

An Experimental Model of Eosinophilic Chronic Rhinosinusitis Induced by Bacterial Toxins in Rabbits

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Abstract

Background: The pathophysiology of chronic rhinosinusitis (CRS) is still not well known due to the multifactorial etiologies involved. Bacteria play a role in the pathogenesis of CRS by various means, including biofilm adhesion, intracellular persistence, or inducing inflammation secondary to toxins. Endotoxins and exotoxins, especially *Staphylococcus aureus* superantigens, can produce significant immune responses in the host and are implicated in patients with CRS. The majority of animal models described for CRS revalidates the pathophysiology of acute sinusitis, ostium occlusion, or foreign body associated infection.

Objectives: To evaluate an experimental model of eosinophilic CRS using prolonged exposure to bacterial toxins. The histological changes in rabbits exposed to *S. aureus* enterotoxin B (SEB), lipopolysaccharide (LPS), or lipoteichoic acid (LTA) were compared.

Methods: After induction with ovalbumin (OVA) sensitization with subcutaneous injection for 2 weeks, rabbits underwent surgery to insert an indwelling catheter into the maxillary sinus. The sinus was irrigated with OVA 3 times weekly for 2 weeks, followed by sinus irrigation with bacterial toxin (SEB: 1 µg/mL, LPS: 100 ng/mL, or LTA: 100 ng/mL) 3 times weekly for 4 weeks. The histological changes in the treated sinus were compared with control rabbits.

Results: Sinuses exposed to bacterial toxins (SEB, LPS, and LTA) produced significant mucosal thickening with infiltration of inflammatory cells, notably eosinophils. SEB was the only toxin that promoted a mixed pattern of inflammation, including eosinophilic and neutrophilic infiltration.

Conclusion: Our experimental model of eosinophilic CRS in rabbits produced significant mucosal thickening and inflammation in the sinuses exposed to bacterial toxins, with histological changes analogous to what is observed in patients with CRS with nasal polyps. This model may serve as a basis for future investigation of the pathogenesis of eosinophilic CRS in relation to bacterial toxins or as a model for testing new therapeutic modalities for this disease.

Keywords

chronic rhinosinusitis, animal model, rabbits, bacterial toxins, *Staphylococcus aureus* enterotoxin B, lipopolysaccharide, lipoteichoic acid, eosinophil, neutrophil, histopathology

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Introduction

Chronic rhinosinusitis (CRS) is a prevalent inflammatory disease of the sinonasal cavities that affects 5% to 15% of the general population. Although the prevalence rate is high, the understanding about CRS pathogenesis is only partial.^{1–4} Current evidence suggests that the underlying mechanisms involved in the genesis or progression of CRS are related to the interaction of exogenous factors and the type of inflammatory responses evoked in the host.^{5,6}

Among the several types of presentation, CRS can be phenotypically classified into those with (CRSwNP) or without nasal polyps (CRSSNP). In Caucasians, for instance, CRSwNP is marked by a mixed pattern of inflammation with a predominance of type 2-derived cytokines and eosinophilic infiltration in the sinonasal mucosa. Moreover, high levels of eosinophilic infiltration are related to more extensive disease, worse symptoms, poorer quality of life scores, and higher rates of nasal polyps recurrence, demonstrating the importance of this critical cell in the pathogenesis and prognosis of this disease.⁷

Before now, multiple groups have demonstrated the importance of microbes in the pathogenesis of CRS, including innate immunity impairment caused by microbiota disruption,^{8,9} sinonasal biofilms,^{10,11} intracellular bacteria,¹² and bacterial endo-/exotoxins (superantigens) inducing mucosal inflammation.^{13,14}

Among the bacterial endotoxins, lipopolysaccharide (LPS) and lipoteichoic acid (LTA) are the major components of the cell wall found in gram-negative and gram-positive bacteria, respectively, that induce robust host inflammatory responses.

LPS is an essential molecule in the majority of gram-negative bacteria that induces potent pro-inflammatory cytokines production, mucus secretion, and upregulation of several inflammatory molecules, mediated by the Toll-like receptor 4 (TLR4)/MyD88 signaling system pathway.^{14–16} LTA is a component of cell wall present in gram-positive bacteria that binds to CD14 and TLR2 in mammals and induces several immune responses, including the production of reactive oxygen and nitrogen species, release of pro-inflammatory cytokines, proteinases, and antimicrobial peptides.^{13,17} Among the bacterial exotoxins, *Staphylococcal* enterotoxins are proteins secreted by *Staphylococcus aureus* and other staphylococcal species that directly binds to major histocompatibility complex class II molecules with T cell receptors, bypassing the specific canonical antigen presenting cell processing. As a result, *Staphylococcal* enterotoxins can cause dysfunction in large populations of T cells and induce a massive inflammatory response in the host.¹⁸ *S. aureus* enterotoxins (SAE) have been recognized as an important molecule in the pathogenesis of

CRS, especially, in patients with CRSwNP.^{19,20} The presence of SAE in CRSwNP is associated with more extensive disease along with a high burden of inflammatory markers such as eicosanoids and type II cytokines.^{21–23}

To date, the majority of animal models described for CRS revalidate the pathophysiology of acute sinusitis, ostium occlusion, or foreign body associated infection, which may not represent the complex and multifactorial CRS pathogenesis.^{24–26} Furthermore, a few groups have described noninfectious models of CRS, especially using long-term exposure to bacterial toxins in the paranasal sinuses.^{27–29}

In this study, we demonstrate an animal model of eosinophilic CRS using chronic exposure to bacterial toxins in the maxillary sinus of previously OVA-sensitized rabbits, which developed similar histopathological changes to what is observed in humans with CRSwNP.

Objectives

This study aims to describe an animal model of eosinophilic CRS in rabbits, utilizing chronic exposure of bacterial toxins to the maxillary sinus.

Material and Methods

This study was approved by the Institutional Human Research Ethics Committee of the Ribeirao Preto Medical School of the University of Sao Paulo (FMRP-USP), with reference number 76/2013.

The surgical procedure performed was similar to that described by Chiu et al.,³⁰ and the protocol used is a modification of the experimentation described by Sejima et al.³¹ We utilized male New Zealand white rabbits (2.0–3.5 kg) that were housed in appropriate cages with food and drinkable water ad libitum. They were kept in-house for at least 3 days before commencement of the experiments.

The animals were split into 5 different experimental groups. In the first group (naïve control), we randomly selected 4 rabbits that were kept in the animal facility for 8 weeks, followed by euthanasia. The remaining rabbits from the other groups were sensitized prior to the experiment with subcutaneous injection containing 1 mL of 2.5% ovalbumin (OVA; Sigma-Aldrich) and 0.4% aluminum hydroxide (Sigma-Aldrich) on days 0 and 7. This was followed by a surgical insertion of an indwelling catheter into the right maxillary on day 14.³² Briefly, the animals were anesthetized with 5 mg/kg of xylazine (Rhobifarma, Hortolândia-SP, Brazil) and 30 mg/kg of dextroketa mine intramuscular (IM) (Rhobifarma), followed by intravenous injections of thiopental sodium

as required (Cristália, 25 mg/mL, ~1 mL every 15–20 min; Thiopentax®, Itapira-SP, Brazil).

