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Laboratory Reptile Surgery: Principles and Techniques

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Reptiles used for research and instruction may require surgical procedures, including biopsy, coelomic device implantation, ovariectomy, orchidectomy, and esophogostomy tube placement, to accomplish research goals. Providing veterinary care for unanticipated clinical problems may require surgical techniques such as amputation, bone or shell fracture repair, and coeliotomy. Although many principles of surgery are common between mammals and reptiles, important differences in anatomy and physiology exist. Veterinarians who provide care for these species should be aware of these differences. Most reptiles undergoing surgery are small and require specific instrumentation and positioning. In addition, because of the wide variety of unique physiologic and anatomic characteristics among snakes, chelonians, and lizards, different techniques may be necessary for different reptiles. This overview describes many common reptile surgery techniques and their application for research purposes or to provide medical care to research subjects.

When considering animals used in research and teaching, the tendency might be to overlook the less common species, such as reptiles. However, reptiles are used frequently in research and teaching. Here we describe many common surgical procedures for reptiles that may be used for research or medical purposes. Some of the more frequently used species include the genus *Anolis*, typically *A. carolinensis*, the green anole, but also *A. sagrei*, the brown anole. Anoles mainly are used as a model of behavioral neuroendocrinology, and studies investigate reproductive behavior, sexual dimorphism, dermal chromatophores, melatonin, and sex steroid hormones. [4,19,37,52,68,85,102,104,105,109] Other examples include species such as the parthenogenic whiptail lizard, *Cnemidophorus uniparens*, a subject of neuroendocrinology studies.[37] The chemosensory function of garter snakes, genus *Thamnophis*, has been studied in its relation to reproduction and feeding behaviors.[37] The green iguana, *Iguana iguana*, often is used for physiologic studies.[37] Red-eared sliders, *Trachemys scripta elegans*, and their relatives, *Chrysemys* turtles, are used in respiratory physiology research because of their apparent resistance to anoxia.[49,101,111-113] Lately, reptiles have been used as environmental biomonitors, particularly because they are sensitive to contaminants, and bioaccumulate or biomagnify contaminants.[20,95] Many free-ranging species, such as the American alligator, *Alligator mississippiensis*, have been used to assess conditions in their environments.[16,17,38,39,108] Reptiles have been investigated for their production of unique substances. Many species of venomous snakes have been used to explore the platelet aggregation inhibitors found in their venom.[21,56,61,62,66] Recently, the saliva of the Gila monster, *Heloderma suspectum*, has rendered a glucagon-like peptide 1, which improves glucose homeostasis in patients with type 2 diabetes.[12,103] Learning has been studied in reptiles. For example, crocodilians have been taught complex tasks,[37] and the spatial learning and memory of a *Geochelone carbonaria* tortoise recently was tested by use of a radial maze and was found to have abilities comparable to those of mammals.[110] Reptiles frequently are used in instruction, such as in natural history exhibits and in biology or ecology classes.

The Public Health Service Policy and the *Guide for the Care and Use of Laboratory Animals*[53] contain guidelines for reptiles used in research and teaching. Therefore, reptile care and use is regulated by the Office of Laboratory Animal Welfare in all institutions that receive NIH funding. The *Animal Welfare Act*[1,2] does not cover reptiles. However, collection and holding of free-living or venomous reptiles may require local, state, or federal permits.

Here we describe various reptile surgical techniques and their application for research purposes and to provide medical care to research subjects. The complexity and breadth of reptile surgery necessitates our incorporation of only the most important factors and most common procedures. Surgical techniques in reptiles can be challenging, due to the wide variety of unique anatomic and physiologic characteristics.[45] Most reptiles lack a diaphragm and have a single coelom, intracoelomic testes, a common cloaca, and so forth. In addition, reptiles have a reduced metabolism, which is approximately 25% to 35% of that of mammals of comparable size.[29] Reptiles are ectothermic and rely on specific temperature ranges for normal physiologic function,[29] including anesthesia metabolism and healing, which are approximately 7 times slower than for a comparable sized mammal. In mammalian healing, the maximum deposition of repair collagen occurs at 2 to 3 wk, and suture removal is recommended at 7 to 10 d.[41] In reptiles, suture removal is recommended at 6 or more weeks after surgery.[71] We recommend that laboratory animal veterinarians and research surgeons first review the anatomy, physiology, and natural history of the specific reptilian species prior to surgery. Knowledge of specific anatomic considerations and unique aspects of patient preparation, positioning, and equipment can facilitate planning and success with surgical procedures. We also recommend several previous publications (specifically references 5, 67, and 70) for further descriptions of these surgical procedures.

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Surgical Equipment

Surgical instrumentation. The need for specialized surgical instruments will depend on the size of the species. For reptiles weighing 5 to 50 kg, most small animal instruments are appropriate. Smaller instruments that are appropriate for mice and hamsters will be sufficient in many situations. However, most exotic patients are less than 5 kg in size, and the majority is less than 1 kg and share general considerations for small-bodied animals. For these small reptiles, microsurgical instruments with their fine, small tips are the optimal instrumentation for this size of patient.

Because one of the most important considerations in surgery is access, retractors must be of the appropriate size and weight for these small animals. Plastic, self-retaining retractors (for example, Lone Star retractor, Lone Star Medical Products, Stafford, TX) can be adjusted to fit different sizes of incisions and do not compromise the ventilation of these patients (Figure 1). Smaller versions of standard abdominal retractors (for example, pediatric Balfour retractors, Haight baby rib spreaders) can be used but are considerably heavier than plastic self-retaining retractors. Figure 2 lists suggested equipment for general surgical packs.

If orthopedic work is anticipated, a variety of surgical drills and saws should be available. Autoclavable or gas-sterilizable models are preferred. For general orthopedic work, the Stryker drill (Stryker, Kalamazoo, MI) offers excellent control and versatility, even for the smallest patients. The oscillating saggital saw attachment to the air-powered 3M minidriver (3M, St Paul, MN) provides fine control and reduced tissue trauma compared with rotating saws. A selection of intramedullary pins, miniature fixator pins, aluminum and carbon fiber clamps, and titanium and carbon fiber support rods complete the orthopedic equipment necessary for reptile surgery. Epoxy resins or low-temperature veterinary acrylics are used for chelonian plastron closures and shell repairs. A 2-polymer orthopedic putty is another very useful aid to external fixation.

Modern rapid absorbable suture materials (for example, polyglactin 910, polyglycolic acid) are recommended for internal soft tissue applications in reptiles. For permanent internal durability, polydioxanone or nylon is required. Monofilament nylon and polydioxanone are favored for skin suturing.

Magnification and illumination. For adequate visualization, appropriate lighting becomes even more important in small patients. Therefore, surgical lights that are sufficiently powerful and can be focused to a small region should be used. Some degree of magnification is recommended, and there are a variety of magnification systems available. Headband or frame-mounted operating loupes (magnification, ×2.5) with a dedicated halogen or xenon light source are affordable, versatile, comfortable, and simple to use. Operating microscopes are costly but versatile.

Hemostasis. Generally, a healthy reptile can tolerate between 0.4 to 0.8 mL blood loss per 100 g body weight.[22] Careful consideration must be given to minimizing hemorrhage. Cotton-tipped spears or applicators are less traumatic and more manageable in small confined spaces than are standard gauze squares and allow the surgeon to apply localized pressure to a small vessel and monitor blood loss. Hemoclips are a convenient and effective way to clamp vessels. The Weck system (Weck Closure Systems, Teleflex Medical, Kenosha, WI) uses autoclavable applicators and clips of various sizes. The application of vascular clips is faster than that for standard suture ligatures, decreasing surgical time. Radiosurgery uses high-frequency radiowaves to cause focal thermal tissue damage. Unlike electrocautery, radiosurgery maintains a cool electrode and offers superior accuracy and reduced collateral damage that rivals that of laser incisions.[28]

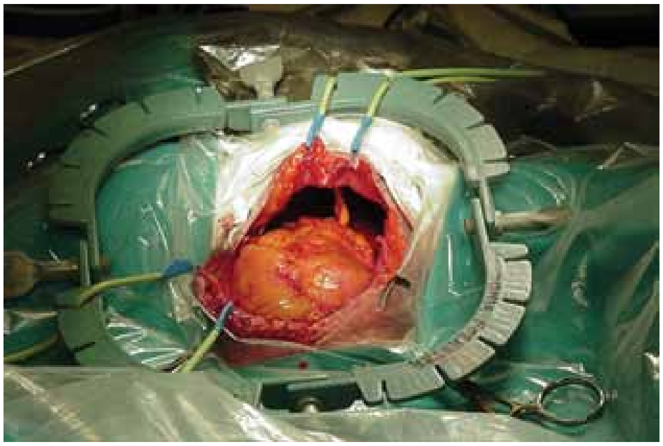

Figure 1. The Lonestar retractor incorporates an adjustable plastic ring to which elastic bands with hooks are attached. This retractor is extremely versatile and works well with a variety of species and surgical procedures.

Preparation of Reptile Surgical Patients

Preparation of reptile surgery patients should include a preanesthetic physical exam, and in some cases, a complete diagnostic workup may be indicated. Reptile subjects must be provided appropriate premedication, and preemptive analgesia is recommended. Inhalant anesthesia is highly recommended, although injectables are commonly used in some reptile species.[10,47,63,93,96] Unlike in mammals, the glottis in reptiles is easy to visualize and access because it is located at the base of the tongue. Reptiles larger than 200 g body weight typically can be intubated with commercially available endotracheal tubes. Smaller species may require ingenuity for fashioning an endotracheal tube from an intravenous catheter, and some species (*Anolis* spp.) may be too small to intubate. We have experience with applying sufficient positive-pressure ventilation to squamates and snakes through a face mask, as long as the seal on the mask is adequate. In some situations, catheterization and application of intraoperative monitoring devices may be indicated.

Appropriate patient positioning will depend on the species and procedure (Figure 3). Considerations for surgical positioning include ensuring that the head and neck position does not interfere with ventilation; avoiding excessive compression of the head, limbs, or coelom to prevent pressure necrosis, visceral rupture, or hypoventilation of the lungs; avoiding extreme and prolonged hyperextension or hyperflexion of any joint; and ensuring that the surgical site is easily accessible and does not require surgeon positioning that will result in fatigue. The use of sandbags, beanbags, foam supports, and adhesive tapes is useful to maintain patient position. In addition, when performing microsurgery, the surgeon's arms and wrists should rest on the surgical table. In most cases, using sandbags or similar objects to support the wrist allows the surgeon the most optimal motor control when incising or suturing delicate tissues.

