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Antiadhesive effect of bioresorbable polylactide film in abraded middle ear mucosa

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ABSTRACT

Background and objective: Polylactide film (PLF) is used to prevent postoperative peridural adhesion in spinal surgery. Up until now, the antiadhesive effect of bioresorbable PLF in ear middle surgery has not been reported. The purpose of this study is to evaluate the antiadhesive effect of PLF in guinea pigs serving as a model for middle ear mucosal trauma.
Materials and methods: The animals were divided into two groups: the PLF group and the silastic sheeting group. There were seven guinea pigs (fourteen ears) in both groups. Under aseptic conditions, the middle ear mucosa was abraded using a pick inserted transbullaly. A PLF or silicone sheet was then placed into the guinea pigs' middle ear cavities. The auditory brainstem responses (ABRs) were assessed preoperatively and at three weeks postoperatively while the animals were under general anesthesia. A histopathological study was performed 3 weeks after the operation.
Results: The difference between the ABR results before the operation and three weeks postoperatively were not statistically significant. The adhesion formation did not appeared in either group. Prominent fibrous capsule formation and inflammation were observed in the silastic sheeting group, but not in the PLF group. Mild fibrous thickening of regenerated mucosa was observed in the PLF group.
Conclusion: From our results, bioresorbable PLF is nonototoxic and biocompatible with the guinea pig's middle ear cavity by short-term evaluation. Further long-term evaluation study is necessary before clinical application.

1. Introduction

To date, numerous factors affecting the surgical outcomes of tympanoplasty have been identified which include; the surgeon's experience, tympanic membrane (TM) graft materials, the causes of perforation and Eustachian tube function. [1–3]. Middle ear packing is often used to support the TM and ossicular prostheses, regeneration of middle ear mucosa, ventilation and hemostasis [4–7].

Gelfoam (Pharmacia & Upjohn, Kalamazoo, MI, USA) is well tolerated when a little abnormality is present in the middle ear cavity with intact mucosa [6], providing reasonable hemostasis [7] whether it is dry or wet [8]. Gelfoam can induce severe connective tissue hyperplasia, which results in adhesions and fibrosis tissue surrounding the TM and ossicular grafts. This subsequently leads to chain distortion and TM retraction, especially with denuded mucosa [7,9].

Silastic sheeting is used to restore a mucosally lined middle ear space and to prevent mucosal adhesions between the medial surface of the tympanic membrane and the promontory after surgical procedures involving the removal of middle ear mucosa [10–12]. The major drawback of this material it is nonresorbable and commonly needs to be surgically removed. Furthermore, the silicone component of silastic sheet may act as a potential nidus of infection, leading to subsequent graft loss and rejection [13].

Polylactide is widely used as a bioabsorbable implant materials in craniomaxillofacial and orthopedic surgery. Recently, Peltonen et al. reported that the biocompatibility characteristics of polylactide in the middle ear were good and not ototoxic [14]. Polylactide can be also formed into sheets for simple and safe surgical placement

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following cardiac surgery. Bioabsorbable polylactide films (PLF) provide the necessary barrier to prevent adhesion formation in the early postoperative period after chest or spine surgery and may act as a scaffold for new tissue growth [15]. To date, the antiadhesive effect of bioabsorbable PLF on abraded middle ear mucosa in an animal model has not been reported. The purpose of this study was to evaluate the antiadhesive effect of PLF in the abraded mucosa of a guinea pig's middle ear.

2. Materials and methods

The animals were divided into two groups: the silastic sheet group and the PLF group. There were seven guinea pigs in each group. All animal experiments followed a protocol approved by the Committee for Animal Experimentation at Chosunl University, Korea (CIACU). General anesthesia was administered by intraperitoneal injection of Zolletil (1:1 combination of tiletamine and zolazepam) and xylazine hydrochloride. Lidocaine (1%) containing 1/100,000 epinephrine was injected into soft tissue over the tympanic bulla prior to retroauricular incision. Under a surgical microscope (Leica M300; Ernst-Leitz-Strasse, Jena, Germany), the tympanic bulla was exposed and a tiny hole was drilled in it. Under aseptic conditions, the middle ear mucosa was abraded using a pick. The 3 mm × 3 mm × 0.05 mm bioresorbable PLF (copolymer 70:30 Poly(L-lactide-co-D,L-lactide, SurgiWrap®, MAST Biosurgery, Inc., CA, USA) covered the abraded mucosa in the experimental group and a silastic sheet (3 mm × 3 mm × 0.02 in., Bioplexus, Inc., CA, USA) was placed over the abraded mucosa in the control group. Care was taken to maintain the continuity of the ossicular chain and the integrity of the tympanic membrane in all animals. Otomicroscopic examination was performed every week.

