UNIVERSITY OF AGRICOL SCIENCES AND VETERINARY MEDICINE
„ION IONESCU DE LA BRAD" FROM IASI
FACULTY OF VETERINARY MEDICINE
DOMAIN: VETERINARY MEDICINE
SPECIALIZATION: NORMAL AND PATHOLOGICAL MORPHOLOGY

Veterinary Mircea LAZĂR

DOCTORAL THESIS

Scientific Leader,
University Professor dr. Octavian Zaharie OPREAN

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MORPHOLOGICAL BASIS OF SWEET WATER FISH DISEASES FROM FISH FARMS

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SUMMARY

The nowadays knowledge analysis within this domain points out that nursery pond cyprinids and salmonids pathology, especially the morph-physiology basis of diseases, was not entirely been studied, mainly within the sphere of systematic arrangements of intensive breeding, in which large fish populations are used as patterns.

The injuries (on morphological basis) caused by different diseases represent point 0 in their pathogenic chain. The ultra structure (electron-microscopic), histological and macroscopic investigations lead to a logical interpretation of disease state, of its reversibility level.

Researches have been performed on approximately 1500 fish from the *Cyprinidae* family: common carp (*Cyprinus carpio)*, silver carp (*Hypophthalmichthys molitrix),* bighead carp *(Aristichtys nobilis)* grass carp (*Ctenopharyngodon idella),* golden carp (*Carassius auratus gibelio)* and from *Salmonidae* family represented by the rainbow trout *(Oncorhynchus mykiss)*, adults and parrs raised in intensive systems (lakes, ponds, vessels, barrier lakes). The specimens were taken from three main farms: two cyprinids farms from Iasi district and one salmonids farm from Neamț district, but specimens were also taken from other nearby districts farms in North - West Romania and salmonids farms from Toscana, Italy.

The fishy material was subdued to a complete ichthyopathology examination, referring in fact to a clinical exam, a necropsy exam, a parasite exam, a bacteriological exam, histopathological exam, immunohistochemical exam and electronic microscopy scanning. (SEM – *scanning electron microscopy*) exam.

The research methods used have been represented by the removal of scrapers on skin level, of flippers and gills, or of scrapers type „squash" (by crushing between blade and lamella) from gills or from pathological formations (knots, cysts etc.); ingathering of tissue and organ fragments to obtain permanently colored contents, through ordinary methods: HEA, HE, PAS,

Gram, Giemsa, Groccot; to identify the bacteriological level, I have used the insemination on usual or special averages, but also quick tests (API 20E tests); also, I have used the immunohistochemical analysis to confirm the disease diagnosis (yersiniosis).

Mucus secretive cells can be found in the epidermis of all teleosteens, but their numbers vary according to their place or species. These cells usually find their origin in the middle layer of epidermis, but within a thin epidermis, and the basis of mucous cells can be observed in the basal membrane. They grow as dimension and release secretions as they get closer to surface.

Starting from this point, I have studied the external structure of skin through electronic microscopy scanning (SEM) on a few species of mirror carp and bighead carp which presented cutaneous ulcer on their body surface, and by microbiological exam, was identified as *Aeromonas spp.*

The results of the research have pointed out the presence of many general pathological processes of organs and systems, caused by viruses, bacteria and parasites.

So, the pathological processes caused by viruses have been noticed on *infectious hematopoietic necrosis*, caused by a *Rhabdovirus,* through the appearance of hemorrhagic focuses, of necrosis, of lymph-histological proliferations and of fiber exudations at peritoneum level, on trout coming from salmonids farms in Toscana, Italy- disease that was firstly confirmed by Zoo Technical Institute of Udine.

The bacteria that have evolved, bacteria taken from the study farms herein mentioned, were represented by: *carp* and *asian cyprinids dermatitis, hemorrhagic bacteria sepsis, yersiniosis*, caused by *Aeromonas* bacteria and *Yersinia ruckeri* bacteria. On examined fish, there have been pointed out, through direct bacterioscopic examinations of cutaneous injuries, bacilli and co bacilli Gram (-). The bacteriological examination was performed by collecting heart blood and by collecting it from the previous kidney which, by insemination, relieved pure colonies of *Aeromonas spp*.

Some histological injures on skin level have been detected, translated by epidermis necrosis, hyperplasia of mucus secretive cells, dystrophy, and within the internal organs (peritoneum, liver, intestine, spleen, kidney) blood circulation disorders, dystrophic and inflammatory processes have been observed.

The parasite diagnosed invasions have been registered in a higher number than the diseases caused by viral and bacteriological infections.

