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THE HELMINTH FAUNA OF ALIEN FISH SPECIES IN THE REPUBLIC OF MOLDOVA

165.05. Parasitology

Summary of the doctoral thesis in biological sciences

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The thesis has been developed within the Doctoral School "Biological sciences" of the State University „Dimitrie Cantemir", Institute of Zoology

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The thesis defense will take place on **23rd of December, 2022**, at **10:00** at the meeting of the Doctoral Commission within the Doctoral School of Biological, Geonomic, Chemical and Technological Sciences, Senate Hall of the State University of Moldova, 3/2 Academiei str., Chișinău. (http://www.usm.md)

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CONTENT

CONCEPTUAL FRAMEWORK OF THE RESEARCH

Actuality and the importance of the research. The studying of the zonal particularities of the helminth fauna of alien fishes is of interest both for the formation of fish productivity in basins, and for solving some general biological problems. Until now, in our country, sufficient ichthyo-helminthological investigations have not been carried out, which could have elucidated the main problems related to this subject. Alien fishes are hosts to a wide variety of helminths, and their parasite fauna is a component of aquatic ecosystems. In recent years, one of the methods used to evaluate aquatic ecosystems is their parasitic status, used as an important bioindicator both of the biocenosis in general and for establishing the level of biological risk of these ecosystems [4].

Alien fishes - as an important component of biocenosis - are of particular scientific interest given the fact that they play the role of intermediate and definitive reservoir hosts for a high diversity of helminths. Thus, in some cases, alien fishes contribute not only to the contamination of domestic and wild animals, but also actively participate in the formation of parasitic zoonoses. This reflects the need to carry out an in-depth and multilateral study on this group of animals in the helminthological aspect [16].

The high diversity of the helminth fauna, the specificity of the biological cycle and the dependence of the helminths on the host species and the environment, as well as the high reproductive capacity of alien fish species are the factors that cause these parasitic worms to be very flexible and to react quickly through mechanisms of adaptation to the stabilization of ecosystems [1, 5].

The purpose of the research consists in the study of helminth fauna of alien and native fishes from various aquatic ecosystems in the Republic of Moldova, the evaluation of the impact of mono- and polyinvasions on some indices of the morpho-functional, productive status and the development of innovative methods to reduce and combat helminthiasis in common carp (*Cyprinus carpio* Linnaeus, 1758).

To achieve the proposed goal, the following **objectives** have been outlined:

1. Establishing the diversity of the helminth fauna of alien and native fish species from various aquatic ecosystems of the Republic of Moldova;

2. Evaluation of the impact of mono- and polyinvasions on some indices of the morpho-functional and biochemical status in Asian carp;

3. Highlighting the impact of mono- and polyinvasions on some productive indices in Asian carp;

4. Development of innovative procedures for the prevention and control of helminth infections in common carp (*Cyprinus carpio* Linnaeus, 1758);

Scientific novelty and originality. For the first time in the Republic of Moldova, a complex science practical study was carried out, which consists in: determination of the helminth fauna of some alien and native fishes from various natural and anthropogenic aquatic ecosystems with the establishment of specific and common helminths both for various species of carnivores, as well as for humans; for the

first time, the impact of mono- and poliyinvasions on some morpho-functional, biochemical, productive indices in Asian carp was evaluated, completed with the development, patenting and implementation of innovative procedures, for prophylaxis and control of their parasites.

Research methodology. Common fishing gear was used for the collection of fish material: stationary nets, drag-net, fishing rods etc. The works of several authors served as methodological support for the determination of fish and helminth species [1, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 17].

Theoretical significance. The diversity of fish helminth fauna in natural and anthropogenic aquatic ecosystems was evaluated, including the helminth fauna of alien fishes, as well as their role as definitive, intermediate, complementary and reservoir hosts for a wide spectrum of helminths, which parasitize in the larval stage in fish, reptiles, mollusks, crustaceans, chironomid larvae, and the adult stage in fish and piscivorous animals, including humans. Also, a major problem in the field of fish farming in the Republic of Moldova was addressed and solved, which made it possible to highlight the result of the action on the host organism of mono- and polyinvasions, to determine their impact on the quality of the products and to develop new procedures of prophylaxis and control of helminthiasis in common carp.

Applicative value. Based on the obtained results a methodological guidebook was edited; an invention patent was developed and innovative measures were implemented to prevent and combat helminthiasis in common carp; were determined innovative procedures for the reduction and recovery of economic damages in the fisheries and zoo veterinary sector, essential both for the reproduction, growth and development of fish in a healthy aquatic ecosystem, as well as for the interruption of the epizootic chain of transmission of common parasitic agents for birds, mammals and humans

Implementation of scientific results. The results of the study were implemented in fish farming practice and used in the training process of students, residents, and master students in various educational institutions.

Approval of results. The scientific results of the thesis were communicated and approved at various specialized scientific forums in the country and abroad: International symposium “Functional ecology of animal”, dedicated to the 70th anniversary from the birth of academician Ion Toderas, 21 September, Chisinau, 2018; The scientific conference of doctoral students “Contemporary trends in the development of science: visions of young researchers”, 8th Edition, June 10, 2019, Chisinau; The scientific conference of doctoral students “Contemporary trends in the development of science: visions of young researchers”, 9th Edition, June 10, 2020, Chisinau; 59th Annual Symposium “Towards a Global Health” 22-23 October, 2020, Iasi, Romania; 10th International Conference of Zoologists ”Sustainable use and protection of animal world in the context of climate change” dedicated to the 75th anniversary from the creation of the first research subdivisions and the 60th from the foundation of the Institute of Zoology, 16-17 September 2021, Chisinau, 2021.

Publications on the thesis topic. Based on the research results 11 scientific papers were published: 3 articles in scientific journals from the National Register of journals, category B; two articles

in scientific journals abroad; one article in proceedings of international scientific conferences (abroad); one article in the proceedings of scientific conferences with international participation (Republic of Moldova); 4 theses presented at scientific conferences (Republic of Moldova); a methodological guide was developed and an patent was obtained.

The volume and structure of the thesis. Annotation presented in Romanian, Russian and English, introduction, five chapters, general conclusions and practical recommendations, bibliography of 198 titles, 15 annexes, 116 pages of basic text, 31 figures, 23 tables.

Key words: helminth fauna, alien fish species, parasitic impact, aquatic ecosystems, productive indices, innovative procedures, prophylaxis, control.

THESIS CONTENT

1. THE DIVERSITY OF HELMINTH FAUNA IN FISH, THE IMPACT ON THE HOST ORGANISM, PROPHYLAXIS AND CONTROL MEASURES (Bibliographic review) represents a general analysis of scientific literature in the field of helminth fauna of fish from aquatic ecosystems in the Republic of Moldova and abroad (Romania, Ukraine, Russian Federation). Also, in chapter 1, the impact of parasites on the host organism and the measures to prevent and control the invasive diseases in fish were described.

Conclusion to the chapter 1

2. RESEARCH METHODOLOGY AND TECHNIQUES

For taxonomic identification of fish, classical methods were used both under field and laboratory conditions. Common fishing gear was used for the collection of fish material: stationary nets, drag-net, fishing rods etc. For the statistical processing of data, STATISTICA 12 and MICROSOFT EXCEL 2019 programs were used. The identification of fish and helminth species was carried out in the Laboratory of Parasitology and Helminthology of the Institute of Zoology, with methodological support from various authors [1, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 17].

Helminthological investigations were carried out on several species of alien and native fish from various natural and artificial water biotopes. The identified helminths examined and described in the thesis were confirmed in Romania (confirmation dated May 24-26, 2021) (table 2.1.).

Table 2.1. Parasitologically examined fishes from various aquatic biotopes

Fish species	Nr. of specimens	Aquatic biotope
Carassius gibelio (Bloch, 1782) – Prussian carp	260	Prut r. (Costesti-Stanca), Dniester r. (Dubasari reservoir), Lozova – "Codrii" Natural Reserve, Bac r. (Chisinau), the

		pond from Fagadau (d. Falesti), the lake from the Rose Valley Park (Chisinau).
Pseudorasbora parva (Temminck & Schlegel, 1846) – Stone moroko	74	Prut r. (Lopatnic r.), Bac r. (Chisinau), the lake from the Rose Valley Park (Chisinau), the lake from Village Museum (Chişinău).
Lepomis gibbosus (Linnaeus, 1758) – Pumpkinseed	87	Dniester r. (Dubasari reservoir /under the dam), the lake from the Rose Valley Park (Chisinau), the lake from Village Museum (Chisinau).
Cyprinus carpio Linnaeus, 1758 – Common carp	69	Lozova – "Codrii" Natural Reserve, the pond from Fagadau v. (Falesti d.).
Hypophthalmichthys molitrix (Valenciennes, 1844) – Silver carp	56	Lozova – "Codrii" Natural Reserve, the pond from Fagadau v. (Fălești d.).
Hypophthalmichthys nobilis (Richardson, 1845) – Bighead carp	51	Nimoreni-Pescarua Danceni S.A, the pond from Fagadau v. (Falesti d.).
Abramis brama (Linnaeus, 1758) – Freshwater bream	14	Dniester r., Dubasari reservoir (Malovata Veche v.).
Rhodeus amarus (Bloch, 1782) – European bitterling	106	Raut r. (Ustia v.).
Perca fluviatilis Linnaeus, 1758 – European perch	52	Prut r. (Racovat r, Draghiste r.), Dniester r. (Dubasari reservoir)
Scardinius erythrophthalmus (Linnaeus, 1758) – Rudd	5	Dniester r. (Criuleni), Dubasari reservoir (Malovata Veche v.).
Blicca bjoerkna (Linnaeus, 1758) – White bream	6	Prut r. (Slobozia Mare v.)
Neogobius melanostomus (Pallas, 1814) – Round goby	12	Dniester r. (Criuleni, Dubasari reservoir)
Rutilus rutilus (Linnaeus, 1758) – Roach	12	Dubasari reservoir (Malovata Veche v.), Prut r. (Costesti-Stanca).
Proterorhinus semilunaris (Heckel, 1837) – Western tubenose goby	25	Dniester r. (Criuleni, Dubasari reservoir)
Neogobius fluviatilis (Pallas, 1814) – Monkey goby	14	Dniester r. (Criuleni, Dubasari reservoir)
Ponticola kessleri (Günther, 1861) – Bighead goby	14	Dniester r. (Criuleni, Dubasari reservoir)
Perccotus glenii Dybowski, 1877 – Chinese sleeper	133	Prut r. (Lopatnic r.).
Alosa immaculata Bennett, 1835 – Pontic shad	60	Dniester r. (s. Olanesti)
Total	1050	

To establish an accurate diagnosis, the method of total evisceration, according to K. I. Skryabin, and the method of parasitological examination, according to V.A. Doghel, and modified by I.E. Bykhovskaya – Pavlovskaya were used [6].

