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Ichthyopathology

# HISTOPATHOLOGICAL CHANGES IN LIVER AND KIDNEY OF THE EEL, ANGUILLA ANGUILLA L., AFFECTED BY PAPILLOMATOSIS ANGUILLARUM

# ZMIANY HISTOPATOLOGICZNE W WĄTROBIE I NERKACH WĘGORZY ANGUILLA ANGUILLA L. PRZY PAPILLOMATOSIS ANGUILLARUM

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Papillomatosis anguillarum causes inflammation, degeneration and necrosis changes in liver and kidney of the diseased eels. The character and profound histopathological changes observed depending upon type and quantity of tumours occurring on the skin of the eel.

#### Introduction

Histopathological structure of the tumour affected by papillomotosis anguillarum was subjected to intensive researches by many authors, as Schäperclaus (1953), Lühmann and Mann (1957), Amlacher (1961) and others. The differences in the structure from the histomorphology of different papillae were investigated by Pilarczyk (1973). He drew the attention to the contributing factors of the skin connective tissues, properly in the course of the formation and increase of papilla. Like Peters et al., (1970), the author also observed the degeneration of papillae building the tumour. Authors Schäperclaus (1953), Koops et al (1969) and Peters et al., (1970) are concerned with the disease etiology, and according to the presumption of Schäperclaus (1953), may be a virus effect of the group halophile. This theory was confirmed by the work of Pfitzner (1969) and Schubert (1969). After the electronic microscope identification, the

authors have been ascertained the presence of a typical virus form in the cells of papillae as well as in the growing cells of the infected blood in diseased eels. Trials for injecting healthy eels by tissue homogenate or transplanting the papillae, do not always give positive results. The diseased eels with papillomatiosis show considerable worse conditions, like the reduction of flesh weight to 40% (Pilarczyk, 1973) weight of internal organs as well as the reduction of the quantity of fat and albuminous substances in liver (Lühmann et al., 1957). Sectional examinations carried out on diseased eels with tumour, showed considerable changes in colour and consistency of internal organs especially in liver and kidney.

## MATERIAL AND METHOD

This study was carried out on eels caught from Szczecin Firth (Zalew Szczeciński) near Trzebież during the period from September to October 1971. The accomplished detailed macroscopic analysis of skins and papillae (differentiating its type such as flat, flaccid as well as stalked and superfacial rugged), and internal organs were taken from 77 fish ranging in length between 29 and 69 cm (l.t.), and weight from 30 to 320 g. The results of morphometric studies of the fish are the same as that in the work of **Pilarczyk** (1973, Table 1.).

For studying histopathological changes in liver and kidney, cross-sections from the internal organs of 25 diseased fish were investigated. The materials were fixed in 10% neutral formalin, mercuric chloride with acetic acid, viz. Susa, Zenker and Bouin fixatives. The histopathological sections were stained in aqueous haematoxylin and eosin according to the method of Dominici Passini and Uny. For the purpose of identification of the coloured deposites occurred in liver and kidney, the sections were additionally stained by ferric salt solution according to Thoms and Lavollay, as well as for detecting the bile pigments, Stein method.

### **RESULTS**

The histopathological examinations of the internal organs of diseased eels, explain the macroscopic changes of liver and kidney.

In most of the observed cases, it was found in liver of eels with flat and flaccid tumour, liver cells which are characterised by dystrophy and fading of cells walls. In some cases, this process is extensive and comprise large parenchyma spaces.

Very often, the regressive changes depend upon the parenchymatous degeneration or the degenerated vacuoles, which at the same time take place with the degeneration changes in nucleus of the cells (Fig. 1 and 2). Most of the nucleus is swollen, with weakly visible and fading nucleolus. This process is happened through the chromatin fragmentation and its condensation at the marginal part of the nucleus, rarely throughout

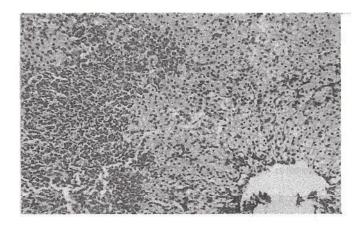


Fig. 1. Necrosis changes in liver of eel with flat and flaccid tumours

Photographed by: mgr M. Strzelichowski

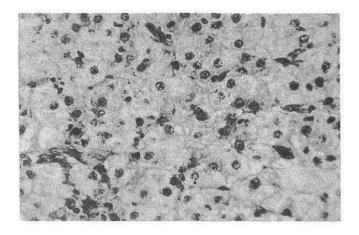


Fig. 2. Focus of colliqative necrosis in liver of eel with flat and flaccid tumours

