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# Evaluation of Absorbable PLA Nasal Implants in an Ovine Model

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**ABSTRACT: Objective:** To examine biocompatibility and absorption profile of a poly (L-lactide-co-D,L-lactide) 70:30 nasal implant.

**Methods:** In an ovine model, 66 rod-shaped absorbable implants were placed in 11 nasal dorsa. The sheep were sacrificed at 1.5 (N = 3), 6 (N = 3), 12 (N = 3), 18 (N = 1), and 24 months (N = 1). The nasal dorsum was harvested from each animal. Gross and histopathological examinations were performed.

**Results:** There were no postoperative complications, signs of infection, or tissue rejection throughout follow-up time points. Upon sacrifice, no abnormalities were identified during gross pathological examinations. The histology of the implant sites at all time points showed the implants were fully encapsulated through 12 months. The inflammatory reaction to the implants was minimal to mild at 1.5, 6, and 12 months. At 18 months the implant material was in the mass loss phase, being actively absorbed. During this phase, the inflammatory reaction within the fibrous connective tissue capsule reached expected moderate levels. By 24 months, the inflammatory reaction had diminished in most implantation sites and complete absorption of the rod implants was noted at some sites with nodular bundles of mature collagenized fibrous tissue replacing the implant, devoid of an inflammatory infiltrate.

**Conclusion:** Biocompatibility of the poly (L-lactide-co-D,L-lactide) 70:30 material was demonstrated when used as a nasal implant in the nasal dorsum ovine model. Absorption of the implant occurred approximately 18 to 24 months postoperatively, and the implant site was replaced with collagenized fibrous tissue.

**Key Words:** Absorbable implant, PLA, biocompatibility, histology, nasal obstruction.

**Level of Evidence:** NA.

## INTRODUCTION

Functional and cosmetic nasal grafting procedures are often focused on adding, removing, or repositioning cartilage in order to achieve the desired nasal structure. Nasal Valve Collapse is a common structural limitation associated with nasal obstruction due to instances where nasal cartilage is weakened by reduction rhinoplasty, aging, trauma, or iatrogenic causes. Weakened lateral nasal structures may result in increased collapse of the nasal sidewall during inspiration. In many cases, autologous cartilage may not be available, suitable, or easily shaped for providing the desired structural support, creating a need for alternative materials. Non-autologous implants used in nasal surgeries include tissues obtained from a human donor and synthetic materials.[1–3] Traditionally, non-absorbable synthetic alloplastic materials such as polyethylene, polytetrafluoroethylene, and silicone, have been associated with complications most notably adverse tissue reaction resulting in significant rejection at both early and late time points. Hence, the use of these aforementioned materials has been reserved as the last choice for implantation.[3] Absorbable polymer implants such as polydioxanone (PDA), and polylactic acid (PLA) copolymers have become a compelling option for nasal graft applications[4–7] as they may reduce the postoperative complications such as extrusions while continuing to provide structural support as the resorbed graft material is replaced with scar tissue.

The biocompatibility of PLA copolymers has been shown in various studies.[8–14] Before degradation takes place, there is minimal tissue reaction.[8] During active mass loss phase there is mild to moderate inflammatory reaction, which constitute part of the normal absorption process as the degradation products are actively removed by inflammatory cells.[9] It is known that macrophages play a prominent role in the absorption process by phagocytosing the debris of the biodegradable implant.[15] As the degradation of the polymers progress, an increase in collagen deposition is observed at the implant site.[8] Absorption kinetics and tissue response to PLA copolymers used as nasal implant have not yet been examined in detail.

Small animal models including rabbits and mice have been frequently used to study the tissue response to absorbable nasal implants.[4,16] However, due to size limitations, these models are not suitable to accommodate human-scale nasal implants in the relevant nasal anatomy. In this study, the poly (L-lactide-co-D,L-lactide)