Presurgical preparation. According to the *Guide*, aseptic surgery should follow established standards, such as surgeons wearing surgical attire and sterile gloves in an appropriately clean room that minimizes unnecessary traffic.[53] The surgical site should be prepared by using chlorhexidine or povidone–iodine concentrate; a sterile toothbrush is particularly useful for cleaning scaled skin. Excessive use of alcohol is not recommended because of increased evaporative heat loss; however,

Standard exotic animal surgery pack	Standard exotic animal microsurgery pack
Plain ophthalmic fine thumb forceps	Mini Gelpi retractor
Adson forceps (1×2 teeth, very fine)	Avian retractor
Small scissors (top, sharp tip; bottom, blunt tip)	K-wire pin driver vice
Castroviejo retractor	Doolen avian bone-holding device
Small suture scissors	Stevens tenotomy scissors
Derf needle holder	2 balanced micro scissors
Strabismus scissors	Doolen Avian spay hook
4 curved ophthalmic mosquito forceps	Extra delicate mosquito forceps, straight
20 gauze sponges (3 x 3 in.)	Extra delicate mosquito forceps, curved
20 cotton tipped applicators	Sontec curved tying forceps
Clear plastic adhesive drape	Ring-tipped thumb forceps with holes
Elastic bandage (2 in.)	2-spring Bulldog clamps
No. 15 scalpel blade	Lonestar retractor

Figure 2. Suggested equipment for surgical packs.

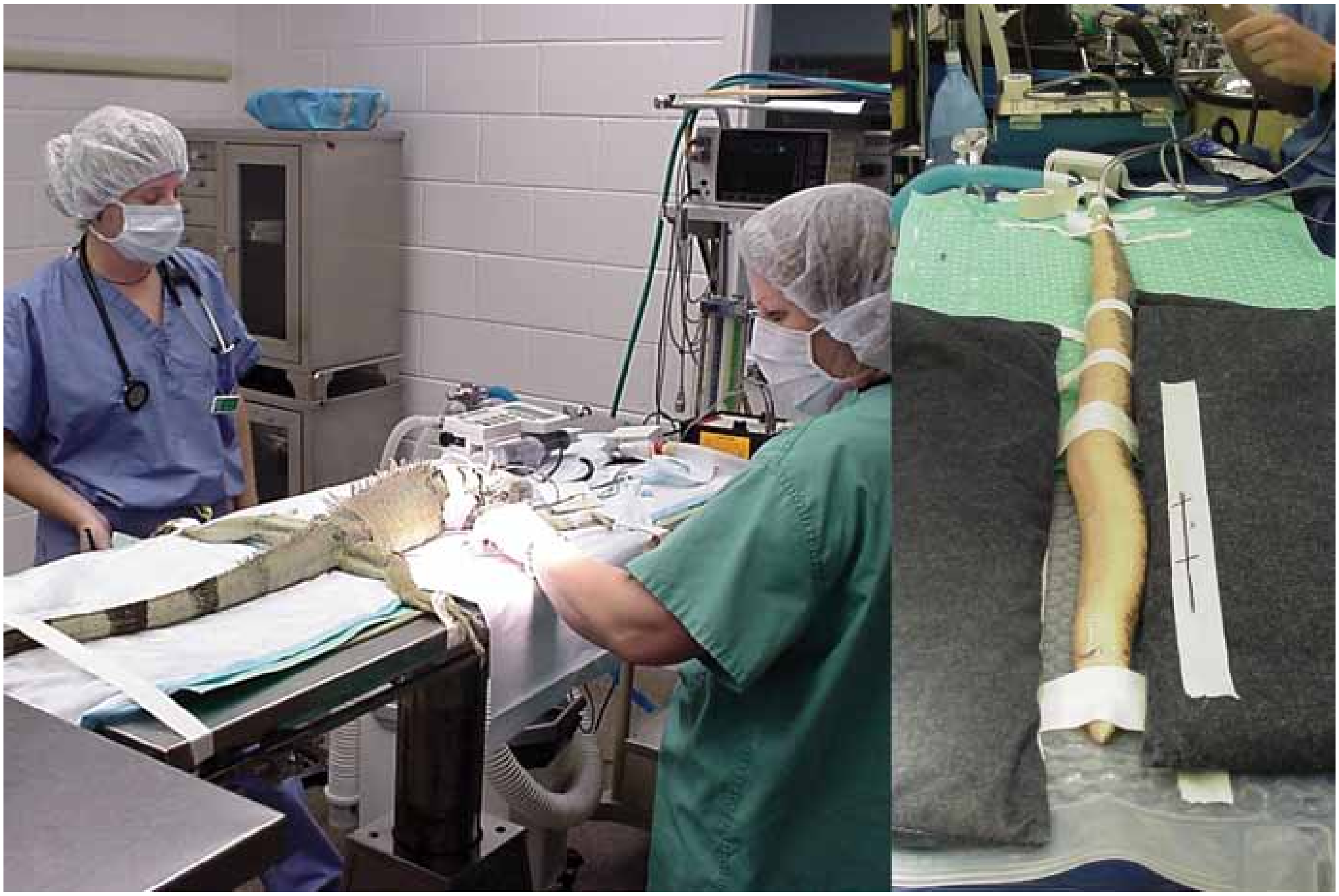

Figure 3. Surgical positioning of lizards and snakes. Left, Sternal positioning of a green iguana in preparation for spinal surgery. Right, Dorsolateral positioning of Kenyan sand boa in preparation for coeliotomy and salpingotomy; note the tape indicating the surgical site.

a final alcohol wipe will ensure a dry, grease-free area to which adhesive drapes will readily stick. Transparent, adhesive drapes have several advantages over cloth drapes, including better visualization of the patient, maintenance of a waterproof barrier, lack of need for towel clamps, and light weight.

Intra- and Postoperative Care

To maximize surgical success, fluid administration during reptile surgery is standard. The standard rate for intraoperative fluid is 3 mL/kg hourly; however, this amount can be modified to account for differences in metabolism (that is, small lizard compared with giant tortoise) and the preoperative condition of the patient. Ideally, catheters are placed before surgery, and fluids are administered parenterally by the intravenous or intraosseous route.[71] Intraosseous catheterization is used most commonly, because this method requires relatively little technical skill and is easy to maintain. An alternative is intraoperative catheterization of the midline abdominal vein or mesenteric veins to administer fluids during the surgical procedure, but this method does not support administration of fluids once the procedure is completed.

The physiology of reptiles is entirely temperature-dependent. Therefore, all metabolic activites, including anesthetic drug metabolism, will rely on maintaining the preferred optimal temperature for the species at hand. Common methods to administer supplemental heat include circulating warm-water blankets, administering warm fluids parenterally, raising the temperature in the operating suite, and forced warm-air blankets (Figure 4). We prefer these blankets, because they appear to provide the best homogenous heat without the possibility of skin burns.

The administration of analgesics before and after surgery should be considered in all reptile patients. The behavioral response to pain of most reptiles cannot be equated to that in mammalian patients: reptiles that do not display the clinical signs associated with pain in mammals should not be assumed to be pain-free. Opioids such as butorphanol and buprenorphine are used in reptiles.[96] Recently, several studies have been published to determine the potential use of opiates as analgesics for reptiles.[86,97,98] Specific options for anesthesia and analgesia in reptiles can be found in reference 10.

Soft Tissue Procedures

Integumental surgery. Skin incision and closure techniques must take into account the nature of reptile skin. Turtle and lizard skin can be tougher than that in mammals, and incisions may have to circumvent osteoderms or other keratinized structures. In snakes, skin incisions should be made between scales, making most surgical wounds scalloped. Incising through the scales is more difficult, causes permanent damage to the scales, and leads to posthealing dysecdysis. The use of radiosurgery or diode laser facilitates surgical incisions in reptiles.[44,46] Incised reptile skin has a tendency to invert. Therefore, everting suture patterns (for example, horizontal or vertical mattress) are recommended to ensure opposition of tissue without future dysecdysis (Figure 5).[5,71] In addition, skin staples have been advocated, because they cause mild eversion of the skin. Sterile surgical tissue adhesive can be useful for closing the skin, although care must be taken to evert the skin when applying the adhesive. Given the length of time needed for reptile wounds to heal, sutures should not be removed until at least 6 wk after surgery.[71] Ecdysis may lead to the premature loss of skin sutures. Dysecdysis is an expected consequence of surgical incisions (particularly in snakes), and future management of the reptile surgical patient should anticipate for this condition.

Skin wounds. Wounds are common in reptiles. Given the caseous nature of reptilian purulent exudates, basic cleaning and irrigation is seldom sufficient, and surgical debridement usually is necessary. Sharp dissection is used to remove all adherent necrotic and infected tissue. Although slow, the ability of reptiles to recover from extensive trauma is remarkable. Allogeneic and xenogeneic grafts have been useful in reptiles.[9,14,27,73,74,114-116] Successful xenogeneic skin grafts using porcine small intestinal submucosa have been reported in reptiles.[9,14]

Subcutaneous abscess removal. Subcutaneous abscesses, which present as a firm, discrete swelling, are very common. A skin incision is made over the entire abscess, the skin reflected, and the abscess removed en block. In some cases, an easily distinguishable capsule can be identified and should be removed with the abscess in toto.[15] A piece of this capsule should be submitted for bacterial and fungal culture. The underlying tissue is irrigated thoroughly with antiseptic solution (for example,

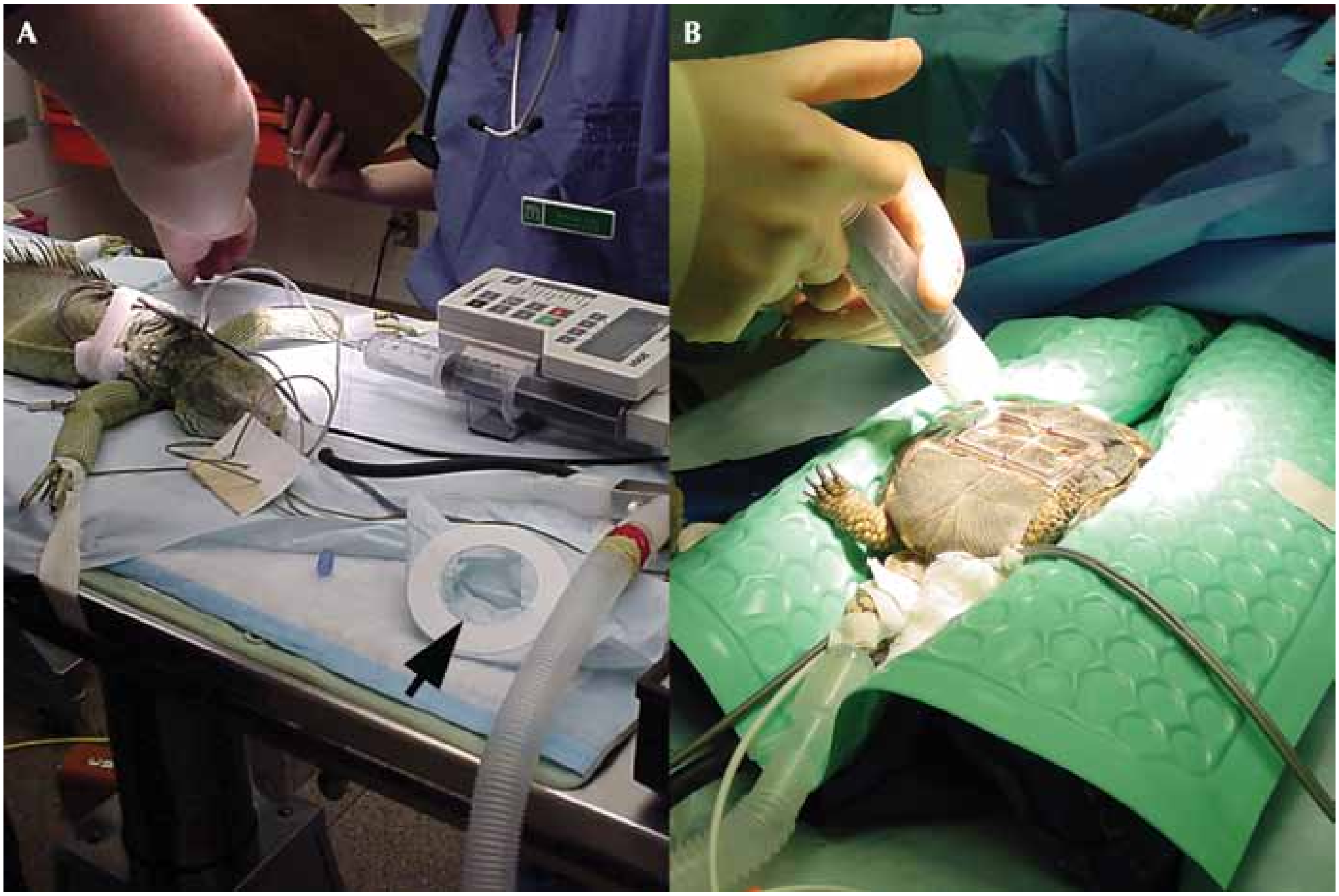

Figure 4. Intraoperative temperature maintenance. (A) Left, A green iguana placed on top of a forced air device; note the warm air inlet to the surgery table blanket (arrow). (B) Right, Box turtle on a warm circulating-water blanket.

