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Emergency procedures in the field: a report of wound treatment and fast healing in the giant ditch frog (Leptodactylus fallax)

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Abstract

*A sub-adult, female, giant ditch frog (*Leptodactylus fallax*) (known locally as a mountain chicken) presented with a puncture of the coelomic cavity with partial intestinal evisceration. Improvised field treatment included replacement of the eviscerated intestines and closure of the wound using cotton-tipped applicator swabs. After seven days, the animal's injury appeared to be healed. During the rapid progression of the healing process, the animal showed no adverse effects. This report demonstrates a novel and successful field technique for wound treatment of a serious penetrating injury in an amphibian. It also shows a practical, feasible, and beneficial procedure that improved this animal's welfare and that might be appropriate under field conditions or if standard medical procedures cannot be followed.*

Keywords: *amphibian medicine, animal welfare, field surgery,* Leptodactylus fallax*, minor surgery, wound healing*

Introduction

When working in the field, researchers are often confronted with situations where veterinary medical procedures are needed, either as part of the project in progress or even in unexpected circumstances. Field surgery is used when it is impractical to transport animals to a laboratory or hospital/rehabilitation centre (Philo *et al* 1981) or when transport would cause more harm. Implantation of transmitters for radiotelemetry studies, and/or tags for identification, are among the most common *in loco* minor surgeries in amphibians (eg Sinsch 1988; Corn 1992; Wright 2001). Besides these, reports on amphibian medical procedures in the field are sparse. In addition, these kinds of actions in the field raise ethical issues. For instance, Kirkwood and Sainsbury (1996) suggested that the decision to intervene to treat (as opposed to euthanasing to prevent further suffering) cases of sick or injured free-living wild animals should not be based on welfare grounds alone but should also consider conservation relevance of a species or population and other factors also.

The giant ditch frog (*Leptodactylus fallax*), known locally as the mountain chicken, is one of the world's largest frogs and currently listed as Critically Endangered (Fa *et al* 2010). Adult individuals may reach more than 1 kg and a maximum snout to vent length (SVL) of 210 mm (Lescure 1979; Garcia *et al* 2007). The species once inhabited at least five major islands in the Lesser Antilles, but occurs now only on two islands: Dominica and Montserrat (Lescure 1979; Schwartz & Henderson 1991; Daltry & Gray 1999; Hedges & Heinicke 2007). Over-hunting, together with habitat loss, and introduction of alien predators are major factors affecting both abundance and distribution (Hedges 1993; Kaiser 1994). However, a new emergent threat, the infectious disease caused by the chytrid fungus (*Batrachochytrium dendrobatidis* [*Bd*]), has been responsible for both pushing the Dominica population to extinction (no positive sightings have taken place in the last few years), and more recently, causing sharp die-offs in Montserrat (Garcia *et al* 2007; Young 2008; Fa *et al* 2010; GM Rosa and A Fernández-Loras, personal observation 2007).

In this short communication we report a minor surgery improvised in the field in a giant ditch frog. We also follow and discuss the healing process under natural conditions.

Description

This work was carried out during ongoing research on the *Bd* outbreak that is threatening the giant ditch frog on Montserrat by Durrell Wildlife Conservation Trust, the Zoological Society of London and Parken Zoo, in collaboration with Montserrat's Forestry Department (Stevens & Waldmann 2001). A sub-adult female *L. fallax* was found within the transect along the Fairy Walk *ghaut*, an area in the Centre Hills used for regular monitoring of long-term

changes in amphibian populations (Garcia *et al* 2007) where giant ditch frogs are currently most abundant (Young 2008).

The Centre Hills (740 m), the largest forested area in the island, is located between the Silver Hills in the north and the Soufrière Hills (including an active volcano) in the South, and is characterised by deep valleys with a radial drainage, holding the majority of Montserrat biodiversity (Stevens & Waldmann 2001; Garcia *et al* 2007; Holliday 2009).

