Introduction: Adhesions and perineural scarring represent major causes of failure after peripheral nerve surgery. For example, carpal tunnel release surgery has a 3% failure rate. Postoperative epineurial and intraneural fibrosis and adhesion are major factors contributing to symptom recurrence. A novel sodium carboxymethylcellulose (CMC)-derived hydrogel in which phosphatidylethanolamine (PE) was introduced into the carboxyl groups of CMC has been developed. CMC is a highly biocompatible polysaccharide that has been experimentally confirmed to decrease intraperitoneal adhesions. PE is a phospholipid (PL), a surfactant-like substance with lubricating properties. The present study investigated the effects of CMC-PE hydrogel in preventing postoperative adhesions and perineural scarring after surgery performed on the sciatic nerve of rats.

Materials and Methods: For CMC-PE preparation, PE was introduced into the carboxyl groups of CMC. CMC-PE powder was sterilized and dissolved in sterilized distilled water. To evaluate the anti-adhesive effects of CMC-PE hydrogels, 62 Lewis rats were used to make an adhesion model as previously described (1). Briefly, the sciatic nerve was dissected from surrounding tissues under operative microscopy. Both the epineurium and perineurium were carefully removed by cutting circumferentially and stripping distally for 15 mm with care to minimize damage to the axons. To stimulate local fibrotic response between the nerve and neural bed, the surface of the biceps femoris was repeatedly burned with a bipolar coagulator while the nerves were gently retracted and protected. The animals were randomly assigned to one of the following groups: CMC-PE(H) (n=27), high-viscosity (306 P) 1 wt% of CMC-PE hydrogel applied around the exposed nerve; CMC-PE(L) (n=27), low-viscosity (86 P) 0.5 wt% of CMC-PE hydrogel; HA (n=27), 1% of sodium hyaluronic acid viscous solution; control (n=27), no treatment; or normal (n=16), non-operated group. In the treatment groups, 0.5 mL of hydrogel or solution was applied around the nerve. Biomechanical analysis: Twenty-four rats were used for biomechanical analysis at 6 weeks after surgery. The nerves were subjected to biomechanical testing to assess ultimate breaking strength as previously described (1). Briefly, the sciatic nerve was exposed under anesthesia, then transected 5 mm proximal to the neurolysis site, taking care not to dissect adhesions around the nerve. The proximal stump of the nerve was mounted on a digital force gauge (Shimpo Co., Kyoto, Japan). The operated limb was firmly fixed to the table of the machine to limit motion to 6 degrees of freedom. The proximal end of the nerve was ligated using a 5-0 suture connected to the load cell. The nerve was subjected to traction at a rate of 2 cm/min until complete detachment of the nerve from the bed, and breaking strength of the nerves was recorded. Data are expressed as mean ± standard deviation. Multiple comparisons between each group were performed using Tukey’s test and values of P<0.05 were considered significant. For histological evaluation, 76 sciatic nerves were harvested from 38 rats, with 12 nerves of 6 rats killed each week in each group beginning at the 1-week time point and continuing to 6 weeks after surgery. All animals were killed by an overdose of sodium pentobarbital and operated nerves and surrounding soft tissues were carefully dissected and kept immersed in 4% paraformaldehyde overnight. Specimens were embedded in paraffin, and 5-µm-thick sections were cut and stained using Masson’s trichrome. All experimental protocols and animal maintenance procedures used in this study were approved by the Animal Ethics Research Committee at Nagoya University.

Results: Figure 1 shows the breaking strength of the perineural adhesion after 6 weeks. Breaking strength was significantly higher for the control group than for the CMC-PE(L), CMC-PE(H) or normal groups. Breaking strengths of the CMC-PE(L) and normal groups were significantly lower than that of the HA group. No significant difference in breaking strength was seen between CMC-PE(L) and normal groups.

Discussion: Our results demonstrate that CMC-PE hydrogels significantly reduced perineural adhesions in a rat model of sciatic nerve adhesion. Biomechanical analysis revealed that low-viscosity CMC-PE hydrogel was more effective than high-viscosity CMC-PE hydrogel or HA. Ikeda et al. reported that 1% HA solution could prevent peripheral nerve adhesion and scar formation in a rabbit neurolysis model, and also emphasized that nerves should be coated with HA throughout the operation to inhibit scarring in the nerve and surrounding tissue (2). In the present study, we applied 1% HA solution after neurolysis, which might have attenuated the anti-adhesive effects of HA. Viscosity of the hydrogel could influence anti-adhesion effects. Use of 3.5% HA is reportedly ineffective for preventing peripheral nerve adhesions compared with 1% HA. In our study, low-viscosity CMC-PE hydrogel was more effective than high-viscosity CMC-PE. A suitable range of viscosities in each material could exist for peripheral nerve surgery.

In conclusion, we have developed a novel CMC-PE hydrogel for preventing perineural adhesions. Low-viscosity CMC-PE hydrogel was superior to 1% HA without any delay in wound healing in a rat model of sciatic nerve adhesion.