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PATHOLOGICAL CHANGES IN KIDNEYS OF INTENSIVELY BRED BROOK TROUT (SALVELINUS FONTINALIS MITCHILL, 1815)

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Abstract

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Description is given of histopathological changes in kidneys of juvenile and generative brook trout (*Salvelinus fontinalis* Mitchill, 1815) under conditions of intensive rearing and feeding with granulated food mixture and meat scraps. Epithelial cells of renal tubuli showed necrosis and dystrophic calcification with subsequent giant cell granulomatous reaction and formation of stones consisting of brushite $CaH(PO_4) . 2 H_2O$, as proved by roentgenometric analysis. An unknown detrimental substance is taken into consideration as pathogenetical agent.

Salvelinus fontinalis, visceral granulomatosis, necrotizing nephrosis, nephrocalcinosis, nephrolithiasis, brushite.

Connected with intensive breeding of Salmonids, there is a rising number of cases reported as nephrocalcinosis (Besse, Kinkelin and Levaditi 1968), visceral granulomatosis (Dunbar and Herman 1971), and necrotizing nephrosis (Vítovec, Vladík and Červinka 1974).

In *Salmo gairdneri* suffering from nephrocalcinosis, white deposits of tricalciumphosphate were described. They were oriented towards the walls of tubuli and surrounded by resorptive granulomas. *Corynebacteria* were isolated from some of the affected kidneys. The authors tend to consider metabolic calcinosis as the ethiological factor. The disease appeared when sea fish was replaced by pelleted food.

Visceral granulomatosis in *Salvelinus fontinalis*, in addition to typical changes on visceral organs, is characterized by granulomas and tubular degeneration resulting in lithiasis in the caudal part of kidneys. The authors, in agreement with Wood and Yasutake (1956) in Dunbar and Herman (1971), failed to isolate any infective agent. They concluded that the disease is an inflammatory reaction to irritating chemical substances in food.

With necrotizing nephrosis in *Salvelinus fontinalis*, dystrophical calcification of desquamated epithelia was described together with a secondary granulomatous reaction to foreign bodies. No grampositive immobile diplobacilli were isolated. Sulfonamides were taken into consideration as etiological factor.

The present report is limited to description of the culminating pathological process in the kidney of *Salvelinus fontinalis* and the resulting grave nephrolithiasis.

Materials and Methods

Post-mortem examination was performed in one generation (4+) brook trout (*Salvelinus fontinalis* Mitchill, 1815) of 440 mm length and 1200 g mass, and one juvenile (1+) brook trout of 180 mm and 75 g, fed with pelleted food mixture of inland provenience and with meat scraps.

The kidney tissue was fixed in 10% neutral formol, decalcified in 0.5 M Chelaton III solution at pH 7–8, and, embedded in paraffin wax. Slides were stained by hematoxyline-eosine, the PAS method, Masson's green trichromium, for reticuline by Gomori's, for calcium by Kossa's, for iron by Perls' and for microbes by Gram's method.

Physico-chemical analyses of concrements were carried out on the diffractometer Chirana GON-3 with a characteristical CoK-alfa radiation, at 28 kV voltage, 14 mA current intensity, 1° per minute goniometer rotations and 600 mm registration shift.

Photographs of the superficial structure of concrements were taken, after coating with gold, by the scanning microscope Jeol, JSM-50 A.*)

Results

The most striking post-mortem finding in the adult fish were protuberancies of a light colour protruding below the renal capsula particularly on the caudal, enlarged part of the kidney (Fig. 1).

After cutting the kidney with a grating noise of the knife, a large number of concrements was removed from the kidney tissue. They had the size from sand particle to 5 mm, were of hard consistency, irregular shape and white to faintly yellowish colour (Fig. 2). No alterations were observed in the rest of the organs.

Microscopical examination of the kidney showed enormously dilated tubular spaces, transformed here and there to cystic formations littered with cylindric, mostly desquamated epithelium (Fig. 5). A layer of mesenchymal cells was below the epithelium, and, a fibrous basophilic mass was inside the dilated tubules. In some of them, granulomatous inflammatory reaction with foreign body giant cells could be observed (Fig. 6, 7).