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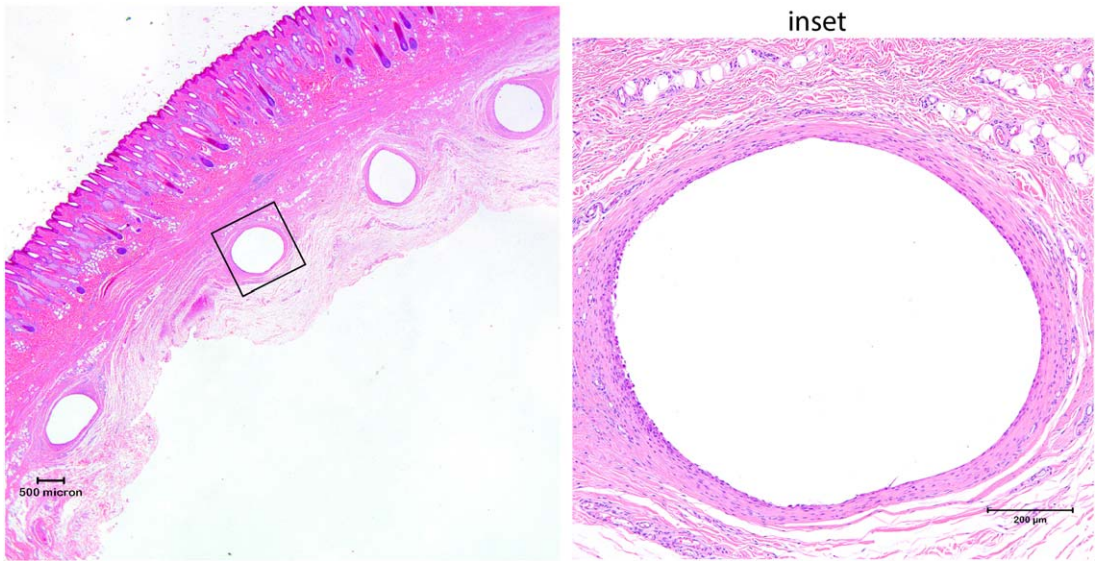


Fig. 1. Histology of the implantation sites at 1.5 months (10x, scale bar 500 $\mu$m). Clear round spaces correspond to the location of implants. Inset (100x, scale bar 200 $\mu$m) shows a thin fibrous connective tissue capsule and mild mixed inflammatory infiltrate within the capsule and at the luminal interface. Inflammation was only seen in the implant sites.

70:30 rod-shaped implants were placed in the ovine nasal dorsum in the proximity of the cartilage and subdermal soft tissue. Biocompatibility of the material at various time points was evaluated.

## MATERIALS AND METHODS

### *Implant Material and Procedure*

The animal study was performed at PMI Preclinical (San Carlos, CA). This study was approved by the Institutional Animal Care and Use Committee at PMI Preclinical. Poly (L-lactide-co-D,L-lactide) 70:30 material was molded into rods and sterilized with Gamma radiation.

The animals were fed a certified diet (Harlan, Madison, WI). On the surgery day, the fasted animals were sedated, weighed, and anesthetized. Anesthesia was induced with 5 mL ketamine (Butler Schein, Dublin, OH) and 5 mL propofol (Abbott Laboratories, North Chicago, IL) dose titrated intravenously. After induction, animals were maintained with 2% to 3% isoflurane (Butler Schein, Dublin, OH) in oxygen (2 L/minute) during the surgical procedure. During the surgery, a total of six rod-shaped implants were placed in the soft tissue overlying the cartilaginous nasal dorsum in each study animal. Placement was performed with an annular device consisting of a 16-gauge needle. All six rod-shaped implants were injected to rest on top of the cartilage just caudal to the frontal bone cartilage junction. A total of 66 rods were placed in 11 animals. Heart rate and rhythm, blood pressure, oxygen saturation, respiratory rate, and body temperature were monitored and corrected throughout the procedure.

The animals were humanely sacrificed at 1.5 months (4 animals, 24 implants), 6 months (2 animals, 12 implants), 12 months (3 animals, 18 implants), 18 months (1 animal, 6 implants), and 24 months (1 animal, 6 implants) postoperatively.

### *Necropsy and Histopathology*

The non-implant organs including kidneys, adrenals, spleen, liver, lungs, salivary glands, brain, lymph nodes, thymus, abomasum, duodenum, pancreas, jejunum, ileum, colon, and esophagus were all examined grossly by carefully sectioning all parenchymal organs at approximately 0.5-cm intervals and noting any changes present. The tissues containing the implants were collected and fixed with 10% neutral buffered formalin. The nasal dorsum tissues were trimmed in two transverse sections (lateral to medial, each including all 6 rod implants). In some cases, recut or retrimmed sections were performed in order to give adequate numbers of implants to evaluate along the nasal dorsum. Thus, the number of implant sites along the length of implanted material varied with the section examined. All sections were processed, embedded in paraffin blocks, sectioned at 4- to 5-micron thickness, and stained with hematoxylin and eosin for light microscopic evaluation.

