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Corn Straw as a Solid Carbon Source for the Treatment of Agricultural Drainage Water in Horizontal Subsurface Flow Constructed Wetlands

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Abstract: Agricultural drainage water with a low C/N ratio restricts the nitrogen and phosphorus removal efficiencies of constructed wetlands. Thus, there is a need to add external carbon sources to drive the nitrogen and phosphorus removal. In this study, the effects of the addition of corn straw pretreated with different methods (acid treatment, alkali treatment, and comminution) on treating agricultural drainage water with a low C/N ratio were investigated in constructed wetlands. The results showed that soaking the corn straw in an alkaline solution was the most suitable pretreatment method according to the release rule of chemical oxygen demand (COD) and the dissolution of total nitrogen (TN) and total phosphorus (TP). The average removal efficiency of TN and TP in constructed wetlands increased respectively by 37.2% and 30.5% after adding corn straw, and by 17.1% and 11.7% after adding sodium acetate when the hydraulic retention time (HRT) was 3 days. As an external carbon source, straw was cheap, renewable, and available. In contrast, the sodium acetate demanded high costs in a long-term operation. Therefore, corn straw had a great advantage in treatment effect and cost, which improved the treatment efficiency of agricultural drainage water using a byproduct of agricultural production as a slow-release carbon source.

Keywords: corn straw; solid carbon sources; agricultural drainage water; low C/N ratio; constructed wetland

1. Introduction

More and more fertilizers have been widely used to increase the agricultural production and alleviate the food crisis caused by population growth. However, due to the low utilization rate, large amounts of unutilized fertilizers run into the surface water along with agricultural drainage water [1]. The agricultural drainage water often contains COD, organic nitrogen, NH₄⁺-N, NO₃⁻-N, and some inorganic phosphates [1]. It has the characteristics of a low C/N ratio, high proportion of nitrate, and the fluctuation of water quality and quantity [2]. The discharged drainage water can easily result in eutrophication [3]. Therefore, an economical and practical treatment technique for agricultural drainage water is desired.

The low C/N ratio in wastewater is often treated with processes such as anaerobic ammonium oxidation and simultaneous nitrification and denitrification, which have the shortcomings of complex operation and management, high energy consumption, and high cost [4,5]. Constructed wetlands (CWs) are known as an ecological technology with a good purification effect, simple process equipment, low setup and maintenance costs, and high ornamental value [6]. CWs have been successfully applied to treat domestic, municipal, and industrial wastewater, as well as contaminated surface water by combining physical, chemical, and biological processes [7–9]. It is well known that biological
nitrogen removal mainly relies on successful ammonia oxidation and nitrate denitrification to nitrogen gas. In the absence of a carbon source, the denitrification will be restricted in CWs [10]. Therefore, an additional carbon source is necessary for treating agricultural drainage water with a low C/N ratio. Studies have shown that traditional liquid carbon sources, including glucose, methanol, ethanol, starch, and sodium acetate, can be used to improve the efficiency of biological denitrification in CWs [11,12]. The liquid carbon source needs to be added continuously, which is high-cost and can easily cause secondary pollution [13]. Compared with liquid carbon sources, solid carbon sources can not only work for a long time, but also create a stable living environment for the denitrifying bacteria [14]. Recent research has indicated that low-cost and renewable natural organic substances, such as corn straw, rice husk, litter, woodchips, sawdust, cotton, maize cobs, seaweed, and bark, can be used as external solid carbon sources to drive denitrification [15]. Shao et al. [16] demonstrated that rice husks can be an economical and effective carbon source and biofilm carrier in the biological denitrification of wastewater in up-flow laboratory reactors. Chen et al. [17] reported that *Typha latifolia* litter addition could greatly improve nitrate removal in subsurface-batch CWs through the continuous input of organic carbon. Yang et al. [18] found that *retinervus luffae fructus*, corn cob, and rice straw were the favorable solid carbon sources and biofilm carriers due to their better carbon release capacity, denitrification potential, and relatively large surface area. Compared to the control membrane bioreactors, TN removal was enhanced by 25.5%, 19.5%, and 38.9%, respectively. Xu et al. [19] and Li et al. [20] also successfully drove denitrification using corn cobs. In addition, corn straw, *Arundo donax*, and *Pontederia cordata* were also proven to be effective carbon sources [11,21]. However, few reports have been reported on the treatment of agricultural drainage water using corn straw as carbon source in CWs, as well as the influence of pretreatment methods on the carbon source release of corn straw.