The nasal dorsum was prepared aseptically followed by local infiltration of 1 mL of lidocaine 2% with epinephrine 1:200.000. The skin and subcutaneous tissues were dissected after a 3 cm vertical incision, preserving a periosteum flap of 2 × 2 cm to cover the catheter at the end of surgery. The anterior wall of the right maxillary sinus was drilled out (2.5 mm) and a polypropylene catheter (10.5 cm × 2.3 mm) was inserted into the sinus, tunneled through the subcutaneous tissue up to the skin between the ears, and fixed with nylon 4-0. Patency of the catheter was confirmed by flushing with 5 mL normal saline. The periosteum flap and skin were sutured in layers using vicryl 4-0 and nylon 4-0, respectively (Figure 1).

After the surgical procedure, the animals received IM antibiotic intraoperatively and 5 days later according to the manufacturer leaflet (0.6 mL of Pentabiótico® Veterinário Pequeno Porte; Eurofarma, São Paulo, Brazil). Animals were monitored weekly as regards

body weight and food ingestion. During the following 2-week postsurgery, the right maxillary sinus was irrigated with 0.5 mL of OVA 2.5% (diluted in NaCl 0.9%), 3 times per week. After this period, the maxillary sinus was irrigated 3 times per week for 4 weeks (0.5 mL) with one of the following solutions, diluted in PBS (Figure 2):

- Phosphate buffer saline (PBS) (group B);
- *S. aureus* enterotoxin B (SEB, 500 ng, 1 µg/mL, Sigma Aldrich) (group C)²¹;
- Lipopolysaccharide from *Escherichia coli* (LPS, 50 ng, 100 ng/mL, Sigma-Aldrich) (group D)^{33,34};
- Lipoteichoic acid purified from *Streptococcus pyogenes* (LTA, 50 ng, 100 ng/mL, Sigma-Aldrich) (group E)³³;

Euthanasia and Specimens Collection

Twenty-four hours after the last sinus irrigation, rabbits were euthanized with an intravenous injection of thio-pental sodium (100 mg/kg, 25 mg/mL) preceded by

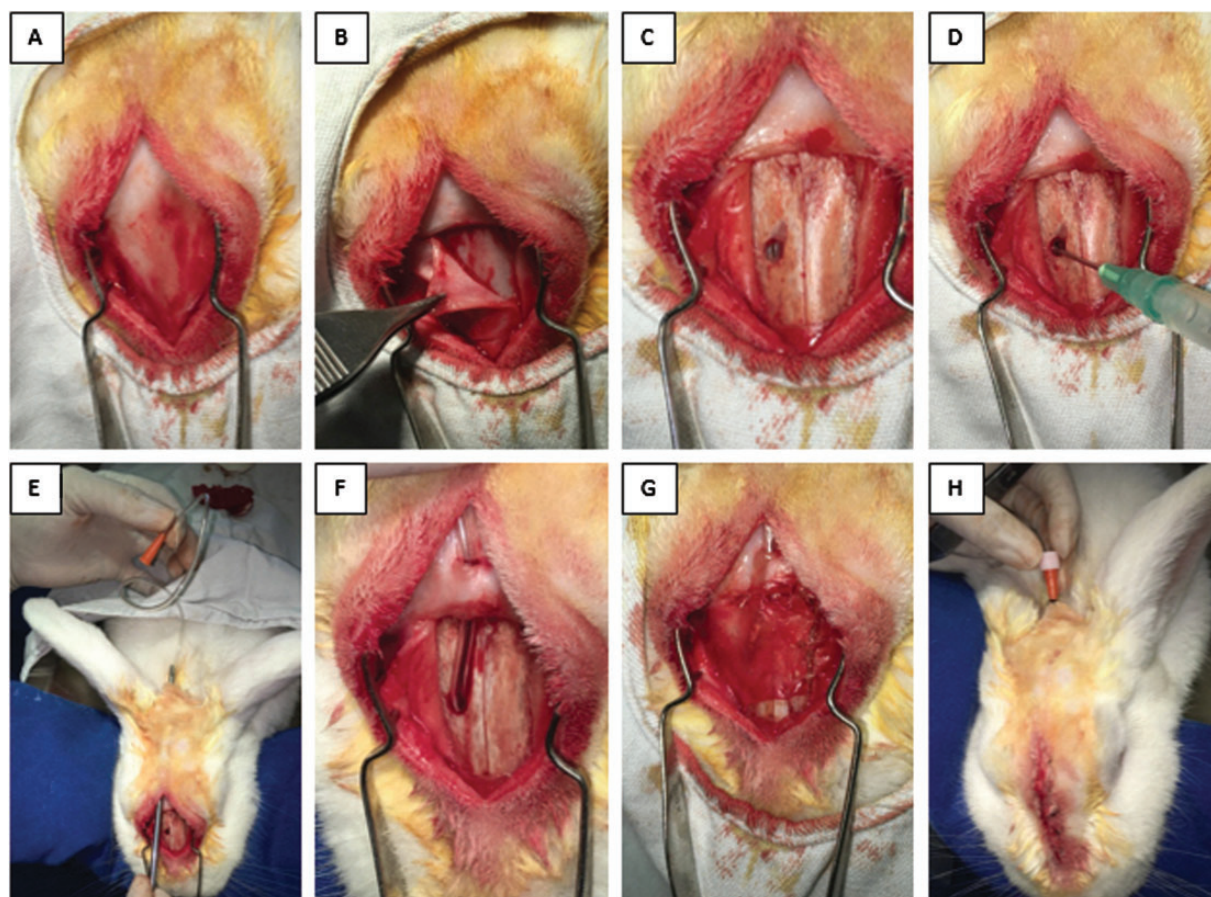


Figure 1. Main surgical steps utilized to implant an indwelling catheter for long-term irrigation of the maxillary sinus: (A) vertical incision, (B and C) “C” shape periosteum flap with 2-mm opening of the anterior wall, (D) sinus lavage with swab culture, (E) subcutaneous positioning of the indwelling catheter, (F) positioning of the catheter into the maxillary sinus, anchored by a periosteum flap, (G) suture of the flap and skin, and (H) final aspect of the surgery with the catheter exiting between ears.

