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IN THIS ISSUE:

Does food provided to captive scavenging wildlife contain veterinary agent residues?

First hand-rearing and translocation of the vulnerable yelkouan shearwater in Malta.

Revisiting the meaning of "science" in the new political climate.

ABOUT THE JOURNAL

THE *Journal of Wildlife Rehabilitation* is designed to provide useful information to wildlife rehabilitators and others involved in the care and treatment of native wild species with the ultimate purpose of returning them to the wild. The journal is published by the International Wildlife Rehabilitation Council (IWRC), which invites your comments on this issue. Through this publication, rehabilitation courses offered online and on-site in numerous locations, and its outreach to those in the profession, the IWRC works to disseminate information and improve the quality of the care provided to wildlife.



On the cover:

Bearded vulture (*Gypaetus barbatus*).

PHOTO © ZWEER DE BRUIN, FLICKR. CC BY-NC-ND 2.0.

Left:

Black bear cub (*Ursus americanus*).

(See *In the News*, page 6.)

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to promote wildlife conservation
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Politics Prompts Reflections on “Science”

Recent news on language the Center for Disease Control was allegedly forbidden from using in describing budget goals led me to reflect on the importance of science to IWRC’s mission. Even the possibility that such an institution, founded with evidence-based human health at its mission’s core, would be barred from using correct technical vocabulary irked me.

Evidence-based practices resulting from true scientific methodology are necessary in advancing our relatively young field of wildlife rehabilitation. The term “science-based” is specifically articulated in IWRC’s mission statement: “Providing science-based education and resources on wildlife rehabilitation to promote wildlife conservation and welfare worldwide.” As such, I took some time to consider why we chose to include science in our mission, a statement in which each word was chosen with care.

Science is integral to IWRC’s efforts in training people in the practice of wildlife rehabilitation and broadcasting updates to practitioners and allies. Our shared use of scientific inquiry provides a way to judge the validity and success of a specific practice.

For example: A rehabilitator sees great vitality in a group of rehab infants given one diet, and speculates that the diet leads to greater fitness and survival to release. Over the next few years, she tracks health markers in infants fed this diet versus those fed a more commonly-used diet. She may ask colleagues to implement a similar protocol and share data. She analyzes results to determine whether the hypothesis proves true. She presents her results at a conference or submits a peer-reviewed paper. Through this, she provides her colleagues the ability to review the process and concur or dispute her analyses.

This example has all the key components necessary to the scientific method: *observation, hypothesis, testing, analysis, and review*. Compare this to a rehabilitator who stops at that first step and *assumes* he has developed a superior diet. This could have been the case regarding increased vitality, but there are

many other possible explanations. Perhaps the parents of those infants were particularly fit and their young inherited genetic traits allowing them to thrive despite a less-than-ideal diet.

I’m intimately familiar with the lack of scientific data available for aspects of wildlife rehabilitation. This makes decision-making difficult for individual practitioners and challenging for instructors and course developers. However, our field is compiling scientific knowledge—piece by piece, animal by animal, datum by datum—structuring a stronger practice of wildlife rehabilitation each year as a result.

Let us take a moment to unpack “science” even further.

Science is a process, not an indivisible fact. Each inquiry refines our understanding of best practices and sets a brick in the foundation of wildlife rehabilitation. A “scientific theory” is not a guess or the result of a single observation. Rather, it is the output of rigorous, structured, repeatable inquiry and testing. This process can cause confusion and consternation among those more familiar with the colloquial use of the word “theory,” meaning an unsubstantiated conjecture or opinion. Such individuals may equate the iterative nature of scientific discovery with weakness, instead of realizing that constant inquiry combined with skeptical analysis, as practiced in science, is a great strength that keeps us from doing things solely because that is the way they have always been done. As we expand our knowledge, we learn, we change, and we improve.

This structured approach to inquiry within a community of individuals with a shared commitment to critical analysis, transparency, and continually-improving care protocols is why science is so central to IWRC’s mission.

This exploration of science and wildlife rehabilitation will be continued in the editorial of 38(2).

— Kai Williams
Executive Director

Turtle Seizure Rehab Needs Elicit Multi-Country Response

IFATY, Madagascar (April 18, 2018)—On Tuesday, April 10, more than 10,000 critically endangered radiated tortoises (*Astrochelys radiata*) were discovered by local police in a nondescript private residence in Toliara, Madagascar. The floors of virtually every room in the house were covered with tortoises that had no access to food or water. As of Friday, April 13, hundreds had died from dehydration and illness. Experts from the Turtle Survival Alliance (TSA) and several zoos and aquariums have been dispatched with medical supplies, and will administer medical and general care for the sick or injured tortoises.

It is not known how long the tortoises have been in the home. Some arrests have been made. The local police, in partnership with Directeur Regional de l'Environnement, de l'Ecologie, et des Forets (DREEF) and the conservation law enforcement authorities in Madagascar, continue their investigation. It is believed that the tortoises were collected for the illegal pet trade, possibly for shipment to Asia where the tortoises' highly-domed shell featuring a brilliant star pattern makes them highly prized. It is estimated that radiated tortoise populations in the wild have declined more than 80 percent in the last 30 years. At this rate of decline, it is estimated that the radiated tortoise could be functionally extinct in the wild in less than two decades.

Currently, triage efforts are being led by a five-member team from the Turtle Survival Alliance's Madagascar staff, Durrell Wildlife Conservation Trust, and Villages des Tortues, who have been working non-stop after relocating the surviving tortoises 18 miles north at SOPTOM-Villages des Tortues, a 17-acre private wildlife facility in Ifaty. While there, each tortoise will receive initial in-processing, health evaluations, hydration, and triage.

"I don't think the word "overwhelming" comes close to describing what the Turtle Survival Alliance is dealing with



Radiated Tortoise (*Astrochelys radiata*). PHOTO © SEBASTIAN NEIDLICH. CC BY-NC-ND 2.0 LICENSE.

here," said Rick Hudson, President of the Turtle Survival Alliance. "We were already caring for 8,000 tortoises in Madagascar; now that number has more than doubled overnight."

Participating organizations accredited by the Association of Zoos and Aquariums (AZA) include Abilene Zoo, Bronx Zoo/Wildlife Conservation Society, Columbus Zoo and Aquarium, Dallas Zoo, Dickerson Park Zoo, Georgia Aquarium, Fort Worth Zoo, New England Aquarium, Oklahoma City Zoo and Botanical Garden, San Diego Zoo Global, Shedd Aquarium, Tennessee Aquarium, Topeka Zoo and Conservation Center, Tulsa Zoo, Utah's Hogle Zoo, Zoo Knoxville, and Zoo Atlanta.

In addition to these AZA organizations, the TSA's efforts are being supported by global conservation partners Aktionsgemeinschaft Artenschutz e.V. (AGA), Durrell Wildlife Conservation Trust, ProWildlife v.E., SOPTOM-Village Des Tortues, Tanganyika Wildlife Park, the Turtle & Tortoise Preservation Group, and the Auckland Zoo in New Zealand, plus a growing number of private donors.

"We are in an "all-hands-on-deck" mentality right now," said Hudson. "For-

tunately, due to our strong relationship with the zoo community, the TSA is well positioned to respond to crises such as this."

"The immediate response of more than 20 AZA-accredited facilities, offering their expertise and assistance to care for thousands of tortoises in Madagascar, is proof we will take whatever action is necessary to address illegal wildlife trade and other threats that put the world's most vulnerable species at risk of extinction," said AZA President and CEO Dan Ashe. "Through programs like SAFE: Saving Animals From Extinction and the U.S. Wildlife Trafficking Alliance, AZA and its members are engaging in critical, coordinated, and needed conservation work."

Given the scale of the rescue efforts, TSA expects to send additional teams of veterinary experts from the United States to Madagascar over the coming weeks and months.

"The support we continue to receive from the global conservation community has been incredible, and we are extremely thankful for the multitude of individuals and organizations that have come forward with donations and supplies," said Hudson. "Yet, the long-term financial impacts

to our Madagascar program are potentially crippling.”

Zoological institutions, private veterinary practices, husbandry technicians, or additional support personnel interested in assisting can directly contact Andrew Walde, Chief Operations Officer, at awalde@turtlesurvival.org.

Bear Rehabilitation Allowed in Alberta

ALBERTA, Canada (April 18, 2018)—The Government of Alberta has introduced a new policy that will allow wildlife rehabilitation facilities in Alberta to accept orphaned black bear cubs.

The new policy allows Alberta Fish and Wildlife staff to work with rehabilitation facilities to ensure orphan black bear cubs are safely returned to the wild whenever possible. Once approved, such facilities will be able to accept black bears less than one year of age.

Shanon Phillips, Minister of Environment and Parks, notes, “Alberta’s orphaned black bear policy is based on the best available scientific research, modern rehabilitation practices, compassion for these animals, and the safety of people. We want black bear cubs to grow up and thrive in the wilds of Alberta.”

The policy includes a draft protocol that sets the requirements surrounding bear feeding, the suitability of the physical space in which the bears are kept, appropriate veterinary care, and kinds of interactions the bears may have with humans.

The draft protocol is the result of more than a year of consultation, research, and engagement. Wildlife management biologists will continue to work with rehabilitation facilities to ensure bears can be returned to the wild without causing issues in populated areas.

This means the bears are able to forage on their own, are appropriately social with other bears, and are less likely to become a part of human-bear conflict.

After being assessed, the bears will be released on or before Oct. 15 of the year they arrived at the facility, and will not be overwintered unless special approval is given.

Released bears will be fitted with monitoring devices, such as ear tags, and will be tracked by scientists to ensure successful reintegration into the wild.

There are more than 40,000 black bears in Alberta. In a given year, there are approximately 10,000 black bear cubs born. British Columbia, Saskatchewan and Manitoba allow the rehabilitation of black bear cubs younger than one year of age. The review of Alberta’s bear rehabilitation protocol began after three black bear cubs were found in a washroom in Banff National Park on April 1, 2017. There are 10 permitted wildlife rehabilitation facilities in Alberta. These facilities provide a service that many Albertans use and value.

After 8 years without bear rehabilitation, centers are in the process of setting up appropriate rehabilitation facilities for orphaned cubs.

Contested WA Deer Released

OLYMPIA, Washington, USA (April 13, 2018)—The owners of a wildlife rehabilitation center in south Thurston County today released 11 young deer into the wild in collaboration with the Washington Department of Fish and Wildlife (WDFW), which previously euthanized four animals at the facility due to concerns about their care.

Claudia and David Supensky, owners of For Heavens Sake Animal Rescue & Rehabilitation, transported eight of the deer from their facility in Rochester and released them on a private thousand-acre land preserve surrounded by wildlands. The other three were released at a nearby location later in the day.

In December WDFW agreed to not euthanize any more deer last winter, following WDFW’s action Nov. 9 to euthanize three fawns and an elk calf removed from the For Heaven’s Sake Animal Rescue & Rehabilitation facility in Rochester, where state wildlife managers found those animals—all males—to be habituated to humans and unfit for release into the wild.

“The department has a responsibility to intercede when animals become too habituated to humans to survive in the wild,” said Eric Gardner, chief of WDFW’s

Wildlife Program. “We removed four animals that displayed signs of severe habituation, but we’ve agreed to work with the owners to find a mutually acceptable solution for the other deer in their care.”

Gardner described the Supenskys, who have been licensed to operate their facility since 2010, as “caring people who work hard on behalf of the animals in their care.” But, responding to reports from concerned citizens, a WDFW veterinarian and other wildlife specialists observed signs of habituation among the deer at their facility during a series of visits starting in August.

“Those animals showed that they had lost their fear of humans and were still looking to be fed at a time when they should have been weaned and avoiding people,” said Gardner, noting that state regulations generally prescribe that such animals be euthanized.

“We are grateful to WDFW for giving the 11 remaining fawns time for evaluation and approving them for release,” said Claudia Supensky. “We appreciate Brian Calkins for including us during this process.”

Calkins said the Supenskys worked hard on behalf of the animals in their care, and have followed the department’s instructions to give the animals the best chance of developing the instincts needed to survive in the wild.

“The public really values the work that our wildlife rehabilitators do, and we want to support them in their efforts to prepare animals to return to the wild,” he said.

