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Optimising growth and physiological performance of carp in polyculture within an integrated multitrophic aquaculture system

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Abstract. The study aimed to determine the impact and evaluate the effectiveness of technological factors in producing a synergistic effect during the feeding and rearing of juvenile cyprinids. The research was conducted using theoretical, experimental, and laboratory methods in fisheries science. The findings indicated that the inclusion of alternative protein sources (*Spirulina platensis*, *Artemia*, and *Hermetia illucens*) within an Integrated Multitrophic Aquaculture (IMTA)

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system significantly enhanced growth performance and reduced dependence on conventional feeds. The cultivation of all components within a single IMTA framework ensured optimal control over both qualitative and quantitative parameters throughout the production cycle. The results demonstrated improved viability in juvenile cyprinids and enhancement of blood composition in experimental Groups A and B. However, Group B exhibited the most pronounced improvements in both qualitative and quantitative parameters, characterised by increased homeostatic regulation and enhanced haematopoiesis. Improvements in haematological profiles, body weight, and muscle tissue composition were closely correlated with elevated protein and carbohydrate metabolism and accelerated ontogenetic development. Fish in the experimental groups displayed superior indicators of mass accumulation and metabolic processes (WG, FCR, HSI, VSI). A strong positive correlation was recorded in experimental Groups A and B: body weight ↔ protein in the muscle tissue ($R^2 = 0.8$-0.9); cholesterol in the blood ↔ fat in the muscle tissue ($R^2 = 0.8$-0.7). In experimental Group A: a higher red blood cell count (RBC) and mean corpuscular haemoglobin (MCH) were recorded in the blood of carp *Cyprinus carpio* (Linnaeus, 1758) ($p \leqslant 0.05$). In the blood of carp *Cyprinus carpio* (Linnaeus, 1758) from Group B: RBC, MCH, total protein and creatinine content ($p \leqslant 0.05$) and haemoglobin (Hb) ($p \leqslant 0.01$) exceeded those of the control group. In the blood of silver carp hybrid *Hypophthalmichthys* molitrix / *Hypophthalmichthys* nobilis of Group B: RBC, Hb, total protein and creatinine content ($p \leqslant 0.05$) also exceeded the control group. The most favourable biochemical composition of muscle tissue was recorded in fish from experimental Groups A and B

Keywords: feeding; cultivation; cyprinids; physiological and biochemical processes; multitrophicity; recirculation systems

Introduction

Aquaculture contributed to the development of high-quality, strategic programmes aimed at ensuring food security in various countries. The priority was to maintain a balance between the ecosystem and each country's globalised food system. In intensive aquaculture systems, inexpensive aquafeed and high-quality water were regarded as critical requirements; therefore, the issue of rationalising resource use became both relevant and practical. In addition, high-quality fish feed was considered essential for aquafarmers, as feeding represented a continuous process in aquaculture technology, and the quality of the feed directly influenced the quality of aquaculture products. At the global level, modern concepts and strategic programmes focused on the sustainable development of aquaculture. Simultaneously, the dominant approach emphasised the efficient use of resources with minimal negative impact on the ecosystem (FAO, 2024). Numerous studies confirmed that the application of chemicals, hormones, and antibiotics remained one of the most urgent problems in fish farming and aquaculture production. Such technological interventions contributed to water pollution and the disruption of ecosystems, as reported by O. Honcharova & V. Bekh (2023), V. Sakharnatsky (2024).

Research within this area revealed and confirmed the positive effects of phytopreparations, essential oils, and probiotics on the general physiological state of the organisms (Silva *et al.*, 2024; Taşcı *et al.*, 2025). Scientific works increasingly substantiated the relevance of developing plant-based feed additives. It was noted that the environmental friendliness of such natural components had become a research focus and was regarded as being of great importance for the sustainable development of aquaculture (Gusmão, 2024; Wang *et* *al*., 2024). The literature contained positive evidence regarding the use of adaptogens, hepatoprotectors, growth stimulants in aquaculture, etc. (Fraijo-Valenzuela *et al*., 2024). The rapid development of technologies implied the need for innovative solutions to correspond with the physiology of aquatic organisms. Therefore, the optimisation of technological schemes for rearing aquatic species and producing aquaculture products remained an open question. At the same time, relatively few combined systems existed in aquaculture. Consequently, the multitrophic aquaculture model was considered a promising approach for addressing the identified challenges. An increasing number of studies from different countries confirmed the rationale and trend towards the use of recirculating and integrated aquaculture systems (Land-based aquaculture report, 2023; Gupta *et al*., 2024). Most authors emphasised the environmentally safe direction of multitrophic aquaculture models, which acquires practical significance and formed prerequisites for the sustainable development of the industry (Alam *et al*., 2024; Meitei *et. al*., 2025). Thus, the multitrophic aquaculture system offered several advantages; however, it required comprehensive and in-depth research in the future. The purpose of the study was to conduct a multicomponent analysis and comparative experiment assessing the impact of several ecologically oriented technological solutions on fish physiological functionality.

Literature Review

Considering the trends and current state of the aquaculture industry, the dominant issue in most scientific works was the search for and development of alternative protein sources for aquatic organisms with an emphasis on their rationality and environmental friendliness (Belluco *et al*., 2013; Honcharova *et al*., 2020; FAO, 2024). Studies by various researchers reflected the possibility of optimising the overall diet of fish through alternative protein sources (Henry *et al*., 2015; Bekh *et al*., 2020). The focus of such research was directed towards achieving less expensive production and reduced environmental impact. At the same time, it was experimentally established that the use of natural additives was aimed at replacing fishmeal. In the scientific works of A. Fraijo-Valenzuela *et al*. (2024) a synergistic effect was observed between the growth of aquaculture production and consumer demand for such products. At the same time, attention was drawn to the importance of comprehensive investigation of each component, taking into account both its beneficial and potentially adverse effects. For instance, in aquaculture feeds, the inclusion of crickets was recommended for study while considering the biological characteristics of aquatic organisms, since their bodies could absorb individual components of such protein supplements differently.