In this study, common corn straw was selected as the solid carbon source. The objectives of this study were: (1) to analyze the effects of pretreatment methods on the amount and rate of carbon released by corn straw; (2) to study the contribution of the solid carbon source to the removal of COD, N, and P in the CW system; and (3) to assess the economy of corn straw used as a slow-release solid carbon source in treating agricultural drainage water with a low C/N ratio.

2. Materials and Methods

2.1. Pretreatment of Corn Straw

Natural corn straw used in all CWs of this study was collected from a local village of Chiping county in Shandong province. The corn straw was cleaned with tap water before drying in the air, cut into 2–3-cm pieces, and then dried at 40 °C to a constant mass. The treated corn straw was kept in a moisture-free container before use.

2.2. Carbon Dissolution Test

Corn straw was treated with four different processes to explore the effects of pretreatment methods (Figure 1). After pretreatment, 2.0 g corn straw was put into 100 mL distilled water and soaked under the water. The collected water samples (once per hour at first and then once a day) were immediately filtered through a 0.45-µm cellulose acetate membrane and analyzed for the dissolution of COD, TN, and TP.

2.3. Characteristics of the Wastewater

Based on the characteristics of agricultural drainage water in different areas [22–24], synthetic wastewater was prepared. The composition of the synthetic wastewater was as follows (mg/L): NH$_4$Cl 5,730, KH$_2$PO$_4$ 6,590, Ca(H$_2$O)$_6$ 26,416, and KNO$_3$ 75,825. This solution represented 28.0 mg/L COD, 1.5 mg/L NH$_4^+$-N, 10.5 mg/L NO$_3^-$-N, 12.0 mg/L TN, and 1.5 mg/L TP. The pH of the synthetic wastewater was 7.2–7.5.
Figure 1. (a) Non-treated corn straw; (b) corn straw treated with acid (solid-liquid ratio 1:20, soaked in 20% HCl for 24 h, washed, and then dried to constant weight); (c) corn straw treated with alkali (solid-liquid ratio 1:30, soaked in 1% NaOH for 24 h, washed, and then dried to constant weight); and (d) comminuted corn straw.

2.4. Wetland Setup and Operation

The five sets of systems established in the greenhouse at Qingdao University were as follows: no added carbon source (W1: HRT = 2 days), added corn straw (W2: HRT = 2 days), added sodium acetate (W3: HRT = 2 days), added corn straw (W4: HRT = 3 days), and added corn straw (W5: HRT = 1 day). The volume of each system was 25 L (length: 0.45 m, width: 0.30 m, height: 0.30 m). All CWs reactors were filled with washed gravel and sand. The top of the CWs were open and the bottom had one opening valve for emptying. In five CWs, multi-dimensional gradation of the gravel was adopted: larger sized gravels (5–8 cm) were placed at the bottom to avoid clogging, on top of which were smaller sized gravels (1–5 cm). Finally, a 5-cm layer of washed sand (particle size < 2 mm) was placed at the top, in which the *Typha latifolia* was planted. Continuous experiments were carried out in an air-conditioned greenhouse at 23 ± 1 °C. After planting, the systems were operated for four weeks with synthetic wastewater until the plant shoots and microorganisms were well established. According to previous research, we chose a proper C/N ratio to achieve a complete denitrification (C/N = 8) [6,25,26]. About 1.87 g sodium acetate was continuously added to W3 daily. Based on the results of the carbon source release, 68.08, 43.57, and 136.16 g corn straw was added to W2, W4, and W5, respectively.

