Postoperative fibrosis after surgical treatment of the porcine spinal cord: a comparison of dural substitutes

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Object. Postoperative adhesion- and fibrosis-induced spinal cord tethering is not uncommon and may be associated with delayed clinical sequelae. Multiple dural substitutes have been used in surgery without a full appreciation of the grafts’ adverse effects. The authors conducted a comparative animal experimental study to evaluate the degree of chronic inflammatory reactions, adhesions, and fibrosis caused by the use of four dural substitutes—Surgicel, Durasis, DuraGen, and Preclude.

Methods. Twenty-six pigs weighing 30 to 40 kg underwent a two-level lumbar laminectomy (a midline durotomy, implantation of a 2-cm dural substitute in the subarachnoid space, and watertight dural closure). After 8 weeks the animals were killed, and two independent neuropathologists blinded to the dural substitute group evaluated several sites along the implants, providing descriptions and quantitative scoring of fibrosis, chronic inflammatory reactions, foreign-body reactions, and spinal cord changes. Kruskal–Wallis one-way analysis of variance for ranks corrected for multiple comparisons was used to examine differences among the materials.

Conclusions. The DuraGen dural substitute produced the least amount of inflammation in the subarachnoid space and Preclude generated the most (p < 0.001). Surgicel and DuraGen were completely resorbed on histological sections, but both produced some inflammation, which diminished gradually from the dural implant center. Histological evaluation of the nonresorbed grafts demonstrated that Durasis caused the least degree of inflammatory reaction (p < 0.001). The Preclude dural substitute consistently demonstrated encapsulation and arachnoidal reaction. There was no evidence of implant-related adverse effects on the underlying pia mater and white matter regardless of the substitute type.

KEY WORDS • dural substitute • spinal cord tethering • inflammation • fibrosis • adhesion • pig

The use of a dural substitute to repair defects within the spinal dura mater or to enlarge the spinal dural sac is not an infrequent requirement in neurosurgery. Some of the more common spinal conditions include Chiari malformations, tethered cord, syringomyelia, and malignant intradural tumors. A postoperative inflammatory reaction followed by fibrosis involving the arachnoid membrane may lead to various complications. When dense adhesions between the spinal cord and the overlying dura mater occur, arachnoiditis, tethering, and neurological deterioration may ensue.

The search for the ideal dura mater substitute continues. Autologous sources include fascia lata and pericranium, and one of the most common is cadaveric dural allograft. Various synthetic or organic grafts have been used with inconsistent results.4,7,12,15,17,18 No single dural substitute has gained wide acceptance.

The ideal substitute produces a minimal inflammatory reaction, has excellent suture-related properties, comes in a variety of shapes and sizes, is inexpensive, and is associated with a low incidence of CSF leakage.

In a porcine model, we compared the adhesion formation, fibrosis, inflammatory response, foreign-body reactions, and spinal cord changes after placement of four commonly used dural substitutes—Surgicel, Durasis, DuraGen, and Preclude—in the lumbar subdural space.

Materials and Methods

Experimental Design and Surgical Procedure

This study was performed in accordance with guidelines from the
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Animal Care and Use Committee at the University of Miami School of Medicine. Twenty-six adult female pigs weighing 30 to 40 kg were used for the study. On arrival from the farm, the animals received 500,000 U/day of procaine penicillin for 3 days and were observed for any illness. No water or food was given the night before surgery. When general anesthesia was induced, each animal received 2.5 g of ceftriaxone, and this was continued for 5 days in twice-daily doses to prevent infection.

Implantation of Material

The upper lumbar region was shaved, prepared with betadine and chlorhexidine, and infiltrated with 1% lidocaine with 1:100,000 epinephrine. After incision of the lumbar fascia, the paravertebral muscles were stripped off the laminae and retracted laterally via a midline lumbar incision. A two-level upper-region lumbar laminectomy exposed the dura for a length of 4 to 6 cm. The porcine spinal cord ends at the lower lumbar levels. Complete hemostasis was achieved at this stage, and 4 to 5 cm of spinal cord was exposed by opening the dura at radially via a linear incision in the midline. A 2 × 1-cm piece of dural substitute material was cut and laid over the spinal cord after opening the arachnoid. At this stage absolute hemostasis was ensured before using a 6-0 Prolene suture to obtain a watertight dural closure. Porcine spinal cords—Surgicel, Durasis, DuraGen, and Preclude—were implanted subdurally. The paravertebral muscles were approximated, and the remaining wound was closed in three layers with a 0 Vicryl suture to the fascia and subcutaneous tissue and a 2-0 Ethilon suture to the skin. All surgical procedures were conducted using aseptic precautions and an operating microscope.