The female individual was first captured by hand using latex gloves, measured, assessed for health status and marked with a PIT (ZooChip, AEG, Germany) tag (small transponder implanted under the skin) on the evening of 31 August 2009. No injuries were recorded and the frog was apparently healthy (tag number L5350, SVL 120 mm, hind-limb length 220 mm). As part of the ongoing research project, the animal was sampled for chytrid fungus, including skin swabs that were taken for analysis before release. According to a pre-established protocol (Garcia *et al* 2007), sterile cotton-tipped swabs (MWV100, Tubed Sterile Dryswab™ Tip, Medical Wire & Equipment Co, Bath, UK) were used to gently but firmly swab the skin of the ventral abdomen, drink patch (area that surrounds the vent), and all legs and feet. The swabs were stored under dry conditions. In order to minimise the risk of transmitting disease between individuals and sites, as well as contamination of samples, an appropriate code of practice for fieldwork was followed (Speare *et al* 2004).

On the evening of 28 October 2009, the same female was found near the place where she was previously observed. The weather was cloudy, warm and humid. The frog was caught by hand, wearing latex gloves. The animal had a penetrating puncture wound (approximately 6 mm in width) of the ventral right coelomic cavity resulting in partial intestinal evisceration (approximately 15 mm) (Figure 1A). The injury may have been a bite wound caused by a predator such as a crab or a rat, judging by the shape of the patch of skin missing (Young 2008; GM Rosa and A Fernández-Loras, personal observation 2007). It was decided to improvise a basic surgical treatment *in loco* since, due to logistic constraints, it was not possible to transport the animal and make a subsequent medical intervention. A physical examination carried out prior to surgery, showed no signs of dehydration, lethargy or abnormal respiratory effort. The wet and exposed environment made sterile surgery a challenge. The wound was washed using only clean running freshwater from a stream to remove all the dirt from the injured area. No anaesthetics or sedatives were applied. One person (GMR) was handling and restraining the frog by both hind legs, with the ventral side exposed to allow easy access to the wound; a second person (AFL, a veterinarian) gently pushed the exposed intestinal tract back into the coelomic cavity with the help of two sterile swabs (as above) (Figure 1B). For the wound's surgical closure, an absorbable suture would normally have been required (Wright 2001; Gentz 2007; Poll 2009). However, in the absence of a medical emergency kit and basic surgery items, and since the researchers were unable to get the two wound rims together enough in order to use surgical glue to close the injury, the tips of two other clean swabs were cut off to cover the opened wound. Surgical cyanoacrylate adhesive (GLUture, Abbott, Illinois, USA) was used to join both swab tips to the outer wound cut (in direct contact with the skin), preventing the intestinal tract from re-eviscerating (Figure 1C). No post-surgical antibiotic was administrated. The intervention was done and the female was released in the same place.

On 30 October 2009, the animal was recaptured in the same location. The identity was confirmed via microchip. The weather was cloudy, warm and very humid. The glued swab tips were gone but the intestinal tract was already held inside the coelomic cavity. The healing process was ongoing with the wound still open at this point (approximately 4 mm in width) but no intestines were exposed (Figure 1D). Wound contraction was obvious and no signs of infection were recorded.

On 2 November the wound was stable, presenting slight remains of the cut on the epidermis, and thus considered almost healed (Figure 1E).

The animal was seen after a further two days in apparent good health and showing no clinical signs or marks of the reported episode (Figure 1F). No weight variation was recorded since the first time the individual was caught (190 g).

The sampling swabs were assessed for the presence of *Bd* using quantitative, real time Polymerase chain reaction analysis (qRT-PCR) conducted using an Applied Biosystems Prism 7700 Sequence Detection System; primers were sourced from MWG Biotech AG (Ebersberg, Germany) and the Taqman MGB2 probe from Applied Biosystems (Foster City, USA), in accordance with the protocol clarified by Kriger *et al* (2006). The results came out negative, ruling out the possible interference of this disease in the injury recovery process.

