

Original Research Articles

Effect of compound Chinese herbs on growth performance, digestion and serum biochemical indexes of Mandarin fish (*Siniperca chuatsi*)

Nan Li¹, Xiaodi Shang¹, Ziyuan Ding¹, Hongqing Sun¹, Mingze Li¹, Na Wang¹, Jian Wang¹, Lin Ma¹, Sudong Xia², Xin Zhang^{3a}

¹ Tianjin Fisheries Research Institute, Tianjin, China, ² Tianjin Agricultural University, Tianjin, China, ³ Tianjin Agricultural Development Service Center, Tianjin, China

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This study investigated the effects of compound Chinese herbs on the growth performance and serum biochemical indexes of *Siniperca chuatsi*. Using commercial finished feed as the basal diet for mandarin fish, a compound traditional Chinese herbal medicine consisting of hawthorn, angelica, and astragalus (in a ratio of 1:2:7) was added at 0%, 1%, and 2% of the basal diet by weight. The experimental diets were fed to fish with an initial body weight of 11.10 ± 0.53 g for a period of 42 days. The results indicated that the weight gain rate of the 1% and 2% groups significantly increased compared to the control group ($P < 0.05$). And the feed conversion ratio, viscera-somatic index, and hepatosomatic index were significantly decreased ($P < 0.05$). In terms of serum biochemical parameters, alanine aminotransferase (ALT) and aspartate aminotransferase (AST) levels were significantly reduced ($P < 0.05$), while high-density lipoprotein cholesterol (HDL-C) levels were significantly increased ($P < 0.05$) in both the 1% and 2% groups; in addition, triglycerides and total cholesterol levels were significantly decreased in the 2% group ($P < 0.05$). Regarding intestinal digestive enzymes, pepsin, trypsin, and lipase activities were significantly enhanced in the 1% and 2% herbal groups ($P < 0.05$). In summary, the addition of 1% and 2% of a compound herbal mixture of hawthorn, angelica, and astragalus (in a ratio of 1:2:7) to the feed can improve the growth performance, digestive ability, and antioxidant capacity of *Siniperca chuatsi*.

INTRODUCTION

Siniperca chuatsi, commonly known as mandarin fish or Chinese perch, belongs to the order Perciformes, family Serranidae, and genus *Siniperca*. It is one of China's "Four Famous Freshwater Fish."¹ Owing to its tender flesh, few bones, rich meat content, and delicious taste, it is highly favored by consumers. Moreover, its rapid growth rate and high economic value have driven the continuous expansion of its aquaculture scale. In 2023, the national aquaculture production reached 478,000 tons, with an output value exceeding 20 billion yuan, and the total value of the entire industry chain approaching 70 billion yuan. As a result, it has become one of the most important species in China's aquaculture industry.²

The mandarin fish exhibits a unique feeding habit, relying exclusively on live prey from when it begins feeding. However, using live bait presents several challenges, in-

cluding high costs, serious disease risks, and an unstable supply of baitfish, all of which significantly hinder the efficient and healthy development of the mandarin fish industry. In recent years, the domestication of mandarin fish and the use of formulated feeds have become key focuses for the sustainable growth of the industry. Farming with artificial feed can reduce environmental pollution, lower production costs, and support the development of intensive aquaculture. Currently, notable progress has been made in using artificial feed for mandarin fish. Still, due to the immaturity of domestication and feeding technologies, the proportion of fish raised on formulated feed remains low. Frequent disease outbreaks and inadequate feed compatibility remain pressing issues that must be addressed.³

Chinese herbal medicines, rich in bioactive components including polysaccharides, alkaloids, flavonoids, volatile oils, organic acids, and tannins, along with multiple nutrients, are widely applied in aquaculture as feeding attractants, growth promoters, antimicrobial agents, and im-

a Corresponding author, Xin Zhang, E-mail: 1832689931@qq.com

Table 1. Nutritional composition of basal feed formula (air dry basis, g/kg)

	Moisture	Crude protein	Crude lipid	Ash
basic diet	82.2	529.1	75.1	183.2

mune enhancers due to their eco-friendly, natural, and safe properties.⁴ Studies have demonstrated that compound herbal mixtures can enhance growth performance and immune function in aquatic species such as largemouth bass (*Micropterus salmoides*),⁵ large yellow croaker (*Larimichthys crocea*),⁶ Pacific white shrimp (*Litopenaeus vannamei*),⁷ and red swamp crayfish (*Procambarus clarkii*).⁸ However, limited research has been conducted on their incorporation into mandarin fish feeds. This experiment investigates the effects of dietary supplementation with a compound Chinese herbal mixture on the growth performance and serum biochemical indices of mandarin fish, aiming to provide references for the development and application of artificial feeds for this species.

