

Structure of the Urinary Bladder in the Pacific Cod

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Abstract The urinary bladder of the Pacific cod, *Gadus macrocephalus* was investigated. A pair of ureters was found to unite and form a sac-like protrusion on the posteroventral aspect of the gonad. This protrusion was confirmed as the urinary bladder based on its histology and the presence of ammonia and urea in the fluid within this structure. The urinary bladder consisted of a central lumen and two lateral expansions found on the right and left sections. The luminal epithelial cells of the Pacific cod urinary bladder, unlike those of other animals, were characterized by microvilli on the free surface, and the presence of a number of vesicles and mitochondria across the apical portion on the cells. This suggests that the luminal epithelium of the urinary bladder might be actively engaged in transportation of water or other materials from the urine. The Pacific cod urinary bladder may therefore be closely associated with osmoregulation by reabsorbing water from the urine. Ammonia-N and urea-N levels of 65–213 $\mu\text{g}/\text{dl}$ and 1.5–3.5 mg/dl were measured in the fluid which filled the urinary bladder.

A considerable body of literature exists concerning the structural organization and physiology of the fish kidney (c.f. Ozaki, 1977). The same is not true, however, regarding our knowledge of the urinary bladder. Indeed, not all fishes possess this structure, e.g. the urinary bladder is present in some gobioid fishes and absent in others (Arai, 1964). The urinary bladder has previously been considered to function solely as a storage chamber for urine. Recently, however, it has been demonstrated that the urinary bladder of the starry flounder, *Platichthys stellatus* also plays an important role in osmoregulation (Hirano *et al.*, 1971, 1973). While participating in groundfish surveys, we had the opportunity to examine specimens of the Pacific cod, *Gadus macrocephalus* and confirmed the presence of a urinary bladder in this fish. This paper describes the gross and histological organization of the urinary bladder of the Pacific cod and presents an analysis of the content of ammonia and urea in the fluid inside the bladder.

Material and method

Specimens of Pacific cod were obtained during Japan-U.S. cooperative groundfish trawl surveys in the Aleutian Islands region, July–September 1983; Gulf of Alaska, August–October 1984 and eastern Bering Sea, May–July 1985. Specimens, varying from 50 to 80 cm in fork length, were

frozen aboard the research vessels and transported to the laboratory for examination of the gross morphology of the urinary bladder and ureters. Some bladders were removed and preserved in formalin for light microscopy. For electron microscopy, small pieces of bladder tissue were pre-fixed in a cold solution of 2% glutaraldehyde, 2% paraformaldehyde, 0.25% calcium chloride and 10% sucrose in 0.1 M cacodylate buffer adjusted to pH 7.4. Fluid was collected by aspiration into syringes from the urinary bladders of both male and female Pacific cod. The fluid was frozen in vials and subsequently analyzed for ammonia-N and urea-N. Urinary bladders preserved in formalin were embedded in paraffin and sections made according to standard procedures. For electron microscopy, small pieces of tissue were post-fixed in 2% osmic acid, dehydrated with ethanol and embedded in Epon 812. Thin sections were double-stained with uranyl acetate and lead citrate and observed on JEM-300 electron microscope. Ammonia-N and urea-N levels were measured by modified techniques of Gips *et al.* (1970) and Wybenga *et al.* (1971).

Results

Gross morphology. Both right and left ureters were invested in a common connective tissue sheath and superficially appeared as a single duct. The ureters extended from the kidney and formed

