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Subconjunctival antimicrobial poloxamer gel for treatment of corneal ulceration in stranded California sea lions (*Zalophus californianus*)

Claire A. Simeone,*,† Carmen M. H. Colitz,‡ Kathleen M. Colegrove,§ Cara L. Field,* Carlos Rios,* Heather L. Chandler¶ and Shawn P. Johnson*

**The Marine Mammal Center, Sausalito, CA 94965, USA; †Office of Protected Resources, National Marine Fisheries Service, National Oceanic and Atmospheric Administration, Silver Spring, MD 20910, USA; ‡All Animal Eye Care, Jupiter, FL 33458, USA; §Zoological Pathology Program, University of Illinois at Urbana-Champaign, Brookfield, IL 60513, USA; and ¶The Ohio State University, Columbus, OH 43210, USA*

Address communications to:

C. Simeone

Tel.: +1 415 480 9665
Fax: +1 415 754 4078
e-mail: simeonec@tmmc.org

Abstract

Objective Corneal ulcers are commonly encountered in pinnipeds. Prolonged oral antibiotics and topical ophthalmic solutions may not be practical to administer, and novel treatment techniques are desired. Thermodynamic gels are a potential solution because they hold antimicrobials at the site of injection, slowly releasing drug. This study investigated the clinical efficacy of antibiotic-impregnated poloxamer gel in management of corneal ulceration.

Animal studied Twenty-six California sea lions undergoing rehabilitation at The Marine Mammal Center.

Procedures A poloxamer gel mixed with 2% enrofloxacin was subconjunctivally injected in the treatment group. Control animals received oral doxycycline. Systemic anti-inflammatories and analgesics were administered as needed. Corneal examinations under general anesthesia were repeated weekly, and included sampling for bacterial culture and corneal cytology, collection of high-quality corneal images, and treatment administration until the ulcers were healed.

Results There was no gross or histologic evidence of a localized tissue reaction to the gel administration in the conjunctiva, and no evidence of systemic reaction to therapy in animals that died due to unrelated causes during the study period (n = 17). In animals that experienced a superficial corneal ulcer involving only epithelium or superficial stroma (n = 12), all lesions resolved completely, in both treatment and control groups. Of those animals with deeper or more complex ulcers involving keratomalacia or descemetoceles (n = 15), four demonstrated complete lesion resolution (all four received gel treatment).

Conclusions This study demonstrates that subconjunctival antibiotic poloxamer gel administration is a safe and effective alternative therapeutic option to traditional treatments for superficial corneal ulceration in pinnipeds.

Key Words: California sea lion, corneal ulcer, marine mammal, poloxamer gel, subconjunctival injection, *Zalophus californianus*

INTRODUCTION

Corneal ulcers are a common medical presentation for both free-ranging and managed pinnipeds.[1–4] Prolonged oral antibiotic administration may be undesirable to treat this localized issue, as some oral antibiotics do not penetrate the cornea, and others require high dosage to observe significant therapeutic efficacy.[5] Topical ophthalmic solutions may not be practical in cases of severe blepharospasm or for managed animals not accustomed to topical medication delivery, and because their application requires typically requires additional restraint, eye drops

may not be practical during periods of high inpatient volume in rehabilitation settings. Novel techniques to treat corneal ulcers are desired.

Poloxamer gels are thermo-reversible carriers, existing in a fluid state when refrigerated, and changing into a solid gel state at body temperature.[6] These gels are in widespread use in the human medical field in a variety of settings, including ophthalmic and cardiovascular use using both topical and intravenous routes.[7–9] Poloxamer gels have been combined with a variety of drugs, and provide prolonged drug plasma concentrations when compared with solo drug therapy.[10–14] Trials suggest that when combined with antibiotic therapies, poloxamer gels hold the antibiotic at the site, releasing the drug slowly.[15–18]

The purpose of this study was to evaluate the effect of an antibiotic-impregnated poloxamer gel on the treatment of corneal ulcers in California sea lions (*Zalophus californianus*). We hypothesized that corneal ulcers treated with antibiotic-impregnated gel injected into the conjunctiva would have more rapid clinical resolution than ulcers treated with systemic antibiotics alone, and would heal without local or systemic adverse reactions.