The parasite diagnosed diseases were: *ichtiobodosis, myxosporidiosis*, *proliferative kidney disease, microsporidiosis, trichodinosis, dactilogirosis, trematodosis, black spot disease, argulosis, sinergasilosis, and lerneosis*. According to the organic affinity of every parasite, the

histopathological injuries have been blood circulation disorders (congestions, bleedings, edema), dystrophic (hydra dystrophy) and inflammatory processes.

Within the research period I've diagnosed a mycosis- *dermocystosis*, which appeared on the golden carp, as some protuberant formations, white and well determined, which, on histopathological skin examination, were discovered to be *Dermocystidium erschovei* cysts, with germs presenting PAS positive cytoplasm and globular nucleus.

Also, hyperplasia of mucus secretive cells have been stated, due to leucocytes infiltrations around cysts with intact walls.

According to the parasitological exams, it was recorded that from 12 species of identified parasites of the examined fish, 10 parasites species (83,3%) were detected on Farm A, then Farm B registered 7 parasites species (58,3%) and Farm C registered 2 parasites species (16,6%).

Bacteria micro flora was also more present in Farm A, where only 3 infectious diseases evolved (70%), as against Farm B where only 2 (20%) were registered and Farm C where only one case (10%) was detected.

The results obtained within 2005 - 2009 on the examined fish convinced us to go further, to the epidemiological analysis of fish species and farms.

The infectious diseases have been tracked down less than parasites invasions and, on the other hand, in our research bacteria prevail. *(Table 1)*.

Table1 1

Infectious diseases diagnosed within 2005 – 2009 in the fish farms studied

No. Crt.	Disease	Affected specie	Farm A	Farm B	Farm C	Other farms
1.	Erytrodermatitis of carp	Crap	53	46	0	17
2.	Erytrodermatitis of silver carp and bighead carp	Silver Carp Bighead carp	37	42	0	0
3.	Hemorrhagic Sepsis	Golden carp	15	0	0	8
4.	Yersiniosis	Rainbow trout	0	0	67	0
5.	Infectious hematopoietic necrosis	Rainbow trout	0	0	0	13
TOTAL			105	88	67	38

Comparing the table data, one can notice a sort of equability of research with reference to those two cyprinid farms, in which carp erytrodermatitis (and of asian cyprinids) prevails as an infectious disease.

Also, in Farm A, it is stated the appearance of hemorrhagic sepsis which had an unspecific occurrence for golden carp species.

Within the salmonids population, a special bacteria was detected which affected all age trout in Farm C, and in one of the farms in Toscana area-Italy the infectious hematopoietic necrosis of the rainbow trout was diagnosed at The Zoo Technical Institute of Udine.

The majority of parasite invasions identified within the fish farms have evolved clinically. When the intensity of parasite invasion has exceeded a certain line (invasion line), the clinical evolution of affections has came in sight.

Researches detected 11 parasite diseases within the fish farms studied, that can be found in *table 2:*

Table 2

Parasite diseases diagnosed within 2005-2009 in the fish farms studied

No.	Disease	Etiological Agent	Affected species	Area	Farm
1.	Ichtiobodosis	*Ichthyobodo necator*	Silver carp2+ Rainbow carp	Skin, gills	Farm A Farm C Other farms
2.	Myxosporidiosis	*Myxobolus spp.*	Carp 1+, 2+	Gas bladder, intestine	Farm A Farm B Other farms
3.	Trichodinosis	*Trichodina spp.*	Carp 2+	Gills	Farm A Farm B Other farms
4.	Dactilogirosis	*Dactylogyrus spp.*	Carp 0+, 1+	Gills	Farm A Farm B Other farms
5.	Black spots disease	*Posthodiplostomum cuticola*	Silver carp 1+, 2+	Skin	Farm A Farm B Other farms
6.	Argulosis	*Argulus foliaceus*	Carp 1+, 2+	Tail	Farm A
7.	Sinergasilosis	*Sinergasilus lieni*	Silver carp 1+ Bighead carp 4+ Grass carp3+	Gills	Farm A Farm B Other farms
8.	Lerneosis	*Lernaea spp.*	Carp, Bighead carp, Silver carp, Grass carp , Golden carp	Skin	Farm A Farm B Other farms
9.	Dermocystosis	*Dermocystidium erschovi*	Golden carp	Skin	Farm A

10.	Proliferative Kidney Disease	*Tetracapsuloides bryosalmonae*	Rainbow trout	Kidneys	Other farms
11.	Microsporidiosis	*Microsporidia incertae*	Rainbow trout	Gills	Farm C Other farms

The pathology of sweet water fish (cyprinids and salmonids) of systematic arrangements is conditioned by water quality, conclusion that was confirmed by the predominance of external parasite invasions in the pathological diagnosed entities.