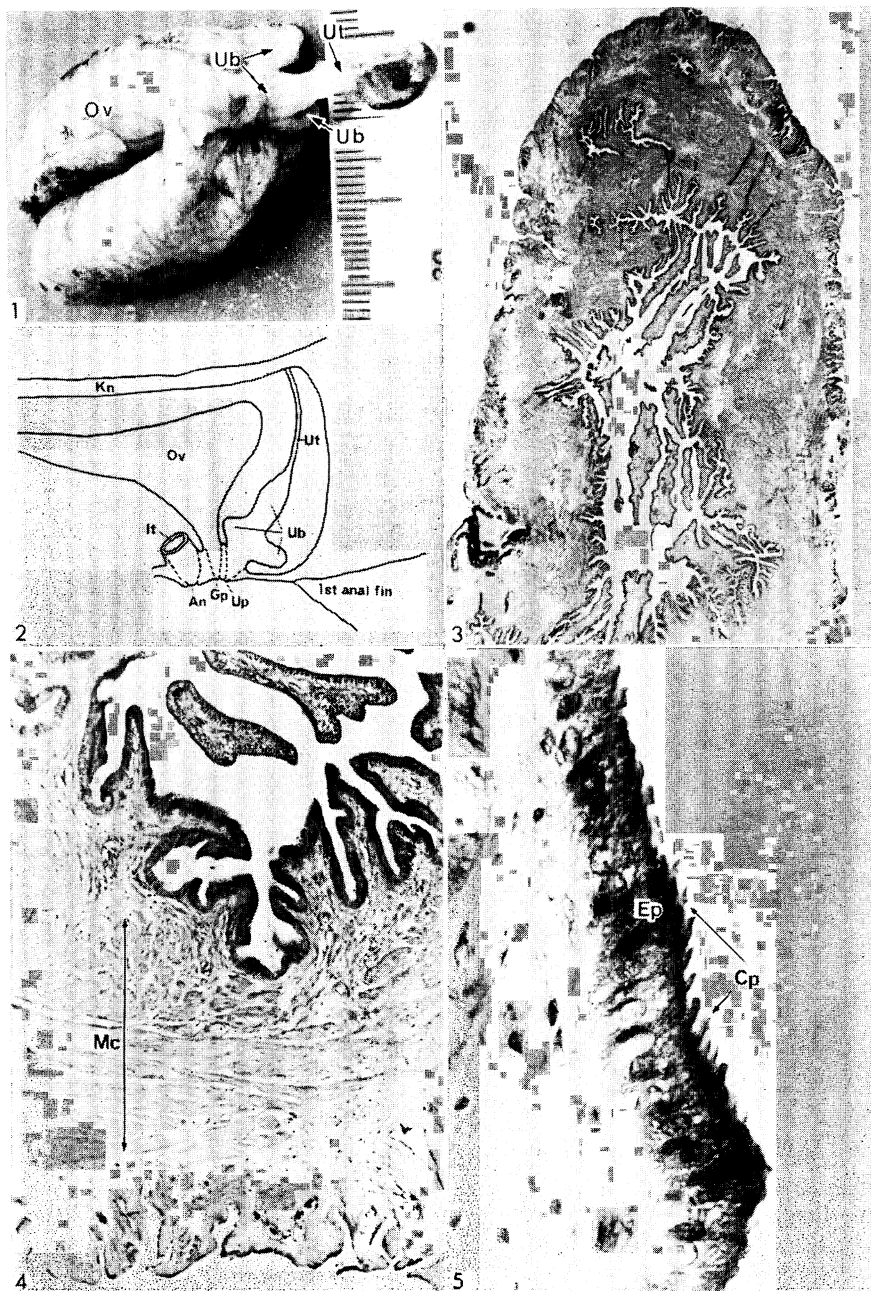


Fig. 1. Urinary bladder found in the Pacific cod, *Gadus macrocephalus*. Ov, ovary; Ub, urinary bladder; Ut, ureter.

Fig. 2. Simplified drawing of the female urogenital system in the Pacific cod. An, anus; Gp, genital pore; It, intestine; Kn, kidney; Ov, ovary; Ub, urinary bladder; Up, urinary pore; Ut, ureter.

Fig. 3. Cross section of the empty urinary bladder of the Pacific cod. Longitudinal folds of mucous membrane were seen in the lumen of the urinary bladder. $\times 3.5$.

Fig. 4. Enlarged view of Fig. 3 showing the thicker muscular coat (Mc) of the urinary bladder wall. $\times 17.5$.

Fig. 5. Mucous epithelium (Ep) of the urinary bladder. Cytoplasmic processes (Cp) were formed on the free surface of the epithelium. $\times 175$.