MATERIALS AND METHODS

EXPERIMENTAL DIETS AND DESIGN

Finished warbling chuatsi feed as base feed (Jie Da feed, floating feed), using Chinese medicine hawthorn, angelica, astragalus compound herbal medicine (1: 2: 7).

To prepare the 2% compound Chinese herbal medicine (CHM) diet, a total of 200 g of dried Chinese herbal materials was used. The herbs were soaked in distilled water at a ratio of 1 L per 100 g of herb for 1 hour. The soaked mixture was then subjected to two rounds of decoction. Each round involved boiling the mixture over high heat for 30 minutes, followed by simmering on low heat for an additional 30 minutes. The decoctions from both rounds were combined and filtered to obtain the final herbal extract.

The resulting medicinal liquid was evenly sprayed onto the basal diet to ensure uniform coating. The coated feed was then oven-dried at $60 \pm 2^\circ\text{C}$ for 3 hours to remove moisture and stabilize the preparation. For the preparation of the 1% CHM diet, the same procedure was followed using 100 g of dried herbs instead of 200 g. Place it into a plastic bag and keep it away from light. The feed was configured every week. The nutritional composition of the feed is shown in [Table 1](#).

The basal diet formulation comprised the following ingredients: fish meal, soybean meal, wheat flour, calcium dihydrogen phosphate, sodium nitrite, vitamin A, vitamin B1, vitamin B2, vitamin B6, vitamin C, vitamin D3, vitamin E, vitamin K3, niacin, folic acid, inositol, and others.

EXPERIMENTAL FISH AND FEEDING MANAGEMENT

The Freshwater Experimental Station of Tianjin Fisheries Research Institute provided the experimental fish, where the aquaculture trial was conducted. Prior to the experiment, the fish were acclimated in culture ponds for 14 days and fed commercial feed for domestication. Feed was with-

held 24 hours before trial initiation. A total of 180 healthy mandarin fish with uniform body size (initial weight: 11.10 ± 0.53 g) were selected and randomly allocated into nine 300-L culture tanks, with 20 fish per tank. The experiment included three treatment groups, each with three replicates (culture tanks).

During the aquaculture trial, each treatment group (with three replicates) was fed to apparent satiation through three sequential feedings per day, with a daily ration equivalent to 2%–3% of the fish body mass. The feed was administered twice daily (at 09:00 and 17:00). Residual feed and feces were removed via siphoning 30 minutes post-feeding, accompanied by partial water exchange. Mortalities were promptly removed, recorded, and weighed. Feeding quantities were adjusted based on observed consumption rates and weather conditions. Throughout the 42-day trial, the water temperature was maintained at $23\text{--}29^\circ\text{C}$, with continuous aeration ensuring that dissolved oxygen levels remained above 5.5 mg/L. Water quality parameters remained within optimal ranges: pH 7.1–7.5, ammonia nitrogen ≤ 0.2 mg/L, and nitrite ≤ 0.1 mg/L.

SAMPLE COLLECTION AND MEASUREMENT

At the end of the culture trial, feeding was stopped for 24 h. The number of fish tails was counted and weighed, which was used to calculate the weight gain rate, feed coefficient, and survival rate. At the time of collection, the whole length, body length, and weight of all test fish were measured with a straightedge. Fifteen fish were randomly selected, and blood was collected from the tail vein using a 1 mL syringe. After removal, the blood was placed at 4°C for 12 h, then centrifuged at 4500 r/min for 10 min, and the supernatant was stored in a refrigerator at -80°C . After blood sampling, the fish were rapidly dissected on a chemical ice pack, the viscera and liver were removed and weighed, and the foregut was isolated and placed in liquid nitrogen for transitional storage, then transferred to -80°C for storage and used for the determination of relevant enzyme activities.

The feeds were subjected to conventional compositional analyses, in which the moisture content was determined by 105°C drying and the constant weight method, the crude protein content was determined by the Kjeldahl nitrogen determination method, the crude fat content was determined by the chloroform-methanol extraction method, and the crude ash content was determined by using the 550°C cauterisation method.