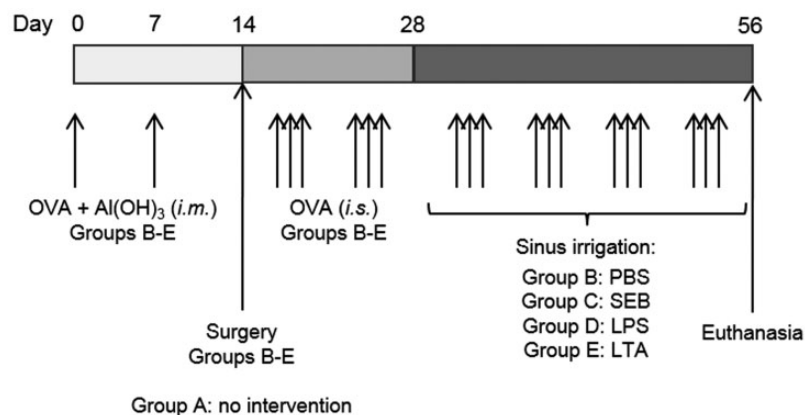


Figure 2. Timetable of experiments among the different groups. Al(OH)₃, aluminum hydroxide; i.s., intra sinus; OVA, ovalbumin; PBS, phosphate-buffered saline; LPS, lipopolysaccharide; LTA, lipoteichoic acid; SEB, *S. aureus* enterotoxin B.

intramuscular sedation with xylazine (5 mg/kg) and dex-troketamine (30 mg/kg).^{35,36} After confirmation of death, the nasal dorsum was dissected, the anterior wall of the maxillary sinus was drilled out, and the presence of polyps was recorded. The medial maxillary wall was removed and processed for histological analysis. The portion analyzed in all samples systematically included the medial portion of the maxillary wall, including the adjacent part of the main natural ostium. The same steps were repeated on the left maxillary sinus to harvest control samples. The sinus mucosa was fixed in formalin 10%, dehydrated, and embedded in paraffin blocks for sectioning. Slices of 6 μ m thickness were obtained and stained with hematoxylin–eosin. Images were acquired in a VS210 Olympus microscope BX61 in 40 $\times$ power magnification. Representative pictures were analyzed according to the following parameters:

1. Epithelial and subepithelial thickness—the thickness was measured every 400 μ m distance, and the mean of all measures in one sample was taken as the representative value.
2. Inflammatory infiltrate in the epithelial and subepithelial space: 3 representative areas of the epithelial space (200 μ m length) and subepithelial region (100 μ m $\times$ 100 μ m; total area of 10 000 μ m²) were defined, and the number of inflammatory cells (eosinophil, neutrophil, lymphocytes, plasma cell, and mast cell) were counted according to their cytological morphology. The density of cells in the epithelial and subepithelial region was expressed as the number of cells per 200 μ m and per 10 000 μ m², respectively.
3. Polyps analysis—polyps were defined as an elevation of the epithelium and stroma toward the lumen, with an intact underlying periosteum. The polyps, when present, were classified as edematous, fibroinflammatory, or with seromucinous gland hyperplasia.³⁷

The histological analyses were performed by 2 blinded investigators (A. A. B. and F. M. F.), taking the mean of the 2 counts when the difference between them was less than 5 cells per field. Whenever discordance was ≥ 5 cells per field, the 2 investigators reviewed the slides and decided together with the most appropriate count.

Swab samples from the maxillary sinus were collected on the day of surgery and at euthanasia; rabbits with positive *Pasteurella multocida* cultures on swab samples were excluded from the study.

Statistical Analysis

To compare the right maxillary sinus between groups, we used the Kruskal–Wallis test followed by Dunn's multiple comparisons test. The histological pattern found in group A (naïve rabbits) was taken as control for comparisons. To compare the right maxillary side (experimental) with the left side (internal control) in each group, we used the Mann–Whitney test. All tests were performed using GraphPad Prism version 6.0 for Windows, GraphPad Software, La Jolla, CA, USA, with a level of significance set at 5%.

Results

Sixty-two rabbits were included in the study, and 37 were eventually examined after exclusions ($n = 16$ developed a subcutaneous abscess or had positive culture; $n = 9$ had the irrigation catheter dislodged or obstructed or presented signs of suffering). We did not observe any association with a particular bacterial toxin inoculation, as about half of exclusions occurred before the sinus irrigation protocol ($n = 12$), and the remaining rabbits

were similarly distributed among groups (PBS = 5, SEB = 5, LPS = 2, LTA = 1).

The 37 rabbits analyzed were distributed as follows: 4 rabbits in the naïve control group (8 sides, nonoperated rabbits, group A); 9 rabbits that underwent only OVA sensitization followed by PBS sinus instillation (group B); 6 animals exposed to SEB (group C); 12 rabbits in the LPS (group D); and 6 rabbits in the LTA group (group E).

The mean body weight at the beginning of the protocol was 2755 g and at the end was 2940 g, with a mean weight gain of 7% throughout the 8 weeks. All groups had similar same body weight gain pattern.

To analyze whether the nonirrigated sinus (left side) was affected by irrigation in the contralateral side, we compared the thickness and inflammatory infiltrate between both sides. We observed that the maxillary sinus (right side) irrigated with PBS or with any other bacterial toxin developed a significant thickening of the epithelium and/or stroma compared to the control side (Figure 3).

Moreover, the sinus irrigated with PBS or bacterial toxins presented with more infiltration of inflammatory cells as against the control side (Figure 4), notably by eosinophils in the stromal compartment (Figure 4(B)). We did not observe an increase in neutrophil and lymphocyte counts in any of the groups exposed to bacterial toxins (Figure 4(C) and (D)).

In our model of CRS, sinus LPS irrigation produced a robust thickening of the epithelial and stromal compartments, whereas SEB and LTA increased the thickness of the epithelium or stroma, respectively. PBS exposure alone increased neither the thickness of the epithelium nor the stroma when compared with that of the naïve control mucosa (Figure 5).

Considering inflammatory cells infiltration, introduction of SEB and LPS into the maxillary mucosa produced a significant infiltration of cells both in the epithelial and stromal compartments, whereas LTA produced a pronounced infiltration of inflammatory cells only in the stroma (Figure 6).

Among the inflammatory cells, the number of infiltrated eosinophils was the highest in the sinus mucosa after bacterial toxin exposure. Notably, SEB instillation produced a robust increase of eosinophils (Figure 7) and neutrophils (Figure 8) both in the epithelium and stroma, whereas LTA produced an increase in eosinophils in the stromal compartment only. None of the toxins induced a significant infiltration of lymphocytes, plasma cells, or mast cells in the sinus mucosa in relation to the naïve sinus control (data not shown).

Considering sinus polyp formation, we identified the presence of microscopic polyps in 5 of 12 animals exposed to LPS and 3 of 6 in the LTA group. Among the LPS group polyps, 3 were fibroinflammatory (Figure 9(A)) and 2 were seromucinous gland hyperplasia (Figure 9(B)). The 3 polyps found in the LTA group were of the fibroinflammatory type (Figure 9(C)).

In summary, sinus exposure to bacterial toxins (SEB, LPS, LTA) significantly thickened the mucosa with infiltration of inflammatory cells, notably eosinophils (Table 1). In our model, SEB was the only toxin that promoted a mixed pattern of inflammation including eosinophilic and neutrophilic infiltration.