In the juvenile fish, there were different greyish mutually communicating strips on the kidney, resembling marble (Fig. 8). Microscopically, there was an apparent

Table 1

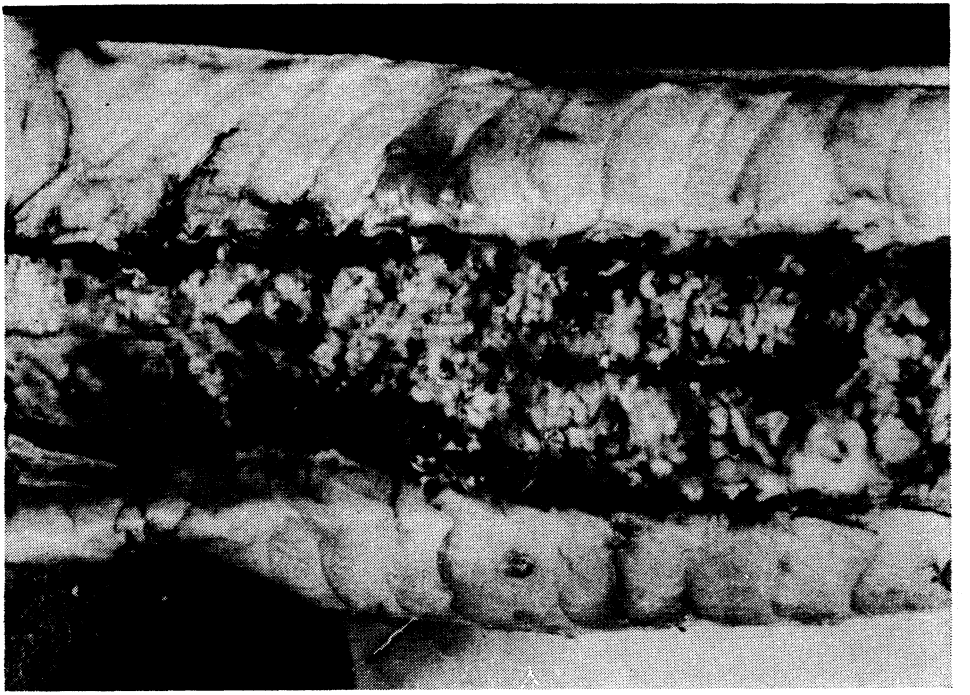
Roentgenometrical analysis of concrements

	Tabellar values			
No. of reflexion	Intensity	*d* (Å)	Intensity	*d* (Å)
1	100	7.583	100	7.624
2	39.3	4.233	23	4.228
3	10	3.790	32	3.768
4	46.4	3.042	48.3	3.036
5	22.6	2.921	9.5	2.920
6	1.8	2.852	4.4	2.813
7	43	2.621	17.4	2.620
8	5.2	2.553	8.4	2.534
9	4.1	2.522	8.4	2.534
10	13.6	2.432	6.1	2.415
11	6	2.266	9.5	2.252
12	16.2	2.167	11.2	2.169
13	14	2.143		
14	12	2.001	12.9	1.996
15	5	1.896	14.6	1.894
16	16.3	1.876	10.1	1.880
17	18.2	1.814	7.8	1.817

*) I am obliged to Dr. J. Králík, School of Mining and Metallurgy, Ostrava, for the analysis and photographs of concrements by the scanning microscope.

Fig. 1.
Enlargement of the caudal part of kidney with numerous concrements in the adult brook trout.

Fig. 2.
Renal concrements.