Photographed by: mgr M. Strzelichowski

chromatin decomposition (karyorrhexis), leading to the necrosis of liver cells. The exacerabation changes causing the origination of different quantity of foci of colliqative necrosis. There are visible cells, only have structureless mass in different degrees of decomposition with few decomposed nuclei, (Fig. 2). In the cytoplasm of liver cells ocurring in the aspect of fine granules, bile pigments (Fig. 3). The few retained bile canals are found to be diminished, with strongly flattened epithelial cells and narrow empty

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lumen. Mostly, close to the injured bile canals, occurring liver cells saturated with bile pigments (Fig. 3). These cells undergo considerable regressive changes leading to necrosis through the decomposition of the cytoplasm and pycnosis of the nucleus (Fig. 3). The majority of blood capillaries are dilated and filled with erythrocytes and swollen endothelial cells. Thereafter, following extravasion of parenchymatous blood, while in the arterial vessels occurring swelling and fragmentation of the external vascular zone. There are also fine-celled infiltrate at the perivascular. Oftenly, the vessels lumen are filled with coagulated plasma in which the resulting blood elements concentrating themselves near to the vessels walls. Considerable regressive changes were observed in the deeper parts of livers from eels having large and stalked papillae, and superfacial rugged. In most of the

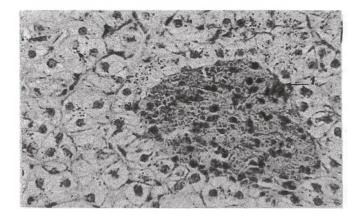


Fig. 3. Intracellular position of bile pigments in liver of eel with flat and flaccid tumours. i. necrosis focus in parenchyma of liver saturated with bile pigments

Photographed by: mgr M. Strzelichowski

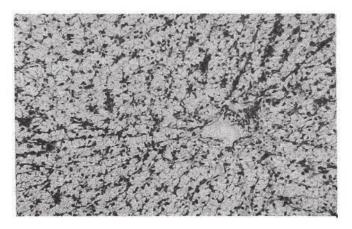


Fig. 4. Cirrhosis liver of eel with large stalked tumour Photographed by: mgr M. Strzelichowski

parenchymatous cells, the cytoplasm decomposition is ascertained (plasmolysis), (Fig. 4). The majority of liver cells are devoid of nucleus, (Fig. 4, 5 and 6). Oftenly the retained nucleus, close to the blood vessels, are pycnotised and excessively saturated with alkaline pigments (Fig. 5 and 6). Bile canals are very rarely observed, as well as the occurrence of bile pigment granules in the cytoplasm of liver cells. As the inflammation processes going deeper, the walls of the large blood vessels swell and then undergoig fragmentation and decompostion (Fig. 5). There are visible fine-celled infiltrates penetrated between the variable liver cells, (Fig. 5 and 6). Then follows the atrophy of capillaries and the decomposition of the endothelial cells. Sometimes a thin layer of cytoplasm retaining itself around the remaining nuclei of liver (Fig. 5). The necrobiotic processes intensifying themselves in liver. As result, fiberous connective tissues remained in few different quantities (Fig. 6).

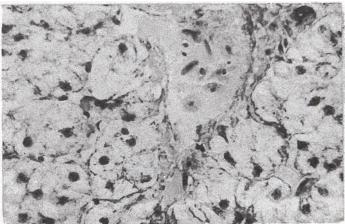


Fig. 5. Fragmentation and decomposition of blood vessels by chronic inflammation in liver of eel with large stalked tumour

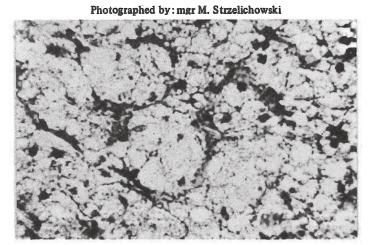


Fig. 6. Development of connective tissues in liver of eel with large stalked tumour Photographed by: mgr M. Strzelichowski