2.5. Sampling and Analysis

The water samples were collected from the reactor and filtered through a 0.45-µm cellulose acetate membrane before analysis. The pH and temperature were monitored by a pH/mV Meter (PHS-3CW, Bante Instrument Co. Ltd., Shanghai, China) and Pen type thermometer (TP101, Shanghai Automation Instrument Factory, Shanghai, China), while conventional pollutants in wastewater (COD, NH$_4^+$-N, NO$_3^-$-N, TN, and TP) were determined according to Chinese standard methods [27]. COD was measured using the potassium dichromate method. NH$_4^+$-N, NO$_3^-$-N, TN, and TP were determined by a UV–vis spectrophotometer (TU-1810, Persee instrument Co. Ltd., Beijing, China) [28].
2.6. Statistical Analysis

All tests in this study were performed in triplicate and the results are expressed as mean ± standard deviation. Experimental data was processed using Microsoft Excel 2013 and the charts were drawn using Microsoft Excel 2013 and Origin 8.0 software.

3. Results and Discussion

3.1. Effect of Pretreatment Methods on the Carbon Source Release of Corn Straw

As shown in Figure 2, the carbon, nitrogen, and phosphorus release rates of the four samples were relatively fast at the early stage, after which they tended to be stable. The COD contents released by four kinds of corn straw were respectively 55.0, 67.5, 94.8, and 242.5 mg/g at the early stage (Figure 2a), due to the release of small molecule organic matter attaching to the surface of the corn straw. In particular, the comminuted corn straw with a larger surface area released a much higher COD content than other treatments. After about 4 days, the small molecule organic matter in the surface was released completely, and most of the COD in water samples were from cellulosic materials, which is main component of corn straw [29]. Alkali treatment was mainly applied to cellulose, which was hydrolyzed to glucose and other monosaccharides, while acid treatment acted mainly on nucleoside bonds that caused fragmentation [30]. Therefore, the released COD contents levels were in the order of c > b > a > d, corresponding to 6.24, 4.17, 2.49, and 1.01 mg/g, respectively. At the early stage, the TN contents released by four kinds of corn straw were in order of d > a > b > c (Figure 2b), and the TP contents were in the order of d > b = c > a (Figure 2c). This was mainly due to the fact that the nitrogen and phosphorus contents in corn straw were high, and the release of nitrogen and phosphorus sped up along with the rapid decomposition of organic matter. The release of nitrogen and phosphorus was similar to the results of previous researchers [1,29]. After 6 days, the TN contents from a, b, c, and d were 0.272, 0.291, 0.233, and 0.266 mg/g, and the TP contents were 0.005, 0.003, 0.003, and 0.005 mg/g, respectively, which were very close to each other. The above results suggested that different treatment methods had no significant effect on the nitrogen and phosphorus release from corn straw.

The C/N values of a, b, c, and d exhibited a sharp decline in the early stage, then rose and finally tended to be stable, at levels of 9.4, 11.8, 24.3, and 4.0, respectively (Figure 3). It was obvious that the corn straw treated by alkali could provide more carbon and little nitrogen as an external carbon source.

Figure 2. Cont.
Figure 2. COD, TN, TP dissolution trends of corn straw pretreated with different methods; (a) COD; (b) TN; and (c) TP.

Figure 3. C/N variation trends of corn straw pretreated with different methods.