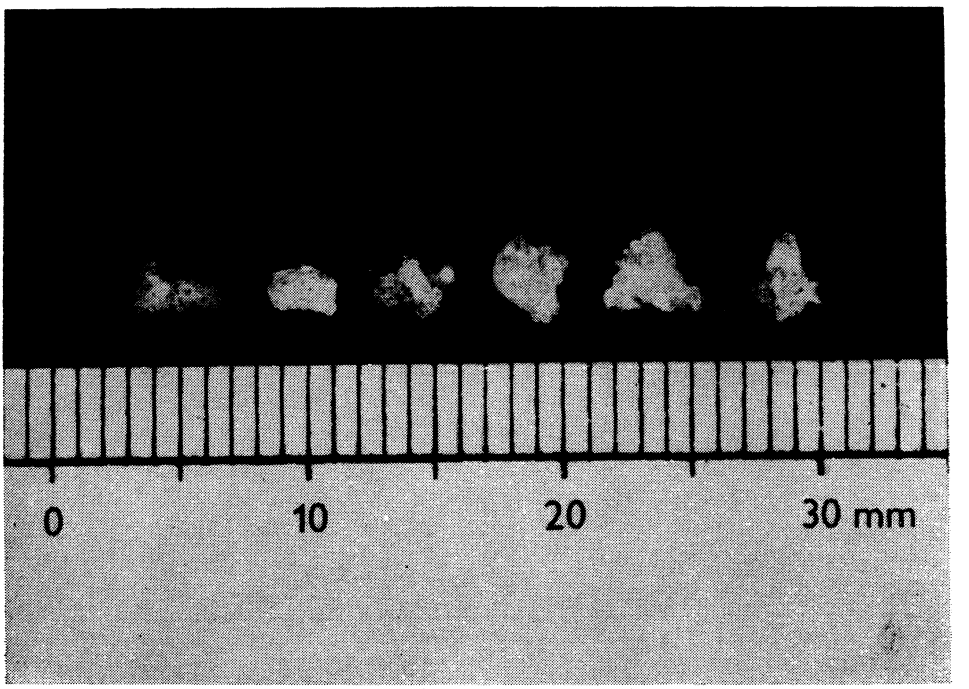

0
10
20
30 mm

Fig. 3.
Brushite crystal on the surface of concrement. Magnification 115×.

Fig. 4.
Brushite crystal on the surface of concrement. Magnification 345×.

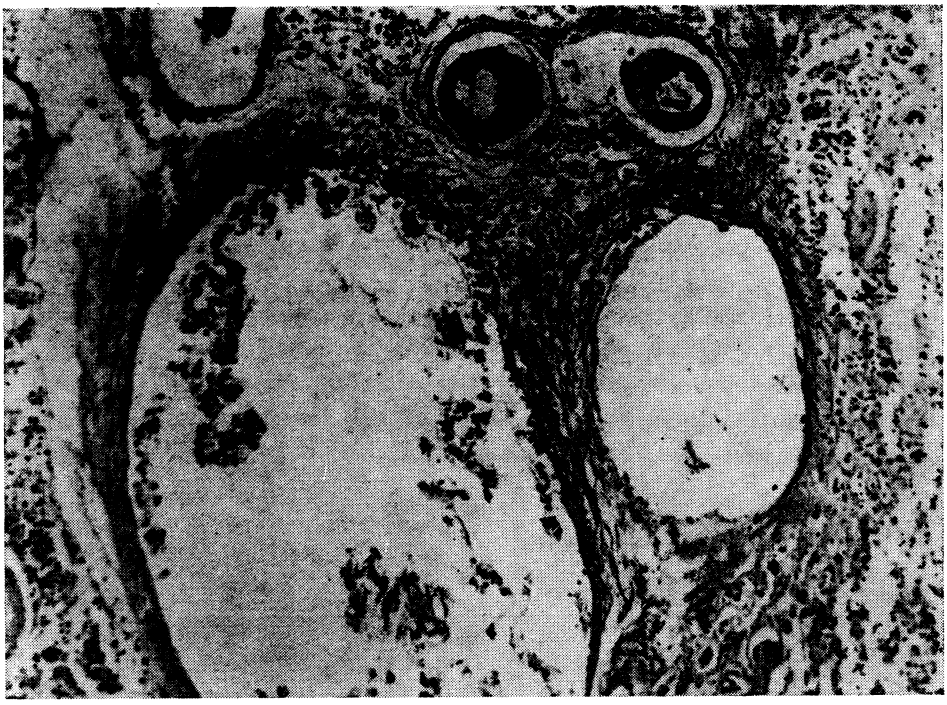


Fig. 5.
Enormously enlarged renal tubules in decalcifited renal tissue. Inflammatory granulomatous reaction evident in the tubules. HE, 65×.

general reduction of renal tubules. They were of irregular shape, enlarged, deformed and showed a distinctly thickened wall. The tubules were filled with dystrophically altered epithelial cells. In the amassed cells were large deposits of calcium salts. Granulation developed around their crystals and filled partly or totally the tubules. In the granulation tissue, histiocytes rich in eosinophilic plasma predominated. There was a minor admixture of fibroblasts, while foreign body polynuclear cells occurred sporadically (Fig. 9, 10, 12). At intervals, thin diaphragms of renal mesenchyma with evident fibroproductive alterations penetrated and formed partititions inside the renal tubuli (Fig. 11). Apart from the altered tubules, there were also tubules with normal epithelium, filled with granular eosinophilic masses. In some areas of the kidney, interstitium showed fibrous alterations of the haemopoetic tissue where decreased pigmentation could be observed. The eosinophilic mass in the tubules as well as the minute grains in the histiocytes of granulomas were stained by the PAS method.

