

CHARACTERISTICS OF LAMELLAR BODIES IN PERITONEUM

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Summary. *We examined 20 healthy persons (kidney donors). Small samples of parietal peritoneum were taken during the surgical operation for kidney extirpation. To avoid artificial damage owing to the extreme fragile of peritoneal tissue, the biopsies were treated with special care. The samples were prepared in the standard way for study by transmission electron microscopy. In order to preserve the fine structure of lamellar bodies, we used a fixative mixture including glutaraldehyde and tannic acid.*

The aim of our study was to investigate the localization and ultrastructure of lamellar bodies of peritoneum in order to contribute to finding out their physiological function and importance for this tissue.

Special round or oval multilaminar structures – lamellar bodies could be recognized among microvilli of peritoneal mesothelial cells apical plasma membrane. Lamellae were organized as concentric bilaminar structures – one of the layers was osmiophilic (dark, electron-dense), the next one was electron-lucent. The lamellar bodies were noticed in mesothelial cells cytoplasm, and were released from the cells by process of exocytosis. For some time after exocytosis, lamellar bodies with regular structure were recognized among the microvilli. Afterwards, probably because of the process of swelling, lamellar bodies had been losing the regular geometrical structure and were recognized on our photomicrographs with only a few lamellae and a free central part. Lamellar bodies could also be seen between two adjacent mesothelial cells, above and in the cellular junctions, possibly contributing to the intercellular sealing and prevention of paracellular transport. We recognized lamellar bodies in lamina propria just below the mesothelial cells, as well as in endothelial cells of peritoneal blood vessels.

Key words: *Mesothelium, endothelium, peritoneum, ultrastructure, lamellar bodies*

Introduction

Lamellar bodies are laminate structures composed of concentric phospholipid bilayers (1,2,3), resembling the onion section. They are investigated in alveolar epithelial cells (4,5), pleura, pericardium and synovia (6,7). It is supposed that these structures are synthesized in Golgi dictiosomes, at first usually in a form of multivesicular bodies (1,4). The main recognized role of lamellar bodies is to provide sliding of adjacent surfaces either in alveoli or of mesothelium covered cavities (pleural, pericardial and peritoneal). This is made possible in two different ways: mechanically, as ball-and-roller bearings while they are on the surface, and as a source of serous fluid, which is entrapped between the lamellae and is released after lamellar bodies destruction (1,3,4).

Aim

The aim of our study was to investigate the localization and ultrastructure of lamellar bodies in healthy people's peritoneum, in order to contribute to finding

out their physiological function and their importance for this tissue.

Subjects and Methods

We examined patient group consisted of 20 healthy kidney donors. Small samples of peritoneal tissue were taken during the surgical operation for kidney extirpation. To avoid artificial damage owing to the extreme fragility of peritoneal tissue, the biopsies were treated with special care. The biopsy was taken immediately after the peritoneum was opened and was gently placed into a fixative prepared in advance (8). A temperature of 4°C was maintained for the entire fixation time. The preparation procedure was standard for routine transmission electron microscopy (TEM) studies of semi-fine and fine sections. Tissue samples were fixed in 4% glutaraldehyde and 1% tannic acid in 0.1 mol/L cacodylate buffer pH 7.4, postfixed in 1% osmium tetroxide in the same buffer, and, after dehydration in alcohols and propylene oxide (9), embedded in Epon resin. The sections were made on ultramicrotome LKB IV. Semi-fine sections were stained with toluidine blue (TB), and fine

sections were contrasted with uranyl acetate and lead citrate. The sections were analyzed on TEM Philips 400.

Results

The peritoneal mesothelium apical plasma membrane appearance in healthy persons varied from smooth surface to surface with great number of microvilli (Fig. 1). The single cilia with yet unknown importance could be noticed. Special round or oval multilaminar structures – lamellar bodies could be recognized among mi-

crovilli (Fig. 1B,C). Lamellae were organized as concentric bilaminar structures – one of the layers was osmiophilic (dark, electron-dense), the next one was electron-lucent.

