

# Electron microscopical studies of the common bile duct in reindeer

**Timo Rahko and Sven Nikander**

College of Veterinary Medicine, Laboratories of Parasitology and Electron Microscopy and Department of Pathology, Postbox 6, SF-00581 Helsinki, Finland.

*Summary:* In a previous publication the authors have described some ultrastructural characteristics of granulated cells in the common bile duct of the reindeer. On the basis of the same material, electron microscopic observations on other tissue elements of bile duct wall are now reported.

The surface and glandular epithelium were composed of tall columnar epithelial cells with villous structures on the luminal surfaces. The parietal cytoplasmic membranes of epithelial cells were equipped with intercellular desmosomes while intraepithelial globule leucocytes did not form any junctional complex with other cells. Apical cytoplasmic areas of superficial epithelial cells showed electron-dense small bodies possibly consisting of mucinous substances. The goblet and deep glandular cells, on the other hand, contained numerous large mucin granules with less electron-dense matrices. It appears that their secretions are more abundant than those in superficial epithelial cells which obviously are absorptive as their main function. The nuclei and other cytoplasmic organelles showed profiles similar to those in epithelial cells generally. The lumen of the bile ducts was usually empty or contained fine-granular or amorphous material. An unusual feature was the presence of parts of globule leucocytes or even almost whole cells occurring freely in ductal secretions.

**Key words:** anatomy, globule leucocyte, mast cell, ultrastructure.

**Rangifer, 10 (1): 17–23**

**Rahko, Timo ja Nikander, Sven.** 1990. Elektronimikroskooppinen tutkimus yhteisen sappikäytävän rakenteesta porolla.

*Yhteenveto:* Aikaisemmassa julkaisussa tekijät kuvasivat poron yhteisen sappikäytävän (ductus hepaticus communis) seinämän jyväsellisten solujen hienorakennetta. Tässä artikkelissa selostetaan saman aineiston perusteella (6 tervettä teurasporoa) elektronimikroskooppisia havaintoja sappikäytäväseinämän muista kudusrakenteista.

Sappikäytäväseinämän pinta- ja rauhasepiteeli koostuu korkeista epiteelisoluista. Pinnallisia epiteelisoluja kattavat säännölliset mikrovillukset, ja niillä on vain vähän ilmeisesti limaa sisältäviä jyväsia solulimassaan. Rauhasepiteelisolujen pinnalliset mikrovillukset sen sijaan puuttuvat. Niiden sytoplasman täyttävät runsaat lipidipalloset, joita solut muodostavat hyvin kehittyneessä Golgin laitteessaan erittäen limaa sappikäytävän onteloon. Erite näkyy hienojyväisenä tai tasa-aineisena seinämiä reunustavana aineena. Poikkeuksellisenä havaintona voidaan pitää keräsolujen kerästen tai lähes kokonaisten keräsolujen esiintymistä sappikäytävänonteloissa mahdollisesti osoituksena solujen vaelluskyvystä. Vaikka epiteelisolut muodostavat lujia solukalvosidoksia toisiinsa, keräsolut eivät kiinnittyneet muihin soluihin. Ilmeistä on, että pinnallisten epiteelisolujen toiminta on pääasiassa absorptiivista, mutta rauhasepiteelisolut ovat erikoistuneet eritystoimintaan.

**Rangifer, 10 (1): 17–23**

**Timo Rahko och Sven Nikander 1990.** En elektronmikroskopisk studie av gallgången hos ren.

*Sammandrag:* Gallgångarnas yt- och körtelepitel bestod av höga epitelceller beklädda med ett regelbundet villuskiikt. Intercellulära desmosomer sågs i epitelcellernas parietala cytoplasmamembraner. De intraepiteliala globulära leukocyterna saknade desmosomer eller andra bindningar med närliggande celler. I de superficiala epitelcellernas apikala cytoplasma fanns elektrontäta små kroppar antagligen bestående av mucin. Bågar- och de djupare belägna körtelecellerna innehöll rikligt med stora mucin granulor med ett mindre elektrontätt matrix. Det föreföll som om dessa celler skulle vara sekreterande och de superficiala epitelcellerna absorberande. Kärnen och andra cytoplasmatiska organeller hade egenskaper jämförbara med epitelceller i allmänhet. Gallgången var oftast tom men ibland sågs ett finkornigt amorft material i den. Som en anmärkningsvärd observation ansees förekomsten av delar och t.o.m. hela globulära leukocyter i gallgången.

**Rangifer, 10 (1): 17–23**

## Introduction

Our understanding of the function of different organs is based on a thorough knowledge of structural features. This also applies to the liver and bile ducts. The relative size and structure of the liver varies among different animal species. Some species such as the rat and the reindeer do not possess a gall bladder.

The ultrastructure of the common bile duct differs between rats and mice (Yamada, 1969, 1970, Luciano, 1972). These differences were attributed to the presence or absence of a gall bladder. To clarify if the reindeer shows some special ultrastructure in the wall of the common bile duct, this was in this study examined by electron microscopy.