According to O. Deren & M. Fedorenko (2023), the introduction of non-traditional feed components for Ukrainian aquaculture was considered relevant. Under conditions involving the use of the black soldier fly (*Hermetia illucens*) in aquaculture, improvements in fishery parameters, feed conversion, and the overall quality and safety of the resulting products were reported. The positive outcomes of using *Tenebrio molitor* were justified by its antioxidant and immunological properties (Shafique *et al*., 2021). Similarly, the incorporation of naked oats as a feed component has demonstrated beneficial effects on growth performance and haematological parameters in carp, with optimal results achieved at 30% inclusion rates (Syrovatka, 2021). In scientific works by H. Wang *et al*. (2011), M. Henry *et al*. (2015) the prospects for utilising biological waste as protein alternatives for aquaculture were presented. Furthermore, increases in the developmental parameters of hydrobionts during ontogenesis were experimentally confirmed. It was emphasised that modular combined aquaculture systems had lower competition for land resources compared with the production of other feed components and additives.

According to R. Chaklader *et al*. (2019) and F. Melenchón *et al*. (2021), the inclusion of mealworm, black soldier fly, and poultry by-product protein in aquaculture diets contributed to enhanced functional-active parameters and histomorphology in fish. At the same time, A. Józefiak & R. Engberg (2017) and S. Nogales-Mérida *et al*. (2019) focused on comprehensive analyses, particularly of chitins and antimicrobial peptides, which not only improved growth but also strengthened the defence mechanisms, immunity, and antimicrobial properties of fish receiving such protein alternatives. A. Gopalakannan & V. Arul (2006) reported that the application of crustacean chitin in low quantities when feeding fish stimulated immune responses and enhanced resistance to pathogens. After analysing available sources and scientific developments, it was noted that the use of natural components and protein alternatives was highly promising. It was also considered advisable to pay attention to technological conditions, as optimisation and adaptation to modern standards ensured the sustainable development of aquaculture.

Furthermore, the hydrochemical regime, particularly within recirculating aquaculture systems (RAS), was recognised as a critical factor (Hrynevych *et al*., 2019). According to E. Can & Ş. Seyhaneyildiz Can (2023), the improvement of technological schemes in aquaculture, for example, through elements of aquaponics and multitrophic aquaculture, was developing dynamically and showed a tendency towards global implementation. Thus, modern directions of optimisation measures in aquaculture were oriented towards the rational use of resources, the maximum environmental sustainability of production, and the high quality of aquaculture products, with particular emphasis on the multitrophic aquaculture model. However, this issue remained open and required further justification and comprehensive evaluation of the effectiveness of such models.

Materials and Methods

The experimental study was carried out at the Scientific Research Laboratory and the "Physiological and Biochemical Research" Laboratory of Kherson State Agrarian and Economic University (KSAEU) and in the conditions of production of Kherson fish enterprise (Ukraine) in 2025. Experiments lasted 120 days. Scientific and research activities were conducted in accordance with generally accepted requirements and standards for the organisation of experiments in fish farming. Compliance with the Directive 2010/63/EU (2010) was ensured. All manipulations involving carp and silver carp hybrids in both experimental and control groups were performed in accordance with the European Convention (1986). All scientific and practical research was based on theoretical (analysis, synthesis, comparison and modelling) experimental, laboratory methods, taking into account the specialisation of the fisheries industry. The functional status of the fish organism was evaluated according to the qualitative and quantitative parameters of biological material in the laboratory of the Department of Aquatic Bioresources and Aquaculture of the KSAEU on the Humalyzer 3000 analyser (Germany) using unified Human GmbH kits.

The study focused on *Cyprinus carpio* (Linnaeus, 1758) and silver carp hybrid *Hypophthalmichthys molitrix / Hypophthalmichthys nobilis*. Fish were reared under recirculating aquaculture system conditions, which included standard water filtration, temperature regulation, and oxygenation systems. The experimental setup incorporated a modular integrated multitrophic aquaculture system, that included the cultivation of natural feed components (Fig. 1). The modular system was constructed on a mobile platform equipped with an environmental monitoring unit and microcrystalline solar panels for supplementary energy. Each module was functionally organised to enable resource recycling, improve energy efficiency, and promote synergistic interactions among aquatic and agricultural components.

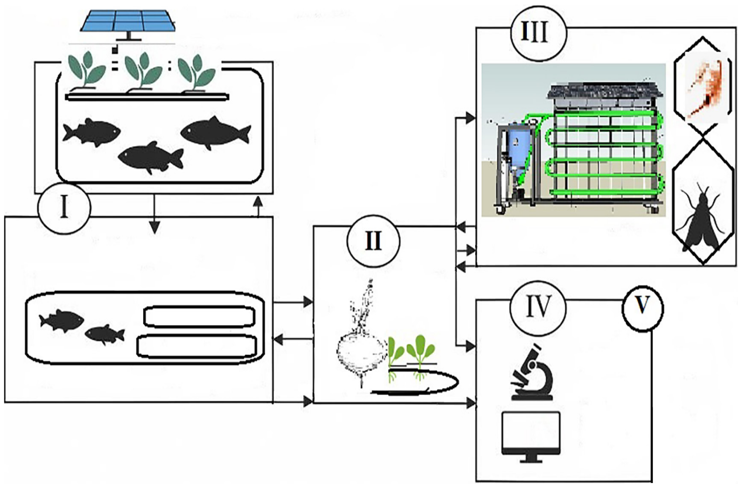

Figure 1. Schematic representation of the modular multitrophic aquaculture system

Note: I – RAS tanks and ponds, combined with elements of biological filtration; II – cultivation units for agricultural raw materials used in feed production; III – cultivation chambers for *Hermetia illucens* (black soldier fly), *Artemia* and *Spirulina*; IV – technological management units for equipment and process control; V – mini-laboratories for rapid hydrochemical and process diagnostics

Source: authors' development

Cyprinus carpio (Linnaeus, 1758) and *Hypophthalmichthys molitrix* / *Hypophthalmichthys nobilis* were used in the experiment. The initial average body weight of fish was 1.5-1.6 mg, and each tank contained 260 individuals. The volume of the rearing tanks was gradually increased as the fish developed through ontogenesis from 150 dm^3 and 250 dm^3 to 500 dm^3. It was taken into account that during days 10-15 of rearing, the carp's body undergoes an active transformation of metabolic processes. The initial daily ration was set at 2% of body weight and was increased to 5% from day 10. The stocking density corresponded to carp culture standards and averaged 25-30 kg/m^3 (FAO *et al.*, 2025).