Discussion

The reported wound can be classified as a full-thickness penetrating wound. The healing process occurred predominantly by granulation tissue formation and re-epithelialisation followed by wound contraction (Brown *et al* 1986; Martin 1997). Even though the injury and repair did not occur under aseptic conditions and there was no administration of systemic antibiotics, the wound was completely sealed without any apparent complications after one week.

Peptides with antimicrobial activity are present in the skin of *L. fallax* individuals, which are believed to protect the animals against invading pathogenic micro-organisms (Rollins-Smith *et al* 2005). These skin secretions are responsible, in part, for reducing the risk of post-operative infections (Wright 2001). In the reported case, the two swab tips were used also as a (precarious) retentive dressing, shielding the wound from further injury and gross contamination. In addition, the glue used to affix the swab tips relieved skin tension, allowing for quicker wound healing. This novel and successful field technique proved to be a practical, feasible, and beneficial procedure that improved the welfare of the individual. We suggest that, where veteri-

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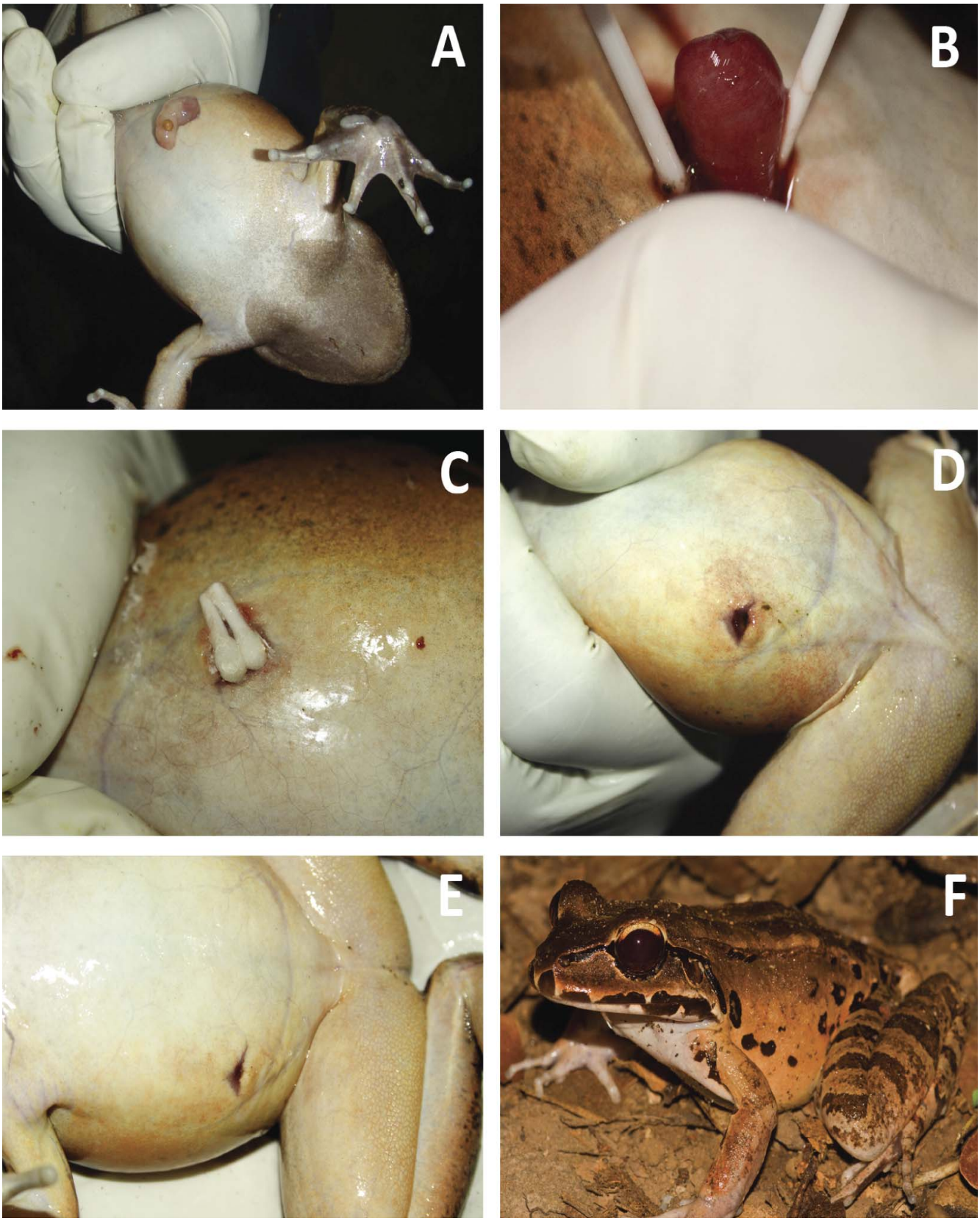
Figure 1



