtain approximately 700 mg/L of chemical oxygen demand (COD) and 60 mg/L of ammonium-nitrogen (NH_4^+-N). The system was operated in batches with three batch cycles applied per day. Each cycle entails 2 hours saturation and 6 hours unsaturation giving a total of 8 hours per cycle with a hydraulic loading rate (HLR) of 0.76 $m^3/m^2\cdot d$. Samples were collected from the influent and effluent in each stage.

Samples were analysed directly *in situ* for pH and temperature by a pH/EC/TDS meter (HANNA HI 991301), whereas each column was instrumented with a MCC-SYSti-18f multiparameter monitoring and control module that provides continuous real time monitoring and measurement of the redox potential (referred to as oxidation reduction potential, ORP) and dissolved oxygen (DO). The module was installed at 20 and 50 cm depths from the surface of the distribution layer of the first, second and third stages, and at 50 cm depth in the fourth stage (see Fig. 1). The reason behind selecting the upper point is because it is close to the surface where you would expect nitrifying bacteria to be predominant; whereas the lower point is deeper where anoxic or anaerobic conditions can take place. The MCC-SYSti module was connected with a computer software system where the data could be easily collected, downloadedand analysed to demonstrate, understand and optimise the performance of the CW system. Chemical oxygen demand (COD), nitrite (NO_2^--N), nitrate (NO_3^--N), ammonium (NH_4^+-N), total nitrogen (TN), orthophosphate (PO_4^{3-}-P) were analysed by using a Hach DR/3900 spectrophotometer according to its standard operating procedures and total phosphorous (TP) was measured by an ICP machine in the laboratory.

3. RESULTS AND DISCUSSION

3.1 Redox potential (ORP) and dissolved oxygen (DO)

The measurement of redox potential is usually used to describe oxidation-reduction conditions of wetland soils (Likens, 2009). In this study, ORP was monitored in the CW at two depths: 20 cm and 50 cm beneath the surface as per data shown in Fig. 2A and 2B. It is well-known that as you go deeper in the CW matrices, ORP decreases. However, after the start-up period (May and June), it was noticed that the first column (upper point) began with negative values due to intense microbial activity, and the rest of the columns were in positive values and below 200 mV which means anoxic condition took place and so did the activity of denitrifying bacteria. Regarding the lower point, the first column remained in negative values due to microbial activity and the depth played a role as well. Regarding DO, this was also monitored at two depths: 20 cm and 50 cm beneath the surface as per data shown in Fig. 3A and 3B. DO decrease with depth as well due to low oxygen availability. It was noticed that in the upper and lower points after the start-up period in the first column, DO varied between 0-1 mg/L and had the lowest reading compared to the other columns due to the bacteria consuming the oxygen in the wastewater; therefore ORP was in negative values. Also, it was noticed that DO readings in the lower points were lower than the upper points in all columns.

In general, positive ORP values indicate the existence of oxygen in the system (Rowell, 1981). Such a situation may be attributed to the plants' ability to oxygenate reed beds by their roots and from the tidal flow process as well. Negative ORP values specify that the oxygen is used by microbes for mineralisation of root exudates (Altor and Mitsch, 2006; Kankaala et al., 2003). Also, ORP could be affected by many factors such as temperature which has a straight effect on the solubility of oxygen in the water and consequently decreases ORP value. However, temperature also influences ORP indirectly by accelerating the plant growth and the microbial activity processes (Kadlec and Reddy, 2001). In addition, an increase in temperature will accelerate biochemical processes including bacterial activity; as a result, oxygen solubility in water decreases and so does ORP. Moreover, the capability of plants to transport oxygen and the release of plant-root exudates into the rhizosphere served as a substrate for heterotrophic microorganisms, causing an increase in microbial activity and consumption of the oxygen, therefore decreasing the ORP value. The depth could also affect the ORP reading which decreased with increasing depth. The different wastewater concentrations that the system has received, e.g. pH and DO, could affect ORP readings, therefore the results varied from one month to another. In general the redox system is driven by the physico-chemical and biological factors. One of the unique advantages of the tidal flow reed bed system is the improvement of oxygen supply (Chang et al., 2014; Sun et al., 2005; Sun et al., 2006), which has been successfully applied during the operational period as shown in Fig. 4 (selected random day combined 3 cycles). The nitrification process requires aeration to take place to remove nitrogen from wastewater. The relationship between ORP and DO and between DO and nitrogen logically indicate a relationship between ORP and nitrogen; nitrogen decreases with the decrease of ORP and DO, because the denitrification process requires anoxic or anaerobic conditions (limited oxygen and also presence of organic carbon to enhance the denitrification process).