3.2. Effects of External Carbon Source and HRT on COD Removal

The five CWs reached a steady state after being cultivated with synthetic wastewater for four weeks (Table 1). After the CWs were stable, an external carbon source was added to W2, W3, W4, and W5. As shown in Figure 4, the influent COD concentrations of W1 and W2 were 26.40–29.30 mg/L and the average COD removal efficiency was 63.2% and 45.0%, respectively. The influent COD concentration of W3 was 95.60–98.60 mg/L, and the average COD removal efficiency was 74.5%. This indicated that the CWs had a low effluent COD concentration after adding corn straw and sodium acetate. The effect of HRT on the COD removal is shown in Figure 4. The influent COD concentrations of W2, W4, and W5 were in the range of 26.40–29.30 mg/L and the average COD removal efficiencies were 45.0%, 54.1%, and 40.7%, respectively, indicating that the HRT had an impact on COD removal in CWs, and that the optimum HRT was 3 days.
**Table 1.** The removal efficiency of COD, NH$_4^+$-N, NO$_3^-$-N, TN, and TP in five constructed wetlands before adding external carbons.

| Reactor | COD Influent (mg/L) | COD Effluent (mg/L) | Average Removal Efficiency (%) | NH$_4^+$-N Influent (mg/L) | NH$_4^+$-N Effluent (mg/L) | Average Removal Efficiency (%) | NO$_3^-$-N Influent (mg/L) | NO$_3^-$-N Effluent (mg/L) | Average Removal Efficiency (%) | TN Influent (mg/L) | TN Effluent (mg/L) | Average Removal Efficiency (%) | TP Influent (mg/L) | TP Effluent (mg/L) | Average Removal Efficiency (%) |
|---------|---------------------|---------------------|-------------------------------|-----------------------------|-----------------------------|-------------------------------|-----------------------------|-----------------------------|-------------------------------|----------------|----------------|-------------------------------|----------------|----------------|-------------------------------|
| W1      | 27.0 ± 1.0          | 10.0 ± 1.0          | 61.4                          | 1.50 ± 0.10                 | 0.60 ± 0.15                 | 61.4                          | 10.5 ± 0.6                  | 4.50 ± 0.20                 | 57.1                          | 12.0 ± 0.5    | 5.90 ± 0.2    | 50.7                          | 1.50 ± 0.15    | 0.80 ± 0.10    | 35.4                          |
| W2      | 28.0 ± 1.5          | 13.0 ± 1.7          | 54.4                          | 1.50 ± 0.25                 | 0.50 ± 0.10                 | 65.6                          | 11.0 ± 0.6                  | 4.28 ± 0.13                 | 60.0                          | 12.0 ± 0.4    | 4.78 ± 0.2    | 53.2                          | 1.50 ± 0.15    | 0.75 ± 0.10    | 37.5                          |
| W3      | 28.3 ± 1.3          | 12.1 ± 2.1          | 55.6                          | 1.35 ± 0.10                 | 0.60 ± 0.30                 | 58.1                          | 10.7 ± 1.0                  | 4.07 ± 0.23                 | 61.6                          | 12.3 ± 1.0    | 4.67 ± 0.3    | 54.4                          | 1.55 ± 0.10    | 0.75 ± 0.10    | 38.3                          |
| W4      | 27.6 ± 0.8          | 9.1 ± 2.0           | 67.5                          | 1.60 ± 0.15                 | 0.50 ± 0.05                 | 65.3                          | 11.0 ± 0.5                  | 3.57 ± 0.50                 | 68.5                          | 12.6 ± 0.5    | 4.07 ± 0.4    | 64.2                          | 1.45 ± 0.10    | 0.65 ± 0.10    | 56.7                          |
| W5      | 26.6 ± 1.8          | 13.4 ± 1.9          | 48.4                          | 1.60 ± 0.10                 | 0.80 ± 0.10                 | 49.3                          | 11.0 ± 0.5                  | 5.40 ± 0.40                 | 50.1                          | 12.6 ± 0.5    | 6.20 ± 0.5    | 36.8                          | 1.45 ± 0.10    | 0.95 ± 0.20    | 30.2                          |

*Values are mean values ± standard deviations.*
3.3. Effects of External Carbon Source and HRT on Nitrogen Removal

