

Evaluation of the histologic reactions to commonly used suture materials in the skin and musculature of ball pythons (*Python regius*)

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Objective—To evaluate histologic reactions to 8 suture materials and cyanoacrylate tissue adhesive (CTA) in the musculature and skin of ball pythons.

Animals—30 hatchling ball pythons.

Procedures—In each snake, ten 1-cm skin incisions were made (day 0). At 8 sites, a suture of 1 of 8 materials was placed in the epaxial musculature, and the incision was closed with the same material. One incision was closed by use of CTA. No suture material was placed in the tenth incision, which was allowed to heal by second intention (negative control). Snakes (n = 5/group) were euthanized for harvest of treatment-site tissues at days 3, 7, 14, 30, 60, and 90. Skin and muscle sections were examined microscopically and assigned a subjective score (0 to 4) for each of the following: overall severity of inflammation, fibrosis, number of macrophages, number of granulocytes, number of perivascular lymphocytes, and degree of suture fragmentation.

Results—Subjective score analysis revealed that CTA did not cause a significant inflammatory response, compared with the negative control. All suture materials caused significantly more inflammation over all time points; for all suture materials, inflammatory response scores were significantly higher than values for the negative control 90 days after implantation. No sutures were completely absorbed by the end of the study period, and several sutures appeared to be in the process of extrusion.

Conclusions and Clinical Relevance—In snakes, CTA can be used to close small superficial incisions or lacerations with minimal inflammatory response, and sutures may undergo extrusion from tissues prior to complete absorption. (*Am J Vet Res* 2011;72:1397–1406)

With the increasing popularity of reptiles as pets over the past 20 years, there has been an increased demand for veterinary expertise in reptile medicine and surgery.^{1,2} The frequency of performing surgery on reptile patients has increased, and many procedures are now commonly performed.^{2–4} The overall process of wound healing in reptiles is similar to that in mammals but slower.^{2,3,5} Certain aspects of wound healing that are unique in reptiles must be taken into consideration as part of their surgical treatment. Healing in reptiles can be influenced by the environmental temperature, the wound orientation, and the nutritional and health status of the animal.^{2,5–7} The inflammatory cells of reptiles may also differ from those of mammals, and they may not have the same proteolytic enzymes needed to break down

ABBREVIATIONS

CG	Chromic gut
CTA	Cyanoacrylate tissue adhesive
FS	Fibrosis score
GS	Granulocyte score
IS	Inflammation score
MN	Monofilament nylon
MS	Macrophage score
PLS	Perivascular lymphocyte score
SFS	Suture fragmentation score

some suture materials.³ These factors may have a role in the selection of appropriate suture material.

Sutures are placed to maintain incised or injured tissue in apposition to allow the tissue to heal.^{8–10} The ideal suture material should have high tensile strength to resist fragmentation and provide sufficient time to allow tissue healing, have good knot security, resist infection, and cause no inflammatory, immunogenic, or carcinogenic reactions. The reaction of tissue to suture materials depends on several variables such as the type, quantity, and duration of suture implantation as well as characteristics of the tissues into which material is implanted.

Various studies^{11–18} have investigated the degree of tissue reaction to different suture materials in various species, and the tissue reaction to suture materials

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in reptiles has been previously investigated in a single study.¹³ That investigation was limited to 4 suture materials, and the histologic reaction was only evaluated at 1 time point, 7 days following surgical sex determination. Although those findings provided some insight into wound healing in reptiles, the study duration was too short to provide information regarding the long-term histologic response to common suture materials. The purpose of the study reported here was to evaluate the histologic reactions to 8 suture materials and CTA in the musculature and skin of ball pythons (*Python regius*) over a period of 90 days. Although there are anatomic and physiologic differences, we hypothesized that the histologic reactions to suture materials in ball pythons would be similar to those that occur in homeothermic vertebrates.

Materials and Methods

Animals—The study protocol was approved by the University of Illinois Institutional Animal Care and Use Committee. Thirty hatchling ball pythons (16 males and 14 females) were obtained from a commercial source^a and housed in conditions approved by the Association for Assessment and Accreditation of Laboratory Animal Care International. The ball pythons were housed in a commercially available rack system specifically designed to house small snakes of this species.^b Mean \pm SD weight of the snakes was 80.2 ± 17.7 g (range, 48 to 117 g), and mean length was 45.3 ± 2.01 cm (range, 40.6 to 49.5 cm). The snakes were provided an ambient daytime temperature range of 25° to 28°C, an ambient night temperature range of 24° to 25°C, a basking temperature range of 32° to 35°C, an environmental humidity range of 50% to 60%, and a 12-hour photoperiod. Every 7 to 10 days, they were offered a thawed frozen mouse of appropriate size for their body diameter. All ball pythons were thoroughly examined on arrival and allowed to acclimate for 7 days prior to initiation of the study.

Anesthesia—To complete the surgical procedures, each snake was anesthetized. Food was not offered to a snake for 48 to 72 hours prior to anesthesia. The ball pythons received butorphanol (1 mg/kg, IM)^c 20 minutes prior to induction of anesthesia. Anesthesia was induced either by means of propofol (10 mg/kg)^d administered via intracardiac injection or by means of direct endotracheal intubation and delivery of 5% isoflurane^e in oxygen. All animals were intubated with endotracheal tubes created from 14- or 16-gauge catheters,^f and anesthesia was maintained with isoflurane in oxygen. Anesthetic depth was monitored by means of ECG and evaluation of the righting reflex, heart rate, and response to a painful stimulus (tail pinch). Positive pressure ventilation was manually performed in snakes with respiratory rates < 1 breath/min. Each ball python was placed on a circulating water blanket set to 40.5°C to maintain its body temperature within the optimal temperature zone. Each snake was placed in ventral recumbency, and the dorsum was prepared for aseptic surgery with alternating washes of 2% chlorhexidine solution and sterile saline (0.9% NaCl) solution. A

sterile plastic fenestrated drape^g was centered over the dorsal midline of each snake.

Surgery—All surgeries were performed by the same surgeon (MSM). In each snake, ten 1-cm full-thickness skin incisions were made over the epaxial musculature (5 incisions on each side of the dorsal midline [day 0]). Incisions were separated by at least 1 cm. At 8 sites, 1 of 8 suture materials was used. A single simple interrupted suture with 2 square knots was placed in the epaxial musculature deep to each of 7 skin incisions by use of 3-0 polydioxanone,^h 3-0 polydioxanone with triclosan,ⁱ 3-0 poliglecaprone 25,^j 3-0 poliglecaprone 25 with triclosan,^k 3-0 polyglactin 910,^l 3-0 MN,^m or 3-0 CGⁿ suture material. A 2- to 3-mm piece of 3-0 stainless steel suture^o without knots was placed in the epaxial musculature exposed at another site. A single horizontal mattress suture of the same material that was implanted in the muscle was placed in the skin over the implanted suture at each of 7 incisions; 1 skin staple^o was placed over the stainless steel suture. At the ninth skin incision, no suture was implanted in the epaxial musculature and the incision was closed with CTA.^p To apply the cyanoacrylate, tension was placed on the wound edges to expose the underlying tissue, the adhesive was applied to the open wound, and the skin edges were then manually apposed and held in place as the adhesive set. Some excess adhesive that was pushed out of the wound during apposition was wiped away. No suture material was placed in the remaining tenth incision, and it was allowed to heal by second intention (negative control). The location of each suture treatment was identical in all snakes. Overall, each experimental treatment was applied to 30 skin incisions.