The modified method according to Doghel consisted in:

Examination of the integument and fins. From the integument and fins mucus was collected with the help of a scalpel, and squash slides were made. The squash slides were examined initially under the

stereoscopic microscope, then under the optical microscope, to detect ectoparasites from the Gyrodactilidae family.

Examination of the gills. Branchial arches were extracted from the branchial cavity to collect parasites visible to the naked eye. To detect ectoparasites from the Dactylogyridae and Diplozoidae families, tissue samples were collected from the gill filaments, squash slides were made and examined under a microscope.

Examination of the eyeball. The eyeball was eviscerated, then, by means of scissors, the eyeball was dissected, from which the lens and the vitreous body were later extracted. The gelatinous layer was scraped from the lens and squash slides were made, which were examined under a microscope, to detect metacercariae of the genus *Diplostomum* (*Diplostomum spathaceum*). Squash slides were prepared from the vitreous body which were examined under a microscope to detect the metacercariae of the genus *Tylodelphys*.

Examination of the abdominal cavity. The abdominal wall was removed by making 3 incisions, so that the internal organs were visible. The first incision was made ventrally from the anus to the pelvic fins, then dorsally to the lateral line, and from the lateral line to the anus. Helminths from the Ligulidae family and nematodes from the *Eustrongylides* genus were detected in the abdominal cavity.

Examination of heart. The heart was removed and placed in a Petri dish with saline solution (0,9%). The heart chambers were dissected, then washed with saline solution, and the resulting sediment was examined under a microscope to detect the trematodes of the genus *Sanguinicola*.

Examination of liver. The liver was separated into small portions, which were used to prepare squash slides. The squash slides were examined under a stereomicroscope, then under an optical microscope. To detect the larvae of the Gryporhynchidae family, the gall bladder was dissected, the bile was collected and examined under the microscope.

Squash slides were prepared from the pancreas, spleen, swim bladder, kidneys, ureters, urinary bladder and examined under the optical and stereo microscope.

Examination of the gonads. To detect parasites, squash slides were prepared from the collected tissue and examined under the optical and stereo microscope.

Examination of the gastrointestinal tract. The esophagus, stomach, pyloric caeca, and intestine were freed from the mass of liver tissue, after which the respective portions, in the order listed above, were sectioned longitudinally. The contents of the compartments of the gastrointestinal tract were used to prepare squash slides, and the helminths detected in the digestive tract were washed from the digestive content in saline solution (0,9%), after which they were preserved in 70% ethanol or 4% formalin.

Examination of muscle tissue. To detect the metacercariae stage, transverse sections were made in the muscle tissue. For this purpose, small portions of muscle tissue were compressed and investigated under a microscope.

In the case of detected macro parasites, the total number was determined, and in the case of monogeneans, metacercariae stage, the total number was determined by counting them in ten microscopic fields. The prevalence and intensity of invasion were determined.

Various chemical substances were used to preserve the parasites. Nematodes were preserved in Barbagallo solution (1000 ml distilled water+30 ml formaldehyde+9 g NaCl). Cestodes, trematodes and acanthocephalans were initially introduced in saline solution, and then passed in 70% ethanol.

In order to evaluate the impact of mono - and polyinvasions on some hematological and biochemical indices in Common carp, Prussian carp, Silver carp and Bighead carp, blood samples were taken by caudal vein puncture. For the hematological examination, the collected blood was preserved in tubes with the anticoagulant EDTA (ethylenediaminetetraacetic acid), and for the biochemical examination, the blood was preserved in serum-separating tubes.

To evaluate the impact of mono- and polyinvasions on some quality indices of the muscle tissue, the following methods were used: Kjeldhal method - for determining the protein content, Soxhlet method for detecting the fat content [3].

Conclusions at the chapter 2

1. Common fishing gear was used for the collection of fish material: stationary nets, drag-net, fishing rods. For helminthological investigations, the fish material was collected between march-may, june-august, september-november.
2. Helminthological investigations were carried out on alien and native fish specimens from various natural and artificial water bodies: Dniester r. (Dubasari reservoir, Olanesti v.), Raut r. (Ustia v.), Prut r. (Costesti Stanca, Slobozia Mare v.), Racovat r. (Branzeni v.), Draghiste r. (Branzeni v.), Lopatnic r. (Briceni), Bac r. (Chisinau), the lake from the Rose Valley Park, the lake from Village Museum (Chisinau), the pond from Natural reserve “Codrii” (Lozova v.), the pond from Pescarus S.A (Nimoreni v.).
3. The helminthological investigations were carried out according to the classical methods accepted in parasitology, after a prior collection of blood from the caudal vein, to carry out the hematological and biochemical examination.
4. For the statistical processing of data, STATISTICA 12 and MICROSOFT EXCEL 2019 programs were used.

3. THE HELMINTH FAUNA OF ALIEN FISH SPECIES FROM VARIOUS AQUATIC ECOSYSTEMS OF THE REPUBLIC OF MOLDOVA

Helminthological investigations were carried out on certain species of alien and native fish from various natural and artificial water bodies. A total of 1050 fish specimens were parasitologically examined.

The study of the helminth fauna of *Carassius gibelio* (Bloch, 1782) – Prussian carp from the Dniester river (Dubăsari reservoir - Criuleni) highlighted the infestation with helminths from: class Monogenea – 2 species: *Dactylogyrus* sp. (EI-38,57%, II-1-64 sp.), *Eudiplozoon nipponicum* (EI-4,28%, II-1 sp.), class Trematoda - 2 species - *Phyllodistomum folium* (EI-2,85%, II-1-3 sp.), *Diplostomum spathaceum* (EI-21,4%, II-1-6 sp.). Monoinvasions were present in 47,07% of cases, and polyinvasions in 52,93% of cases. As a result of the study of the helminth fauna of Prussian carp from Prut river (Costești-Stânca) the following species of helminths were detected: class Monogenea – 1 species: *Dactylogyrus* sp. (EI-27,27%, II-1-5 sp.), class Cestoda 3 species - *Paradilepis scolecina* (EI-11,36%, II-2-6 sp.), *Khawia sinensis* (EI-2,27%, II-1 specimen), *Bothriocephalus opsariichthydis* (EI-2,27%, II-1 specimen); class Chromadorea – 1 species: *Raphidascaris acus* (EI-6,81%, II-1 specimen). Parasitic invasions represented by only one species of helminths were present in 25,0% of cases, and in 75,05% of cases polyinvasions were detected. The polyinvasions in Prussian carp from Prut river (Costești-Stânca) were formed by associations of two species of helminths (*Dactylogyrus* sp. + *Paradilepis scolecina*) (figure 3.1.).

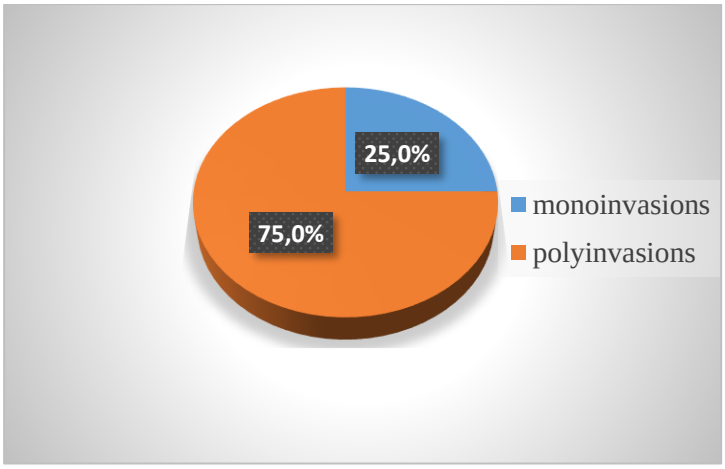

Fig. 3.1. Mono – and polyinvasions in *Carassius gibelio* (Bloch, 1782) – Prussian carp, from the Prut river (Costesti-Stanca)

The helminth fauna of Prussian carp from Bâc river consisted of helminths from the following classes: class Monogenea – 3 species: *Dactylogyrus* sp. (EI-100%, II-29-153 sp.), *Gyrodactylus* sp. (EI-57,89%, II-4-15 sp.), *Diplozoon paradoxum* (EI-36,84%, II de 1-7 sp.); class Trematoda – 1 species: *Diplostomum spathaceum* (EI-63,15%, II-1-3 sp.); class Enoplea – 1 species: *Pseudocapillaria tomentosa* (EI-18,94%, II-2-5 sp.). Of the total species of helminths detected in the specimens of Prussian carp from Bîc river, 60% were represented by geohelminths (*Dactylogyrus* sp., *Gyrodactylus* sp., *Diplozoon paradoxum*), and 40% - by biohelminths (*Diplostomum spathaceum*, *Pseudocapillaria tomentosa*).

As a result of parasitological examination of fishes from the pond located in Fagadau village (Falesti district), was established that the helminth fauna of Prussian carp specimens consisted of: class Monogenea – 2 species: *Dactylogyrus extensus* (EI-100%, II-1-45 sp.), *Eudiplozoon niponicum* (EI-16,0%, II-1-2 sp.); class Trematoda – 3 species: *Diplostomum spathaceum* (EI-20,0%, II-1-8 sp.),

Phillodistomum folium (EI-0,25%, II-1 specimen), *Aspidogaster limacoides* (EI-4,0%, II-1 specimen); class Cestoda – 1 species: *Khawia sinensis* (EI-4,0%, II-1 specimen.); class Chromadorea – 1 species: *Philometroides sanguinea* (EI-4,0%, II-2 sp.). It was also noted that monoinvasions predominated (53,3%) in the examined specimens.

The species *Pseudorasbora parva* (Temminck, Schlegel, 1846) – Stone moroko from Bac river was infested with the cestodes *Paradilepis scolecina*, *Valipora campylancristrota* and the nematode *Pseudocapillaria tomentosa*. *Paradilepis scolecina* and *Valipora campylancristrota* were found in the intermediate host as larvae. The extensivity and intensity of invasion with these helminths was: *Paradilepis scolecina* (EI-30,0%, II-1-2 sp.), *Valipora campylancristrota* (EI-30%, II-1 specimen), *Pseudocapillaria tomentosa* (EI-70,0%, II-1-3 sp.). Parasitic invasions in 90,0% of cases were represented by monoinvasions (in one case *Pseudocapillaria tomentosa* was detected, and in another one *Valipora campylancristrota*), and in 10,0% of cases by polyinvasions (*Paradilepis scolecina*+*Valipora campylancristrota*+*Pseudocapillaria tomentosa*).

Parasitic invasions in Stone moroko from Lopatnic river and the Village Museum lake were represented by monoinvasions. In the captured specimens, sporadic cases of infestation with the trematode *Posthodiplostomum cuticola* and the nematode *Schulmanela petruschewski* were found.

