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A survey on the ratio of effluent algal BOD concentration in primary and secondary facultative ponds to influent raw BOD concentration

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ABSTRACT

It is well known that algae play an important role in treatment of municipal wastewater with facultative ponds. The objective of the present study was to determine the ratio of effluent algal BOD concentration in facultative ponds to influent BOD concentration. This descriptive analytical study was carried out on a full-scale wastewater stabilization pond system located in Islamabad Gharb, northeast Iran. A total of 138 grab samples were collected from Primary and Secondary Facultative Ponds (PFPs and SFPs) twice per month over a period of one year and analyzed for algal cell number, chlorophyll a, and BOD_5 concentrations. The mean ratios of effluent algal concentration of PFP_5 and SFP_7 to influent BOD_5 of PFP_5 were 1.11 ± 0.53 and 1.59 ± 0.66, respectively. Also, the mean ratios of effluent algal concentration of PFP_6 and SFP_8 to influent BOD_5 of PFP_6 were 1.20 ± 0.63 and 1.57 ± 0.68, respectively. Except PFP_6, significant differences were found in the means of the respective ratio in PFP_5, SFP_7, and SFP_8 between cold and warm seasons ($p < 0.05$). Although there was a significant difference in the means of the respective ratio between primary and secondary facultative ponds ($p < 0.05$), there was no significant difference between the same ponds in modules ($p > 0.05$). Lower effluent algal concentration to influent BOD_5 ratio found in cold months indicates strong dependency of algal photosynthetic activity to sunlight and temperature. Also, the effluent algal concentration in secondary facultative ponds was higher than that in primary facultative ponds, since better condition is provided by secondary facultative ponds for algal–bacterial activity.

Keywords: Algal BOD concentration; Influent BOD_5; Chlorophyll a concentrations; Primary and secondary facultative pond

1. Introduction

Stabilization ponds are natural systems favored for municipal and industrial wastewater treatment in Iran, where climatic conditions are suitable and large land is available [1]. Facultative ponds, the most common type of wastewater stabilization pond (WSP), consist of an aerobic zone in top layer and an anaerobic zone in very bottom layers. In top zone, there is a mutualistic relationship between algae and facultative and aerobic bacteria in a way that algae generate alga cells and oxygen through photosynthesis in the presence of

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sunlight, using minerals produced through the activity of aerobic bacteria. The bacteria oxidize organic matter (suspended and dissolved BOD) found in wastewater using dissolved oxygen generated by algae. This process can result in the biological decomposition and stabilization of organic matter and the growth of bacteria. Heterotrophic bacteria such as Pseudomonas, Flavobacterium, Achromobacter, and Alcaligenes play a dominant role in this process [2–7].

Metabolic activity of aerobic bacteria yields CO_2, PO_4, and NO_3. Algae use carbon, phosphorus, nitrogen, and other minerals during photosynthesis and generate new alga cells and oxygen [3,8,9]. Regarding mutualistic relationship noted above, algae have a key role in wastewater treatment in facultative ponds. In the top layers of the ponds (known as photic zone) photosynthetic activity is carried out by a wide range of algal species (mostly green algae, green–blue algae, and diatoms) producing 10–66 g algae/m^2/day [10]. The most common algae types encountered in facultative ponds are Chlorella, Scenedesmus, Micractinium, Oscillatoria, Microcystis, Chlamydomonas, and Euglena [10–13]. Algal photosynthetic activity that is strongly temperature and light-dependent can cause pH to increase, especially if wastewater alkalinity content is low. This may render condition favoring the removal of nutrient. In high pH condition, phosphorus is precipitated as calcium phosphate and ammonium release into the atmosphere as ammonia. The concentration of algae in facultative pond effluents usually ranges between 500 and 2,000 g/l (chlorophyll a basis) depending on organic loading and temperature [2,14].

Marais (1970) estimated that around 30% of influent BOD leaves primary facultative ponds (PFP) in the form of methane [3]. Moreover, as a result of algal–bacterial activities described earlier, a high proportion of the influent BOD that does not leave the pond as methane finally ends up to algal cells. Thus, sewage BOD is converted into "algal BOD" in secondary facultative pond and upper layers of PFP. Depending on operational parameters as well as the parameters like local climate and season, effluent algal BOD of both primary and secondary facultative ponds may be higher than influent total BOD [15]. Few literature has been reported that 70–90% of the BOD in final effluent of facultative ponds is due to their algae content [8,16], although, there is insufficient empirical evidence to support it. There is also no comprehensive report on BOD contribution of the algae in primary and secondary facultative pond effluents separately. Some researchers now believe that the range noted above may be changed depending on many factors such as local climatic conditions, ambient temperature, wastewater constituents (especially the level of biodegradable organic matter), retention time, and operation and maintenance status. Present study was conducted to determine the ratio of effluent algal BOD (biodegradable and non-biodegradable) concentration to BOD concentration of raw wastewater entering to primary and secondary facultative ponds in Islamabad-e-Gharb wastewater treatment plant.