The chemical analysis of concrements was based mainly on the results of diffraction of the roentgenometric examination. Maxima were satisfactorily relevant to tabellar values for brushite (calciumhydrogenphosphate $CaH(PO_4) . 2 H_2O$, with the exception of the comparatively intensive reflexion of 2.143 d-value which is not listed in Schneider's tables, but is to be found in the atlas (1974). Microscopical examination showed tubular crystals of brushite (monoclinical system) with evidently good fissility (010), reminding of gypsum (Fig. 3, 4).

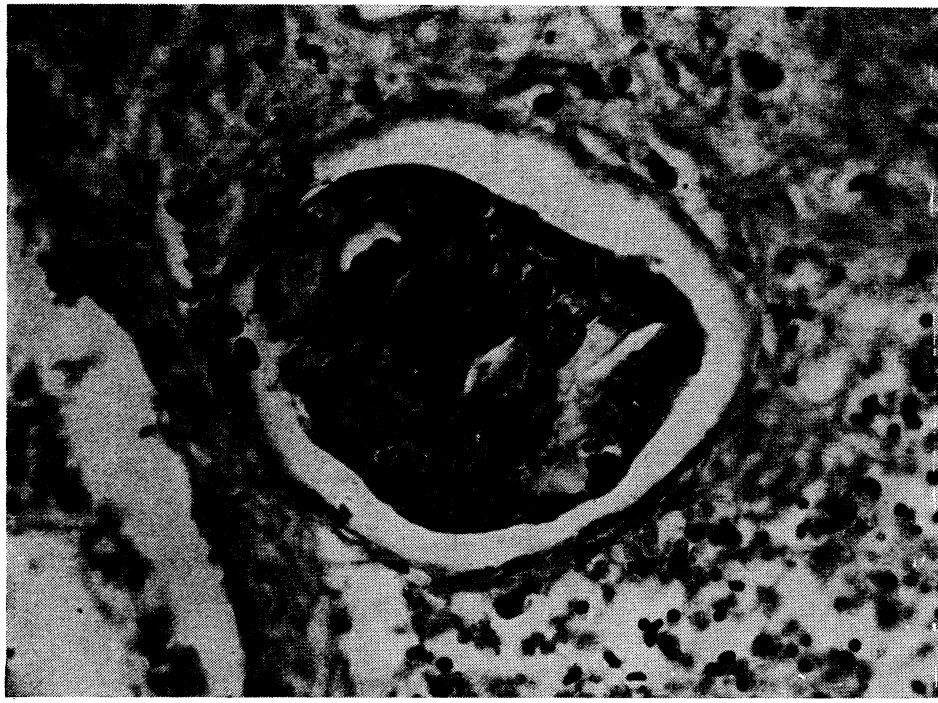


Fig. 6.
Transformed renal tubule. Histiocytes and giant cells around spaces left by dissolved calcium salts (decalcified kidney). HE, magnification 170×.