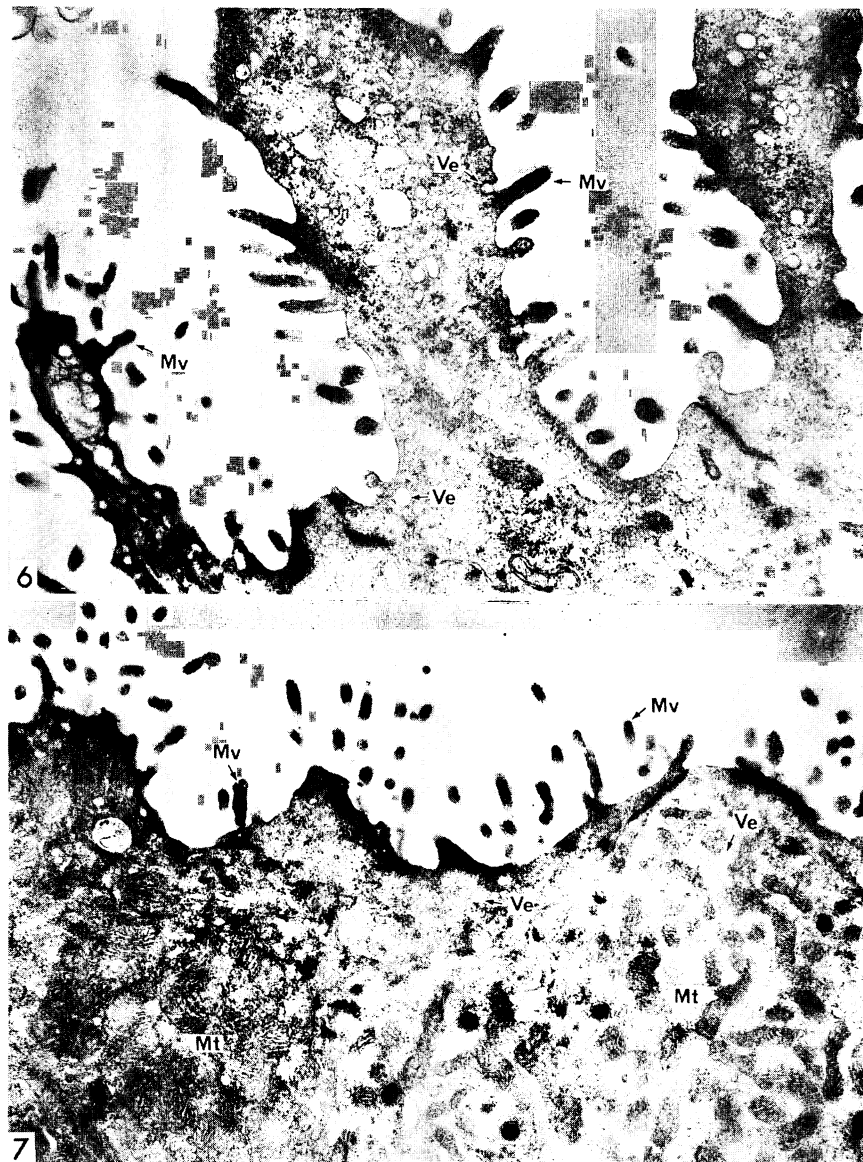


Fig. 6. Electron micrograph of the epithelial cell of the urinary bladder. Note protruded cytoplasmic process with microvilli (Mv) and number of vesicles (Ve) throughout the cytoplasmic process. $\times 19,600$.

Fig. 7. Electron micrograph of the urinary bladder epithelium. The epithelial cell possessed microvilli (Mv) on the free surface, numerous vesicles (Ve) and mitochondria (Mt) in the apex of the cell. $\times 14,300$.

a sac-like protrusion on the posteroventral aspect of the gonads. This protrusion corresponded to the urinary bladder.

The urinary bladder consisted of a central lumen and two lateral expansions (Figs. 1, 2). In many live specimens observed aboard the vessels all three portions of the urinary bladder were distended with fluid. We confirmed the identity

of the urinary bladder based on its histology and the presence of ammonia and urea in the fluid within the structure. Three apertures were situated in front of the 1st anal fin, viz. anus, genital pore and urinary pore, with the anus being the most rostral and the urinary pore the most caudal (Fig. 2).

The left lateral expansion was persistently larger

than the right whether it was filled with fluid or empty. Relative weight of the left expansion was 10–20 mg per 100 g body weight. When fully distended with fluid, the left expansion measured 3 cm in length by 2 cm in width in adult fish weighing 5 kg. The size ranged from 7 mm in length by 2 mm in width in young fish weighing 130 g.

Histological features. Both the ureters and urinary bladders were lined by a simple columnar mucous epithelium. Beneath the epithelium was lamina propria contributing to the mucosa. A submucosa was lacking. External to the mucosa was a well developed muscular layer covered by mesothelium of the peritoneum (Figs. 3–5).

The mucous membrane was organized into longitudinal folds in both the urinary bladder and ureter (Figs. 3, 4) and protruding cytoplasmic processes were formed on the epithelium of the urinary bladder (Figs. 5, 6). The luminal epithelial cell of the urinary bladder was characterized by microvilli on the free surface, numerous vesicles and mitochondria across the apical portion of the cell (Figs. 6, 7).