2. Materials and methods

2.1. Study site

Islamabad Gharb city is located between north latitudes of 34°-6′ and 34°-7′ and west longitudes of 51°-4′ and 51°-33′ in northwest of Iran. The city is situated at an altitude of 1,330–1,380 meter above sea level and has a predominantly semi-arid desert climate. Wastewater treatment plant in the Islamabad Gharb was originally constructed in 2005 and consisted of six stabilization ponds in two modules operated in parallel. The ponds in each module arranged in three stages, one anaerobic pond as the first stage followed by one PFP and one secondary facultative pond (SFP) (see Fig. 1). Design characteristics of PFPs and SFPs are given in Table 1. The existing wastewater treatment facility currently handles wastewater produced from 90,000 people and treats approximately 27,000 m^3 sanitary sewage per day (13,500 m^3 per day per module). It was projected that the plant will fulfill the wastewater management needs of 120,000 people in the future.

2.2. Sampling

In a descriptive analytical study, samples were taken twice per month for one year. A total of 138 grab samples were collected from facultative ponds (46 samples at the PFPs outlet, 46 samples at the SFPs outlet, and 46 samples at the PFPs inlet) and analyzed for algal cell number, chlorophyll a, and BOD_5 concentrations. Among all 23 collected samples at each location, 12 samples (samples No. 2–13) were collected from April to September (representing warm months), and 11 samples (samples No. 1 and 14–23) were collected from October to March (representing cold months). All samples were taken using a 1L polyethylene bottle and preserved with lugol's solution for later analysis.

2.3. Analysis and enumeration

Algal cells were enumerated by Sedgwick–Rafter microscopic slide based on the standard methods [17]. The samples were concentrated through sedimentation

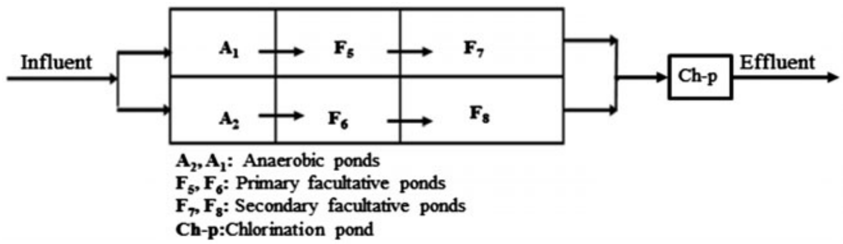

Fig. 1. Schematic diagram of WSP system in Islamabad Gharb.

Table 1
Design characteristics of the primary and secondary facultative ponds

Pond system	HRT (day)	Area (m^2)	Volume (m^3)	Organic loading (kg BOD/ha)	Width to length ratio	Mean depth (m)
PFP	7.98	67,914.24	107,688	180	1:7.43	1.54
SFP	3.13	33,271.00	42,219	87	1:3.54	1.50

before analysis. First, a volume of 1L sample from pond effluents was allowed to sediment over 24 h. Then top layer of sample (95% volume) was carefully siphoned off without disturbing the settled algae. The remaining sample was shaken gently and an aliquot of 1 ml was immediately transferred to a labeled 100 ml sample bottle with a large bore disposable plastic pipette. Based on algal cells density, the sample was diluted by a factor of 100 using distilled water. Afterward an aliquot of diluted solution (1 ml) was transferred to a Sedgwick–Rafter slide (50 × 20 × 1 mm) for final examination. Sedgwick–Rafter cell was etched with a calibrated grid of 50 × 20 equal sized squares (1 mm). Before counting, the full Sedgwick-Rafter slide was left for 15–20 min to ensure the sample has adequately settled. Finally, it was placed directly under the upright microscope included the 10 × ocular in combination with 4 × objective (a total 40 × magnification). The number of algal cells per milliliter was calculated by Eq. (1).

$$C\,[\text{cells/ml}] = \frac{N \times 1000\,\text{mm}^3}{A \times D \times F} \quad (1)$$

where: N = number of cells counted, A = area of a field (mm^2), D = depth of a field (Sedgwick–Rafter chamber depth) (here 1 mm), and F = number of fields counted (here 5).