Under their agreement with WDFW, the Supenskys minimized contact with the remaining deer, avoided hand-feeding them, and weaned any of them that were still bottle-feeding. If they choose to keep taking ungulates, the agreement commits them to work with WDFW to develop a corrective action plan for their facility.

Gardner noted that there are 30 licensed wildlife rehabilitation facilities in Washington, most of which are registered non-profit organizations that rely on donations and grants to cover their operating expenses. Many of the animals that wind up in those facilities were “rescued” by

CONTINUED ON PAGE 28

First hand-rearing and translocation of Vulnerable yelkouan shearwaters, *Puffinus yelkouan*, in Malta

Nicola Piludu¹, Edward Jenkins¹, Julia Gulka,² and Eurydike Kovacs^{1,3}



Chick 2 being fitted with a ring before its release. PHOTO ©HSBC MALTA.

Introduction

Recent taxonomic changes have led to the splitting of shearwater species, including the yelkouan shearwater *Puffinus yelkouan* and the Balearic shearwater *P. mauretanicus*, which were recently recognized as separate from the Manx shearwater, *P. puffinus*.¹⁻³ Since this taxonomic elevation, the Critically Endangered Balearic shearwater has been a major research focus⁴⁻⁶ despite remaining knowledge gaps in the life history of the Vulnerable yelkouan shearwater.⁷

The yelkouan shearwater is endemic to the Mediterranean basin, with known colonies either in decline⁸ or understudied.⁹ The species has established colonies of approximately 1,660–1,980 breeding pairs (about 10% of the world population) on cliffs across the Maltese archipelago and has been the target of extensive research and conservation efforts by BirdLife Malta (2016).

¹BirdLife Malta, Xemxija, Malta

²University of Manitoba, Department of Biological Sciences, Winnipeg, Manitoba

³Vetcare Animal Clinic, Misrah Lourdes, San Gwann SGN2010, Malta

ABSTRACT: Gaps exist in our knowledge of the life history of the Vulnerable yelkouan shearwater *Puffinus yelkouan*, which is endemic to the Mediterranean basin. This work documents the first instance of hand-rearing and translocation of two yelkouan shearwater chicks rescued off the coast of Malta during the 2016 breeding season. The chicks were hand-reared from estimated ages of 5 and 33 days, respectively, fed fish daily, and translocated to artificial burrows around the ages of 77 and 80 days, respectively. Both showed healthy growth curves and, based on condition and behavior, are presumed to have fledged naturally. While the cause of the chicks' abandonment remains unknown, this experience provides insight into yelkouan shearwater chick growth and a method for hand-rearing the species, especially in the context of stranding and translocation.

KEYWORDS: conservation management, nest box, *Puffinus yelkouan*, rehabilitation, seabird, translocation, yelkouan shearwater.

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Seabirds face various threats; population decline has been reported for 47% of species worldwide.¹¹ Yelkouan shearwaters in Malta are no exception.¹² The species is threatened by anthropogenic disturbances (including chemical, light, and sound pollution), predation by non-native mammals, fishery bycatch,¹³ and microplastics.¹⁴ On-going conservation efforts include controlling rats at key colony sites, reducing light pollution from industrial sites, addressing disturbances by marine traffic, and raising public awareness of the species.¹⁰

Due to the Vulnerable status of the population, ensuring chick survival is imperative. Hand-rearing abandoned seabird chicks, especially after the death of their parents at sea, is a common practice for species of concern to conservationists.¹⁵ Furthermore, translocating hand-reared chicks to colonies in the wild, often using artificial nest sites for housing chicks prior to fledging, has proven to be a successful technique for population restoration and is now widespread.^{16–20} Although hand-rearing and translocation have been successfully employed for other shearwater species,²⁰ neither had been used for yelkouan shearwaters.

During the 2016 breeding season, two chicks were found by members of the public during two separate incidents at Blue Grotto, Qrendi, on Malta. This prompted the first hand-rearing of yelkouan shearwaters. This paper includes a case study of the experience, provides a method for hand-rearing, translocating, and releasing yelkouan shearwaters, and presents data on fledgling growth, development, and condition.

Methods

Recovery

The Blue Grotto (35°821'N, 14°456'E) is a southern Maltese cave system surrounded by steep limestone cliffs. It hosts colonies of both yelkouan and Scopoli's shearwaters *Calonectris diomedea*. The area is a popular tourist destination and is highly disturbed by human activity (e.g., boat traffic, snorkeling, and eateries). Com-

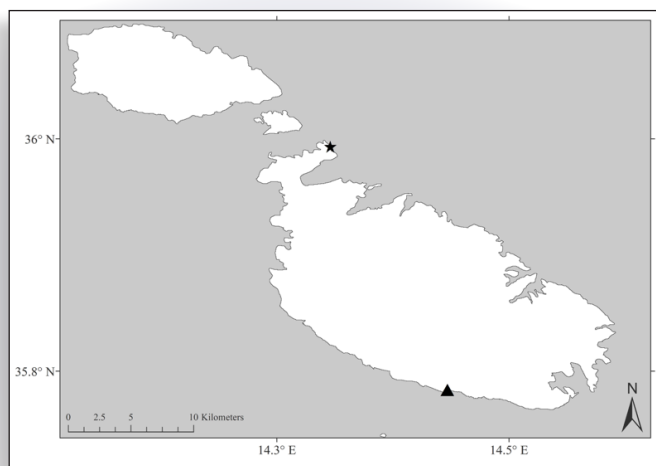


FIGURE 1. Recovery and translocation sites for two yelkouan shearwater chicks on Malta, 2016. Recovery occurred at Blue Grotto, Qrendi (▲). Chicks were translocated and released at Rdum tal-Madonna (★).

mercial boat operators rescued two yelkouan shearwater chicks in this area on the first and 18th of May (Fig. 1) and contacted the BirdLife Malta bird rescue team. Both chicks were discovered floating in the sea below known nesting areas. The age of the first chick (hereafter, Chick 1) was estimated at five days, and the age of the second (hereafter, Chick 2) was estimated at 33 days. Because the chicks' specific nest locations were unknown, the chicks could not be returned to their natal site.

Diet and feeding

Both chicks were moderately active and warm at rescue. They were immediately hydrated with 5 ml of water administered through a syringe gently inserted into the birds' throats. This amount of water was chosen based on previous experiences with similarly-sized birds under veterinary supervision. The chicks were then left to rest for two hours. As Chick 1 did not show signs of stress (e.g., regurgitation), it was then fed 20 g of white fish (*Gadus sp.*) dipped in sunflower oil and water. This was administered by gently pinching the chick's upper mandible and inserting small pieces of fish into its mouth. The chick's feathers were partially contaminated by the oil. Syringe-feeding (rather than hand-feeding) is recommended for avoiding contamination. After the first day, 100 g tins of sardines in vegetable oil (72% sardines and 28% oil) were the main feed. The contents of each tin was combined with 50 ml of water and mixed to form a liquid paste that could be administered with a syringe. Food was prepared every other day and kept refrigerated in a sealed plastic container. As the feeding protocol was already in place when Chick 2 was rescued, the second chick was fed 20 g of sardines via syringe following hydration.

As a sudden switch from a fresh fish diet in the wild to a tinned sardine diet could be fatal to debilitated seabird chicks, caution was employed. The chicks were initially fed a total of 20 g²¹ which was gradually increased according to the chicks' weight gain to 60 g, following a conservative approach based on the method of Gummer and Adams.²¹ At 35 days of age, when the chicks could swallow larger portions, whole frozen big-scale sand smelt *Atherina boyeri* was slowly integrated into the tinned sardine diet. A total of 10 g of smelt was given to each chick on their 35th day, followed by 20 g daily over the following four days and then 30 g daily until their release.

Feeding Technique

It was immediately noted that the chicks responded positively to handling, and although restraint during feeding is suggested by Gummer and Adams,²¹ it was not deemed necessary. The chicks were placed on a flat surface covered with paper. Handlers administered the food to each chick by gently pinching the upper mandible, extending the bird's head up and outwards, and gently inserting the feeding tube, avoiding the trachea opening.^{20,21} A 10 ml syringe fitted with a 5 cm plastic feeding tube was used to deliver feedings. Chunks of whole fish were offered, which the chicks actively took and swallowed.

Feeding Schedule

Growth data were collected beginning on the third day of the birds' hand-rearing, when the protocol was implemented. This was immediately applied when Chick 2 was rescued. The chicks were weighed prior to their first feeding of each day with a 500 g Pesola scale. Four meals were delivered daily for the first month. This was reduced to three daily meals for the 31st through the 44th days of age and reduced again to two at 45 days of age. After translocation, the chicks were fed once every two days. See Appendix A for the full feeding schedule.

Veterinary care

During hand-rearing, both chicks encountered minor health problems. Chick 1 initially presented with an abnormal skin condition in the neck area, which spread to the face and eyes and resulted in feather loss and eyes closing at 20 days of age. Although oil contamination may have exacerbated the condition, this was determined to be stress-related, and the chick was prescribed Canina Petvital[®] vitamin mix for birds^a and eye antibiotic ointment (chloramphenicol, containing 1% posifenicol). Including vitamin and calcium supplements is recommended, especially in the context of a frozen fish-based diet. Additionally, because ointment can contaminate feathers, using eye drops instead is recommended. Both products were administered during feeding. The antibiotic was discontinued at age 27 days because the condition improved. The vitamin supplement was continued throughout the hand-rearing period.

Similarly, Chick 2 was rescued with a small mass on its upper left eyelid. The mass was removed by a veterinary surgeon the day after rescue, and the eye was treated with the same antibiotic ointment used with Chick 1. Discharge was noted that evening only and was not present afterwards, and the treatment was interrupted on the fourth day of hand-rearing at an estimated age of 33 days. Chick 2 was also administered Canina Petvital vitamin mix throughout the hand-rearing period.

Housing conditions

During the rearing stage before translocation, the chicks were kept in partially closed cardboard boxes. Simulating the darkness of the natural burrow while allowing ventilation was important. A layer of newspaper was laid down and changed each day to avoid contamination of feathers by feces. However, contamination occurred, especially in the cloacal area. Using sand instead of newspaper as a substratum is therefore recommended. If used, paper without ink, such as newspaper ink, is preferable because of the ink's potential negative effects on natural waterproofing.

^aComposition: Plant oils and fats, emulsifying agent, vitamin premix. Additives per 100 ml: Vitamin A 50,000 IU, Vitamin D3 10,000 IU, Vitamin E (as alpha-tocopherol acetate) 1,000 mg, Vitamin K3 100 mg, Vitamin B1 600 mg, Vitamin B2 1,000 mg, Vitamin B6 250 mg, Vitamin B12 1,000 mcg, nicotine acid 4,000 mg, pantothenic acid 1,000 mg, folic acid 10 mg.



Chick 1 at two weeks of age. PHOTO BY BIRDLIFE MALTA.

Translocation site

An existing colony of yelkouan shearwaters at Rđum tal-Madonna (35.993°N, 14.372°E), in northeast Malta, was the translocation site (Fig. 1). Lying about 30 km from the recovery site, the colony site faces northeast (in contrast to the south-facing recovery site). The site was chosen for its logistics, its pre-existing predator control regime, and its status as the most-studied colony in Malta. It is also the largest colony in Malta, with a population of 398–602 breeding pairs.²² These efforts have led to the successful fledging of yelkouan shearwater chicks, including those transferred from pre-existing artificial burrows (unpublished). Guidelines for translocating fluttering shearwaters *P. gavia* were used for reference.²¹

Artificial burrow design

Two artificial burrows were built from 12 mm of marine plywood, according to an original design (Metzger pers. comm; Fig. 2). Assembled on-site, each contained a large interior chamber and a smaller exterior chamber with entrances 10 cm wide and 9 cm high. These dimensions were intended to prevent larger prospecting Scopoli's shearwaters from disturbing or ejecting the chicks.²³ Five days before translocation, the artificial burrows were placed on a flat rock in a shallow cave to provide shade, among boulders near nest crevices (distance <1 m) used by wild birds. A shallow layer (<1 cm) of sand from the immediate area was added to simulate the eroded soft limestone found in the deep natural crevices.