During recovery, each snake was maintained on a circulating water blanket and heart rate, respiratory rate, righting reflex, and response to painful stimuli (tail pinch) were monitored. If the snake's respiratory rate was < 1 breath/min, positive pressure ventilation was manually performed. When each snake was spontaneously breathing and had regained the righting reflex, it was returned to its enclosure.

The snakes were randomly assigned to 6 groups of 5 snakes. The groups of snakes were euthanized with pentobarbital sodium^q (100 mg/kg administered intracoelomically) at days 3, 7, 14, 30, 60, or 90. Skin sutures were removed at day 42 in all snakes that had not been euthanized.

Histologic examination of skin and muscle samples—Immediately following euthanasia, the dorsal skin and underlying muscle (to the level of the body wall) of each snake were harvested, and sections containing sutures (or scars if sutures were no longer in place) were fixed in neutral-buffered 10% formalin. Tissues were trimmed, routinely processed, embedded in paraffin, and serially sectioned at 5 μ m. Sections were stained with H&E stain and evaluated via light microscopy. All histologic evaluations were performed by a single pathologist (MJK) with extensive experience in exotic animal pathology who was unaware of the type of suture used and postoperative interval for each sample. The sections were examined and assigned a subjective score for each of the following: overall severity

of inflammation (IS), fibrosis (FS), number of macrophages (MS), number of granulocytes (GS), number of perivascular lymphocytes (PLS), and degree of suture fragmentation (SFS). The severity of each of the aforementioned variables was rated on an ordinal scale from 0 (virtual absence of inflammatory cells, fibrosis, or fragmentation) to 4 (most severe reaction or fragmentation observed). Sections were also evaluated for the presence or absence of an organized granuloma, bacteria, or foreign material (excluding suture material). For purposes of analysis, presence or absence of bacteria or granuloma formation in a section was converted into a numeric assessment value (1 or 0, respectively); for a given treatment, the section assessment values were summed to provide an overall value for comparisons.

Statistical analysis—A general linear model was used for the multivariate analysis to evaluate the effect of suture and the pathological changes over time. The Kruskal-Wallis test was used to determine whether there were differences in the various pathological findings by suture type while controlling for time. When differences were detected, a Mann-Whitney *U* test was used to evaluate direct comparisons between suture types. Statistical software[†] was used to analyze the data. A value of $P \leq 0.05$ was considered significant.

Results

All 30 snakes underwent the surgical procedure (ie, 10 skin incisions, each of which was treated with

Table 1—Significant differences* (*P* values) in IS, MS, GS, FS, and presence or absence of bacteria and granuloma formation determined histologically at incision sites that received experimental treatments (1 treatment/incision [day 0]) with 1 of 8 suture materials (3-0 polydioxanone, 3-0 polydioxanone with triclosan, 3-0 poliglecaprone 25, 3-0 poliglecaprone 25 with triclosan, 3-0 polyglactin 910, 3-0 MN, 3-0 CG, or stainless steel) in 30 ball pythons (*Python regius*), compared with the negative control findings, over all time points (days 3 to 90).

Suture material	IS	MS	GS	FS	Bacteria	Granuloma
Polydioxanone	0.001	0.001	—	0.001	—	0.04
Polydioxanone with triclosan	0.001	0.001	—	0.001	—	0.04
Poliglecaprone 25	0.001	0.001	—	0.001	—	—
Poliglecaprone 25 with triclosan	0.001	0.001	—	0.001	—	—
Polyglactin 910	0.001	0.001	0.001	0.001	0.05	0.002
CG	0.001	0.001	0.001	0.001	—	0.002
MN	0.001	0.001	0.001	0.001	—	—
Stainless steel	0.001	—	—	—	—	—

For each treatment involving suture material, a single simple interrupted suture was placed in the epaxial musculature; a single horizontal mattress suture of the same material that was implanted in the muscle or skin staple was used to close the incision. At days 3, 7, 14, 30, 60, and 90 after treatment, 5 snakes were euthanized and incision sites were excised and examined histologically; IS, MS, GS, and FS were rated on an ordinal scale from 0 (virtual absence of inflammatory cells or fibrosis) to 4 (most severe reaction observed). Sections were also evaluated for the presence or absence of an organized granuloma, bacteria, or foreign material (excluding suture material); presence or absence of bacteria or granuloma formation in a section was converted into a numeric assessment value (1 or 0, respectively), and a summed assessment value was calculated for treatments for purposes of comparison. At 3, 7, 14, and 30 days, each treatment was performed and evaluated in 5 snakes at each time point. At 60 and 90 days, each treatment was evaluated in 4 snakes because of the death of 1 snake at each time point. At 60 days, a sample of MN was not evaluated because of loss of the suture material. At 90 days, 2 samples of MN and 1 sample of 3-0 poliglecaprone 25 with triclosan were not evaluated because of loss of the suture materials.

*When a difference was present, the score or assessment was significantly ($P \leq 0.05$) higher for the suture material, compared with the negative control finding.
— = Not significant.

Table 2—Significant differences* (*P* values) in IS, MS, GS, FS, and presence or absence of granuloma formation determined histologically at incision sites that received experimental treatments (1 treatment/incision [day 0]) with 1 of 8 suture materials in 30 ball pythons, compared with the negative control findings, at days 7, 14, 30, 60, and 90.

Suture material	Day													
	7			14		30		60			90			
	IS	MS	GS	IS	MS	IS	MS	IS	MS	FS	IS	MS	FS	G
Polydioxanone	—	—	—	—	—	0.01	0.01	0.001	0.001	0.01	0.0001	0.001	—	—
Polydioxanone with triclosan	—	—	—	—	—	—	—	0.007	0.013	—	0.0001	0.001	0.02	0.007
Poliglecaprone 25	—	—	—	0.01	0.01	—	—	0.007	0.03	—	0.0001	0.001	0.02	—
Poliglecaprone 25 with triclosan	—	—	—	—	—	—	—	0.017	0.03	—	0.0001	0.001	—	0.02
Polyglactin 910	0.02	—	0.001	—	—	0.02	0.04	0.003	0.001	0.005	0.0001	0.001	0.002	—
CG	0.008	0.04	—	0.002	0.002	0.001	0.002	0.017	0.03	0.005	0.0001	0.001	—	0.007
MN	—	—	—	0.005	0.03	—	—	0.04	—	—	0.0001	0.002	—	0.04
Stainless steel	—	—	—	0.03	0.03	—	—	—	—	—	0.007	0.004	—	—

There were no significant differences in any score or assessment, compared with the negative control finding, at day 3 after suture implantation. There were no significant differences in GS at time points other than day 7 and no significant differences in FS at days 7, 14, or 30. Significant differences in the assessment of granuloma formation (G) were detected only at day 90.