Discussion

In the past decades, several groups have proposed different animal models of CRS to better understand the pathophysiology of this disease and to potentially test

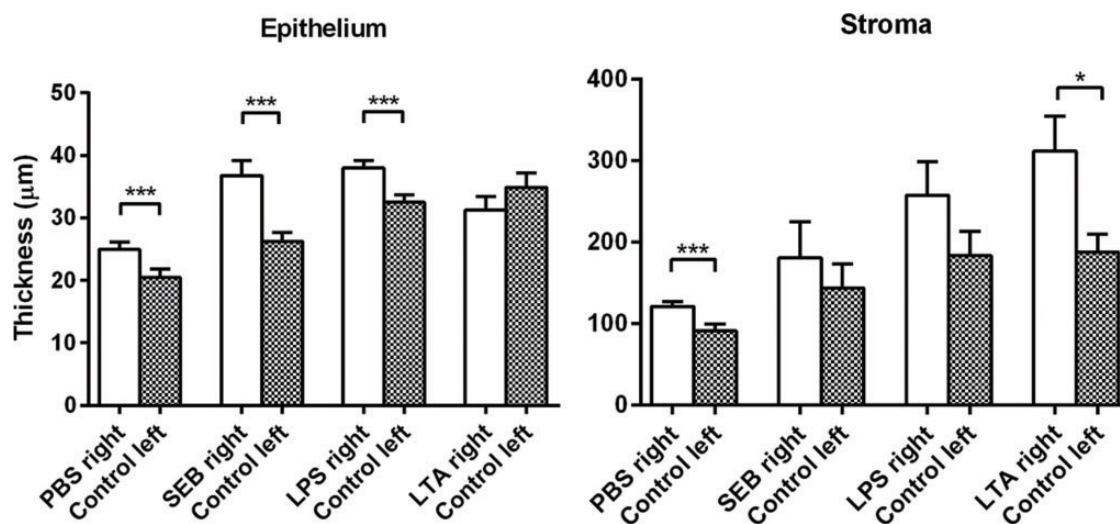


Figure 3. Epithelial and stromal thickness compared between the treated (right side) and control (left) sides. Data expressed as mean $\pm$ SEM. * $P < .005$; *** $P < .001$. LPS, lipopolysaccharide; LTA, lipoteichoic acid; PBS, phosphate-buffered saline; SEB, *S. aureus* enterotoxin B.

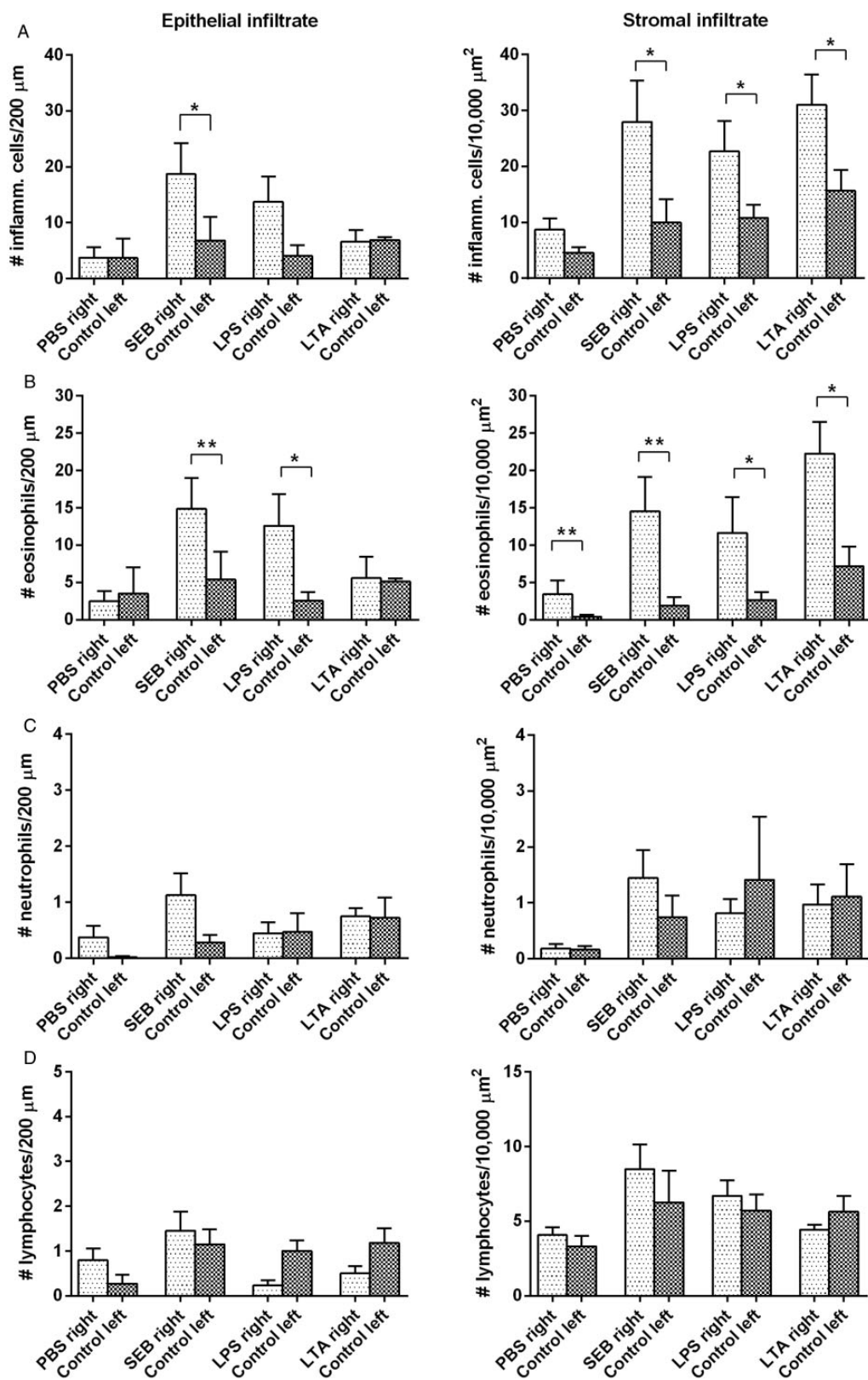


Figure 4. Inflammatory cell infiltration compared between the treated (right side) and the control side (left side). Data expressed as mean $\pm$ SEM. * $P < 0.05$; ** $P < 0.01$. LPS, lipopolysaccharide; LTA, lipoteichoic acid; PBS, phosphate-buffered saline; SEB, *S. aureus* enterotoxin B.