MATERIALS AND METHODS

California sea lions (CSLs) with active corneal ulcers, undergoing rehabilitation at The Marine Mammal Center (TMMC) in Sausalito, CA, USA, were included in the study. From June 2014 to September 2015, 26 CSLs were included in the study (18 in the treatment group, eight in the control group). All animals were housed in saltwater pools with a semi-closed filtration system. Water quality was routinely monitored. Animals were randomly assigned to a treatment group. Criteria for inclusion included an active corneal ulcer as evidenced by fluorescein stain retention and lack of discernable abnormalities of the anterior chamber (e.g., hyphema, hypopyon, anterior lens luxation). Eight animals were excluded from the study because they had a corneal lesion that did not retain fluorescein stain. Anesthetic induction was achieved with either injectable drugs or inhalant anesthesia according to protocols routinely used in pinnipeds, and subsequently maintained on isoflurane.[19] An ophthalmic examination was performed, including evaluation of the cornea and anterior chamber. If globe rotation occurred secondary to the anesthetic and prevented evaluation of the cornea, a retrobulbar block was employed to improve visualization of the cornea, with 4 mg/kg lidocaine HCl using a two-point (ventrolateral and ventromedial) transpalpebral injection with a 20-ga, 1.5-inch needle.[20] A sterile swab of the cornea was collected for cytology and microbiology. Fluorescein stain was subsequently used to confirm active ulcers. High-quality images of the cornea were taken at each examination, and daily if the cornea was visible. All photographs were evaluated retrospectively for corneal ulcer size, depth, type and severity of ocular discharge, and severity of corneal edema, by individuals (CS, CC, HC) masked to the treatment group, and given a corneal grade between 0 and 9, with 0 being normal and 9 being severe. Ulcers were categorized as superficial if they involved only epithelium or superficial stroma, and complicated if they involved deep stroma, Descemet's membrane, or perforation of the cornea.

Control animals received oral doxycycline (10 mg/kg BID), the standard of care for corneal ulcers in pinnipeds at TMMC. Doxycycline was selected for its ability to be secreted in tear film, as well as for its ability to inhibit the matrix metalloproteinases responsible for corneal malacia and stromal loss, and to promote corneal re-epithelialization through upregulation of growth factors such as in the TGF-B family.[21–24] Treatment animals received poloxamer 409 gel compounded with enrofloxacin at a 2% concentration (Thermaffix Gel, Med Specialties Rx, Yorba Linda CA, USA). The antibiotic gel was administered via subconjunctival injections of 0.2 mL at each of two sites in the dorsolateral and ventrolateral bulbar conjunctiva with a 22-gauge needle (Fig. 1). No treatment animals received doxycycline. Antibiotic choice and dosing in the gel were established using two criteria: (1) based on culture and sensitivity results of the most common microbes retrieved from multiple anatomic sites in the TMMC rehabilitation population and (2) based on consultation regarding which antibiotics mix best with Thermaffix gel, taking into account the vehicle, pH, and concentration, as well as previous clinical successes in companion animals. Doxycycline should not be mixed with the gel, as it is water-soluble and will quickly diffuse out of the gel upon injection (M. Gonzalez, personal communication).

Systemic anti-inflammatories and analgesics were administered as needed to both groups. Carprofen (4.4 mg/kg PO SID) and tramadol (4 mg/kg PO TID) were administered if blepharospasm was noted, and sustained-release buprenorphine (0.12 mg/kg SQ q72 h) was administered if discomfort was not alleviated. Anesthetic examinations were repeated weekly, with culture, cytology, and gel treatment administered until the ulcer was considered healed, which ranged between one and four doses. A healed ulcer was defined as an absence of fluorescein stain uptake, although corneal edema and fibrosis may have persisted.

In cases of patient mortality, a gross necropsy was performed, and tissues were collected in 10% neutral-buffered formalin for histopathologic evaluation. Microbiology was performed at TMMC in fifteen of eighteen treatment animals and four of eight control animals. Bacterial cultures were performed using a swab to inoculate a blood agar plate (nonselective media), a MacConkey agar plate (selective for Gram-negative bacteria), and a Columbia CNA agar plate (selective for Gram-positive bacteria). Plates were incubated for 24 h and checked for bacterial growth. If no growth occurred at 24 h, plates were