See Table 1 for remainder of key.

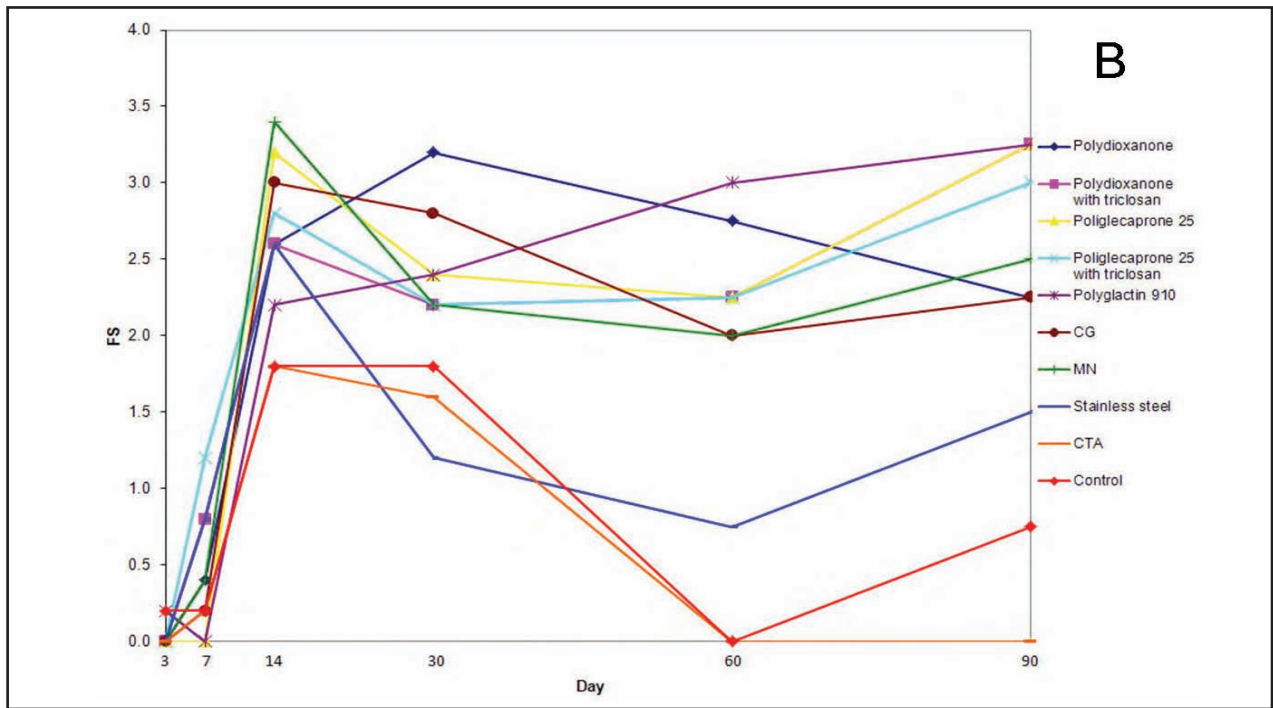
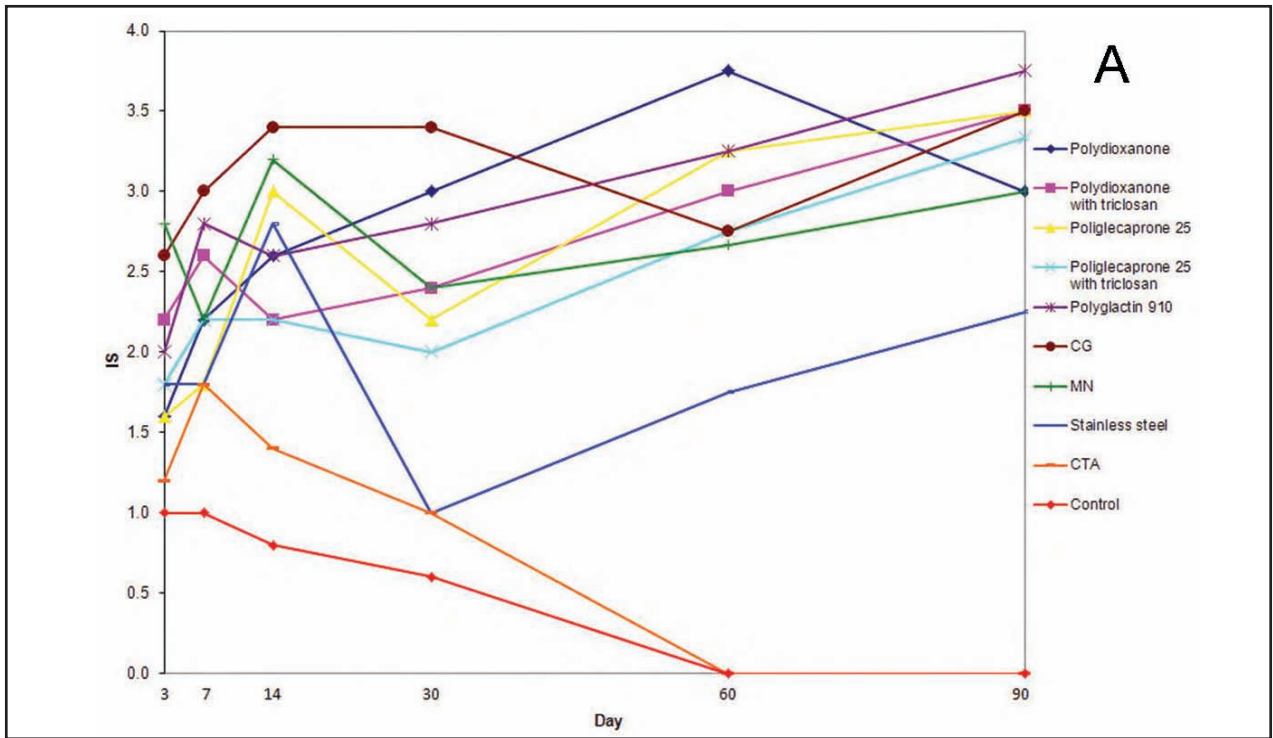


