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## Construction of Water-saving Ecological Aquaculture Model in Ponds in North China

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Abstract [Objectives] To explore the ecological aquaculture model in ponds in North China, several single techniques were assembled into the same culture system. [Methods] Three ponds were selected, the species and stocking rate were exactly the same, the water was not changed during the culture period, and the water loss due to evaporation and leakage was recovered. Since the middle of May, the hydrochemical indicators such as ammonia nitrogen, nitrite nitrogen, water temperature, dissolved oxygen and pH were monitored every 10 d. According to the monitoring results of ammonia nitrogen, carbon sources were added to the culture pends to adjust the ratio of C to N, and carbon sources were added 9 times during the culture period. The stocking rate and yield per unit area were accurately measured at the beginning and end of the experiment, and no less than 30 fishes were randomly sampled to calculate the relevant growth indicators and feed coefficients. [Results] Except that the nonionic ammonia in pond 3# exceeded the standard by 10.3% on July 25, all other hydrochemical indicators met the Fisheries Water Quality Standard, and there was no significant difference in all hydrochemical indicators at the same time (P>0.05). The survival rate in 3 ponds was more than 95.0%, the average body weight of individuals out of the pond had no difference (P>0.05), and the feed coefficient was 1.41 – 1.43. There was no disease during the culture period, and the water was saved by 46.6% compared with the traditional culture model. [Conclusions] This study can provide a basis for the construction of a new model suitable for ecological aquaculture in ponds in North China. Key words Ponds in North China, Water Saving, Ecology, Model, Construction

#### 1 Introduction

uaculture.

Over the past 40 years of reform and opening up, aquaculture has developed rapidly, in which freshwater pond culture plays an important role in aquaculture<sup>[1]</sup>. However, due to the excessive pursuit of profit maximization, the traditional aquaculture industry makes excessive use of land and water resources and increases the use of inputs while increasing the output. High density and high input lead to frequent diseases, deterioration of aquaculture water quality, and environmental pollution of water caused by frequent water exchange. Since the 18th CPC National Congress, the national fishery system has taken accelerating the green development of fishery as the fishery center, vigorously developing healthy aquaculture, and the mode of industrial development is undergoing revolutionary changes<sup>[2]</sup>. Therefore, changing the traditional extensive production mode and optimizing and adjusting the pond green ecological aquaculture model is the fundamental way to solve the problem of pond culture<sup>[3]</sup>. At present, all provinces and cities in the south are vigorously promoting new culture models such as container fish culture<sup>[4]</sup>, pond circulating water fish culture<sup>[5]</sup>, fish and vegetable symbiosis [6]. The northern region is limited by climatic conditions and natural environmental conditions and other factors, so it is difficult to popularize and apply many new models.

Therefore, it is necessary to explore an efficient, simple and easy-to-popularize pond green ecological aquaculture model.

Since the implementation of the national bulk freshwater fish industry technology system construction project in 2018, a number of new pond culture technologies have been studied and established to meet the needs of healthy culture<sup>[7]</sup>. A number of technologies have been demonstrated and popularized, such as immune prevention and control of two-year-old grass carp, biological floculation technology to regulate and control water quality, accurate feeding, comprehensive disease prevention and control, micropore oxygen enrichment in feeding areas. The purpose of this study is to integrate the above technologies into the same culture system to construct a new model suitable for ecological aquaculture in ponds in North China, which has the "ecological, clean and accurate" characteristics, so as to lay a foundation for the rapid development of freshwater aquaculture.