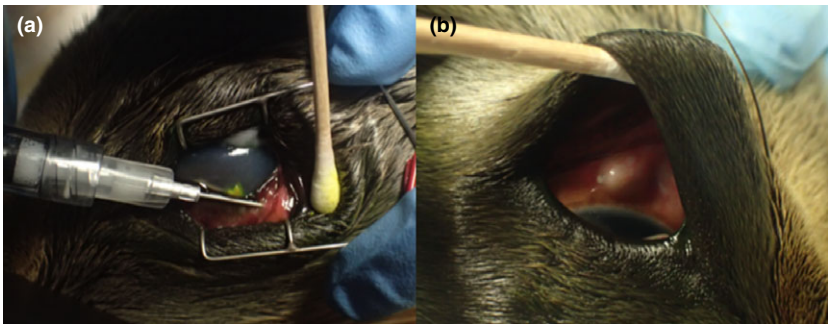

Figure 1. Subconjunctival injection of poloxamer gel in the bulbar conjunctiva of a California sea lion (a) and the bleb that forms following injection (b).

incubated for 24 more hours. Plates with bacterial growth were worked up depending on the organism(s). Gram-negative bacteria (enteric and nonenteric) were isolated and identified with the use of biochemical and sugar reactions using the TMMC laboratory flowchart for Gram-negative bacteria. Gram-positive bacteria (primarily *Staphylococcus*, *Streptococcus*, and *Enterococcus*) were isolated and identified with the use of Gram stain and other reactions specific for gram positives using the TMMC laboratory flowchart for Gram-positive bacteria. Bacteria were identified at least to the genus, and to the species if possible. Kirby–Bauer testing was pursued on the most dominant bacterial colonies, or on a bacterial colony of particular pathogenic concern to the CSL cornea, to determine antibiotic sensitivity to enrofloxacin ($n = 5$ in treatment group) or doxycycline ($n = 2$ in control group). Sensitivity testing was not pursued if the animal died prior to completion of the culture.

Statistical analysis

Confidence intervals for mean weight and length of treatment were estimated using nonparametric bootstrap, specifically the adjusted bootstrap percentile (BCa) interval, with 10 000 replications. Dependence between treatment and length of treatment, as well as between treatment and final corneal status, was assessed using permutation tests. While animals were visually evaluated daily, confirmed ulcer healing was only assessed during anesthetic events that occurred once a week. This limited the ability to statistically assess true healing times. Due to the small sample size, power is low and true differences between treatment and control would need to be large to be detected in a statistical test. All analyses were performed in the statistical software R.[25]

RESULTS

Table 1 summarizes the characteristics of the animals included in the study (sex, age class, weight, corneal ulcer characterization) and course of therapy (total treatment days, number of gel treatments, final corneal status, and final disposition). The treatment group consisted of five males (27.8%) and 13 females (72.2%), while the control group consisted of three males (37.5%) and five females (62.5%). This gender distribution is similar to the sex ratio of total admitted CSLs per year (1557 total animals; 45.7% males, 54.3% females). Animal age class ranged from pups (<1 year old) to adults (sexually mature). The mean weight for all treatment animals was 30.5 kg (95% confidence interval (CI) 20.3–47.8), while the mean weight for control animals was 41.5 kg (95% CI 22.9–72.1). Seven of eighteen ulcers in the treatment group were classified as superficial, and five of eight ulcers in the control group were classified as superficial. Nine of eighteen ulcers in the treatment group and five of eight ulcers in the control group were diagnosed at admission; nine of eighteen ulcers in the treatment group and three of eight ulcers in the control group developed during rehabilitation. The mean length of treatment did not significantly differ among groups (treatment animals: 10.4 days (95% CI 7.7–15.3); control animals: 13.3 days (95% CI 7.6–28.6, Approximative two-sample Fisher–Pitman permutation test, $Z = -0.668$, $P = 0.55$). Of the treatment animals, ten CSLs had complete ulcer resolution (55.6%), four died with an active ulcer (22.2%), and four had treatment failure and globe perforation (22.2%). Of the control animals, five had complete ulcer resolution (62.5%), one ended with an active ulcer (12.5%), and two experienced treatment failure and globe perforation (25%). There was no significant difference in final corneal status between treatment and control animals (approximative general permutation independence test, $P = 0.87$). All animals that presented with a superficial corneal ulcer ($n = 12$) experienced complete ulcer resolution, in both treatment and control groups. Anti-inflammatories and analgesics were administered to 20 animals ($n = 12$, $n = 8$ controls). The mean length of treatment with analgesics was 8 days. The length of treatment corresponded to the severity of the lesion, regardless of treatment group.