Figure 1—Mean IS (A) and FS (B) determined histologically at incision sites that received experimental treatments (1 treatment/incision [day 0]) with 1 of 8 suture materials (3-0 polydioxanone, 3-0 polydioxanone with triclosan, 3-0 poliglecaprone 25, 3-0 poliglecaprone 25 with triclosan, 3-0 polyglactin 910, 3-0 MN, 3-0 CG, or stainless steel) or CTA or that were untreated (negative control) in 30 ball pythons (*Python regius*). For each treatment involving suture material, a single simple interrupted suture was placed in the epaxial musculature; a single horizontal mattress suture of the same material that was implanted in the muscle was used to close the incision. For the stainless steel treatment, a 2- to 3-mm piece of 3-0 stainless steel suture without knots was placed in the epaxial musculature and the skin was closed with a single skin staple. At 1 site, no suture was implanted in the epaxial musculature and the incision was closed with CTA. Untreated incisions were allowed to heal by second intention. At days 3, 7, 14, 30, 60, and 90 after treatment, 5 snakes were euthanized and incision sites were excised and examined histologically; IS and FS were rated on an ordinal scale from 0 (virtual absence of inflammatory cells or fibrosis) to 4 (most severe reaction observed). For the treatments, differences in ISs were apparent at days 7, 14, 30, 60, and 90, and differences in FSs were apparent at days 60 and 90, compared with the negative control. At 3, 7, 14, and 30 days, each treatment was evaluated in 5 snakes at each time point. At 60 and 90 days, each treatment was only evaluated in 4 snakes because of the death of 1 snake at each time point. At 60 days, 1 sample of MN was not evaluated because of the loss of the suture material. At 90 days, 2 samples of MN and 1 sample of 3-0 poliglecaprone 25 with triclosan were not evaluated because of loss of the suture materials.

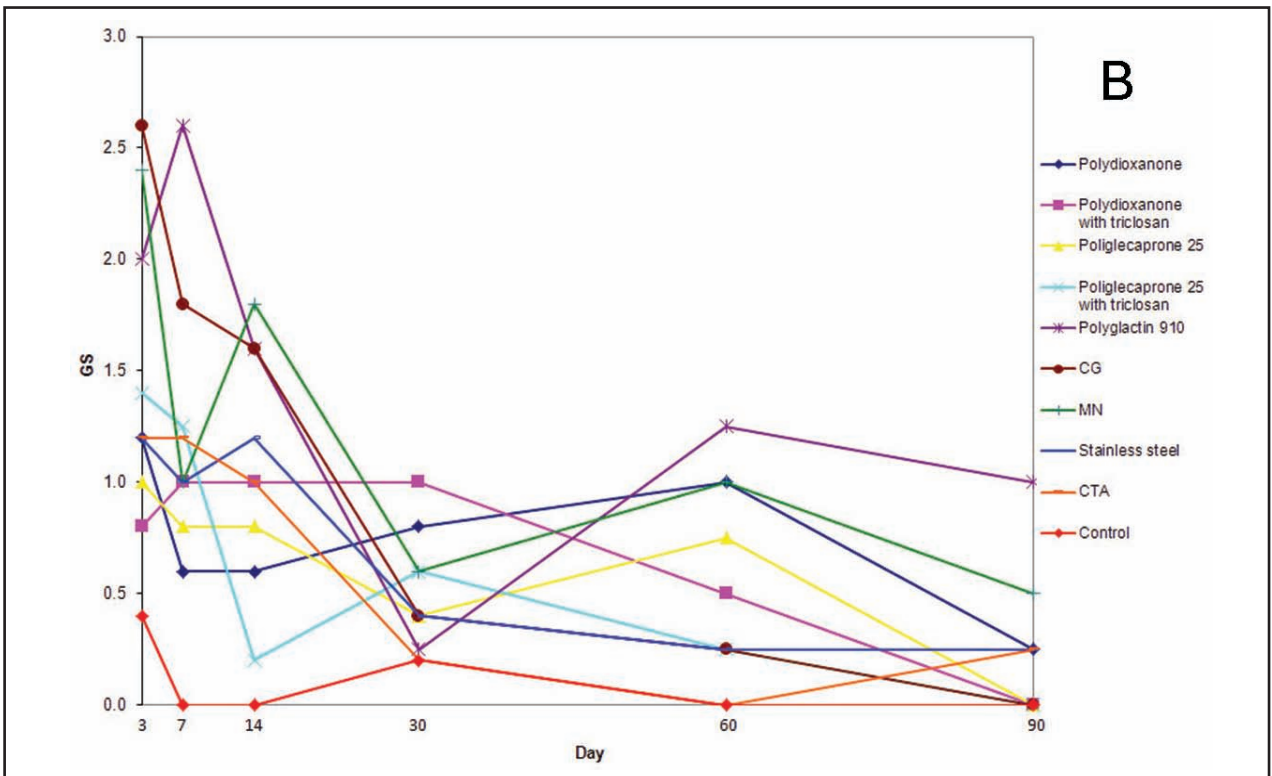
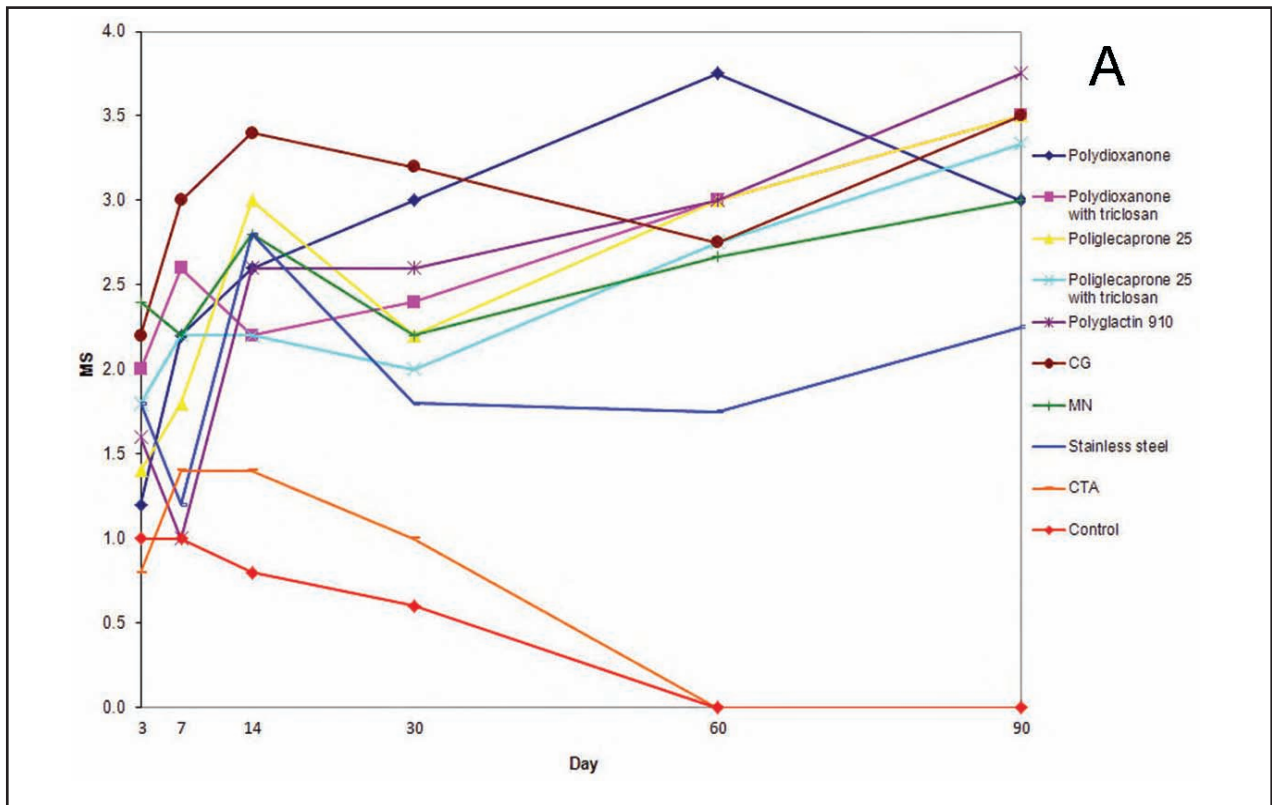