The foregut samples were removed from the refrigerator at -80°C and weighed, thawed on an ice bath, 9 times the volume of pre-cooled saline (0.85% NaCl) was added, the samples were homogenized in an ice bath, centrifuged at 4°C , 10000 r/min for 10 min, and the intestinal supernatant

was taken to determine the activities of digestive enzymes including pepsin activity, trypsin activity, amylase activity, lipase, and the sera were taken to determine the biochemical indexes, including alanine aminotransferase (ALT), aspartate aminotransferase (AST), high-density lipoprotein (HDL) cholesterol (CH), low-density lipoprotein (LDL) cholesterol (LDL) cholesterol, triglyceride (TG), total cholesterol (TC), total protein (TP), albumin (ALB), globulin (GLOB). All of the above indicators were measured using a full-wavelength enzyme marker kit produced by the Nanjing Jianjian Bioengineering Institute.

CALCULATION FORMULAE

Survival rate (SR)= Number of fish at the end of the breeding experiment/number of fish at the beginning of the experiment $\times 100\%$;

Weight gain (WG)= (final mean body mass - initial mean body mass)/initial mean body mass $\times 100\%$;

Feed conversion rate (FCR)= ingested feed dry matter mass/(final body mass - initial body mass);

Viscera-somatic index (VSI)= Final hepatopancreatic weight/final body weight $\times 100\%$;

Hepatosomatic index (HIS)= Final visceral weight/final body weight $\times 100\%$;

DATA PROCESSING

The data were expressed as mean $\pm$ standard deviation (mean $\pm$ SD), statistically analyzed by SPSS25.0 software, one-way ANOVA, Tukey's multiple comparisons, and $P < 0.05$ as the criterion of significant difference.

RESULTS

GROWTH PERFORMANCE AND MORPHOLOGICAL INDICATORS

As shown in Table 2, the weight gain rate was significantly higher in the compound herbal 1% and 2% groups compared with the control group ($P < 0.05$), and the feed coefficients were significantly lower by 0.38 and 0.5, respectively ($P < 0.05$). Among the morphological indexes, the visceral body ratio and liver body ratio were significantly decreased in the 1% and 2% groups compared to the control group ($P < 0.05$).

SERUM BIOCHEMICAL INDICATORS

As shown in Table 3, in serum, alanine aminotransferase, aspartate aminotransferase were significantly lower ($P < 0.05$), HDL cholesterol was significantly higher ($P < 0.05$), and triglycerides and cholesterol were significantly lower ($P < 0.05$) in the compound Chinese herbal medicine 1% and 2% groups compared to the control group.

INTESTINAL BIOCHEMICAL INDICATORS

As shown in Table 4, in the intestine, pepsin activity, trypsin activity, and lipase activity were significantly increased in the compound herbal medicine 1% and 2% groups compared to the control group ($P < 0.05$). There was no significant difference in amylase activity ($P > 0.05$).

DISCUSSION

EFFECT OF COMPOUND HERBS ON THE GROWTH PERFORMANCE OF *SINIPERCA CHUATSI*

In this experiment, the growth performance and digestive ability of the 1% and 2% compound Chinese herb groups were significantly improved compared to the control group, which may be related to the fact that Chinese herbs contain many nutrients and bioactive components. Studies have shown that hawthorn has many active ingredients, mainly flavonoids: quercetin, apigenin, kaempferol, etc.; organic acids: citric acid, etc.; triterpenoids: hawthorn acid, ursolic acid, oleanolic acid, etc. Oleanolic acid, etc.; Phenolic acid: Chlorogenic acid, etc.; There are also some other compounds such as polysaccharides,⁹ of which the content of organic acids in hawthorn is second only to flavonoids, including oxalic acid, malic acid, citric acid, tartaric acid, quinic acid, palmitic acid, stearic acid, oleic acid, linoleic acid, linolenic acid, succinic acid, etc., in which Citric acid, quinic acid and malic acid have the richest content,¹⁰ Vc, VB2, carotene and many kinds of organic acids in hawthorn can promote gastrointestinal peristalsis, make digestive enzyme activity and secretion increase,^{11,12} and at the same time, regulate the gastrointestinal acidity, and protect intestinal digestive bacterial flora. It also has 56 essential oil volatile substances,¹³ which can stimulate the senses of taste, smell, touch and other senses of aquatic animals. It is conducive to the stimulation and secretion of digestive juices of aquatic animals, thus enhancing the appetite of aquatic animals, promoting the digestion and absorption of nutrients, and promoting the growth of animals. In this experiment, pepsin activity, trypsin activity, lipase activity were significantly increased in the 1% and 2% groups of the compound herbal medicine, and the weight gain rate was significantly increased, which coincided with the addition of 0.5-1% hawthorn extract to the feed to enhance the digestive ability of carp (*Cyprinus carpio*) by Abdul-Hassan et al.¹⁴ The main chemical components of Astragalus are polysaccharides, saponins, flavonoids and other chemicals, etc. Among them, Astragalus polysaccharide (APS) is the main medicinal active ingredient in Astragalus, and Astragalus polysaccharide can significantly enhance the growth performance of aquaculture animals, and its effects on the growth of eels (*Monopterus albus*),¹⁵ largemouth bass (*Lepomis macrocephalus*),¹⁶ and many other fish species, which may be related to the improvement of intestinal health of aquaculture animals by astragalus polysaccharide, by promoting the proliferation of beneficial bacteria such as lactic acid bacteria, bifidobacteria, and yeasts in the intestinal tract, which promotes fish to make an increase in digestive