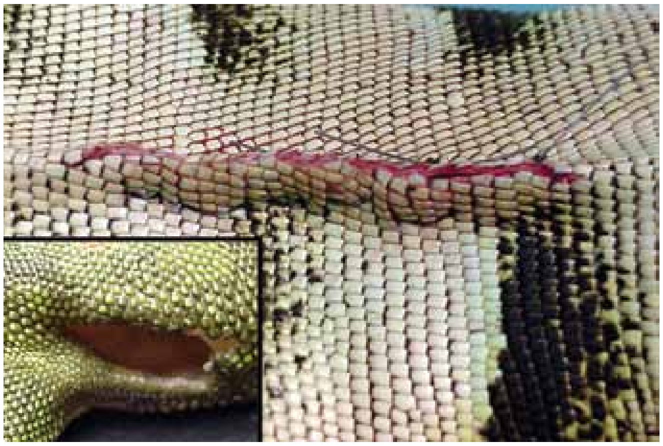

Figure 5. Routine coeliotomy skin closure in a green iguana, with monofilament nylon in an everting horizontal mattress pattern. Insert, Open surgical wound after abscess removal in a giant day gecko; note the inverting wound edges.

chlorhexidine, povidone–iodine). If the entire abscess was removed, the skin can be closed in a routine manner. In the case of an abscess that has progressed to osteomyelitis, antibiotic-impregnated beads can be implanted in the surgical site.[25]

Aural abscess removal. Aural abscesses in chelonians are very common and have been associated with hypothermia, malnutrition, and environmental pollutants.[81] The anesthetized chelonian should be in sternal recumbency, with the head and neck extended and rotated so that the tympanic abscess is uppermost. A ventral, semicircular incisional flap is made over the distended tympanum in a craniocaudal direction. The skin is retracted gently to allow the insertion of a curette around the edge of the caseous material. If further exposure is necessary, a horizontal incision across the tympanum permits half of the tympanum to be removed.[81] In extreme cases, the entire tympanum can be removed, creating a complete circular defect. The Eustachian canal must remain patent to prevent recurrence; often fine-tipped forceps are needed to remove material from within this canal. The cavity should be flushed copiously with an antiseptic solution followed by sterile saline. The site is left open for postoperative care. Postoperative care includes correcting the underlying causes (for example, hypothermia, malnutrition, environmental pollutants) and flushing the site daily with antiseptic solution.

Ophthalmic Surgery

Subspectacular abscess removal. Blockage of the lacrimal duct in snakes (and lizards that possess spectacles) results in fluid accumulation between the spectacle and cornea. The fluid often becomes infected with bacteria, inspissates, and must be removed. A 30° to 90° wedge is removed from the ventral aspect of the spectacle. The caseous material is removed for cytology and microbiology and the subspectacular space thoroughly flushed.[65] The patency of the lacrimal duct must be ensured. It is generally easier to catheterize the buccal opening of the duct, because it emerges close to the cranial margin of the palatine teeth, and to flush retrograde. The wedge incision in the spectacle is left open, and topical ophthalmic medication is recommended. In reptiles with persistent lacrimal duct blockage that fails to respond to spectacular wedge resection, a conjunctivoralostomy can be performed.[77] At the medial canthus edge of the spectacle, a 30° incision allows the introduction of a needle that is forced from the inferior fornix of the subspectacular space through the roof of the mouth to emerge between the palatine and maxillary teeth. Sialistic tubing is threaded through the needle, and the needle removed. The tubing is sutured to the roof of the buccal cavity and to the periocular skin to maintain patency. The tube is removed 4 to 6 wk postoperatively.

Enucleation. Removal of the globe is required where irreversible ocular damage, prolapse, or pain persist, or for the collection of tissues for research. In snakes (and lizards that possess spectacles), a circular incision is continued around the entire spectacle, which then is removed. The globe is grasped with forceps, gently elevated, and dissected free from its attachments. Optical vessels are ligated by using a small vascular clip and curved applicator, although suture can be used. As with enucleation in any species, extensive hemorrhage can result if sufficient hemostasis is not attained. Given the lower blood pressure of reptiles compared with mammals, radiosurgery is often effective in smaller reptiles where ligation of the optic vessels is not possible. The ocular deficit is left open, and topical ophthalmic antibiotics should be applied to the area. Healing typically takes 4 wk. Alternatively, the globe may be eviscerated, leaving the sclera and bony ossicles. The advantage of evisceration is that the globe does not collapse and result in a sunken appearance. In chelonians, crocodilians, and most lizards, it is possible to close the orbit by tarsorrhaphy. In very small reptiles, a small radiosurgery loop can be used to eviscerate the globe.

Thyroidectomy. The reptilian thyroid plays an intimate role in ecdysis, reproduction, tail regeneration, growth, endurance, metabolic rate, and oxygen consumption.[55,57-59] Many studies demonstrate the role of the thyroid through the use of experimentally thyroidectomized reptiles.[58,59] The complete cervical region of the dorsally recumbent reptile is aseptically prepared and draped. The hyoglossum (dewlap) is retracted laterally. A ventral midline incision from the manubrium is extended cranially. Retraction of the skin reveals the left and right constrictor colli muscles. These muscles originate from dorsal aponeuroses and insert as a midventral fascia. The ventral midline connection between these muscles is incised and reflected with little bleeding. The deeper muscle fibers of the omohyoideus and, more caudally, episternocleidomastoideus run in a craniocaudal direction from the clavicles and suprascapulas to the posterior aspect of the hyoglossum and occipital bones, respectively. The muscle fibers are separated bluntly at the ventral midline. After retraction of these muscles and the ventral fascia, the well-circumscribed thyroid is visible. The thyroid is encapsulated within a friable membrane. Arterial supply consists of left and right thyroid arteries and left and right laryngotracheal arteries. A single thyroid vein drains into the right tracheal vein. The position of the vagus nerve, ventral to the thymus where the external carotid crosses ventral to the jugular vein, and inferior laryngeal nerve, adjacent to the trachea, are identified and preserved. The thyroid is removed intact after ligating the 4 supply arteries and single vein by using small vascular clips. The parathyroid glands are not identified reliably at the time of surgery. After removal of the thyroid, the muscle layer closed by using absorbable braided synthetic polyglactin 910 suture. The skin is closed in a routine manner.[48]

Coeliotomy

Surgical celiotomy provides access to most of the major internal organs and therefore is useful for a range of surgical procedures including exploration, implantation, and biopsy. Surgeons should remember that the visceral organs of most reptiles are more delicate and friable than are their mammalian counterparts. The technique is simplest to perform in the uncompartmentalized coelom of most lizards, more difficult in snakes

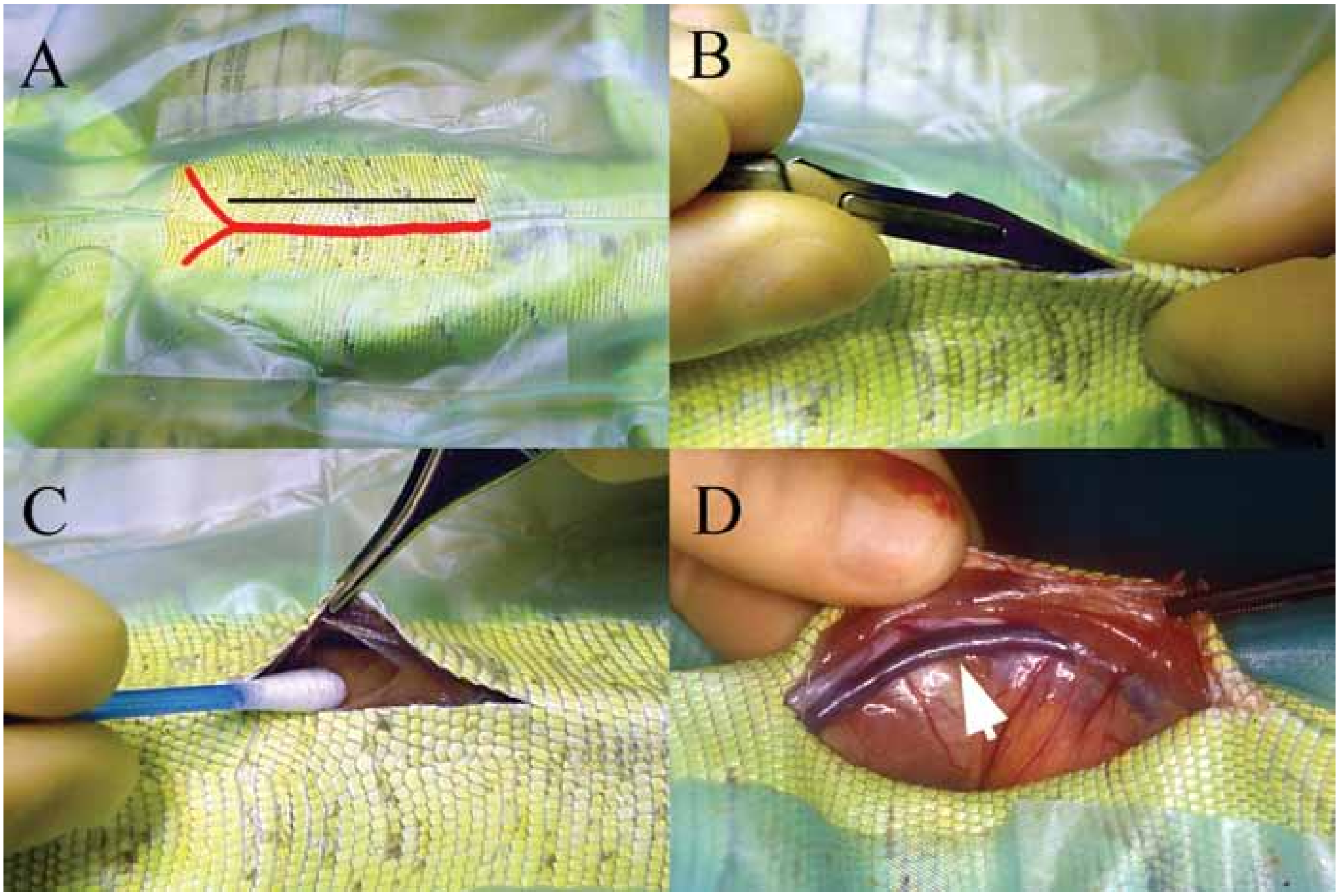

Figure 6. Coeliotomy in a green iguana. (A) The ventral midline abdominal vein (red line) and proposed paramaedian skin incision (black line). (B) Skin incision made by using a reversed scalpel blade to avoid inadvertent damage to deeper structures. (C) Dissection through the thin abdominal musculature by using cotton-tipped applicators. (D) Exposure of the coelom and visualization of the ventral midline abdominal vein.

with diffuse fat bodies and fascial planes, and most difficult in chelonians and crocodilians due to their bony integument and compartmentalized coelomic cavities.