Figure 2 shows representative photos of a superficial, uncomplicated corneal ulcer that healed with gel treatment. Seven treatment animals had superficial ulcers, all of which healed with one gel treatment and in fewer than 9 days (mean 6.1, 95% CI 4.1–7.6). Five controls had

Table 1. California sea lions with active corneal ulcers treated with subconjunctival poloxamer gel (treatment) or oral doxycycline (control)

Animal ID	Treatment group	Sex	Age class	Classification	Grade (0–9)	Total Tx days	# Gel Tx	Final corneal status	Final disposition	Noted on admit
CSL-11441	Case	M	Subadult	Complicated	7	6	1	Healed	Released	Yes
CSL-11569	Case	F	Subadult	Complicated	9	14	2	Active ulcer	Euthanized	No
CSL-11612	Case	M	Pup	Complicated	8	26	4	Active ulcer	Released	No
CSL-11623	Case	M	Yearling	Complicated	9	33	4	Perforation	Euthanized	No
CSL-11659	Case	F	Pup	Complicated	7	15	2	Perforation	Died	Yes
CSL-11685	Case	M	Pup	Complicated	9	9	1	Perforation	Died	Yes
CSL-11691	Case	M	Pup	Superficial	5	3	1	Healed	Euthanized	Yes
CSL-11729	Case	F	Yearling	Superficial	3	3	1	Healed	Died	No
CSL-12061	Case	F	Pup	Complicated	7	14	2	Active ulcer	Died	No
CSL-12099	Case	F	Pup	Complicated	6	7	1	Perforation	Euthanized	No
CSL-12119	Case	F	Pup	Complicated	5	7	1	Healed	Euthanized	Yes
CSL-12314	Case	F	Yearling	Superficial	1	5	1	Healed	Euthanized	No
CSL-12612	Case	F	Adult	Superficial	4	8	1	Healed	Released	No
CSL-12644	Case	F	Adult	Superficial	1	9	1	Healed	Released	No
CSL-12671	Case	F	Subadult	Superficial	4	8	1	Healed	Released	Yes
CSL-12677	Case	F	Adult	Complicated	5	9	1	Healed	Euthanized	Yes
CSL-12698	Case	F	Subadult	Complicated	4	5	1	Active ulcer	Euthanized	Yes
CSL-12722	Case	F	Yearling	Superficial	1	7	1	Healed	Released	Yes
CSL-11213	Control	F	Yearling	Complicated	9	47	N/A	Perforation	Euthanized	Yes
CSL-11220	Control	F	Adult	Superficial	6	10	N/A	Healed	Euthanized	Yes
CSL-11225	Control	F	Adult	Superficial	6	12	N/A	Healed	Released	Yes
CSL-11230	Control	F	Subadult	Superficial	6	4	N/A	Healed	Released	No
CSL-11266	Control	M	Yearling	Superficial	6	6	N/A	Healed	Died	No
CSL-11862	Control	M	Pup	Complicated	8	7	N/A	Active ulcer	Died	No
CSL-12080	Control	M	Pup	Complicated	9	8	N/A	Perforation	Died	Yes
CSL-12105	Control	F	Pup	Superficial	3	12	N/A	Healed	Released	Yes

M, Male; F, Female; Tx, Treatment.

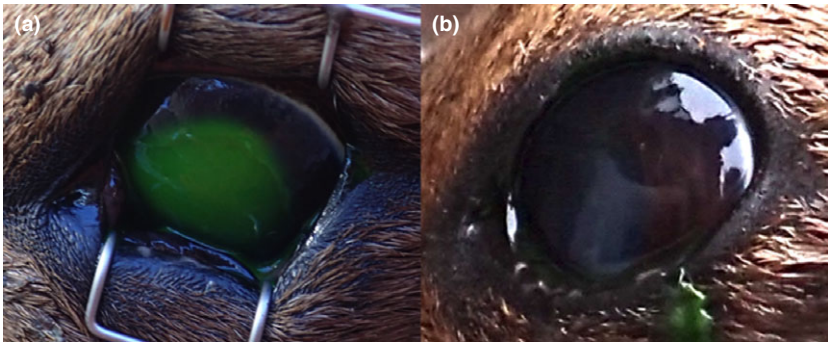

Figure 2. Representative photographs of a superficial, uncomplicated corneal ulcer at initial examination (a) and at recheck 7 days later (b). Note large, superficial ulcer with large lip that takes up fluorescein stain (a). No evidence of corneal edema, defect, or fluorescein stain uptake at 7 days, when ulcer was considered resolved (b). [CSL-12677]

superficial ulcers, which all resolved in fewer than 12 days (mean 8.8, 95% CI 5.2–10.8).

