

*Original Research*

# The Optimal Planting Coverage Rate and Wastewater Concentration for Water Spinach Floating Bed on Geese Wastewater Purification

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*Received: 23 March 2021*

*Accepted: 16 July 2021*

## Abstract

The goal of this study was to explore the optimal planting coverage rate and wastewater concentration required by water spinach floating bed system for the purification of geese farm wastewater. The results showed that the floating bed with water spinach in high concentration wastewater had better removal rates on ammonia nitrogen ( $\text{NH}_4^+\text{-N}$ ), total nitrogen (TN) and total phosphorus (TP) ( $P < 0.05$ ). There was no significant difference on the removal of nutrients except turbidity (NTU) among the different plant coverage rates ( $P > 0.05$ ). The floating bed system with water spinach removed heavy metals (Hg, As and Zn) in different concentration wastewater, with removal rates of 22.21%~68.20%, 40.00%~52.94% and 100.00%. Water spinach in low concentration wastewater had significant higher root length, stem height and weight gain ( $P < 0.05$ ) than medium and high concentration wastewater. The TN and TP contents of water spinach stem leaf in high concentration wastewater were significantly higher than other groups ( $P < 0.05$ ). The concentration of heavy metals (Pb, Cd, As and Hg) in water spinach in all groups was far below the national food safety standard. In general, the water spinach floating bed is suitable for the purification of medium and low concentration wastewater, and the optimal coverage rate is 40%~60%.

**Keywords:** wastewater concentration, coverage rate, water spinach, plant floating bed

## Introduction

In 2020, the annual slaughter number of geese was about 740 million worldwide, with an increase of about

17.5% compare with that in 2019, and Asia accounts for about 96% of total amount and China possess the largest number of geese sold in Asia [1]. Nowadays, the expansion of the scale of geese breeding, coupled with the physiological habits of playing with water and washing water for cleaning house, leading to a large increase in breeding wastewater [2]. Wastewater from livestock and poultry breeding contains large amounts

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of nutrients, such as phosphorus, nitrogen, potassium and so on, which cannot be directly discharged and used in agriculture. The feces and urine of geese are alkaline, and the concentration of pollutants are different from those of livestock and other poultry. The wastewater treatment method of geese farm in the early stage is roughly the same as that of other livestock and poultry farms, and there may be some differences in the further purification treatment of biogas slurry after subsequent anaerobic fermentation. However, there are few researches concentrating on the treatment of wastewater from geese farm, especially the follow-up biogas slurry treatment [3]. In addition, many traditional treatment methods are difficult to be applied in practice because of high cost and low efficiency [4, 5]. Therefore, it will be great meaningful and valuable to explore how to treat wastewater of geese farm economically and efficiently.

As an innovative wastewater treatment technology, plant floating bed system consists of aquatic or terrestrial plants growing in a hydroponic manner with buoyant frames floating on the surface of wastewaters [6-8]. Plant floating island system is usually used to treat lakes and rivers water, which has the characteristics of low cost, convenient operation and no secondary pollution [9]. The key role of plants as part of the natural processes which remove nutrients and other pollutants from the wastewater. Many researchers also have proven that plant floating island could effectively reduce the content of nitrogen, phosphorus and other nutrients from wastewater [10-13]. Previous researchers compared the purification performances of plant floating bed for nitrogen and phosphorus removal under a condition with a macrophyte coverage rate of 12.8%, and the results showed that the plant floating bed system combined with calamus seedlings can decline the TN, TP, and  $\text{NH}_4^+\text{-N}$  of eutrophic water in summer, and the highest removal rate is 36.3%, 35.7% and 44.3% respectively [14]. The average removal rates in an integrated floating island system for TN,  $\text{NH}_4^+\text{-N}$ , TP in summer and autumn season were 36.9%, 44.8% and 43.3% respectively [15].