## Materials and methods

The same tissue blocks, from six healthy reindeer, were used in this study. The ultrastructure of granulated cells of bile ducts in reindeer have been described elsewhere (Nikander & Rahko, 1990).

## Results

The general architecture of the mucosal layer of the common bile duct showed a regular epithelium supported by proprial tissues. The epithelium was of two types; the surface epithelial cells and glands with goblet cells (Figures 1 to 6).

Superficial epithelial cells were characterized by abundant and regular villous projections (Fig. 3). Their secretory granules appeared small and electron dense. The glands, on the other hand, did not carry regular microvilli and ap-

peared to be secretory in nature (Figs. 1 and 2). Their cytoplasm showed an abundance of large electron opaque granules and globules of obviously mucinaceous composition. Golgi complexes appeared prominent in the glandular cells without profound mucin contents (Fig. 1).

The mucosal layer was surrounded by connective tissue elements containing blood capillaries and different types of migratory cells such as mast cells.

Ultrastructural features of cytoplasmic organelles in the superficial epithelial cells were fairly regular (Figs. 3 to 5). The cells were tall columnar with regular oval or elongated basal nuclei showing mainly marginal chromatin and a smooth nuclear envelope. Nucleoli were not distinct. The cytoplasm of the cells showed typical structures of epithelial cells such as prominent Golgi complex usually in the supranuclear area, numerous small mitochondriae, short prothyls of granular endoplasmic reticulum and free ribosomes. No glycogen particles were identified in the cytoplasmic matrix, which was rich in microfilaments (Figs. 3 and 4). The general contrast of the cytoplasmic matrix varied between different cells. Those with great contrast and dark cytoplasmic matrix contained an abundance of microfilaments (Fig. 5). In basal areas of apical villous projections the microfilaments were concentrated in small bundles supporting the microvilli. Also in areas adjoining the intercellular connections the microfilaments were abundant (Figs. 3 and 4).

The intercellular spaces between the cells were empty. The cellular membranes formed villous projections also into the intercellular spaces (Figs. 4 and 5) but the basal cell membrane was smooth, resting on the basal lamina (Fig. 2).

The lumen of the bile ducts was either empty or contained fine granular or amorphous substances particularly in areas adjoining the walls (Figs. 7 and 8). Additionally, parts of globule leucocytes with several intact globules and mitochondria were seen to occur freely in the lumen, indicating a possible migratory capacity of the cells into the lumen (Figs. 7 and 8).

Certain areas of the walls contained epithelial structures interpreted as pancreatic acini. These epithelial cells contained numerous granules similar to the zymogen granules in the exocrine pancreas and an abundance of granular endoplasmic membranes.

The globules in the globule leucocytes varied in structure and size, the largest being even 6–7 microns in diameter. The globules were surrounded by smooth double membranes. They mostly showed diffusely electron dense, but some contained fine granular or dissolving matrices (Fig. 6). The cells were migratory in nature and though they were closely apposed to the epithelial cells, intercellular connections were never observed.

## Discussion

The bile duct epithelium of the reindeer is composed of two types of cells; absorptive superficial and secretory glandular cells. The presence of microvilli on the luminal surface is thought to indicate the absorptive function of the superficial cells. Obviously, the mucin granules and globules and mucin products of ordinary goblet cells in the deep glands are secreted into the ducts. Similar evidence for absorptive and secretory function has previously been presented in the rat (Yamada, 1969, 1970), the mouse (Yamada, 1969, 1970, Rahko, 1971) and cattle (Rahko, 1973). Also the rat displayed intervillous depressions in superficial epithelium as signs for pinocytosis (Yamada, 1969, 1970).

In a previous histological and histochemical study the authors showed the presence of mucin on the surface epithelium (Rahko & Nikander, 1990 a, b). It appears that the surfactant, which was histochemically positive by stainings for neutral and weakly acidic mucins, is seen in the present material by electron microscopy as fine granular or amorphous material on the luminal surface of the *lamina epithelialis*.

Superficial epithelial cells appeared as either light or dark in electron micrographs. Also the

mouse and the rat show differences in cytoplasmic contrast of the cells (Yamada, 1969, 1970). Yamada proved that in small rodents the darkness of cytoplasm in certain cells is produced by an abundance of microfilaments in the cells. An especially large amount of microfilaments has been described in the so-called brush cells of the bile duct walls of the mouse and rat (Luciano & Reale, 1969, Luciano, 1972). According to the present findings in the reindeer, obviously not only the richness of microfilaments but also profuse amounts of free and attached ribosomes contribute to the electron density of the dark epithelial cells. It remains uncertain whether the differences in the cellular ultrastructure indicate different functional status.

Globule leucocytes and mast cells are considered to be migratory cells (Gregory, 1979, Morales & al., 1980, Toledo & Morales, 1981, Morales, 1983). In the present material globule leucocytes never carried desmosomes on their cytomembranes. The cells were usually located in basal areas of the epithelium. However, in the present study it was shown that luminal contents of the common bile duct may show intact globules of globule leucocytes and even cytoplasmic areas with globules and mitochondria (see Figs. 7 and 8). The presence of parts of globule leucocytes in the lumen of the ducts may be an indication of the migratory capacity of the cells. However, in this kind of study it is impossible to determine if the presence of such cells in the lumen is only an artificial phenomenon following a possible mechanical rupture of the *lamina epithelialis* during the process of slaughter and sampling of the tissue specimens. Freely migrating globule leucocytes have not previously been described by electron microscopy in luminal sites of other mucosal tissues.