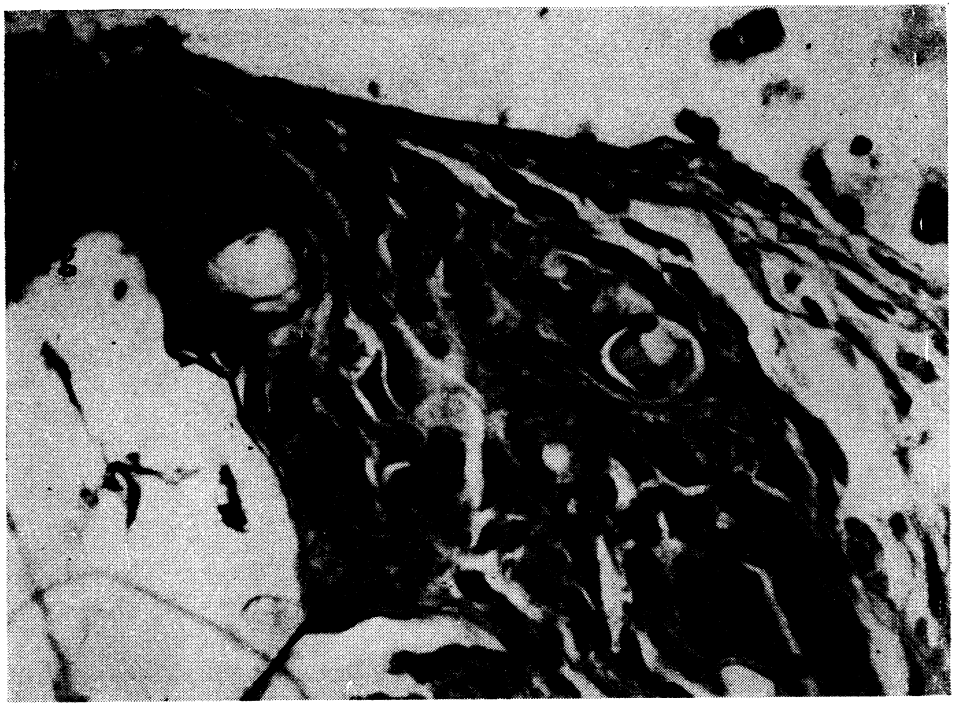
Discussion

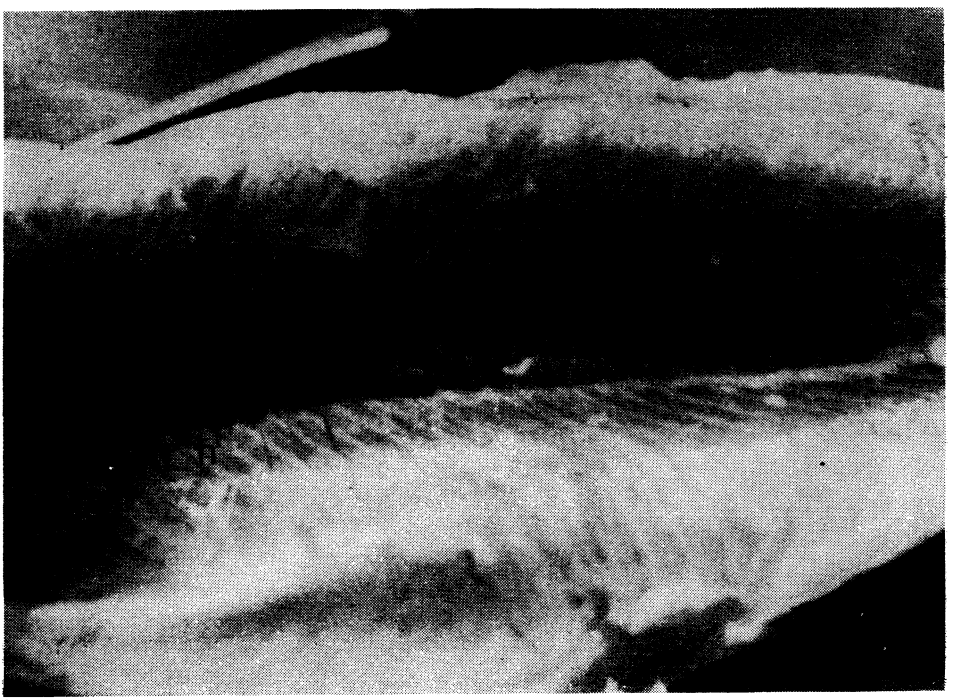
The described morphological alterations in kidneys of both post-mortem examined fish are identical with the changes reported by Vítovec, Vladík and Červinka (1974) in necrotising nephrosis. The histopathological picture corresponds to findings described with visceral granulomatosis in *Salvelinus fontinalis* (Dunbar and Herman 1971) and partly also with nephrocalcinosis in *Salmo gairdneri* (Besse, Kinkelin and Levaditi, 1968). Similarly as Vítovec et al. (1974), we proved no grampositive diplobacilli in histological sections, while Vladík et al. (1974), succeeded in cultivating them from altered kidneys.