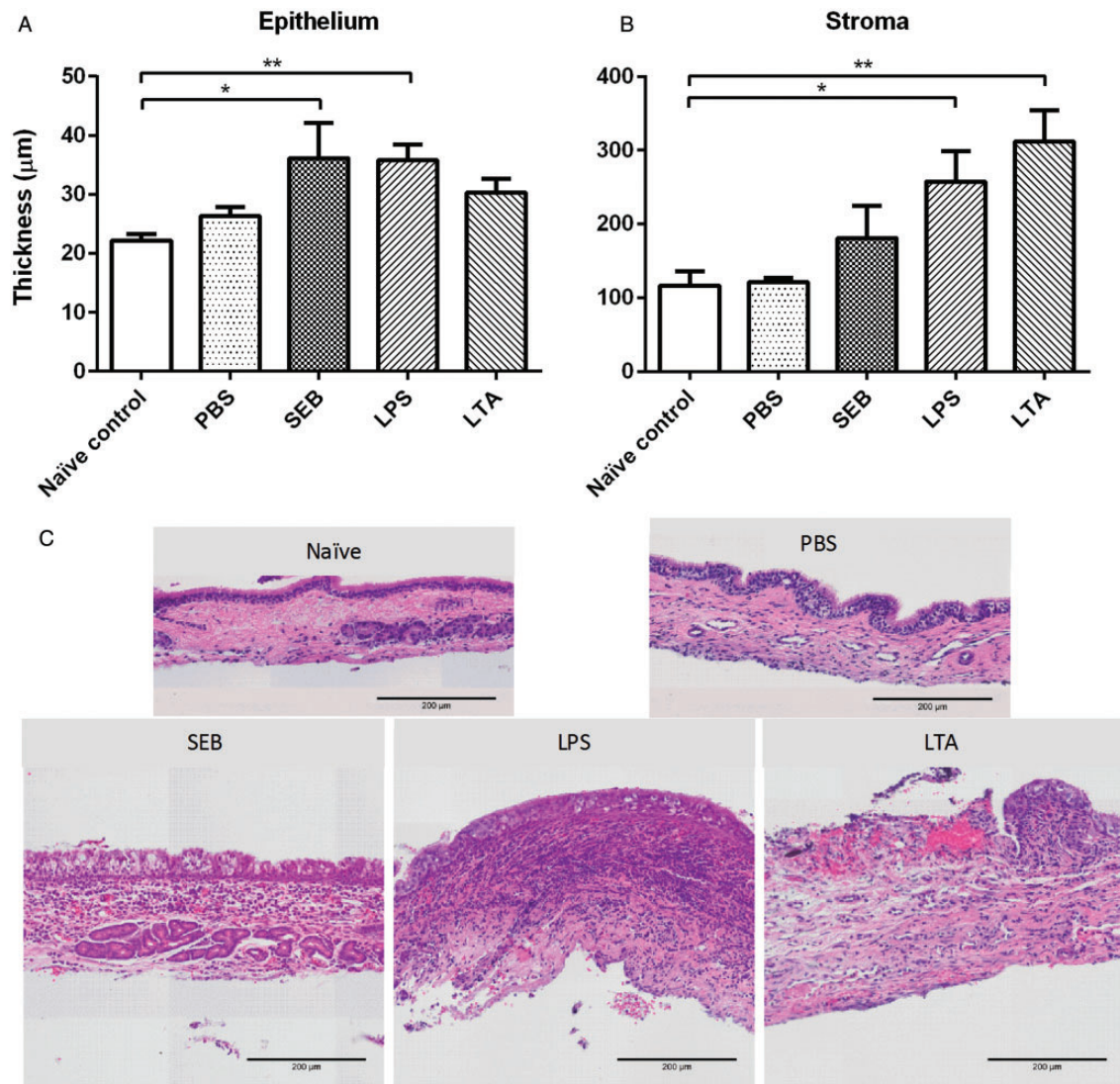


Figure 5. Thickness of the epithelium (A) and stromal compartment (B) of the maxillary mucosa exposed to different toxins (data expressed as mean $\pm$ SEM, * $P < .05$; ** $P < .01$). C, Representative images showing the epithelial and stromal thickening observed in each group. LPS, lipopolysaccharide; LTA, lipoteichoic acid; PBS, phosphate-buffered saline; SEB, *S. aureus* enterotoxin B.

new therapeutic approaches.^{38,39} In our model, we observed that the exposure of bacterial toxins, notably SEB, to the sinus mucosa induces mucosal thickening and inflammatory cell infiltration (mainly eosinophils), closely related to the pathology found in patients with CRSwNP.

In our study, we concentrated on having appropriate controls to evaluate the actual effects of bacterial toxins in the sinus mucosa. We analyzed whether the surgical disruption of the mucoperiosteum of the maxillary sinus, systemic effect of OVA-sensitization, mechanical irrigation and presence of an indwelling catheter, or even the possible reflux from one nasal cavity to the contralateral sinus could have influenced our outcomes. Interestingly, the PBS-irrigated side presented a thicker epithelium and stroma as well as higher eosinophilic infiltration in the

stroma relatively to the naïve contralateral side, demonstrating that the mechanical irrigation or the surgical manipulation itself promotes inflammation. We cannot overlook the potential additional or synergistic effects of the surgical/sinus manipulation on the inflammation promoted by bacterial toxins in our experiments. However, the inflammation caused by SEB, LPS, and LTA is notable in comparison to the tissue response to PBS, especially in regard to the total number of inflammatory cells and eosinophils. These findings strengthen the established fact of the pro-inflammatory action of these bacterial toxins on the sinonasal mucosa.

OVA sensitization used as an allergic inflammatory model in several other studies did work in our model as a facilitator for eosinophilic inflammation after exposure to bacterial toxins. The control group that was

