Light and Electron Microscopic, and Immunohistochemical Analysis of Deposits in the Proximal Ends of Ventriculoperitoneal Shunt Catheters

Ventrikülo-Peritoneal Şantların Proksimal Kataterlerinde Toplanan Depozitlerin Işık, Elektron Mikroskopik ve İmmunohistokimyasal Olarak İncelenmesi

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Abstract: The ventriculoperitoneal shunt is accepted as the simplest and most effective treatment for hydrocephalus. The major problem with these devices is obstruction of the proximal end of the tubing. In this study, we examined deposits in the proximal ends of Codman shunts from 20 of our patients whose catheter failed. Light microscopic examination revealed that the deposited material contained fibrin, other proteinaceous material, erythrocytes, lymphocytes, macrophages, eosinophils, leukocytes, giant cells, and evidence of neovascularization. Electron microscopy confirmed the presence of fibrin, other proteinaceous material, many phagocytic cells, giant cells, and bacteria within the cytoplasm of these two types of cells. Immunohistochemically, the deposited material stained intensely for IgA, moderately for IgG, and weakly for IgM. This report discusses on ventriculoperitoneal shunt failure and the types of deposits that accumulate at the proximal end of the catheter, and reviews the relevant literature. The authors conclude that such deposits do cause shunt failure.

Key Words: Deposits, electron microscope, immunohistochemistry, light microscope, shunt failure


Anahtar kelimeler: Depozitler, ışık mikroskop, elektron mikroskop, immünohistokimya, şant malfonksiyonu
INTRODUCTION

Shunt surgery is considered the simplest and most effective way to treat hydrocephalus. The major problem with this therapy is obstruction of the proximal end of the shunt tubing, an outcome that is particularly common in pediatric patients. Various types of material have been known to block these shunts, including brain tissue, tumor cells, choroid plexus tissue, blood cells, infectious agents, and cerebrospinal fluid (CSF) debris (1,3,4,6,8,12-17). The purpose of this study was to characterize the components of deposits found in nonfunctional ventriculoperitoneal shunts using light and electron microscopy (EM), and immunohistochemical methods. We also sought to make further conclusions about the possible causes of this problem.

MATERIALS AND METHODS

We examined the proximal ends of the Codman shunts in 20 patients whose ventriculoperitoneal shunts had failed. The study group consisted of 6 females and 14 males who underwent shunt surgery between 1994 and 1997 in the Department of Neurosurgery at Cukurova University Medical School. A 5 cm length of the proximal end of each failed shunt was cut into two pieces. In each case, we fixed one of the pieces in 10% buffered neutral Formalin and processed it for histological and immunohistochemical examination. For histological evaluation, sections of silicone catheter and lumen deposits were stained with hematoxylin-eosin (H-E) and periodic acid-Schiff stain. For immunohistochemistry, we used the indirect streptavidin-biotin peroxidase (ABC) method, and paraffin sections were incubated overnight in primary antibody at 4°C. Rabbit anti-human immunoglobulins IgA, IgG, IgM (Immunon, USA) and rabbit anti-human lysozyme (Dako, Denmark) antibodies were applied. After the immunohistochemical reaction was completed, sections were stained with hematoxylin only. Photographs were taken using 21 DM/100 ASA film.

After cutting the original length of tubing, we immediately placed the second half of each piece in 5% glutaraldehyde buffered to pH 7.4 with Millonig's phosphate buffer (11) for 1 hour. We then made a cut along the length of the silicone tubing and carefully removed the accumulated material. Next, the deposits were fixed again in the same solution for 2 hours, and then postfixed in 1% osmic acid for 2 hours. Finally, the samples were dehydrated in graded ethanol baths, embedded in araldite, and then processed for transmission EM using conventional methods.

RESULTS

Macroscopically, some specimens of silicone tubing were filled with spongy tissue, and others were packed with gray-brown material. Light microscopic examination of the shunt deposits revealed fibrin, other proteinaceous material, erythrocytes, leukocytes, some glial cells, and evidence of neovascularization (Figure 1). All shunt material stained intensely for IgA and lysozyme, but less strongly for IgG and IgM (Figures 2,3,4). EM examination of the spongy material revealed fibroblasts, collagen fibers, and polymorphonuclear leukocytes (Figure 5). Some of the spongy specimens consisted of fibroblasts, collagen fibers, and proteinaceous material (Figure 6). In some cases, we also found an abundance of phagocytic cells in the deposit material, indicating the presence of an inflammatory reaction. Examination of the ultrastructure of the gray-brown material indicated that this was infected tissue, containing abundant granulocytes, mononuclear cells, fibrin, other proteinaceous material, erythrocytes, and scattered giant cells (Figure 7). Many specimens exhibited scattered bacteria and groups of bacterial organisms within the cytoplasm of the neutrophils and macrophages that were present (Figure 8). In addition, we also observed free intact bacteria, a few erythrocytes and platelets, fibrin, other proteinaceous material, and free cytoplasmic debris in the specimens.