In this study, the globules of the globule leucocytes showed different size and ultrastructural appearances. Also some giant globules even 6 to 7 microns in diameter were observed. The inner matrix of the large globules was faintly electron-dense and fine-granular as also Baert (1989) has described in the rat.

The authors have tried to study by immunohistochemical methods the intracytoplasmic antigenicity of mast cells and globule leucocytes of the reindeer but the commercial antisera so far applied have failed to show positive immunoreactivity (Nikander and Rahko, unpublished data).

## References

- Baert, J.** 1989. Ultrastructure of globule leucocytes isolated from rat tracheal epithelium. - *J. Submicr. Cytol. Pathol.* 21: 765-769.
- Gregory, M. W.** 1979. The globule leucocyte and parasitic infection - a brief history - *The Vet. Bull.* 49: 821-827.
- Luciano, L.** 1972. Die Feinstruktur der Gallenblase und der Gallengänge II. Das Epithel der extrahepatischen Gallengänge der Maus und der Ratte. - *Z. Zellforsch.* 135: 103-114.
- Luciano, L. & Reale, E.** 1969. A new cell type («brush cell») in the gall bladder epithelium of the mouse. - *J. Submicr. Cytol.* 1: 43-52.
- Morales, C. R.** 1983. Argentaffin and migrating mast cells in the bovine gall bladder epithelium. - *Anat. Anz. Jena.* 154: 419-423.
- Morales, C. R., Pereyra, L. A., Toledo, O. M. S. & Montes, G. S.** 1980. Histochemical and morphological characterization of migrating mast cells in the bovine gall bladder epithelium. - *Histochemistry* 68: 159-168.
- Nikander, S. & Rahko, T.** 1990. Ultrastructure of granulated cells in the bile duct of reindeer. - *Rangifer Special Issue No. 3* (in press).
- Rahko, T.** 1971. Studies of the Pathology of Bovine and Murine Liver Infected with *Fasciola Hepatica* with Reference to the Mast Cell and Globule Leucocyte. Thesis. - *Ann. Acad. Sci. Fenn. Ser. A. V. Medica N:o* 148.
- Rahko, T.** 1973. On the ultrastructure of epithelial cells in bile ducts of cattle chronically infected with *Fasciola hepatica*. - *Acta Vet. Scand.* 14: 233-244.
- Rahko, T. & Nikander, S.** 1990a. Macroscopical and microscopical studies of the common bile duct in the reindeer. - *Rangifer* 10(1): 3-8 (this issue).
- Rahko, T. & Nikander, S.** 1990b. Histochemical studies of the common bile duct in the reindeer. - *Rangifer* 10(1): 9-15 (this issue).
- Toledo, O. M. S., Morales, C. R., Pereyra, L. A., Jorado, T. & Montes, G. S.** 1981. Migrating mast cells in the gall bladder epithelium of cattle and sheep. - *Histochemistry* 72: 433-442.
- Yamada, K.** 1969. Fine structure of rodent common bile duct epithelium. - *J. Anat.* 105: 511-523.
- Yamada, K.** 1970. The glands of the common bile duct in the rat and mouse. An electron microscope study. - *Acta Anat.* 77: 438-453.

Manuscript delivered 26 March, 1990.

Fig. 1. Electron micrograph, showing glandular epithelial cells devoid of regular microvilli. The Golgi (G) lamellae and vesicles of the cells are prominent. A goblet cell filled with mucin globules is marked with an arrow. (GER = granular endoplasmic reticulum). x 4.600.

Fig. 2. General appearance of glandular epithelial cells containing numerous mucinaceous granules in the cytoplasm (BM = basal membrane). x 4.600.

Figures 3-5, see page 22

Fig. 3. and 4 are electron micrographs of superficial epithelial cells carrying microvilli on the luminal surface and showing projections of cytoplasmic membranes into the intercellular spaces (arrows). Note the abundance of microfilaments and dense desmosomes (D). Fig. 3 x 60.000, Fig. 4 x 30.000.

Fig. 5. The cytoplasmic matrix of superficial epithelial cells is either light (on the left) or dark (on the right). Both contain small rounded granules (e.g. arrows) in apical cytoplasmic areas but the electron density of the dark cell is mainly produced by more abundant microfilamentous structures. The dark cell also contains numerous free or attached ribosoms. (D = desmosomes). x 18.400.

Figures 6-7, see page 23

Fig. 6. Two globule leucocytes (GL) showing either regular globules of usual size and structure or some giant globules with fine-granular matrices (arrows). x 4.600.

Fig. 7. and 8 are electron micrographs of the lumen of the bile duct, showing cytoplasmic structures of globule leucocytes (GL) surrounded by different luminal secretions of fine-granular composition. Fig. 7 x 15.000, Fig. 8 x 9.000.