Fourteen total animals (55.6%) had complicated ulcers. Figure 3 shows representative photos of a complicated corneal ulcer with severe malacia that progressed to globe perforation while receiving gel treatment. Nine total animals (33.3%) experienced globe perforation ($n = 4$ treatment, $n = 2$ control). A variety of organisms were cultured from the corneal swabs (Table 2). *Pseudomonas* sp. were the most common bacteria cultured ($n = 6$ treatment, $n = 2$ control), followed by alpha-hemolytic *Streptococcus* sp. ($n = 6$ treatment). All four treatment animals and one of two control animals had positive cultures for *Pseudomonas sp.* with intermediate susceptibility to enrofloxacin, and severe corneal malacia noted on examination.

Thirteen animals ($n = 10$ treatment, $n = 3$ control) either died or were euthanized for reasons unrelated to ophthalmic disease or treatment during the study period, while four animals were euthanized or died as a direct result of globe perforation ($n = 2$ treatment, $n = 2$ control). Malnutrition and pneumonia were the most common causes of death ($n = 8$ treatment, $n = 3$ control), and two

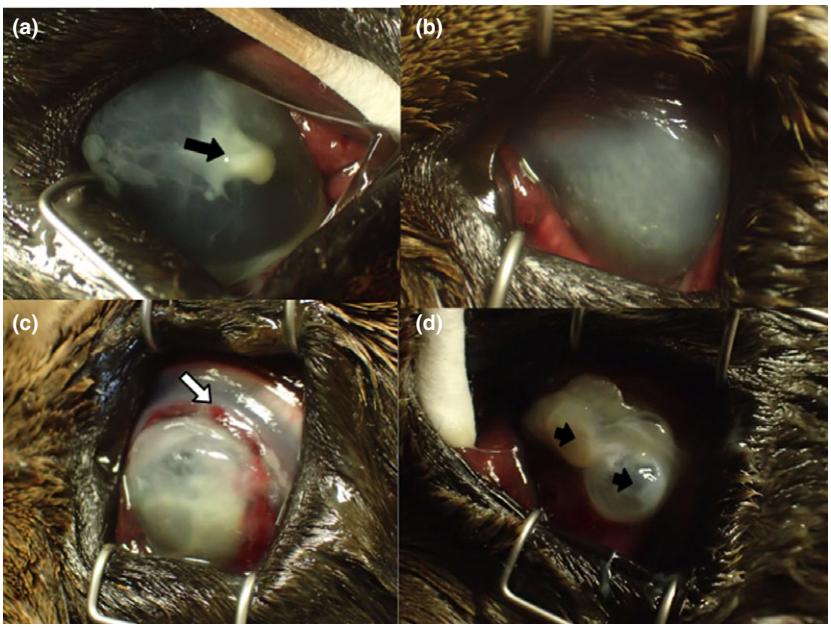

Figure 3. Representative photographs of a complicated, malacic corneal ulcer that progressed to descemetocele and eventual globe perforation. Note sloughing, necrotic stroma (black arrow) at initial examination (a). Marked corneal edema and malacia present at three-week recheck examination (b). Note the large area of white blood cell infiltrates surrounded by blood and fibrin (white arrow) at four-week recheck examination (c). At 5 weeks (d), multiple bullae are present (arrow heads), which progressed to corneal perforation 3 days later. [CSL-11623]

treatment animals were euthanized due to persistent seizures secondary to domoic acid intoxication. There was no gross or histopathologic evidence of a localized tissue reaction to the gel administration in the conjunctiva and no evidence of systemic reaction to the therapy in any animals. In all cases, histopathology confirmed clinical diagnosis of healed or complicated corneal ulcers.