The effect of the external carbon source on NH$_4^+$-N removal is shown in Figure 5a. The influent NH$_4^+$-N concentrations of W1, W2, and W3 were in the range of 1.45–1.55mg/L, and the average NH$_4^+$-N removal efficiencies were respectively 67.2%, 74.8%, and 75.2%, indicating that the external carbon source improved the NH$_4^+$-N removal and that corn straw was more efficient than sodium acetate. The first reason for this was that the denitrifying bacteria grew vigorously in enough carbon sources, which also required a certain amount of nitrogen during growth. Secondly, the CW was a mixed culture system, where nitrifying bacteria, autotrophic bacteria, and denitrifying bacteria coexisted. Therefore, nitrification, denitrification, and anaerobic ammonia oxidation might occur simultaneously [11]. In W2, the corn straw pretreated by alkali might release ammonia while providing a carbon source. Thus, the NH$_4^+$-N in W2 was slightly larger than that in W3.

The NO$_3^-$-N concentrations of the influent in W1, W2, and W3 were in the range of 9.90–11.40 mg/L, and the average effluents were 4.89, 1.69, and 1.72 mg/L, respectively (Figure 5b). In W1, denitrifying bacteria could not obtain a sufficient carbon source, so nitrogen removal efficiency was poor. However, denitrifying bacteria could obtain enough carbon sources in W2 and W3 after the addition of an external carbon source. Therefore, an external carbon source can improve the denitrification efficiency of CWs [17,31]. Corn straw showed a better promotion than sodium acetate, because the corn straw could not only increase the available carbon source, but also provide places for microbial attachment, thus increasing the biomass [32,33]. Moreover, the influent TN concentrations in W1, W2, and W3 were in the range of 11.35–12.90 mg/L, and the average TN removal efficiencies
were 50.8%, 71.9%, and 67.9%, respectively (Figure 5c). It was obvious that the nitrification and denitrification progress were promoted in W2 and W3 after adding the external carbon source. However, the removal efficiency in W2 was 4.0% higher than that in W3. This indicated that as a solid slow-release carbon source, corn straw was a better choice for treating low C/N wastewater in CWs than sodium acetate.

![Figure 5](image)

HRT had an influence on nitrogen removal. The NH$_4^+$-N concentrations in W2, W4, and W5 were in the range of 1.45–1.55 mg/L; and the average removal efficiencies were 74.8%, 79.3%, and 65.6%, respectively (Figure 6a). The above result indicated that HRT had an influence on NH$_4^+$-N removal, and the NH$_4^+$-N removal efficiency in W4 (HRT = 3 days) was significantly higher than those in W2 and W5. The HRT of 3 days was optimum for microbial nitrification, which meant better NH$_4^+$-N removal. The influent NO$_3^-$-N concentrations of W2, W4, and W5 were in the range of 9.90–11.40 mg/L, and the effluent NO$_3^-$-N concentrations were 1.69, 0.72, and 3.65 mg/L, respectively (Figure 6b). This suggested that the HRT had a great influence on denitrification [34]. In addition, the TN removal efficiencies in W2, W4, and W5 varied with the HRT, which was 71.9%, 87.9%, and 52.3%, respectively (Figure 6c). The HRT of 3 days was also optimum for the removal of TN in CWs [35].

**Figure 5.** Effects of external carbon source on nitrogen removal; (a) NH$_4^+$-N; (b) NO$_3^-$-N; and (c) TN.
3.4. Effects of External Carbon Source and HRT on Phosphorus Removal

The phosphorus removal in CWs includes biotic processes (e.g., the uptake and growth of plants and microorganisms) and abiotic processes (e.g., settling, sorption on substrates, co-precipitation with minerals, adsorption, and precipitation) [36,37]. Phosphorus is mostly removed through precipitation/adsorption in the media. Plant uptake and biological assimilation are limited processes. Since the CWs were relatively new, the increase in phosphorus removal observed with the addition of the carbon source was attributed to the faster build-up of biofilm within the system and to the clean and unsaturated gravel media. In this study, the phosphorus removal through the uptake and growth of microorganisms was different in the same five CW systems. In the aerobic zone, phosphate-accumulating organisms (PAOs) could oxidize organics material to obtain energy, resulting in an increase of phosphate concentration in the water. Under aerobic conditions, excessive phosphorus accumulated by PAOs was converted into polyphosphate using oxygen as an electron acceptor and then stored inside the cells.