#### 2 Materials and methods

**2.1 Test materials** The test time is from May 6 to October 14, 2021, and the test site is Jinyuan Aquatic Farm, Changyi District, Jilin City. There are three experimental ponds (pond 1#, pond 2#, pond 3#), the area is 10 667 m², the depth of the pond is 3.2 m, the maximum depth of water is 2.8 m, and the source is groundwater. The fries of two-year-old grass carp and two-year-old *C. carpio* 2 were cultivated on this farm, and the average body weight was 164.747 2 and 147.336 6 g, respectively. The fries of *P. fulvidraco* were purchased locally, with an average body weight of 19.782 g. Silver carp and bighead carp were introduced from South China, and the average body weight was 0.210 07 and

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0.260 06 g, respectively. Sucrose molasses was selected as the carbon source, and the content of active constituent was 48%. Quadruple vaccine was selected as grass carp vaccine (enteritis, gill rot, red skin disease, hemorrhagic disease). The oxygen enrichment equipment was the combination of microporous aeration equipment (Roots blower + microporous aeration plate) and traditional aeration equipment (paddle wheel aerator). Positive pressure 360° air-assisted feeding machine was selected as feeding equipment.

**2.2 Experimental design** The experiment was conducted with two-year-old grass carp as the main fish for culture, combined with the fries of carp, *P. fulvidraco*, silver carp and bighead carp for mixed culture. The stocking situation of fries in 3 experimental ponds was exactly the same, and the stocking quantity and specifications are shown in Table 1.

Table 1 Stocking of experimental fish

Fish species	Specifications // g	Quantity//pc/667 m <sup>2</sup>
Grass carp	$164.70 \pm 47.20$	480
C. carpio 2	$147.30 \pm 36.60$	200
P. fulvidraco	$19.70 \pm 8.20$	150
Silver carp	$0.21 \pm 0.07$	1 200
Bighead carp	$0.26 \pm 0.06$	800

Two-year-old grass carp were released for breeding after injection of quadruple vaccine, and the vaccine was injected in the same pond within the same day. Bleach or chlorine dioxide was used to disinfect the pond water on the same day or the next day after vaccination. From the middle of May, the physical and chemical indicators such as ammonia nitrogen ( $NH_4^{\,+}-N)$ , nitrite nitrogen ( $NO_2^{\,-}-N)$ , dissolved oxygen (DO) and pH were monitored once a week. According to the results of ammonia nitrogen determination, carbon sources were added every 10 d on June 5, and the carbon source addition was calculated as follows  $^{[8]}$ :

$$\Delta_{\it CH} = 20 \times H \times S \times C_{\it ammonia nitrogen}$$
 (1) where  $\Delta_{\it CH}$ —carbon source (molasses) addition, g;  $\it H$ —culture pond water depth, m;  $\it S$ —water area, m²;  $\it C_{\it ammonia nitrogen}$ —NH<sub>4</sub><sup>+</sup>-N concentration in water, mg/L.

The water level was about 1.5 m before the middle of June, and 2.5 - 2.8 m from the middle of June to September 20. After September 20, the water level gradually lowered with the drop of temperature, in preparation for coming out of the pond. There was no change of water during the whole test period, only the water lost due to evaporation and leakage was added. 100 m<sup>2</sup> microporous aeration facility was installed under the feeding table, the microporous aeration equipment was turned on during feeding, to increase the dissolved oxygen in the feeding area, promote the feeding of fish and improve the feed utilization rate. Feed nutrition and feeding were mainly for grass carp. According to the principle of accurate feeding, the feeding characteristics, living habits and metabolic rules of grass carp were studied to determine the feed nutrition, grain size, feeding frequency, feeding amount and so on. When the average body weight was lower than 250 g, the feed protein content was 29%, it was fed 4 times a day, and the grain size of feed was 3.5 mm. When the average body weight was more than  $250~\rm g$ , the feed protein content was 27%, it was fed 4 times a day, and the grain size of feed was  $4.0-6.0~\rm mm$ . The feeding amount should be flexibly controlled according to the weather and the intake of fish, until most of the fishes were full.

#### 2.3 Data monitoring

**2.3.1** Monitoring of hydrochemical indicators. The hydrochemical indicators were monitored three times a day, and the sample monitoring time was about 8:00, 12:00 and 16:00, respectively, taking the average.  $NH_4^+$ -N and  $NO_2^-$ -N (determined by ultraviolet spectrophotometer, Agilent Cary60). 3-5 sampling sites were selected in the pond feeding area, upper and lower air outlet, and 1 L water sample was collected at 0.5-1.0 m water level below the water surface. After the water samples were mixed uniformly at each sampling point, 1 L of samples were taken to monitor the relevant data. The monitoring sites of pH, DO and water temperature were the same as the above sampling sites (on-site monitoring by American YSIproplus multi-parameter water quality analyzer), and the average values of several sampling points were taken.