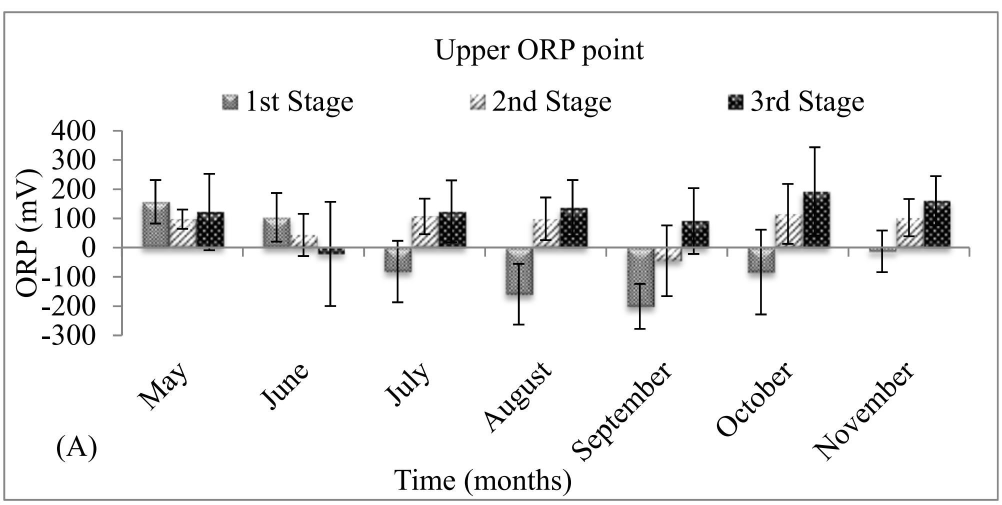

Figure 2A (to be continued)

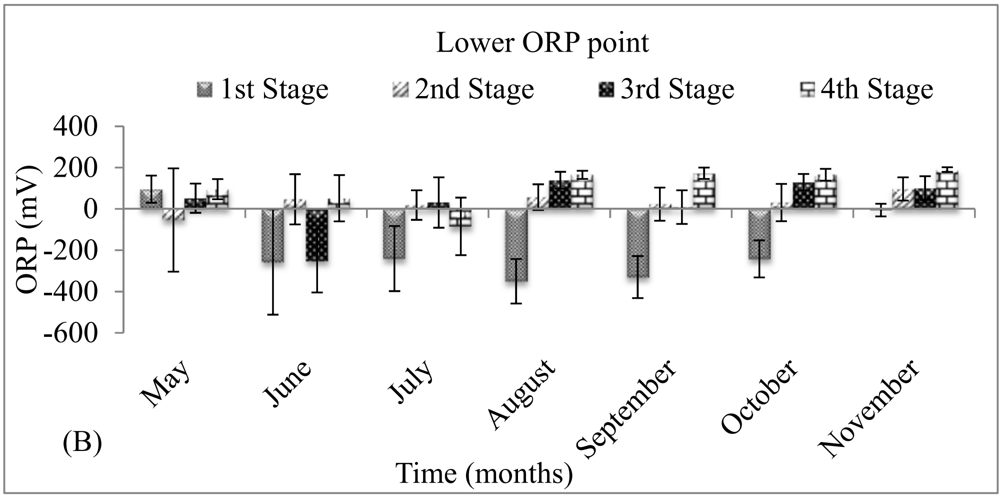

Figure 2 Average oxidation reduction potential (ORP) values at (A) upper point (20 cm depth) and (B) lower point (50 cm depth)

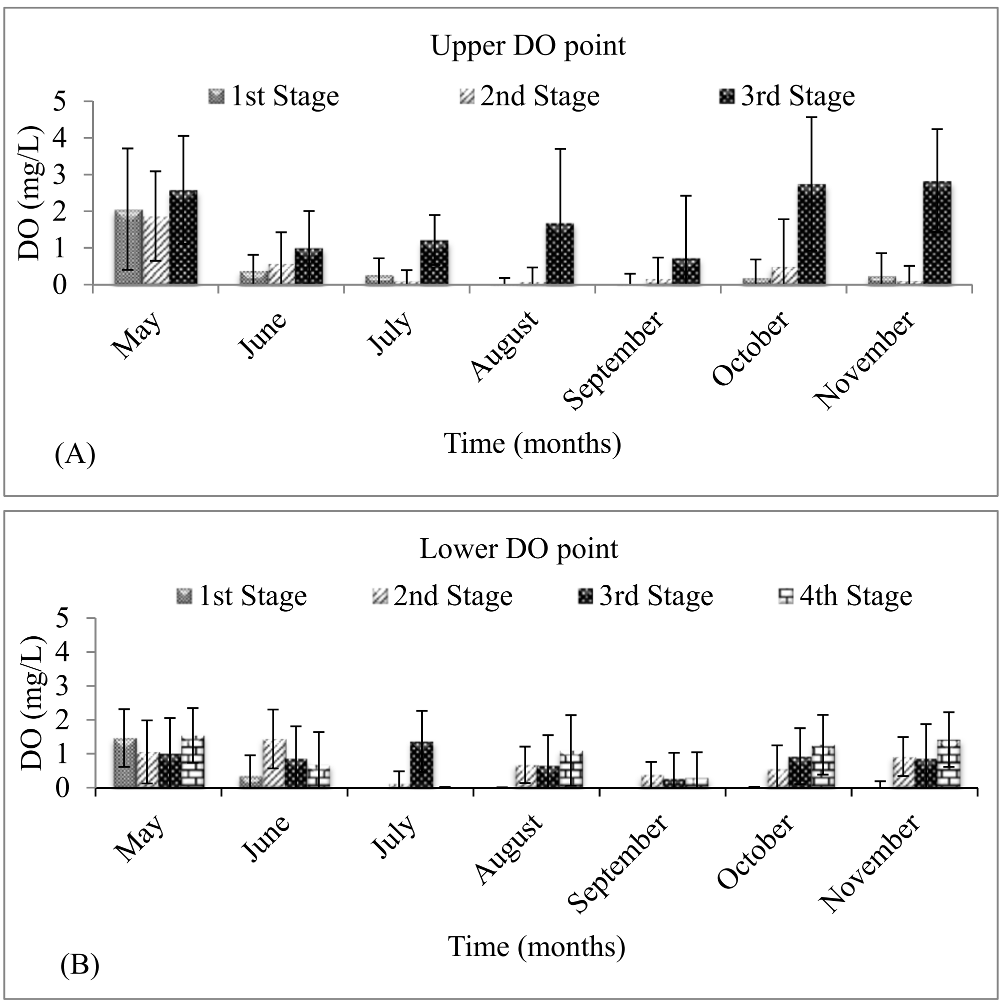

Figure 3 Average Dissolved Oxygen (DO) values at (A) upper point (20 cm depth) and (B) lower point (50 cm depth)