The helminthological study of 133 de specimens of *Perccottus glenii* Dybowski, 1877 – Chinese sleeper from Lopatnic river revealed the presence of the nematode *Schulmanela petruschewski* Schulman, 1948, which was detected in the liver and intestine. The prevalence was 53,58% and the intensity of invasion, 5-20 specimen per fish.

As a result of parasitological examination of *Lepomis gibbosus* (Linaeus, 1758) – Pumpkinseed collected from Village Museum lake, was established a level of infestation with the following helminths: *Diplostomum spathaceum* located in the lens of the eyeball (EI-2,98%, II-1 specimen) and *Schulmanela petruschewski* (EI-58,20%, II-1-7 sp.). The parasitic invasions, consisting of a species of helminths, were present in 5,13% (cases of infestation with *Schulmanela petruschewski*), and in 94,97% of the examined specimens, the parasitic invasions were represented by polyinvasions.

In the specimens of Pumpkinseed fish collected from the Rose Valley lake, unlike those collected from the Village Museum lake, only the helminth *Diplostomum spathaceum* was detected (EI-81,25%, II-1-7 sp.).

The study of the helminth fauna of the species *Hypophthalmichthys molitrix* (Valencienne, 1844) – Silver carp from the pond of "Codrii" Natural Reserve, highlighted its infestation with the larval stage of trematodes of the genus *Diplostomum* von Nordman, 1832 with localization in the lens of the eyeball. The extensivity of invasion with this helminth reached 60,0%, and the intensity of invasion 1-130 specimens per fish. In the specimens of Silver carp from the pond of Fagadau v. (Falesti d.) the following helminths were detected: class Monogenea – 2 species: *Dactylogyrus* sp. (EI-92,30%, II-1-45 sp.), *Eudiplozoon niponicum* (EI-38,46%, II-1-7 sp.); class Trematoda – 2 species: *Diplostomum spathaceum*

(EI-30,76%, II-1-4 sp.), *Posthodiplostomum cuticola* (EI-7,69%, II-5 sp.); class Cestoda – 2 species: *Valipora campylancristrota* (EI-7,69%, II-1 specimen), *Khawia sinensis* (EI-46,15%, II-1-13 sp.).

As a result of the parasitological study the helminth fauna of the species *Hypophthalmichthys nobilis* (Richardson, 1845) – Bighead carp from the fishery Pescarus S.A it was established the infestation with the following helminths: class Monogenea – 1 species: *Dactylogyrus* sp. located on the gill filaments (EI-91,67%, II-2-28 sp.); class Trematoda – 1 species: *Diplostomum spathaceum* located in the lens of the eyeball (EI-44,44%, II-2-38 sp.); class Cestoda – 2 species: *Paradilepis scolecina* located in the liver (EI-8,33%, II-1 specimen), *Valipora campylancristrota* located in the gall bladder (EI-44,44%, II-1-6 ex.). Parasitic invasions consisting of a species of helminths were present in 27,27% of the examined fishes, and in 72,73% of the examined fishes, the parasitic invasions were represented by polyinvasions (figure 3.2.).

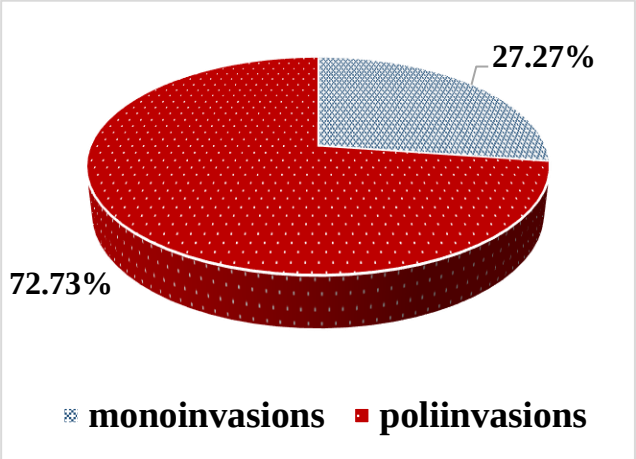

Fig. 3.2. Mono- and polyinvasions in *Hypophthalmichthys nobilis* (Richardson, 1854) – Bighead carp, from the fishery Pescarus S.A

In Bighead carp specimens collected from the pond fishery from Fagadau (Fălești d.) helminths were detected from following classes: Monogenea – 1 species: *Dactylogyrus* sp. (EI-100%, II-74-160 sp.); Trematoda – 1 species: *Diplostomum spathaceum* (EI-100%, II-1-92 sp.); Cestoda 3 species – *Valipora campylancristrota* (EI-44,44%, II-1-3 sp.), *Bothriocephalus opsariichthydis* (EI-9,09%, II-1 specimen), *Schyzocotyle acheilognathi* (EI-9,09%, II-1 specimen). All specimens were infested as polyinvasion.

Investigation of the helminth fauna of *Cyprinus carpio* Linnaeus, 1758 – Common carp from the pond of Fagadau (Falesti) allowed to established invasions with parasites from: class Monogenea – 2 species: *Dactylogyrus extensus* (EI-92,30%, II-1-45 sp.), *Eudiplozoon nipponicum* (EI-38,46%, II-1-7 sp.); class Trematoda – 2 species: *Diplostomum spathaceum* (EI-30,76%, II-1-4 sp.), *Posthodiplostomum cuticola* (EI-7,69%, II-5 sp.); class Cestoda – 2 species: *Valipora campylancristrota* (EI-7,69%, II-1 specimen), *Khawia sinensis* (EI-46,15%, II-1-13 sp.). Of all the parasitologically examined specimens, 92,3% were polyinfected.

In the examined specimens of the species *Abramis brama* (Linnaeus, 1758) – Freshwater bream from Dubasari reservoir, unique cases of infestation with the cestode *Ligula intestinalis*, and the trematode *Diplostomum spathaceum* were detected. The intensity of invasion with *Diplostomum spathaceum* and *Ligula intestinalis* in the examined Freswater bream was 2-6 specimens and, respectively, 3 specimens per fish.

In the specimens of *Blicca bjoerkna* (Linnaeus, 1758) – White bream, collected from the Beleu lake, infestations with the trematodes from the family *Diplostomidae* - *Diplostomum spathaceum* (II-10-50 sp.) and *Posthodiplostomun cuticola* (II-3-18 sp.) were identified.

In a specimen of *Rutilus rutilus* (Linnaeus, 1758) – Roach collected from the Costesti-Stanca reservoir the trematodes *Diplostomum spathaceum* (II-9 sp.), *Tylodelphys clavata* (II-9 ex.) (located in the vitreous body of the eyeball), *Aspidogaster limacoides* (II-6 sp.) (found in the intestine) were registered. In another specimen of Roach collected from the Dubasari reservoir (Malovata-Veche) the intensity of invasion with these helminths was: *Diplostomum spathaceum* (II-100-120 ex.), *Tylodelphys clavata* (II-48 ex.), *Aspidogaster limacoides* (II-13 ex.).

The helminthological examination carried out on the specimens of *Rhodeus amarus* (Bloch, 1782) – European bitterling from the Raut river - highlighted the infestation with helminths from: class Trematoda – 4 species: *Diplostomum spathaceum* located in the lens of the eyeball (EI-50,47%, II 1-8 sp.), *Clinostomum complanatum* located in subcutaneous tissue and muscle tissue (EI-18,1%, II-1-7 sp.); *Apophallus* sp. found on the caudal fin, in approximately the majority of the examined specimens, with an intensity of invasion of 1-7 sp.; *Phyllodistomum* sp. located in the ureter (EI-0,95%, II-3 sp.)); class Enoplea – 1 species: *Pseudocapillaria tomentosa* located in the intestine (EI-79,04%, II-1-9 sp.)). Out of the total examined species, 12,90% were monoinfected (specimens infested only with *Apophallus* sp.), and 79,04% of specimens were polyinfected (figure 3.3.).

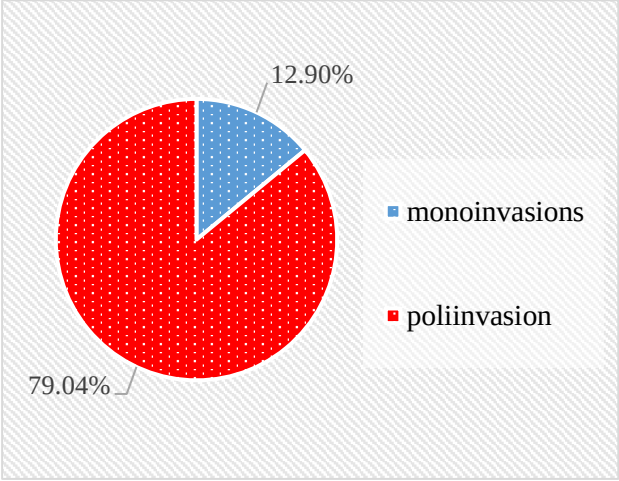

Fig. 3.3. Mono- and polyinvasions in *Rhodeus amarus* (Bloch, 1782) – European bitterling from the Raut river

As a result of the study of the helminth fauna of the species *Perca fluviatilis* Linnnaeus, 1758 - European perch from the Dubasari reservoir - it was established that it was infested with parasitic agents from: class Trematoda – 5 species: *Bunodera luciopercae* located in the pyloric caeca an instestine (EI-10,52%, II-1-2 sp.), *Ichthyocotylurus* sp. located on the walls of the swim bladder and on the kidneys (EI-89,47%, II-1-24 sp.), *Apophallus* sp. located on the skin, fins, opercula's, in the superficial muscle tissue, with EI-78,94% and II-dozens of specimens, *Diplostomum spathaceum* located in the lens of the eyeball (EI-10,52%, II-1 specimen), *Tylodelphys clavata* in the vitreous body of the eyeball (EI-26,31%, II-3-9 sp.); class Enoplea – 1 species: *Eustrongylides* sp. encapsulated on the mesentery and internal organs (EI-21,05%, II-1-9 sp.).

The study of the helminth fauna of the European perch from the Racovat river highlighted its infestation with parasitic agents classified as: class Trematoda – 2 species: *Ichthyocotylurus* sp. located on the walls of the swim bladder and on the kidneys (EI-16,66%, II-1-2 sp.), *Diplostomum spathaceum* located in the lens of the eyeball (EI-5,55%, II-1 specimen); class Palaeacanthocephala – 2 species: *Acanthocephalus lucii* located in the pyloric caeca and intestine (EI-38,88%, II-1-4 sp.), *Pomphorhynchus laevis* located in the intestine (EI-16,66%, II-3-11 sp.). Parasitic invasions formed by a single species of helminths were present in 58,34% of the examined specimens, and in 41,66% of the specimens the invasions were represented by polyinvasions. The helminth fauna of European perch from the Draghiște river was represented by the species *Clinostomum complanatum* located in the abdominal cavity (EI-25,0%, II-1 specimen), *Ichthyocotylurus* sp. located on the walls of the swim bladder and on the kidneys (EI-37,5%, II-2-12 sp.), *Acanthocephalus lucii* located in the intestine (EI-37,5%, II-1-2 sp.). Monoinvasions were found in 33,33% of cases, and polyinvasions in 66,67% of cases.