Feed factor. The diet of carp fish was optimised in accordance to generally accepted recommendations (Yevtushenko & Khyzhniak, 2019; Yevtushenko *et al.*, 2022; The International Aquaculture Feed Formulation Database, n.d.). At the beginning of early ontogenesis, cyprinids were fed with finely dispersed flour (0.1-0.2 mm) in accordance with the recommendations of the standard diet in fish farming. Feeding was provided at a rate of 2% of body weight; after 14 days, the feeding rate was increased to 4.8-5%. The main oriented periods of transition to another diet were identified: initial with an active phase for 10-15 days, transitional and growth. In the experimental groups, the fish received a general diet and supplements in accordance with the study scheme. The composition of the base diet, %, was as follows: fish meal (13), meat and bone meal (5), grass meal (3), wheat (9.5), sunflower meal (40), peas (10), feed yeast (18.5), premix (1). Natural feed components were obtained from in-house production within the autonomous sectors of the modular multitrophic aquaculture system. The technological scheme of cultivation provided appropriate equipment and optimal conditions for each sector (Fig. 1). The design of the experimental groups is presented in Figures 2 and 3.

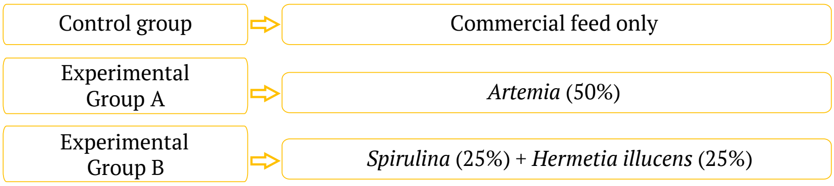

Figure 2. Scheme for introducing natural components to the fish diet
Source: authors' development

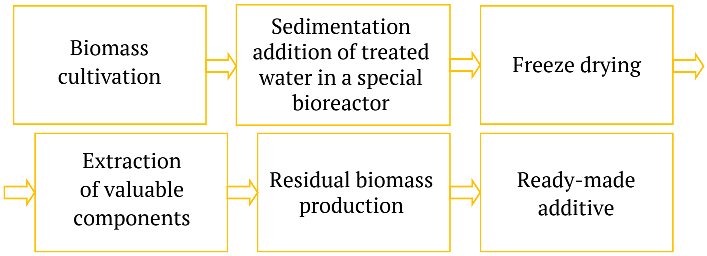

Figure 3. Technological aspects of the preparation of feed ingredients for addition during feeding to the experimental group
Source: authors' development

The day before the experiment, the optimal ratio of natural components was experimentally established and subsequently used in the present study:

➢ *Hermetia illucens* were cultured using cereal bran and fruit/vegetable residues. Cereal bran was selected as the optimal substrate based on prior trials. Rearing conditions (temperature, humidity, light) were maintained in accordance with standard rec ommendations. Composition (%): moisture – 68.4; protein – 43.8; dry matter – 31.6; fat – 25.9; ash – 6.5.

➢ *Spirulina* biomass was cultivated in a closed-loop bioreactor using an energy-saving technology developed by the authors. The harvested *Spirulina* biomass was green and mildly saline. Composition (%): moisture – 10; dry matter – 90; protein – 62; carbohydrates – 14.7; fat – 4; fibre – 3; ash – 6.3.

➢ *Artemia nauplii* were reared under standard hydrochemical conditions (temperature: 25-28°C; salinity: 30-80‰. *Nauplii* measured 0.3-0.6 mm in length and weighed approximately 0.01-0.06 mg per individual). Composition (%): moisture – 12; dry matter – 88; protein – 53; carbohydrates – 10; fat – 13.2; ash – 4.0. *Artemia* were used primarily during the early feeding stages of carp larvae. The feeding substrate for *Artemia* consisted of *Spirulina* and *Chlorella* cultures obtained from the multitrophic aquaculture system. To obtain *nauplii*, cysts were hydrated at a density of approximately 3 g L^{-1}. After hatching, aeration was discontinued and the *nauplii* were allowed to settle for 5 minutes before being filtered. Stocking density was maintained at 10-12 *nauplii* mL^{-1} in the rearing tanks.

Study of blood composition. Blood samples were collected from the caudal vein using Pasteur needle and heparinised syringes. Morpho-functional and biochemical parameters were determined for cyprinid blood: red blood cell count (RBC), haematocrit (Ht), haemoglobin (Hb), mean corpuscular volume (MCV), mean corpuscular haemoglobin concentration (MCHC) and mean corpuscular haemoglobin (MCH). Biochemical indices in plasma: total protein, albumin, cholesterol, creatinine and glucose, were quantified photometrically using a Humalyzer 3000 biochemical analyser (Germany, 2010) with commercial kits (Human GmbH, Germany).