Considering the character of alterations and the failure to find any infective agent (Wood and Yasutake, see Dunbar and Herman 1971), intoxication by some unknown food component seems to be the more probable etiological factor. A detrimental effect of sulphonamides, however, was reliably excluded by scanning performed earlier.

Fig. 7.
Margin of granulomatous process with histiocytes and foreign body giant cells (g). Decalcified kidney, HE, magnification 270×.

Fig. 8.
Altered kidney from juvenile brook trout. Macroscopically intact part in foreground (h).





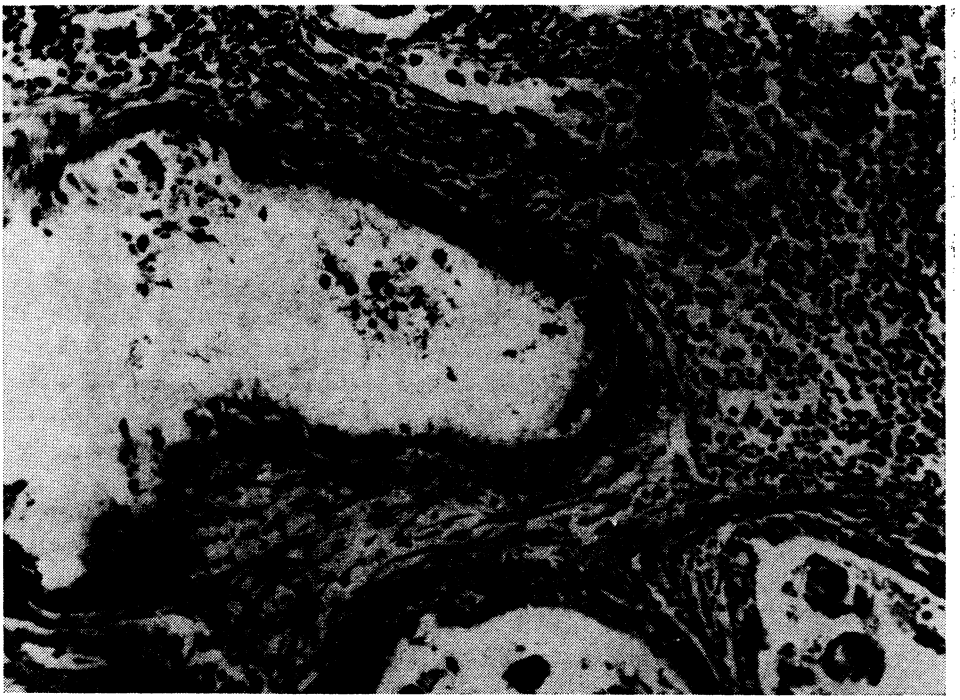


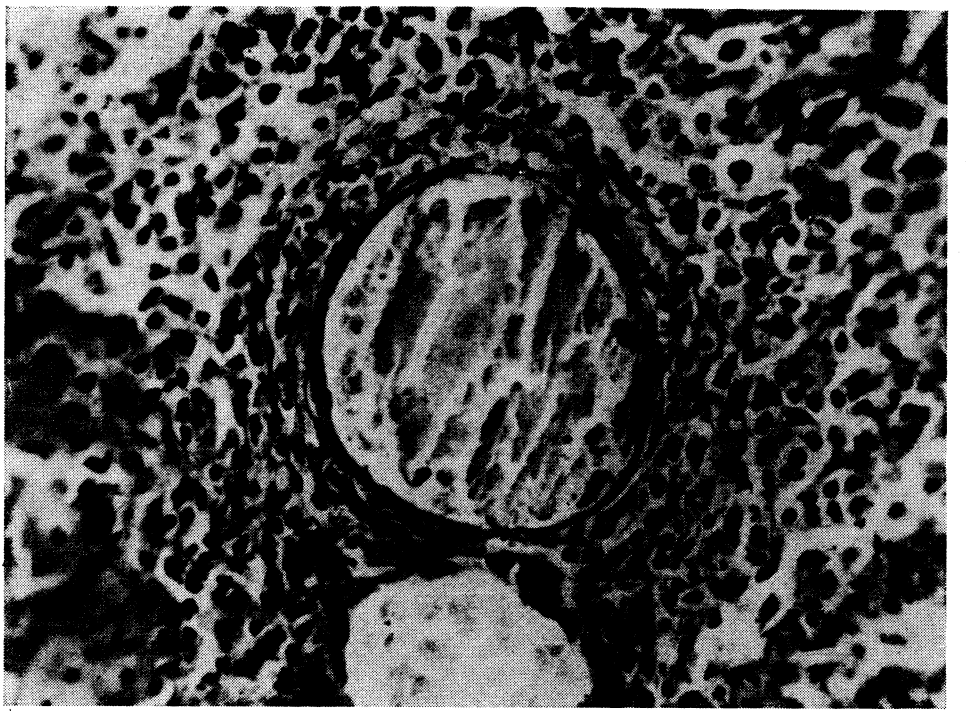
Fig. 9.
Enlarged and irregular renal tubule with dystrophic epithelial cells and altered haemopoietic tissue between enlarged tubules. HE, magnification 90×.