DISCUSSION

Accumulation of intraluminal debris is an important factor in shunt obstruction. Shunt material, the nature of the CSF, and length of time the shunt has been in place all contribute to the deposition of material in the tubing. Denatured protein, in addition to lipid and mineral deposits, damage the inner surface of the catheter through mechanical and immunological processes (3,7,8,14,15,17). Infection also increases the risk of shunt obstruction (9,10,15).

Fibrin, other proteinaceous material, red blood cells, lymphocytes, neutrophils, macrophages, eosinophils, giant cells, platelets, immunoglobulins, lipid, inorganic film, mucin, minerals, and
neovascularization within the sediment are all known to accumulate in ventriculoperitoneal shunts. Deposition of these materials on the tubing wall creates an irregular surface that encourages bacterial adhesion and colonization. Also, various built-up minerals, oligosaccharides, and proteins supply energy to such bacteria (4,5,8,14,15). Cultures of the material from the ends of our patients’ failed shunts were negative, thus, based on these results, none of our cases were infected. Still, EM examination revealed fibrous tissue composed of fibroblasts, collagen fibers, granulocytes, phagosomes within phagocytes, and macrophages. As mentioned above, some samples contained abundant phagocytic cells. Mononuclear cells and giant cells were noted in the gray-brown material that predominated in some specimens. It should noted that some of the components of this debris are found in normal CSF, and in normal, uninfected tissue. The observation of lymphocytes, eosinophils, macrophages, and giant cells can be considered to reflect bacterial colonization or, at minimum, a hypersensitive reaction to silicone (8,15,21).

Gower et al. stated that infection is the most significant problem associated with shunt failure. In accord with our findings, they detected macrophages, platelets, and lymphocytes on their EM examination of the proximal ends of ventriculoperitoneal shunts (7,8). Studies done on silicone-based prostheses, shunts, and contact lenses have demonstrated the effects that tears, cosmetics, mechanical stress, and environmental in constant factors have on this material. Contact lenses, in particular, which are contact with tears, become completely coated with a thick layer of mucoprotein,
Figure 5: The ultrastructural appearance of the spongy tissue found in some of the shunts. We observed fibroblast (F) and collagen fibers (+), as well as large intercellular spaces (+) and the presence of polymorphonuclear leukocytes (arrow). (X6,000)

Figure 6: The spongy tissue contained fibroblasts (F), polymorphonuclear leukocytes (L), and proteinaceous material (arrows). The nucleus (n) and the intercellular spaces (+) are also identified in this image. (X8,800)

Figure 7: The gray-brown shunt material contained granulocytes (arrows) and erythrocytes (arrowheads). Fibrinous material (+) is also identified in this photo. (X8800)

Figure 8: This image of granulomatous tissue shows a neutrophil (NL) that has phagocytosed many bacteria (arrows). Fibrinous material (FM), cellular debris (+), and the cell's nucleus (n) are indicated. (X14,000)

We suggest that most of factors, except for the CSF environment, affect ventriculoperitoneal catheters in similar ways. Gower et al. (8) observed lipid, and calcium deposits within 8 hours of wear, and are easily penetrated by Pseudomonas aeruginosa (2,18,19,20).
lymphocytes, platelets, and especially macrophages due to Subarachnoid Hemorage in silicone-based catheters, and Skholl et al. (16) detected bits of suture debris and giant cells containing foreign bodies in occluded ventriculoperitoneal shunt devices. In our study, immunohistochemical studies of the materials in the end of the tubing identified immunoglobulins. We found that lysozyme, macrophages, and IgA were present in all specimens, and detected IgM in six cases.

As Gower et al. (8) suggested, the elimination of bacteria from the deposits, or the induction of delayed hypersensitivity by contact with silicone and plastic, stimulates the body's immunologic mechanisms. This leads to the accumulation of macrophages, giant cells, and antibodies, all factors that can damage silicone. An examination of patients with infected shunts by Waytt and coworkers (21) revealed nephritis, which had resulted from activation of the complement system by antigen-antibody complexes and deposits. We did not observe nephritis in any of our cases. The fact that we detected IgG, even in culture-negative cases, suggests that the immunologic reaction we observed in these patients is one of hypersensitivity. On this basis, we conclude that, in addition to infection-related causes, shunt malfunction is also linked to delayed hypersensitivity. In this scenario, deposits of various materials and antigen-antibody complexes accumulate on the tubing walls, damaging the surface, encouraging bacterial proliferation, and ultimately occluding the shunt lumen.

REFERENCES


Bağdaşılık: Ventrikuloperitoneal Shunts and Deposits