Lizards. Most lizards are placed in dorsal recumbency, but some laterally compressed species (for example, chameleons) may be better placed in left or right lateral position with transection through several ribs. There are 2 major approaches, paramedian and midline, to the coelom of most lizards.[5,71] The incision may have to extend from the xiphoid process to just cranial to the pelvis to provide sufficient exposure for certain procedures.

The advantage of the paramedian approach is avoidance of the large midline abdominal vein (Figure 6). However, incision through abdominal musculature may produce considerable postoperative pain. A craniocaudal incision is made parallel to, but lateral from, the ventral midline. The abdominal musculature is thin and is easily incised by radiosurgery, laser, or blunt dissection by using cotton-tipped applicators. Sharp incision by using scalpel or scissors should not be used, because it results in increased hemorrhage. The coelomic membrane is very thin and is penetrated easily by using cotton-tipped applicators. The advantages of the midline approach are equal accessibility to both sides of the coelom and decreased muscle trauma; however, greater surgical skill is required to preserve the abdominal vein than is necessary with the paramedian approach. The midline approach requires a careful incision that starts at, or caudal to, the umbilical scar and extends craniad. Careful incision through the skin and underlying linea alba reveals the abdominal vein that should be preserved and gently retracted laterally.

Once the coelomic cavity is accessed, care is required to identify a distended bladder and avoid accidental rupture on entry into the coelom. Coeliotomy closure in larger lizards should be in 2 layers with the muscle closed by using a simple interrupted or continuous pattern with absorbable suture. In small lizards (less than 50 g), suturing the skin only may be acceptable, given that the abdominal musculature might be too thin to manipulate.

Snakes. The elongated nature of snakes makes it impossible to make a single coeliotomy incision to view all major organs. Therefore, it is vital that the precise surgical site is determined accurately ahead of time by using anatomy references, palpation, clinical pathology, and diagnostic imaging.[26,76] In general, the incision is made between the first and second rows of lateral scales,[71] taking care to incise between the scales whenever possible (Figure 7). This strategy ensures that the incision is positioned laterally and off the floor when the recovered snake is ambulatory. Radiosurgery or laser or blunt dissection is continued through the muscle layer, just ventral to the ribs. Entry into the coelom is achieved by creating an opening by moving between the ribs and ventral musculature. It is often necessary to navigate through multiple fascial layers to gain access to the coelom in snakes, and extensive fat bodies can hinder visualization. A 2-layer closure is routine, with the muscle layer closed by using absorbable suture in a simple interrupted or continuous pattern.

The use of biotelemetry and radiotelemetry has widespread use in snake field studies.[100] The transmitters are implanted in the coelomic cavity by means of coeliotomy, frequently with a

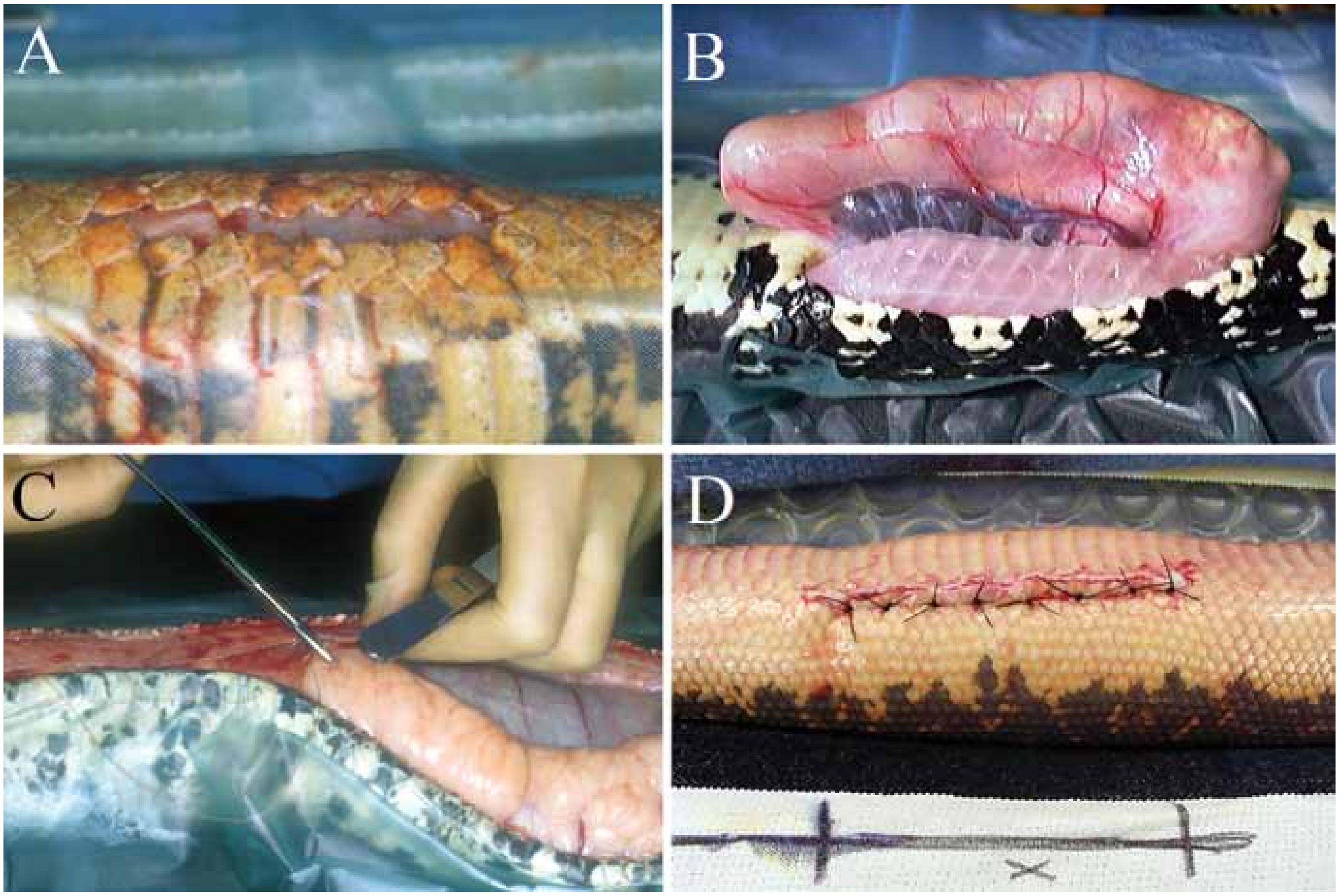

Figure 7. Coeliotomy in snakes. (A) Initial skin incision between the first and second row of lateral scales. (B) Entry into the coelom just ventral to the ribs to permit exteriorization of inspissated retained ova in a kingsnake. (C) Closure of the muscle layer in a boa constrictor by using a simple continuous pattern and absorbable suture. (D) Routine skin closure in a Kenyan sand boa, demonstrating typical skin eversion due to horizontal mattress sutures.

subcutaneous antenna. The transmitter should be sterilized by gas sterilization.[83] A skin incision is made between the first and second dorsal scale rows at a location just anterior to the gonads.[92] One way to estimate the correct incision site is to measure anteriorly from the vent to one third of the total distance from vent to snout.[83] Then the ventral abdominal muscles are incised immediately ventral to the costal cartilages. The transmitter is implanted in the coelom through this incision. The antenna is placed subcutaneously by pulling it through a cannula tunnelled subcutaneously. Options for a cannula include a 16-gauge needle[92] or sterile copper tubing.[83] Another method describes implanting the transmitter between the skin and epaxial muscles (not within the coelomic cavity) and antenna subcutaneously by using multiple stab incisions through the skin.[30]

Chelonians. There are 2 major approaches, transplastron and prefemoral,[5,71] to the coelom of chelonians, and the more appropriate choice depends on the species and organ of interest. The traditional transplastron coeliotomy approach requires a temporary osteotomy through the ventrum of the chelonian. In contrast, the prefemoral approach is a soft tissue technique that involves entry into the coelom in front of a pelvic limb.

For a transplastron celiotomy, the chelonian is positioned in dorsal recumbency and aseptically prepared (Figure 8). The 2 lateral, cranial, and caudal plastron incisions are planned. An oscillating saggital saw couples accuracy with reduced collateral damage and minimal soft tissue trauma. The saw is held at 45° to bevel the shell incisions.[5,71] The 2 lateral cuts are made first, and a 25-gauge needle is used to ensure that the cuts are full-thickness. The caudal cut and finally the cranial cut are made in a similar fashion. Penetration of the shell over the heart is made last. By using a periosteal elevator, the cranial edge of the plastron segment is lifted to expose the muscle attachments. Staying as close to the plastron segment as possible, blunt dissection separates the plastron osteotomy segment from the underlying soft tissues.[5] The paired abdominal veins are associated closely with the plastron, and it may be necessary to strip away carefully the periosteum of the flap to preserve these vessels with the rest of the soft tissues (Figure 9). The caudal soft tissue attachments can be left intact as the plastron flap is reflected caudally and covered with moistened sterile gauze. A 2-layer closure is routine. The coelomic membrane is closed in a simple interrupted or continuous pattern by using absorbable suture. The plastron's healing process depends directly on the quality of the repair (Figure 10). Accurate reduction of the bony section and plastron will improve the likelihood of first- or second-intention healing. However, in most cases the, plastron flap becomes a sequestrum that provides temporary protection to the new bone developing beneath. Three or 4 stabilizing sutures (polydioxione for small specimens, wire for adults of larger species) are placed through predrilled holes to anchor the bony section to the plastron. The shell is cleaned carefully with an alcohol swab to remove any grease, before epoxy resin or an acrylic (for example, polymethylmethacrylate) is used to seal the area. It is helpful to use masking tape to protect adjacent areas before applying the repair material. Any excess repair material can then be removed easily along with the tape, creating a clean,