Figure 2—Mean MS (A) and GS (B) determined histologically at incision sites that received experimental treatments (1 treatment/incision [day 0]) with 1 of 8 suture materials or CTA or that were untreated (negative control) in 30 ball pythons. The MS and GS were rated on an ordinal scale from 0 (virtual absence of inflammatory cells) to 4 (most severe inflammatory cell response observed). For the treatments, differences in MSs were apparent at days 7, 14, 30, 60, and 90, and differences in GSs were apparent at day 7, compared with the negative control. At 3, 7, 14, and 30 days, each treatment was evaluated in 5 snakes at each time point. At 60 and 90 days, each treatment was only evaluated in 4 snakes at each time point because of the death of 1 snake. At 60 days, a sample of MN was not evaluated because of loss of the suture material. At 90 days, 2 samples of MN and 1 sample of 3-0 poliglecaprone 25 with triclosan were not evaluated because of loss of the suture materials. See Figure 1 for remainder of key.

placement of 1 of 8 suture materials in the epaxial muscle prior to skin closure with the same suture material, untreated prior to closure with CTA, or untreated and allowed to heal by second intention) and recovered from anesthesia without complication. Placement of sutures was completed in all instances; however, following application of the CTA onto the underlying tissues, some of the CTA was expelled from the wounds as the skin edges were apposed. Initially, 5 snakes were allocated to each of the 6 groups (ie, groups that would be euthanized for harvest of incision site tissues at days 3, 7, 14, 30, 60, and 90).

Two pythons were found dead in their cages during the study period; 1 snake in the group assigned to be euthanized at day 60 was found dead 48 days after surgery, and 1 snake in the group assigned to be euthanized at day 90 was found dead 70 days after surgery. Because those snakes did not reach their predetermined euthanasia time point and substantial tissue autolysis had developed between time of death and time of discovery of the dead snakes, necropsies were not performed and data for those snakes were not included in the analysis.

Complications or adverse effects associated with the incisions were not noted for any snake; all incisions healed in the 30-, 60-, and 90-day groups, and all snakes in those groups went through normal ecdysis cycles for pythons of this species and age. At 3 incision sites closed with MN suture, there was no identifiable suture in the deeper tissues at day 60 (1 site) and at day 90 (2 sites). At 1 incision site closed with poliglecap-

rone 25 with triclosan, there was no identifiable suture in the deeper tissues at day 90. At each of these 4 sites, there was only evidence of a healed skin incision with minimal fibrosis. Because the time from loss of suture material to evaluation was unknown, an accurate IS, FS, MS, PLS, or SFS could not be assigned, and accurate assessment of granuloma formation could not be determined. Thus, all data for the experimental treatments at those 4 sites were excluded from analysis.

Over all time points, there were no significant differences in IS, MS, GS, FS, PLS, or presence or absence of bacteria, granuloma formation, or foreign material between the incisions treated with CTA and the negative control. The IS, MS, GS, FS, PLS, and presence or absence of bacteria, granuloma formation, or foreign material for the 8 suture materials (polydioxanone, polydioxanone with triclosan, poliglecaprone 25, poliglecaprone 25 with triclosan, polyglactin 910, CG, MN, and stainless steel) were compared with findings for the negative control (incision sites with no suture material placement that were allowed to heal by second intention) over all time points (Table 1). When a difference was present, the score or assessment value was significantly ($P \leq 0.05$) higher for the suture type, compared with the negative control finding. There was no significant difference in PLS score or assessment value for foreign material between any treatment and the negative control.

Comparisons among individual suture types over all time points revealed that the IS for the stainless