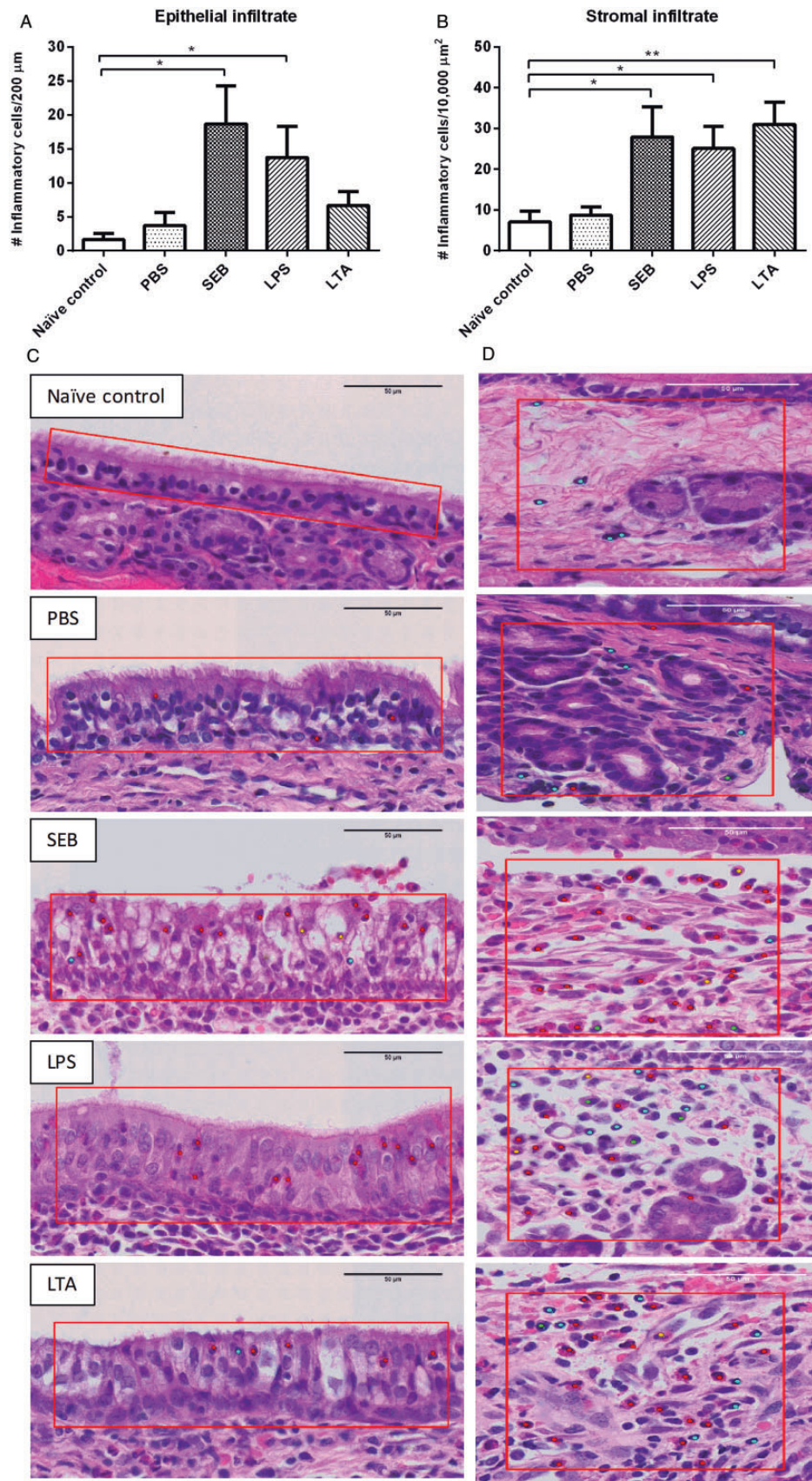


Figure 6. Infiltration of inflammatory cells in the epithelium (A) and stromal compartments (B) of sinus mucosa in response to different toxins (data are expressed as mean $\pm$ SEM, * $P < .05$, ** $P < .01$). Representative histological images of the epithelium (C) and stroma (D) among different groups. Red, eosinophil; yellow, neutrophil; blue, lymphocyte; green, plasma cell; white, mast cell. LPS, lipopolysaccharide; LTA, lipoteichoic acid; PBS, phosphate-buffered saline; SEB, *S. aureus* enterotoxin B.

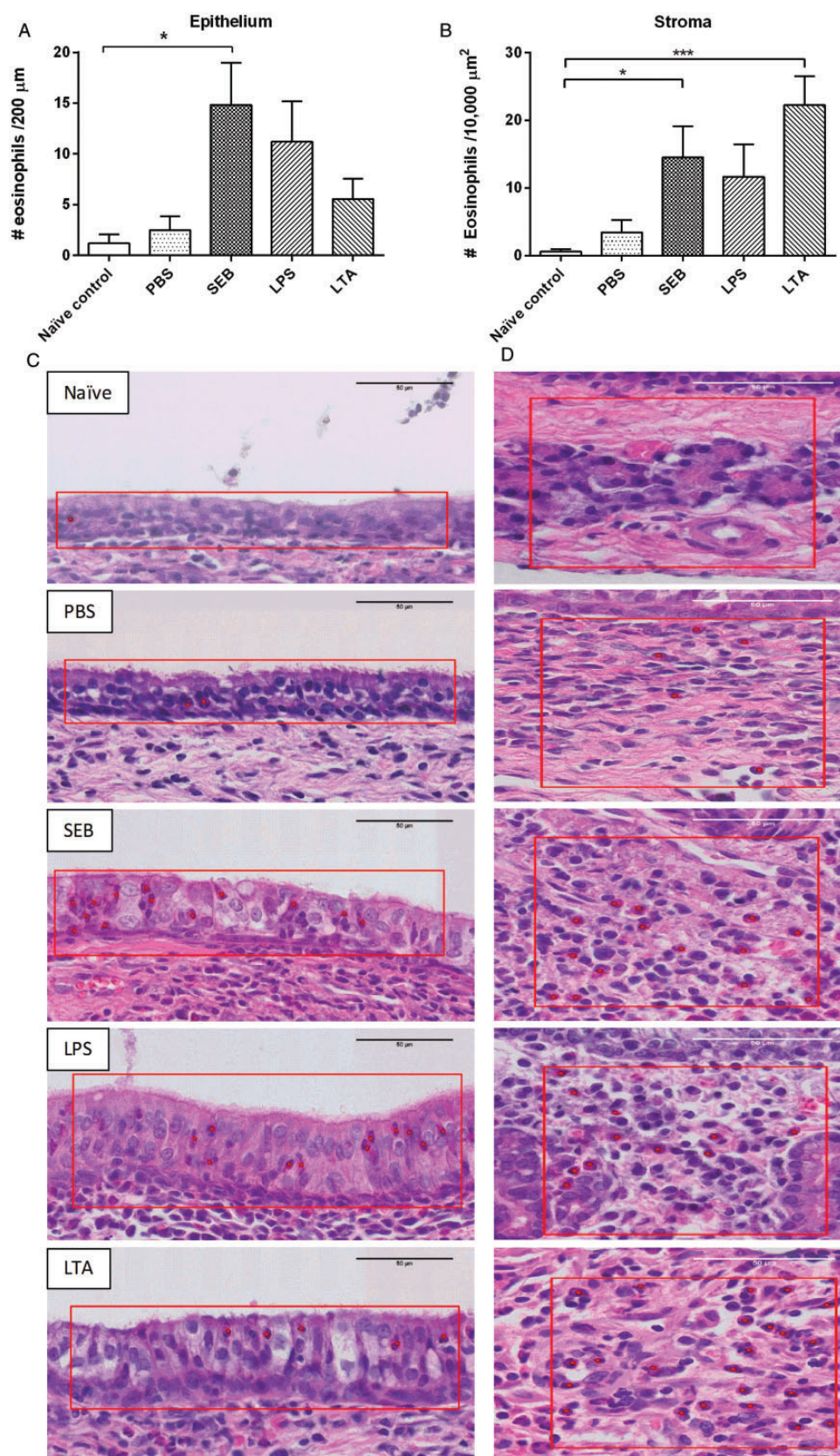


Figure 7. Infiltration of eosinophils in the epithelium (A) and stromal compartments (B) of sinus mucosa in response to different toxins (data are expressed as mean $\pm$ SEM, $*P < .05$, $***P < .001$). Representative histological images of the epithelium (C) and stroma (D) among different groups. LPS, lipopolysaccharide; LTA, lipoteichoic acid; PBS, phosphate-buffered saline; SEB, *S. aureus* enterotoxin B.