Similarly as in liver, inflammation and degeneration changes in kidney are intensified in different degrees. In kidney of eels with flat and flaccid papillae are observed inflammable changes covering in the first place the renal glomeruli, in less degree the renal canals, the interstitial tissues as well as the blood vessels (Fig. 7). In the enlarged swollen renal glomeruli occurring proliferation of the endothelial cells loops. In few renal glomeruli have been observed the exudative inflammation, moreover the symptoms of proliferation. In these glomeruli following the thickening of the basement zone of the glomerulus loops. In the spaces of Bowman capsules occurring a fiberous discharge with separate blood elements. The changes in glomeruli accompanying with different intense degenerated changes in the renal canals. The parenchymatous degeneration characterising

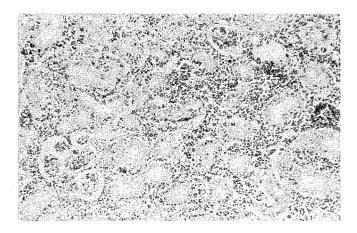


Fig. 7. Renal glomeruli in different degrees of degeneration in kidney of eel with flat and flaccid tumour Photographed by: mgr M. Strzelichowski

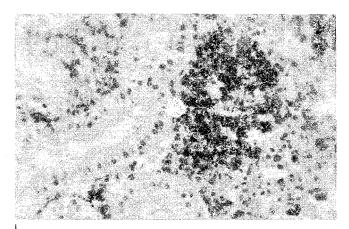


Fig. 8. Hemosiderin deposites in the kidney interstitial tissue of eel with flat and flaccid tumour. Degeneration changes in renal canals

Photographed by: mgr M. Strzelichowski

itself in the cytoplasm of the epithelial cells as well as the swollen and decomposed nucleus of the cells. In the lumen of the fine renal canals occurring fragments of desquamated epithelial cells. The fine capillaries surrounding the renal canals are dilated and filled with erythrocytes. The occurring fine-celled infiltrate, oftenly concentrating around the vessels and sometimes penetrates between the swollen tissue cells. Besides, in the interstitial tissues occurring pigments concentration in the form of goldenbrown granules, sometimes agglutinating itself in masses and forming large pigmented area. Similarly, pigment deposites occurring close to the blood vessels, rarely between the stratified fiberous walls of the blood vessels (Fig. 7 and 8). This pigment gives positive

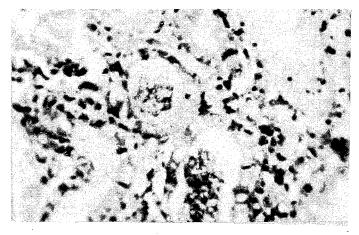


Fig. 9. Degeneration of parenchymatous epithelial cells of renal canals in kidney of eel with large stalked tumour. (In the lumen of the canals are present granules of albuminous masses)

Photographed by: mgr M. Strzelichowski

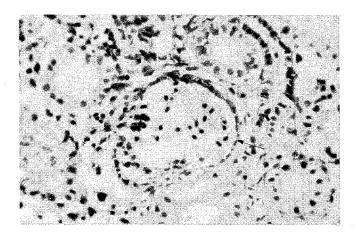


Fig. 10. Decomposition of renal glomerulus (erythrocytes are visible in the Bowman capsules)

Photographed by: mgr M. Strzelichowski

reaction with iron. This is probably hemosiderin. Near to these places occurring macrophagi abundantly filled with the same granules. In places where hemosiderin deposites are formed, the cells of parenchymatous tissues undergoing considerable regressive changes leading to necrosis.

In kidney of eels with large stalked papillae, the observed inflammable changes leading to the decomposition of most of blood vessels.

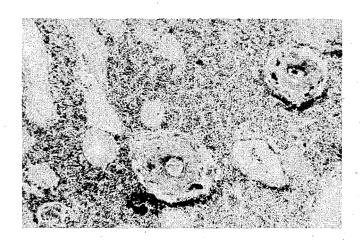


Fig. 11. Fragementation and decomposition of the arteries walls in kidney of eel with stalked tumour Photographed by: mgr M. Strzelichowski

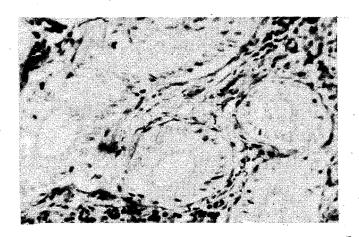


Fig. 12. Development of connective tissues in the interstitial tissue of kidney from eel with stalked tumour

Photographed by: mgr M. Strzelichowski

The intensive process of illness in the renal glomeruli leading to their decomposition and necrosis (Fig. 10). The majority of the renal canals of flatten epithelial cells having the lumen intensively filled by these cells, rarely coagulated protein mass of colloidal appearance (Fig. 9). At the same time, in the arteries being ascertained the increasing inflammation and fragmentation of the membranes. Sometimes, in separate cases leading to decomposition and necrosis of the arteries, (Fig. 11). In this damaged tissue following the proliferation of connective tissues, particularly visible in various places of renal glomeruli nd canals (Fig. 12). Where hemosiderin deposites occurring in large amount to that contributed in the damaged parenchymatous tissues of kidney.