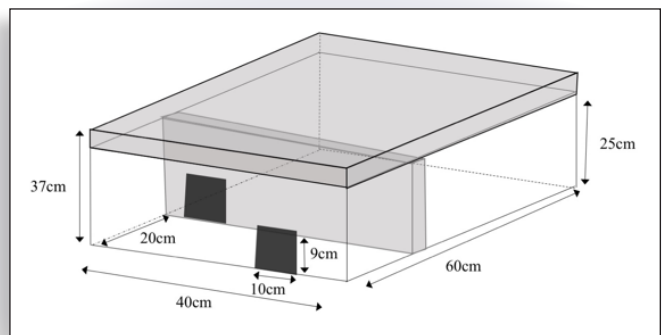


FIGURE 2. Artificial burrow design used for translocated yelkouan shearwater chicks at the Rđum tal-Madonna breeding colony.

TABLE 1. Recovery and fledging measurements of yelkouan shearwater chicks recovered from Blue Grotto, Qrendi, Malta in May 2016. Fledging measurements represent the last time chicks were found in artificial burrows prior to presumed fledging.

RECOVERY		FLEDGING			
ESTIMATED AGE (DAYS)	MASS (G)	ESTIMATED AGE (DAYS)	WING LENGTH (MM)	MASS (G)	DATE (2016)
6	35	77	207	340	6TH JULY
33	81	80	214	330	4TH JULY

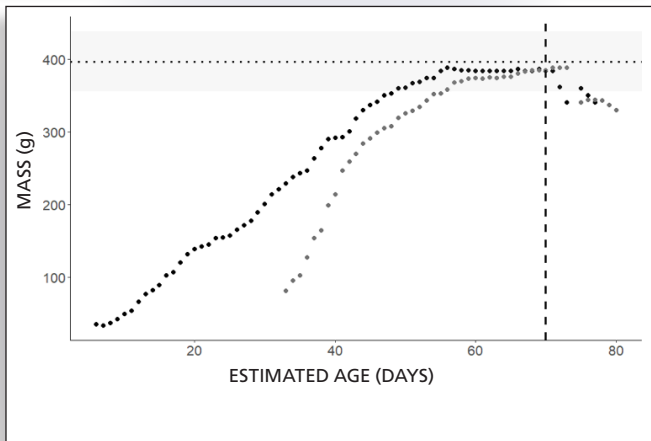


FIGURE 3. Mass growth data for two individual yelkouan shearwater chicks over hand-rearing period. Chick 1 (black) and Chick 2 (grey) were rescued at ages 6 and 33 days respectively. Dotted horizontal line represents mean adult weight with shading \pm standard deviation. Dotted vertical line represents age of translocation and reduced feeding rate from daily to every two days until fledge. Note a decrease in mass prior to fledging is common in Procellariiformes and is associated with a reduced provisioning rate²⁴.

Translocation and fledging

Prior to translocation, both chicks were ringed with uniquely numbered metal rings provided by the Valletta Ringing Scheme, run by Birdlife Malta. Chick 1 was translocated at an estimated age of 71 days and a mass of 384 g; Chick 2 was translocated at an estimated age of 73 days and 388 g. A 200 mm long wing cord was a criterion in the choice of translocation date.

The box entrance was covered with plastic mesh blockades to allow the chicks to become accustomed to their surroundings without leaving prematurely.²¹ The mesh was removed 4–5 days later.

After translocation, camera traps (Bushnell Trophy Cam HD Aggressor No-Glow[®]) were placed at the entrance of each artificial burrow to monitor the emergence and fledging behavior of the chicks. Yelkouan shearwater chicks in the Rdum tal-Madonna colony and elsewhere in Malta were being monitored concurrently in a separate, long-term study to allow researchers to compare the two hand-reared chicks with their wild counterparts (BirdLife Malta, unpublished).

Results

Growth rate

Before translocation, Chick 1 gained an average of 5.37 ± 0.62 g (SE) daily, while Chick 2 gained 7.67 ± 1.40 g daily. Although both chicks were alert and reactive upon rescue, Chick 2 was extremely emaciated. This resulted in much quicker weight gain for Chick 2. The daily feeding regimen resulted in weight gain for both chicks until they were 55 ± 5 days old, when their weight leveled off at approximately 370–380 g (Fig. 3). After translocation, feedings occurred once every two days. Overall, mass decreased by an average of 7.3 ± 5.9 g for Chick 1 and 8.5 ± 4.2 g for Chick 2 during this period, when times of visits and feedings could not be consistent because of field conditions.

Fledging

The chicks were translocated when they were approximately the same age and size (Table 1). Photo analysis revealed that both spent nights near the entrance of the box during the mesh-on period, retreating out of view during the day. Once the mesh was removed, both chicks spent the first night outside the burrows (presumably training their wings) and returned to their artificial burrows during the day. Both chicks were in the artificial burrows for 2–3 days following mesh removal and presumably fledged naturally. The chicks appeared healthy until fledging, as gauged by the quality and cleanliness of their plumage, their alertness, and their vociferous reaction to the researchers. Once the chicks were found to have left the burrow, the immediate area was thoroughly examined to make sure that the chicks had not been preyed upon or moved to another crevice.

Discussion

Chick discovery

Given that the BirdLife Malta rescue team had not experienced similar events before, the team did not have a protocol for the rehabilitation of yelkouan shearwater chicks when Chick 1 was retrieved. For the first two days, care was provided according to the best available knowledge, which was fragmentary. Diet for the species had been studied,²⁵ but at the time of the first rescue, no data on the species' development or guidelines on its hand-rearing were available. Although dead yelkouan shearwater chicks, presumably abandoned, have been found in burrows across the island (Metzger, pers. comm.), these were the first live chicks retrieved outside of their nest. A third chick had been found alive in similar circumstances on an unspecified date between the two incidents but had not been reported; the fate of the chick is unknown, but it is presumed to have drowned. Ascertaining the cause of the chicks' ejection from their burrows was not possible, and finding two young chicks in this manner had not been previously reported. Starvation, abandonment by inexperienced breeders, or loss of a parent can cause seabird chicks as young as one week old to leave their nest site despite having limited mobility. Nevertheless, the extremely low weight of Chick 2 at rescue strongly suggests chick abandonment and resultant starvation before stranding.

Other possible causes have been identified. Black rats *Rattus rattus* are present in the area and predate eggs and young chicks.²² Brown rats *R. norvegicus* are also found in Malta, but their impact on breeding seabirds is unknown. Yelkouan shearwaters in Malta often nest in crevices that extend out directly from a central cavity and are adjacent to a shared entrance. Adults fighting in these areas have been recorded by infrared camera (BirdLife Malta, unpublished). This fighting can dislodge chicks occupying less protected sites. Interspecific competition for nest cavities (between the later-breeding and larger [505–795 g] Scopoli's shearwater and the smaller [349–416 g] yelkouan shearwater) is possible. This competition could also lead to chick ejection, although occurrences have not been recorded.²⁶ Similar studies have found that the closely related Cory's shearwaters *C. borealis* eject smaller Procellariiformes when nesting sympatrically.²⁷ The effects of major anthropogenic disturbance at the site (i.e., engine sound pollution, voices directly below the colony, and light pollution at night) have not been thoroughly investigated but may affect chick behavior. Other possibilities include rock movement and cavity collapse.

The chicks' having come from the same location in southwest Malta can be explained by the high level of human activity in Blue Grotto relative to colony sites on more remote cliffs, and their coincident rescue should not be interpreted as evidence that the area has a higher rate of chick strandings. The cause of the strandings remains unknown and further research is recommended, especially if similar cases continue to occur.

Diet and Growth

Adaptations of shearwaters and other Procellariiformes are beneficial to hand-rearing chicks. These adaptations include the build-up of large reserves of lipids as buffers against irregular food intake²⁸ and the rapid development of homeothermic capabilities, which free adults from brooding and allow them to forage earlier in chick development.²⁹ Yelkouan shearwater feeding rates vary with resource availability and other oceanic factors, thus these adaptations are beneficial given the variable rate, time, and energetic content of feedings that can occur when hand-raising chicks. Despite these variations, the chicks appeared to follow a predicted growth curve when compared to fluttering and Manx shearwaters.^{21,30} Chick mass plateaued at approximately 388 g, and chick mass at fledging was between 330 and 340 g. While there is no information on the fledging mass of yelkouan shearwater chicks, weight loss prior to fledging of shearwaters and other seabirds is common.²⁴ Manx shearwater chicks lose an average of 14% of their body mass prior to fledging,³⁰ thus the yelkouan shearwater chicks' loss of 12–15% body mass prior to fledging suggested that they fledged at healthy weights.³ Emergence and fledging behavior, including emerging from the burrow for several nights prior to fledging, followed a trajectory similar to that recorded for conspecifics,³⁰ suggesting that the chicks fledged successfully rather than wandered off or fledged prematurely (i.e., having left immediately after mesh removal). While the 2–3 day period between mesh removal and fledging was short in comparison to

the 9-day desertion period of the closely related Manx shearwater,³⁰ this may be a product of the artificial feeding regime or specific to yelkouan shearwaters in Malta.¹⁴

Conclusions

This paper presents the method employed during the first instance of yelkouan shearwater hand-rearing and translocation. Although the sample is very small, the paper is intended to guide conservationists in rescues by providing a protocol for hand-rearing and translocation, given the absence of development data for yelkouan shearwater chicks. We conclude that the hand-rearing and translocation of the yelkouan shearwater chicks was a success. The fates of the chicks will be known only if they are recaptured or recovered, events that may happen given that the species is strongly philopatric. Satellite tagging studies have shown that chicks fledging from the same colony disperse and range over large areas of the eastern Mediterranean,³¹ potentially returning at three years of age.¹² We encourage researchers to consider hand-rearing if similar events occur, especially for understudied species with declining populations.

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Appendix A

Feeding schedule for hand-reared yelkouan shearwater chicks including estimated age, weight at 8 AM prior to the first feeding of the day, amount of food given, and number of feedings

per day along with diet. Diet items include sardines (*Clupidae* sp.) and big-scale sand smelt (*Atherina boyeri*). Asterisk indicates date of translocation.

CHICK 1

AGE (DAYS)	WEIGHT (G)	FEED (G)	TIMES	DIET
5	-	20	4	SARDINES
6	35	20	4	SARDINES
7	33	20	4	SARDINES
8	37	20	4	SARDINES
9	42	25	4	SARDINES
10	49	25	4	SARDINES
11	54	30	4	SARDINES
12	66	27	4	SARDINES
13	77	25	4	SARDINES
14	82	30	4	SARDINES
15	89	30	4	SARDINES
16	102	30	4	SARDINES
17	107	37.5	4	SARDINES
18	120	37.5	4	SARDINES
19	132	30	4	SARDINES
20	139	30	4	SARDINES
21	142	40	3	SARDINES
22	145	40	3	SARDINES
23	154	30	3	SARDINES
24	155	40	3	SARDINES
25	157	50	3	SARDINES
26	165	50	3	SARDINES
27	171	50	3	SARDINES
28	178	50	3	SARDINES
29	189	50	3	SARDINES
30	201	50	3	SARDINES
31	214	50	3	SARDINES
32	221	50	3	SARDINES
33	229	50	3	SARDINES
34	238	50	3	SARDINES
35	243	50	3	40G SARDINES, 10G SMELT
36	247	50	3	30G SARDINES, 20G SMELT
37	263	50	2	30G SARDINES, 20G SMELT
38	278	50	2	30G SARDINES, 20G SMELT
39	290	50	2	30G SARDINES, 20G SMELT
40	292	60	2	30G SARDINES, 30G SMELT
41	293	60	2	30G SARDINES, 30G SMELT
42	301	60	2	30G SARDINES, 30G SMELT
43	318	60	2	30G SARDINES, 30G SMELT
44	330	60	2	30G SARDINES, 30G SMELT
45	337	60	2	30G SARDINES, 30G SMELT

AGE (DAYS)	WEIGHT (G)	FEED (G)	TIMES	DIET
46	341	60	2	30G SARDINES, 30G SMELT
47	350	60	2	30G SARDINES, 30G SMELT
48	353	60	2	30G SARDINES, 30G SMELT
49	360	60	2	30G SARDINES, 30G SMELT
50	361	60	2	30G SARDINES, 30G SMELT
51	367	60	2	30G SARDINES, 30G SMELT
52	369	60	2	30G SARDINES, 30G SMELT
53	374	60	2	30G SARDINES, 30G SMELT
54	374	60	2	30G SARDINES, 30G SMELT
55	384	60	2	30G SARDINES, 30G SMELT
56	388	60	2	30G SARDINES, 30G SMELT
57	386	60	2	30G SARDINES, 30G SMELT
58	385	60	2	30G SARDINES, 30G SMELT
59	385	60	2	30G SARDINES, 30G SMELT
60	384	60	2	30G SARDINES, 30G SMELT
61	384	60	2	30G SARDINES, 30G SMELT
62	384	60	2	30G SARDINES, 30G SMELT
63	384	60	2	30G SARDINES, 30G SMELT
64	384	60	2	30G SARDINES, 30G SMELT
65	384	60	2	30G SARDINES, 30G SMELT
66	386	60	2	30G SARDINES, 30G SMELT
67	384	60	2	30G SARDINES, 30G SMELT
68	384	60	2	30G SARDINES, 30G SMELT
69	386	60	2	30G SARDINES, 30G SMELT
70	384	60	2	30G SARDINES, 30G SMELT
71*	384	90	1	SARDINES
72	-	-	-	
73	340	90	1	SARDINES
74	-	-	-	
75	360	90	1	SARDINES
76	-	-	-	
77	340	80	1	SARDINES

CHICK 2

AGE (DAYS)	WEIGHT (G)	FEED (G)	TIMES	DIET
33	81	20	2	SARDINES
34	95	30	3	SARDINES
35	102	40	3	40G SARDINES, 10G SMELT
36	127	50	3	30G SARDINES, 20G SMELT
37	154	50	3	30G SARDINES, 20G SMELT

CONTINUED

CHICK 2 (cont.)