The helminth fauna of *Neogobius fluviatilis* (Pallas, 1814) – Monkey goby from the Dniester river (Dubasari reservoir) was represented by helminths from: class Trematoda – 2 species: *Diplostomum spathaceum* located in the lens of the eyeball (EI-7,14%, II-219 sp.), *Paracoenogonimus ovatus* located in the muscle tissue (EI-21,42%, II-3-18 sp.); class Enoplea – 1 species: *Eustrongylides* sp. located in the abdominal cavity, on the organs of the digestive system (EI-7,14%, II-1 specimen); class Palaeacanthocephala – 1 species: *Pomphorhynchus laevis* located in the abdominal cavity, encapsulated on the internal organs of the digestive system (EI-21,42%, II-1-13 sp.) Monoinvasions were present in 87,5% of the examined specimens, and in 12,5% of the examined specimens the parasitic invasions were represented by polyinvasions.

The study of the helminth fauna of the species *Ponticola kessleri* (Günther, 1861) – Bighead goby revealed the infestation with the following helminths: class Trematoda – 5 species: *Diplostomum spathaceum* (EI-64,28%, II-9-20 sp.), *Paracoenogonimus ovatus* located in the muscle tissue (EI-71,42%, II- several dozen specimens), *Asymphylodora imitans* located in the intestine (EI-85,71%, II-83-128 sp.), *Phyllodistomum angulatum* located in the urinary blade (EI-7,14%, II-1 specimen); class Enoplea – 1 species: *Eustrongylides* sp. located in the abdominal cavity, on the internal organs (EI-64,28%, II-1-4 sp.);

class Palaeacanthocephala – 1 species: *Pomphorhynchus laevis* (EI-85,71%, II-1-20 sp.). All the examined specimens of Bighead goby were polyinfected.

The helminth fauna of the species *Neogobius melanostomus* (Pallas, 1814) – Round goby from the Dniester river (Dubasari reservoir) was represented by helminths from: class Trematoda – 2 species: *Diplostomum spathaceum* (EI-66,67%, II-3-14 sp.), *Paracoenogonimus ovatus* (EI-33,33%, II-9-21 sp.); class Enoplea – 1 species: *Eustrongylides* sp. located in the abdominal cavity, on the internal organs (EI-8,33%, II-2 sp.); class Palaeacanthocephala – 1 specie: *Pomphorhynchus laevis* (EI-83,33%, II 3-45 sp.). Monoinvasions were present in 33,3% of the examined specimens, and in 66,7% of the examined specimens the parasitic invasions were represented by polyinvasions.

The ichthyo-parasitological researches carried out on the species *Proterorhinus semilunaris* (Heckel, 1837) – Western tubenose goby from the Dniester river (Dubasari reservoir) highlighted the infestation with the following parasites: class Trematoda – 4 species: *Diplostomum spathaceum* (EI-36,0%, II 2-160 sp.), *Paracoenogonimus ovatus* (EI-20,0%, II-22-40 sp.), *Ichthyocotylurus* sp. encapsulated in the eyeball (EI-4,0%, II-4 sp.); *Asymphylodora imitans* located in the stomach (EI-28,0%, II-3-56 sp.); class Enoplea – 1 species: *Eustrongylides* sp. (EI-40,0%, II-1-10 sp.); class Palaeacanthocephala – 1 species: *Pomphorhynchus laevis* (EI-72,0%, II-3-17 sp.). Monoinvasions were present in 16,67% of the examined specimens, and in 83,3,7% of the examined specimens the parasitic invasions were represented by polyinvasions

As a result of the parasitological examination of 60 specimens of *Alosa immaculata* Bennet, 1835 – Pontic shad captured from the lower Dniester, 3 species of helminths were detected: 2 species of trematodes - *Pronoprymna ventricosa* (Rudolphi, 1819) Poche, 1926, *Lecithasther confusus* Odhner, 1905 which were found in the pyloric caeca, and 1 species de nematodes - *Hysterothylacium aduncum* (Rudolphi, 1802) found in the stomach. The degree of infestation with these helminths was: *Pronoprymna ventricosa* (EI-20,0%, II-2-18 sp.), *Lecythaster confusus* (EI-7,8%, II-1-12 sp.), *Hysterothylacium aduncum* (EI-85%, II-1-80 sp.).

Conclusions at the chapter 3

1. It was established that the helminth fauna of alien and native fish species in the analyzed water basins is represented by 31 species of helminths systematically classified into different classes: class Monogenea – 4 species: *Dactylogyrus* sp., *Gyrodactylus* sp., *Diplozoon paradoxum*, *Eudiplozoon nipponicum*; class Trematoda – 15 species: *Asymphylodora imitans*, *Bunodera lucioperca*, *Nicolla skrjabini*, *Aspidogaster limacoides*, *Phyllodistomum angulatum*, *Pronoprymna ventricosa*, *Lecithaster confusus*, *Phyllodistomum folium*, *Diplostomum spathaceum*, *Posthodiplostomum cuticola*, *Tylodelphys clavata*, *Apophallus* sp., *Clinostomum complanatum*, *Ichthyocotilurus* sp., *Paracoenogonimus ovatus*; class Cestoda – 5 species: *Ligula intestinalis*, *Khawia sinensis*, *Bothriocephalus opsariichthydis*, *Paradilepis scolecina*, *Valipora*

campylancristrota; class Enoplea – 3 species: *Hepaticola petruschewskii*, *Pseudocapillaria tomentosa*, *Eustrongylides* sp.; class Chromadorea – 2 species: *Raphidascaris acus*, *Hysterothylacium aduncum*; class Palaecanthocephala – 2 species: *Acanthocephalus lucii*, *Pomphorynchus laevis*.

2. Of the total number of helminths detected in the examined alien and native fish specimens predominated: *Diplostomum spathaceum* (Prussian carp from the Dubasari reservoir (EI-21,4%), Prussian carp from the Bac river (EI-63,15%), Prussian carp from the pond of Fagadau v. (EI-20%), Pumpkinseed fish from the Village Museum lake (EI-2,98%), Pumpkinseed fish from the Rose Valley lake (EI-81,25%), Silver carp from the "Codrii'' Natural Reserve (EI-60%), Silver carp from the pond of Fagadau v. (EI-30,76%), Bighead carp from the fishery Pescarus S.A. (EI-44,44%), Bighead carp from the pond of Fagadau v. (EI-100%), Common carp from the pond of Fagadau v. (EI-30,76%), European bitterling from the Raut river (EI-50,47), European perch from the Dubasari reservoir (EI-10,52%), European perch from the Racovat river (EI-5,55%), Monkey goby from the Dubasari reservoir (EI-7,14%), Bighead goby from the Dubasari reservoir (EI-64,28%), Round goby from the Dubasari reservoir (EI-66,67%), Western tubenose goby from the Dubasari reservoir (EI-36,0%)), *Dactylogyrus* sp. (Prussian carp from the Dubasari reservoir (EI-38,57%), Prussian carp from the Prut river (EI-27,27%), Prussian carp from the Bac river (EI-100%), Prussian carp from the pond of Fagadau v. (EI-100%), Silver carp from the pond of Fagadau v. (EI-92,30%), Bighead carp from the fishery Pescarus S.A. (EI-91,67%), Bighead carp from the pond of Fagadau v. (EI-100%), Common carp from the pond of Fagadau v. (EI-92,30%)), *Eustrongylides* sp. (European perch from the Dubasari reservoir (EI-21,05%), Western tubenose goby from the Dubasari reservoir (40,0%), Monkey goby from the Dubasari reservoir (EI-7,14%), Bighead goby from the Dubasari reservoir (EI-64,28%), Round goby from the Dubasari reservoir (EI-8,33%)), *Pomphorhynchus laevis* (European perch from the Racovat river (EI-16,66%), Monkey goby from the Dubasari reservoir (EI-21,42%), Bighead goby from the Dubasari reservoir (EI-85,71%), Round goby from the Dubasari reservoir (EI-83,33%), Western tubenose goby from the Dubasari reservoir (EI-72%)).
3. The most infested native fishes were European perch – 9 species (*Bunodera luciopercae*, *Diplostomum spathaceum*, *Tylodelphys clavata*, *Clinostomum complanatum*, *Apophallus* sp., *Ichthyocotylurus* sp., *Eustrongylides* sp., *Acanthocephalus lucii*, *Pomphorhynchus laevis*), Bighead goby – 7 species, (*Diplostomum spathaceum*, *Paracoenogonimus ovatus*, *Asymphylodora imitans*, *Nicolla skrjabini*, *Phyllodistomum angulatum*, *Eustrongylides* sp., *Pomphorhynchus laevis*), Western tubenose goby – 6 species (*Diplostomum spathaceum*, *Paracoenogonimus ovatus*, *Asymphylodora imitans*, *Ichthyocotylurus* sp., *Eustrongylides* sp., *Pomphorhynchus laevis*).
4. The most infested alien fishes were Prussian carp – 12 species (*Dactylogyrus* sp., *Gyrodactylus* sp., *Diplozoon paradoxum*, *Eudiplozoon nipponicum*, *Phyllodistomum folium*, *Diplostomum*

spathaceum, *Bothriocephalus opsariichthydis*, *Khawia parva*, *Paradilepis scolecina*, *Philometroides sanguinea*, *Pseudocapillaria tomentosa*, *Raphidascaris acus*) and Stone moroko – 5 species (*Posthodiplostomum cuticola*, *Paradilepis scolecina*, *Valipora campylancristrota*, *Pseudocapillaria tomentosa*, *Shulmanela petruschewski*). The less infested alien fishes were silver carp – 2 species (*Ligula intestinalis*, *Diplostomum spathaceum*), Pumpkinseed fish– 2 species (*Diplostomum spathaceum*, *Shulmanela petruschewskii*), Chinese sleeper – 1 species (*Shulmanela petruschewskii*).