Water spinach is a vining plant, consumed as a leafy vegetable with high tolerance to heat and wet [16, 17]. It likes fertilizers and water, has strong regeneration ability, short picking cycle and long growth cycle spanning spring, summer and autumn. Researchers

have found that water spinach, as a floating bed plant, has a good effect on the purification of fishpond water, with the removal rates of total nitrogen and total phosphorus were 27.07%~69.84% and 48.48%~76.67% respectively [18, 19], which can fully recycle the aquaculture wastewater. In addition, water spinach is one of the main edible vegetables in the south region of middle and lower reaches of the Yangtze River. As we know, geese are herbivorous birds, which have a great demand for pasture in geese farm. Therefore, it also can be beneficial to harvest water spinach in floating bed system for feed of geese. At present, there are few studies on the wastewater purification of geese farm, especially on the appropriate related parameters, such as the planting density of floating bed plants and the tolerance to wastewater with different concentrations. To investigate the effect of the water spinach floating bed system on the pollutants in the wastewater of geese farm, the present study was carried out on geese farm for 40 days. In this work, the concentration of pollutants in wastewater and growth of water spinach was monitored to find optimal plant coverage and wastewater concentrations.

## Materials and Methods

### Experimental Materials and Floating Bed Design

The experiment was performed in the poultry scientific research base of Chongqing Academy of Animal Science. The wastewater was obtained from biogas liquid after precipitation in the base, and the average concentration of pollutants are shown in Table 1. The water spinach was planted by the base. The water spinach grew in the fields for 20 days until they reached the ideal size to be cut into segments and transferred to the defined location of the experiment.

The self-made floating bed system was composed of double polyethylene net (1×1 cm) and PVC tube, sized with 70×55×5 cm, 64×45×5 cm and 48×40×5 cm horizontal and vertical. The experiment was carried out in plastic cartons, sized with 80×60×50 cm horizontal and vertical.

Table 1. The initial concentration of nutrients in geese wastewater<sup>1</sup>.

WCON <sup>2</sup>	COD (mg/L)	BOD <sub>5</sub> (mg/L)	$\text{NH}_4^+\text{-N}$ (mg/L)	TP (mg/L)	TN (mg/L)	NTU
Low	110.00±0.32	28.65±0.49	47.05±1.07	6.60±0.06	56.35±0.26	40.23±0.75
Medium	170.50±0.87	32.90±0.81	77.75±0.43	10.05±0.03	97.55±9.50	68.55±1.30
High	338.01±2.31	66.65±0.61	152.00±2.31	20.15±0.32	207.00±9.81	137.13±1.08

<sup>1</sup>Data are shown as mean ± SEM and each mean represent 3 samples;

<sup>2</sup>means concentration of wastewater (high/medium/Low- 3/6/9 times diluent from biogas).

### Testing Design

The water spinach segments were rinsed with tap water, then divided into homogenous groups and acclimatized for 5 days in the same environmental conditions. Three plants with different weight (60 g, 45g and 30 g,) and quantities (48, 36 and 24) were respectively planted into the floating bed at 80%, 60% and 40% coverage, then put into plastic cartons filled with biogas liquid dilution by 3, 6 and 9 times respectively. A total of 9 treatment groups were set up in the experiment according to the wastewater concentration and plant coverage, each of 3 replicates. The information of group was as following: (1) L-CON: low-concentration wastewater without plant; (2) L-40%: low-concentration wastewater with 40% coverage plant; (3) L-60%: low-concentration wastewater with 60% coverage plant; (4) L-80%: low-concentration wastewater with 80% coverage plant; (5) M-CON: medium-concentration wastewater without plant; (6) M-40%: medium-concentration wastewater with 40% coverage plant; (7) M-60%: medium-concentration wastewater with 60% coverage plant; (8) M-80%: medium-concentration wastewater with 80% coverage plant; (9) H-CON: high-concentration wastewater without plant; (10) H-40%: high-concentration wastewater with 40% coverage plant; (11) H-60%: high-concentration wastewater with 60% coverage plant; (12) H-80%: high-concentration wastewater with 80% coverage plant. The experiment was initiated from August 2019 to September 2019 and lasted for 40 days. During the experiment period, the temperature ranged from 20.1°C to 34.2°C and the mean temperature was approximately 27.4°C.

### Pollutants Determination

At day 0 and 30, water samples were collected to detect the concentration of pollutants which were composed of TN, TP, turbidity,  $\text{NH}_4^+\text{-N}$ , 5-day biochemical oxygen demand ( $\text{BOD}_5$ ), chemical oxygen demand (COD) and heavy metal (Hg, As, Zn, Cr, Cd, Se, Cu, and Pb). Plant growth parameters (plant height, root length and relative growth rate), nutritional parameters (nitrogen and phosphorus content) and safety indicators (Pb, Cd, As, Hg and  $\text{NaNO}_2$ ) were also measured to monitor the growth of plants. The pollutants from wastewater were measured according to the standard methods for water and wastewater monitoring and analysis (APHA, 2005), And the relative indexes in plant tissues were determined according to previous methods [20, 21].