#### DISCUSSION

Histopathological examination showing in different degrees, the intensive regressive and degeneration changes in liver and kidney of diseased eels affected by papillomatosis. The partial changes are related mostly to type of papillae occurring on skin of diseased eels It was ascertained in most of the cases affected by flat and flaccid papilla, that the inflammation changes in the observed internal organs, generally, taking place in sharp forms. In liver, they are comprising in the first place to parenchyma cells and bile canals, and in few degrees the blood vessels. The regressive changes in the cytoplasm of liver cells (fading parenchyma and degenerating vacuole) as well as the nucleus, causing the formation of colliqative necrosis focus. In vessels lumen, positive congestion is observed which favours the formation of slow blood circulation, injured the endothelial as well as the same blood. In some parenchymatous areas reaching to injury the blood capillaries and the outflow of interstitial blood occurs. As a result of the severly injured liver cells, are observed the disturbance in the process of altering the organ pigments chiefly in the bile. In the performed pathological process in kidney, evident changes of inflammation and degeneration covering in the first place the renal glomeruli and in less degree the remaining elementary organs. In glomeruli, the proliferation of cells elements in glomerulus and the vascular loops, oftenly is caused by the occlusion of blood vessels brought to glomerulus. Owing to the occurred inflammation and degeneration changes, the remaining glomeruli series are excluded with the excreation processes. This in turn leading to secondary degeneration changes in the cytoplasm of the epithelial cells of renal canals. In consequence to its accumulation in it, harmful transformed metabolic substances occurring in the glomeruli and renal canals. Excess quantity of hemosiderins are evident, probably form the defected processes of blood circulation.

Pathological changes in kidney, considerably increasing in eels with large stalked papillae and superfacial rugged. They are characterising themselves as chronic inflammation forms which combine with severly changes in blood vessels as well as in the growing connective tissues in the destroyed areas, decomposed renal canals and glomeruli.

Also inflammation changes in liver, of the same type of papillae were evident in the form of chronic inflammation. The decomposition of the cytoplasm of liver cells as well

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as changes in blood vessels and their atrophy in the latter cases are contributed to the considerable defection of the metabolic processes of liver and properly damaging its function. Then, following the development of connective tissues leading to partial fiberous organs.

The degrees and profound inflammation, degeneration and necrosis changes in liver and kidney of diseased eels affected by papillomatosis anguillarum would be indicated by the action of strongly existing factor which perhaps be virus. The character of virus illness affecting the structure of tumours stated by **Schäperclaus** (1953), as well as the results from the work of **Pfitzner** (1969), and **Schubert** (1969). The authors isolated the virus with tumour papillae as well as with the growing cells of infected blood of diseased eels and defined its structure **Schubert** (1969).

Changes of the same characters in the internal organs of *Ichtalurus punctatus* are observed by Wolf et al., (1972) through experimental creation of virus disease (CCVD).

The authors obtained the material for histopathological examinations after 48–72 hours of intramuscular injection of homogonized filtrate of growing virus.

The profuound degrees of the observed pathological changes in liver and kidney of eels with papillae, probably due to the long action of disease and partially perhaps to the influence of unsuitable environment in Szczecin Firth.