Algae mass was determined by a modified spectrophotometer method based on Almasi experiments in 2009. It was assumed that chlorophyll a constitutes 1% of dry weight of algae mass. After measuring chlorophyll a, algae mass was calculated using algorithm Eq. (2).

$$A = \frac{C \times 100 \times Q}{S} \quad (2)$$

where A is algae biomass in gram per m^2, C is chlorophyll a concentration in gram per m^3, 100 is algae to chlorophyll a dry weight ratio, Q is influent wastewater flow rate in m^3 per day, and S is surface area of facultative pond in m^2 [18].

Concentration of BOD_5 was analyzed according to Standard Methods [17]. The mean of algal BOD_5/influent BOD_5 ratio between cold and warm seasons was compared using the non-parametric Mann–Whitney U test at significance level of $\alpha = 0.05$. Independent sample t-test was also used to compare respective ratio between primary and secondary facultative ponds (PFP_5 to SFP_7 and PFP_6 and SFP_8) in each module and between PFP (PFP_5 and PFP_6) and secondary facultative ponds (SFP_7 and SFP_8). Statistical analyses were performed using SPSS version 11.5 software.

3. Results and discussion

BOD_5 concentration in PFP_5 influent and algal cell and chlorophyll a concentrations in effluent of both PFP_5 and SFP_7 as well as the ratios of effluent algal concentration in the PFP_5 and SFP_7 to influent BOD_5 concentration of PFP_5 are shown in Table 2 and Fig. 2. The respective parameters for PFP_6 and SFP_8 are presented in Table 3 and

Table 2
Influent BOD_5 and effluent algal concentration and their ratio in PFP_5 and SFP_7

Sample	BOD_5 concentration in the PFP_5 influent (mg/l)	Chlorophyll a concentration in the PFP_5 effluent (mg/l)	Total algal concentration in the PFP_5 effluent (mg/l)	Chlorophyll a concentration in the SFP_7 effluent (mg/l)	Total algal concentration in the SFP_7 effluent (mg/l)
1	95	890	89	1,890	189.0
2	110	1,260	126	2,670	267.0
3	115	780	78	2,690	269.0
4	168	1,900	190	2,430	243.0
5	110	2,340	234	2,332	233.2
6	130	987	98.7	2,870	287.0
7	109	2,050	205	2,549	254.9
8	107	760	76	1,980	198.0
9	96	980	98	2,768	276.8
10	143	2,030	203	1,876	187.6
11	159	2,100	210	1,865	186.5
12	115	1,870	187	2,456	245.6
13	121	1,800	180	2,589	258.9
14	135	980	98	2,145	214.5
15	115	1,100	110	1,765	176.5
16	119	1,400	140	1,567	156.7
17	104	1,230	123	1,237	123.7
18	125	1,320	132	1,230	123.0
19	142	1,400	140	1,345	134.5
20	151	1,120	112	1,347	134.7
21	169	545	54.5	1,089	108.9
22	110	450	45	679	67.9
23	163	500	50	856	85.6
Mean	126.5 ± 22.9	$1,364.9 \pm 626.7$	$1,36.5 \pm 62.7$	$1,922.8 \pm 657.5$	192.3 ± 65.7

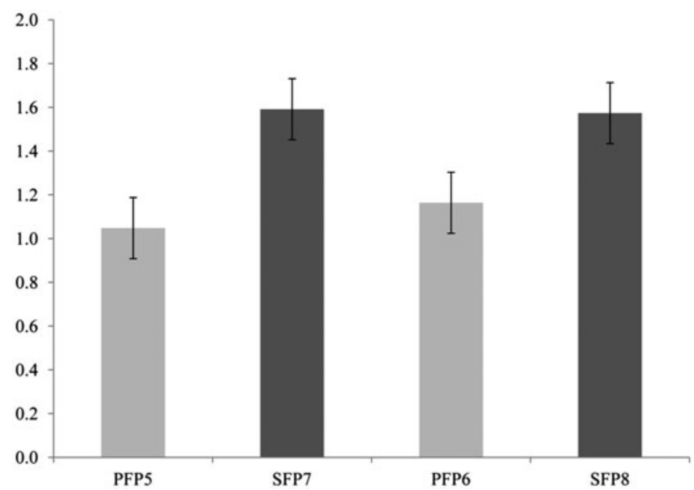

Fig. 2. Mean of effluent algal concentration to the influent BOD_5 concentration ratio in the primary and secondary facultative pounds.