Hearing sensitivities were measured using auditory brainstem response (ABR) thresholds. ABRs were assessed preoperatively and three weeks after surgery. Animals were placed in a soundproof booth and given an intraperitoneal injection Zolletil/xylazine to anesthetize them to keep them in the sound proof field. Auditory brainstem response (ABR) was recorded using an evoked potential system (Tucker-Davis Technologies, Alachua, FL, USA) and a Samsung computer. Stimuli were digitally synthesized using Siggen© software and presented through an ER-2 insert earphone (Etymotic Research, Elk Grove Village, IL, USA). Acoustic stimuli consisting of click (low frequencies less than 4 kHz) and 4, 8 and 16 kHz tone bursts were produced. Tone bursts consisted of a 3 ms envelope consisting of a 1 ms ramp onset, 1 ms plateau and 1 ms decay. ABR was recorded through Grass® stainless steel needle electrodes placed subcutaneously at the vertex (active), right cheek (inverting) and left cheek (common). The resulting signal was band-pass filtered (100–3000 Hz), amplified (10,000 × g) and digitized by a TDT Bioamp (Tucker Davis Technologies). Responses were collected and averaged at 30 presentations per second for up to 512 times. The stimulus was presented at 90 dB SPL and progressed downward in 10 dB steps until no response was identifiable. A separate model was used for each frequency. The ABR threshold shift at each frequency was compared among groups using one-way analysis of variance (ANOVA) and a 2-tailed t test was used to identify differences between the individual groups. A p-value <0.05 was defined as statistically significant.

After measurement of ABR, the animals were sacrificed and the temporal bones were harvested. After sitting in 4% paraformaldehyde solution for 24 h, the fixed specimens were placed in decalcifying solution (RapidCal™) for 2 days. Each specimen was then cut according to the standard histologic technique. The paraffin-embedded sections were then stained with hematoxylin and eosin and Masson–Trichrome. A light microscope was used to look for fibrous thickening and inflammation of the middle ear mucosa. The degree of these change was graded as none (grade 0, mild (grade 1), moderate (grade 2) or prominent (grade 3).

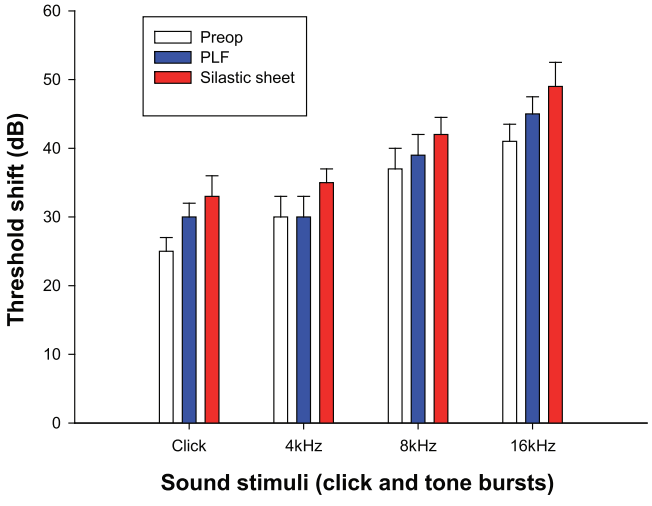

Fig. 1. Mean auditory brainstem response (ABR) thresholds before and three weeks after the insertion of the silastic sheet and PLF in the middle ear.

3. Results

All animals survived the duration of the experiment. None of these animals exhibited middle ear or inner ear infections. Otomicroscopically, there was no evidence of infection such as hyperemia or otorrhea. The differences in ABR results preoperatively and three weeks after surgery were not statistically significant different (Fig. 1). The systematic comparative histologic findings were summarized in Table 1. PLF group had a slight increase in the thickness of the mucous membrane but there was no new bone formation. Mild subepithelial thickening was observed at the wound site and compared between the PLF group and silastic sheet group. There was no subepithelial thickening in the silastic sheet group but a prominent thickened fibrous capsule formation over the silastic sheet was noted (Figs. 2 and 3). Neither adhesions nor inflammation of the healing mucosa was observed in the PLF group. Inflammation of the healing mucosa was observed six of 14 ears in the silastic sheet group (Fig. 4).

4. Discussion

In this study, we demonstrated that PLF did not induce adhesions in the middle ear cleft after abrasion of the mucosa. Along with the ABR results, this study indicates that PLF is biocompatible. Histologically, PLF did not induce fibrous capsule formation when compared to silastic sheet. The presence of adhesions after middle ear surgery is not uncommon. Excessive

Table 1
Histologic findings of silastic sheet and polylactide film (PLF).

Grade	Silastic sheet (n = 7)	PLF (n = 7)
Inflammation		
Grade 0	0	5
Grade 1	4	2
Grade 2	3	0
Grade 3	0	0
Fibrous thickening		
Grade 0	2	0
Grade 1	5	5
Grade 2	0	2
Grade 3	0	0