After monitoring, the NH<sub>3</sub><sup>+</sup>-N data were converted into nonionic ammonia (NH<sub>3</sub>-N). It was converted according to the percentage of nonionic ammonia in the aqueous solution of ammonia in Appendix A of the *Fisheries Water Quality Standard*<sup>[9]</sup> (GB11607-1989).

**2.3.2** Growth indicators monitoring. No less than 30 fishes were randomly selected during stocking and catching, the body weight of each fish was measured one by one, and the average body weight during stocking and catching was calculated. During the catching, all kinds of fish were weighed separately, the yield per unit area was calculated, and the catch was calculated according to the average body weight and yield per unit area. Each growth indicators was calculated according to formula (2) - (4):

$$N_t = W/W_t \tag{2}$$

$$SR(\%) = 100 \times N_{t}/N_{0}$$
 (3)

$$FCR = W_{feed}/(W_{yield} + W_{death} - W_{stocking})$$
 (4) where  $SR$  is the survival rate, %;  $FCR$  is the feed conversion rate;  $N_t$  and  $N_0$  are the number of fishes during catching and stocking, respectively;  $W$  and  $W_t$  are the yield per unit area and the average body weight at the end of the trial, respectively,  $g$ ;  $W_{feed}$ ,  $W_{yield}$ ,  $W_{stocking}$  and  $W_{death}$  are feed amount, total yield of carp and grass carp, total stocking amount of carp and grass carp, and total death amount during culture of carp and grass carp, respectively,  $g$ .

**2.4 Data analysis** The data were analyzed by Excel, and the data were expressed as mean  $\pm$  standard deviation (Mean  $\pm$  S. E.). The significance was tested by single factor analysis of variance (ANOVA).

#### 3 Results and analysis

**3. 1** Change of hydrochemical indicators In mid-May, hydrochemical indicators such as NH<sub>4</sub><sup>+</sup>-N (converted into NH<sub>3</sub>-N), NO<sub>2</sub><sup>-</sup>-N, DO and pH were monitored. In May, the water temperature of the ponds in North China was only about 15 °C, the amount of feed was small, the change of water quality was relatively sta-

ble, and all hydrochemical indicators were in the normal range (Table 3). In June, the water temperature gradually increased, the amount of feed increased, and the hydrochemical indicators

changed obviously. In order to maintain the relative stability of aquaculture water quality, carbon source was applied every 10 d for 9 times on June 5 (Table 2).

Table 2 Addition time and amount of carbon source (kg/667 m<sup>2</sup>)

Date (month/day)									
Pond	06 - 05	06 – 15	06 - 25	07 - 05	07 – 15	07 - 25	08 - 04	08 – 14	08 – 24
1#	15.9	18.2	13.6	18.8	17.0	20.1	19.0	32.5	33.9
2#	10.8	15.3	14.5	18.6	18.6	18.1	20.9	30.8	25.3
3#	12.2	12.0	15.7	16.9	20.5	19.8	17.5	23.5	23.7

0.15

0.10

0.05

NO<sub>2</sub>-N // mg/L

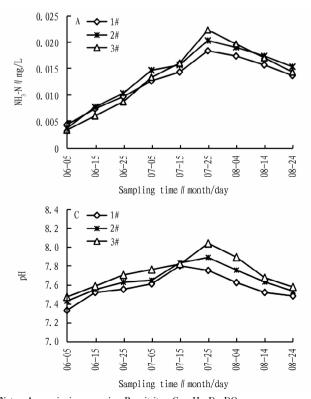
Fig. 1A reflects the changes of  $\mathrm{NH_3}\text{-N}$  in three experimental ponds, showing a trend of rising first and then descending. The content of  $\mathrm{NH_3}\text{-N}$  in all three ponds reached peak on July 25, and the ranges were 0.004 3 – 0.018 3, 0.003 7 – 0.020 3, 0.003 3 – 0.022 3 mg/L, respectively. Only the content of nonionic ammonia in pond 3 was 0.022 3 mg/L on July 25, exceeding the standard by 10.3%. After reaching the peak, nonionic ammonia decreased slowly. There was no significant difference in the change of  $\mathrm{NH_3}\text{-N}$  at the same time point (P > 0.05).