Fig. 2. The ranges of effluent algal BOD concentration/influent BOD_5 concentration ratio in PFPs and SFPs for cold and warm seasons are given in Fig. 3.

Table 3
Influent BOD_5 and effluent algal concentration and their ratio in PFP_6 and SFP_8

Sample	BOD_5 concentration in the PFP_6 influent (mg/l)	Chlorophyll a concentration in the PFP_6 effluent (mg/l)	Total algal concentration in the PFP_6 effluent (mg/l)	Chlorophyll a concentration in the SFP_8 effluent (mg/l)	Total algal concentration in the SFP_8 effluent (mg/l)
1	156	1,030	103.0	1995	199.5
2	207	1,255	125.5	2,650	265.0
3	149	890	89.0	2,879	287.9
4	173	1,920	192.0	2,220	222.0
5	110	2,345	234.5	2,331	233.1
6	130	1,013	101.3	2,865	286.5
7	93	2,089	208.9	2,559	255.9
8	102	1,230	123.0	1,987	198.7
9	99	1,114	111.4	2,760	276.0
10	134	2,045	204.5	1,890	189.0
11	147	2,134	213.4	1,880	188.0
12	115	1,876	187.6	2,545	254.5
13	112	1,890	189.0	2,570	257.0
14	132	1,136	113.6	2,135	213.5
15	115	1,250	125.0	1,867	186.7
16	123	1,545	154.5	1,590	159.0
17	79	1,320	132.0	1,378	127.8
18	76	1,445	144.5	1,342	134.2
19	122	1,340	134.0	1,243	124.3
20	153	1,010	101.0	1,223	122.3
21	184	543	54.3	987	98.7
22	123	543	54.3	765	76.5
23	195	456	45.6	790	79.0
Mean	1,26.9 ± 31.6	1,449.5 ± 616.6	144.9 ± 61.7	1,894.3 ± 680.5	189.4 ± 68.1

Two-group independent sample *t*-test indicates that a statistically significant difference exist in the means of effluent algal BOD concentration/influent BOD_5 concentration ratios between PFP_5 and SFP_7 ($p = 0.003$) and between PFP_6 and SFP_8 ($p = 0.022$). However, no significant difference was observed in the means of effluent algal BOD concentration/ influent BOD_5 concentration ratios between PFP_5 and PFP_6 ($p = 0.542$) and between SFP_7 and SFP_8 ($p = 0.928$).

The results of two-group independent sample *t*-test indicated that the mean of effluent total algal BOD concentration to influent BOD_5 concentration ratio in PFP_5 (136.5 ± 62.7) has significant difference with the respective ratio in SFP_7 (192.3 ± 65.7) (p-value < 0.05). Similar difference was observed between PFP_6 (144.9 ± 61.7) and SFP_8 (189.4 ± 68.1) (p-value < 0.05).

Significant differences in algal bulk between primary and secondary facultative ponds in both modules and higher algal concentration in the effluent of SFPs illustrates optimum performance of the ponds.

The regression between the influent BOD and alga BOD concentration obtained using equations following (Eqs. (3) and (4)).

$$\text{Algal BOD}_{\text{effluent of primery FP}} = -0.548[\text{influent BOD}_5]_{\text{influent of primery FP}} + 208.74$$

$$\text{Algal BOD}_{\text{effluent ofprimery FP}} = 0.116[\text{influent BOD}_5]_{\text{influent of primery FP}} + 0.525[\text{influent BOD}_5]_{\text{influent of primery FP}} + 100.38 \quad (4)$$

As a result of the algal-bacterial activities described earlier, a high proportion of sewage BOD was converted into algal BOD. This process is mainly limited to upper layers of PFP, while it can be extended to the deeper layers of secondary facultative ponds [18,19]. Therefore, effluent algal BOD concentration in secondary facultative ponds was higher than that in PFP. Since the generated algal BOD in PFP is introduced into the secondary facultative pond, it is evident that the effluent algal levels in secondary facultative pond are higher than in PFP.