Study of ontogenesis. Growth performance was assessed through regular biometric monitoring of a representative subsample of 40 fish per group (measurements were taken on: 1 – day 0; 2 – day 30; 3 – day 60; 4 – day 90; 5 – day 120). For each specimen, total length and body weight were recorded, and the following indices were calculated: weight gain (WG), specific growth rate (SGR), Fulton's condition factor (FC), feed conversion ratio (FCR), and survival rate. Furthermore, the liver and viscera of 12 randomly selected fish per group were excised and weighed to compute the hepatosomatic index (HSI) and viscerosomatic index (VSI). After dissection, fillets were skinned, homogenised, and analysed to determine muscle composition.

$$\text{WG, g} = \text{Final body weight (g)} - \text{Initial body weight (g)}. \quad (1)$$

$$\text{Fulton's Condition Factor} = [\text{body weight (g)}/\text{total length (cm)}^3] \times 100. \quad (2)$$

$$\text{SGR, \%} = \left[\left(\frac{\log(\text{final body weight,g}) - \log(\text{intial body weight,g})}{\text{number of feeding days}}\right)\right] \times \times 100 \quad (3)$$

$$\text{FCR} = \text{total feed consumed per tank biomass cyprinids (g)}/\text{weight gain (g)}. \quad (4)$$

$$\text{Survival, \%} = (\text{number of fish initial}/\text{number of fish final}) \times 100. \quad (5)$$

The liver and viscera of *Cyprinus carpio* (Linnaeus, 1758) and *Hypophthalmichthys molitrix / Hypophthalmichthys nobilis* from each group (12 specimens per tank) were separated and their weights were measured. HSI and VSI (%) were calculated:

$$\text{HSI, \%} = [(\text{liver weight (g)}/\text{body weight (g)}] \times 100. \quad (6)$$

$$\text{VSI, \%} = [(\text{viscera weight (g)}/\text{body weight (g)}] \times 100. \quad (7)$$

Study of the chemical composition of carp muscle tissue. Biological samples were transported using a ThermoMix unit (USA). Fillets were skinned, homogenised, and prepared for the determination of chemical composition. The analysis of muscle tissue was performed in triplicate. Moisture content was determined by drying samples at 105°C to constant weight. Crude protein was quantified using the Kjeldahl method, while ash content was measured by standard incineration. The fat content in muscle tissue was determined using the classical gravimetric extraction method with organic solvents.

Study of hydrochemical parameters. During the entire period of the experiment, the hydrochemical regime corresponded to generally accepted standards for carp with appropriate age characteristics of cyprinids. Water quality was monitored throughout the experimental period using a Palintest 7500 photometer (UK) and corresponding reagent kits, following aquaculture guidelines (Yevtushenko & Khyzhniak, 2019). Dissolved oxygen, pH, and temperature were measured daily, while ammonium (NH_4^+), nitrite (NO_2^-), and nitrate (NO_3^-) concentrations were analysed three times per week. The rearing environment was maintained within optimal ranges: temperature (22.1-24.2°C), dissolved oxygen (5.8-6.1 mg/L), pH (7.2-7.4), NO_3^- (1.0-1.2 mg/L), and NO_2^- (0.08-0.012 mg/L). Measurements were performed using both rapid on-site tests and laboratory analyses to ensure the reliability of data.

Statistical analysis. All data were expressed as mean ± standard deviation (SD). Statistical comparisons were conducted using one-way analysis of variance (ANOVA). Differences between treatments were considered statistically significant at $p \leq 0.05$ with comparisons made relative to the control group.

Results and Discussion

The study of the growth and development of cyprinids in ontogenesis demonstrated that under the conditions of using elements of multitrophic aquaculture in two experimental groups (A and B), the parameters were higher than the control values (Fig. 4).

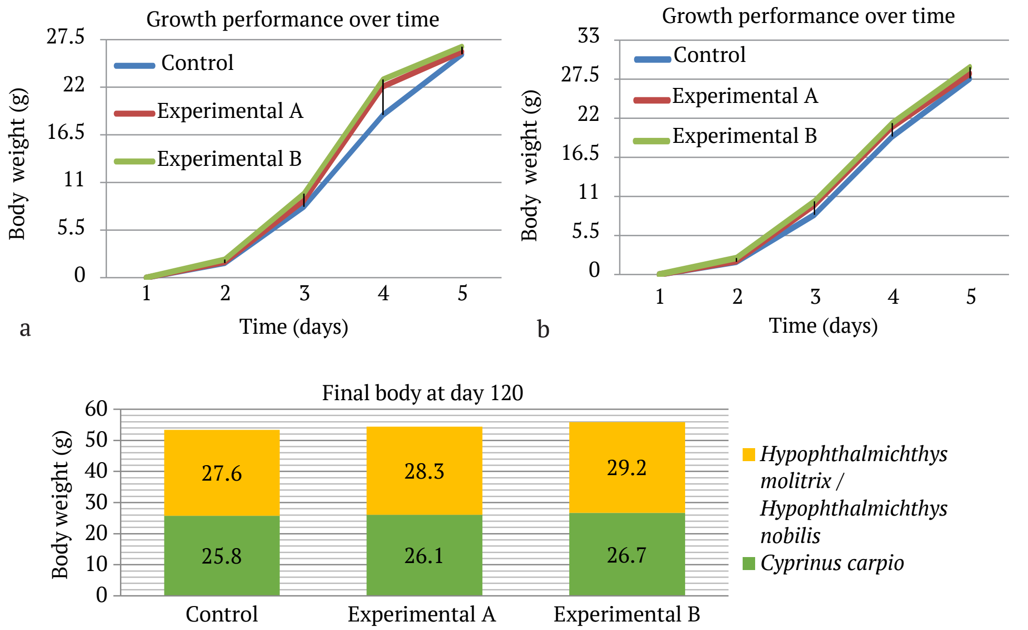

Figure 4. Study of the growth rate of *Cyprinus carpio* (a) and *Hypophthalmichthys molitrix* / *Hypophthalmichthys nobilis* (b) in ontogenesis under the influence of technological factors ($x \pm SD$)
Note: 1 – day 0; 2 – day 30; 3 – day 60; 4 – day 90; 5 – day 120
Source: authors' development

In experimental Group B, compared with the control group, significantly higher values of metabolic activity were observed than in the other groups. This was reflected in several key parameters, including Fulton's condition factor, average body weight, weight gain, and feed conversion ratio. At the end of the experimental period, the average body weight of carp in Group B exceeded that of the control group by 3.4%, WG by 6.9%, and FCR by 14.6% ($p \leq 0.05$). In experimental Group A, the average body weight of carp surpassed the control group by 1.21%, WG by 2.9%, and FCR by 9.2%. Overall, carp and silver carp in experimental Group B demonstrated more active weight accumulation and a more efficient utilisation of physiological potential compared with Group A and the control group (Fig. 5).