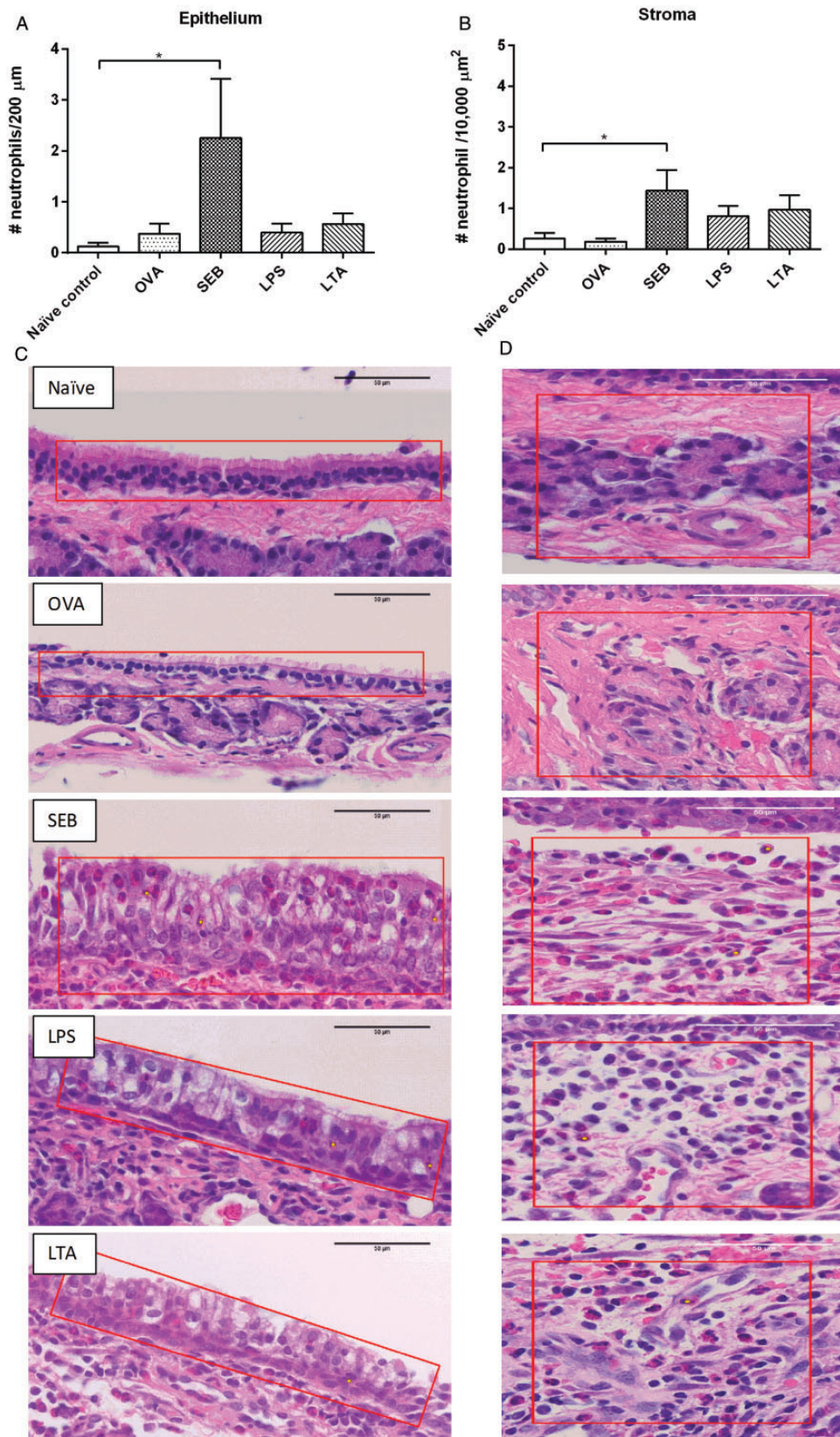


Figure 8. Infiltration of neutrophils in the epithelium (A) and stromal compartments (B) of sinus mucosa in response to different toxins (data are expressed as mean $\pm$ SEM, $*P < .05$). Representative histological images of the epithelium (C) and stroma (D) of the sinus mucosa among different groups. LPS, lipopolysaccharide; LTA, lipoteichoic acid; PBS, phosphate-buffered saline; SEB, *S. aureus* enterotoxin B.