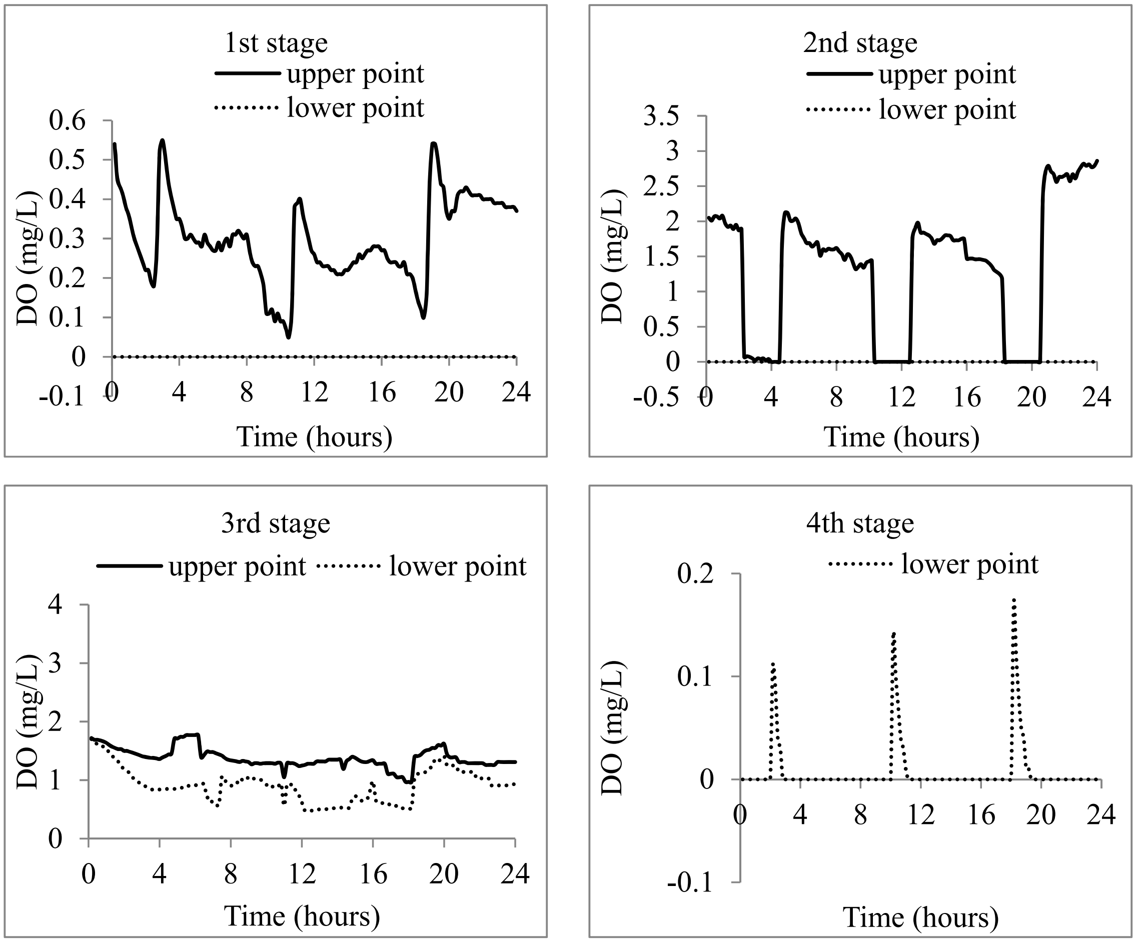

Figure 4 Oxygen diffusion into the system during the filling and draining period

Overall, the results highlight enhanced ammonium removal performance of the redox-stratified constructed wetland with > 96% due to the oxygen that entered into the matrices bed at a high rate (see Table 2). As a result, nitrification took place; because the nitrification process is an aerobic process which prefers aeration (2 hours saturated and 6 hours unsaturated). In the first stage, it is clear that most of the ORP values were negative, which means that the microorganisms were active in the contaminant and nutrient removal in low DO concentrations. In the second stage, most ORP values were below 100 mV with low DO concentration which was suitable for the denitrification process (anoxic). In the last two stages (third and fourth), ORP was below 200 mV with low DO concentration; as a result, partial nitrification took place. More details will be discussed in the nitrogen removal section.

3.2 Treatment performance

3.2.1 COD removal

The overall treatment efficiency for COD during the study period was 97% with the average effluent concentration from the final stage of 21 mg/L as presented in Table 2. COD removal during the whole operation period is illustrated in Fig. 5. Concentrations of COD decreased in all of the four stages, with the highest removal in the first stage just after the startup period (63 days) as shown in Fig. 6. The significant COD removal obtained during the experiment is predominantly due to the enhanced oxygenation ability of the tidal flow (Chang et al., 2014; Hu et al., 2012; Zhao et al., 2004) and the intensive microbial activities (Dušek et al., 2008). The time of filling and draining of the CWs ensured that the oxygen present in the beds was frequently replenished (see Fig. 4).