The histology slides were evaluated for the tissue response to the implants. This evaluation included assessment of encapsulation of the material and the presence, type, and severity (minimal, mild, moderate, and severe) of inflammation. Severity of inflammation was defined by counting the number of macrophages and multinucleated giant cells: minimal (scattered, 1–10 cells/40x objective); mild (11–40 cells/40x objective); moderate (41–80 cells/40x objective) and severe (>80 cells/40x objective). The presence of calcification, erosion through the overlying epidermis, and any adverse effect of the material on the surrounding tissues were assessed.

## RESULTS

### *Animal Study, Necropsy, and Gross Pathology*

All animals were in good health for the duration of the study and maintained or gained body weight during their in-life phase. During daily observations, all animals were bright, alert, and responsive. For all the study animals sacrificed at different time points, no gross lesions or abnormalities related to the implantation materials were identified in any of the peripheral organs or tissues examined grossly at necropsy. None of the implants were associated with any infection or

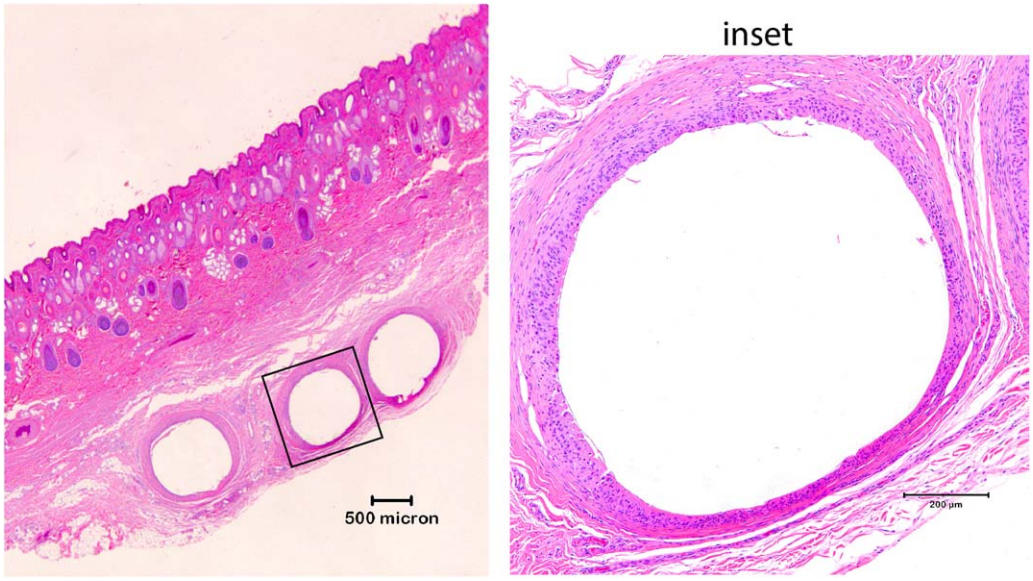


Fig. 2. Histology of the implantation sites at 6 months (10x, scale bar 500 $\mu$m). Clear round spaces correspond to the location of implants. All implant sites are encapsulated and the overlying dermis is devoid of an inflammatory infiltrate. The epidermis is intact. Inset (100x, scale bar 200 $\mu$m) shows a fibrous connective tissue capsule and mild inflammatory infiltrate (macrophages, a few multinucleated giant cells) within the capsule and primarily at the luminal interface.

swelling; no nasal discharge or an exudate over the nasal dorsum was present. There were no epidermal erosions at the implant sites.

### *Histology of Implantation Sites at Nasal Dorsum*

At 1.5 months postsurgery, the rod implants appeared as clear round spaces during microscopic examination of tissue sections. All implants were discretely encapsulated and surrounded by a minimal to mild mixed inflammatory infiltrate (macrophages, lymphocytes, multinucleated giant cells), expected microscopic findings for the type and location of the implanted material. Thin connective tissue capsules surrounded the implanted rods, and the observed inflammatory reaction was limited to the immediate area (connective tissue capsule and interface with the material) (Fig. 1). The surrounding and overlying dermis and epidermis were free of inflammation. The