5. Polyinvasions were estimated to be prevalent in the following fish species: Prussian carp from the Dniester river (monoinvasions – 47,07%, polyinvasions – 52,93%), Prussian carp from the Prut river (monoinvasions – 25,0%, polyinvasions – 75,0%), Stone moroko from the Bac river (monoinvasions – 10,0%, polyinvasions – 90,0%), Pumpkinseed from the Village Museum lake (monoinvasions – 5,13%, polyinvasions – 94,87%), Bighead carp from the fishery S.A Pescarus (monoinvasions – 27,17%, polyinvasions – 72,73%), European bitterling from the Raut river (monoinvasions – 13,0%, polyinvasions – 79,0%), European perch from the Dniester river (Dubasari reservoir) (monoinvasions – 10,52%, polyinvasions – 89,47%), European perch from the Racovat river (monoinvasions – 58,33%, polyinvasions – 41,66%), European perch from the Draghiste river (monoinvasions – 33,35%, polyinvasions – 66,67%), Monkey goby from the Dniester river (Dubasari reservoir) (monoinvasions – 87,55%, polyinvasions – 12,5%), Bighead goby from the Dniester river (Dubasari reservoir) (polyinvasions – 100%), Round goby from the Dniester river (Dubasari reservoir) (monoinvasions – 33,3%, polyinvasions – 66,67%), Western tubenose goby from the Dniester river (Dubasari reservoir) (monoinvasions – 16,67%, polyinvasions – 83,3%).
6. Of the detected helminth species, common to human and carnivorous mammals were: *Clinostomum complanatum* and *Apophallus* sp. found in European perch and European bitterling specimens; *Paracoenogonimus ovatus* and *Eustrongylides* sp. found in Monkey goby, Bighead goby, Round goby and Western tubenose goby specimens.

4. THE IMPACT OF MONO- AND POLYINVASIONS ON SOME INDICES OF THE MORPHO-FUNCTIONAL AND BIOCHEMICAL STATUS IN SOME ASIAN CARPS

In order to evaluate the impact of mono- and polyinvasions on the hematological and biochemical indices, blood was collected from 4 species of fish: Common carp, Prussian carp, Silver carp and Bighead carp. After the blood has been collected the parasitological examination of the internal organs and muscle tissue was carried out to establish the degree of infestation. The Common carp and Prussian carp specimens were divided into four groups: group I – uninfested, group II – specimens infested with the monogenean *Dactylogyrus extensus* (monoinvasions), group III – specimens infested with the cestode

Khawia sinensis (monoinvasions), group IV – specimens infested with *Dactylogyrus extensus* and *Khawia sinensis* (polyinvasions).

Also, the specimens of Silver carp and Bighead carp were divided in four groups: group I – uninfested, group II – specimens infested with the monogenean *Dactylogyrus* sp. (monoinvasions), group III – specimens infested with the trematode *Diplostomum spathaceum* (monoinvasions), group IV – specimens infested with *Dactylogyrus* sp. and *Diplostomum spathaceum* (polyinvasions).

For this purpose, the following hematological and biochemical indices were analyzed: hemoglobin, red blood cells, hematocrit, white blood cells, procalcitonin, total protein, triglycerides, cholesterol, glucose, bilirubin, uric acid, ALT and AST.

As a result of the evaluation of the impact of mono - and polyinvasions on the hematological indices in the examined specimens of Asian carp, it was established that, in the polyinfested groups, the level of hemoglobin, hematocrit, procalcitonin, as well as the number of red blood cells decrease, compared to the non-infested group. In the polyinfested group, significant decrease in the number of red blood cells was established compared to group I. Thus, the number of red blood cells was reduced by 26,8% in Common carp, by 41,45% in Prussian carp, by 15,02% in Silver carp and by 29,06 %, in Bighead carp (figure 4.1.).

The level of procalcitonin in the blood of polyinfested specimens was reduced by 55,56% in Common carp, by 50,0% in Prussian carp, by 12,22% in Silver carp and by 28,0% in Bighead carp.

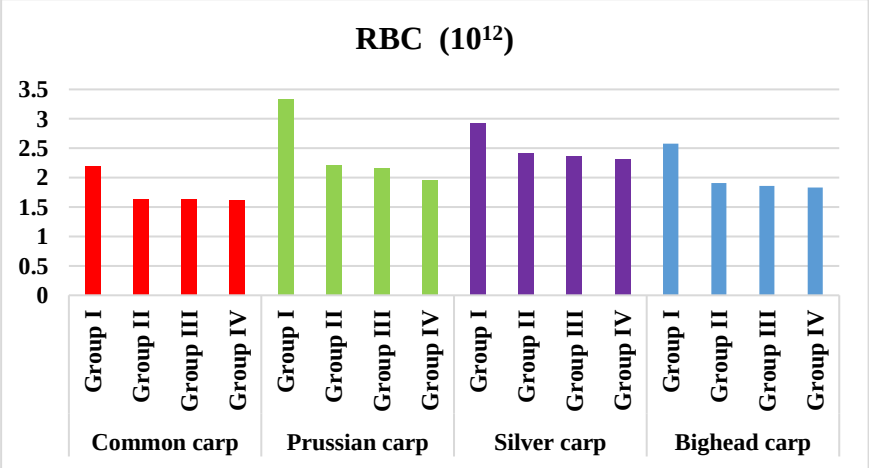

Fig. 4.1. Variation in the number of red blood cells depending on the level of infestation in Asian carp

As a result of the biochemical examination of the blood of mono – and polyinfested specimens, a decrease in total protein, triglycerides and glucose was established compared to the uninfested group. Among these indices, the lowest level was that of glucose, which decreased by 43%, and total protein, which decreased by approx. 25% compared to the uninfested group (figure 4.2.).

Contrary to the indices mentioned above, higher values of transaminases (ALT, AST) were established. The level of ALT and AST, unlike the uninfested group , was higher by 25,0% and 8,0%, respectively (figures 4.3-4.4.).

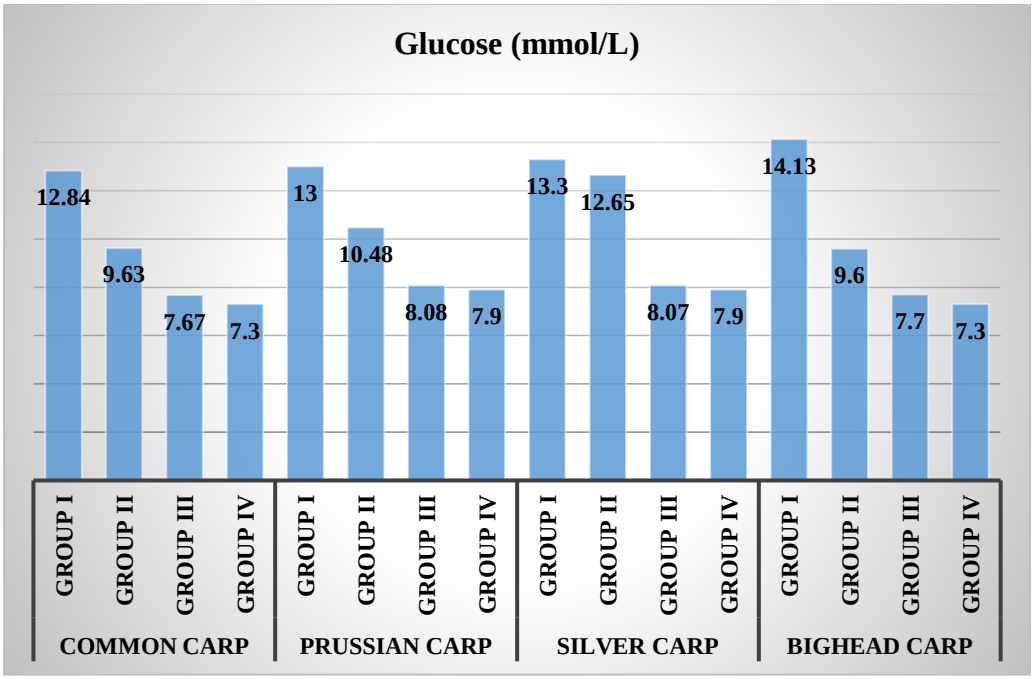

Fig. 4.2. Variation of glucose depending on the level of infestation in Asian carp