Table 2 Overall treatment performance of multistage vertical constructed wetlands excluding the startup period

Parameter	Inflow	1st Stage	2nd Stage	3rd Stage	4th Stage	Overall treatment efficiency (%)
pH	7.50 (±0.04)	7.47 (±0.13)	7.21 (±0.05)	7.34 (±0.07)	7.62 (±0.15)	-
NH_4^+-N (mg/L)	57.77 (±0.49)	40.21 (±5.47)	17.46 (±6.57)	5.96 (±3.64)	1.88 (±1.71)	97
NO_2^--N (mg/L)	0.021 (±0.017)	0.003 (±0.002)	0.015 (±0.01)	0.351 (±0.1)	0.471 (±0.089)	-
NO_3^--N (mg/L)	0 (±0)	3.99 (±2.11)	0.98 (±0.76)	3.49 (±1.22)	3.98 (±1.25)	-
TN (mg/L)	61.64 (±0.37)	NM*	NM*	NM*	15.34 (±2.98)	75
COD (mg/L)	699.28 (±5.37)	296.98 (±154.22)	67.82 (±21.69)	40.68 (±15.23)	21.46 (±11.59)	97
TP (mg/L)	10.30 (±0.59)	9.34 (±1.21)	7.29 (±2.35)	8.18 (±2.69)	9.10 (±1.99)	12
PO_4^{3-}-P (mg/L)	9.32 (±0.9)	8.53 (±1.22)	6.52 (±2.12)	7.32 (±2.6)	8.16 (±2)	13

NM*: Not Measured

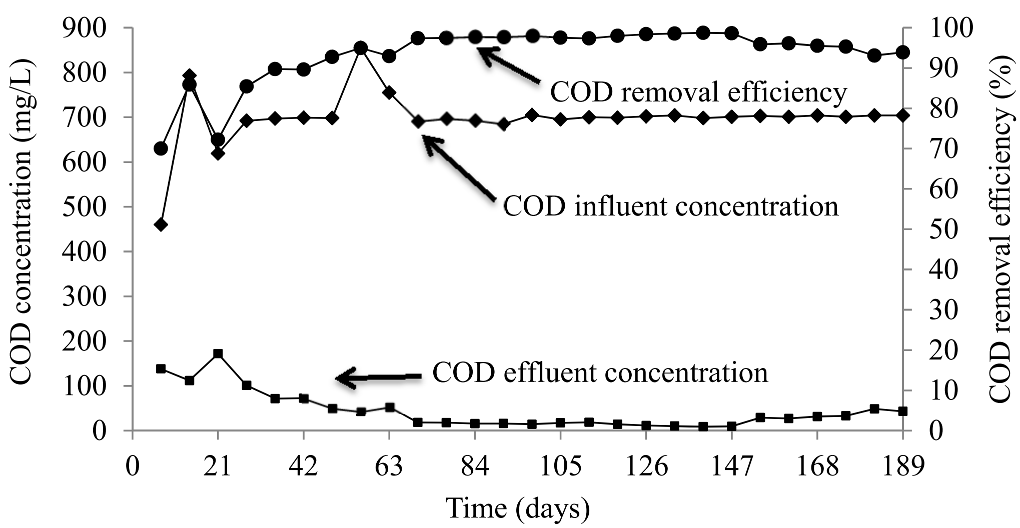

Figure 5 COD removal during the operation period

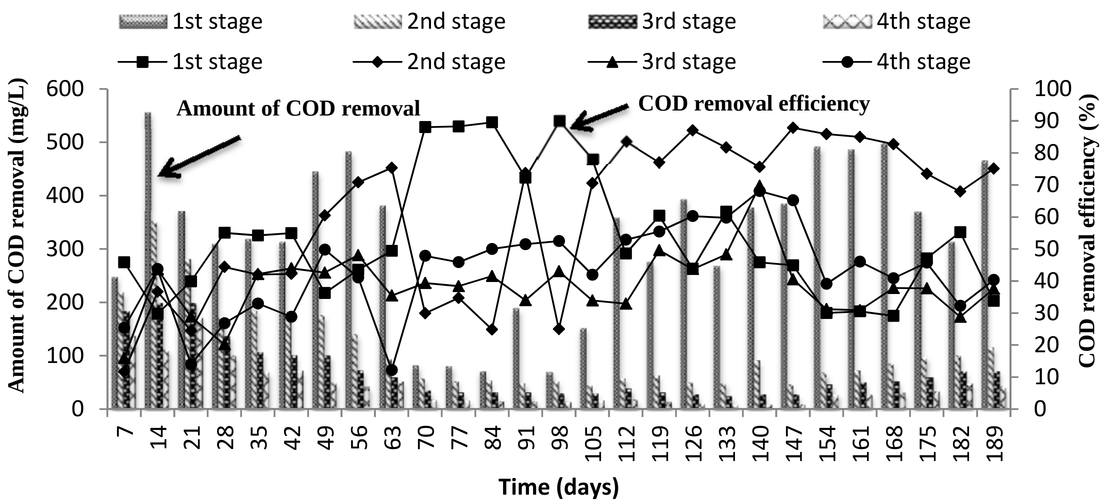

Figure 6 Contribution of individual stage to overall removal of COD during the operation period

3.2.2 Phosphorus removal

As the system was not designed for phosphorus removal, total phosphorus (TP) and ortho-phosphate (PO_4^{3-}-P) removal efficiencies were observed at 12% and 13%, respectively. Fig. 7 shows the removal of both TP and PO_4^{3-}-P through the stages; from the beginning of the experiment phosphorus increased during the startup through all the stages, because the biological activities in the CWs may have limited impact and influence on the phosphorus removal from the system. Also, because the microorganisms release phosphorus when the anaerobic condition takes place (Likens, 2009). After 63 days TP began to decrease slightly.