Plants were washed with tap water at the start and end of the experiment, and the average fresh plant weight was measured. The relative growth rate (RGR) was calculated as:

$$\text{RGR (\%)} = 100 \times (W_e - W_i)/W_i$$

...where  $W_e$  is the average fresh weight of plants at the end of experiments,  $W_i$  is the average initial fresh weight of plants.

The removal rate (RR) of pollutants in this study was calculated as:

$$\text{RR (\%)} = 100 \times (C_i - C_f)/C_i$$

...where  $C_f$  is the average concentration of pollutants after treatment,  $C_i$  is the average initial concentration of pollutants.

### Statistical Analysis

All data calculation as averages and standard error of mean (SEM) were performed in Microsoft Excel 2016. Statistical analyses were carried out using Analysis of variance (ANOVA) in Software SPSS 20.0 (SPSS Inc., Chicago, IL, USA) including Bartlett's test for homogeneity of variances analysis and Duncan's test for differences between means. All statements of differences were based on a significance level of  $P < 0.05$ .

## Results

### Removal Efficiencies of Nutrients by Water Spinach Floating Bed System

The removal performances of plant floating bed system on nutrients in geese wastewater from geese farm are shown in Table 2. The water spinach floating bed has a significant effect on the removal of pollutants in wastewater compared with the control group. The wastewater concentration has significant influence on the removal rate of pollutants in wastewater by water spinach floating bed system ( $P < 0.05$ ). As the growth time of floating bed plants goes on, the pollutants in wastewater were decreased. The removal rate of COD and  $\text{BOD}_5$  in low-concentration wastewater was significantly lower than medium and high concentration wastewater ( $P < 0.05$ ), which were 51.41%, 59.99%, 59.27% and 58.72%, 58.94%, 59.09%, respectively. There was no significant effect on the removal of pollutants between medium and high concentration wastewater ( $P > 0.05$ ). The removal effect of  $\text{NH}_4^+\text{-N}$  in high concentration wastewater is better than other groups ( $P < 0.05$ ), which were 99.93%, 99.84% and 99.70%, respectively. For total phosphorus and total nitrogen, the removal rate decreased significantly with the decrease of wastewater concentration ( $P < 0.05$ ), and the removal rate in high concentration wastewater was the highest in our studies, which were 63.47, 61.67%, 43.47% and 80.37%, 74.59%, 57.74% respectively. The removal effect of the turbidity in low and medium concentration wastewater were higher than that in high concentration wastewater ( $P < 0.05$ ), which were 75.77%, 77.29% and 65.60%, respectively. In general,

Table 2. Influence of concentration and plant coverage rate on pollutant removal rate in wastewater (%)<sup>1</sup>.

WCON <sup>2</sup>	High			Medium			Low			SEM <sup>4</sup>	P <sup>5</sup>				
	Control	40%	60%	80%	Control	40%	60%	80%	Control		40%	60%	80%	WCON	PCOV
PCOV <sup>3</sup>	37.45	60.06	55.62	62.13	34.51	62.66	58.94	58.36	41.39	52.73	50.30	51.21	0.08	0.00	0.04
COD	40.85	57.94	58.90	60.44	33.38	62.52	57.75	56.54	46.09	59.47	57.97	58.72	0.08	0.00	0.09
BOD <sub>5</sub>	83.78	99.93	99.92	99.94	70.35	99.85	99.83	99.85	89.36	99.77	99.73	99.61	0.00	0.00	0.00
NH <sub>4</sub> <sup>+</sup> -N	36.78	65.91	60.00	64.50	11.69	65.87	56.82	62.32	13.84	42.17	37.53	50.71	0.02	0.00	0.26
TP	54.53	81.82	78.66	80.63	51.37	78.54	74.20	71.02	43.67	59.06	54.21	59.95	0.01	0.00	0.19
TN	21.12	62.59	66.43	67.78	67.40	71.96	77.67	82.25	64.65	72.94	76.17	78.21	0.01	0.00	0.00

<sup>1</sup>Data are shown as mean and each mean represent 3 samples;

<sup>2</sup>means concentration of wastewater (high/medium /Low- 3/6/9 times diluent from biogas);

<sup>3</sup>plant coverage rate;

<sup>4</sup>means standard error of mean;

<sup>5</sup> WCON/ PCOV: mean main effects of water concentration/plant coverage rate; WCON\* PCOV: means interaction of water concentration and plant coverage rate.