The results of the independent sample *t*-test revealed that there was no significant difference in average concentration of effluent algal BOD /influent BOD_5 ratios between PFP_5 (136.5 ± 62.7) and PFP_6

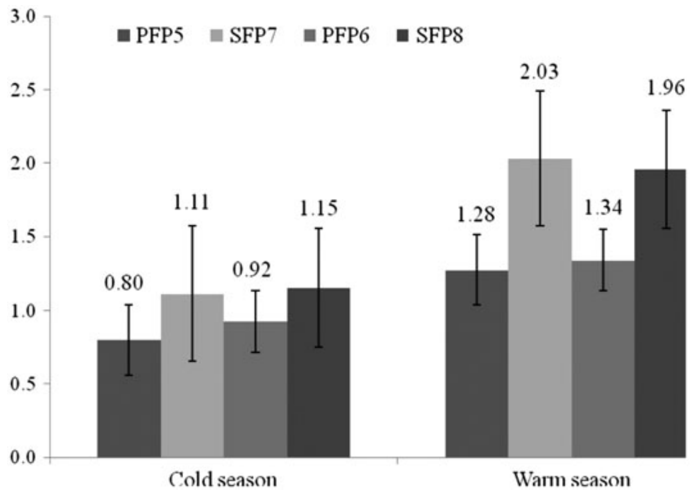

Fig. 3. Mean of effluent algal concentration to the influent BOD_5 concentration ratio, within the warm (15–27˚C) and cold (5–15˚C) seasons.

(144.9 ± 61.7) and between SFP_7 (192.3 ± 65.7) and SFP_8 (189.4 ± 68.1) (p-values > 0.05). This is expected because of the similarity in arrangements of ponds in both modules (Fig. 1). Based on the non-parametric Mann–Whitney *U* test, significant differences were found in average concentration of effluent algal BOD to influent BOD_5 ratios between cold (temperature range 5–15˚C) and warm (temperature range 15–27˚C) seasons ($p < 0.05$) for PFP_5, SFP_7, and SFP_8. However, such a difference was not found for PFP_6 (p-value > 0.05).

Facultative pond performance is affected by ambient temperature for two main reasons. Low temperatures slow down microbial activity and the colder months associated with short day length and weak solar radiation. Therefore, algal photosynthesis is strongly dependent on temperature and sunlight [11], so algal photosynthesis and microbial metabolism increase with increasing temperature and vice versa [2]. As a rule, biological activity will double if temperature increases by 10˚C. All ponds, particularly facultative ponds, perform well on a sunny and cloudless day at an air temperature above 20˚C and mild wind conditions [3,20].

On the other hand, lower algal photosynthetic activity in cold weather conditions can result in lower oxygen production that affects bacterial growth and consequently algal growth. Insufficient oxygen leads to reduced activity of heterotrophic Bactria (Pseudomonas, Flavobacterium, Achromobacter, and Alcaligenes) and the presumptive symbiotic cycle between the bacteria and algae may be disturbed in facultative ponds [21].

The presence or absence of sunlight is other important parameter influencing facultative pond performance. Sunlight influences oxygen generation indirectly via changing photosynthetic activity. Therefore, intensity and duration of sunlight have a major effect on algal-bacterial interactions in facultative ponds, particularly colder months in which day length is shorter and solar radiation is weaker than those in warm months. Photosynthesis rate at sunlight intensities of higher than a certain point reaches to a plateau. However, researchers were found that duration of sunlight is more influential than its intensity on photosynthetic activity of algae [22–26].

It must be remembered that the effluent BOD of facultative ponds is very high and its nature is different from influent BOD. However, BOD levels in the effluent of facultative ponds must be complying with regulatory effluent requirements established by Iranian Institute of Standard and Industrial Research and Environmental Protection Agency. The facultative ponds apparently do not have sufficient efficiency in removal of sewage BOD, but it must be remarked that “algal BOD” is very different in nature to “sewage BOD.” It should be emphasized that this effluent may be advantageous to reuse in agriculture. Algae essentially act as slow-release fertilizers and promote plant growth via increasing the soil humus content and improving its water-holding capacity [20]. Accordingly, the importance of blue-green algae as one of the major components of the nitrogen fixing biomass in rice paddy fields has long been recognized [21]. Therefore, in monitoring of WSP effluents unlike other effluents, filtered samples are used to determine BOD and COD, which are therefore the residual non-algal BOD values [3]. Unfortunately, some laboratories do not consider this fact and yet report effluent total (algal and non-algal) BOD and COD for WSPs. Since in WSP effluents, the algae comprise most of the suspended solids (> 80%), in the European Union, pond effluents can contain up to 150 mg SS/l, whereas effluents from other treatment processes must contain only 35 mg SS/l. This regulatory requirement reflects the inherent difference between algal BOD and sewage BOD (or COD) and SS [2].

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