Table 2. Effects of compound Chinese herbal medicine on growth performance and morphological parameters of *Siniperca chuatsi*

Parameters	0	1%	2%
IBW(g)	10.45±0.08	10.39±0.57	10.66±0.56
FBW(g)	15.22±0.37 ^a	17.20±1.06 ^b	18.59±1.03 ^b
SGR(%)	78.33±2.89	81.67±2.89	78.88±3.33
WG(%)	45.6±1.71 ^a	65.5±3.20 ^b	74.4±3.53 ^b
FCR	1.57±0.21 ^a	1.19±0.14 ^b	1.07±0.15 ^b
HSI(%)	2.50±0.9 ^a	1.74±0.14 ^b	1.62±0.45 ^b
VSI(%)	8.56±0.5 ^a	7.16±1.15 ^b	6.37±0.13 ^b

Note: In the same line, letters with different superscripts indicate significant differences ($P<0.05$)

Table 3. Effects of compound Chinese herbal medicine on serum biochemical indexes of *Siniperca chuatsi*

Parameters	0	1%	2%
Serum			
Alanine aminotransferase (ALT)	181.75±7.95 ^a	110.55±19.63 ^b	62.3±6.39 ^c
Aspartate aminotransferase (AST)	505.95±113.35 ^a	326.73±84 ^b	219.63±20.3 ^b
High-density lipoprotein cholesterol (HDL-CH)	2.35±0.18 ^a	2.73±0.27 ^b	2.82±0.13 ^b
Low-density lipoprotein cholesterol (LDL-CH)	0.15±0.02	0.14±0.05	0.17±0.02
Triglycerides (TG)	2.54±0.47 ^b	2.38±0.16 ^{ab}	2.16±0.18 ^a
Total Cholesterol (TC)	3.85±0.25 ^b	3.45±0.42 ^{ab}	3.22±0.26 ^a
Total protein (TP)	35.83±1.73	32.92±1.8	34.45±2.58
Albumin (ALB)	11.18±0.68	10.93±0.21	11.92±0.63
	24.65±1.05	21.98±1.71	22.53±2.07

Note: In the same line, letters with different superscripts indicate significant differences ($P<0.05$)

Table 4. Effects of compound Chinese herbal medicine on intestinal biochemical indexes of *Siniperca chuatsi*

Parameters	0	1%	2%
Pepsin activity (U/mgprot)	2.53±0.19 ^a	3.42±0.28 ^b	3.72±0.13 ^b
Trypsin activity (U/mgprot)	937.33±28.99 ^a	1516.69±184.50 ^b	1690.97±75.21 ^b
Amylase activity (U/gprot)	72.56±6.72	78.03±10.62	77.73±1.71
Lipase activity (U/gprot)	26.02±0.68 ^a	34.16±2.31 ^b	34.89±3.05 ^b

Note: In the same line, letters with different superscripts indicate significant differences ($P<0.05$)

enzyme activity and secretion,¹⁷ In addition, the APS can improve the length of the intestinal villi in tilapia (*Oreochromis niloticus*) by increasing the length of the crypt depth and muscularis layer thickness, improving the intestinal structure and enhancing the growth performance of the organism.¹⁸