Minor surgery in the field and healing process in *Leptodactylus fallax*. Showing A) a ventral view showing the puncture with part of the intestinal tract exposed, B) the intestinal tract being gently pushed back into the coelomic cavity, C) the improvised dressing of two tips of clean swabs joined by surgical cyanoacrylate adhesive, D) the wound in healing process two days after the surgical procedure, E) the wound five days after the procedure, almost healed, presenting just slight remains of the cut on the epidermis and F) the individual completely recovered after seven days. Images courtesy of GM Rosa.

nary facilities are not available, field workers consider carrying appropriate dressings and treatments (such as pain relief) to enable emergency treatments.

Amphibian medicine is still an emerging area, particularly regarding emergency medicine and critical care, with little published information in peer-reviewed journals (Clayton & Gore 2007). However, working and developing emergency medicine can be worthwhile, especially when dealing with endangered species or populations and interventions might be considered preferable. We strongly believe that researchers and veterinarians can contribute to an improvement of methodologies and techniques through the dissemination of their own experiences reporting empirical procedures and results, even when negative (Anderson & Talcott 2006).

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References

Anderson CD and Talcott M 2006 Clinical practice versus field surgery: a discussion of the regulations and logistics of implanting radiotransmitters in snakes. *Wildlife Society Bulletin 34*: 1470-1471. http://dx.doi.org/10.2193/0091-7648(2006)34[1470:CPVFSA]2.0.CO;2

Brown GL, Curtsinger L, Brightwell JR, Ackerman DM, Tobin GR, Polk HC, George-Nascimento C, Valenzuela P and Schultz GS 1986 Enhancement of epidermal regeneration by biosynthetic epidermal growth factor. *Journal of Experimental Medicine 163*: 1319-1324. http://dx.doi.org/10.1084/jem.163.5.1319

Clayton LA and Gore SR 2007 Amphibian emergency medicine. *Veterinary Clinics of North America. Exotic Animal Practice 10*: 587-620. http://dx.doi.org/10.1016/j.cvex.2007.02.004

Corn PS 1992 Laboratory and field evaluation of effects of PIT tags. *Froglog 4*: 2

Daltry JC and Gray G 1999 Effects of volcanic activity on the endangered mountain chicken frog (*Leptodactylus fallax*). *Froglog 32*: 1-2

Fa J, Hedges B, Ibéné B, Breuil M, Powell R and Magin C 2010 *Leptodactylus fallax*. *IUCN Red List of Threatened Species. Version 2011.2*. http://www.iucnredlist.org

Garcia G, Cunningham AA, Horton DL, Garner TWJ, Hyatt A, Hengstberger S, Lopez J, Ogrodowczyk A, Fenton C and Fa JE 2007 Mountain chickens *Leptodactylus fallax* and sympatric amphibians appear to be disease free on Montserrat. *Oryx 41*: 398-401. http://dx.doi.org/10.1017/S0030605307001012