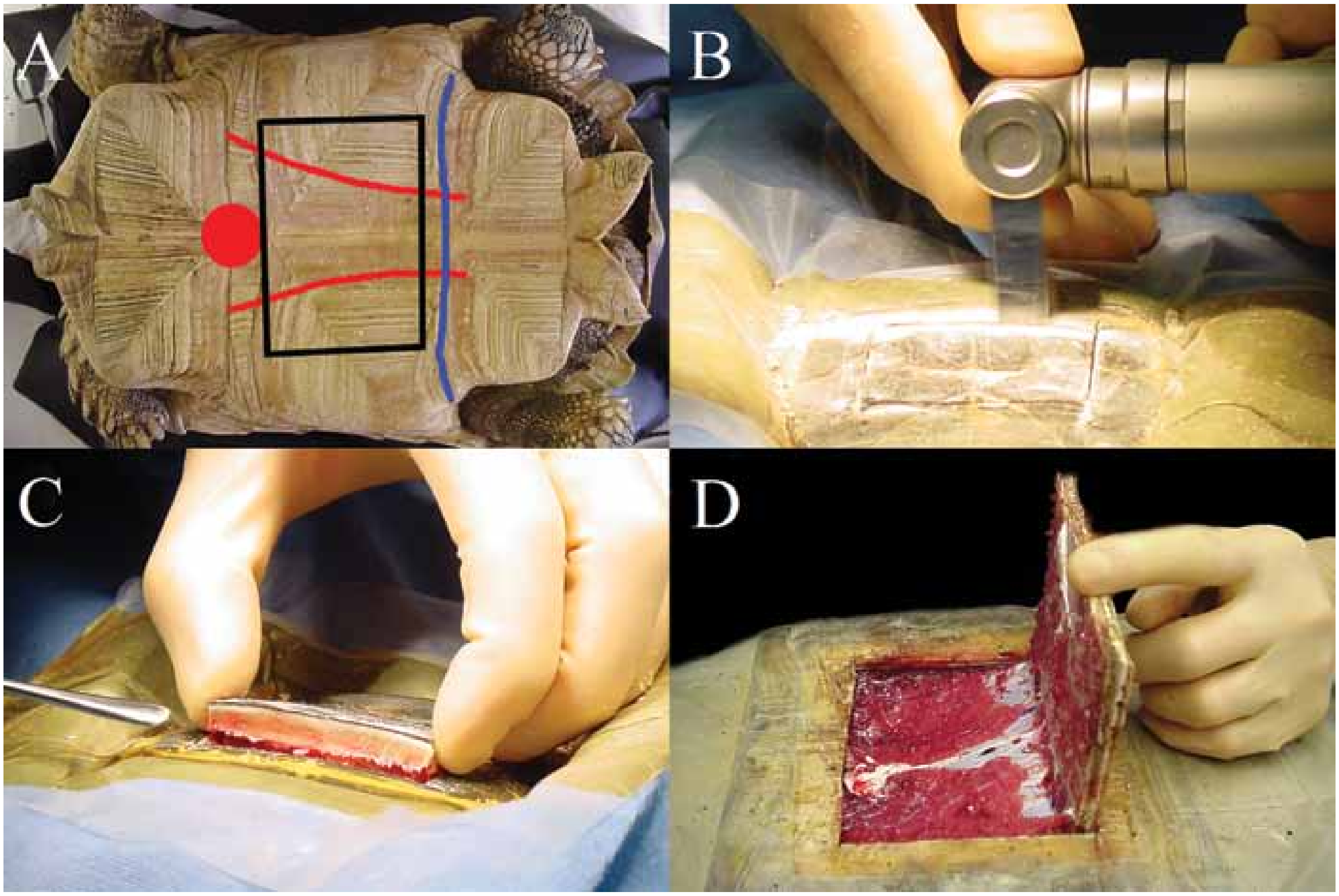

Figure 8. Transplastron coeliotomy in chelonians. (A) View of the ventrum of a tortoise, illustrating the position of the heart (red circle), paired ventral abdominal veins (red lines), caudal plastron hinge (blue line), and the 4 proposed osteotomy incisions (black square). (B) Making a beveled incision in the plastron by using an oscillating saggital saw. (C) Elevating the plastron flap from its cranial edge. (D) Reflection of the plastron flap after blunt dissection between the bone and underlying soft tissues.

cosmetic finish. The plastron heals in 12 to 18 wk. The covering material (that is, epoxy) should be left in place for 6 to 12 mo before removal. Radiography is always helpful in the assessment of healing, because ossified callous formation seldom occurs.

Prefemoral coeliotomy (Figure 11) provides limited lateral access to the coelom, but might be sufficient in species with a large prefemoral fossa.[79] With the chelonian in lateral or laterodorsal recumbency, the pelvic limb is secured caudally to expose the prefemoral fossa.[5] After aseptic preparation, a craniocaudal incision is made in the middle of the fossa. Blunt dissection cranial to the sartorius and ventral to the iliacus muscles reveals the coelomic aponeurosis of the transverse and oblique abdominal muscles. This coelomic aponeurosis is fibrous, and some force may be required to breach the membrane. Stay sutures through the skin, subcutaneous tissues, coelomic membrane, and visceral organs improve surgical exposure. Closure routinely is accomplished in 2 layers, the coelomic membrane and the skin.

Crocodilians. The thick integument of crocodilians contains osteoderm, making coeliotomy difficult. In these species, an incision directly over the organ of interest is easier to make than is a standard midline coeliotomy approach for assessing multiple organs.

Reproductive System

Reproductive disease is common in captive reptiles and represents one of the most common medical reasons for surgery.[24,33,35,36,60,70,79,88] In addition, manipulation of an animal's endocrine or reproductive system is commonly necessary in research. Many of these procedures have been described previously.[67]

Orchidectomy and vasectomy. ***Lizards.*** In lizards, the testes generally are located adjacent to the dorsal renal veins and adrenal glands, in the mid- to caudal coelom. The testes are suspended by a short broad ligament from the dorsal body wall, within which the testicular artery and veins are located. The left adrenal gland is positioned between the left testis and left renal vein,[71] and care must be exercised to identify and preserve the gland during orchidectomy. The right adrenal is below the renal vein and is, therefore, less likely to be damaged.[71] It is easier to use vascular clips to ligate the vessels, rather than suture, and then dissect the testes free distal to the vascular clips. One method is to pierce the testis with a tuberculin syringe needle and use the syringe as a handle to retract the testis enough to allow the vessels to be visualized for clipping or cauterization[43] (Figure12).

Snakes. In snakes, testis location varies with family. However, the surgery is straightforward and similar to that in lizards once the location of each testis is identified. Vasectomy in the garter snake has been reported.[117] The technique required a 5-cm incision from the 45th to 40th ventral scale (from the cloaca). The ducti deferentes were located on either side of the intestinal tract, and a 3-cm segment was excised from each duct.

Chelonians. Orchidectomy is extremely difficult in chelonians due to the testes' caudodorsal location and intimate relationship

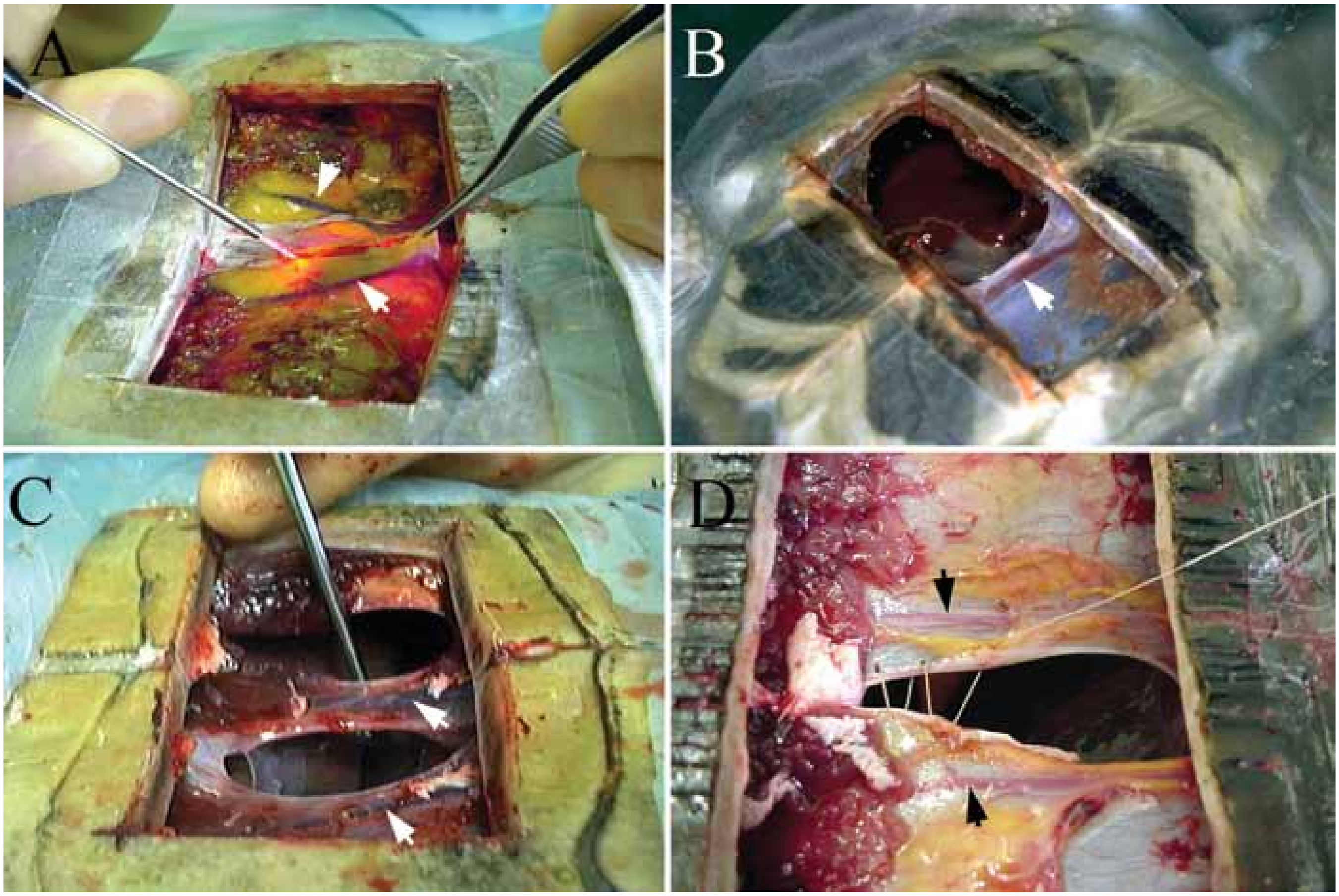

Figure 9. Transplastron coeliotomy in chelonians. (A) Incision through the coelomic membrane, between the paired abdominal veins (arrows). (B) Ligation and trisection of an abdominal vein to create a large access flap in the coelomic membrane; the remaining intact abdominal vein is shown (arrow). (C) Two coelomic membrane incisions—one between the abdominal veins (arrows), and the other lateral to the abdominal veins—to provide improved access to the lateral coelom. (D) Routine closure of the coelomic membrane by using absorbable suture in a simple continuous pattern.

with the kidneys. Endoscopic vasectomy has been performed by one of the authors.