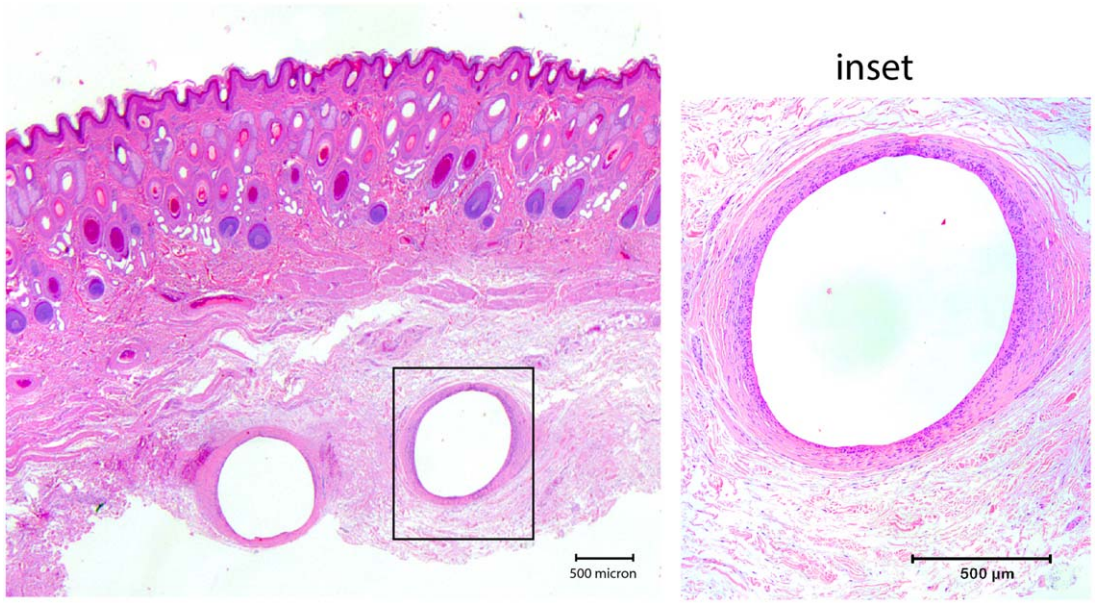


Fig. 3. Histology of the implantation sites at 12 months (10x). Clear round spaces correspond to the location of implants. All implant sites are encapsulated and the overlying dermis is devoid of an inflammatory infiltrate. The epidermis is intact. Inset (40x) shows a fibrous connective tissue capsule and mild inflammation (macrophages, few multinucleated giant cells) within the capsule and primarily at the luminal interface. Scale bars, 500 $\mu$m.