The ultrastructure of peritoneal mesothelial cells showed euchromatic nuclei, well-developed rough endoplasmic reticulum, Golgi dictiosomes, and a great number of mitochondria, pinocytotic vesicles, lipid inclusions and multivesicular bodies (Fig.1). The lamellar bodies were also noticed in mesothelial cells cytoplasm,

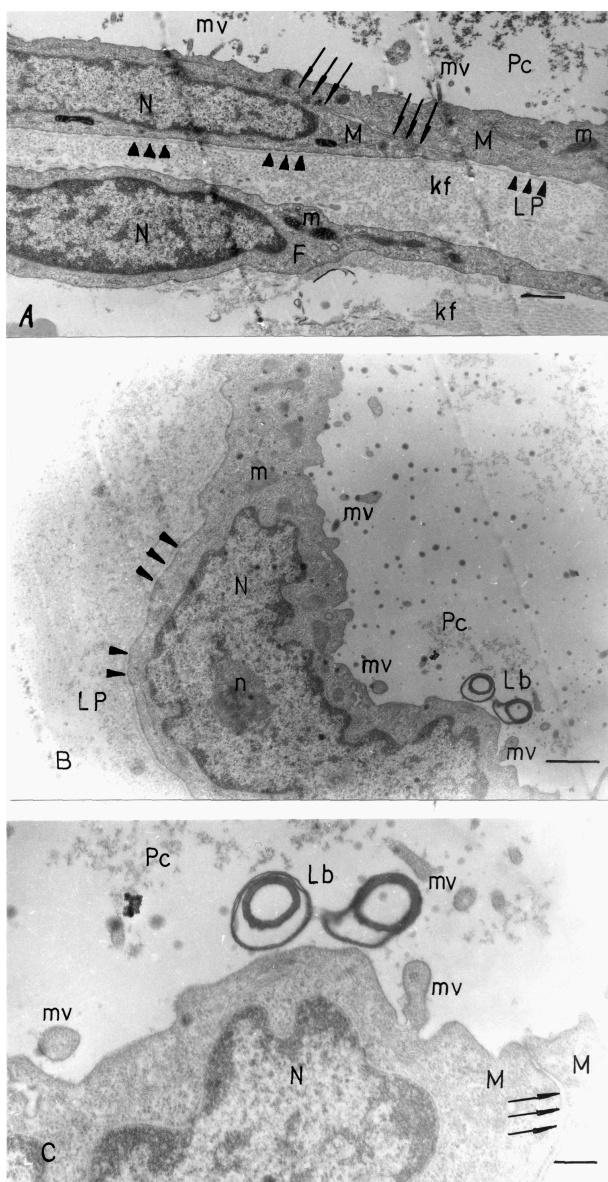


Fig. 1. Peritoneal mesothelium of healthy person
 A) Mesothelial cells and fibroblast, TEM (bar - 1µm)
 B) Mesothelial cell lamellar bodies, TEM (bar - 1µm)
 C) Detail from B, TEM (bar - 300nm)
 Pc - peritoneal cavity, M – mesothelium, LP – lamina propria, N – nucleus, n – nucleolus, mv - microvilli, arrowheads – basement lamina, m – mitochondria, Lb – lamellar body, kf – collagen fibers, arrows - junction of two adjacent mesothelial cells, F – fibroblast.