Ovariectomy. Because reproductive disease is common in female reptiles, ovariectomy is recommended frequently as a preventive measure. In addition, ovariectomy will prevent breeding in communal groups, alter the hormones present for research reasons, and may be part of the treatment for reproductive disease.[24,69,79,84,87]

Lizards. In nonreproductively active female lizards, the ovaries are generally small and located adjacent to the dorsal renal veins and adrenal glands, in the mid to caudal coelom. Ovary removal can be accomplished safely by first applying vascular clips along the ovarian ligament to ligate the small vessels, before dissecting the gonad free. It is not necessary to remove the oviducts if they are normal. In cases of preovulatory ova stasis, the active ovaries are considerably enlarged and immediately obvious, because they resemble clusters of yellow-orange grapes. Each ovary is elevated to expose the suspensory ligament containing 3 to 8 large vessels that branch off the aorta and renal veins. Hemoclips or suture is used to ligate these vessels, before the ovary is dissected free. The oviducts are usually small and involuted and do not need to be removed unless diseased.

Snakes and chelonians. Routine ovariectomy is seldom recommended in snakes or chelonians because of the invasive nature of surgery and their reduced chances of spontaneous reproductive disease compared with that in lizards. Ovariectomy in chelonians is performed as part of dystocia surgery to prevent future breeding—a transplastron or bilateral prefemoral coeliotomy is required. Chelonian ovaries originate close to the ventrolateral aspect of the kidneys, but in mature female chelonians the ovaries are large and extend into the central coelom. The ovarian suspensory ligament is extensive, and great care is required to ensure that the entire ovary has been isolated, before vascular clips are applied and the ovary removed.

Salpingotomy and salpingectomy. *Lizards and snakes.* In cases of postovulatory egg stasis, the thin oviducts full of eggs are immediately obvious on entry into the coelom of lizards and snakes.[24] Multiple salpingotomy incisions can be made to remove the eggs in an effort to maintain future breeding capacity; however, surgery time is extended greatly. In most lizards, bilateral ovariosalpingectomy is recommended but is not performed routinely in snakes because of the extensive length of the serpentine ovaries and oviducts. The large, numerous blood vessels that supply each oviduct must be ligated with suture or vascular clips.[71] Vascular clips and laser greatly reduce surgery time, and often several vessels can be clamped with a single clip. The oviducts are ligated close to their insertion with the cloaca by using a circumferential transfixing ligature and removed. In lizards, coelomitis secondary to follicles ovulated into the coelomic cavity requires removal of the ovaries. In snakes, single or multiple coeliotomy and salpingotomy incisions are preferred, with efforts made to manipulate more cranial and caudal eggs out of the same incision.[36,78,88]

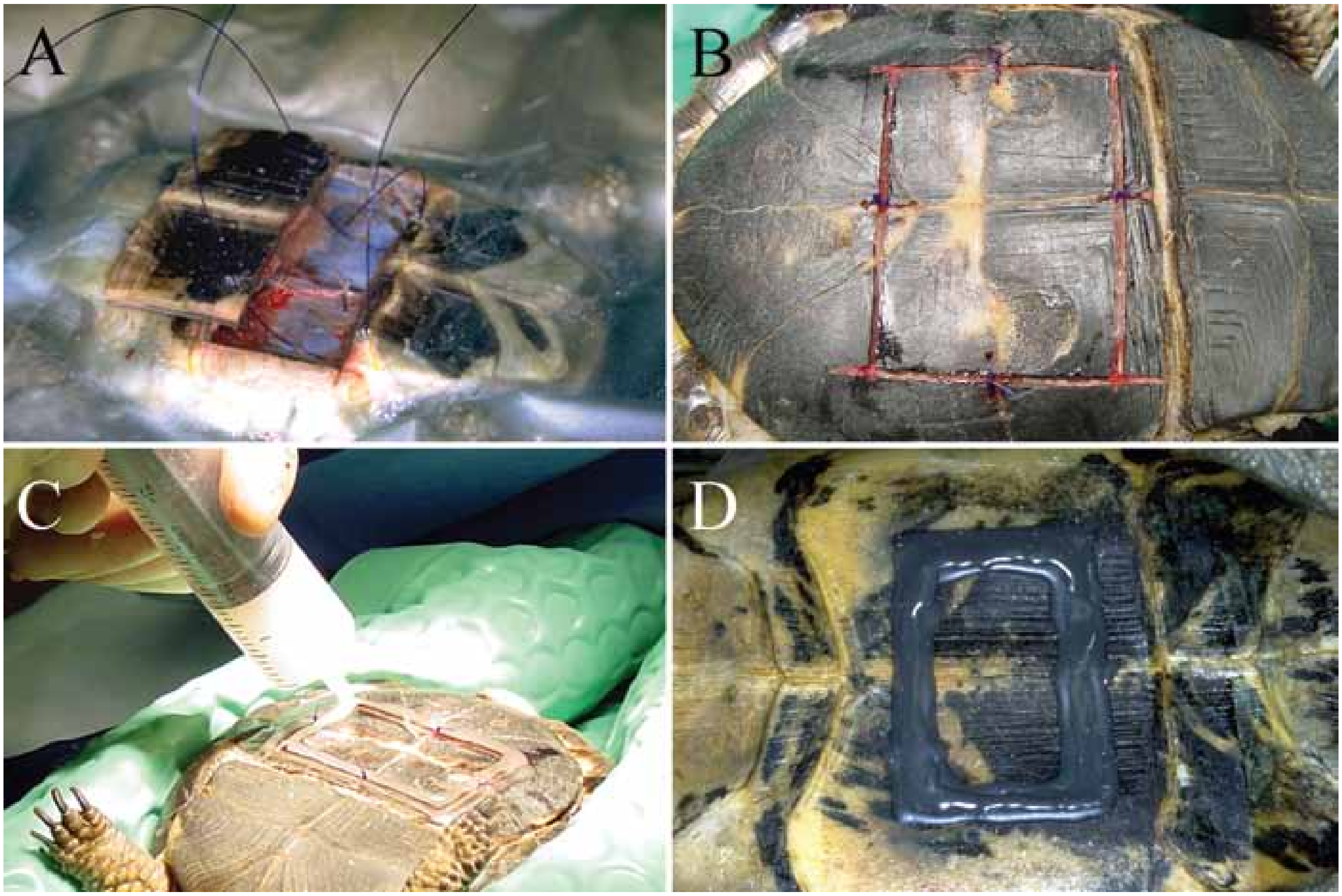

Figure 10. Transplastron coeliotomy in chelonians. (A) Suture placed through predrilled holes in the plastron flap and shell in preparation for repositioning flap. (B) Plastron flap repositioned and held in place by using 4 sutures, with the incision lines filled with antibiotics. Note the overscoring at the corners due to the use of a large rotating blade. (C) Application of low-temperature polymethylmethacrylate (Technovit, Jorgensen Laboratories, Loveland, CO) by syringe. (D) Final repair, with the incision and sutures buried beneath the repair material.

Chelonians. Chelonian oviducts are thicker than those in lizards and snakes. The surgeon often has the viable option of performing multiple salpingotomy incisions and preserving the reproductive status of the animal or choosing unilateral or bilateral ovariosalpingectomy.[50,86,110-112] Endoscope-assisted ovariectomy can be performed in chelonians.

Urinary System

Cystotomy and cystectomy. A common indication for bladder surgery is the removal of bladder stones, which are common in lizards and chelonians and may be quite large.[7,31] Partial or total cystectomy may be undertaken as a research procedure to investigate the postrenal urinary physiology of reptiles.

Coeliotomy allows manipulation of the bladder, which is located in the caudoventral coelom. Often stay sutures are used to prevent coelomic contamination by nonsterile urine, and fluid contents should be aspirated prior to incision. Scalpel, radiosurgical, or laser incision permits entry into the bladder. The bladder should be flushed with sterile saline and closed by using fine absorbable suture. The bladder wall usually is thickened when a stone has been present and should be closed in 2 layers.[5] In cases where the bladder wall is very thin, such as in a normal, healthy bladder, it can be impossible to perform a 2-layer closure. In such cases, single continuous closure followed by an inverting pattern provides assurance against leakage.

Gastrointestinal System

Esophagostomy tube placement. *Lizards and snakes.* Because lizards and snakes are readily tube-fed by the oral route, the indications for placement of an esophagostomy tubes are rare. If performed in these species, the initial approach to the esophagus is made caudal to the tympanic scale: in lizards, half way to the shoulder, and in snakes, at least 2 cm from the head.

Chelonians. In situations where repeated stomach tubing is required, the placement of an esophagostomy tube has both practical and welfare benefits and is used often in anorectic chelonians. A soft feeding catheter is premeasured from the cranial rim of the plastron to the junction of the pectoral and abdominal scutes, and marked with a nontoxic permanent marker. With the chelonian in lateral recumbency (Figure 13), curved hemostats are introduced into the mouth, down the esophagus, and tented up against the caudolateral aspect of the neck, as far down the neck as possible. A small skin incision is made through the aseptically prepared skin, over the point of the hemostats, to move the closed jaws out through the esophagus and the skin incision. Beware of the dorsolateral jugular vein and ventrolateral carotid artery. The distal end of the feeding catheter is grasped, pulled through the incision into the lumen of the esophagus, redirected down toward the stomach, and advanced into the stomach, to the predetermined mark on the tube. The catheter is secured to the neck by using either a butterfly tape sutured to the skin of the dorsolateral aspect of the caudal neck or a finger-trap suture. In chelonians, the tube should also be attached to the carapace. The tube is cut to length and capped.