Fig. 1B reflects the change of  $NO_2^-$ -N in three experimental ponds, and the change trend of  $NO_2^-$ -N was consistent with that of  $NH_3$ -N. The peak value of  $NO_2^-$ -N appeared on July 25, which was 0.098 mg, and the peak value of pond 2#, 3# was 0.093 and 0.094 mg/L on July 15, respectively. The amplitude of variation of  $NO_2^-$ -N in 3 ponds was 0.033 – 0.098, 0.026 – 0.093 and 0.031 – 0.094 mg/L, respectively. There was no significant

difference in  $NO_2^-$ -N at the same time point (P > 0.05).

Fig. 1C reflects the change of pH in three ponds. The change rule of pH was completely the same, rising first and then decreasing, but the peak time was different. The peak value of pond 1# was 7.80 on July 15, and the peak value of pond 2#, 3# was 7.89 and 8.04 on July 25, respectively. After the peak value appeared in three ponds, the pH gradually decreased with the experiment. There was no significant difference in the change of pH in three ponds at the same time  $(\it{P}\,{>}\,0.05\,)$ .

Fig. 1D reflects the change of DO in three ponds. From the change curve, there was no obvious change rule, and the range of variation was 4.99-6.00, 5.38-6.11 and 5.27-5.99 mg/L, respectively, and the lowest value was 4.99 mg/L, which met the requirements of fishery water quality standards for dissolved oxygen in aquaculture water (close to or greater than 5.0 mg/L).



06 - 1506-25 07-05 07-15 07-25 08-04 08 - 1408 - 24Sampling time # month/day 7.00 - 2# 6.50 **△** 3# 6.00 DO // mg/L 5.50 5.00 4.50 4.00 06 - 1590-90 06-25 07 - 0508-24 Sampling time // month/day

Note: A. nonionic ammonia; B. nitrite; C. pH; D. DO.

Fig. 1 Change of hydrochemical indicators

**3.2** Growth indicators and feed utilization The growth indicators and feed utilization were shown in Table 2. There was no significant difference in individual average body weight of all kinds

of fish caught in 3 ponds (P > 0.05). Other growth indicators and feed coefficients were very similar.

Table 3 Growth indicators and feed utilization (667 m<sup>2</sup>)

Fish species	1#			2#			3#		
	$W_t /\!\!/ g$	SR // %	<i>W</i> ///kg	$W_{\iota}/\!/\mathrm{g}$	SR // %	<i>W</i> ///kg	$W_{\iota}/\!/\mathrm{g}$	SR // %	W//kg
Grass carp	1 710.4 ± 234.6 <sup>a</sup>	97.3	798.6	1 693.7 ± 219.2 <sup>a</sup>	96.7	785.9	1 675.6 ± 241.8 <sup>a</sup>	98.1	790.9
Carp	1 365.6 ± 254.6 <sup>a</sup>	97.0	264.9	$1\ 428.3 \pm 241.7^{a}$	95.5	272.8	1 328.8 ± 233.7 <sup>a</sup>	97.5	259.1
P. fulvidraco	96.4 ± 18.1 a	94.7	13.7	$95.8 \pm 16.4^{a}$	96.0	13.8	$98.3 \pm 19.2^{a}$	95.3	14.1
Silver carp	$93.7 \pm 12.3^{a}$	96.4	108.4	$90.8 \pm 14.1^{a}$	95.2	103.7	$94.8 \pm 12.6^{a}$	96.6	109.9
Bighead carp	$120.4 \pm 23.4^{a}$	95.3	91.8	$126.7 \pm 31.4^{a}$	96.2	97.5	$121.8 \pm 22.4^{a}$	97.2	94.7
Total			1 277.4			1 273.7			1 268.7
FCR			1.41			1.42			1.43

Note: Values with the same letter superscripts in the same line indicate that the difference is not significant (P > 0.05).