Table 3. The heavy metals content in initial and treated wastewater<sup>1</sup>

WCON <sup>2</sup>	Initial value	High			Medium			Low								
		Control	40%	60%	80%	Initial value	Control	40%	60%	80%	Initial value	Control	40%	60%	80%	
PCOV <sup>3</sup>	2.35	1.75	0.75	0.83	0.88	1.35	1.30	1.05	1.30	1.35	1.30	1.35	1.30	0.83	0.73	0.91
Hg(μg/L)	0.85	0.55	0.40	0.40	0.40	0.60	0.40	0.30	0.30	0.50	0.30	0.30	0.30	-	-	-
As(μg/L)	0.10	-	-	-	-	0.07	-	-	-	0.05	-	-	-	-	-	-

<sup>1</sup>Data are shown as mean and each mean represent 3 samples;

<sup>2</sup> means concentration of wastewater (high/medium /Low- 3/6/9 times diluent from biogas);

<sup>3</sup> plant coverage rate.

Table 4 The heavy metals and nitrites contents of water spinach in each treatment group<sup>1</sup>.

WCON <sup>2</sup>	High			Medium			Low		
	40%	60%	80%	40%	60%	80%	40%	60%	80%
PCOV <sup>3</sup>									
Pb(mg/kg)		-	0.027	-	-	0.035	-	0.035	-
Cd(mg/kg)		-	0.002	0.002	-	-	-	-	-
Hg(mg/kg)		-	-	-	-	-	-	-	-
As(mg/kg)		0.016	0.013	0.017	0.012	0.014	0.016	0.012	0.013
NaNO <sub>2</sub> (mg/kg)		10.0	20.0	-	2.5	2.2	-	1.0	1.0

<sup>1</sup> Data are shown as mean and each mean represent 3 samples;

<sup>2</sup> means concentration of wastewater (high/medium /Low- 3/6/9 times diluent from biogas);

<sup>3</sup> plant coverage rate.

the removal effect of pollutants reached the best in high concentration wastewater.

The coverage rates of water spinach had significant effects on the removal of COD, BOD<sub>5</sub>, NH<sub>4</sub><sup>+</sup>-N, TP and TN in wastewater (P<0.05). There was no significant difference on the removal of nutrients among the treatment groups (P>0.05). The removal effect of the turbidity with 80% and 60% coverage rates were higher than that with 40% coverage rates (P<0.05), which were 76.08%, 73.42% and 69.16%, respectively. On the whole, the coverage rate of water spinach should be 40%~60% and the planting cost can be reduced.

In our studies, there was a synergistic effect between the two factors on the removal effect of COD, NH<sub>4</sub><sup>+</sup>-N and NTU (P<0.05), while there was no synergistic effect on TP and TN.

#### Removal Efficiencies of Heavy Metal in Wastewater by Water Spinach Floating Bed System

The heavy metal content in wastewater before and after the experiment are shown in Table 3. Some important heavy metals (Hg, As, Zn, Cr, Cd, Se, Cu and Pb) were measured, just three heavy metals (Hg, As and Zn) were found in wastewater. The floating bed system of water spinach reduced the concentration of heavy metals (Hg, As and Zn) in wastewater with different concentrations, which are 22.2%~68.2%, 40.0%~52.94% and 100.0% respectively. There was not obvious relation between the removal effect and the plant coverage rate. The removal effect is mainly affected by wastewater concentration.

#### Effects on Plant Growth

Wastewater concentration and plant coverage rate had significant effects on root length, stem height, weight gain and growth rate of water spinach, and the results were showed in Table 6. Under the coverage rates of 40% and 80%, the stem height, weight gain and growth rate of water spinach were inversely

proportional to the wastewater concentration (P<0.05). At 60% coverage rate, which of low concentration groups was higher than medium concentration and high concentration groups (P<0.05).