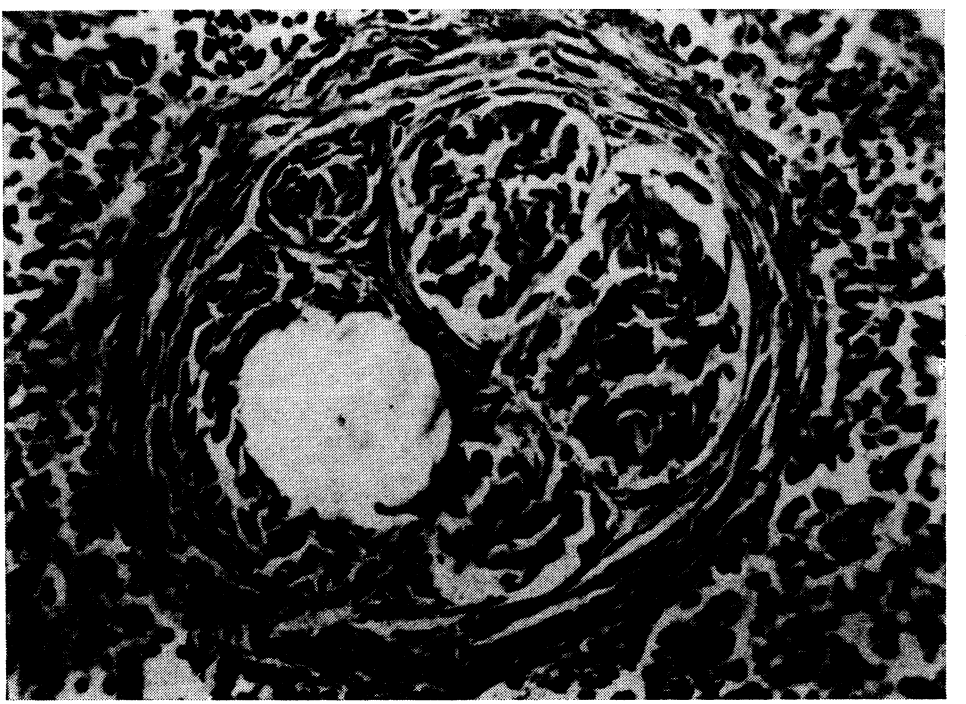
Patologické změny na ledvině sivena amerického (Salvelinus fontinalis Mitchill, 1815) v intenzívním chovu

V práci jsou popsány histopatologické změny v ledvinách juvenilního a generačního sivena amerického (*Salvelinus fontinalis* Mitchill, 1815) z podmínek intenzivního odchovu při výživě granulovanou krmnou směsí s masitými odpady. Byla nalezena nekróza buněk epitelu ledvinných kanálků a jejich dystrofická kalcifikace s následnou obrovskobuněčnou granulomatosní reakcí a dále tvorba kamenů složených z brushitu (kalciumhydrogenfosfát $CaH(PO_4) . 2 H_2O$), jak prokázala rentgenometrická analýza. V etiopatogenezi se uvažuje o možném toxickém působení neznámé škodliviny v krmivu.

Fig. 10.
Renal tubule filled with precipitation calcium salts, initiating histiocytic reaction on periphery. HE, magnification 170×.

Fig. 11.
Thickening of wall and fibrotic partitions of renal tubule. HE, magnification 170×.