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Translated: I Soliman

## ZMIANY HISTOPATOLOGICZNE W WĄTROBIE I NERKACH WĘGORZY ANGUILLA ANGUILLA (L.) PRZY PAPILLOMATOSIS ANGUILLARUM

#### Streszczenie

Badania histopatologiczne wątroby i nerek pobranych z 25 sztuk węgorzy chorych na brodawczycę, wskazują na różny stopień nasilenia zmian zapalnych i zwyrodnieniowych w tych narządach, uzależniony częściowo od typu i wielkości narośli występujących na skórze ryby. Stwierdzono, że przy naroślach płaskich, rozlanych, zmiany zapalne w badanych narządach przebiegają najczęściej w formie ostrej. W wątrobie prowadzą one do powstania ognisk o charakterze martwicy rozpływnej, zaburzeń w krążeniu oraz do upośledzenia procesów przemiany barwnikowej, głównie żółciowej. W nerkach, zmiany zapalne obejmują przede wszystkim kłębki nerkowe, w mniejszym stopniu pozostałe elementy narządu. W kłębkach nerkowych proliferacja elementów komórkowych w gromelurus, lobulacja pętli naczyniowych, rzadziej wysięk w przestrzeni torebki Bowmana, świadczą o toczących się procesach zapalnych i zwyrodnieniowych, powodujących wyłączenie wielu kłębków z czynnego procesu wydalania. Prowadzi to do wtórnych zmian zwyrodnieniowych w cytoplazmie komórek nabłonkowych kanalików nerkowych, wskutek gromadzenia się w niej szkodliwych substancji przemiany metabolicznej.

W nerkach węgorzy o naroślach dużych, szypułkowatych lub o postrzępionej powierzchni, zmiany zapalne przebiegają przeważnie w formie zapalenia przewlekłego. Wiąże się to z cięższymi zmianami w naczyniach krwionośnych, a także rozrostem tkanki łącznej w miejscu zniszczonych kanalików i kłębuszków nerkowych. Również w wątrobie tych węgorzy obserwowane objawy wskazywałyby na przewlekłą formę toczącego się zaplenia. W narządzie tym obserwuje się znaczny rozplem tkanki łącznej zrębu. W krańcowych przypadkach w komórkach wątrobowych następuje rozpad cytoplazmy oraz zanikanie naczyń krwionośnych.

Stopień i charakter zmian zapalnych, zwyrodnieniowych i martwiczych w wątrobie i nerkach węgorzy chorych na brodawczycę, wskazywałby na działanie czynnika silnie toksycznego, jakim może być wirus. Różny stopień nasilenia zmian obserwowany przy różnych typach brodawek spowodowany jest, prawdopodobnie, dłuższym działaniem tego czynnika na organizm ryby.

# FUCTORATAJOFNYECKNE N3MEHEHNA B REYEHN N ROYKAX JFPEN ANGUILLA ANGUILLA (..) RPN PAPILLOMATOSIS ANGUILLARUM

#### Резюме

Гистопаталогические исследования печени и почек, взятых из 25 экземпляров угрей, больных папилооматозом, указывают на разную степень интенсификации воспалительных и легенеративных изменений в этих органах, которая зависит частично от типа и размеров опухолей, образующихся на кожном покрове рыб. Установлено, что при плоских, разлившихся опухолях воспалительные изменения в исследуемых органах протекают чаще всего в острой форме. В печени они приводят к возникновению очагов колликвационното некроза, к напушениям в кровообращении, а также к недоразвитию процессов пигментационного обмена, главным образом в жёлчном пузыре. В почках воспалительные изменения охватывают прежде всего почечные клубочки,
в меньшей степени — остальные элементы органа. В почечных клубочках про-

лиферация клеточных элементов в gromelurus, лобуляция сосудистых петель, реже — экссудат в области капсулы Боумана, свидетельствуют о протекающих воспалительных и дегенеративных процессах, вызывающих выходиногих клубочков из активного процесса выделения. Это приводит к вторичным дегенеративным изменениям в цитоплазме эпителиальных почечных каналов вследствие накопления в ней вредных веществ метаболического обмена.

В почках угрей с большими опухолями, сосочковидными или с неровной поверхностью, воспалительные изменения протекают преимущественно в виде хронического воспаления. Это связано с более сложными изменениями в кровеносных сосудах, а также с разрастанием соединительной ткани в области повреждённых почечных каналов и клубочков.

Также и в печени этих угрей наблюдаемые явления могут указывать на хроническую форму протекающего воспаления. В этом органе наблюдается значительное разрастание соединительной ткани субстрата. В исключительных случаях в клетках печени начинается распад цитоплазмы, а также отмирание кровеносных сосудов.

Степень и характер воспалительных, дегенеративных и некротических изменений в печени и в почках угрей, больных папилломатозом, могут указывать на действие тожсического фактора большой силы, каким может быть вирус. Разная степень интенсификации изменений, отмечаемая при разных типах папилломы, вызвана, вероятно, длительным воздействием этого фактора на организм рыбы.

Address: Received 1 III 1973

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