Postoperative Care and Functional Assessment

Before the animals were awakened from the procedure, 0.01 to 0.05 mg/kg buprenorphine was administered subcutaneously for pain control and continued 8 to 12 hours postoperatively. Immediate postoperative care included placement on a special rubber pad and continuous monitoring of heart rate, respiration, and temperature in the intensive care unit. Once the animal was fully awake, an initial neurological assessment was performed to rule out any deficit (hindlimb paresis, standing difficulties, and bowel/bladder incontinence). The next morning, the animals were transferred to a temperature-controlled room (28°C) that housed one pig per cage with a 12-hour light/dark cycle, for 7 to 10 days. The animals were monitored daily for neurological deficits, appetite, and fluid intake. Intravenous fluids were discontinued once the animal was drinking, and able to stand normally. Once the wound was healed, each animal was transferred to a larger cage with four others. The pigs were weighed weekly until they reached the end stage of the study.

Specimen Collection, Histological Preparation, and Evaluation

After 8 weeks, the animals were killed with a ketamine and barbiturate overdose. The vertebral canal was opened using an electric saw/Kerrison punch and sharp dissection, and taking great care, the spinal cord was removed and its covering was harvested. The specimen was placed in 10% buffered formalin for 2 to 3 days and then placed in 1% phosphate-buffered saline for a few days before we undertook standard sectioning and staining. Each specimen was cut into five blocks (Fig. 1), and every block was sliced into 8-μm serial sections at right angles to the dura mater–spinal cord interface and perpendicular to the anastomotic lines. Stained (H & E) tissues were prepared for histological evaluation. Two independent observers (a board-certified neuropathologist and a neurosurgeon with a doctorate in neuropathology) made all microscopic and histological evaluations in a blinded fashion.

Grading System

A quantitative grading system was used to score the tissue response to the implants (0, none; 1, minimal; 2, moderate; and 3, severe). Fibrosis, chronic inflammation, foreign-body reactions, and spinal cord changes were evaluated. A microscopic assessment for adhesions, encapsulation, and neomembrane formation was also undertaken for each specimen.

Statistical Analysis

The results of the grading from the histological examination were evaluated to determine differences between the dural substitute materials. Kruskal-Wallis one-way analysis of variance for ranks corrected for multiple comparisons was used to examine differences among the materials based on the scores of the tissue. A probability value less than 0.05 was considered statistically significant.

Results

A total of 26 pigs underwent implantation of dural substitute material. Six animals developed complications and were killed earlier than Week 8 and were thus excluded from the study. Of these six animals, one developed perioperative anesthesia-related complications, one died of pneumonia in the perioperative period, and three developed hindlimb paresis due to a deep wound infection on the 5th to 7th postoperative day. Each of these three animals was from a different implant group. One animal suffered an iatrogenic spinal cord injury and developed neurological dysfunction while it coughed and tried to jump off the table due to light anesthesia. None of the animals had a wound dehiscence, CSF leak, or pseudomeningocele.

Microscopic analysis of the sections revealed no adhesions between the implants and the pia of the spinal cord, nor were there any changes within the spinal cord itself (Table 1). Surgicel and DuraGen were completely resorbed, as the original matrix could no longer be visualized on tissue sectioning. In the case of DuraGen, no inflammatory cells were seen on any section. Comparisons of histological findings (fibrosis, inflammatory response, foreign-body reactions, and spinal cord changes) for each material are summarized in Table 2. Significant (p < 0.001) chronic inflammatory reactions, foreign-body reactions, and fibrosis were observed in our histological evaluation of Preclude-treated specimens compared with the other dural substitutes. In particular, these comparisons were highly significant at the upper, center, and lower sections of the implant (Fig. 1). Encapsulation of Preclude was a consistent finding in every section as a neomembrane formed around the material.