AGE (DAYS)	WEIGHT (G)	FEED (G)	TIMES	DIET
38	164	50	3	30G SARDINES, 20G SMELT
39	199	50	3	30G SARDINES, 20G SMELT
40	214	50	3	30G SARDINES, 30G SMELT
41	247	50	3	30G SARDINES, 30G SMELT
42	259	50	3	30G SARDINES, 30G SMELT
43	270	50	3	30G SARDINES, 30G SMELT
44	284	50	3	30G SARDINES, 30G SMELT
45	291	50	3	30G SARDINES, 30G SMELT
46	299	50	2	30G SARDINES, 30G SMELT
47	305	50	2	30G SARDINES, 30G SMELT
48	308	50	2	30G SARDINES, 30G SMELT
49	319	50	2	30G SARDINES, 30G SMELT
50	325	50	2	30G SARDINES, 30G SMELT
51	329	60	2	30G SARDINES, 30G SMELT
52	334	60	2	30G SARDINES, 30G SMELT
53	343	60	2	30G SARDINES, 30G SMELT
54	352	60	2	30G SARDINES, 30G SMELT
55	353	60	2	30G SARDINES, 30G SMELT
56	358	60	2	30G SARDINES, 30G SMELT
57	368	60	2	30G SARDINES, 30G SMELT
58	370	60	2	30G SARDINES, 30G SMELT
59	373	60	2	30G SARDINES, 30G SMELT
60	374	60	2	30G SARDINES, 30G SMELT
61	373	60	2	30G SARDINES, 30G SMELT
62	375	60	2	30G SARDINES, 30G SMELT
63	374	60	2	30G SARDINES, 30G SMELT
64	376	60	2	30G SARDINES, 30G SMELT
65	376	60	2	30G SARDINES, 30G SMELT
66	380	60	2	30G SARDINES, 30G SMELT
67	383	60	2	30G SARDINES, 30G SMELT
68	385	60	2	30G SARDINES, 30G SMELT
69	385	60	2	30G SARDINES, 30G SMELT
70	385	60	2	30G SARDINES, 30G SMELT
71	388	60	2	30G SARDINES, 30G SMELT
72	388	60	2	30G SARDINES, 30G SMELT
73*	388	60	2	30G SARDINES, 30G SMELT
74	-	-	-	
75	340	70	1	SARDINES
76	344	90	1	SARDINES
77	-	-	-	
78	343	90	1	SARDINES
79	-	-	-	
80	330	80	1	SARDINES

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Case study: detection of two nonsteroidal anti-inflammatory drugs (NSAIDs) in the eggs of captive-reared bearded vultures at a breeding center in southern Spain

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Introduction

Large amounts of time and money are invested in the breeding, rearing, and rehabilitation of imperiled scavenging species in order to boost or stabilize local populations. Such is the case with Old World vulture species in Europe, particularly the bearded vulture (*Gypaetus barbatus*). In the wild, the main threats to the species have been identified as: unintentional poisoning via consumption of pesticide-laced baits placed in retaliation against predators,^{1,2} collisions with power lines, and human disturbances.¹ Currently broadly listed as “Near Threatened” by the IUCN Red List,³ the European bearded vulture population is categorized as “Endangered.”

Reintroduction efforts for the species were initiated during the late 1970s, with the opening of a captive breeding center in Harensee, Austria. Releases began in the mid-1990s, focusing initially on the Austrian Alps, but eventually extending over the entire mountain range into France, Italy, and Switzerland. Europe now boasts three captive breeding centers and 21 “supporting” facilities—mostly zoos—which host breeding pairs and their offspring. Collectively, these facilities are regarded as a mainstay of the future of this species, having yielded 59 eggs and 27 fledglings in 2016. In Spain, the first of the two captive breeding centers was built in Andalucía (Andalusia) (Cazorla; southern

ABSTRACT: The nonsteroidal anti-inflammatory drugs (NSAIDs) ketoprofen and/or meloxicam were qualitatively detected in two of three unfertilized eggs from the nests of two female bearded vultures (*Gypaetus barbatus*) at a breeding center in southern Spain in 2014. One egg contained trace levels of lead but not cadmium. No residues of organochlorine, carbamate, organophosphorus, or pyrethroid compounds were found in any of the eggs. Both birds were captive-born and reared, and never administered either drug. Investigation suggests provisioned food was the most likely source of exposure. In Europe, considerable investments are being made in efforts to conserve the bearded vulture. We recommend that funding be allocated to the comprehensive, harmonized safety testing of carcasses provisioned to breeding pairs, and encourage all facilities that source carcasses for scavengers and predators to enact similar verification to ensure these animals are receiving the highest possible standards of care.

KEY WORDS: NSAID, nonsteroidal anti-inflammatory drug, meloxicam, ketoprofen, diclofenac, egg, captive breeding, wildlife rehabilitation, scavenger, bearded vulture, *Gypaetus barbatus*, safety testing.

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Spain) in 1996, with releases starting in 2006. The other center, in Cataluña (Catalonia) (Vallcalent; northeastern Spain), opened in 1998. The sourcing and provision of high quality food items that closely mirror the species' natural dietary predilections represents a cornerstone in these efforts. In the wild, bearded vultures favor the bones of various wild ungulates and extensively graze-reared livestock, particularly sheep.⁴ While nestlings are being reared, about 25% and 5% of their diet consists of soft tissue and of skin, respectively,⁵ which parent birds both provision and consume themselves. Across their European range, including in Spain, wild avian scavengers may unwittingly feed on medicated animals,⁶ with subsequent nestling exposure (e.g., to anti-microbial residues).⁷ Still, the extent to which carcasses bearing veterinary agent residues are in fact available to avian scavengers in Spain and across Europe remains largely unquantified.^{1,8}

The role of the non-steroidal anti-inflammatory drug (NSAID) diclofenac in driving three species of Old World *Gyps* vultures—once considered the world's most populous birds of prey—to near extinction on the Indian subcontinent has now been thoroughly documented.^{9,10,11,12} Yet in recent years several European countries, including the Czech Republic, Italy, and Spain, approved the use of veterinary diclofenac (e.g., Diclovet).¹² In 2013, the Agencia Española de Medicamentos y Productos Sanitarios (Spain) approved a request from Italian drug manufacturer FATRO and its Spanish subsidiary to register diclofenac for use in pigs and cattle in Spain.¹³ Approval was granted on the grounds that the degree to which medicated livestock carcasses were available to Asian vultures on the Indian subcontinent was unparalleled.¹³ Nonetheless, Spain does manage numerous traditional carcass dumps and feeding stations, called *muladares*, for avian scavengers, particularly vultures, that are stocked with wild- and farm-sourced livestock carcasses. Spain is a major stronghold for Europe's vultures,¹ and the licensing was widely condemned.^{12,14,15} In parallel, our team also reported on the first known detection of the NSAID flunixin (with presence of ketoprofen) and fatal visceral gout in a wild Eurasian griffon vulture (*Gyps fulvus*) found within miles of a *muladar* in southern Spain.¹³ Although this bird was recovered for analysis in July, 2012, the detection of flunixin and ketoprofen was not published or divulged until after the registration of veterinary diclofenac.

In summary, four overarching factors have prompted personnel at the Center for Analysis and Diagnosis of Wildlife (CAD) to actively monitor a selection of veterinary drug residues in samples recovered during routine wildlife forensic investigations:

1. An existing dearth of information about the extent of medicated carcasses available to avian scavengers, and about the range (type and level) of residues therein;
2. Licensing of veterinary diclofenac;
3. The griffon vulture flunixin and ketoprofen detection case;
4. Emergence of other NSAIDs (e.g., carprofen, ibuprofen, ketoprofen, aceclofenac, nimesulide) potentially, or confirmed as being, harmful to (Old World) vultures.^{16,17,18}

These opportunistically analyzed samples are typically collected from suspected scenes of wildlife crime, under the Andalusian government's anti-poisoning/poaching strategy—the Estrategia Andaluza contra el Veneno (EAV)—with the aim of protecting vulnerable wildlife, especially endangered or imperiled species. However, there is now a dawning recognition that captive wildlife such as the bearded vulture, bred and rehabilitated at considerable cost to ensure the continuity of wild populations, represents a special subset that also merits a similar monitoring effort. Whether recovered from the wild or in captivity, eggs are a suitable sample with which to monitor contaminants, because residue distribution therein tends to be more homogenous than in other samples (e.g. feathers).¹⁹ Unfertilized eggs can also be collected noninvasively and the results of analyses can sometimes be directly related to reproductive effects and success.^{21–25}

Note that failed eggs and dead nestling bearded vultures have previously been collected from wild nests in the Spanish Pyrenees (2005–2008) for analysis of veterinary agents and investigation of exposure in nest failure. The detection of multiple NSAID and anti-microbial residues was reported; but this study²⁰ was retracted, so the veracity of these findings remains in question.

With this in mind, three unfertilized eggs were retrieved from the nests of two adult female bearded vultures at the captive breeding center in Cazorla, southern Spain, to determine whether any contaminants might be detectable therein. Following a series of analyses for heavy metals, pesticides, and veterinary agent residues, the NSAIDs meloxicam and ketoprofen were detected (both qualitatively, and rather unexpectedly) in two of the three eggs. Yet these two adult females have lived out their entire lives in captivity and have never been treated with either drug, raising questions as to the origin(s) of exposure. Here, we outline what a qualitative finding means, discuss the line of inquiry which led to our surmising that the birds' food was the source of exposure, and briefly consider the broader implications and applications to scavenger species in rehabilitation, zoos, and captive breeding settings.

Methods

In the winter of 2014, three unfertilized bearded vulture eggs were recovered from two separate nests (attended by Females BG 360 and BG 317) at the Centro de Cría Guadalentín (CCG) in Cazorla. This breeding center is operated by the Fundación Gypaetus (<http://www.gypaetus.org/>), with the aim of reintroducing the species within Andalucía and strengthening the stability of the local population. The eggs, which the females had incubated between 42 and 48 days, were collected 31 January and 6 February 2014, and refrigerated (4°C) until their transport. In March of that same year, the eggs were sent to the CAD for inspection and toxicological analysis, to investigate possible reasons for the birds' breeding failure. The CAD is Spain's only fully dedicated wildlife forensics facility, and all investigations are conducted in accordance with ISO–IEC guidelines (17025:2005: General requirements for the competence of testing and calibration laboratories; <https://www.iso.org/standard/39883.html>).