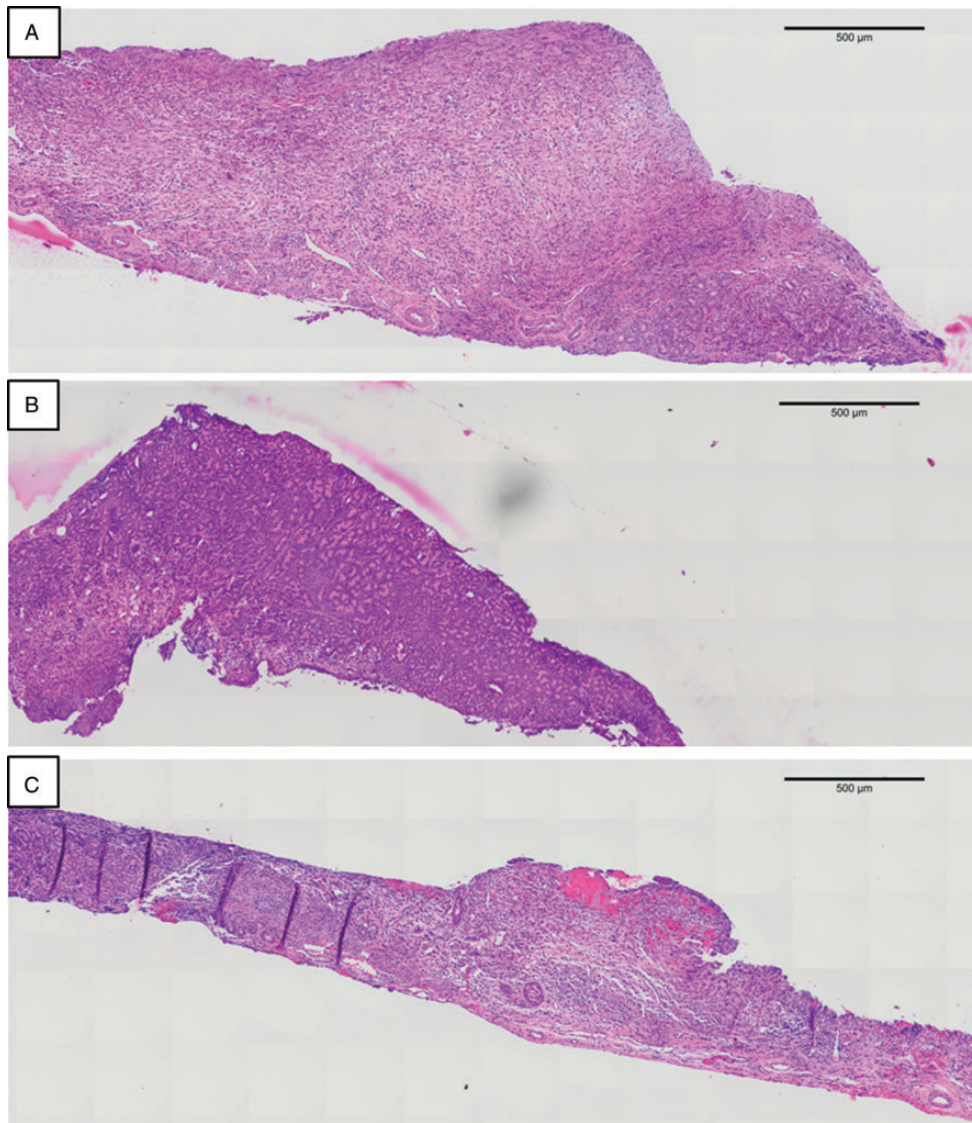


Figure 9. Histological images of maxillary sinus polyps observed in the LPS (A and B) and LTA groups (C).

Table 1. Summary of the Main Histological Differences Found in Each Group When Compared With the Naïve Control Mucosa.

	PBS	SEB	LPS	LTA
Epithelium				
Thickness		*	*	
Inflammatory cells infiltration (total)		*	*	
Eosinophilic infiltration		*		
Neutrophilic infiltration		*		
Lymphocytes, plasma cells, mast cells				
Stroma				
Thickness			*	**
Inflammatory cells infiltration (total)		*	*	**
Eosinophilic infiltration		*		***
Neutrophilic infiltration		*		
Lymphocytes, plasma cells, mast cells				

Abbreviations: LPS, lipopolysaccharide; LTA, lipoteichoic acid; PBS, phosphate-buffered saline; SEB, *S. aureus* enterotoxin B.
* $P < .05$; ** $P < .01$; *** $P < .001$.

subjected to OVA sensitization did not show histological differences from the naïve rabbits, demonstrating that the possible mucosal changes due to allergic mechanisms subsided during this period.

Previous studies have also reported relevant effects of LPS and LTA on the nasal mucosa, such as thickening of the mucosa, cilia loss, and increase of goblet cells, similarly to our findings.^{33,34} In a murine model of CRS, Wang et al. demonstrated that chronic nasal exposure to LPS (5 and 10 µg) induces neutrophilic nasal polyps, with a dose-dependent secretion of IFN- γ , TNF- α , and IL-17, but not IL-4. To the best of our knowledge, this is the first study to report the effects of long-term exposure of LPS or LTA on the sinus mucosa in rabbits.

Among the polyps found in 8 rabbits exposed either to LPS or LTA, the majority had fibroinflammatory characteristics, and none had mucosa with interstitial edema and loose stroma. This type of polyp may represent a reactive granuloma rather than a typical nasal polyp found in humans with CRSwNP. Similar findings were obtained by Kanda et al.⁴⁰ in a murine model of eosinophilic airway inflammation using nasal transference of activated splenic eosinophils.

Previous studies have reported the effects of SEB on the nasal mucosa both in vitro and in other animal models. Using human nasal epithelial cells culture, Huvenne et al.²¹ demonstrated that SEB induces the production of chemokines and growth factors in a dose-dependent manner. In disrupted cells from inferior turbinate controls and nasal polyps, stimulation with SEB (0.5 µg/mL) produces a secretion of several pro-inflammatory cytokines, from both Th1 and Th2 subsets.⁴¹ In a murine model of CRS using chronic exposure to SEB (103 days), Kim et al.⁴² and Khalmuratova et al.⁴³ demonstrated that low doses of SEB (5–10 ng) could induce nasal polypoid lesions with infiltration of eosinophils and lymphocytes in the respiratory mucosa. Interestingly, higher doses of SEB (500 ng) did not evoke nasal polypoid formation or inflammatory cell infiltration in relation to controls.⁴² On the other hand, the dose of SEB utilized in our study (500 ng) did produce significant inflammatory changes in the sinus mucosa of rabbits, with a mixed pattern of eosinophilic and neutrophilic infiltration. There are some pieces of evidence pointing to the fact that the type of immune response is dependent on the concentration of the superantigen: low levels skew to a Th2-type pattern of response, whereas high doses polarize to a Th1-type inflammation.^{44,45} In our rabbit model, the dose of superantigen utilized in rabbits (500 ng) causes a mixed pattern of Th1 (neutrophilic) and Th2 (eosinophilic) inflammation in a shorter time of exposure (4 weeks). One of the limitations of this study was that we did not use different doses of toxins to test dose-dependent responses, they were

empirically chosen from prior data in the literature. Despite the low dose of LPS and LTA used in this study (50 ng), it was sufficient to observe significant mucosal changes and inflammatory cells infiltration. However, we were not able to verify whether different doses of toxins would promote various types of inflammatory responses, such as neutrophilic or eosinophilic inflammation, or even a more robust polyp formation. Furthermore, we did not explore different timeframes of bacterial toxins exposure, which could have changed our findings. If bacterial toxins play a role in CRS in humans, we believe that low-dose exposure of bacterial toxins in a persistent or prolonged manner might be the most probable scenario, rather than acute and intense exposures. Another limitation of this study is that we did not evaluate other mechanisms of CRS pathogenesis described in humans, including mucociliary clearance changes, inflammatory molecular biomarkers, and microbiota related issues. Finally, we unfortunately had a significant loss of animals throughout the experiments, mainly due to local infectious complications or issues with the positioning of the catheter.

The quest for an optimal animal model of CRS is still a challenge. However, we believe that this aseptic model of eosinophilic CRS, which did not use foreign body or sinus ostium occlusion, may be helpful in the future to study bacterial toxin related mechanisms in CRS, with potential therapeutic implications.

Conclusions

In our experimental model of eosinophilic CRS in rabbits, chronic exposure to bacterial toxins produced significant mucosal thickening and infiltration of inflammatory cells, especially eosinophils, analogous to what is observed in humans with CRSwNP. This model, especially involving SEB exposure, may serve as a basis for future investigation of the pathogenesis of CRS related to bacterial toxins or as a model for testing new therapeutic modalities in this disease.

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Declaration of Conflicting Interests

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