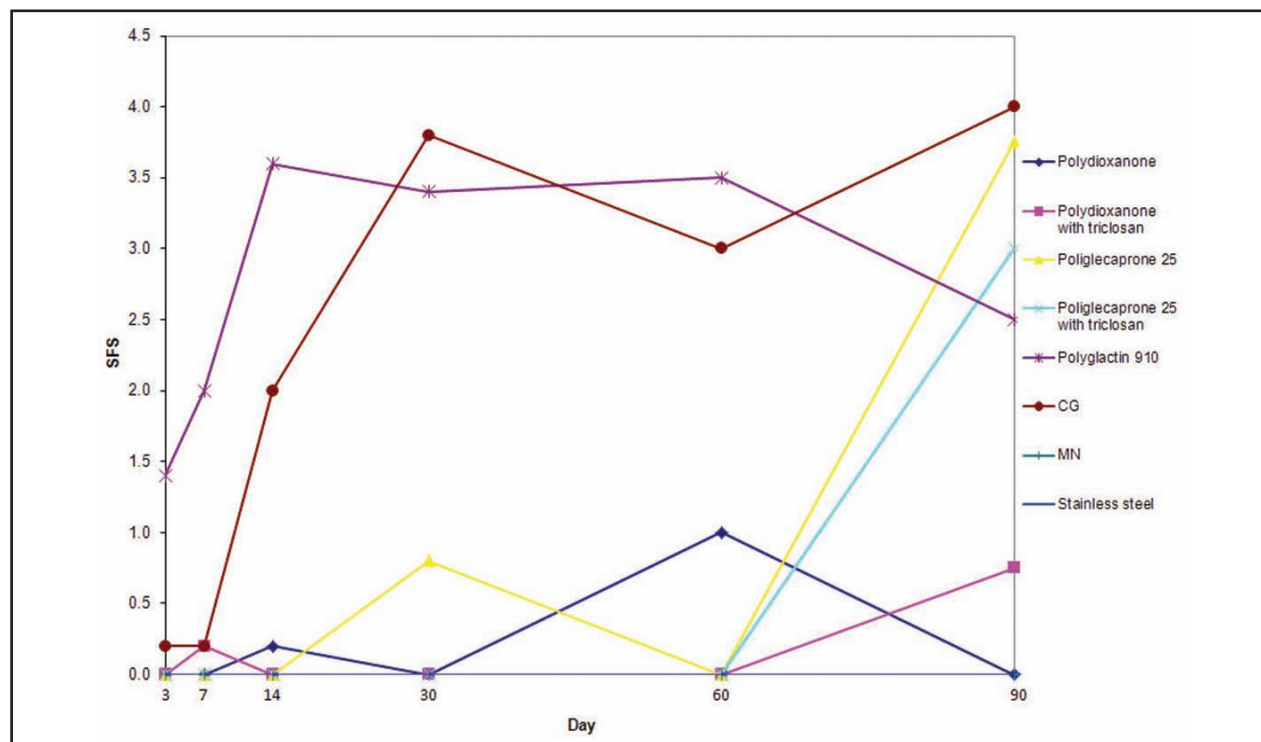


Figure 3—Mean SFS determined histologically at incision sites that received experimental treatments (1 treatment/incision [day 0]) with 1 of 8 suture materials in 30 ball pythons. At day 90, the SFSs for poliglecaprone 25 and poliglecaprone 25 with triclosan differed significantly from the scores for polydioxanone and polydioxanone with triclosan. At 3, 7, 14, and 30 days, each treatment was evaluated in 5 snakes at each time point. At 60 and 90 days, each treatment was only evaluated in 4 snakes because of the death of 1 snake at each time point. At 60 days, a sample of MN was not evaluated because of loss of the suture material. At 90 days, 2 samples of MN and 1 sample of 3-0 poliglecaprone 25 with triclosan were not evaluated because of loss of the suture materials. See Figure 1 for remainder of key.

steel sutures was significantly lower than the ISs for the polyglactin 910 sutures ($P = 0.03$) and CG ($P = 0.001$) sutures. The IS for the poliglecaprone 25 with triclosan sutures was significantly ($P = 0.003$) lower than the score for the CG sutures. Macrophage score was also significantly ($P = 0.001$) lower for the stainless steel sutures, compared with the MS for the CG sutures. Granulocyte score was significantly higher for the polyglactin 910 sutures, compared with the GSs for the poliglecaprone 25 sutures ($P = 0.03$) and the poliglecaprone 25 with triclosan sutures ($P = 0.002$). The assessment value for granuloma formation was significantly ($P = 0.04$) higher for the CG sutures, compared with the finding for the stainless steel sutures.

At each time point, there were no significant differences in IS, MS, GS, FS, PLS, or presence or absence of bacteria, granuloma formation, or foreign material between the incisions treated with CTA and the negative control. The IS, MS, GS, FS, PLS, and presence or absence of bacteria, granuloma formation, or foreign material for the 8 suture materials (polydioxanone, polydioxanone with triclosan, poliglecaprone 25, poliglecaprone 25 with triclosan, polyglactin 910, CG, MN, and stainless steel) were compared with findings for the negative control sites at days 3, 7, 14, 30, 60, and 90 (Table 2). When a difference was present, the score or assessment value was significantly ($P \leq 0.05$) higher for the suture type, compared with the negative control finding. There was no significant difference in PLS score and the bacteria and foreign material assessment values between treatments and the negative control. There were no significant differences for any score or assessment value, compared with the negative control findings, at 3 days after suture implantation (data not shown).

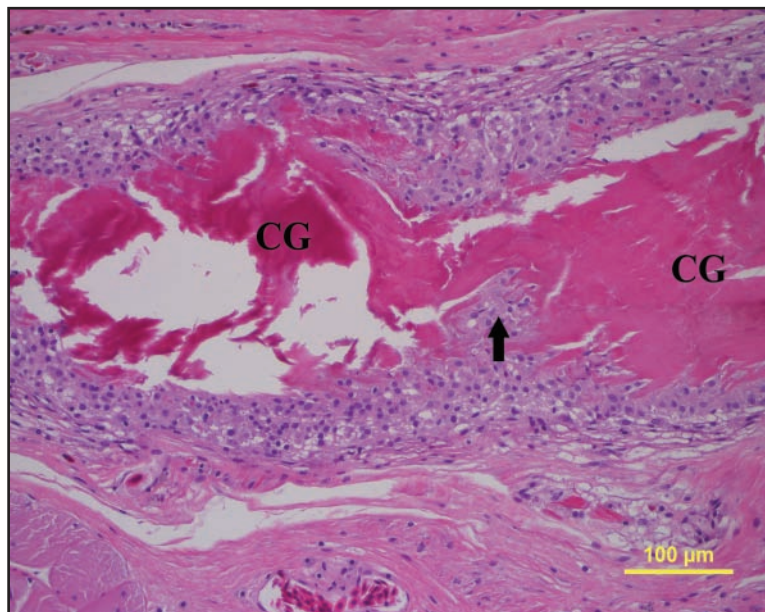


Figure 4—Photomicrograph of a tissue section obtained from an incision site that was implanted with a CG suture and closed by use of the same suture material in a ball python. The suture (CG) in the musculature has induced a severe inflammatory response (IS = 4 [scale of 0 to 4]), and there is evidence of suture fragmentation (SFS = 4 [scale of 0 to 4]). An organized granuloma is present around the suture, and macrophages are invading clefts within the suture tract (arrow). H&E stain; bar = 100 μ m.

The mean ISs, FSs, MSs, and GSs for each of the 10 experimental treatments over time were compared (Figures 1 and 2). The only significant ($P = 0.03$) difference among the experimental treatments when controlling for time was in the GS for polyglactin 910, which was higher than the GS for polydioxanone at day 7 following suture implantation.

Mean SFS for CG suture was significantly ($P = 0.001$) higher, compared with the 7 other suture materials except polyglactin 910, from 3 to 60 days (Figure 3). Suture fragmentation score for CG was evident by infiltration of inflammatory cells within the suture tracts (Figure 4). The SFSs for polyglactin 910 suture were also significantly ($P = 0.001$) higher than the SFSs for all other experimental treatments except CG over that same time period. At 90 days, the SFS for poliglecaprone 25 and poliglecaprone 25 with triclosan increased from the day 60 values. At 90 days, the degree of suture fragmentation for poliglecaprone 25 and poliglecaprone 25 with triclosan was significantly different from findings for polydioxanone ($P = 0.02$) and polydioxanone with triclosan ($P = 0.04$), but not different from findings for CG ($P = 0.3$) or polyglactin 910 ($P = 0.07$). Poliglecaprone 25 and poliglecaprone 25 with triclosan also had significantly ($P = 0.01$) higher SFSs than the values for MN and stainless steel sutures at 90 days. Polydioxanone and polydioxanone with triclosan had minimal suture fragmentation at any time point.