Immediately prior to transport, each egg was placed in an unused latex glove and housed within a new polystyrene container. Sheep's wool was placed between the eggs to protect them from breakage; however, there was no direct contact between the wool and the eggs. This is noteworthy because residues of the NSAID flunixin have been detected both on the surface of, and in, sheep's wool.²⁶

Examination and measurements

The three eggs were refrigerated at the CAD (4°C) until they were processed for analyses. Each egg was first externally examined for signs of structural imperfection, weakness, and breakage. During this initial examination, a fecal stain was observed on the egg labeled "BG 317; 31/01/2014." All eggs were weighed to the nearest 0.1 mg using a balance (Model Q-Weight Adam Equipment, England) and their length and breadth measured to the nearest 0.1 mm using calipers (Fisher Scientific, Thermo Fisher Scientific Inc.). The eggs were then cut open at the distal (pointed) end and their contents were removed and placed into chemically clean jars (250 ml; 250 Schott Duran, DURAN Group, Germany). The empty eggshells were cleaned with 70% ethanol and air-dried. Their thicknesses were measured using the same calipers. These measurements were used to calculate the Ratcliffe index²⁷ to estimate relative thickness, as follows: eggshell index = eggshell weight (mg) / length (mm) x breadth (mm).

Pesticides screening

A 5.0 g mixture of eggshell and egg contents was homogenized in a mixer (Agimatic-N, Selecta, Spain). Each aliquot was screened for 278 pesticides, including organophosphorus (OP), carbamate (CM), organochlorine (OC) pesticides, and pyrethroids (PYR).^{28,29} Briefly, the mixtures were ground in a mortar with anhydrous sodium sulphate (VWR VDH PROLABO, New Zealand) and the pesticides were extracted with dichloromethane (DCM; Fluka Analytical, Italy). Sample cleanup was performed using C18 solid phase extraction cartridges (Sharlau, Sharlab, Spain). The resultant extract aliquots were first assessed by thin layer chromatography (TLC) at the CAD, then screened once via gas chromatography–mass spectrometry (GC–MS/MS) by ion trap (IT) and triple quadrupole (QqQ) analyzers, and/or via ultra-performance liquid chromatography mass spectrometry (UHPLC–MS/MS) with triple quadrupole (QqQ) analyzer at the *Laboratorio Analítico Bioclínico* (LAB) in Almería, Spain. For GC-amenable pesticides, the limits of detection (LODs) ranged from 0.001 to 0.436 mg/L and the limits of quantification (LOQs) from 0.003 to 1.452 mg/L. For LC-amenable pesticides, the



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Captive bearded vulture chick in wool-lined nest

LODs ranged from 0.003 to 1.048 mg/L and the LOQs ranged from 0.011 to 3.494 mg/L.³⁰

Metals analyses

The eggshells were analyzed for lead and the egg contents were analyzed for cadmium, both at the CAD. Samples were dried at 105°C in an oven, then chopped with a grinder. An aliquot (0.5 g taken from approximately 10 g of total sample) was acid-digested with nitric acid (HNO₃ 65% Panreac 473255.1611) and hydrogen peroxide (H₂O₂ 30% p/v of 100 volume, Panreac 141076.1214), 7 mL HNO₃:H₂O (2:1) + 2 mL H₂O₂, topping the volume to 10 mL with distilled water, in a microwave. The extracts obtained were analyzed once, using Atomic Absorption Spectrometry (AAS) with graphite furnace using a wavelength of λ=283.3 nm for lead and λ=228.8 nm for cadmium, with LODs of <0.15 mg/kg and <0.04 mg/kg, respectively.

Antibiotic/Antimicrobial and NSAID screening

A 5.0 g mixture comprised of eggshell and egg content was homogenized in a mixer and extracted (at LAB) with acetonitrile (Merck, USA) and mixed for two minutes in an orbital mixer, after which

TABLE 1. Measurement parameters recorded for unfertilized bearded vulture eggs (n = 3) recovered from the Centro de Cría Guadalentín (Cazorla, Spain) in early 2014

SAMPLE NAME	WEIGHT ¹ OF ENTIRE EGG	LENGTH X WIDTH ²	WEIGHT ¹ OF SHELL ALONE	SHELL THICKNESS ²	RATCLIFFE INDEX ³
BG 360; 6–11/02/2014	149.83 ⁴ 193.5 ⁵	85 X 64 ⁴ 86.5 X 67 ⁵	32.72	0.8–1.0	6.01
BG 317; 6/02/2014	166.68 ⁴ 167.9 ⁵	79 X 63 ⁴ 80.5 X 64 ⁵	23.67	0.9–1.0	4.75
BG 317; 31/01/2014	137.91 ⁴ 140 ⁵	83 X 65 ⁴ 83 X 67 ⁵	29.52	0.9–1.1	5.47

¹Reported in grams (g)

²Reported in millimeters (mm)

³Eggshell index = eggshell weight (mg) / length (mm) x breadth (mm)

⁴Value as measured at CAD, pre-analysis

⁵Value as measured at CCG, post-hatching

TABLE 2. Reported bearded vulture egg (n=182) parameters from the Centro de Cría Guadalentín (Cazorla, Spain) (1996–2017)

ID	N	LENGTH ¹ (MIN)	LENGTH ¹ (MAX)	LENGTH ¹ (MEDIAN)	LENGTH ¹ (AVG)	LENGTH (STANDARD DEVIATIONS)	N	WIDTH ¹ (MIN)	WIDTH ¹ (MAX)	WIDTH ¹ (MEDIAN)	WIDTH ¹ (AVG)	WIDTH ¹ (STANDARD DEVIATIONS)
ALL CRG ³ EGGS	66	73	94	87	86	4.2	66	58	70	66	66	2.1
ALL BG 317	14	78	89	82	82	2.8	14	63	67	66	65	1.6
ALL BG 360	11	81	88	86	85	2.4	11	65	69	67	67	1.1
ALL FAILED EGGS	20	73	92	86	86	4.3	20	58	69	66	66	2.7
FAILED BG 317	2	80.5	83	NA	82	1.8	2	64	67	NA	66	2.1
FAILED BG 360	4	82	88	86	86	2.6	4	65	67	67	66	1.0
ALL HATCHED EGGS	46	78	94	87	86	4.2	46	63	70	66	66	1.8
HATCHED BG 317	12	78	89	82	82	3.0	12	63	67	66	65	1.6
HATCHED BG 360	7	81	87	86	85	2.4	7	66	69	67	67	1.0

¹Reported in millimeters (mm). ²Reported in grams (g). ³CCG = Centro de Cría Guadalentín.

TABLE 3. Lead and cadmium residue levels detected in unfertilized bearded vulture eggs (n = 3) recovered from the Centro de Cría Guadalentín (Cazorla, Spain) in early 2014

SAMPLE NAME	METAL ¹	WEIGHT ANALYZED ²	HUMIDITY ³	RESIDUES ⁴
BG 360; 6-11/02/2014	LEAD	10.33	12.99	0.15
BG 317; 6/02/2014		10.00	13.87	NOT DETECTED ⁵
BG 317; 31/01/2014		11.25	20.66	NOT DETECTED ⁵
BG 360; 6-11/02/2014	CADMIUM	10.38	81.45	NOT DETECTED ⁶
BG 317; 6/02/2014		10.13	82.20	NOT DETECTED ⁶
BG 317; 31/01/2014		11.61	76.63	NOT DETECTED ⁶

¹Lead analyzed in egg shell, cadmium analyzed in egg contents

²Reported in grams (g)

³Reported as % gravimetry

⁴Reported in mg/kg, dry weight

⁵< 0.15 mg/kg LOQ

⁶< 0.04 mg/kg LOQ

magnesium sulphate, di-sodium citrate and tri-sodium citrate were added (using a modified Quechers method for pesticide analysis). Fatty acids in the extracts were cleaned up with 400 mg primary secondary amine (PSA), and passed through a 400 mg C18 SPE cartridge (Sharlau, Sharlab, Spain) and through a Florisil® column. Antibiotics and antimicrobials (ABs and AMs; see Appendix A) were analyzed via two ultra-high pressure liquid chromatography (UHPLC) mass spectrometry (MS) systems (Waters Acquity UHPLC with tandem quadrupole MS; Transcend 600 UHPLC with single stage orbitrap MS), with LOQ of 0.01 mg/kg. NSAID residues (see Appendix B) were analyzed

qualitatively, i.e., as presence–absence, via UHPLC–Orbitrap.³¹ Control (i.e., “spike”) solutions of 250 µg/L concentration were used. All samples for both sets of analyses were analyzed twice.

Results

The observed egg parameters—thickness, length x width, and weight—are summarized in Table 1. An absence of historic shell thickness measurements for the bearded vulture precludes further comparison with the calculated Ratcliffe indexes. However, the measurement values are considered to fall within normal parameters when compared to eggs with similar incubation times (i.e., spanning 42 to 48 days; Table 2).

Only one of the three eggs contained trace levels of lead, at the limits of detection. Neither cadmium nor lead were detected in the remaining two eggs (Table 3). For comparison and perspective, Table 4 provides lead and cadmium levels observed in other bird species found in Spain.

No residues of OP, CM, OC, PYR or AB–AM agents were detected in any of the three eggs. However, the NSAID meloxicam was qualitatively detected (i.e., via UHPLC–Orbitrap) in the sole unfertilized egg recovered from breeding female BG 360 (Table 5, Figures 3 and 4). Meloxicam and the NSAID ketoprofen were qualitatively detected in one of the two eggs collected from female BG 317 (6/02/2014; Table 5, Figures 1 and 2). No residues of any kind were found in the fecal stain observed on BG 317; 31/01/2014, the only egg in which neither meloxicam nor ketoprofen was detected.

A qualitative finding means that the compound of interest is present within established detection limits, but not quantifiable.^{26,34} As with a quantitative detection, identification rests on the presence of characteristic (or diagnostic) ion fragments (m/z) seen at associated retention times. In this case, the residue levels

N	WEIGHT ² (MIN)	WEIGHT ² (MAX)	WEIGHT ² (MEDIAN)	WEIGHT ² (MEDIAN)	WEIGHT (STANDARD DEVIATIONS)
62	126	238	186	187	31.7
14	140	188	168	169	13.5
9	179	194	189	189	9.3
18	126	237	189	188	28.2
2	140	168	NA	154	19.7
2	179	194	NA	186	10.1
45	156	238	184	187	33.1
12	156	188	172	171	11.5
7	179	207	189	189	9.8

TABLE 4. Lead and cadmium residue levels reported in the eggs ($n = 110$) of wild birds of prey from two nature reserves in southern Spain

SPECIES	N	LEAD ¹		CADMIUM ¹	
		MEAN	RANGE	MEAN	RANGE
LESSER KESTREL ² (<i>Falco naumanni</i>)	41	0.89	0.10–2.8	0.71	0.38–1.1
COMMON BUZZARD ³ (<i>Buteo buteo</i>)	69	NOT RECORDED	0.32–3.5	NOT RECORDED	0.02–0.16
BOOTED EAGLE ³ (<i>Aquila pennata</i>)					
GOLDEN EAGLE ³ (<i>Aquila chrysaetos</i>)					
BLACK KITE ³ (<i>Milvus migrans</i>)					
PEREGRINE FALCON ³ (<i>Falco peregrinus</i>)					

¹Reported in mg/kg

²Ref. 32, from the province of Guadalquivir

³Ref. 33, from Doñana and Castilla La Mancha

lie below the established LOQ of 0.01 mg/kg. Ketoprofen was positively identified in sample 6955 (i.e., BG 317; 6/02/2014) based on the presence of m/z ion fragments [255] and [209] (Figures 1 and 2, respectively). Meloxicam was positively identified in samples 6955 and 6954 (i.e., BG 317; 6/02/2014 and BG 360; 6-11/02/2014, respectively) based the presence of m/z ion fragments [352] and [286] (Figures 3 and 4, respectively). Neither NSAID was observed in sample 6956 (i.e., BG 317; 31/01/2014).