- 3.3 Water saving and emission reduction During the experiment, there was no change of water, fresh water was added 8 times in 3 ponds, the average fresh water (16.0 cm) was added each time, the total depth of fresh water was 1.28 m, and the amount of water added per 667 m² was 853.8 m³. In the traditional culture model in the northern region, the water was added and changed 12-13 times in the production season, with water of about 20.0 cm each time. The average annual water replacement per 667 m² was about 1600.0 m³ [10]. Compared with the traditional culture model, this culture model saved water 46.6% a year.
- **3.4** Occurrence of fish disease After the middle of July, due to the increase of water temperature, the large feeding amount and the deteriorating water quality of culture ponds, regular testing found that some carp and grass carp suffered from gill rot disease. Fresh water was added for 2 consecutive days and aeration equipment ran for a long time from July 26 to 27, and no disease was found in early August. During the whole culture period, except for regular inspection of fish disease and sampling, only a few sporadic dead individuals were found in each pond, and the fish was found to be rotten and the cause of death could not be examined. Only the survival rate of *P. fulvidraco* cultured in pond 1# was slightly lower than 95.0% (94.7%), and the survival rate of other fishes in pond 1# and all fishes in pond 2#, 3# was greater than 95.0%.

#### 4 Discussion

**4.1** Effects of ecological aquaculture on hydrochemical indicators 
Excessive  $NH_3$ -N and  $NO_2^-$ -N in high-density pond culture is a common problem in pond culture, which is difficult to degrade [11]. The main reason is that after a large amount of feeding, feed remnants and feces were decomposed to produce nitrogen-containing intermediates, most of which exist in the form of ammonia. Secondly, due to the lack of oxygen content in aquaculture water for a long time, ammonia could not be transformed into nitrite under the action of nitrifying bacteria, and the pH value and the concentration of  $NH_3$ -N and  $NO_2^-$ -N increased in aquaculture water. Excessive concentrations of  $NH_3$ -N and  $NO_2^-$ -N in aquaculture water would destroy gill tissue, reduce blood oxygen carrying capacity, make tissue lack oxygen, lose balance, and even die [12].

From the results of Fig. 1(A), except that the highest value

of  $\mathrm{NH_3}\text{-N}$  in pond 3# was 0.022 3 mg/L on July 25, the  $\mathrm{NH_3}\text{-N}$  values of other monitoring points in pond 1#, 2#, 3# were all within the range of 0.02 mg/L stipulated in *Chinese Fisheries Water Quality Standard* <sup>[9]</sup>. There was no significant difference in  $\mathrm{NH_3}\text{-N}$  values among the monitoring points in the three ponds (P>0.05). During the whole culture period, there was only one temporary over-standard point in pond 3#, and it only exceeded the standard by 10.3%, which would not cause any harm to cultured fish. From the results of Fig. 1(B), the highest value of  $\mathrm{NO_2}^-\text{-N}$  in the three test ponds was 0.098 mg/L, which would not harm the cultured fish <sup>[13]</sup>, and there was no significant difference among the three ponds at the same monitoring time point (P>0.05).

During the culture period, the NH<sub>3</sub>-N value was basically in the normal range, and the NO2-N value was in the normal range, which was mainly due to the joint action of several single technologies integrated into the same culture system. The combination of microporous aeration and traditional oxygen enrichment made the oxygen supplied in the upper and lower pond at the same time, and the three-dimensional oxygen supply of the pond was sufficient<sup>[7]</sup>. From the results of Fig. 1(D), the lowest oxygen content was 4.69 mg/L in the pond during the experiment period, resulting in rapid decomposition of organic matter, reducing the accumulation of NH<sub>3</sub>-N and NO<sub>2</sub>-N, and ensuring the healthy and stable operation of the pond ecosystem<sup>[14]</sup>. The application of biological flocculation technology was mainly for the rapid heterotrophic transformation of NH<sub>4</sub><sup>+</sup>-N. Through the addition of carbon source, it increased the ratio of C to N in culture water, and led to the rapid reproduction of heterotrophic bacteria, thus reducing the accumulation of  $NH_3$ -N and  $NO_2^-$ - $N^{[15]}$ .