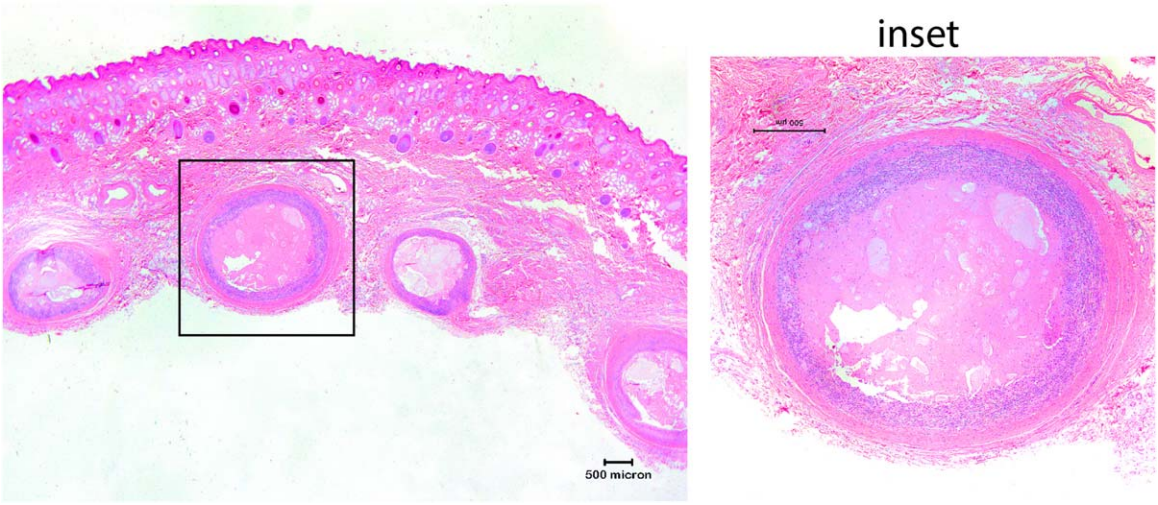


Fig. 4. Histology of the implantation sites at 18 months (10x). Round spaces correspond to the location of implants. All implant sites are encapsulated and the overlying dermis is devoid of an inflammatory infiltrate. The epidermis is intact. The spaces have been filled in with central amorphous eosinophilic material. Inset (40x) shows a moderate mixed inflammatory infiltrate is present at the interface of the implant capsule and the degrading material. Clusters of multinucleated giant cells admixed with lymphocytes and macrophages are associated with the degrading materials. Scale bars, 500 $\mu$m.

implant sites were free of calcification and no evidence of epidermal erosion was noted. There was no adverse effect of the materials on the overlying skin and subcutaneous musculature.

At both 6 months and 12 months postsurgery, the implants still appeared microscopically as clear round spaces. The material had not yet entered the mass loss phase where structural integrity of the implant would likely be compromised. The rod implants were well encapsulated and elicited a similar minimal to mild mixed inflammatory response when compared to 1.5 months postoperatively (Figs. 2 and 3).

At 18 months postsurgery, mass loss was observed as microscopically, the round spaces indicating the implant sites were filled in with central amorphous eosinophilic (Fig. 4). Most of the implant sites had a moderate mixed inflammatory response, which was an expected reaction with near complete structural degradation and active absorption.

At 24 months postsurgery, material absorption had progressed and the inflammatory response was waning. While evidence of the implanted material was still seen in some implant sites, occasional focal discrete areas of nodular collagen deposition indicated complete absorption had

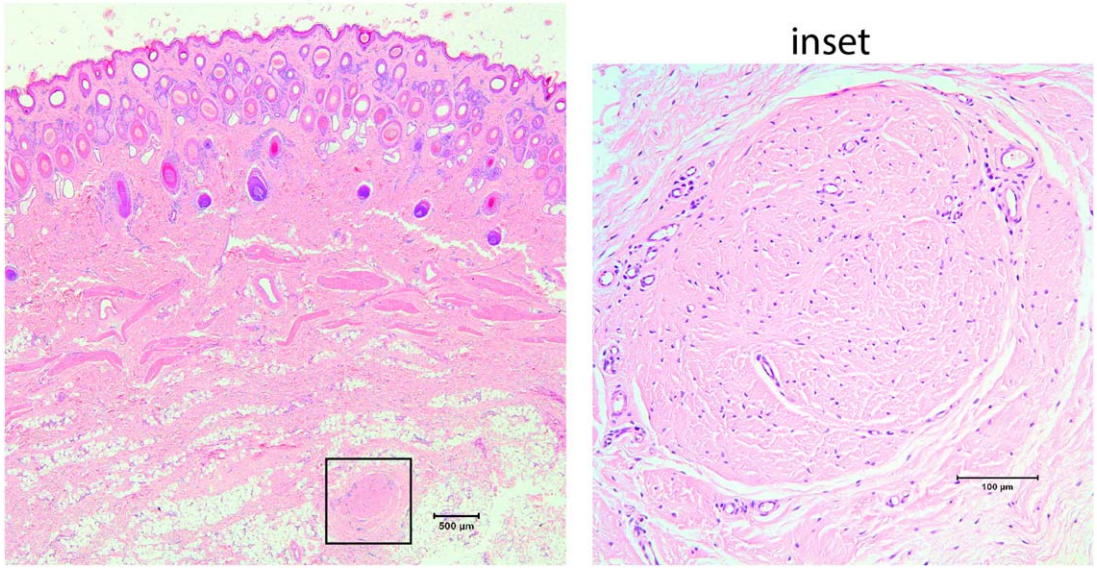


Fig. 5. Histology of the implantation sites at 24 months (20x, scale bar 500 $\mu$m). A nodular solid site corresponds to the location of implant. Inset (200x, scale bar 100 $\mu$m) shows the implant site replaced by mature collagenized connective tissue devoid of inflammation and any evidence of degraded material (complete resolution of implant site).