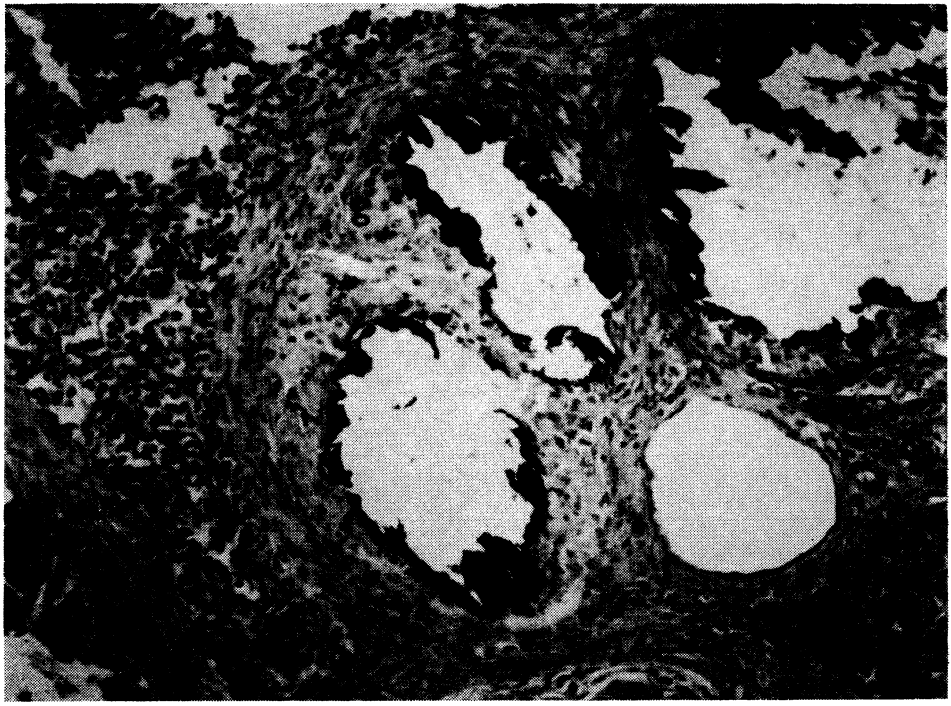


Fig. 12.
Amorphous mass containing calcium surrounded by giant cell reaction and its fibrotic transformation. (g = foreign body giant cell). Kossa, magnification 90×.

Патологические изменения почки американской пальи (Salvelinus fontinalis Mitchill, 1815) в интенсивном разведении

Приводится описание гистопатологических изменений почек молодых и взрослых представителей пальи американской [*Salvelinus fontinalis* Mitchill, 1815] в условиях интенсивного разведения с питанием гранулированной кормовой смесью и мясными отходами. Происходит некроз клеток эпителия почечных канальцев и их дистрофическая кальцификация с последующей крупноклеточной грануломатозной реакцией и образование камней из брушита [кальцийгидрогенфосфат $CaH(PO_4) \cdot 2H_2O$], соответственно рентгенометрическому анализу. В этиопатогенезисе предполагается токсическое воздействие неизвестного вредного вещества в корме.

References

BESSE, P. — de KINKELIN, P. — LEVADITI, J. C.: La Néphrocalcinose de la truite arc-en-ciel (Salmo irideus, Gibbons, 1855). Bill. Off. int. Epiz., **69** (9—10), 1968: 1391—1399.
DUNBAR, C. E. — HERMAN, R. L.: Visceral Granuloma in Brook Trout (Salvelinus fontinalis). J. Nutr., **101**, 1971: 1445—1452.
SCHNEIDER, H. J.: Technik der Harnsteinanalyse. VEB Georg Thieme Leipzig, 1974.
VÍTOVEC, J. — VLADÍK, P. — ČERVINKA, S.: Nekrotizující nefróza s granulomy a četnými

cystopapilárními adenomy při hromadném onemocnění sivena amerického a pstruha duhového. Vet. medicína (Praha), č. 5, 19 (XLVII), 1974: 283–287.
VLADÍK, P. – VÍTOVEC, J. – ČERVINKA, S.: Taxonomie grampozitivních nepohyblivých diplobacilů izolovaných z nekrotizujících nefróz sivena amerického a pstruha duhového. Vet. medicína (Praha), **19**, č. 4 (XLVII), 1974: 233–238.
Selected Powder Difraction Data for Minerals Published by the Joint Committee on Powder Difraction Standards. Philadelphia, 1974 (atlas).