At the end of the experiment, the growth and development rate indicators in silver carp demonstrated higher values in experimental Group B compared with both the control and experimental Group A. Specifically, the average body weight exceeded that of the control by 7.0% ($p \leq 0.001$) and that of Group A by 3.3% ($p < 0.01$). Weight gain was higher by 6.8%

($p \leqslant 0.05$) and 5.2%, while the feed conversion ratio improved by 7.6% ($p \leqslant 0.001$) and 4.9%, respectively. Parameters such as the specific growth rate, viscerosomatic index, and hepatosomatic index in carp and silver carp were also higher in both experimental groups compared with the control group, correlating positively with mass accumulation indicators.

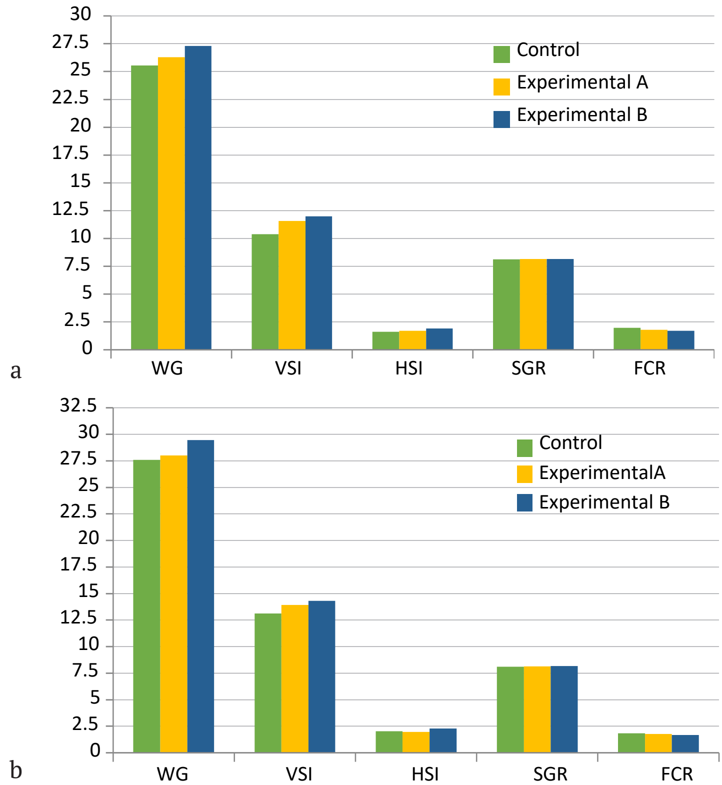

Figure 5. Study of the growth rate of *Cyprinus carpio* (a) and *Hypophthalmichthys molitrix* / *Hypophthalmichthys nobilis* (b)

Source: authors' development

Given that the activity and efficiency of physiological and biochemical processes in fish are synergistically dependent on the parameters of homeostatic balance, the morpho-functional composition of blood in experimental fish was analysed (Fig. 6). The total erythrocyte count and haemoglobin content were higher in both experimental groups than in the control, remaining within the physiological range for cyprinids. The obtained results suggest that redox reactions and oxygen-synthesising processes in the organism of experimental fish occurred more actively. In carp blood, the erythrocyte content in experimental Group A exceeded the control value by 30.4% ($p \leqslant 0.05$), while in Group B the difference reached 34.8% ($p \leqslant 0.05$). The haemoglobin concentration and MCH followed a similar trend, indicating enhanced oxygen transport efficiency and metabolic activity in the experimental groups. In carp from this group, haemoglobin levels exceeded those of the control by 6.7% ($p \leqslant 0.01$), while MCH values were higher by 22.9% ($p \leqslant 0.05$) (Fig. 6).