3.2.3 Nitrogen removal

The NH_4^+-N and TN of the synthetic wastewater were reduced by 97% and 75% respectively through four stages of the system. Most of the oxygen was consumed by nitrifying bacteria which is a slow process compared to COD reduction reaction. Consequently, under high organic loading, most of the available oxygen was used to reduce the COD of the influent. Fig. 8 shows NH_4^+-N removal efficiency during the whole operation period, whereas Fig. 9 illustrates the contribution of the individual stage to overall removal of NH_4^+-N during the whole operation period and it shows continuous reduction of NH_4^+-N through each stage due to microbial activities. In CWs the reduction of NH_4^+-N usually results in increase in NO_2^--N and NO_3^--N levels as NH_4^+-N can be bio-oxidised by nitrifying bacteria to nitrite (NO_2^--N) and further to nitrate (NO_3^--N). The removal of NH_4^+-N in the current study may result from a combination of numerous processes, which include nitrification, adsorption, assimilation associated with decomposition of organics, volatilisation and plant uptake. From Fig. 10, the concentration levels of NO_2^--N and NO_3^--N in the effluents were slightly higher than that in the influents in some cycles, which indicated that the nitrification process was occurring during the experiment due to the aerobic condition (see section 3.2.3.1. mass balance). Note that nitrification and denitrification processes may have occurred simultaneously in the system and this resulted in a decrease of NO_2^--N and NO_3^--N level in the effluent which contributed to the NH_4^+-N reduction. TN removal through the study period is illustrated in Fig. 11.

3.2.3.1 Mass balance

In order to explore the characteristics of nitrogen removal in individual stage under different operational conditions, the contributions of each stage to overall nitrogen removal were estimated by mass balance. The net mass flux of NH_4^+-N reduction (φNH_4, Re.) and NOx^--N (NO_2^--N + NO_3^--N) accumulation (φNOx, Acc.) in each stage was calculated (Table 3). Here, the calculation was based on the simplifying assumption that NH_4^+-N reduction was due to nitrification, thus the nitrogen loss by biomass assimilation and plant uptake was negligible (Hu, 2012). The negative value for φNH_4, Re. implies that the amount of NH_4^+-N produced via ammonification exceeds the amount of NH_4^+-N reduced due to nitrification. Accordingly, the negative value for φNOx, Acc. suggests that the amount of NOx^--N removed via denitrification is above the amount of NOx^--N produced through nitrification.