TABLE I.
Implant Structural Degradation, Inflammatory Tissue Response and Encapsulation (Healing Reaction) at Different Time Points During the Study Period.

| Duration of implantation (months) | Mass Loss | Severity of inflammation at majority of the implantation sites | Capsule Formation |
|---|---|---|---|
| 1.5 | None | Mild | Capsule formation |
| 6 | None | Mild | Stable capsule |
| 12 | None | Mild | Stable capsule |
| 18 | Active Mass Loss | Moderate | Active absorption within capsule |
| 24 | Some sites fully absorbed | Mild | Collagen deposition |

occurred (Fig. 5). These sites showed implants replaced by mature collagenized fibrous tissue devoid of any inflammation and degraded material.

## DISCUSSION

This study utilized an ovine nasal dorsum model for exploring tissue response to an absorbable poly (L-lactide-co-D,L-lactide) 70:30 nasal implant. The study characterized the tissue response at multiple timepoints through 24 months (Table I). The rod implants were fully encapsulated and the inflammatory reactions to the rod implants were minimal to mild at 1.5, 6, and 12 months postoperatively. Rapid mass loss and near complete structural degradation was observed at 18 months, accompanied by an expected increase in an inflammatory reaction surrounding the implant during active absorption. By 24 months, complete absorption of the rod implant was noted with nodular bundles of mature collagenized fibrous tissue replacing the implant site, devoid of an inflammatory infiltrate.

In past decades, autologous implants derived from patients have been the most preferred materials in nasal reconstructive surgery. However there are several disadvantages. Autologous implant materials require harvesting which not only adds time to the surgery, but often requires a second harvest site for rib or ear cartilage. The harvested materials require reshaping to fit the anatomy and extra care needs to be taken as any error in cutting may ruin the implant. Absorbable synthetic materials provide a promising alternative to autologous implants, as they are biocompatible, readily available in pre-shapes, and can be easily adapted to any desired shape or size for a particular patient.

This study demonstrated that the absorbable nasal implant composed of poly (L-lactide-co-D,L-lactide) 70:30, is biocompatible. Human data suggest that non-absorbable alloplastic materials have undesired complications including extrusions or infections following their use in nasal surgeries. Ramakrishnan and colleagues reported an extrusion rate of 21% (4 of 12 patients and 5 of 24 implants) using porous polyethylene as the implant material.[17] Winkler et al. reported an extrusion rate of 12% in 662 patients with implantations of high-density polyethylene and polytetrfluoroethylene for nasal reconstruction.[18] This study demonstrated that the rod implants made of PLA copolymers elicited expected and contained inflammatory response, with no extrusions observed through 24 months (total of 66 rods placed). The rod implant in this study was well into the mass loss phase at 18 months. Once the rod implant was completely absorbed at 24 months, it was replaced with scar tissue. As scar tissue is essentially collagenized fibrous tissue, it may continue to provide mechanical support for nasal structures at the original implant site beyond 24 months.[19]

This study is the first to use an ovine model to examine the tissue response surrounding a nasal implant. Sheep have similar nasal anatomy to humans, and it has been found to be the most useful animal model for hands-on training in endoscopic nasal and sinus surgery.[20,21] Here, an ovine nasal model was utilized to allow assessment of the tissue responses to nasal implants for as long as 24 months postoperatively.

A few limitations of the present study included small numbers of animals evaluated at 18 and 24 months postoperatively. Although very minimal variation in tissue response across different animals and implant sites was observed in earlier time points, it would be ideal to include more animals at later time points. Additionally, the findings in the animal model may not be completely representative of the response in humans. Future clinical studies in humans may provide additional information on tissue response to the implant material and the clinical relevance of the implant structural degradation and the material absorption profile.

Absorbable materials that are biocompatible, readily available, customizable in shape and size, and have different absorption kinetics, may be viable options for nasal implants rendering nasal surgeries less difficult, less time-consuming and may result in more positive surgical outcomes.[1,3] Future research exploring more types of absorbable material and long term follow up studies in humans will benefit patients in need of nasal construction.

## CONCLUSION

This study established an ovine nasal dorsum model for examining tissue response surrounding a rod-like nasal implant composed of poly (L-lactide-co-D, L-lactide) 70:30. It was found to be biocompatible and in the active mass loss phase by 18 months. Evidence of complete absorption at some sites, with collagenized fibrous scar tissue replacing the implant site, was observed at 24 months. Although the material absorption was not

complete at all sites, the expected inflammatory response was waning significantly at this time point and indicated ongoing absorption and fibrous scar replacement.

## CONFLICT OF INTEREST

SB and MR are employees of Spirox. BAS is a consultant to Spirox.


## FUNDING

Spirox.


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