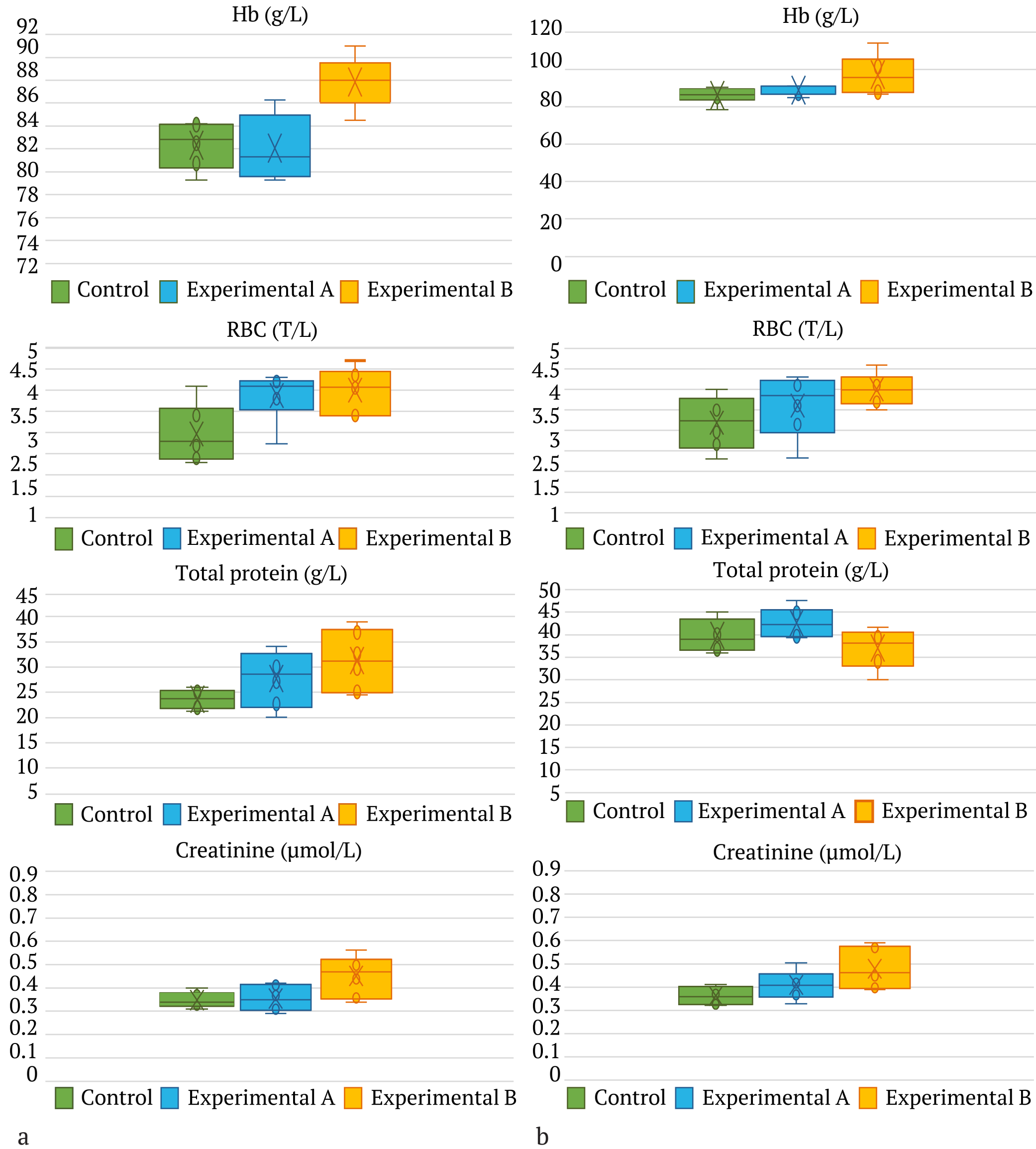

Figure 6. Study of the growth rate of *Cyprinus carpio* (a) and *Hypophthalmichthys molitrix* / *Hypophthalmichthys nobilis* (b) in ontogenesis under the influence of technological factors ($x \pm SD$)

Source: authors' development

In experimental Group B, the erythrocyte and haemoglobin contents in the blood of silver carp exceeded those in the control group by 25.3% ($p \leq 0.05$) and 12.9% ($p \leq 0.05$), respectively. In experimental Group A, the average erythrocyte count and haemoglobin content were also higher than in the control group; however, the differences were not statistically significant. The concentrations of total protein and creatinine, which are indicative of protein synthesis, amino acid catabolism, and energy metabolism, were higher in all experimental groups compared with the control. The total protein content in the serum of carp and silver carp from experimental Group A exceeded that of the control by 17.1% and 4.5%, respectively. In experimental Group B, these parameters in silver carp and carp were higher than in the control by 15.4% ($p \leqslant 0.05$) and 32.3% ($p \leqslant 0.05$), respectively. Correlation analysis of parameters with the highest R^2 values indicated a synergistic effect of the technological factor (natural feed components) on the physiological status of carp and silver carp in both experimental groups. However, the values recorded in Group B were higher while remaining within the physiological norm for cyprinids (Tables 1, 2) A high level of correlation was established with the specified parameters, which was within the limits of $R^2 = 0.6\text{-}0.9$ (A) and $R^2 = 0.8\text{-}0.9$ (B) (Table 1) and $R^2 = 0.8\text{-}0.9$ (A) and $R^2 = 0.7\text{-}0.9$ (B) (Table 2).

Table 1. *Coefficient of determination of mass accumulation and blood parameters of carp in polyculture*

Parameters	Experiment A	Experiment B
	Cyprinus carpio	
creatinine	$R^2 = 0.7771$	$R^2 = 0.8499$
	$y = 11.588\ln(x) + 38.17$	$y = 8.5236\ln(x) + 33.635$
total protein	$R^2 = 0.7954$	$R^2 = 0.8974$
	$y = 8.6543\ln(x) - 2.5546$	$y = 9.2401\ln(x) - 4.9808$
red blood cell count (RBC)	$R^2 = 0.5797$	$R^2 = 0.863$
	$y = 9.0266\ln(x) + 13.935$	$y = 13.333\ln(x) + 8.307$
haemoglobin (Hb)	$R^2 = 0.8661$	$R^2 = 0.9112$
	$y = 54.353\ln(x) - 213.49$	$y = 68.903\ln(x) - 281.65$
Hypophthalmichthys molitrix / Hypophthalmichthys nobilis		
creatinine	$R^2 = 0.8151$	$R^2 = 0.9468$
	$y = 9.9406\ln(x) + 37.235$	$y = 5.72\ln(x) + 33.25$
total protein	$R^2 = 0.7127$	$R^2 = 0.9169$
	$y = 15.566\ln(x) - 29.031$	$y = 13.652\ln(x) - 22.257$
red blood cell count (RBC)	$R^2 = 0.8934$	$R^2 = 0.8787$
	$y = 6.5648\ln(x) + 19.968$	$y = 10.044\ln(x) + 15.077$
haemoglobin (Hb)	$R^2 = 0.9396$	$R^2 = 0.7785$
	$y = 57.569\ln(x) - 230.12$	$y = 8.4743\ln(x) - 9.7891$

Source: developed by the authors

Table 2. *Coefficient of determination of parameters of chemical composition of muscle tissue and indicators of mass accumulation and blood of carp in polyculture*

Parameters	Experiment A	Experiment B
	Cyprinus carpio	
Body weight ↔ protein in muscle tissue	$y = 82.287\ln(x) + 201.74$	$y = 31.112\ln(x) + 60.316$
	$R^2 = 0.7697$	$R^2 = 0.9376$
Moisture ↔ fat in muscle tissue	$y = 34.912\ln(x) + 145.98$	$y = 11.483\ln(x) + 44.467$
	$R^2 = 0.7517$	$R^2 = 0.7026$
Cholesterol in the blood ↔ fat in muscle tissue	$y = 4.2002\ln(x) + 15.481$	$y = 3.2918\ln(x) + 10.727$
	$R^2 = 0.7855$	$R^2 = 0.6591$
Hypophthalmichthys molitrix / Hypophthalmichthys nobilis		
Body weight ↔ protein in muscle tissue	$y = 54.587\ln(x) + 131.24$	$y = 28.289\ln(x) + 54.84$
	$R^2 = 0.8991$	$R^2 = 0.8748$

Table 2. Continued

Parameters	Experiment A	Experiment B
	Cyprinus carpio	
Moisture ↔ fat in muscle tissue	y = 11.028ln(x) + 44.954	y = 21.7ln(x) + 91.032
	$R^2 = 0.9532$	$R^2 = 0.8625$
Cholesterol in the blood ↔ fat in muscle tissue	y = 1.6325ln(x) + 3.9733	y = 2.6439ln(x) + 7.9951
	$R^2 = 0.9762$	$R^2 = 0.9029$

Source: developed by the authors

After analysing the leading parameters of protein and carbohydrate metabolism, was noted the positive effect of the technological factor with elements of multitrophic aquaculture on the blood composition of cyprinids. At the same time, in the blood of carp and silver carp, the creatinine concentration in experimental Group B significantly exceeded the value in the control by 30% $p \leqslant 0.05$ and by 32% $p \leqslant 0.05$, respectively (Tables 3, 4).