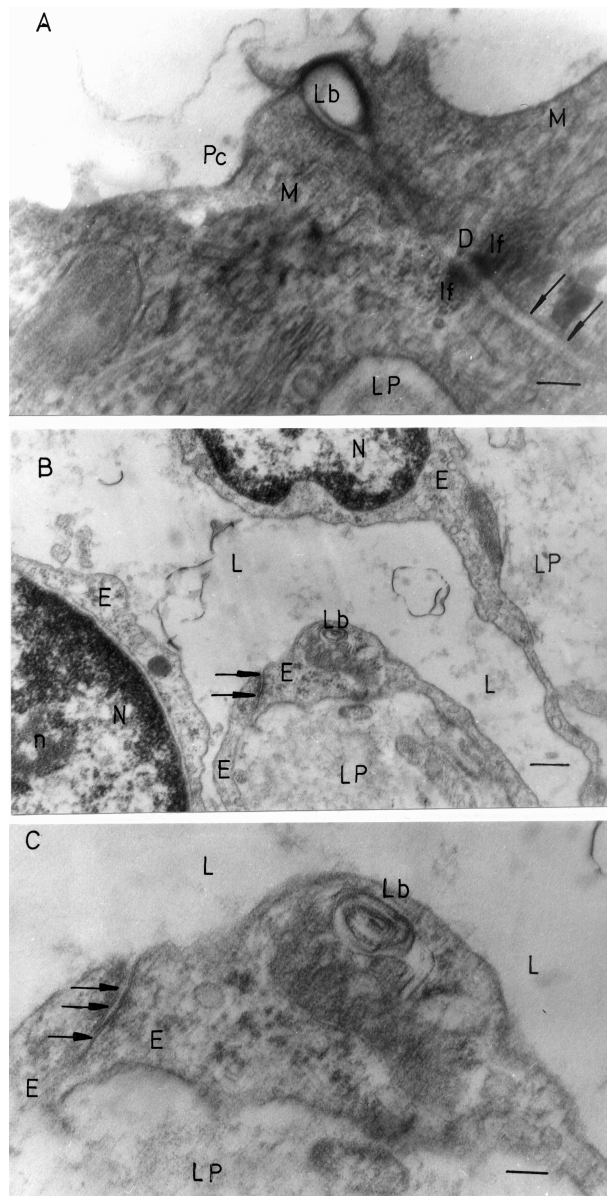


Fig. 2. Peritoneum of healthy person
 A) Peritoneal mesothelial cells, TEM (bar 100nm)
 B) Blood vessel in lamina propria, TEM (bar - 300nm)
 C) Detail from B, TEM (bar - 100nm)
 Pc – peritoneal cavity, M – mesothelium, BL – basement lamina, D – desmosom, If – intermedial filament, E - endothelial cell, N – nucleus, n - nucleolus, arrows - junction of two adjacent endothelial cells, Lb – lamellar body, LP – lamina propria, L – lumen of blood vessel.

and were released from the cells by process of exocytosis, as could be recognized on biopsies of our patients. For some time after exocytosis, lamellar bodies with regular structure were recognized among the microvilli. Afterwards, probably because of the process of swelling, lamellar bodies had been losing regular geometrical structure and we recognized them on our photomicrographs with only a few lamellae and a free central part, (Fig. 1B,C).

Lamellar bodies could also be seen between two adjacent mesothelial cells, above (Fig. 2A) and in the cellular junctions, possibly contributing to the intercellular sealing and prevention of paracellular transport.

We recognized lamellar bodies in lamina propria just below the mesothelial cell, as well as in endothelial cells of peritoneal blood vessels (Fig. 2B,C).

Discussion

Lamellar bodies are synthesized in peritoneal mesothelial cells (1,5), especially when fixated in tannic acid.

It is documented that lamellar bodies are structures of 1.5µm in diameter. Lamellae are organized as concentric bilaminar structures – one of the layers is osmiophilic (dark, electron-dense), the next one is electron-lucent (4,10). The lamellae are phospholipid in their nature, noticed in all mesothelial cells (1). The structure of these lamellae was also compared to the structure of lamellae in lung alveoli epithelial cells. Lamellar bodies are also noticed in pleura and pericardium too.

There are many speculations on lamellar bodies synthesis. Some of them are based on investigations of pneumocytes type II (1,4). It is shown that in these cells lamellar bodies originate from multivesicular bodies. Multivesicular bodies are often found in peritoneal mesothelial cells in our patients. In pneumocytes type II, the maturing process of multivesicular bodies towards lamellar bodies takes place in rough endoplasmic reticulum and in Golgi apparatus. Since the ultrastructure of these cells is very similar to peritoneal mesothelial cells including euchromatic nucleus, marked nucleoli, well developed rough endoplasmic reticulum, Golgi apparatus, a great number of mitochondria, pinocytotic vesicles, lipid inclusions and multivesicular bodies (3,

11) it could be supposed that lamellar bodies of peritoneal mesothelial cells have a similar way of maturation and similar function like a pneumocytes type II.