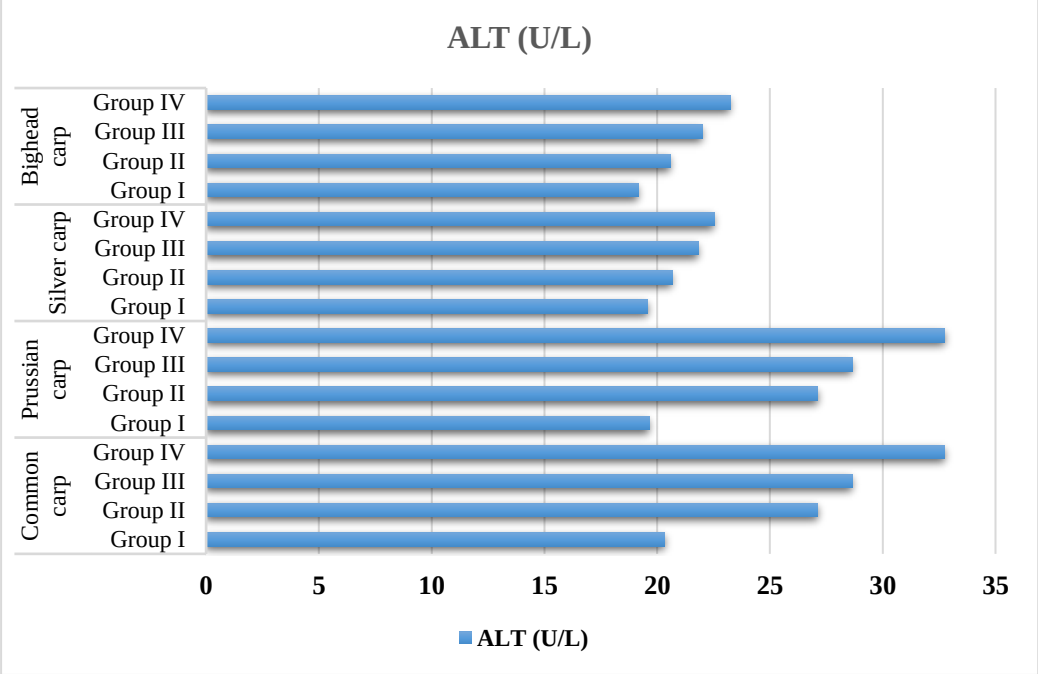

Fig. 4.3. Variation of ALT depending on the level of infestation in Asian carp

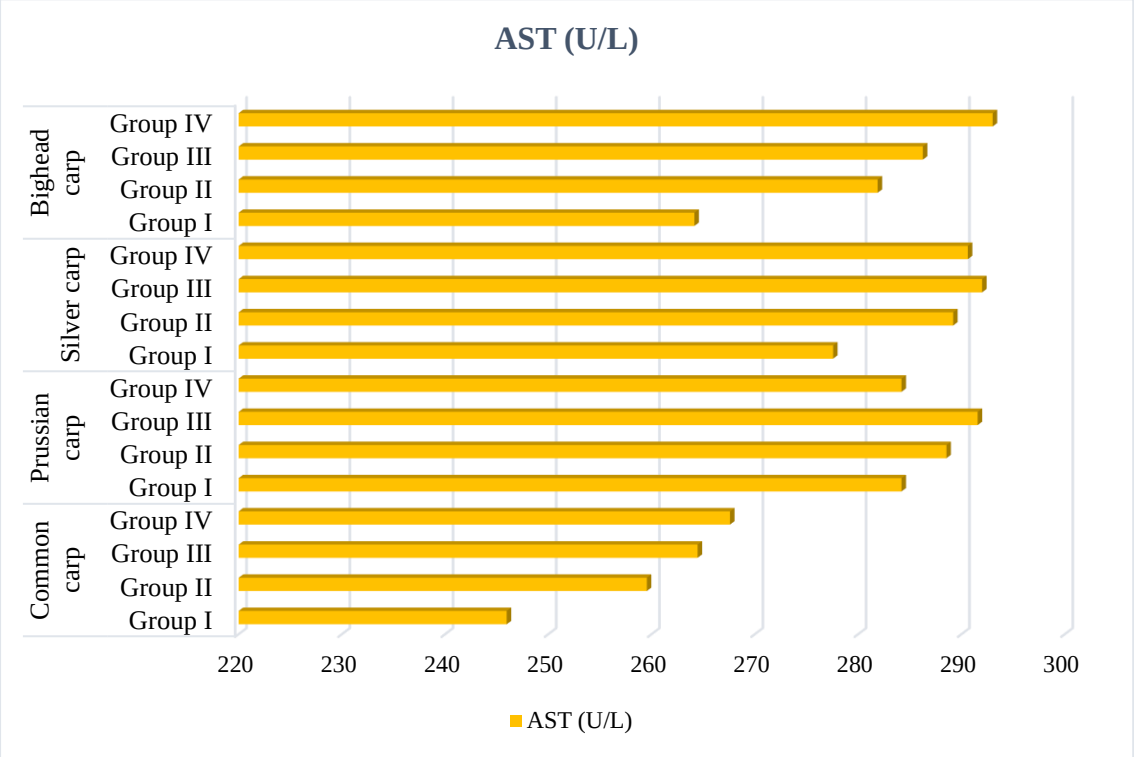

Fig. 4.4. Variation of AST depending on the level of infestation in Asian carp

As a result of meat quality examination, it was established that in infested specimens the protein and fat content decreases and humidity and pH increases, compared to non-infested specimens. Thus, the level of protein and fat in the muscle tissue decreased by 14,68% and 42,86% in Common carp, by 19,46% and 28,12% in Prussian carp, by 17,38% and 36,94% in Silver carp, by 10,61% and 56,8% in Bighead carp. Humidity and pH in the mono- and polyinfested specimens increased insignificantly (on average by 2,4% and 2,6%) (table 4.1.).

Table 4.1. The influence of mono- and polyinvasions on the variation of the chemical composition of muscle tissue in Asian carp

Group	Number of the examined fish	Proteins, (%)	Fat, (%)	Humidity, (%)	pH
Cyprinus carpio					
I	10	14,58±0,39	4,27±0,21	81,39±0,60	6,46±0,045
II	10	14,59±0,97	3,96±0,75	81,81±0,50	6,50±0,015
III	10	12,16±3,73	3,32±0,49	82,52±0,52	6,56±0,048
IV	10	12,44±0,59	2,44±0,17	82,95±0,57	6,62±0,060
Carassius gibelio					
I	10	16,65±0,38	2,17±0,17	77,68±0,26	6,47±0,043
II	10	15,99±0,43	2,06±0,12	77,87±0,21	6,50±0,016
III	10	14,56±1,16	1,82±0,17	79,25±0,75	6,56±0,045
IV	10	13,41±0,64	1,56±0,17	80,02±0,57	6,65±0,048
Hypophthalmichthys molitrix					
I	10	18,65±0,55	2,22±0,26	79,08±0,68	6,46±0,059
II	10	18,29±0,63	2,10±0,17	78,97±0,58	6,50±0,013

III	10	16,56±0,74	1,43±0,34	79,92±0,92	6,56±0,03
IV	10	15,41±0,92	1,40±0,33	80,97±0,56	6,68±0,051
Hypophthalmichthys nobilis					
I	10	18,58±0,466	1,62±0,22	79,28±0,67	6,48±0,045
II	10	18,00±0,464	1,17±0,24	80,09±0,54	6,50±0,010
III	10	17,66±0,783	0,93±0,23	80,65±0,50	6,63±0,041
IV	10	16,61±0,762	0,70±0,22	81,52±0,58	6,68±0,041
Reference: *-P<0,05; **-P<0,01; ***-P<0,001					

Conclusions at the chapter 4

1. As a result of the evaluation of the impact of mono - and polyinvasions on the hematological indices, it was determined that the level of hemoglobin, hematocrit, procalcitonin, as well as the number of red blood cells in the polyinfested groups decreases significantly, compared to the uninfested group. Thus, the level of hemoglobin decreased by 13,5% in Common carp, by 19,31% in Silver carp, by 22,61% in Bighead carp; the hematocrit level decreased by 17,98% in Common carp, by 22,66% in Prussian carp, by 17,41% in Silver carp, by 13,89% in Bighead carp; the level of procalcitonin decreased by 55,56% in Common carp, by 50,0% in Prussian carp, by 12,22% in Silver carp, by 28,0% in Bighead carp; the number of red blood cells by 26,82% in Common carp, by 41,45% in Prussian carp, by 15,02% in Silver carp, by 29,06% in Bighead carp. It was also established that the values of the hematological indices recorded in the monoinfested groups vary insignificantly compared to group I.
2. It was determined that the number of white blood cells increased by 21,64% in Common carp, compared to the uninfested group, by 17,75% in Prussian carp, by 18,44% in Silver carp, and by 22,16% in bighead carp. The number of white blood cells increases in polyinfested group, as a result of the response of the immune system to the aggressive action of the parasitic agents.
3. The biochemical examination of the blood of mono- and polyinfested specimens, reveals a decrease in total protein, triglycerides and glucose compared to uninfested groups. Among these indices, the lowest level was that of glucose, which decreased by 43%, and total protein, which decreased by approximately 25% compared to group I. Increased values of transaminases (ALT, AST) were recorded. The level of ALT and AST, unlike group I, was higher by 25,0% and 8,0%, respectively.
4. It was determined that the level of protein and fat in muscle tissue decreased by 14,68% and 42,86% in Common carp, by 19,46% and 28,12% in Prussian carp, by 17,38% and 36,94% in Silver carp, by 10,61% and 56,8% in Bighead carp. The value of humidity and pH in mono- and polyinfested groups increased insignificantly.

5. DEVELOPMENT OF INNOVATIVE PROCEDURE FOR PROPHYLAXIS AND TREATMENT OF HELMINTHIASIS IN COMMON CARP (*Cyprinus carpio* Linnaeus, 1758)

The prevention and control method is related to fish farming, in particular, to the protection of cultured carp and can be widely applied in practice for the dehelminthization of carp in various ponds, lakes etc. It is known that, for the purpose of dehelminthization of Common carp, the anti-parasitic preparation Râbolic is used, which, in one gram of anti-parasitic preparation, contains: Praziquantel-35 mg, Fenbendazole-70 mg and Levamisol-20 mg. According to the instructions a feed mixture with the addition of an antiparasitic preparation is made: 1 kg of the preparation is mixed well with 99 kg of feed (the recipe corresponding to each age group). The therapeutic (daily) dose of antiparasitic preparation is 1,5% of the estimated weight of the fish. The daily dose is divided into 5-6 regular portions, administered at certain feeding places, during the day, at an interval of 1-2 hours. Another fodder during the dehelminthization period is no longer used. The problem, which the proposed method solves, consists in the use of an effective, harmless, cheap and simple composition for dehelminthization and complementary feeding of common carp, which contains: sunflower press cake - 38.5%; corn meal – 30%; soybean press cake – 15%; alfalfa meal – 10%; meat and bone meal - 5%; antiparasitic preparation Rabolic -1.5%.

The result of this procedure consists in carrying out a complex and effective dehelminthization during April-May, as well as additional feeding with the use of nutritive briquettes rich in vitamins, trace elements, assimilable concentrated minerals, which allows the extensivity of parasitic invasions produced by cestodes *Khawia* and *Bothriocephalus* to decrease by 75-80%, thus stimulating the process and effectiveness of reproduction, increased weight gain, viability and resistance to environmental conditions and predators.

Also, the obtained result is due to the use of briquetted components, which have the role of supplementing the deficiency of vitamin-mineral substrate, with a complex anti-parasitic and immunostimulatory effect (Praziquantel, Fenbendazole - anti-parasitic preparations and Levamisol - anti-parasitic preparation with immunostimulatory effect).

Considering the fact that the daily additional food ration for common carp must be 3-5% of the body weight, a briquetted feed mass with the addition of anti-parasitic preparation was formed of 150 kg, respectively, 15,0 kg/ day within 10 days for 300 kg of live common carp mass.

The briquetted fodder (figure 5.1.) has the following composition: sunflower press cake – 55,5 kg (38,5%); corn meal – 45,0 kg (30%); soybean press cake – 22,5 kg (15%); alfalfa meal – 15,0 kg (10%); meat and bone meal – 7,5 kg (5%); antiparasitic preparation Rabolic – 4,5 kg (1,5%).

Fig. 5.1. Briquetted fodder

The Rabolic antiparasitic preparation is registered in the Nomenclature of Medical - Veterinary products from the Republic of Moldova and sold in the network of veterinary pharmacies in the country, and contains: Praziquantel – 35 mg; Fenbendazole – 70.0 mg and Levomisol – 20 mg. All the obtained mass of 150,0 kg is well mixed and then briquetted in a special device for the production of briquettes.

The result of complementary feeding and dehelminthization of Common carp, through the addition of complementary food containing anti-parasitic preparations, consists in compensating the deficit in a vulnerable period of the annual cycle - April-May (recovery period after "winter rest" and the pre-reproductive period), with vitamins, trace elements , assimilable concentrated minerals, as well as in their dehelminthization by means of broad-spectrum antiparasitic preparations, with immunostimulatory action. These actions allow strengthening the general condition of the body and preserving the high reproductive potential. Also, this procedure allows to use efficiently and economically both briquetted fodder and anti-parasitic preparations with an immunomodulatory effect.

For the experimental control of the proposed composition, 3 different variants were prepared with food content and briquetted antiparasitic preparation (table 5.1.).

Tab. 5.1. The experimental variants and the effectiveness of dehelminthization of Common carp

Nr. of variants	Nr. of the pond	The total content of the briquetted fodder mass (kg)	The content of the antiparasitic preparation in the briquetted fodder (kg)	Alive carp mass content (kg)
V 1	1	140,0	3,5	300
V2	2	150,0	4,5	300
V3	3	160,0	5,5	300

The population and the density of the fish in three ponds were determined, after which a detailed helminthological examination of the fishes was carried out for the presence of helminths. Several ponds were examined, but only three were included in the experiment, which contained the same mass of living

common carp and a similar level of infestation. Based on the biomass values, established in those three ponds, a briquetted complementary food mass is prepared with the addition of an anti-parasitic preparation with an immunostimulatory effect, which is distributed in the ponds, corresponding to the three variants (in pond no. 1 the briquetted fodder containing anti-parasitic preparation from version 1 was distributed, briquetted fodder from variant 2 was administered in pond no. 2, and respectively, briquetted fodder was used in pond no. 3, according to variant no. 3).