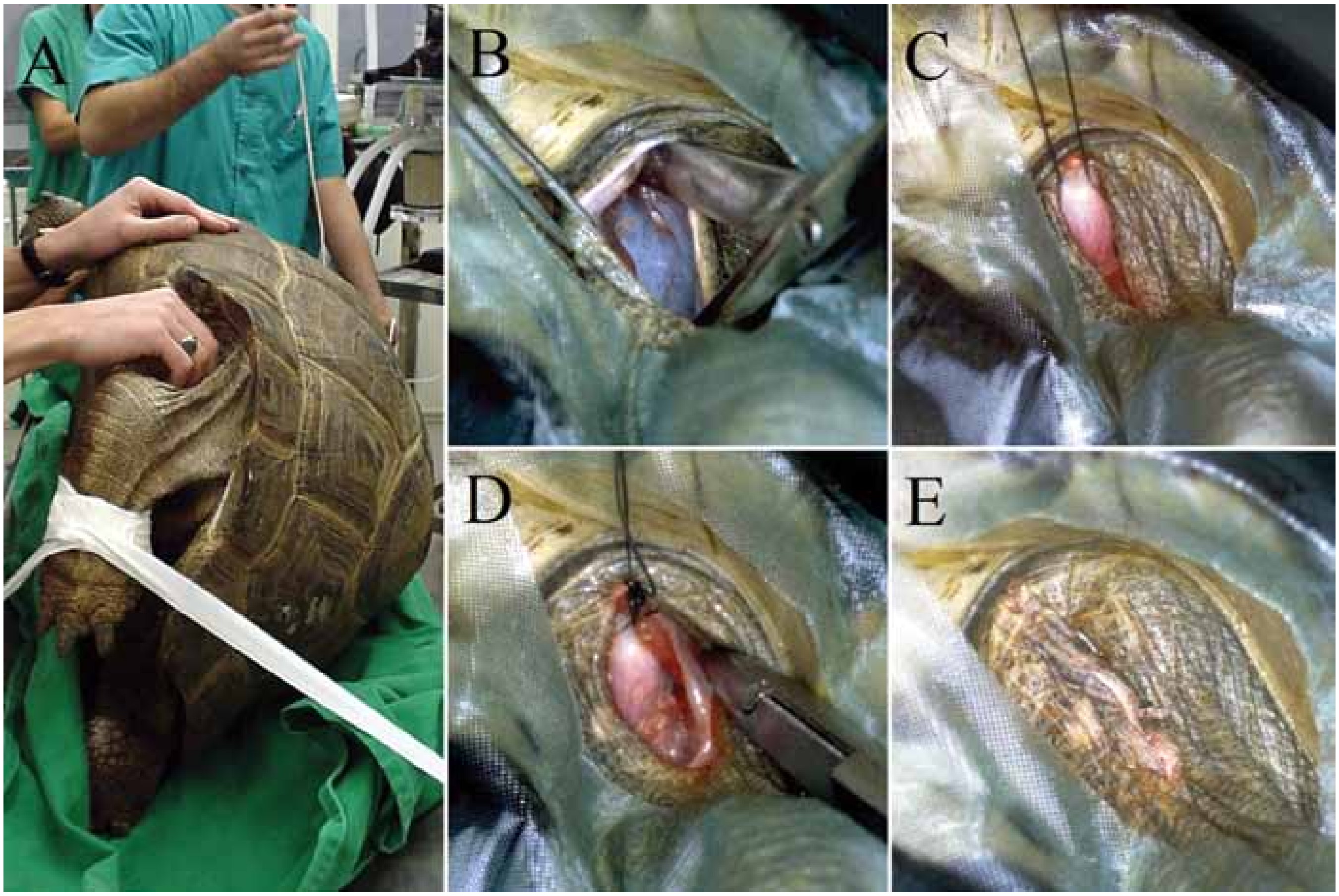

Figure 11. Prefemoral coeliotomy in chelonians. (A) Giant Aldabra tortoise in lateral recumbency ready for prefemoral coeliotomy. (B) After skin incision the soft tissues are bluntly dissected to reveal the fibrous aponeurosis of the abdominal musculature. (C) Stay suture restraint of the colon in a spur-thighed tortoise. (D) The colon is sutured to the coelomic aponeurosis and transverse abdominal muscle to anchor the colon and prevent future prolapse. (E) Routine skin closure at the prefemoral site; healing takes approximately 6 wk.

Gastrotomy, gastrectomy, enterotomy, and enterectomy. Gastrointestinal surgery is similar to that in mammals, although tissues are often thin and friable in reptiles and call for the use of fine suture, atraumatic forceps, and needles. In general, fine suture and standard intestinal closure techniques are used. The mesentery that suspends the gastrointestinal tract is variable and may prevent exteriorization through the coeliotomy incision. Packing the exteriorized gastrointestinal tract with sterile gauze to trap any inadvertent leakage is useful. In addition, copious irrigation with fluids prior to closure is recommended. Foreign body removal, resection–anastomosis, and anastomosis for colorectal atresia have been performed successfully in reptiles.[23,34,42,64]

Cloacal organ prolapse. Various organs have been reported to have prolapsed through the vent, including phallus or hemipenes, oviduct, cloaca, colon, bladder, and even kidney;[70] therefore, it is vital that the precise anatomy of the prolapse is identified as treatment options vary dramatically. Only the most common types of prolapse are covered here.

Phallus and hemipenes. The penis of a crocodilian or chelonian is a single organ and usually is retracted into the cranioventral cloaca. Snakes and lizards possess paired hemipenes that retract into the ventral tail, caudal to the cloaca. Penile prolapse is a common condition of reptiles. If the prolapsed tissue appears viable, it can be cleaned, moistened, and replaced gently. The squamate hemipenes are replaced caudally from the vent, whereas the crocodilian and chelonian penis is replaced cranially into the urodeum of the cloaca. Two simple interrupted sutures can be placed across the lateral margins of the vent to prevent immediate recurrence of prolapse for 5 to 10 d. A purse suture should not be used, because it is likely to damage the musculature and innervation to the vent. It is important that suture placement allows for normal urination and defecation. If the tissue appears necrotic, penile amputation is the treatment of choice.

Figure 12. Celiotomy with castration of an anolis. A small needle is used to pierce the testis to enable its retraction from the coelomic cavity.

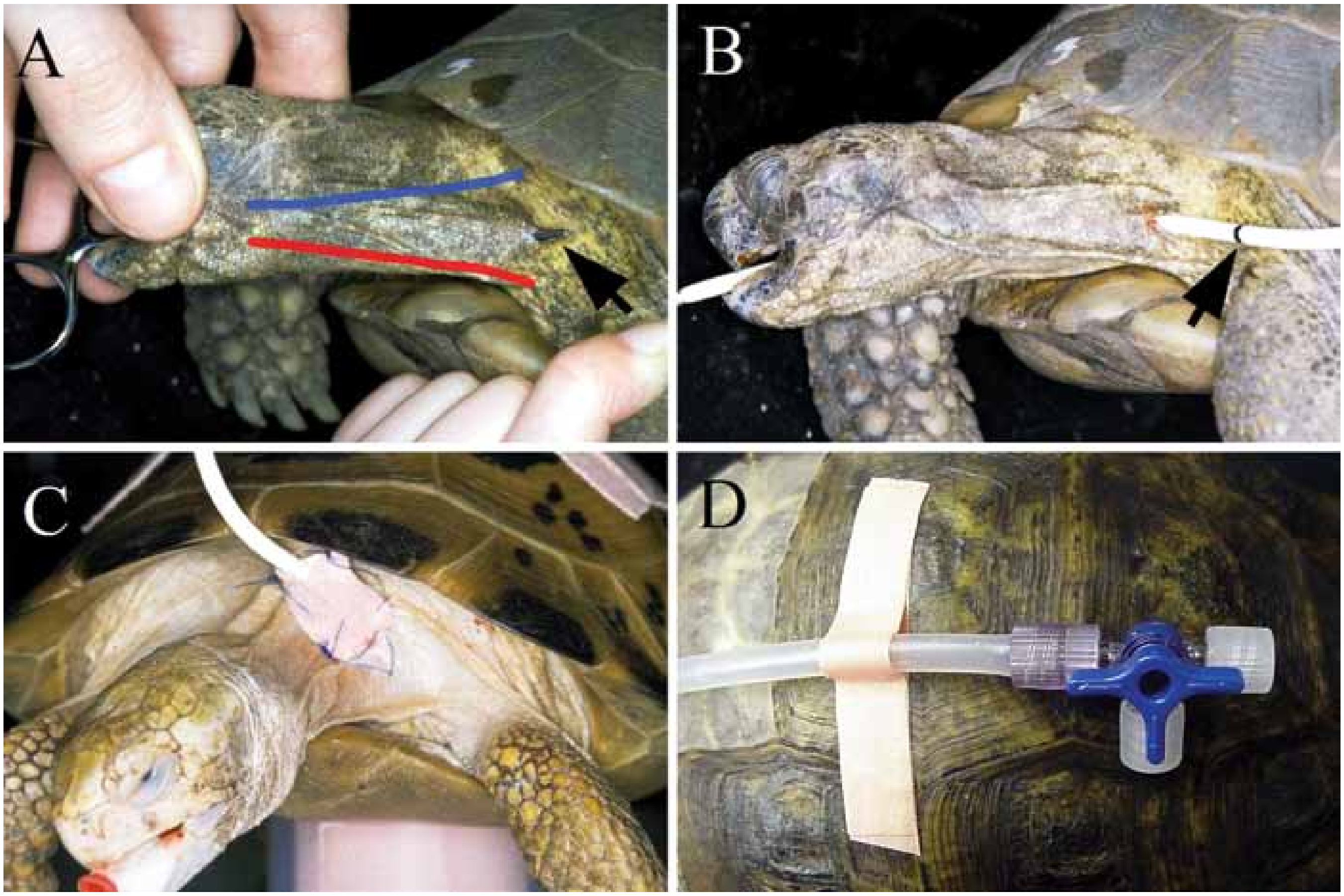

Figure 13. Oesophagostomy tube placement in chelonians. (A) A small skin incision has permitted the jaws of the haemostats to penetrate through the lateral skin of the caudal neck (arrow). Note the positions of the jugular vein (blue) and carotid artery (red). (B) The end of the feeding catheter is grasped and pulled through the skin into the lumen of the esophagus, and redirected down the esophagus to the predetermined mark on the tube (arrow). (C) Butterfly tape attached to the tube and sutured to the caudal skin of the neck. (D) The end of the tube, glued and taped to the carapace, with a 3-way tap attached.

The reptilian copulatory organ does not contain a urethra; therefore, mattress sutures or circumferential ligatures can be placed at the base, and the organ safely resected.[6] Obviously, this procedure will compromise future breeding, although snakes and lizards with a single remaining hemipenis can still reproduce.

Cloaca and colon. Prolapse of the cloaca and distal colon are not uncommon. The underlying cause (for example, obesity, renomegaly, constipation, dystocia, intestinal parasitism, secondary nutritional hyperparathyroidism, cloacitis, colitis) must be corrected. A minor prolapse can be replaced, and 2 simple interrupted sutures can be placed near the edges of the vent to prevent recurrence. More severe prolapse is treated by transcutaneous cloaco- or colopexy.[6] On rare occasion, this procedure will fail, and a coeliotomy is required to anchor the colon to the last 1 to 3 ribs by using nylon.[8]

Respiratory System

In most lizards and snakes, the lack of a functional diaphragm enables the surgeon to directly approach and enter the lungs through a standard coeliotomy for the purposes of debridement, resection, parasite removal, or tissue collection. In chelonians, access to the lungs requires a dorsal osteotomy through the carapace.

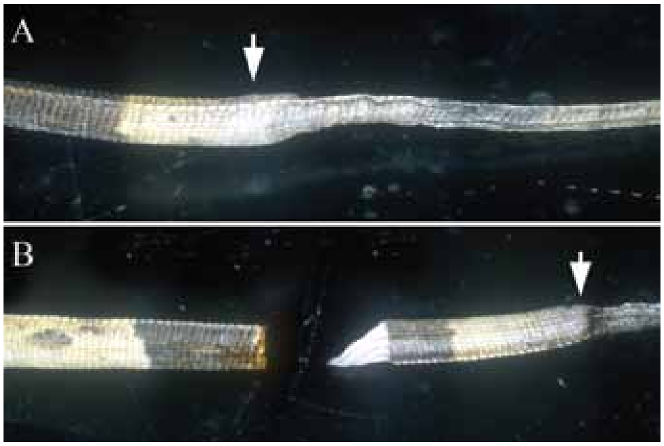

Figure 14. Tail amputation in a green iguana. (A) Tail with distal avascular necrosis at presentation. The visible margin between visibly normal and abnormal tissue is marked (arrow). (B) After aseptic preparation, the tail is amputated by snapping and twisting the caudal tail cranial to the beginning of the necrosis (arrow) to ensure that all diseased tissue is removed. The muscle strands of the proximal tail have been trimmed.