There were no significant differences in stem height, root length, weight gain and growth rate of water spinach among different coverage rates (40%, 60% and 80%) in high concentration wastewater (P>0.05). In medium concentration wastewater, the stem height with the coverage rate of 60% is the best, followed by 80% and the last 40% (P<0.05). Weight gain with 60% and 80% coverage rates were higher than 40% (P<0.05). The growth rate of water spinach with 80% coverage rate were higher than 60% and 40% (P<0.05). There was no significant difference in root length among different coverage rates. In low concentration wastewater, the stem height with the 40% coverage rate was higher than 60% (P<0.05), root length with the 40% coverage rate was higher than 60% and 80% (P<0.05), Weight gain with 80% coverage rate were higher than 60% and 40% (P<0.05), The growth rate of water spinach with 60% and 80% coverage rates were higher than 40% (P<0.05). For stem height, root length, weight gain and growth rate, there was a significant interaction between biogas slurry dilution ratio and plant coverage rate (P<0.05).

The influence of water concentration and plant coverage rate on nutrient content of water spinach are shown in Table 7. Wastewater concentration has a significant effect on the TN and TP contents of water spinach stem leaf and root but has no significant effect on water content. The growth of water spinach in high concentration wastewater was slow and some died, which led to insufficient samples in the 40% and 60% coverage groups at the end of the experiment and no measurement was carried out. The TN and TP content of water spinach stem leaf in high concentration wastewater were significantly higher than those in medium and low concentration wastewater (P<0.05). The TN and TP content in root is proportional to the wastewater concentration. Coverage rate had no significant effect on TN and TP content in stem leaf of

Table 5. The heavy metals and nitrites content standards in vegetables.

mg/kg	Standard	Pollution-free	Green vegetables	Detection limit
Pb	0.3	0.2	0.1	0.02
Cd	0.2	0.05	0.05	0.001
Hg	0.01	0.01	0.01	0.003
As	0.5	0.5	0.2	0.01
NaNO <sub>2</sub>	1	4	1	1

water spinach but had effect on TN and TP content in root. The TN content of 80% coverage rate is higher than 60%, and the TP content is higher than 60% and 40%. Overall, there was no significant interaction between wastewater concentration and plant coverage rate on moisture content, TN and TP content of stem leaf ( $P>0.05$ ).

In addition, this experiment measured the moisture content and TN and TP content (88.07%, 2.11 mg/kg, 0.50 mg/kg and 87.14%, 1.97 mg/kg, 0.41 mg/kg) in the stem leaf and root of water spinach cultivated in soil (it grows for about 60 days), and found that the moisture content and TN content were significantly lower than that in hydroponic culture (90.05%, 3.41 mg/kg, 0.47 mg/kg and 88.56%, 3.47 mg/kg, 0.41 mg/kg) ( $P<0.05$ ).

### Water Spinach Safety Assessment

The heavy metal and nitrite content in water spinach were determined and shown in Table 4. The content standards of heavy metals and nitrites in vegetables and the corresponding minimum detection limits in this experiment are shown in Table 5. The concentration of several heavy metals (Pb, Cd and As) in water spinach in all groups is far below the national food safety standard and Hg was not detected at 40 day. The results showed that the higher the concentration of wastewater, the higher the nitrite content of water spinach. Therefore, water spinach cultured in high concentration wastewater cannot meet the safety standards of edible vegetables in China.

## Discussion

### The Nutrients Removal by Water Spinach Floating Bed System

The plant floating system could form a biofilm with a large surface area for the purification of water by both epiphyte and microbes [20]. The epiphyte could contact wastewater to absorb, transform and degrade the pollutants, and secrete large amount of enzyme and organic acid to accelerate the decomposition of the macromolecular pollutants and improve the bioavailability of nitrogen and phosphorus in

wastewater [21, 22]. Meanwhile, plant floating system will bring various bacteria involved in pollutants degradation and removal into wastewater [23]. In the present study, the plant floating bed system with different coverage rates could significantly improve the removal rates of COD, BOD<sub>5</sub>, NH<sub>4</sub><sup>+</sup>-N, TP, TN and turbidity in different concentrations wastewater compare with control in winter. This was similar to previous studies on the purification of rivers by plant floating bed systems, which can effectively remove pollutants from the water [24-26].