Although there were no significant differences in assessment values for foreign material between treatments and the negative control or among treatments, foreign material was detected in some sections. Keratin fragments were present in 2 sites treated with CG suture (at days 30 and 90), 1 site treated with poliglecaprone 25 suture (at day 3), 1 site treated with polyglactin 910 suture (at day 14), 1 site treated with stainless steel suture (at day 90), and 1 site treated with CTA (at day 30). A fragment of epidermis was present in 1 site treated with polyglactin 910 suture (at day 14). Plant material (from paper towels) was present in 1 site treated with CG suture (at day 14) and 1 site treated with MN suture (at day 60).

Discussion

Suture material is a foreign body in a surgical wound and can potentiate infection and inflammation.¹⁹ The nature and extent of the reaction are dependent on many variables, including the nature of the suture material, the tissue into which the suture is implanted, interval since implantation, and the amount of suture to which the tissue is exposed. Surface area can greatly increase foreign material-to-tissue contact in multifilament suture.^{9,10} Once implanted, there are various final outcomes for the foreign material. Some sutures, particularly skin sutures, are ultimately removed. Suture material that is implanted in deeper tissues will undergo absorption via hydrolysis or phagocytosis, extrusion by the body, or incor-

poration in a fibrous capsule.⁹ These outcomes ultimately depend on the type of suture material.

Chromic gut is an absorbable suture material composed of purified connective tissue (mostly collagen) derived from either bovine or ovine intestines. It is absorbed via phagocytosis and enzymatic digestion by macrophages.^{8,9} Chromic salts are used to coat the suture to help delay absorption, increase tensile strength, and decrease tissue reactivity. Chromic gut suture is completely absorbed in approximately 60 to 90 days following implantation in mammals. Results of previous studies^{11,12,14,15,17,18,20} in many species have indicated that CG suture causes a more severe inflammatory response, compared with other commonly used suture materials. In the present study, CG suture caused an early and sustained inflammatory response, migration of macrophages to the surgical site, and increased formation of granulomas and fibrosis, compared with the findings for the negative control (incision sites with no suture material placement that were allowed to heal by second intention). It also caused more inflammation, had a higher MS, and had a higher incidence of organized granuloma formation, compared with findings for stainless steel suture material, and caused more inflammation than did poliglecaprone 25 with triclosan. The early and sustained inflammatory response was expected on the basis of the results of previous studies; however, there is some debate about the use of CG suture in reptiles. In another study,²¹ intact CG suture material was detectable 12 weeks (84 days) following implantation in a rhinoceros viper (*Bitis nasicornis*). The assumption was that snakes may not have a mechanism for absorbing CG suture because of a lack of appropriate proteolytic enzymes in reptilian macrophages. Chromic gut suture was still present in tissues at 90 days following implantation of suture materials in the present study, but when the suture materials were assessed for fragmentation, CG had significantly higher mean SFS, compared with values for all other suture materials, with the exception of polyglactin 910. Organized granulomas were present around samples of CG with macrophages invading clefts within the suture material. This suggests that reptile macrophages are capable of phagocytosis of CG, but that the time required for complete absorption is longer than that in mammals. This is supported by results of a study¹¹ on suture reaction in birds, which indicated that CG suture was still intact 120 days following implantation.

Polyglactin 910 is a multifilament suture material composed of a copolymer of lactic acid and glycolic acids that are coated with calcium stearate and a second copolymer of glycolide and lactide. Absorption of polyglactin 910 is through hydrolysis, and complete absorption in mammals takes approximately 60 days.^{8,9,22} In the present study, there was an acute reaction to polyglactin 910, which resulted in significantly more overall inflammation over all time points and an increased GS at day 7 following suture implantation, compared with the negative control findings. Polyglactin 910 also caused significantly more inflammation than did stainless steel suture and a significant increase in GS, compared with poliglecaprone 25 and poliglecaprone 25 with triclosan, over all time points and compared with

polydioxanone at day 7 only. After day 7, there was a decrease in IS and MS for polyglactin 910 at 14 days, then a gradual increase in IS and MS at 30, 60, and 90 days following suture implantation. The multifilament nature of polyglactin 910 increases the surface area of suture exposed to tissue and may explain the difference in inflammatory responses and increase in the number of granulocytes at the site of implantation. Our findings were similar to those of other studies in nonmammalian species in which polyglactin 910 was found to cause a substantial inflammatory response in rock doves,¹¹ koi,¹⁴ and loggerhead sea turtles.¹³

The SFSs for polyglactin 910 also differed significantly from SFSs for all other suture materials, except for CG, at days 3 to 60. Similar to CG, the SFS for polyglactin 910 at day 90 indicated that the suture material was in the process of breaking down, but that the time to complete absorption is longer in snakes than it is in mammals. Polyglactin 910 was the only suture material that differed significantly from the negative control with regard to the presence of bacteria over all time points. Capillarity of multifilament sutures leads to movement of fluid and bacteria into the interstices of multifilament fibers. These interstices are too small to be penetrated by inflammatory cells, which allow the bacteria to persist.²²

Monofilament nylon is a polyamide suture material that is nonabsorbable although it does undergo some degradation and loses 20% of its tensile strength/y. In the present study, MN had significantly higher IS and MS, compared with the negative control findings, at day 14 following implantation. At day 30, the IS and MS were decreased from the day 14 values, but there was a gradual increase in each score at days 60 and 90. When used in koi, the reaction at 7 and 14 days after implantation of MN was greater than that associated with other synthetic sutures.¹⁴ The exact cause of the increase in inflammation is unknown, but nylon suture buried in tissue has sharp stiff ends that can lead to frictional irritation.⁹

Poliglecaprone 25 and polydioxanone are monofilament, synthetic absorbable polymers. Poliglecaprone 25 and polydioxanone are also available (as is polyglactin 910) impregnated with the antimicrobial triclosan. After implantation in mammals, poliglecaprone 25 is absorbed between 90 and 100 days, and polydioxanone is absorbed at approximately 180 days.⁸ In the present study, the ISs and MSs for all synthetic absorbable polymers were significantly different from the negative control findings at days 60 and 90. For polydioxanone, the IS and MS were also significantly greater than the negative control values at day 30. This suggests that these sutures lead to a chronic inflammatory response, and any acute inflammation caused by the suture materials themselves was not significantly different than the negative control. All synthetic monofilament absorbable sutures were intact at day 90 with the exception of 1 suture of poliglecaprone 25 with triclosan (no material was found at the incision site). The SFSs were significantly higher for poliglecaprone 25 and poliglecaprone 25 with triclosan, compared with polydioxanone and polydioxanone with triclosan, 90 days following implantation. This suggests that the breakdown of poliglecaprone 25 in reptiles begins between 60 and

90 days. There was minimal evidence of fragmentation of polydioxanone or polydioxanone with triclosan at any time point. Overall, the reaction to the synthetic absorbable polymer suture material was a chronic inflammatory response that generally increased from days 30 to 90 after implantation. Prolonged absorption times may have led to a chronic inflammatory response, or the duration of the present study may not have been sufficient to detect decreases in inflammatory response over longer periods as the sutures were slowly broken down.