As shown in Figure 7a, the influent TP concentrations in W1, W2, and W3 were in the range of 1.40–1.55 mg/L, and the average TP removal efficiencies were 34.9%, 49.7%, and 46.6%, respectively. For agricultural drainage water with a low C/N ratio, the release of phosphorus was suppressed. Therefore, W2 and W3 were better than W1 in the treatment of phosphorus after the addition of a carbon source. The TP removal efficiencies in W2, W4, and W5 varied with the HRT, which were 49.7%, 65.4%, and 33.4%, respectively (Figure 7b). Phosphorus removal in the CWs consisted of physical and chemical reactions, plant uptake, and microbial assimilation. With the increasing HRT, the above progresses played out more fully. Thus, the HRT of 3 days was optimum for the removal of TP in CWs.
Figure 7. Effects of carbon source and HRT on TP removal: (a) carbon source; (b) HRT.

3.5. The Cost Analysis of Different External Carbon Sources

The average removal efficiencies of TN after adding corn straw and sodium acetate were 71.9% and 67.9%, respectively. It was obvious that corn straw and sodium acetate were good choices as external carbon sources to remove nitrogen from the wastewater. The cost of corn straw and sodium acetate as the external carbon source was calculated according to the wastewater treatment capacity of 1 t/day. One liter of agricultural drainage water required 0.14 g sodium acetate, which was about 2.20 RMB/kg. The cost of sodium acetate was about 43.20 RMB for 20 days. Conversely, 1 L agricultural drainage water needed 5.30 g corn straws, costing little money. Before use, the corn straw needed to be pretreated with NaOH, requiring about 3.30 RMB/kg. The cost of corn straw and NaOH was about 10.36 RMB for 20 days, which was lower than that of sodium acetate under the same conditions.

In the north of China, corn is a common crop. As a byproduct of corn, corn straw is widely abundant and its price is very low (about 300.00 RMB/t). Therefore, enough corn straw can be provided for full-scale CWs. Corn straw used in full-scale wetland systems need to be purchased, transported, pretreated, and distributed. The following cost calculation was conducted according to the wastewater treatment capacity of 100 t/day. The overall cost was about 1105.62 RMB, including the purchasing cost of 190.80 RMB, transportation cost of 200.00 RMB, pretreatment cost of 314.82 RMB, and labor cost of 400.00 RMB. Given that corn straw can efficiently work 30 days at least, the treating cost of a ton of waste water is about 0.37 RMB. Due to the low cost, sufficient quantity, and good effect, it is feasible to use corn straw as an external carbon source in CWs.

4. Conclusions

This study investigated the effects and economy of using corn straw as an external carbon source on treating agricultural drainage water with a low C/N ratio. Through different pretreatments (acid treatment, alkali treatment, and comminution), some organic matter, N and P, was released from the corn straw in the dissolution process. Compared with the other two pretreatment methods, the alkali treatment could provide more COD with lower N and P release by hydrolyzing the cellulose material of corn straw to glucose and other monosaccharides. The removal efficiencies of TN and TP were significantly promoted by adding straw and sodium acetate as the external carbon sources, compared with the control test. The effects of the HRT on the removal of N and P were also studied. It was revealed that the optimum HRT was 3 days. As an agricultural byproduct, the corn straw was cheap and easily obtained, which presented great advantages in treating agricultural drainage water with a low C/N ratio in CW. The long-term effects of corn straw as an external carbon source on treating agricultural drainage water in CWs will be studied in the future.
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