In this experiment, the removal effect of water spinach floating bed in high-concentration wastewater was better than that in low-concentration wastewater, possibly because the more nutrients in the high concentration wastewater were absorbed by plants [27, 28]. In addition, there is a problem that the plants in a long-term over-nutrition environment will lead to water loss and even death, which indicates that it is not necessarily proportional between decontamination effect and plant growth. It is also closely related to the species and physiological characteristics of plant. In our study, there was no significant difference in the removal effect of many pollutants (COD, BOD<sub>5</sub>, NH<sub>4</sub><sup>+</sup>-N, TP, TN) among floating beds of water spinach with different coverage rates, which may be related to the planting scale of floating bed plants and the amount of wastewater. Our study proved that the coverage rate is proportional to the turbidity removal rate and consistent with the former study, the removal rates of turbidity with 80% plant coverage rate by the water spinach floating bed system (76.05%) was more than 1.05 times of that by 40% (69.16%) and 60% (73.42%).

The floating bed plants are capable of absorbing, metabolizing and enriching heavy metals, and the content of heavy metals within the plants increased as the reduction of the water's pollution level, so plant floating bed system was regarded as an efficient way of reducing heavy metal pollution in wastewater [29-31]. The contents of heavy metals in the wastewater of each treatment group were reduced by 23%~68% of Hg, 32.59%~56.25% of As and 100% of Zn, respectively, indicating that water spinach floating bed can effectively reduce heavy metals in wastewater. In practical application, water spinach can continuously remove the heavy metal from wastewater because of its long growth cycle to avoid the environmental and land

Table 6. Influence of concentration and plant coverage rate on the growth of water spinach<sup>1</sup>.

WCON <sup>2</sup>	High			Medium			Low			SEM <sup>4</sup>	P <sup>5</sup>		
	40%	60%	80%	40%	60%	80%	40%	60%	80%		WCON	PCOV	WCON *PCOV
PCOV <sup>3</sup>	8.33	7.77	11.11	12.89	13.89	13.11	16.55	15.22	15.22	0.37	0.00	0.09	0.00
Root length	21.97	22.44	21.89	26.44	35.56	30.78	35.44	31.11	33.45	0.82	0.00	0.13	0.00
Stem height	24.03	32.66	20.33	110.83	170.33	162.33	186.50	182.00	251.33	10.74	0.00	0.01	0.01
Growth rate	48.07	43.56	20.33	221.67	227.11	162.33	373.00	242.67	251.33	13.79	0.00	0.00	0.01

<sup>1</sup> Data are shown as mean and each mean represent 3 samples;

<sup>2</sup> means concentration of wastewater (high/medium /Low- 3/6/9 times diluent from biogas);

<sup>3</sup> plant coverage rate;

<sup>4</sup> means standard error of mean;

<sup>5</sup> WCON/ PCOV: mean main effects of water concentration/plant coverage rate; WCON\* PCOV: means interaction of water concentration and plant coverage rate.

Table 7. Influence of concentration and plant coverage rate on the nutrient content of water spinach<sup>1</sup>.

WCON <sup>2</sup>	PCOV <sup>3</sup>	Soil culture	Water culture									SEM <sup>4</sup>	P <sup>5</sup>		
			High			Medium			Low				WCON	PCOV	WCON * PCOV
			40%	60%	80%	40%	60%	80%	40%	60%	80%				
moisture content (%) stem leaf		88.07	-	-	89.56	80%	89.97	90.38	90.36	89.93	0.32	0.54	0.65	0.75	
root		87.14	-	-	88.75	80%	88.46	88.32	88.75	88.75	0.14	0.29	0.39	0.43	
N(mg/kg) stem leaf		2.11	-	-	4.34	3.39	3.25	3.31	3.13	3.08	0.03	0.00	0.47	0.86	
root		1.96	-	-	4.05	3.50	3.58	3.48	3.41	3.31	0.29	0.00	0.01	0.02	
P(mg/kg) stem leaf		0.50	-	-	0.60	0.46	0.45	0.47	0.44	0.41	0.06	0.00	0.86	0.50	
root		0.41	-	-	0.52	0.40	0.40	0.40	0.37	0.38	0.01	0.00	0.88	0.90	

<sup>1</sup> Data are shown as mean and each mean represent 3 samples;

<sup>2</sup> means concentration of wastewater (high/medium /Low- 3/6/9 times diluent from biogas);

<sup>3</sup> plant coverage rate;

<sup>4</sup> means standard error of mean;

<sup>5</sup> WCON/ PCOV: mean main effects of water concentration/plant coverage rate; WCON\* PCOV: means interaction of water concentration and plant coverage rate.

pollution caused by excessive content of heavy metals in wastewater after long-term deposition.