Orthopedic Procedures

For unexpected medical conditions requiring orthopedic procedures, the welfare of the animal may necessitate euthanasia. However, minor injuries or injuries to valuable individuals or

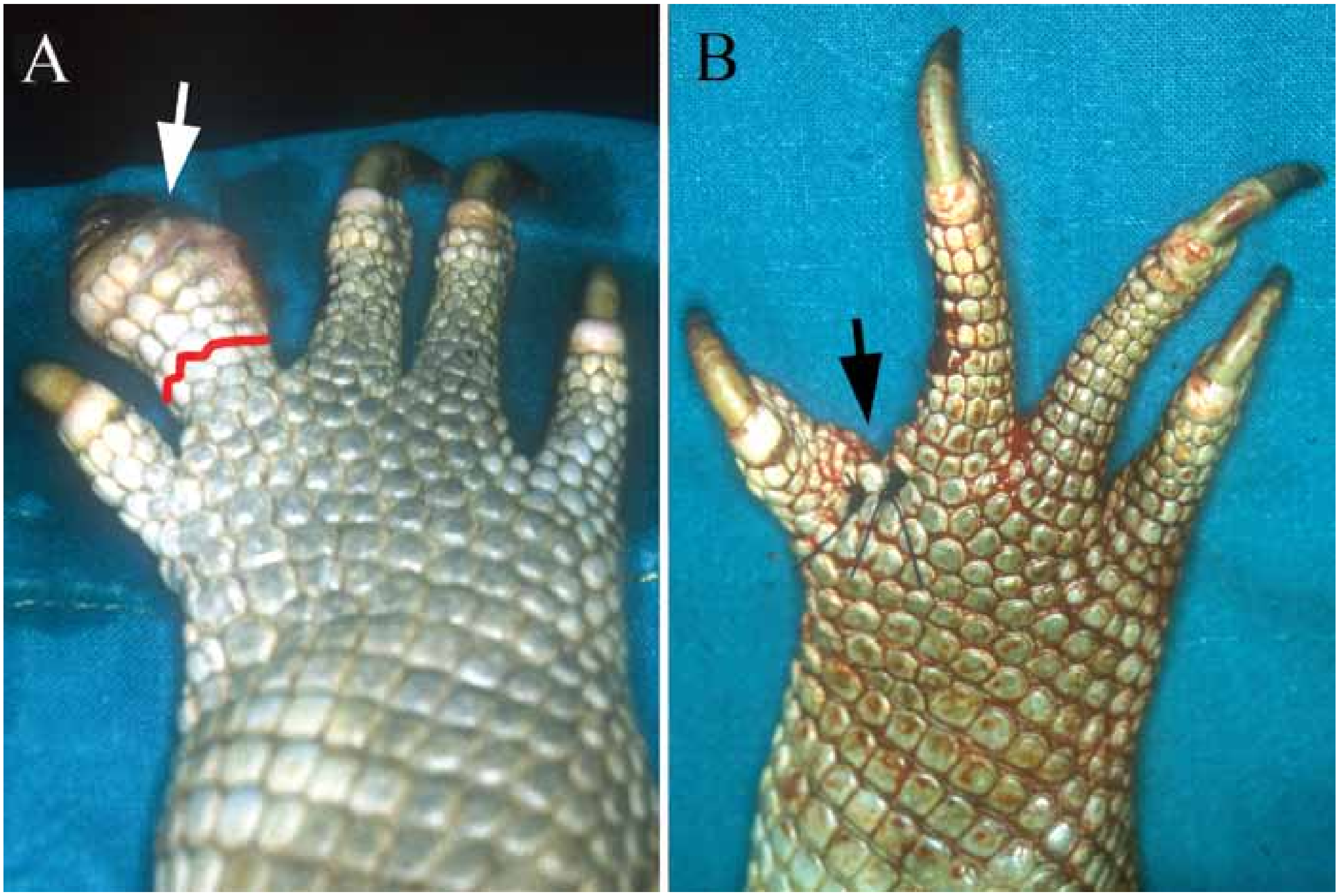

Figure 15. Digit amputation in a Bosc monitor. (A) Preoperative view of the left forelimb, illustrating the planned incision between the scales at the proximal base of digit 2. (B) After amputation, 2 mattress sutures close the defect; any excess skin is trimmed.

endangered species may require humane repair. In addition, research procedures may require orthopedic manipulations such as bone biopsy, grafting, and controlled fracture induction. With orthopedic procedures in reptiles, considerable postoperative care and analgesia must be anticipated.

Amputation. ***Tail amputation.*** Tail amputation is indicated when a reptile presents with a necrotic or diseased tail that fails to respond to medical management. To confirm that the site of amputation is well cranial to all abnormal tissue, radiography is advisable. In lizards that have the capability to perform autotomy (tail loss to evade predation), tail amputation can be most easily accomplished by incising the skin caudal to the site of amputation and simply snapping and twisting the tail through a fracture plane of a coccygeal vertebra[71] (Figure 14). Even though autotomy is a natural phenomenon, anesthesia and aseptic preparation are mandatory. The muscle strands are trimmed, local antiseptic can be applied to the open wound. In iguanids, the flaps are not sutured closed, to allow tail regrowth. For other lizards, which do not have regenerative tails, the skin is closed. More substantial tails that do not lend themselves to autotomy will require a more detailed approach. The surgeon should ensure that sufficient skin is preserved for closure once the soft tissues and vertebra have been transected. 'Tail' (body distal to cloaca) amputation of a snake is not common but can be performed.

Digit and limb amputation. Digits often are traumatized and infected, therefore perhaps necessitating resection. Amputation of the affected digit is recommended whenever osteolysis is evident or chronic inflammation or infection and pain cannot be managed medically. The amputation should be to the level of the metacarpus or metatarsus [5,71](Figure 15). If disease has progressed to include the carpus or tarsus, complete or partial limb amputation should be performed. Partial amputation preserves a stump that a small lizard may continue to use for ambulation; however, stump trauma and infection are common complications in larger reptiles and chelonians. In those cases, complete amputation through the scapulohumeral or coxofemoral joints is recommended.[5,71] The skin incision should include a large skin flap to permit closure. The soft tissues are best transected by using radiosurgery or laser, and although the position of major blood vessels may be unknown, a proximal tourniquet will reduce hemorrhage until major vessels are identified and ligated by using vascular clips or suture. A prosthesis can be provided[71] to assist with ambulation in chelonians, and furniture coasters and various wheels have been used successfully for this application.[11]

Fracture repair. Limb fractures are common in reptiles.[72,80,89-91] Immediate stabilization entails a limb immobilized against the body or tail or inside the shell to prevent further damage. The reptile's overall nutritional and health status must be assessed critically, because bones affected by secondary nutritional hyperthyroidism will not support internal or external fixation.

External coaptation. Radiographs are essential, and multiple views may be required. General anesthesia is required for fracture manipulation as well as repair. Fractures involving the digits can be immobilized by using a ball bandage. A cotton ball is placed in the center of the plantar or palmar aspect. The digits are curled around the ball and taped in position, encasing the entire foot and lower limb in the bandage.[71] Small lizards and crocodilians with fractures of the radius, ulnar, tibia, or fibula can also be treated by using external coaptation.[80] Forelimbs are extended and strapped to the lateral body wall, whereas pelvic limbs are strapped to the tail base.[80] The inclusion of a small wooden splint (for example, wooden tongue depressor) between limb and body or tail helps prevent movement. Alternatively a large syringe case can be placed over the entire limb and bandaged in place.

Fractures of the femur and humerus can be treated in a similar manner, but reduction and healing are usually less than perfect due to movement at the scapulohumeral or coxofemoral joints. Better healing can be expected if a cast is used to immobilize the pectoral or pelvic girdle. In chelonians, restricting a limb within the shell can often produce sufficient fracture reduction to enable a functional repair. Healing typically takes 6 to 12 wk, but physical and radiographic reevaluations should be performed before removing support materials. Callus formation is often not readily appreciated on radiographs because initial healing may be by fibrous union.

External fixation. Modern carbon fiber external fixators are extremely strong and lightweight. They can be used in reptiles by following general surgical principles.[13,99] It is usually impossible to use external fixators for limb repairs in chelonians because of the close proximity of the shell. However, small external fixators have been used to repair chelonian mandibular fractures.[45,75]

Internal fixation. A variety of internal fixation techniques can be used to manage fractures in reptiles. In general, internal fixation follows the same general principles as in mammalian patients.[18,40,94,99] More detailed descriptions for internal fixation in reptiles have been published previously.[71,80]

Shell repair. Shell fractures are common in chelonians,[32,33,60,80,82] and detailed descriptions of management of shell injuries have previously been published.[106,107] Prognosis depends on whether the shell fracture extends into the coelomic cavity and whether the wound is infected. Infected wounds with extensive communication with the coelomic cavity carry the worst prognosis. Separate surgeries may be required to manage the coelomic defect and then repair the shell. If the trauma occurred within a few hours of surgery, the wound is assumed to be contaminated but not infected. The wound can be irrigated with an antiseptic solution, rinsed with physiologic saline, and closed. If the interval has been longer than a few hours, the wound is assumed to be contaminated and infected.[3] In this case, the wound is irrigated with antiseptic solution and managed as an open wound. Covering the wound with a sterile bandage minimizes further contamination. Standard wet-to-dry bandaging will assist the removal of debris over several days, and both systemic and local antimicrobial agents should be considered. Once the wound is free of infected tissue, shell repair can be completed. Radiographs are used to assess whether the bony portion of the shell has been affected. Because the margins of the scutes do not match those of the underlying bony plates, fractures of the bony shell can be missed. Displaced fractures are reduced by using either screws and wire, plates, or other devices. Once the bone fragments are aligned, the shell is cleaned with alcohol or acetone to remove any greasy debris or algae. Small crevices are filled with sterile antibiotic ointment to prevent repair material from penetrating below the shell. Light sanding can help generate a strong bond between shell and repair material. Large defects can be bridged with autoclaved fiberglass patches or car-repair aluminum mesh. Masking tape can be used to protect areas adjacent to the repair site. Epoxy resin and acrylic often are used.[32] Repair material is applied to cover the deficit and allowed to harden. The repair provides fracture immobilization and waterproofing of the defect. Aquatic turtles typically are kept out of water for 24 h after shell repair.

These shell repairs should be considered like a splint. Because they may prevent correct growth of the shell, especially in juveniles, these repair structures should be removed after healing.[51,54] In general most shell repairs are left in place for 6 to 12 mo, although uncomplicated healing of reduced fractures typically takes only 12 to 18 wk.

Reptiles are valuable as models in biomedical research and as subjects in ecology and wildlife studies. This article provides guidance regarding some of the surgical procedures that these animals may require. We hope that this information will enhance the humane care and use of reptiles in research and instruction.

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