One juvenile male, CSL-12581 presented with fungal keratitis that progressed to corneal perforation (Fig. 4). Due to the fungal nature of the infection, he was not included in the case series, but was treated with the gel. In addition to enrofloxacin, fluconazole (0.2%) was mixed with poloxamer gel. Gel was administered for 7 weeks (four of which included fluconazole). Whole blood was collected at each anesthetic event for generation of platelet-rich plasma, which was also applied topically to the cornea. Fluorescein stain uptake was negative at the five-week recheck, and 8 weeks following the initial examination, a small area of focal corneal edema remained, but with no evidence of blepharospasm or discomfort. The animal was able to track and catch live fish and was cleared for release into the wild.

DISCUSSION

Corneal ulcers occur in both captive and wild pinnipeds, and while the etiology of the condition is being investigated in both populations, physical trauma, changes in water quality, excessive sunlight, viral infections, underlying uveitis, and other factors have been attributed to ocular surface damage.[3,26–28] In the rehabilitation setting at TMMC, corneal ulcers are most frequently attributed to trauma. CSLs that have ingested domoic acid, a potent marine neurotoxin produced by some diatom species of the genus *Pseudo-nitzschia*, exhibit neurologic signs such as seizures, ataxia, and coma.[29] Intoxicated animals often present with superficial abrasions of the cornea, thought to be secondary to rolling along the beach when neurologically inappropriate. Although they were a minority of the total number of cases in the present study, these ulcers are an example of an ideal clinical use for the poloxamer gel: they typically involved only the superficial epithelium with minimal associated corneal edema (Fig. 2), did not exhibit growth on aerobic culture, and resolved with a single gel application in all cases when rechecked 7 days later.

Antibiotic therapy alone has variable success in managing complicated ulcers.[30,31] In addition to topical medications such as hyperosmotic solutions and polysulfated glycosaminoglycans, surgical interventions including debridement, and keratotomy or keratectomy are often indicated for chronic epithelial erosions to reduce healing times, which therapies such as cross-linking and tectonic or conjunctival graft placement may be indicated for deeper stromal ulcers.[32–36] It is not surprising that eleven of the fifteen complicated cases did not resolve with antibiotic treatment alone, particularly in the face of a *Pseudomonas aeruginosa* infection with intermediate sensitivity to enrofloxacin or doxycycline (two treatment, one control). Furthermore, every animal in the study was undergoing rehabilitation for a variety of issues unrelated to the

Table 2. Aerobic culture and sensitivity results for corneal swabs of California sea lions with active corneal ulcers

ID	Treatment group	Organism	Sensitivity
CSL-11213	Control	*Citrobacter amalonaticus*	None run
		Enterococccus sp.	–
CSL-11441	Case	*Enterococcus faecalis*	Susceptible
		Corynebacterium sp.	–
CSL-11569	Case	*Proteus* sp.	None run
		Escherichia coli	–
		Streptococcus sp. (alpha hemolytic)	–
		Diptheroids	–
		Staphylococcus sp.	–
CSL-11612	Case	*Citrobacter amalonaticus*	None run
		Staphylococcus sp.	–
CSL-11623	Case	*Pseudomonas aeruginosa*	Intermediate
		Streptococcus sp. (non-hemolytic)	–
CSL-11659	Case	*Klebsiella* sp.	Resistant
		Pseudomonas aeruginosa	Susceptible
		Enterococcus sp.	–
CSL-11685	Case	*Pseudomonas aeruginosa*	None run
CSL-11691	Case	*Staphylococcus* sp.	None run
		Enterococccus sp.	–
CSL-11729	Case	*Pseudomonas* sp.	Intermediate
		Staphylococcus sp.	–
CSL-11862	Control	*Pseudomonas aeruginosa*	Susceptible
		Diptheroids	–
CSL-12061	Case	*Arcanobacterium phocae*	None run
		Streptococcus sp. (alpha hemolytic)	–
		Pseudomonas aeruginosa	–
		Escherichia coli	–
CSL-12080	Control	*Pseudomonas aeruginosa*	Intermediate
		Staphylococcus sp.	–
CSL-12099	Case	*Pseudomonas aeruginosa*	None run
CSL-12105	Control	Diptheroids	None run
CSL-12119	Case	*Moraxella lacunata*	None run
		Psychrobacter phenylpyruvicus	–
CSL-12314	Case	Non-enteric gram negative rod	None run
		Streptococcus sp. (alpha hemolytic)	–
CSL-12581	Case	Non-enteric gram negative rod	None run
		Yeast	–
CSL-12644	Case	Non-enteric gram negative rod	None run
		Streptococcus virudans	–
CSL-12677	Case	*Psychrobacter phenylpyruvicus*	Susceptible
		Streptococcus sp. (alpha hemolytic)	Intermediate
CSL-12698	Case	*Psychrobacter phenylpyruvicus*	Susceptible