The daily amount of fodder with the addition of anti-parasitic preparation is divided into 5-6 regular portions, which is then administered in certain well-lit feeding places, during the day, at an interval of 1-2 hours. For 10 days, while the complementary feeding and dehelminthization lasts, no other food is used. The procedure was applied in three experimental fish farms, from various areas of the Republic of Moldova, pre-selected according to the mass of living common carp and the level of infestation, during April-May period, when there is a sudden warming of the weather and water in ponds (temperatures within the limits of 18-22℃) and at the same time there is a major need for complementary food, the natural trophic base not being sufficiently developed. Based on the recommendations of the leaflet of the Rabolic anti-parasitic preparation included in the briquettes, the dehelminthization of Common carp is carried out only once a year.

After carrying out the complementary feeding and dehelminthization (which lasts for 10 days), in 2-3 weeks the biological samples are taken and analyzed to establish the effectiveness of the dehelminthization. For complementary feeding, dehelminthization and prophylaxis of invasions caused by the cestodes of the genus *Khawia* and *Bothriocephalus*, we recommend for use the fodder with the antiparasitic content of variant 2 whose composition has demonstrated more optimal results (figure 5.1.).

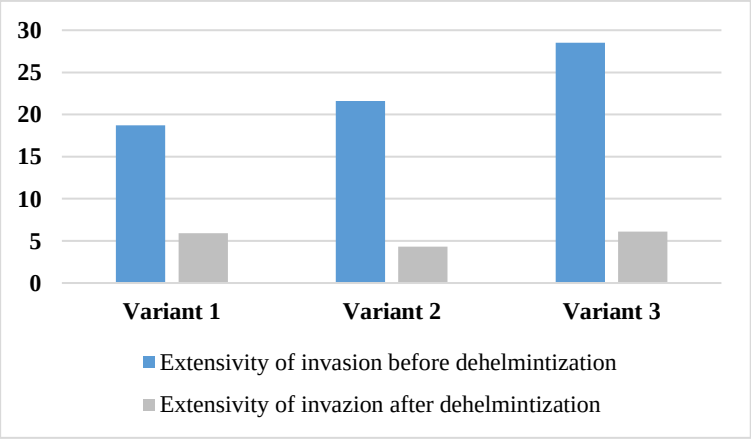

Fig. 5.1. Extensivity of invasion before and after dehelminthization

Based on the research results with the purpose of developing innovative procedures to reduce and control the helminthiasis in Common carp (*Cyprinus carpio* Linnaeus, 1758), was obtained: an invention patent (Patent MD 1590 Y 2022.01.31:"Composition and procedure for dehelminthization and additional feeding of Common carp") and 4 implementing acts.

Conclusions at the chapter 5

1. The optimal temperature of water in ponds for the application of the procedure of prophylaxis and control of endohelminths is 18-22℃ and summarizes the major need for complementary food in the conditions of an insufficiently developed trophic base.
2. As a result of the experimental control of the ratio of food content and the active substance in the briquettes, we determined that the most effective was variant no. 2, which contains 150 kg of concentrated fodder and 4,5 kg of antiparasitic preparation.
3. It was established that the Rabolic preparation not only has antiparasitic action against intestinal cestodes, trematodes and nematodes, as indicated in the manufacturer's instruction, but being administered in briquetted form, it manifests a relevant action against the ectoparasites from the class Monogenea (*Dactylogyrus extensus, D. vastator, Diplozoon paradoxum*), which were identified in Common carp (high extensivity of invasion).
4. The results of the hematological and biochemical research carried out on Common carp allowed us to establish that, during April-May, in the body of the common carp, the greatest insufficiency of vitamins, micro- and macroelements is registered, being also considered the most vulnerable period in their annual development cycle, a period when is strictly necessary to intervene both with dehelminthization and complementary feeding.
5. The carried out research allowed us to establish that the developed and patented procedure "Composition and procedure for dehelminthization and additional feeding of Common carp (*Cyprinus carpio* Linnaeus, 1758)" allows reducing the extensivity of invasion with endoparasites in Common carp by 75-80% and, at the same time, ensures the supply of the organism with vitamins, trace elements, assimilable minerals, thus stimulating the process and effectiveness of reproduction, increased weight gain, viability and resistance to environmental conditions and predators.
6. The undertaken procedure for prophylaxis and control allowed us to use efficiently and economically both briquetted fodder and antiparasitic preparations with an immunomodulatory effect.
7. The obtained results both due to the components of the fodder and the briquetted form allowed keeping all the ingredients together for a long time and their maximum consumption.

GENERAL CONCLUSIONS

1. It was established that the helminth fauna of alien and native fish species in the analyzed water basins is represented by 31 species of helminths systematically classified into different classes: class Monogenea – 4 species: *Dactylogyrus* sp., *Gyrodactylus* sp., *Diplozoon paradoxum*, *Eudiplozoon nipponicum*; class Trematoda – 15 species: *Asymphylodora imitans, Bunodera lucioperca, Nicolla skrjabini, Aspidogaster limacoides, Phyllodistomum angulatum,*

Pronoprymna ventricosa, *Lecithaster confusus*, *Phyllodistomum folium*, *Diplostomum spathaceum*, *Posthodiplostomum cuticola*, *Tylodelphys clavata*, *Apophallus* sp., *Clinostomum complanatum*, *Ichthyocotilurus* sp., *Paracoenogonimus ovatus*; class Cestoda – 5 species: *Ligula intestinalis*, *Khawia sinensis*, *Bothriocephalus opsariichthydis*, *Paradilepis scolecina*, *Valipora campylancristrota*; class Enoplea – 3 species: *Schulmanela petruschewskii*, *Pseudocapillaria tomentosa*, *Eustrongylides* sp.; class Chromadorea – 2 species: *Raphidascaris acus*, *Hysterothylacium aduncum*; class Palaecanthocephala – 2 species: *Acanthocephalus lucii*, *Pomphorynchus laevis*.

2. Polyinvasions were estimated to be prevalent in the following fish species: Prussian carp from the Dniester river (monoinvasions – 47,07%, polyinvasions – 52,93%), Prussian carp from the Prut river (monoinvasions – 25,0%, polyinvasions – 75,0%), Stone moroko from the Bâc river (monoinvasions – 10,0%, polyinvasions – 90,0%), Pumpkinseed from the Village Museum lake (monoinvasions – 5,13%, polyinvasions – 94,87%), Bighead carp from the fishery S.A Pescăruș (monoinvasions – 27,17%, polyinvasions – 72,73%), European bitterling from the Răut river (monoinvasions – 13,0%, polyinvasions – 79,0%), European perch from the Dniester river (Dubăsari reservoir) (monoinvasions – 10,52%, polyinvasions – 89,47%), European perch from the Racovăț river (monoinvasions – 58,33%, polyinvasions – 41,66%), European perch from the Draghiște river (monoinvasions – 33,35%, polyinvasions – 66,67%), Monkey goby from the Dniester river (Dubăsari reservoir) (monoinvasions – 87,55%, polyinvasions – 12,5%), Bighead goby from the Dniester river (Dubăsari reservoir) (polyinvasions – 100%), Round goby from the Dniester river (Dubăsari reservoir) (Dubăsari reservoir) (monoinvasions – 33,3%, polyinvasions – 66,67%), Western tubenose goby from the Dniester river (Dubăsari reservoir) (monoinvasions – 16,67%, polyinvasions – 83,3%).
2. Of the detected helminth species, common to human and carnivorous mammals were: *Clinostomum complanatum* and *Apophallus* sp. found in European perch and European bitterling specimens; *Paracoenogonimus ovatus* și *Eustrongylides* sp. found in Monkey goby, Bighead goby, Round goby and Western tubenose goby specimens.
3. Evaluating the impact of mono - and polyinvasions on the hematological indices, it was determined that the level of hemoglobin, hematocrit, procalcitonin, as well as the number of red blood cells in the polyinfested groups decreases significantly, compared to the uninfested group. Thus, the level of hemoglobin decreased by 13,5% in Common carp, by 19,31% in Silver carp, by 22,61% in Bighead carp; the hematocrit level decreased by 17,98% in Common carp, by 22,66% in Prussian carp, by 17,41% in Silver carp, by 13,89% in Bighead carp; the level of procalcitonin decreased by 55,56% in Common carp, by 50,0% in Prussian carp, by 12,22% in Silver carp, by 28,0% in Bighead carp; the number of red blood cells by 26,82% in Common carp, by 41,45% in Prussian carp, by 15,02% in Silver carp, by 29,06% in Bighead carp. It was also established that

the values of the hematological indices recorded in the monoinfested groups vary insignificantly compared to group I.

4. The biochemical examination of the blood of mono- and polyinfested specimens, reveals a decrease in total protein, triglycerides and glucose compared to uninfested groups. Among these indices, the lowest level was that of glucose, which decreased by 43%, and total protein, which decreased by approximately 25% compared to group I. Increased values of transaminases (ALT, AST) were recorded. The level of ALT and AST, unlike group I, was higher by 25,0% and 8,0%, respectively.
5. It was determined that the level of protein and fat in muscle tissue decreased by 14,68% and 42,86% in Common carp, by 19,46% and 28,12% in Prussian carp, by 17,38% and 36,94% in Silver carp, by 10,61% and 56,8% in Bighead carp. The value of humidity and pH in mono- and polyinfested groups increased insignificantly.
6. The optimal temperature of water in ponds for the application of the procedure of prophylaxis and control of endohelminths is 18-22℃ and summarizes the major need for complementary food in the conditions of an insufficiently developed trophic base.
7. As a result of the experimental control of the food ratio content and the active substance in the briquettes, we determined that the most effective was variant no. 2, which contains 150 kg of concentrated fodder and 4,5 kg of antiparasitic preparation.
8. The Rabolic preparation, in the form of briquetted fodder mass, led to a decrease in the extensivity of invasion with endoparasites in Common carp by 75-80% and, at the same time, ensured the supply of the organism with vitamins, trace elements, assimilable minerals, thus stimulating the process and effectiveness or reproduction, increased weight gain, viability and resistance to environmental conditions and predators.

PRACTICAL RECOMMENDATIONS

1. The data obtained regarding the helminth fauna of alien and native fishes can be used for the development of measures to prevent and control parasitic diseases of fish in the aquatic biotopes of the Republic of Moldova.
2. It is recommended to inform the population about parasitic diseases and the potential danger posed by infested fish, to protect the health of humans and carnivorous animals.
3. The scientific results obtained regarding the helminth fauna of alien fishes, the influence of mono- and polyinvasions on some hematological, biochemical, and productive indices, the measures of prophylaxis and control of helminthiasis in fish, can be implemented in fish farming practice, and used in the training process of students and master's students from various educational institutions.