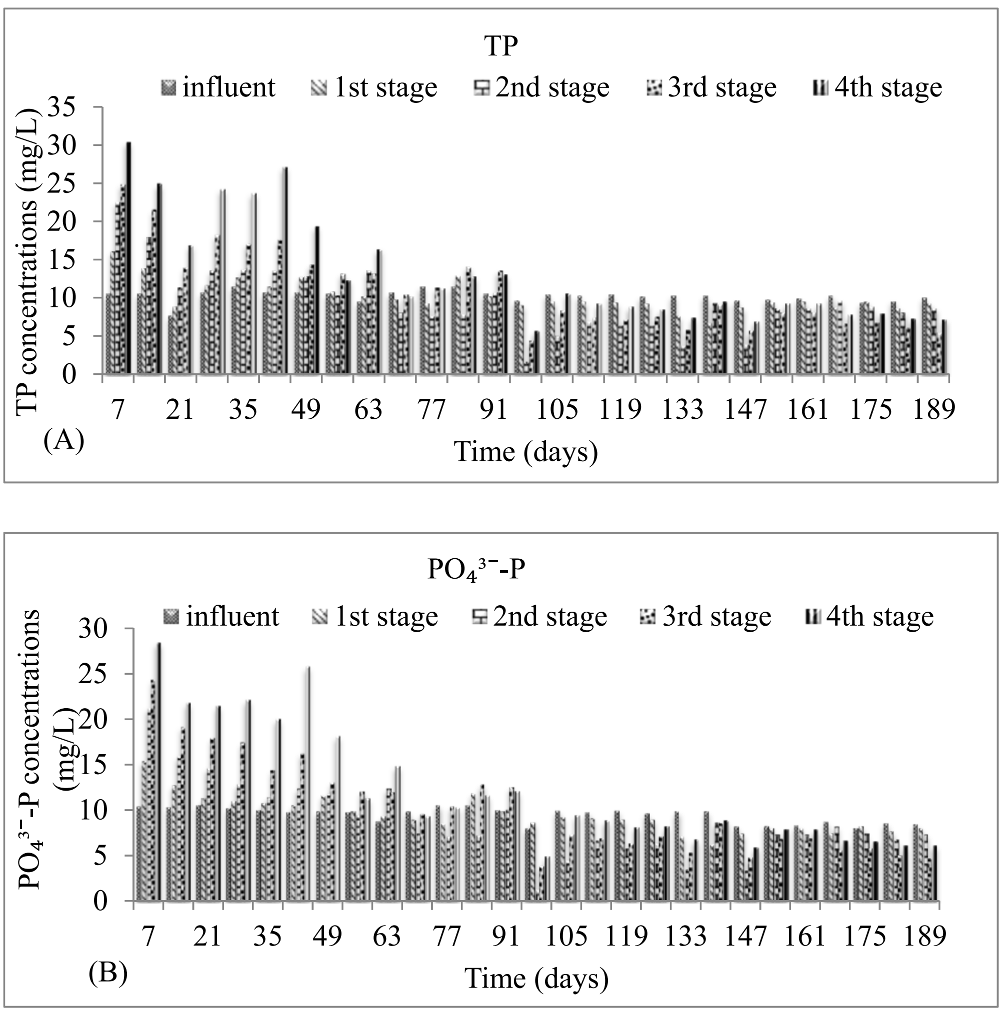

Figure 7 (A) TP and (B) PO_4^{3-}-P removal through multistage VFCW

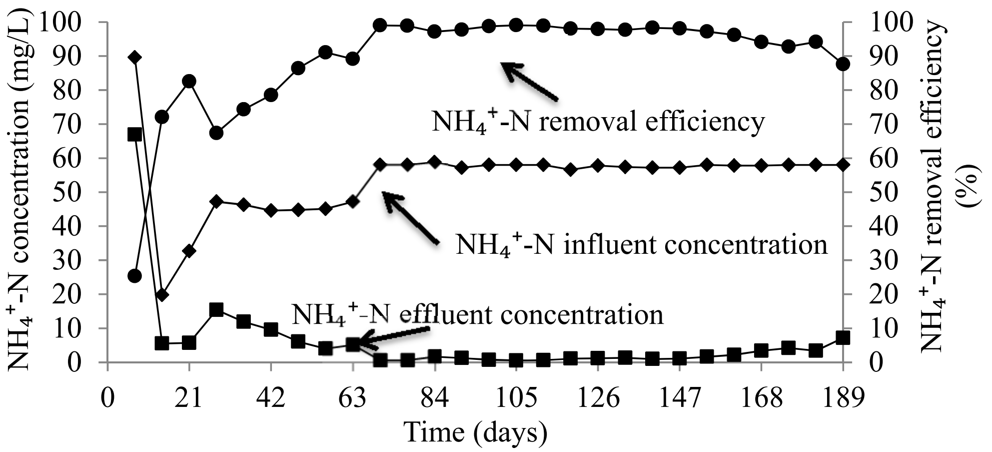

Figure 8 NH_4^+-N removals during the operation period

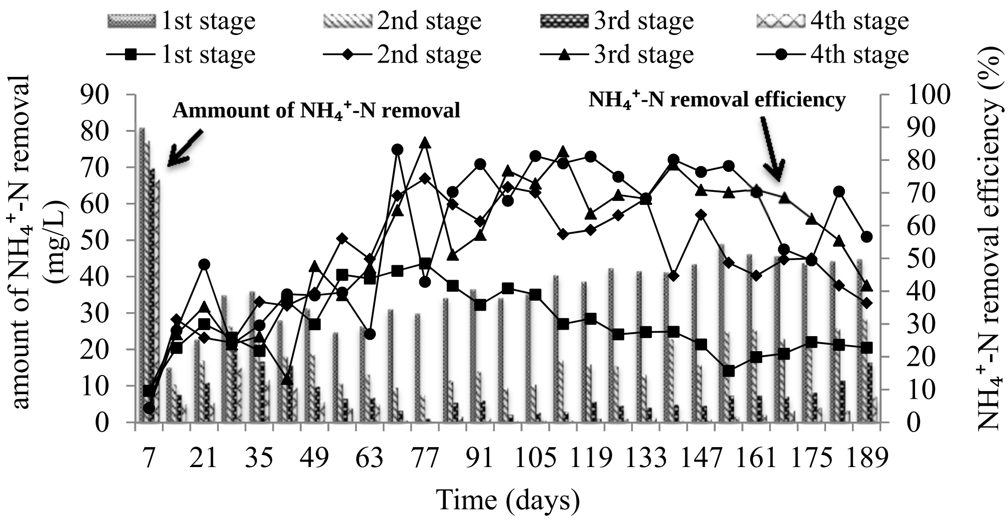

Figure 9 Contribution of individual stage to overall removal of NH_4^+-N during the operation period

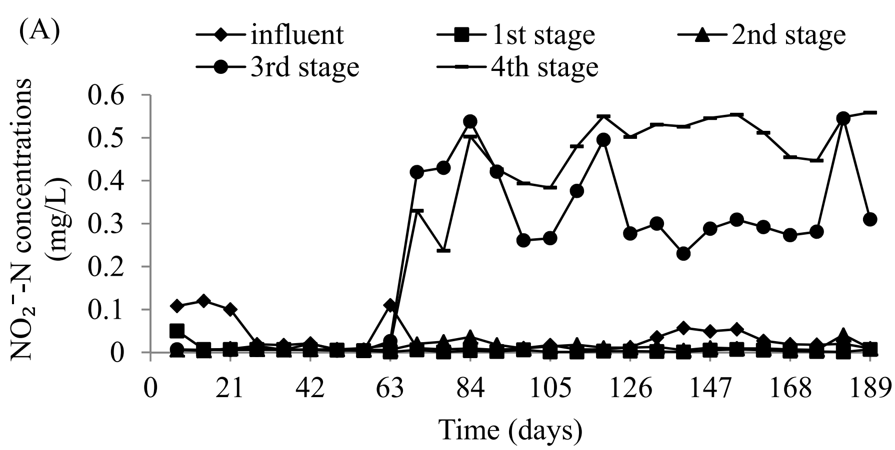

Figure 10A (to be continued)