Only initial culture is noted, although culture was repeated if ulcer was not resolved at recheck exam. Kirby–Bauer testing was performed for most abundant bacterial colonies on the aerobic culture, or if a particular pathogen of concern was noted. Sensitivity testing was not performed if the animal died prior to completion of the culture (noted as 'none run').

corneal ulcer, potentially creating immunocompromised conditions. With the exception of the adults being treated for acute domoic acid intoxication (n = 5), all animals had a degree of malnutrition, ranging from mildly underweight to severely emaciated. Protein depletion, even in cases of mild nutritional deficiencies, impairs wound healing and slows the recovery process when compared with normally nourished patients.[37,38]

Pseudomonas aeruginosa and *Streptococcus* spp. were most commonly isolated from the corneal ulcers, with varying sensitivity to enrofloxacin. Historically, corneal ulcers have not frequently been cultured at TMMC, and initial antibiotic selection was chosen based on culture results from CSLs from a variety of sites, where *E. coli*, *Klebsiella* spp., *Enterococcus* spp., and *Staphyloccocus* spp. are the most commonly isolated bacteria.[39] Based on these results and the ease and availability of compounding, enrofloxacin appeared to be an appropriate broad-spectrum antibiotic of choice to use for the study. Based on the common isolates from this study, other antibiotic choices such as newer fluoroquinolones may be more appropriate in some cases. Because the poloxamer gel can be mixed with a variety of antimicrobials, an appropriate course of action would be to tailor antibiotic therapy to individual culture and sensitivity results.

There was one animal in which the poloxamer gel was utilized as part of a multimodal therapy to treat a complicated ulcer (Fig. 4). CSL-12581 presented with complicated malacic ulcers that progressed to corneal perforation. Topical platelet-rich plasma and subconjunctival injections of gel mixed with antibiotic and antifungal medications were applied over the course of 7 weeks. After 29 days of treatment, the ulcers were considered resolved and corneal edema continued to clear through the date of their release 8 weeks after admission. This case is an example of antimicrobial-impregnated gel as part of intensive management of complicated corneal ulcers.

Thirteen animals died or were euthanized due to causes unrelated to their ophthalmic disease, primarily due to malnutrition and pneumonia, or domoic acid intoxication. As a result, these corneal ulcers could not be followed to their potential resolution, therefore reducing the total number of healed ulcers in the study. Complete gross necropsy and ocular histology on each case showed that there was no evidence of a local or systemic evidence of toxicity or conjunctival inflammation in either the gel or control group. Histopathology was also able to confirm that seven of the ulcers had resolved. Blepharospasm was a common clinical finding in both treatment and control animals, and lack of gross or histologic evidence of inflammation in the conjunctiva suggested that the blepharospasm was due to the ulcer itself, and not the gel.

It is important to note that fifteen of the animals (55%) had ulcers that were noted on admission, meaning that the date of initial insult was unknown. While daily photographs were collected when possible, animals were also only evaluated under anesthesia once weekly; thus, the actual date of resolution was unknown and was estimated to the closest 7 days. These two facts make it difficult to