From the results of Fig. 1(C), the change of pH was relatively stable, increasing slowly and decreasing slowly, and the amplitude of variation of pH in three ponds was 7.33 – 7.82, 7.43 – 7.89, 7.47 – 8.04, respectively. The reason for the stable pH change is that the formation of biological flocs should be carried out under alkaline conditions, requiring reduction of pH, while with the increase of temperature, photosynthesis of phytoplankton and aquatic plants converted  $HCO_3^-$  into  $CO_3^{2^-}$ , and pH increased  $^{[16]}$ . This decrease and increase made pH more stable and suitable for fish growth.

**4.2** Effects of ecological aquaculture on fish growth and feed utilization The growth of fish is affected by many factors. Under the condition of human culture, the main controllable factors are culture environment and feed nutrition [7]. In this experiment, ecological aquaculture was adopted, and many technologies such as biological flocculation technology, three-dimensional oxygen enrichment,  $360^{\circ}$  air-assisted feeding and precision feeding were integrated to promote the growth of fish and improve feed utilization rate. Biological flocculation technology could not only regulate the water quality of aquaculture, but also transform harmful substances such as  $NH_4^+$ -N and  $NO_2^-$ -N in aquaculture water into bacterial components, which not only reduced the content of  $NH_4^+$ -N and  $NO_2^-$ -N, but also flocculated into bacterial protein by bacteria, which was used by farmed fish to promote fish growth and reduce  $FCR_2^{[17]}$ .

Through 360° air-assisted feeding, the feed was distributed evenly, and the feeding area was large, which avoided the intense fish intake and physical energy consumption. The microporous aeration in the feeding area satisfied the feeding needs of fish under the condition of high oxygen environment, and the feed conversion rate was improved. According to the results of culture in Table 3, the average specification and total yield of all kinds of fish in three ponds were not lower than those under the traditional culture mode in North China. Through the precise feeding combination, the feeding amount, feeding frequency, feeding mode and nutritional requirements at each growth stage were effectively controlled, so that the feeding rate was close to the best intake rate under the condition of maximum benefit<sup>[18]</sup>, and the feed utilization rate was improved. The FCR was low, only 1.41 – 1.43, which was significantly lower than that under the traditional culture model<sup>[19]</sup>.

4.3 Effects of ecological aquaculture on water saving, emission reduction and fish diseases The biological immunity and disease prevention technology of two-year-old grass carp, the interspecific immunity technology and the technology of biological flocs to control water quality played a key role in the prevention and control of fish disease. Biological immunity was applied before stocking of two-year-old grass carp, and injection of "quadruple vaccine" of grass carp could prevent common diseases of grass carp, reduce the use of drugs in production, and improve the quality and safety of farmed aquatic products. The mixed culture of multi-species in the pond was based on the principle of "ecological complementarity and symbiosis" among species [20]. Without increasing the cost input, it made full use of bait resources and water space, which saved not only water but also bait. It is a model of "raising fish with water and controlling water with fish". The application of biological flocculation technology could not only reduce NH<sub>4</sub><sup>+</sup>-N and NO<sub>2</sub><sup>-</sup>-N, but also decompose organic pollutants, purify water quality and realize pollution-free culture. The application of the above technologies had an obvious effect on disease prevention of farmed fish, and there was no water change and disease during the culture period. The survival rate of all kinds of fish reached more than 95.0%, and various physical and chemical indicators of water quality basically met the Fisheries Water Quality Standard. Compared with the traditional aquaculture model, water was saved by 46.6%, and it made aquaculture ecological, clean and accurate, which was worth popularizing in North China.

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