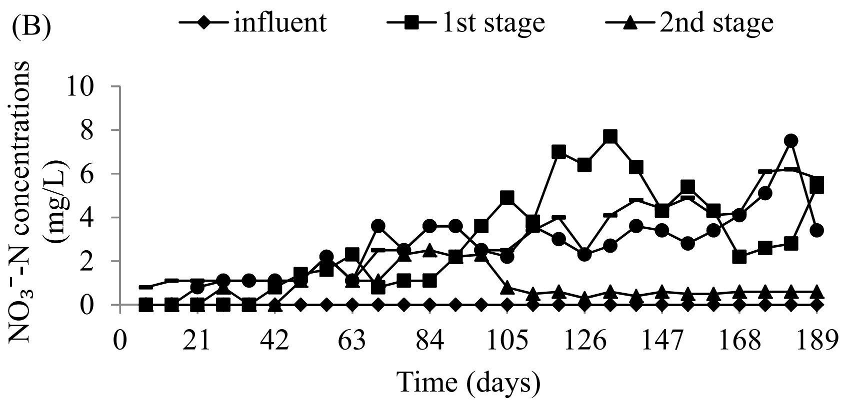

Figure 10 (A) NO_2^--N and (B) NO_3^--N conversion through the multistage VFCW system

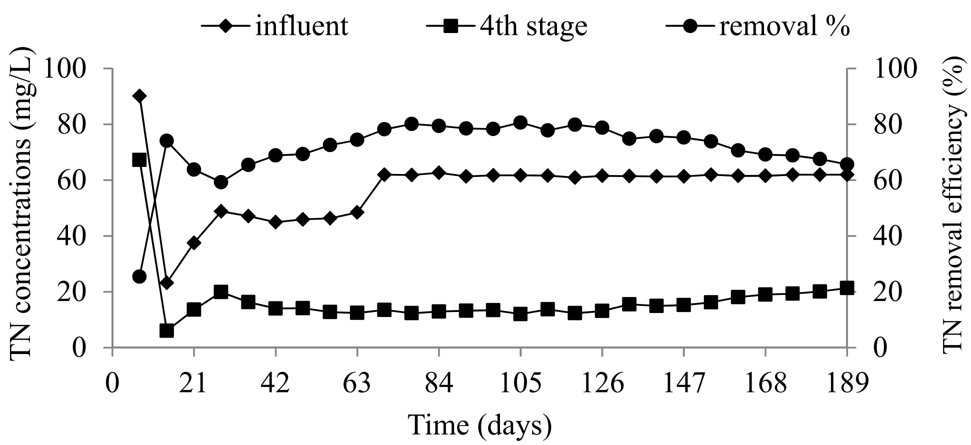

Figure 11 TN removals during the operation period

Table 3 Mass flux of NH_4^+-N reduction (φNH4, Re.) and NOx^--N accumulation (φNOx-N, Acc.) in each stage (mg/d)

Stages	1	2	3	4
φNH_4^+-N, Re.	105	137	69	25
φNOx-N, Acc.	24	-18	17	4

Q: flow rate, l/d; Q = 6, NH_4, in, NOx, in: influent NH_4^+-N and NOx^--N, mg/L, NH_4, Si, NOx, Si: effluent NH_4^+-N and NOx^--N of each stage, mg/L; i = 1-4

From Table 3, it can be concluded that the net mass flux of NH_4^+-N reduction for first, second, third and fourth stages equal 105, 137, 69 and 25 mg/d, respectively. NOx^--N accumulation for first, second, third and fourth stages equal 24, -18, 17 and 4 mg/d, respectively.

From Tables 2 and 3, it can be concluded that in the first stage the nitrification process took place under aerobic process; as a result, NH_4^+-N converted to a small amount of NO_2^--N and increased the amount of NO_3^--N in the effluent. The effluent from the second stage, NO_2^--N increased slightly; whereas NO_3^--N sharply decreased, which indicates that nitrification and denitrification simultaneously happened (the amount of NOx^--N removed via denitrification is above the amount of NOx^--N produced through the nitrification process, -18 mg/d). In both the third and fourth stages, the NO_2^--N and NO_3^--N levels of the effluent was significantly increased indicating the nitrification process didn't fully take place due to the increase of nitrite and nitrate in the effluent of each stage and lack of denitrification process due to the low amount of available organic carbon source (nitrate increased).

CONCLUSIONS

The vertical flow constructed wetland that applied tidal flow with HRT of 8 hours proved that the system was able to deal with high concentration levels and efficiently remove organics (expressed as COD) and nitrogen. The overall removal efficiencies for COD, NH_4^+-N and TN were 97%, 97% and 75%, respectively. The improvement of removal efficiency was attributed to the aeration of the CW during the prolonged unsaturated time that enhanced the oxygen transfer. With improved and enhanced oxygen transfer ability, the tidal flow technique can be used to reduce the high strength level of pollutants in wastewaters. It was clearly demonstrated that with a shorter saturated time and longer unsaturated time in the tidal flow the CWs systems were more efficient in the removal of various pollutants due to enhanced oxygen supply into the CW.

REFERENCES

Altor, A.E. and Mitsch, W.J. (2006). Methane flux from created riparian marshes: relationship to intermittent versus continuous inundation and emergent macrophytes. *Ecological Engineering*, 28(3), 224-234.

Barbera, A.C., Cirelli, G.L., Cavallaro, V., Di Silvestro, I., Pacifici, P., Castiglione, V., Toscano, A. and Milani, M. (2009). Growth and biomass production of different plant species in two different constructed wetland systems in Sicily. *Desalination*, 246(1-3), 129-136.