Table 3. *Functional status of the Cyprinus carpio organism under the influence of the feed factor under the conditions a modular multitrophic aquaculture system, (x ± SD)*

Parameters	Groups		
	Control	Experiment A	Experiment B
Red blood cell count, T/L	2.967 ± 0.680	3.870 ± 0.583*	4.001 ± 0.5195*
Haemoglobin content, g/L	82.333 ± 1.962	82.073 ± 2.848	87.833 ± 2.295**
MCV, mkm³	124.649 ± 36.541	104.453 ± 30.058	102.903 ± 19.819
MCH, pg	28.912 ± 6.278	21.715 ± 4.159*	22.294 ± 3.1900*
MCHC, %	23.839 ± 3.810	21.493 ± 4.117	22.048 ± 3.675
Glucose, µmol/L	97.983 ± 12.001	102.117 ± 13.206	110.708 ± 12.4708
Total protein, g/L	23.667 ± 1.838	27.733 ± 5.495	31.317 ± 5.9975*
Creatinine, µmol/L	0.347 ± 0.033	0.355 ± 0.054	0.451 ± 0.0890*
Cholesterol, mg/dL	114.983 ± 5.115	112.400 ± 4.767	113.967 ± 2.658

Note: *0.05 ($p \leqslant 0.05$); **0.01 ($p \leqslant 0.01$); ***0.01 ($p \leqslant 0.001$); MCV – mean corpuscular volume; MCH – mean corpuscular haemoglobin; MCHC – mean corpuscular haemoglobin concentration
Source: developed by the authors

Table 4. *Functional status of the organism Hypophthalmichthys molitrix / Hypophthalmichthys nobilis under the influence of the feed factor under the conditions a modular multitrophic aquaculture system, (x ± SD)*

Parameters	Groups		
	Control	Experiment A	Experiment B
Red blood cell count, T/L	3.190 ± 0.655	3.613 ± 0.763	3.998 ± 0.3907*
Haemoglobin content, g/L	86.117 ± 4.144	88.980 ± 2.428	97.233 ± 10.7306*
MCV, mkm³	122.231 ± 33.935	114.652 ± 24.598	105.798 ± 21.2466
MCH, pg	27.841 ± 4.984	25.680 ± 6.050	24.347 ± 1.7034
MCHC, %	23.547 ± 4.512	22.771 ± 4.588	23.692 ± 4.3003
Glucose, µmol/L	102.745 ± 7.820	111.750 ± 11.401	115.398 ± 12.7321
Total protein, g/L	37.033 ± 4.371	38.717 ± 8.007	42.750 ± 4.0412*
Creatinine, µmol/L	0.363 ± 0.038	0.409 ± 0.061	0.478 ± 0.0856*
Cholesterol, mg/dL	91.300 ± 4.839	90.883 ± 3.311	88.533 ± 3.168

Note: *0.05 ($p \leqslant 0.05$); **0.01 ($p \leqslant 0.01$); ***0.01 ($p \leqslant 0.001$)
Source: developed by the authors

In carp that received natural feed components within the multitrophic aquaculture system in experimental Group A and Group B, the concentrations of glucose, creatinine, and cholesterol in the blood differed from those observed in the control group. At the same time, the correlation between the biochemical composition of the muscle tissue and that of the blood exhibited a high R^2 value (Tables 1, 2). From a physiological and biochemical perspective, these findings were consistent with the activation of compensatory mechanisms within the cyprinid organism.

Modern scientific and applied research increasingly demonstrated the relevance of combined systems in aquaculture, with particular emphasis on their environmental friendliness (Dyudyaeva, 2021). Experimental findings published by various researchers highlighted the growing recognition of the multitrophic approach in aquaculture (Can & Seyhaneyildiz Can, 2023). The importance of reducing nutrient emissions into the environment and rationally utilising the potential of such systems was emphasised. However, investigations in this area, particularly those concerning marine bioremediation, remained largely at the pilot stage. Analysis of the available publications indicated that the majority of multitrophic systems were developed under conditions of predominantly marine aquaculture, whereas land-based systems of this type exhibited greater efficiency compared with open-water systems (Batır *et al.*, 2025).

A combined approach in aquaculture met the modern requirements of the industry. One of its notable advantages was the potential to reduce the use of fishmeal and fish oil in aquaculture diets (Cao *et al.*, 2018; Harmantepe & Yılmaz, 2025). Previous research confirmed the positive effects of incorporating *Spirulina* and *Chlorella* (for example, replacing up to 25% of fishmeal) in *Cyprinus carpio* diets, resulting in enhanced growth, nutrient conversion, and digestive enzyme activity. General physiological parameters of the blood, as well as the immunity and resistance of juveniles, improved accordingly. The studies of H. Sabetmand *et al*. (2024) also demonstrated an increase in growth rate and immune response ($p < 0.05$), alongside a reduction in the feed conversion ratio, when *Spirulina* sp. and citric acid were used in carp diets. The influence of natural components on these parameters was similarly confirmed experimentally in the present study.