Management implications of these findings

In early 2014, three unfertilized eggs were recovered from two adult bearded vultures housed at a captive breeding facility in southern Spain. Table 6 provides relevant additional information about the reproductive success of BG 317 and BG 360 for per-

TABLE 5. Antibiotics/antimicrobials and non-steroidal anti-inflammatory drugs detected in unfertilized bearded vulture eggs ($n = 3$) recovered from the Centro de Cría Guadalentín (Cazorla, Spain) in early 2014

SAMPLE NAME	WEIGHT OF SHELL EGG + CONTENTS ¹	ANTIBIOTICS / ANTIMICROBIALS ^{2,3}	NSAIDS ^{4,5}
BG 360; 6-11/02/2014 (6954 IN FIGS 1-4)	51.22	NOT DETECTED ⁶	MELOXICAM
BG 317; 6/02/2014 (6955 IN FIGS 1-4)	50.82	NOT DETECTED ⁶	MELOXICAM KETOPROFEN
BG 317; 31/01/2014 (6956 IN FIGS 1-4)	50.96	NOT DETECTED ⁶	NOT DETECTED ⁷

¹Reported in grams (g)

²Reported in mg/kg

³See Appendix I for a list of screened ABs/AMs

⁴Nonsteroidal anti-inflammatory drugs

⁵See Appendix II for list of screened NSAIDs

⁶< 0.01 mg/kg LOQ

⁷< 0.01 mg/kg LOQ

TABLE 6. Summary information about breeding success in bearded vulture pairings ($n = 9$) at the Centro de Cría Guadalentín (Cazorla, Spain) between 1996 and 2017

FEMALE ID	MALE ID	TOTAL NUMBER EGGS LAID	TOTAL NUMBER HATCHED	PERCENT HATCH RATE
153	286	12	7	58
290 ¹	172	4	0	0
	410	4	3	75
317	337	14	12	86
329 ¹	124	6	5	83
	223	1	1	100
330	313	11	8	73
360	391	11	7	64
389	362	8	3	38
TOTALS		71	46	

¹Paired with two males

TABLE 7. Annual bearded vulture pair ($n = 9$) productivity at the Centro de Cría Guadalentín (Cazorla, Spain) between 1996 and 2017

FEMALE x MALE ID	2009	2010	2011	2012	2013	2014	2015	2016	2017
153 x 286	1	2	2	1	2	2	2		
290 x 172		2	2						
290 x 410							1	2	1
317 x 337		2	2	2	2	3	2	1	2
329 x 124							4	2	
329 x 223				1					
330 x 313		1	2			2	4	1	1
360 x 391		2	2			1	3	1	2
389 x 362				2	1	1		4	

spective against the other breeding females at the center. Prior and subsequent to the recovery of these eggs, both birds successfully hatched young. Although in 2014 BG 360 only laid a single egg, one of the three eggs laid by BG 317 hatched. Furthermore, the number of eggs laid and percent hatch rates recorded for these two females are among the highest for the birds housed at the facility (Table 6). The distribution of annual pair productivity on a per year basis is shown in Table 7.

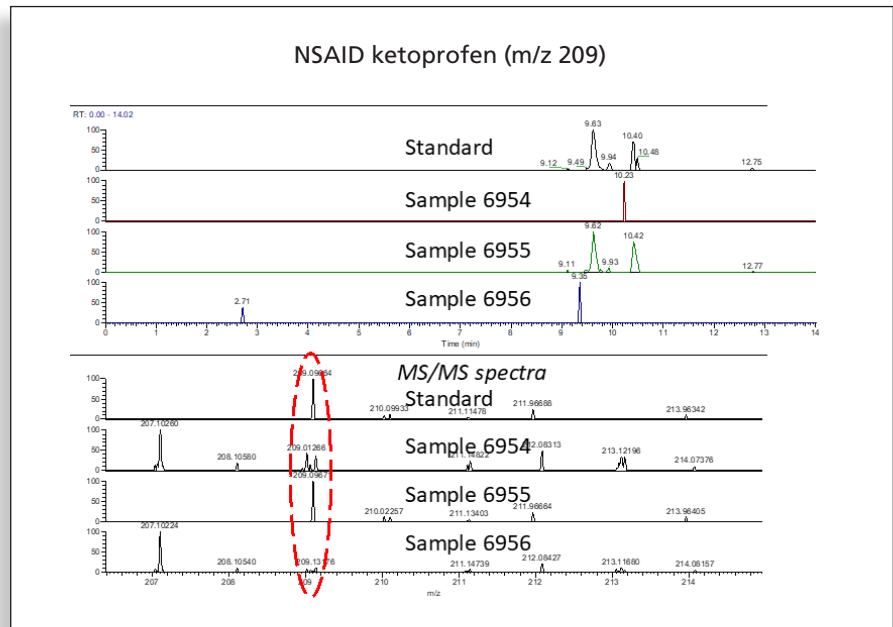
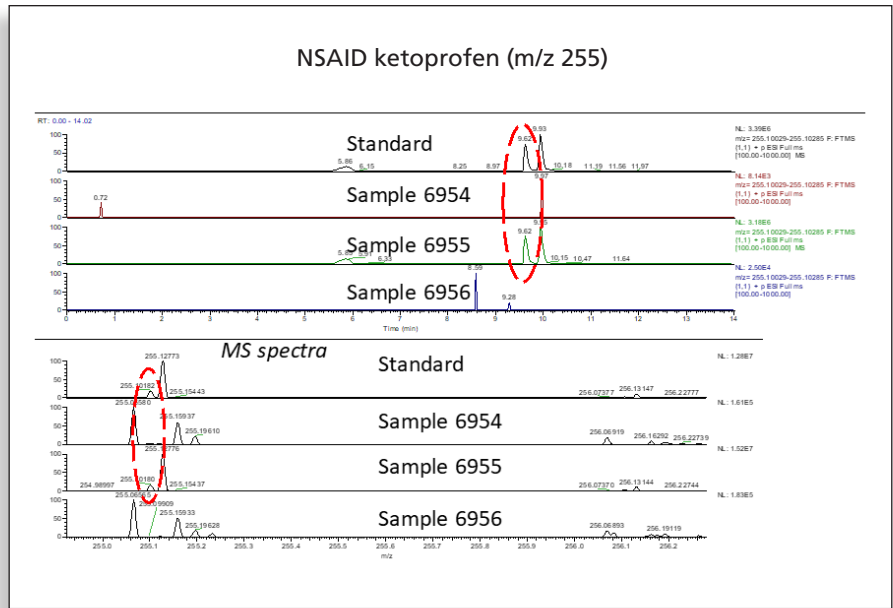
When eggs fail, eggshell thinning is invariably one of the first factors assessed. This phenomenon is commonly associated with OC exposure, particularly DDE, a highly persistent breakdown product of DDT.^{32,35} For example, two failed eggs were analyzed from wild Corsican bearded vultures (measuring 62 x 48 mm and 67 x 50 mm) that were abandoned after 69 days of incubation.³⁶ While trace levels of pesticides were found, residues of p,p'-DDE (0.42 and 0.61 ppm), hexachlorobenzene (HCB), and Clophen A60 (a PCB, or polychlorinated biphenyl) were detected. It has also been suggested that exposure to the OP diazinon, used as an anti-parasiticide (AP) in livestock, may impair thermoregulation in bearded vultures, reduce egg hatching success, and compromise nestling success.²

None of the 278 pesticides screened²⁹—including diazinon as well as DDT and its suite of environmentally relevant metabolites (DDE and DDD)—were detected in these eggs. Nor did the AB–AM analysis reveal the presence of any of these agents (see Appendix A). The measured eggshell lengths and widths reported for these three eggs (Table 1) lie within the commonly cited European measurements (75 x 56 mm,³⁷ and Table 2). Note that bearded vulture females BG 317 and BG 360 are of the (larger) Corsican subspecies (*G. barbatus barbatus*).

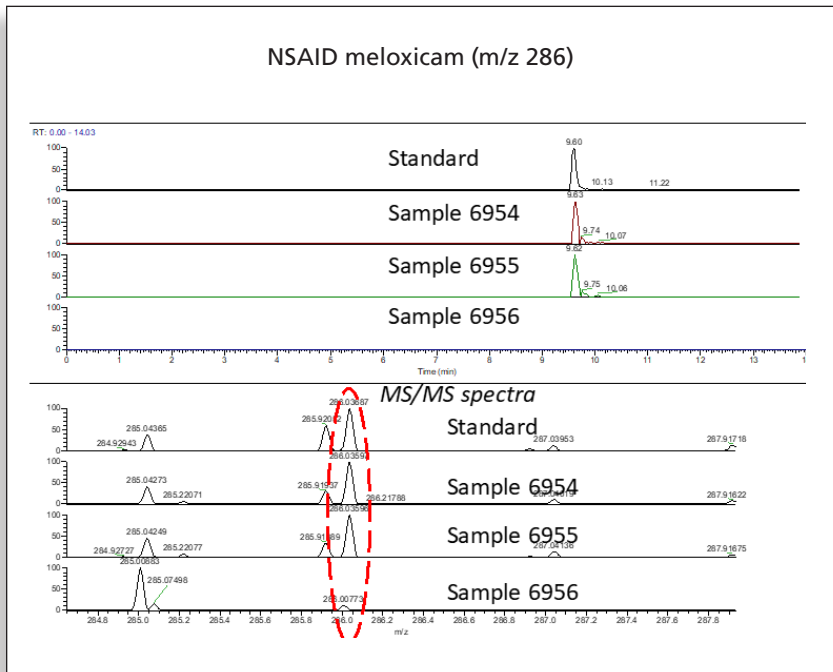
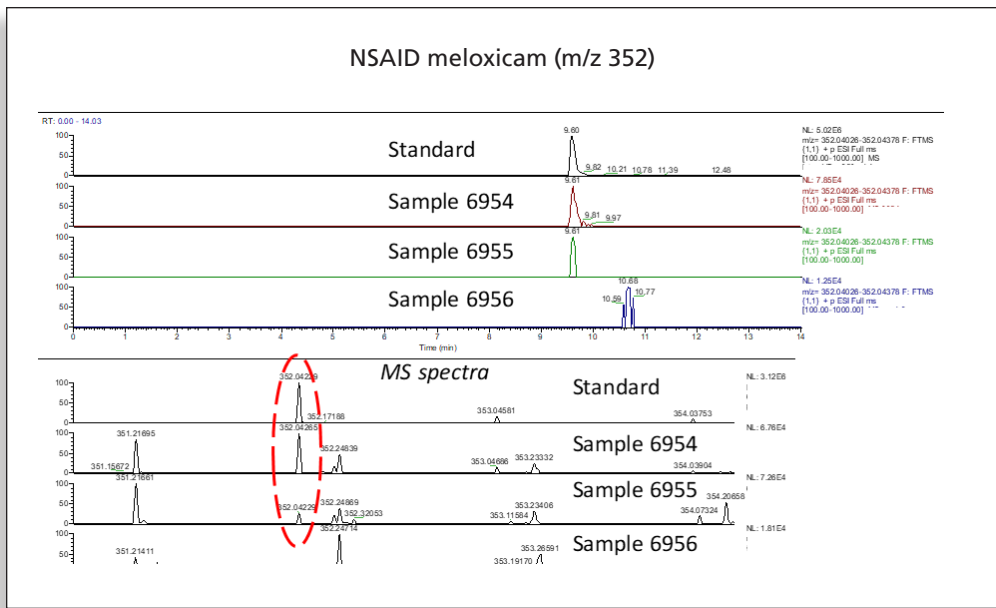
Exposure to heavy metals may also cause reproductive repercussions. However, no cadmium and only trace lead levels in a single egg were detected (Table 3, and Table 4 for comparison of levels reported locally in the eggs of other raptorial species). Nonetheless, given that metals and other persistent contaminants can occur in drinking water, and that such exposure has been shown to adversely affect reproduction of captive birds,³⁸ the breeding facility's water was verified as an additional precaution. The water tested negative for lead contamination, and all the pipes carrying

water provided to the birds are plastic. Additional concerns of captive vultures being exposed to lead in soil contaminated by flecks of lead-based paint (e.g., sanded from aviaries during enclosure upkeep)³⁹ are not applicable here.