EFFECT OF COMPOUND HERBS ON BIOCHEMICAL INDICES OF *SINIPERCA CHUATSI*

Alanine aminotransferase (ALT) and aspartate aminotransferase (AST) are reliable indicators of liver cell damage, as shown in Table 3; the ALT and AST levels in the 1% and 2% compound herbal medicine treatment groups are significantly lower than those in the control group. This suggests that compound herbal medicine protects the liver, on the one hand, by enhancing the body's antioxidant power

and reducing the occurrence of inflammation. Hawthorn contains a large number of flavonoids, such as isoosmetin, quercetin, etc., organic acids, such as ascorbic acid, etc., and proanthocyanidins, which can effectively scavenge oxygen radicals in the body.¹⁹ In addition, hawthorn pectin and hawthorn total organic acid inhibit oxidative stress-induced damage by increasing antioxidant enzyme activity and reducing oxidation product generation, while hawthorn flavonoid components mainly regulate Nrf2 signaling pathway-related proteins and factors to alleviate oxidative stress.²⁰ The main components of *Angelica sinensis* include essential oils, flavonoids, organic acids, polysaccharides, etc. *Angelica sinensis* polysaccharide (ASP) is the main water-soluble component, which has a good scavenging ability of free radicals, significantly improves the activity of antioxidant enzymes in the body and up-regulates the

expression of antioxidant genes, and the study showed that *Angelica sinensis* polysaccharide can significantly improve the activity of grouper (*Epinephelus malabaricus*), ovigerous pompano jack (*Trachinotus ovatus*) juvenile fish antioxidant capacity.^{21,22} Astragalus polysaccharide (APS) in *Astragalus membranaceus*, as a kind of plant polysaccharide, has similar antioxidant power, and the addition of appropriate amount of APS to the feeds of crucian carp²³ and tilapia²⁴ have significantly improved the antioxidant power of the organism.

On the other hand, compound herbs inhibit lipid synthesis and enhance lipid uptake to avoid fat accumulation in the liver and reduce liver damage. The role of HDL cholesterol is to transport cholesterol from non-hepatic cells to the liver, thereby reducing serum cholesterol levels. In contrast, LDL cholesterol has the opposite effect; serum cholesterol and triglyceride levels reflect the organism's lipid metabolism to a certain extent. As shown in Table 3, triglycerides and cholesterol levels were significantly reduced in the 2% group of compound herbs. Studies have shown that hawthorn has a good lipid-lowering effect, and the total flavonoids in hawthorn inhibit the secretion of leptin and plasma fibrinogen activator inhibitor (PAI-1) from mature adipocytes,²⁵ in addition, proanthocyanidins, chrysin, ursolic acid, oleanolic acid, corosolic acid and hawthornic acid in hawthorn are also considered to be one of the effective components in lowering blood lipids.²⁶⁻³⁰ *Angelica* polysaccharides can reduce lipid content, inhibit fat regeneration, and improve hepatic lipodystrophy.³¹ ASP also inhibits hepatic stellate cell activation via the IL-22/STAT3 pathway, which may reduce hepatic fibrosis and promote the regeneration and repair of damaged hepatocytes.³² *Astragalus* polysaccharides may protect the liver by up-regulating the expression of HO-1 protein in liver tissues, inhibiting pro-inflammatory factors, exerting anti-oxidative free radical effects, and promoting the production of anti-inflammatory factors, among other mechanisms.³³

CONCLUSION

The combination of Hawthorn, *Angelica sinensis*, and *Astragalus membranaceus* (in a 1:2:7 ratio) can significantly enhance the growth performance, digestive ability, and antioxidant capacity of *Siniperca chuatsi*. Considering the growth performance of *Siniperca chuatsi* and the serum biochemical indexes, it is recommended to add hawthorn, *An-*

gelica sinensis, and *Astragalus membranaceus* (1:2:7 ratio) at a rate of 2%. Further farming trials with multiple inclusion levels will be conducted subsequently to determine the optimal inclusion ratio.

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AUTHORS' CONTRIBUTION PER CREDIT

Methodology: Nan Li; Formal analysis and investigation: Xiaodi Shang, Ziyuan Ding, Hongqing Sun; Writing - original draft preparation: Mingze Li, Na Wang; Writing - review and editing: Jian Wang, Sudong Xia; Funding acquisition: Xin Zhang; Resources: Lin Ma.

COMPETING INTEREST – COPE

The authors declare no potential conflict of interest.

ETHICAL CONDUCT APPROVAL – IACUC

All animal care protocols have been approved by the Animal Care and Use Committee of the Tianjin Fisheries Research Institute, and all authors clearly state that they have followed these guidelines.

INFORMED CONSENT STATEMENT

All authors and institutions have confirmed this manuscript for publication.

DATA AVAILABILITY STATEMENT

All are available upon reasonable request.

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