Gentz EJ 2007 Medicine and surgery of amphibians. *ILAR Journal 48(3)*: 255-259

Hedges SB 1993 Global amphibian declines: a perspective from the Caribbean. *Biodiversity and Conservation 2*: 290-303. http://dx.doi.org/10.1007/BF00056674

Hedges SB and Heinicke MP 2007 Molecular phylogeny and biogeography of West Indian frogs of the genus *Leptodactylus* (Anura, Leptodactylidae). *Molecular Phylogenetics and Evolution 44*: 308-314. http://dx.doi.org/10.1016/j.ympev.2006.11.011

Holliday SH 2009 *Montserrat: A Guide to the Centre Hills*: West Indies Publishing: St John's, Antigua

Kaiser H 1994 *Leptodactylus fallax*. Catalogue of American Amphibians and Reptiles. *Society for the Study of Amphibians and Reptiles 583*: 1-3

Kirkwood JKK and Sainsbury AW 1996 Ethics of interventions for the welfare of free-living wild animals. *Animal Welfare 5*: 235-243

Kriger KM, Hines HB, Hyatt AD, Boyle DG and Hero JM 2006 Techniques for detecting chytridiomycosis in wild frogs. Comparing histology with real-time Taqman PCR. *Diseases of Aquatic Organisms 71*: 141-148. http://dx.doi.org/10.3354/dao071141

Lescure J 1979 Étude taxonomique et éco-éthologique d'un Amphibien des petites Antilles: *Leptodactylus fallax* Müller, 1926 (Leptodactylidae). *Bulletin du Muséum National d'Histoire Naturelle, Paris 1*: 757-774. [Title translation: Taxonomic and eco ethologic study of an amphibian of the lesser antilles *Leptodactylus fallax* (Leptodactylidae)]

Martin P 1997 Wound healing: aiming for perfect skin regeneration. *Science 276*: 75-81. http://dx.doi.org/10.1126/science.276.5309.75

Philo LM, Follmann EH and Reynolds HV 1981 Field surgical techniques for implanting temperature-sensitive radio transmitters in Grizzly bears. *Journal of Wildlife Management 45*: 772-775. http://dx.doi.org/10.2307/3808719

Poll CP 2009 Wound management in amphibians: etiology and treatment of cutaneous lesions. *Journal of Exotic Pet Medicine 18*: 20-35. http://dx.doi.org/10.1053/j.jepm.2008.10.005

Rollins-Smith LA, King JD, Nielsen PF, Sonnevend A and Conlon JM 2005 An antimicrobial peptide from the skin secretions of the mountain chicken frog *Leptodactylus fallax* (Anura: Leptodactylidae). *Regulatory Peptides 124*: 173-178. http://dx.doi.org/10.1016/j.regpep.2004.07.013

Schwartz A and Henderson RW 1991 *Amphibians and Reptiles of the West Indies: Descriptions, Distributions, and Natural History*. University of Florida Press: Gainesville, USA

Sinsch U 1988 Temporal spacing of breeding activity in the natterjack toad, *Bufo calamita*. *Oecologia 76*: 399-407

Speare R, Berger L, Skerratt LF, Alford R, Mendez D, Cashins S, Kenyon N, Hauselberger K and Rowley J 2004 *Hygiene Protocol for Handling Amphibians in Field Studies*. http://www.jcu.edu.au/school/phtm/PHTM/frogs/field-hygiene.pdf

Stevens M and Waldmann G 2001 Animal biodiversity of the Lesser Antillean island of Montserrat (British West Indies): an annotated checklist of terrestrial and freshwater animals. *Archiv Zoologischer Publikationen Band 6*: i-viii, 1-145

Wright KM 2001 Surgical techniques. In: Wright KM and Whitaker BR (eds) *Amphibian Medicine and Captive Husbandry* pp 273-283. Kerala Krieger: Malabar, Florida

Young RP 2008 *A Biodiversity Assessment of the Centre Hills, Montserrat*. Durrell Conservation Monograph No 1. Durrell Wildlife Conservation Trust: Jersey, Channel Islands

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