Lamellar bodies could be seen in the cytoplasm of peritoneal mesothelial cells (1,10) and are released from the cells by the process of exocytosis, as could be recognized on biopsies of our patients. For some time after exocytosis, lamellar bodies with regular structure are recognized on the mesothelial cells surfaces, and after that the process of lamellar bodies swelling is very intensive and leads to the loss of regular geometrical structure and, finally, to complete destruction. Besides the lamellar bodies completely filled with concentric lamellae, we also noticed on our photomicrographs lamellar bodies with only several lamellae and a free central part of the structure. The complete destruction of lamellar bodies is the last phase in which the content of these structures is released over the cellular surface.

It is supposed that lamellar bodies, noticed between two adjacent mesothelial cells, above and in the cellular junctions (1), contribute to the intercellular sealing and prevent paracellular transport.

Our finding of lamellar bodies in lamina propria and endothelial cells of peritoneal blood vessels agrees with other authors findings (1). Their hypothesis is that endothelial cells, as well as macrophages and fibroblasts synthesize these structures. Our speculation on mesothelial cells exocytosis of lamellar bodies through the basal part of the cell is not in agreement with the data from literature that basement lamina is the unpermeable barrier for these structures (1).

The main role of lamellar bodies in human peritoneum is reduction of friction between visceral and parietal layers (1). This is made possible in two different ways: mechanically, as ball-and-roller bearings while they are between the microvilli, and as a source of serous fluid which is entrapped between the lamellae (it is released after lamellar bodies destruction) (1, 4). The purpose of lamellar bodies synthesis by cells in peritoneal lamina propria remains unknown. The fate of those lamellar bodies is also unknown. In further investigations, it would be important to find out the possible relationship between the lamellar bodies of mesothelial cells origin and lamellar bodies of cells in lamina propria origin.

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KARAKTERISTIKE LAMELARNIH TELA U PERITONEUMU

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Kratak sadržaj: *Biopsije peritoneuma uzete su od 20 zdravih dobrovoljaca, donora bubrega za transplantaciju. Isečci tkiva dobijani su tokom hirurškog zahvata uzimanja bubrega. Uzorci su standardno pripremljeni za transmisionu elektronsku mikroskopiju (TEM). Da bi se sačuvala struktura lamelarnih tela, koristili smo mešavinu fiksativa glutar aldehida i taninske kiseline.*

Cilj ovog istraživanja bio je da se detaljno ispita morfologija i lokalizacija lamelarnih tela peritoneuma zdravih osoba da bi se doprinelo utvrđivanju njihove uloge.

Multilaminarne strukture, lamelarna tela, okruglog ili ovalnog oblika, zapažala su se između mikrovila apikalne membrane mezotelnih ćelija peritoneuma. Lamelle su bile organizovane kao koncentrične bilaminarne strukture – jedna lamela bila je tamna (veće elektronske gustine), dok je susedna bila svetla (male elektronske gustine). Lamelarna tela su nadjena i u citoplazmi mezotelnih ćelija, iz kojih su sekretovana egzocitozom. Neko vreme nakon egzocitoze, između mikrovila su zapažena lamelarna tela regularne strukture, a zatim, verovatno kao posledica procesa bubrenja, lamelarna tela gube pravilnu geometrijsku strukturu i mi ih, na fotomikrografijama, uočavamo sa samo nekoliko lamela na periferiji i praznim centralnim delom. Lamelarna tela mogu takodje biti zapažena između dve susedne mezotelne ćelije, iznad ili u okviru međućelijske veze, sa mogućom ulogom sprečavanja paracelularnog transporta. Lamelarna tela takodje su nadjena u vezivnom tkivu kao i u endotelnim ćelijama krvnih sudova lamine proprije peritoneuma.

Ključne reči: *Mezotel, endotel, peritoneum, ultrastruktura, lamelarna tela*