### Effects on the Growth and Security of Water Spinach

The efficiency of pollutants removal by the plant floating bed systems depends upon plant biomass production and pollutants concentrations in wastewater [29, 30]. In this experiment, the water spinach in high concentration wastewater grew slowly and some plants even died in the interim and late stage of the experiment, which indicated that the high concentration wastewater would inhibit the growth of plants. This is consistent with previous research [31]. The average stem height and root length of water spinach in low concentration group were 1.08 and 1.51 times and 1.18 and 1.73 times of those in medium and high concentration groups, respectively. The average weight gain and growth rate of water spinach in low concentration were 1.95 and 8.05 times and 1.40 and 7.74 times of those in medium and high concentration group. As a crucial factor, the concentration of wastewater played an important role in plant growth [28], and low concentration wastewater was more suitable for plant growth in floating boat system. The water spinach with 40% coverage grew best in low concentration wastewater, and water spinach with 60% and 80% coverage was better in medium concentration wastewater. This may be because the nutrients in low concentration wastewater could not meet the need of long-term growth of water spinach with high coverage rate, and there was a competitive mechanism among plants, while the medium concentration could fully meet the nutritional requirements. Therefore, wastewater concentration and plant coverage rate have a co-regulation effect on plant growth.

Food safety has always been the most concerned issue for consumers. As a common vegetable, it is essential to conduct safety tests of water spinach produced by hydroponics in this experiment to avoid excessive content of heavy metals and other harmful substances to human and animals [32]. The heavy metal content of water spinach in this experiment is far lower than the industry standard, and the contents of Pb, Cd and As were 0.175, 0.046 and 0.032 times of the standard, respectively. The floating bed water spinach growing in the low concentration wastewater can reach the standard of green vegetable, and the water spinach growing in the middle concentration wastewater can reach the standard of pollution-free vegetable. The nitrite content of water spinach was proportional to the concentration wastewater, which also indicated that high concentration wastewater could not be directly used for hydroponic water spinach. In addition, water spinach belongs to a vine, with shallow root distribution, beard root and strong regeneration ability. In the practice, the harvesting cycle of stem and leaves can

be shortened to about 20 days to reduce the deposition of heavy metals [33]. Meanwhile, the hydroponic fluid should be renewed timely to avoid excessive deposition of nitrite to ensure the food safety.

As a green leaf vegetable, water spinach is rich in nutrients, such as nitrogen and phosphorus [34]. In our study, the water content and total nitrogen content in stem leaf and root of water spinach in hydroponic culture were significantly higher than those in soil. This is probably because water spinach in hydroponic culture has more developed roots than that in soil, it is easier to absorb nutrients from wastewater [35]. The content of TP in stem, leaf and root of water cabbage in medium and low concentrations were slightly lower than those in soil, which might be related to the TP content in soil and growth cycle. Even if the wastewater concentration is proportional to the content of nitrogen and phosphorus in water spinach, there was no direct relationship between the wastewater concentration and the nutritional quality of water spinach. The wastewater concentration is high that plants can't survive and produce large amounts of nitrites, which are harmful to humans.

### Conclusion

The water spinach floating bed system had a better removal effect on pollutants in high concentration wastewater from geese farm compared to medium and low concentration groups, while the growth of part plants was inhibited or even dead in late stage. Low concentration wastewater was more beneficial for the growth of water spinach and the yield was the highest among three concentration wastewaters. The water spinach with 40% coverage rate in low concentration wastewater has highest growth rate in all groups. The water spinach cultured in low concentration wastewater reached the standard of green vegetable, and those cultured in medium concentration wastewater reached the standard of pollution-free vegetable.

In general, the water spinach floating bed is suitable for the application of medium and low concentration wastewater, and the optimal coverage rate is 40%~60%.

### Acknowledgments

This study was supported by China Agriculture Research System of MOF and MARA (CARS-42-22), and the Key R & D Project in Agriculture and Animal Husbandry of Rongchang (cstc2019ngzx0017).

### Conflict of Interest

The authors declare no conflict of interest.

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