According to A. van Huis (2020), increasing attention in aquaculture was directed towards the use of the black soldier fly (*Hermetia illucens*) as a component capable of converting organic waste into nutritionally valuable feed ingredients, thereby enhancing the organism's metabolic activity. In the present experiment, the total protein level in the blood of cyprinids in the experimental groups increased simultaneously with protein accumulation in muscle tissues. This could be attributed to elevated protein synthesis and stimulated amino acid metabolism in fish receiving natural feed supplements. Meanwhile, cholesterol levels in the blood of the experimental fish were lower than those in the control and other experimental groups, which was also reflected in the reduced lipid content of the muscle tissue. A positive correlation was therefore established between body weight and blood composition parameters of cyprinids. Similar results were reported by P. Bryant & A. Matty (1980) and V. Serra *et al*. (2024), who demonstrated improved health, welfare, growth performance, and fillet quality in fish. These findings were further supported by research demonstrating enhanced product quality and sustainable protein utilisation in aquaculture diets (D'Souza *et al.*, 2006; Hua *et al.*, 2019). S. Maiolo *et al.* (2020) highlighted the practical significance of using natural protein-enriched components as alternative energy sources within environmentally sustainable production systems. Such approaches facilitated the development of aquaculture while simultaneously reducing environmental pressure, thereby contributing to the sustainable development of the industry. The findings of R. Chaklader *et al.* (2019) further substantiated that metabolic processes in aquatic organisms could be modified through dietary optimisation. The rational use of resources in aquaculture, alongside consideration for ecosystem balance and biodiversity, was confirmed by both scientific and applied studies in this field (Samarathunga *et al.*, 2023; Parsa Khanghah & Can, 2024). In addition to optimising technological processes in aquaculture, particular attention was devoted to feeding strategies and multitrophic production models.

Recent studies revealed positive effects of herbal preparations, beneficial bacteria, and probiotics on the general physiological condition of aquatic organisms. Positive findings in aquaculture and biotechnology literature emphasised the synergistic potential of such natural components. Furthermore, the implementation of integrated aquaculture management systems substantially increased production efficiency compared with traditional approaches and farming models.

Conclusions

The application of the proposed modular system incorporating elements of multitrophic aquaculture contributed to the production of aquaculture outputs that were as close as possible to environmentally friendly and safe standards. This effect was attributed to the use of natural components cultivated autonomously within a single modular system, which allowed full control of cultivation conditions and quality parameters throughout the entire production cycle. The integration of components with different trophic levels within the system provided a synergistic effect that promoted the rational use of resources. The inclusion of carp-based components as supplementary feed in experimental Group B resulted in higher developmental rates during ontogenesis. Overall, improvements in physiological and biochemical processes were observed in comparison with other experimental groups. The multitrophic system model demonstrated a high potential for stimulating crustacean development. Compared with the control group, the actual performance indicators in experimental Group A and Group B were higher, with Group B showing the most pronounced improvement. Increases relative to the control were recorded as follows: average body weight (1.21-7%), weight gain (2.9-6.9%), and feed conversion ratio (4.9-14.6%). In parallel, the blood composition of fish in the experimental groups improved, particularly in haematopoietic and protein-synthesising functions. The total number of erythrocytes and haemoglobin content increased by 13-36% and 3-13%, respectively, compared with the control. Total protein content increased by 5-32%. Correlation analysis confirmed strong relationships among the studied parameters (R^2 = 0.7-0.9). Future research should focus on a more detailed analysis of the biochemical composition of fish muscle tissue under the influence of technological factors, along with a comprehensive histomorphological evaluation.

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Conflict of Interest

The authors declare that they have no potential conflict of interest regarding the authorship or publication of this article.

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Оптимізація розвитку та фізіологічного статусу коропа в полікультурі за умов впровадження елементів мультитрофічної аквакультури

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Анотація. Метою дослідження було визначення впливу та аналіз ефективності використання технологічних факторів для формування їх синергічного ефекту при годівлі та вирощуванні молоді коропових риб. Дослідження базувалося на теоретичних та експериментальних лабораторних методах у рибництві. Результати дослідження продемонстрували, що альтернативні джерела білка (*Spirulina platensis*, *Artemia* та *Hermetia illucens*), включені до інтегрованої мультитрофічної системи аквакультури (МТА), значно покращують темпи росту та зменшують залежність від традиційних кормів. Культивування всіх компонентів в єдиній системі МТА забезпечувало максимальний контроль над якісними та кількісними характеристиками протягом усього циклу. Результати продемонстрували підвищення життєздатності організму молоді коропових риб, покращення складу їхньої крові в дослідних групах А та В. Однак, група В продемонструвала вищі якісні та кількісні параметри, що характеризувалося активацією гомеостатичної регуляції та процесів кровотворення. Покращення гематологічного профілю, маси тіла та складу м'язової тканини тісно корелювало з параметрами активації білкового та вуглеводного обміну та прискоренням онтогенезу. Риби в дослідних групах відрізнялися кращими параметрами, що визначають накопичення маси, метаболічні процеси (WG, FCR, HSI, VSI). Позитивна висока кореляція була зафіксована в дослідних групах А та B: маса тіла ↔ білок у м'язовій тканині (R^2 = 0,8-0,9); холестерин у крові ↔ жир у м'язовій тканині (R^2 = 0,8-0,7). В дослідній групі А: у крові коропа *Cyprinus carpio* (Linnaeus, 1758) зафіксовано вищий вміст еритроцитів та загального білка ($p \leqslant 0{,}05$). У крові коропа *Cyprinus carpio* (Linnaeus, 1758) групи В: вміст еритроцитів, МСН, загального білка, креатиніну ($p \leqslant 0{,}05$) та гемоглобіну (Hb) ($p \leqslant 0{,}01$) перевищував контрольну групу. У крові товстолобика *Hypophthalmichthys molitrix* / *Hypophthalmichthys nobilis* групи В: вміст еритроцитів, Hb, загального білка та креатиніну ($p \leqslant 0{,}05$) перевищував контрольну групу. Найкращий біохімічний склад м’язової тканини зафіксовано у риб з дослідних груп А та В

Ключові слова: годівля; вирощування; коропові; фізіолого-біохімічні процеси; мультитрофність; рециркуляційні системи