Chang, Y., Wu, S., Zhang, T., Mazur, R., Pang, C. and Dong, R. (2014). Dynamics of nitrogen transformation depending on different operational strategies in laboratory-scale tidal flow constructed wetlands. *Science of the Total Environ*, 487(14), 49-56.

Cui, L., Feng, J., Ouyang, Y. and Deng, P. (2012). Removal of nutrients from septic effluent with re-circulated hybrid tidal flow constructed wetland. *Ecological Engineering*, 46(9), 112-115.

De Boer, W., Gunnewiek, P.K. and Troelstra, S. (1990). Nitrification in Dutch heathland soils. *Plant and Soil*, 127(2), 193-200.

Dušek, J., Picek, T. and Čížková, H. (2008). Redox potential dynamics in a horizontal subsurface flow constructed wetland for wastewater treatment: Diel, seasonal and spatial fluctuations. *Ecological Engineering*, 34(3), 223-232.

Hu, Y. (2012). *High rate nutrients removal from wastewater with alum sludge-based constructed wetlands system*. Ph.D. Dissertation, University College Dublin, Dublin, Ireland.

Hu, Y., Zhao, X. and Zhao, Y. (2014a). Achieving high-rate autotrophic nitrogen removal via Canon process in a modified single bed tidal flow constructed wetland. *Chemical Engineering Journal*, 237(5), 329-335.

Hu, Y., Zhao, Y. and Rymszewicz, A. (2014b). Robust biological nitrogen removal by creating multiple tides in a single bed tidal flow constructed wetland. *Science of the Total Environment*, 470-471(2), 1197-1204.

Hu, Y.S., Zhao, Y.Q., Zhao, X.H. and Kumar, J.L. (2012). Comprehensive analysis of step-feeding strategy to enhance biological nitrogen removal in alum sludge-based tidal flow constructed wetlands. *Bioresource Technology*, 111(5), 27-35.

Hunt, P. and Poach, M. (2001). State of the art for animal wastewater treatment in constructed wetlands. *Water Science and Technology*, 44(11-12), 19-25.

Ju, X., Wu, S., Huang, X., Zhang, Y. and Dong, R. (2014a). How the novel integration of electrolysis in tidal flow constructed wetlands intensifies nutrient removal and odor control. *Bioresource technology*, 169(5), 605-613.

Ju, X., Wu, S., Zhang, Y. and Dong, R. (2014b). Intensified nitrogen and phosphorus removal in a novel electrolysis-integrated tidal flow constructed wetland system. *Water Research*, 59(4), 37-45.

Kadlec, R.H. and Reddy, K. (2001). Temperature effects in treatment wetlands. *Water environment research*, 73(5), 543-557.

Kankaala, P., Mäkelä, S., Bergström, I., Huitu, E., Käki, T., Ojala, A., Rantakari, M., Kortelainen, P. and Arvola, L. (2003). Midsummer spatial variation in methane efflux from stands of littoral vegetation in a boreal meso-eutrophic lake. *Freshwater Biology*, 48(9), 1617-1629.

Li, C., Wu, S. and Dong, R. (2015a). Dynamics of organic matter, nitrogen and phosphorus removal and their interactions in a tidal operated constructed wetland. *Journal of Environmental Management*, 151, 310-316.

Li, L., He, C., Ji, G., Zhi, W. and Sheng, L. (2015b). Nitrogen removal pathways in a tidal flow constructed wetland under flooded time constraints. *Ecological Engineering*, 81, 266-271.

Likens, G.E. (2009). *Encyclopedia of inland waters*. Academic Press, Salt Lake City, USA.

Liu, M., Wu, S., Chen, L. and Dong, R. (2014). How substrate influences nitrogen transformations in tidal flow constructed wetlands treating high ammonium wastewater? *Ecological Engineering*, 73, 478-486.

Rowell, D. (1981). Oxidation and reduction. *The chemistry of soil processes*, 401-461.

Sun, G., Zhao, Y. and Allen, S. (2005). Enhanced removal of organic matter and ammoniacal-nitrogen in a column experiment of tidal flow constructed wetland system. *Journal of Biotechnology*, 115(2), 189-197.

Sun, G., Zhao, Y., Allen, S. and Cooper, D. (2006). Generating "tide" in pilot-scale constructed wetlands to enhance agricultural wastewater treatment. *Engineering in Life Sciences*, 6(6), 560-565.

Vymazal, J. and Kröpfelová, L. (2015). Multistage hybrid constructed wetland for enhanced removal of nitrogen. *Ecological Engineering*, 84, 202-208.

Wang, R., Baldy, V., Périssol, C. and Korboulewsky, N. (2012). Influence of plants on microbial activity in a vertical-downflow wetland system treating waste activated sludge with high organic matter concentrations. *Journal of Environmental Management*, 95(2), 158-164.

Weather, B. (2016). *Weather Cardiff*. Available at http://www.bbc.co.uk/weather/2653822.

Wu, S., Dong, X., Chang, Y., Carvalho, P.N., Pang, C., Chen, L. and Dong, R. (2015). Response of a tidal operated constructed wetland to sudden organic and ammonium loading changes in treating high strength artificial wastewater. *Ecological Engineering*, 82, 643-648.

Zhao, Y.Q., Sun, G. and Allen, S.J. (2004). Purification capacity of a highly loaded laboratory scale tidal flow reed bed system with effluent recirculation. *The Science of the Total Environment*, 330(1-3), 1-8.

Zhi, W. and Ji, G. (2014). Quantitative response relationships between nitrogen transformation rates and nitrogen functional genes in a tidal flow constructed wetland under C/N ratio constraints. *Water Research*, 64(7), 32-41.