Stainless steel is the strongest yet least reactive suture material.^{8,9} In the present study, there was significantly less inflammation with steel, compared with polyglactin 910 and CG. Compared with the negative control findings, the IS and MS for stainless steel suture were significantly different at days 14 and 90 following implantation. The reaction to the stainless steel suture may be associated with its stiffness, which can cut adjacent tissue and lead to necrosis of tissue near the suture ends.^{8,9}

The n-butyl CTA^p that we used polymerizes in the presence of moisture, produces a strong flexible bond, is nonabsorbable, and can lead to granuloma formation and fistulization.⁸ In the present study, the CTA caused the least inflammatory reaction. There was no significant difference between the CTA and negative control findings for any variable evaluated, and the adhesive caused significantly less inflammation than the various suture materials. The lack of a response may have been associated with the method of application. During application, tension was placed on the wound edges and the CTA was placed directly onto the underlying tissues. This was done to ensure that the underlying musculature and subcutaneous tissues were exposed to the adhesive. As the skin edges were apposed, some of the CTA was expelled from the wound. This ultimately led to little contact between the CTA and the underlying tissues. Because the volume of foreign material can affect the reaction, the small volume of CTA in the subcutaneous tissues may have resulted in the limited inflammatory response that we detected, although a true difference in reactivity is possible because little reaction was evident in the tissues that were in contact with the CTA.

Interestingly, there were 3 sites treated with MN suture and 1 site treated with poliglecaprone 25 with triclosan suture in which no suture material was present in the tissues at day 60 or day 90 after implantation. In all samples, all tissues under the skin incisions to the level of the body wall were submitted for evaluation, and all submitted tissues were trimmed and serial sections examined. For the MN-treated sites, 1 site evaluated at day 60 and 2 sites evaluated at day 90 had no suture material in the deeper tissues and only fibrosis and evidence of a healed skin incision. Similar findings were noted at day 90 for the 1 site treated with poliglecaprone 25 with triclosan suture. Our goal was to evaluate the tissue reaction to suture material, and in the absence of suture material in the incision site, a score of the surrounding tissue would not be an accurate assessment of the reaction to the suture. Therefore, data for these sites were excluded from the statistical analysis.

The ultimate outcome for these sutures was believed to be absorption, extrusion, or migration. Because MN is nonabsorbable, absorption of those 3 sutures was not likely. Absorption of the poliglecaprone 25 with triclosan suture at 90 days was possible because there was a substantial increase in SFS for poliglecaprone 25 with triclosan from day 60 to day 90. However, the site in which no suture was found only had a small area of fibrosis with no residual suture or suture tract; if this suture had been completely absorbed, some residual inflammatory reaction would be expected. Migration of MN or poliglecaprone 25 with triclosan that was sutured to the underlying musculature was not likely, especially because adjacent sections of tissues with other suture types showed no evidence of migration. It seemed most likely that these sutures were extruded at the 4 sites.

Suture extrusion occurs when a foreign body irritant is moved through the tissues toward the skin surface and expelled.²³ Extrusion of subcuticular sutures is problematic in human surgery. Factors affecting extrusion of subcuticular sutures in pigs have been investigated,²³ and it appears that knot volume, mechanical motion, suture type, and elapsed time from implantation all contribute to suture extrusion. Suture extrusion from deeper tissues has also been reported.^{24,25} Suture extrusion in reptiles has been reported.¹³ In that study, full-thickness simple interrupted sutures were used to close skin defects; the investigators reported that sutures were extruded by 14 days, but did not suggest potential mechanisms or provide support for extrusion versus suture breakage or dehiscence. In the present study, sutures that were placed in deep tissue with the skin closed over them appeared to have been extruded. In addition to the sites in which no suture was present, granulomas that appeared to be contiguous with the skin were detected in association with other suture materials at different time points, as if the suture was in the process of being extruded from the underlying tissues.

In the present study, all experimental treatments caused significantly more inflammatory changes than the negative control treatment. The CTA was the least reactive, compared with the suture materials, but this may have been due to limited tissue contact. The only significant difference among suture types at different time points was that polyglactin 910 caused more granulocytes to migrate into the wound, compared with polydioxanone at day 7 following implantation. Over all time points, polyglactin 910 and CG caused more inflammatory changes, compared with other suture types. All suture materials, but not CTA, appeared to cause a chronic inflammatory response, as determined by changes in the assessed variables. For several suture materials, inflammation appeared to increase between days 30 and 90. Unfortunately, we did not extend the study to include time points > 90 days to determine whether the chronic inflammation persisted until absorption or expulsion of the suture or subsided once the suture material was encapsulated in a granuloma. All types of suture material were still present at day 90 following implantation, including suture types that are typically completely absorbed in mammals over a

similar period. Further studies in reptiles to evaluate the long-term effects of suture materials after 90 days would help determine absorption times (if complete absorption occurs) and evaluate whether sutures are extruded from tissues.

The results of the present study indicated that small superficial skin incisions or lacerations in reptiles can be closed with CTA. The evidence that absorption of suture material is slower than that in mammals and that extrusion of the suture may occur suggests that use of suture materials that are rapidly absorbed may be more appropriate in reptiles than use of suture materials that are nonabsorbable or may take several months or possibly years to be absorbed (eg, polydioxanone). Of the suture materials evaluated, poliglecaprone 25, poliglecaprone 25 with triclosan, and CG, which have the shortest times to complete absorption in mammals, all showed evidence of fragmentation within the 90-day study period. Another rapidly absorbed suture material is a variation of polyglactin 910,⁵ which is completely absorbed in approximately 40 days in mammals.

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- a. Ben Siegel Reptiles, Deerfield Beach, Fla.
 - b. PVC Cages, Grand Haven, Mich.
 - c. Fort Dodge Animal Health, Fort Dodge, Iowa.
 - d. Abbott Laboratories, North Chicago, Ill.
 - e. IsoFlo, Abbott Laboratories, North Chicago, Ill.
 - f. Angiocath, Becton-Dickinson, Sandy, Utah.
 - g. Large Surgical Drape, Veterinary Specialty Products, Mission, Kan.
 - h. PDS, Novartis Animal Health, Greensboro, NC.
 - i. PDS Plus, Novartis Animal Health, Greensboro, NC.
 - j. Monocryl, Novartis Animal Health, Greensboro, NC.
 - k. Monocryl Plus, Novartis Animal Health, Greensboro, NC.
 - l. Vicryl, Novartis Animal Health, Greensboro, NC.
 - m. Ethilon, Novartis Animal Health, Greensboro, NC.
 - n. Novartis Animal Health, Greensboro, NC.
 - o. Reflex One, Conmed, Utica, NY.
 - p. Vetbond, 3M, Saint Paul, Minn.
 - q. Sleepaway, Fort Dodge, Madison, NJ.
 - r. SPSS, version 16.0, SPSS Inc, Chicago, Ill.
 - s. Vicryl Rapide, Novartis Animal Health, Greensboro, NC.
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