In Spain, the availability of veterinary agent residues (particularly ABs–AMs) to adult vultures in the carcasses of medicated livestock^{6,7} and potential effects on embryo development and nestling exposure^{40,20} are increasingly being scrutinized. To our knowledge, this is the first time that the presence of NSAIDs has been reported in the eggs of an Old World vulture species *in captivity*. In the continued absence of concrete data linking NSAID exposure to egg failure, we are unable to determine whether or not the presence of these agents compromised the viability of these



FIGURES 1 AND 2. Chromatograms showing presence of the NSAID ketoprofen in one (6955) of the three bearded vulture eggs recovered from the Centro de Cria Guadalentin (Cazorla, Spain) in 2014 (ion fragments monitored: m/z 255 and m/z 209).



FIGURES 3 AND 4. Chromatograms showing presence of the NSAIDs meloxicam in two (6954 and 6955) of the three bearded vulture eggs recovered from the Centro de Cria Guadalentin (Cazorla, Spain) in 2014 (ion fragments monitored: m/z 352 and m/z 286).

particular eggs in any way. Nevertheless, the presence of residues in these eggs raises important questions as to its origin and, more broadly, to overall safety provisions. Residues were most likely transferred to the egg when the oviduct was in the egg formation stage. In terms of possible sources through which the adult females may have been exposed, neither bird was ever administered meloxicam or ketoprofen at any point during her life; nor are either of these agents known to be *in vivo* metabolic transformation products for other NSAIDs (as is the case with aceclofenac, recently discovered to be a prodrug of diclofenac¹⁸ and, similarly, suxibuzone metabolizing to phenylbutazone). This leaves ingestion

of residues, then, as the most likely route of exposure to these two NSAIDs.

Exposure via incidental ingestion

Ibuprofen and diclofenac—two of the most frequently detected NSAIDs in water samples across Europe—have been qualitatively detected both inside and on the surface of Eurasian otter (*Lutra lutra*) pelage, suggesting exposure via immersion in the waterways these animals spend so much time in.³⁴ Although meloxicam is described as being “practically insoluble in water,”³⁴ there is

evidence that the injectable formulation is stable in aqueous solution for 7 days.⁴² Ketoprofen does not readily dissolve in water either, though its solubility can apparently be increased with some manipulation.⁴³ Interestingly, residues of ketoprofen have also been detected in river sediments.⁴⁴ We were unable to find any reported instances of meloxicam detection in aquatic or associated samples, though this may be because it is not among the NSAIDs currently screened for in this medium.

Captive bearded vultures, including the two females whose eggs are reported on here, are typically provisioned with wool for lining their nests. Flunixin residues have been detected on the surface of sheep’s wool and flunixin readily dissolves in water.²⁶ Since neither the facility’s drinking water nor the wool used by these particular birds has been tested for NSAID or AB-AM residues, we are unable to assess whether either of these contributed to the residues observed in the eggs.

Exposure via primary ingestion pathways

In nature, bearded vultures feed preferentially upon the bones of various wild ungulates and extensively grazed or reared livestock, which comprises more than 60% of their diet.⁴ In Andalucía, for example, these are predominantly Iberian ibex (*Capra pyrenaica*) and sheep. Captive bearded vultures at breeding centers are fed a diet that closely mirrors that of their wild counterparts to the degree it is possible to procure such food items. The bearded vultures at the CCG, including females BG 317 and BG 360, are sometimes provisioned with ungulate bones, but most often receive one lamb leg six days out of every week, the equivalent

of roughly 300 grams of food each at a time. The birds typically consume the legs in their entirety, including the hoof.

In theory, captive rearing and breeding removes risks that bearded vultures would face in the wild, (e.g. exposure to pesticides used to deliberately poison predators). Throughout the region, specially appointed rangers at the adjacent and local natural parks (Sierras de Cazorla, Segura y Las Villas and Sierra de Castril) are tasked with ensuring that the bones sourced on behalf of the CCG are from wild ungulates hunted with copper rather than lead ammunition (lead intoxication being another known threat to this species).⁴⁵ And, in principle, livestock carcasses or carcass parts sourced from slaughterhouses (and therefore also intended for human consumption) only originate from farms where the use of potentially toxic or harmful veterinary agents like ABs-AMs and NSAIDs is tightly controlled. It is for this reason that the CCG always sources lamb legs from the same slaughterhouse, and, as per recommendations discussed below,² washes them with water and freezes them before they are fed to the birds at the facility. However, to the best of our knowledge, none of the carcass parts currently provided for bearded vulture consumption at any of the captive or supporting facilities are analytically residue tested, meaning their safety for consumption is not, in fact, guaranteed (for bearded vultures, but also potentially for people).

Mateo et al.² investigated the presence of topical APs in lamb legs and pig feet and assessed the potential associated exposure risk posed to bearded vultures in northeastern Spain. Residues were detected in 71% of the lamb legs analyzed ($n = 24$) but none in any of the pig feet ($n = 24$). Not factoring in dermal absorption, washing the lamb legs with water reduced but did not completely eliminate AP levels, leaving open the possibility of chronic exposure. It is noteworthy that these authors concluded wild bearded vultures may be more frequently affected by accidental poison from ingesting legally used OPs as APs in ovine livestock, than by consumption of illegally placed pesticide-poisoned baits. They also found significant differences between the occurrences of AP residues among slaughterhouses, with higher incidences in more remote, mountainous areas.

Important gaps remain in our knowledge of the extent to which residues are present in free-ranging and intensively reared livestock carcasses, and the range of compounds used for veterinary purposes available to avian scavengers therein in Spain.^{12,13} This is relevant to both wild and captive birds. The repercussions posed to *Gyps* vultures by the NSAID diclofenac and others (e.g., ketoprofen, ibuprofen, nimesulide, phenylbutazone, aceclofenac) have now been well documented,^{9,18,46,47} as has the relative vulture “safety” of meloxicam.^{8,48} There is also emerging concern about the vulnerability of other scavenging species such as steppes eagle (*Aquila nipalensis*), Egyptian vulture (*Neophron percnopterus*), red kite (*Milvus milvus*), and Spanish imperial eagle (*Aquila adalberti*), as well as the bearded vulture.^{e.g.,12,16,49} Ketoprofen has been banned for use in several parts of the Asian subcontinent (both in 2015 and in 2017),⁸ and conservationists are calling for a precautionary ban on veterinary diclofenac in Spain, promoting the use of

meloxicam instead.¹² In Spain and other European countries, ketoprofen is registered for veterinary use in cows, pigs, and horses, and meloxicam is registered for cows, pigs, sheep, goats, and horses,¹³ among others.

Within the European Union, a range of policies, legislative instruments and residue standards are being developed to address environmental contamination, protect human health, and safeguard wildlife species.^{50,51} In Andalucía alone, roughly 9.5 million euro—in excess of 10 million USD—has been spent on bearded vulture conservation efforts since 1996, primarily for research on reintroduction suitability and the captive breeding program itself. In the last five years, approximately 1 million euro has been invested in “anti-poison” efforts within the region, with 70% of this specifically tailored to protect the bearded vulture. And yet, the presence of both a potentially toxic and a relatively safe NSAID in the eggs of captive-bred birds indicates a necessary precaution that remains to be addressed in these extensive collaborative efforts: safety testing of provisioned food. Green et al.¹² recommended immediate development and implementation of programs to monitor and identify incidences of NSAID exposure and toxicity in vultures in Spain. Studies like the ones conducted by Casas-Díaz et al.⁶ and, more recently, Gómez-Ramírez et al.,⁵² are particularly valuable because they clearly distinguish between 1) the presence of residues in carcasses available to vultures, 2) evidence of the vultures’ exposure from consuming these carcasses, contrasted against 3) the actual implications (e.g., exposure at levels too low to likely be toxic in themselves, but sufficiently chronic to result in antibiotic resistance).

The cost of each of the analyses we have reported on is approximately as follows, for a total of up to 360 euro per sample:

- pesticides: 95 euro
- metals: 75 euro
- antibiotics and antimicrobials: 95 euro
- NSAIDs: 95 euro

Given these costs relative to the amount already expended and currently being expended to safeguard the future of the bearded vulture population, we recommend that a subset of available funding be allocated to the development and implementation of comprehensive, strategic and harmonized analytically-based safety testing of the carcasses and carcass parts provisioned to breeding pairs. We also believe that identifying all potential incidental sources of exposure at a given facility (e.g., wool, enrichment materials, and drinking water), and analytically ruling these out, represents a worthy investment and line of inquiry.

These results and their implications have been provided from a European perspective, yet the concerns we outline are just as applicable to North American practitioners seeking to ensure that the captive scavengers under their care are provided with carcasses and carcass parts that are safe for consumption. The recent accidental deaths of several captive animals fed the meat of a horse later determined to have been euthanized with sodium pentobarbital at a rehabilitation facility in Nevada,⁵³ and a follow-up review⁵⁴ which uncovered 126 cases of poisoning affecting a total of 433

animals (with 75% of the reported cases not in the published literature) lend weight to the need to implement safety testing and to increase the number of potentially harmful veterinary agents that are screened. That said, we are also aware that wildlife rehabilitation and captive facilities in North America typically operate under very tight budgets. Who will devise and implement such programs? And who will pay for them? These

are all important considerations which are outside the scope of this paper, but must nonetheless be addressed. Fundamentally, as Casas Díaz et al.⁶ stated: "...wildlife should not have access to food contaminated with pharmaceuticals." This is especially the case for captive animals, who have no choice in what they consume and whose safety and well-being therefore lies entirely in our hands.

Supplemental Material

APPENDIX A. List of antibiotic/antimicrobial agents screened in three unfertilized bearded vulture eggs recovered from the Centro de Cria Guadalentín (southern Spain) in 2014

CATEGORY OF ANTIBIOTIC/ANTIMICROBIAL	NAME
ANTHELMINTIC	ABAMECTIN ALBENDAZOLE MEBENDAZOLE OXFENDAZOLE
B-LACTAM	AMOXICILLIN AMPICILLIN
MACROLIDE	EMAMECTIN (BENZOATE) ERYTHROMYCIN FENBENDAZOLE IVERMECTIN JOSAMICYN LEVAMISOLE TILIMICOSIN
FUNGICIDE, PARASITICIDE	TIABENDOZOLE
BROAD SPECTRUM	TYLOSIN
FLUOROQUINOLONE	CYPROFLOXACIN DANOFLOXACIN DIFLOXACIN ENROFLOXACIN FLUMEQUINE MARBOFLOXACIN
QUINOLONE	OXOLINIC ACID SARAFLOXACIN
SULFONAMIDE	SULFACHLOROPYRIDIZNE SULFADIAZINE SULFADIMETHOXINE SULFADIMIDINE (OR SULFAMETHAZINE) SULFAQUINOXALINE TRIMETHOPRIM SULFAMETHOXAZOLE
TETRACYCLINE	CHLORTETRACYCLINE DOXYCYCLINE OXYTETRACLINE TETRACYCLINE

APPENDIX B. List of nonsteroidal anti inflammatory drugs (NSAIDs) screened in three bearded vulture eggs recovered from the Centro de Cria Guadalentín (southern Spain) in 2014

DICLOFENAC AND METABOLITES / BREAKDOWN PRODUCTS	
3'-HYDROXY-4'-METHOXYDICLOFENAC	
3'-HYDROXY DICLOFENAC	
4',5-HYDROXY DICLOFENAC	
4'-HYDROXY DICLOFENAC	
5-HYDROXY DICLOFENAC	
CARPROFEN	MELOXICAM
FLUNIXIN MEGLUMINE	NAPROXEN
INDOMETHACIN	NIMESULIDE
KETOPROFEN	

Addendum from the authors:

While our paper was in press, a study by Hernández et al. (2018) emerged, titled “Changes in eggshell thickness and ultrastructure in the bearded vulture (*Gypaetus barbatus*) Pyrenean population: A long-term analysis.” Here, the authors examined selected eggshell parameters—including thickness—and their trends in samples collected from 1989 to 2012 from wild bearded vultures in Spain and France, compared with museum specimens submitted from 1858 to 1911. Levels of organochlorines were also measured. Hernández et al. (2018) observed a decrease in the Ratcliffe Index and eggshell thickness in the eggs of wild BVs collected since 2001 and suggested that a corresponding decrease in the availability and quality of food resources in the wild—which may arise following a tightening in phytosanitary regulations—could be responsible. They also observed significant variability in both wild- and museum-sourced eggs to a degree which they believe is unique to the species and reflects its capacity to adapt to varying environmental circumstances. DDE levels appeared to increase but remained below levels associated with reduced breeding success. While in theory the concerns outlined about nutritional stress and reduced food availability should affect captive birds to a much lesser extent, such factors nonetheless are part of the larger conversation about ensuring the availability and prevalence of safe, nutritious food for this and other scavenging species. The observed inherent variation between the eggshell parameters of individual BVs reported by Hernández et al. (2018) must also be factored into future efforts focused on monitoring the eggs of this species.