Discussion

The search for an ideal dural substitute in cases of tumor,
Spinal cord retethering after intradural spinal surgery is not an uncommon phenomenon, and its prevention is a technical challenge. Numerous techniques have been developed to reduce the incidence of retethering, based on preventing contact between the spinal cord and dural sac. These techniques are intended to allow the spinal cord to be circumferentially surrounded by CSF and thereby prevent adhesions and retethering. Numerous dural substitutes are being used in animal models and humans and their success rates are conflicting with regard to prevention of postoperative adhesions and fibrosis.1

We evaluated the degree of fibrosis, inflammation, and adhesions caused by Surgicel, Durasis, DuraGen, and Preclude in a porcine model after implanting the dural substitutes between the dura mater and spinal cord and closing the dura over the implant in a watertight fashion. Surgicel (oxidized regenerated cellulose) is widely used, it may exert pressure on neural tissues and result in paralysis and/or nerve damage. In our study, Surgicel was completely replaced with endogenous tissue with only a moderate chronic inflammatory response and arachnoid reaction (Fig. 2 upper left, Table 2). No adhesion was noted between the implant and pia mater of the spinal cord, but there was obvious dural thickening.

Durasis is made from porcine small intestinal submucosa. This dural substitute is produced by Cook Biotech, Inc. (West Lafayette, IN). In animal models, it has been shown to form occasional adhesions between the implant and the cerebral cortex. Authors of histological studies have shown increased implant site thickness, increased vascularity, no adverse immunological responses, and a distinctive remodeling of the Durasis including incorporation into the host. Investigators conducting initial clinical studies found no infection or CSF leakage. Durasis is currently undergoing a multicenter prospective trial as a cranial or spinal dural substitute in humans and is pending Food and Drug Administration approval. In our experience in the porcine model, we found no adhesions between the Durasis implant and the pia of the spinal cord, as well as no changes within the spinal cord itself. The dural substitute was visible in all the sections, and there was evidence of a mild chronic inflammatory response (Fig. 2 upper right, Table 2).

The third material used as an investigational implant was DuraGen (Integra LifeSciences, Plainsboro, NJ). The information brochure published by DuraGen is described as a collagen matrix that is fully resorbed within 8 to 12 months of implantation, followed by complete tissue closure of the dural defect. The material is soft, easy to cut to any size, and it molds instantly to the brain surface. Suturing is not required, but tensionless stay sutures can be used. There is no related reported encapsulation or neomembrane formation after implantation. DuraGen’s infection rate (6.1%) is comparable to other materials for dural closure (5.5%), and its incidence of CSF leakage is 1.46%. Investigators have shown minimal adhesion formation and a lack of any foreign-body reaction clinically. Our microscopic and histological findings demonstrated that DuraGen was completely resorbed within 8 weeks as the implant was not visible on any section. There was no evidence of fibrosis, chronic inflammatory reaction, foreign-
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Body reaction, or spinal cord change (Fig. 2 lower left, Table 2).

The fourth implant was Preclude (W. L. Gore & Associates, Newark, DE). In the manufacturer information, Preclude is described as a second-generation material composed of inert expanded polytetrafluoroethylene with a comparably inert elastomeric fluoropolymer in a three-layer construct. Its mean porosity is less than 1/10,000 μm, which gives it excellent conformability and handling and, thus, minimizes fibrous tissue ingrowths. Because it is a synthetic material, this dural substitute is nonabsorbable, nonpyrogenic, nonhemolytic, nonirritating, and disease free. Histological analysis, however, has revealed the formation of a thin encapsulating neomembrane, which is a two-cell-thick layer and mainly composed of fibroblasts. Clinical evaluation has shown a CSF leakage rate of 3.3%, an infection rate of 1.4%, and minimal scarring and tethering associated with the use of Preclude dural substitute.2,10,11,13,22–24 Our experience with Preclude confirmed that a multilayer neomembrane and graft encapsulation form after implantation. Moreover, there were significant chronic inflammatory reactions, foreign-body reactions, and fibrosis observed in the histological evaluation (Fig. 2 lower right, Table 2).

Conclusions

Of the four dural implants tested in a porcine model—Surgicel, Durasis, DuraGen, and Preclude—we observed varying degrees of inflammation and fibrosis without evidence of tethering. At 8 weeks postimplantation, the DuraGen dural substitute produced the least amount of inflammation and Preclude generated the most. Durasis demonstrated the least degree of inflammatory cell infiltration of the grafts that were not resorbed.

Disclosure

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References


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