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2. GOLOGAN, I. *The helminth fauna of some invasive fishes from various natural and artificial water bodies from the Republic of Moldova*. Scientific Papers Journal VETERINARY SERIES, vol.63, nr.2. 2020. pp.136-141. ISSN 2393 – 4603. (Cat. B+ CNCSIS)

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Patents

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ADNOTARE

Gologan Ion „Helmintofauna speciilor alogene de peşti în condiţiile Republicii Moldova”, teză de doctor în științe biologice, Chișinău, 2022.

Structura tezei: introducere, cinci capitole, concluzii generale și recomandări, bibliografia din 198 de titluri, 19 anexe, 119 de pagini de bază, 42 figuri, 19 tabele. Rezultatele obținute sunt publicate în 11 lucrări ştiinţifice la tema tezei.

Cuvinte-cheie: helmintofaună, specii de pești alogeni, impact parazitar, ecosisteme acvatice, indici productivi, procedee inovative, profilaxie, combatere.

Scopul. Stabilirea helmintofaunei și a particularităților impactului acesteia asupra organismului-gazdă, în scopul elaborării procedeelor inovative de profilaxie și combatere a helmintozelor la unele specii de pești din grupul ciprinidelor asiatice.

Obiective. Evidențierea biodiversității helmintofaunei speciilor alogene de peşti din diverse ecosisteme acvatice ale Republicii Moldova; evaluarea impactului mono- şi poliinvaziilor asupra unor indici ai statusului morfo-funcţional şi biochimic la unele specii de pești din grupul ciprinidelor asiatice; evidențierea impactului mono- şi poliinvaziilor asupra unor indici productivi la unele specii de pești din grupul ciprinidelor asiatice; elaborarea procedeelor inovative de profilaxie şi combatere a helmintozelor la unele specii de pești din grupul ciprinidelor asiatice.

Noutatea şi originalitatea ştiinţifică. Pentru prima dată în Republica Moldova, a fost realizat un studiu științifico-practic complex ce constă în: determinarea helmintofaunei la speciile de pești alogene și autohtone din diverse ecosisteme acvatice naturale și antropizate cu stabilirea speciilor de helminți specifici și comuni atât pentru diverse specii de animale carnivore, cât și pentru om; în premieră a fost evaluat impactul mono- şi poliinvaziilor asupra unor indici morfo-funcţionali, biochimici, productivi ai organismului-gazdă la unele specii de pești din grupul ciprinidelor asiatice, finalizat cu elaborarea, brevetarea și implementarea procedeelor innovative, de profilaxie și combatere a parazitozelor acestora.

Rezultatele principale. S-a stabilit rolul speciilor alogene de peşti din diverse ecosisteme acvatice ale Republicii Moldova în formarea, menţinerea și vehicularea focarelor de agenţi parazitari comuni atât pentru diverse specii de animale carnivore, cât și pentru om. Pentru prima dată au fost elucidate modificările indicilor morfo-funcţionali la infestarea mono-și poliparazitară și elaborate procedee noi de tratament antiparazitar complex în dependență de specificul de infestare la unele specii de pești din grupul ciprinidelor asiatice.

Semnificația teoretică. Pentru prima data a fost abordată și soluționată o problemă majoră în domeniul pisciculturii din Republica Moldova, fapt care a permis de a evidenția rezultatul acțiunii asupra organismului-gazdă a mono-, poliinvaziilor, a determina impactul acestora asupra calității produselor și elaborarea de noi procedee de profilaxie și combatere a parazitozelor la crap (*Cyprinus carpio* Linnaeus, 1758).

Valoarea aplicativă. În baza rezultatelor obținute a fost elaborat un Ghid metodologic, brevetate și implementate măsuri inovative de profilaxie și combatere a parazitozelor la unele specii de pești din grupul ciprinidelor asiatice, elaborate procedee inovative de diminuare și redresare a prejudiciilor economice în sectorul piscicol și zooveterinar, care sunt esenţiale atât pentru înmulţirea, creşterea şi dezvoltarea speciilor productive de pești într-un ecosistem acvatic sănătos, cât și pentru întreruperea lanțului epizootic de vehiculare a focarelor de agenţi parazitari comuni; acestea se referă la diverse specii de păsări și mamifere, cât și pentru om.

Implementarea rezultatelor științifice. Rezultatele studiului realizat au fost implementate în practica piscicolă și folosite în procesul de instruire a studenţilor, rezidenţilor și masteranzilor din diverse instituții de învățământ.

ANNOTATION

Gologan Ion "The Helminth fauna of alien fishes in the Republic of Moldova", PhD thesis in biological sciences, Chisinau, 2022.

Thesis structure: introduction, five chapters, general conclusions and recommendations, bibliography of 198 titles, 19 annexes, 119 basic pages, 42 figures, 19 tables. The obtained results are published in 10 scientific papers.

Keywords: helminth fauna, alien species, parasitic impact, aquatic ecosystems, productive indices, innovative procedures, prophylaxis, control.

Aim. To establish the helminth fauna and the particularities of its impact on the host organism, to develop innovative procedures for prophylaxis and control of helminthiasis in some Asian carp species.

Objectives. Establishing the biodiversity of the helminth fauna of alien fishes from various aquatic ecosystems of the Republic of Moldova; evaluation of the impact of mono- and polyinvasions on some indices of the morpho-functional and biochemical status in some Asian carp species; highlighting the impact of mono- and polyinvasions on productive indices in some Asian carp species; elaboration of innovative procedures for prophylaxis and control of helminths in some Asian carp species.

Scientific novelty and originality. For the first time in the Republic of Moldova, a complex scientific-practical study was conducted consisting in: determining the helminth fauna in alien and native fish species from various natural and anthropogenic aquatic ecosystems with the establishment of specific and common helminths for various species of birds and mammals, as well as for humans; evaluation of the impact of mono- and polyinvasions on morpho-functional, biochemical, productive indices of the host organism in some Asian carp species, finalized with the development, patenting and implementation of innovative procedures, prophylaxis and control of parasites.

Main results. The role of alien fishes from various aquatic ecosystems of the Republic of Moldova was established in the formation, maintenance, and circulation of outbreaks of common parasitic agents for various species of birds and mammals, as well as for humans. For the first time, the changes in the morpho-functional indices for mono- and polyparasitic infestation were elucidated and new procedures for complex antiparasitic treatment were developed depending on the specificity of the infestation in some Asian carp species.

Theoretical significance. For the first time a major problem in the field of fish farming in the Republic of Moldova was addressed and solved, which allowed to highlight the results of the action on the host organism of mono-, polyinvasions, determine their impact on product quality and development procedures for prophylaxis and control of parasitosis in some Asian carp species.

Applicative value. Based on the obtained results, a Methodological Guidebook was developed, innovative measures for prophylaxis and control of parasitosis in some Asian carp species were implemented and patented, innovative procedures to reduce and recover economic damage in the fisheries and livestock sector were developed, essential both for the reproduction, growth and development of productive fishes in a healthy aquatic ecosystem, and for the interruption of the epizootic chain of outbreak of common parasitic agents, both for various species of birds and mammals, and for humans.

Implementation of scientific results. The results of the study were implemented in fishing practice and used in the training of students, residents, and masters in various educational institutions.

АННОТАЦИЯ

Гологан Ион "Гельминтофауна аллогенных видов рыб в условиях Республики Молдова", кандидатская диссертация по биологическим наукам, Кишинев, 2022.

Структура диссертации: введение, пять глав, общие выводы и рекомендации, библиография из 198 наименований, 19 приложений, 119 основных страниц, 42 рисунок, 19 таблиц. Полученные результаты опубликованы в 10 научных статьях по теме диссертаций.

Ключевые слова: гельминтофауна, аллогенные виды, паразитарное воздействие, водные экосистемы, продуктивные показатели, новаторские меры, профилактика, контроль.

Цель. Йзучение гельминтофауны и особенностей ее воздействия на организм хозяина с целью разработки новаторских методов профилактики и борьбы с гельминтозами у некоторых видов азиатских карповых.

Задачи. Установление биоразнообразия гельминтофауны аллогенных видов рыб из различных водных экосистем Республики Молдова; оценка влияния моно- и полиинвазий на некоторые показатели морфо-функционального и биохимического статуса у некоторых видов рыб из группы азиатских карповых; выявление влияния моно- и полиинвазий на продуктивные показатели некоторых видов рыб группы азиатских карповых; разработка новаторских методов профилактики и борьбы с гельминтозами у некоторых видов рыб из группы азиатских карповых.

Научная новизна и оригинальность. Впервые в Республике Молдова проведено комплексное научно-практическое исследование, заключающееся в следующем: определении гельминтофауны у аллогенных и аборигенных видов рыб из различных природных и антропогенных водных экосистем с установлением специфических и распространенных гельминтозов для различных видов птиц и млекопитающих, а также для человека; оценка влияния моно- и полиинвазий на морфофункциональные, биохимические, продуктивные показатели организма хозяина у некоторых видов рыб из группы азиатских карповых, завершенная разработкой, патентованием и внедрением новаторских методик профилактики и борьбы с паразитами в них.

Основные результаты. Установлена роль аллогенных видов рыб из различных водных экосистем Республики Молдова в формировании, поддержании и циркуляции вспышек общих паразитарных агентов для различных видов птиц и млекопитающих, а также для человека. Впервые выявлены изменения морфофункциональных показателей при моно- и полипаразитарной инвазии и разработаны новые методы комплексного противопаразитарного лечения в зависимости от специфики инвазии у некоторых видов рыб из группы азиатских карповых.

Теоретическая значимость. Впервые рассмотрена и решена крупная проблема в области рыбоводства в Республике Молдова, что позволило выделить результат действия моно- и полиинвазий на организм хозяина, определить их влияние на качество продукций и разработка методов профилактики и борьбы с паразитозами у некоторых видов рыб группы азиатских карповых.

Прикладное значение. На основании полученных результатов разработано, внедрено и запатентовано Методическое пособие по новаторских мероприятиям по профилактике и борьбе с паразитозами у некоторых видов рыб группы азиатских карповых, разработаны новаторские методы по снижению и возмещению экономического ущерба в рыбном хозяйстве и в секторе животноводства, необходимый для размножения, роста и развития продуктивных видов рыб в здоровых водных экосистемах.

Внедрение научных результатов. Результаты исследования внедрены в рыболовную практику и использованы при обучении студентов, ординаторов и магистров различных учебных заведений.

GOLOGAN ION

THE HELMINTH FAUNA OF ALIEN FISH SPECIES IN THE REPUBLIC OF MOLDOVA

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