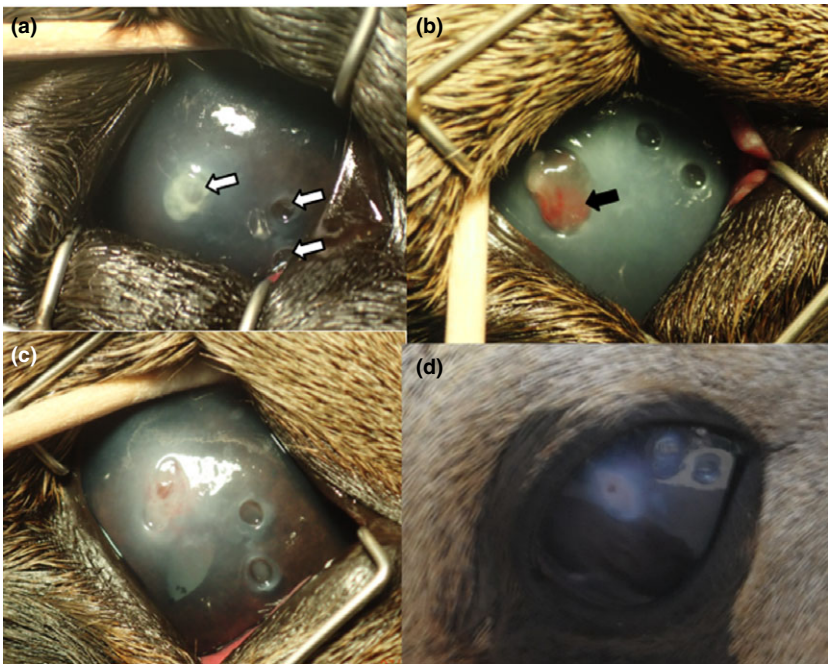

Figure 4. Case progression for CSL-12581, with corneal trauma that progressed to fungal keratitis. Note during initial examination (a), three ulcers of approximately 50% stromal depth (white arrows), including an area of marked cellular infiltrates temporally. At three-week recheck examination (b), the temporal ulcer has perforated (black arrow) and the two other ulcers are 90-95% stromal depth. At five-week recheck (c), corneal edema has partially cleared, allowing visualization of cataractous lens. There is no fluorescein stain uptake on any part of the cornea at this point, and the ulcers are considered healed. Eight weeks following initial examination (d), a small area of focal edema remains, but with no evidence of blepharospasm or discomfort.

compare the exact healing times of treatment groups. Upon receiving treatment, there was no statistical difference between treatment and control group mean healing times; however, a larger sample size and more precise measurement of healing time are needed to accurately compare antibiotic poloxamer gel therapy to oral doxycycline administration.

This study incorporated a small sample size, which is common in studies using marine mammals, but reduces the ability to evaluate the results in a statistically robust manner. There was also no control over other systemic medications being used, other than antibiotics, because the animals had other issues that warranted therapy during their rehabilitation. While the study was designed to eliminate any drugs with known interactions with either doxycycline or enrofloxacin, there is a possibility that medication interactions occurred. The pharmacokinetics of poloxamer 407 and several drugs has been investigated in a variety of species and suggest an increased half-life and prolonged plasma concentrations.[10,12,15,16] Unfortunately, the pharmacokinetics has not been studied in marine mammals, and therefore, this study cannot demonstrate the duration of drug retention in the tissue. However, despite these limitations, the gel demonstrated similar healing outcomes for superficial ulcers compared to the current standard of care.

Antimicrobial-impregnated poloxamer gels may be used in a variety of situations. In a managed care setting, subconjunctival injections may be helpful for intractable pinnipeds, or in cases of severe blepharospasm, where eye drops are not a useful treatment modality. Poloxamer gel treatment holds particular promise in the event of an oil spill, as petroleum contains substances such as hydrogen sulfide that are extremely irritating to the eye, leading to severe conjunctivitis, corneal erosions, and ulcers.[40] Traditional treatments, including prolonged oral antibiotic therapy and administration of eye drops multiple times per day, are often not feasible in wild animals undergoing rehabilitation. Targeted administration of antimicrobial therapy for focal bacterial or fungal infections could revolutionize the treatment of corneal ulcerations in wildlife by potentially reducing the number of sedative or anesthetic procedures a single patient experiences; by decreasing stress to the animals; and by reducing the risk to both animal care personnel and the stress to the animals associated with daily handling.

In total, 27 California sea lions with corneal ulcers were treated in this investigation. All 12 animals that had a superficial ulcer experienced complete resolution. There was no gross, histologic, or systemic reaction to the poloxamer gel. Subconjunctival antibiotic poloxamer gels proved safe and were equally effective as the standard-of-care therapeutic option for treating superficial corneal ulceration in California sea lions. As healing was clinically equivalent between the two therapies, antibiotic poloxamer gel can be used as a substitute for or in combination with

doxycycline. The poloxamer gel should not be used alone for complicated, deep stromal, or melting ulcers, but shows promise as part of a multimodal treatment regimen.

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