The lamina propria consisted of loose connective tissue and was thin in both the urinary bladder and ureter. The muscular coat was more well developed in the urinary bladder than in the ureter. Large blood vessels and nerves were distributed in the connective tissue between the muscular coat and the mesothelium. The mesothelium of both the urinary bladder and ureter consisted of a single layer of cuboidal cells.

Results of quantitative analyses of the fluid collected from the urinary bladders of both male and female Pacific cod indicated that urea-N levels (1.5–3.5 mg/dl; average, 2.7 mg/dl) were much higher than ammonia-N (65–213 μ g/dl; average, 141.5 μ g/dl).

Discussion

We verified that a urinary bladder occurred in the Pacific cod. It was present as a central dilated lumen with two lateral expansions. Its gross position and shape corresponded to the urinary bladder of other teleosts (Goodrich, 1930 cited in Kent, 1965; Orr, 1976). The urinary bladder of the Pacific cod differs from that of other animals, e.g. rat (Sandborn, 1974) in the following histological characteristics; 1) the luminal epithelium consisted of a single layer of columnar cells instead of

transitional epithelial cells and 2) the epithelial cell possessed microvilli on its free surface and numerous vesicles and mitochondria were densely distributed in the apex of the cell. Analysis of the fluid contained in the urinary bladder revealed ammonia-N and urea-N. These are among the nitrogen-containing compounds excreted as urine by fishes (Wood, 1958).

In the Pacific cod, the urinary bladder opened independently as the urinary pore and this arrangement is the same as that for a teleost shown by Goodrich (1930 cited in Kent, 1965). In some teleosts, e.g. *Hippocampus* and *Amia*, the oviduct or spermiduct and ureter unite posteriorly and form a urogenital pore (Orr, 1976). In some male elasmobranchs, e.g. *Chlamydoselachus anguineus*, *Squalus sucklii* and *Mustelus* species, the posterior portions of the seminal vesicle and ureter are modified to form a urogenital papilla (Daniel, 1928; Gilbert, 1943; Teshima, 1981).

As described above the urinary bladder is present in some fishes and absent in others. The same phenomenon is reported to occur in other animals, e.g. lizards (Beuchat, 1986).

Hirano *et al.* (1971) described the urinary bladder of the starry flounder, *Platichthys stellatus*, as playing an important role in osmoregulation by modifying the concentration of the urine. If water is reabsorbed from the urine, this could make a contribution to the hydromineral balance. We observed apical microvilli, numerous vesicles and mitochondria in the epithelial cells of the Pacific cod urinary bladder and it might be suggested that water or other materials were being actively reabsorbed. It may also be true that water is being transported alternatively via the intercellular spaces, as in the amphibian urinary bladder (Berridge and Oschman, 1972). In either case, the urinary bladder of the Pacific cod may be closely associated with osmoregulation.

Teleosts are usually considered as ammonotelic animals in which the major components of the nitrogenous excreta are ammonia-N and urea-N. The amount of ammonia-N has been reported to occupy 50–80% of the total amount excreted (Wood, 1958; Oguri, 1970). In three species of marine teleosts, viz. sculpin, *Leptocottus armatus*, starry flounder, *P. stellatus* and blue sea-perch, *Taeniotoca lateralis*, the amount of ammonia-N excreted (mg) is reported to exceed that of urea-N by about 1.5 to 7.5 times (Wood, 1958). Con-

versely, in the Pacific cod, the amount of urea-N was about 20 times larger than ammonia-N.

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マダラの膀胱

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著者等は、マダラの測定及び解剖中に、尿管の下端部が膨張し囊状構造を呈していることを見出した。形態学的観察並びに囊状部内液の定量分析結果より、この囊状構造は膀胱と考えられた。膀胱は、左右一対の尿管がその後端部で融合して形成される正中部の囊状構造とここから突出する左右一対の囊状構造により構成される。マダラの膀胱の上皮細胞は、他の動物のものと異なり、その自由表面に形成される微絨毛、細胞質上部に密に分布する多数の小胞とミトコンドリアにより特徴付けられる。これらの細胞学的特徴から、マダラの膀胱上皮細胞は活発に吸収活動を行っていたことが考えられ、このため、マダラの膀胱は尿から水を再吸収し、浸透圧調節に寄与している可能性が示唆された。船上で凍結して持ち帰った膀胱内液から、尿の代表的成分である 65-213 $\mu\text{g}/\text{dl}$ のアンモニア-N と 1.5-3.5 mg/dl の尿素-N が検出された。

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