Citation:

Hernández M, Colomer À, Pizarro M, Margalida A. Changes in eggshell thickness and ultrastructure in the bearded vulture (*Gypaetus barbatus*) Pyrenean population: A long-term analysis. *Science of the Total Environment*. 2018;624:713-721. <https://doi.org/10.1016/j.scitotenv.2017.12.150>.

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Ngaiio Richards has consulted on wildlife mortality and contaminants monitoring cases with CAD since 2012. She has a PhD in Forensic Science (Anglia Ruskin University, UK), an MSc in Natural Resource Sciences (applied wildlife biology and ecotoxicology; McGill University, Québec) and a BSc in Environmental Science (Acadia University, Nova Scotia). Currently an Assistant Adjunct Professor at the University of Florida (Gainesville) in the Veterinary Forensic Sciences program, she has authored numerous papers on repercussions of veterinary agents to wildlife and the environment, wildlife pesticide poisoning and noninvasive monitoring methods. She also serves as the Forensics and Field Specialist for Working Dogs for Conservation, a Montana-based nonprofit.

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Mónica Calvino is a veterinarian (Autonoma University of Barcelona) for Pechu Venus, a wildlife rehabilitation center run by the Ministry of Environment of Andalucía. She also works for CAD where she provides veterinary forensic capacity and expertise.

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News

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well-meaning citizens who unwittingly separated the animals from their mothers.

In a February 6th update, Eric Gardner, WDFW wildlife program director, said the department initiated a periodic review of the rules governing those facilities last October and now wants to include other perspectives.

“Since then, there have been times when the department, wildlife rehabilitators, and some members of the public have disagreed about the treatment of wild animals at certain facilities,” Gardner said. “We will look to this advisory group to help us clarify the rules to make sure we are all working toward a common goal of preparing these animals for release back into the wild.”

To represent the public interest, Gardner said the department encourages applicants with an interest in wildlife rehabilitation and the ability to communicate their perspectives in a productive manner. Applicants do not have to be affiliated with an organized group, and WDFW hopes to attract applications from both eastern and western Washington.

Members of the advisory group will serve until the state’s wildlife rehabilitation rules are adopted, unless both the department and the group’s members agree to extend the period of service.

Record Number of Sea Turtles Cared For

CORPUS CHRISTI, Texas (January 11, 2018)—From January 1 through January 10, the Texas State Aquarium has admitted and cared for nearly 800 cold-stunned sea turtles, releasing more than 700 sea turtles back into their natural habitat. This monumental undertaking—which took in more than a third of sea turtles recovered in Texas’ recent cold snap—marks an all-time record for the Aquarium’s wildlife rehabilitation team.

As cold weather swept through Texas in early January, over 2000 green sea turtles, afflicted by a hypothermic reaction called cold-stunning, were found by rescuers throughout the Lone Star State.

Dr. Donna Shaver, Chief of the Padre Island National Seashore’s Division of Sea Turtle Science and Recovery, collaborated with the Aquarium and other conservation partners to receive, maintain, and analyze records of cold-stunned turtles throughout the state. Between January 1 and January 6, 755 of these sea turtles—more than a third of those found in Texas—were taken in by the Texas State Aquarium’s Wildlife Rescue and Recovery Center. Staff and volunteers at the Aquarium worked around the clock to transport sea turtles from the Padre Island National Seashore and other facilities, document their health conditions, and treat them for cold-stunning. The sea turtles were housed in the Aquarium’s specialized rehabilitation center where their condition could be closely monitored and they could gradually recover to a healthy body temperature.

Thanks to the skill and dedication of the Aquarium’s wildlife rehabilitation team, the majority of the cold-stunned sea turtles made a full recovery and were soon ready to be released. On Friday, January 5, nearly 200 sea turtles were released offshore with the help of the U.S. Coast Guard. On January 6, 343 sea turtles were released at Padre Island National Seashore. Another 130 green sea turtles were released at Padre Island National Seashore on January 7 at an event that drew more than a thousand members of the public. Several smaller-scale releases also took place on January 8 and 9. Between January 5 and January 9, a total of 716 sea turtles taken in and treated at the Aquarium were returned to their natural habitat.

As of January 10, 21 sea turtles in critical condition remain at the Wildlife Rescue and Recovery Center. These sea turtles have various ailments ranging from wounds to missing flippers which require additional supportive care. Many sea turtles also show symptoms of fibropapillomatosis (FP), a disease that afflicts sea turtles with benign but debilitating tumors caused by the herpesvirus. Some of these sea turtles will undergo laser surgery to remove the most harmful tumorous growths before the sea turtles are released into their natural habitat.

Warm-blooded Animals More Likely than Cold-blooded to Adapt to Climate Changes

VANCOUVER, BC (January 29, 2018)—New research that analyzed more than 270 million years of data on animals shows that mammals and birds—both warm-blooded animals—may have a better chance of evolving and adapting to the Earth’s rapidly changing climate than their cold-blooded peers, reptiles and amphibians.

“We see that mammals and birds are better able to stretch out and extend their habitats, meaning they adapt and shift much easier,” said Jonathan Rolland, a Banting postdoctoral fellow at the biodiversity research centre at UBC and lead author of the study. “This could have a deep impact on extinction rates and what our world looks like in the future.”

By combining data from the current distribution of animals, fossil records, and phylogenetic information for 11,465 species, the researchers were able to reconstruct where animals have lived over the past 270 million years and what temperatures they needed to survive in these regions.

The planet’s climate has changed significantly throughout history, and the researchers found that these changes have shaped where animals live. For example, the planet was fairly warm and tropical until 40 million years ago, making it an ideal place for many species to live. As the planet cooled, birds and mammals were able to adapt to the colder temperatures so they were able to move into habitats in more northern and southern regions.

“It might explain why we see so few reptiles and amphibians in the Antarctic or even temperate habitats,” said Rolland. “It’s possible that they will eventually adapt and could move into these regions but it takes longer for them to change.”

Rolland explained that animals that can regulate their body temperatures, known as endotherms, might be better able to survive in these places because they can keep their embryos warm, take care of their offspring, and migrate or hibernate.

“These strategies help them adapt to cold weather but we rarely see them in

the ectotherms, or cold-blooded animals,” he said.

Rolland and colleagues argue that studying the past evolution and adaptations of species might provide important clues to understand how current, rapid changes in temperature impact biodiversity on the planet.

The study was a collaboration between scientists at UBC and in Switzerland and Sweden. It was published today in *Nature Ecology & Evolution*: <http://nature.com/articles/doi:10.1038/s41559-017-0451-9>.

White-Nose Syndrome sensitive to UV light

MADISON, Wisconsin, USA (January 2, 2018)—The fungus behind white-nose syndrome, a disease that has ravaged bat populations in North America, may have an Achilles’ heel: UV light. White-nose syndrome has spread steadily for the past decade and is caused by the fungus *Pseudogymnoascus destructans*, known as *P. destructans* or *Pd*.

In the course of genomic analyses of *P. destructans*, a team of scientists from the U.S. Forest Service, U.S. Department of Agriculture and the University of New Hampshire found that the fungus is highly sensitive to UV light. *P. destructans* can only infect bats during hibernation because it has a strict temperature growth range of about 39-68 degrees Fahrenheit. However, treating bats for the disease during hibernation is challenging, so any weakness of the fungus may be good news to managers trying to develop treatment strategies.

In a study published on Jan. 2 in the journal *Nature Communications* titled “Extreme sensitivity to ultra-violet light in the fungal pathogen causing white-nose syndrome of bats,” the research team suggests that *P. destructans* is likely a true fungal pathogen of bats that evolved alongside bat species in Europe and Asia for millions of years, allowing Eurasian bats to develop defenses against it. In the course of comparing *P. destructans* to six closely related non-pathogenic fungi, researchers discovered that *P. destructans* is unable to repair DNA damage caused by UV light, which could lead to novel treatments for the disease. The study, which was funded

by the U.S. Fish and Wildlife Service, is available at: <https://www.nrs.fs.fed.us/pubs/55557>.

“This research has tremendous implications for bats and people,” said Tony Ferguson, Director of the Forest Service’s Northern Research Station and the Forest Products Laboratory. “Bats play a key role in the health of forests as well as the production of food in the United States, and developing an array of tools with which we can treat bats for white-nose syndrome is important to preserving these very important species.”

The research team generated annotated genomes for *P. destructans* as well as six non-pathogenic *Pseudogymnoascus* species in an effort to gain insight into the origins and adaptations of the fungal pathogen of WNS. Using comparative genomics, the research team noticed that *P. destructans* lacked a key DNA repair enzyme, prompting them to expose the fungi to DNA damaging agents, including different wavelengths and intensities of UV light. They found that a low dose exposure of UV-C light resulted in about 15 percent survival of *P. destructans* while a moderate dose exposure resulted in less than 1 percent survival. These values translate to only a few seconds of exposure from a hand-held UV-C light source.

“It is unusual that *P. destructans* appears to be unable to repair damage caused by UV light,” said Jon Palmer, a research botanist in the Northern Research Station’s lab in Madison, Wis., and the lead author of the study. “Most organisms that have been found in the absence of light maintain the ability to repair DNA caused by UV light radiation. We are very hopeful that the fungus’ extreme vulnerability to

UV light can be exploited to manage the disease and save bats.”

“White-nose syndrome is the single biggest threat to many North American bat species and one of the most pressing conservation challenges facing America’s wildlife today,” said Jeremy Coleman, national white-nose syndrome coordinator, U.S. Fish and Wildlife Service. “Investing in defeating WNS must be a priority, and the results from this study and contributing research give us hope that we can develop the tools to more effectively manage the fungus that causes the disease.”

Research on potential treatment using UV light is under way. Daniel Lindner, a research plant pathologist with the Northern Research Station in Madison and the corresponding author on the study, is leading follow-up research to determine if UV light can be used as a treatment for bats suffering from white-nose syndrome.



Scanning electron micrograph of *P. destructans* in culture, isolated from a little brown bat (*Myotis lucifugus*). Characteristic curved conidia are visible in purple (false color).

The study will measure the survival of little brown bats during hibernation after being treated with UV light compared to control groups. The researchers are also exploring whether there are any non-target effects by measuring changes in bat skin microbiome (both fungal and bacterial communities). The study, funded by a grant from the National Fish and Wildlife Foundation Bats for the Future Fund began late last year. Co-authors on the study include Kevin P. Drees and Jeffery T. Foster of the University of New Hampshire. ■

PHOTO © DISPRING, CC BY-NC-ND 3.0 LICENSE.

Strategic acoustic control of a hummingbird courtship dive

CJ Clark and EA Mistick. *Current Biology*. 23 April 2018;28(8):1257–64. <https://doi.org/10.1016/j.cub.2018.03.021>

Male hummingbirds court females with a high-speed dive in which they “sing” with their tail feathers. The male’s choice of trajectory provides him strategic control over acoustic frequency and pressure levels heard by the female. Unlike related species, male Costa’s hummingbirds (*Calypte costae*) choose to place their dives to the side of females. Here we show that this minimizes an audible Doppler curve in their dive sound, thereby depriving females of an acoustic indicator that would otherwise reveal male dive speed. Wind-tunnel experiments indicate that the sounds produced by their feathers are directional; thus, males should aim their tail toward females. High-speed video of dives reveal that males twist half of their tail vertically during the dive, which acoustic-camera video shows effectively aims this sound sideways, toward the female. Our results demonstrate that male animals can strategically modulate female perception of dynamic aspects of athletic motor displays, such as their speed.

Fatal swine acute diarrhea syndrome caused by an HKU2-related coronavirus of bat origin

P Zhou, H Fan, T Lan, XL Yang, WF Shi, W Zhang, Y Zhu, YW Zhang, QM Xie, S Mani, et al. *Nature*. 2018;556:255–258. <https://doi.org/10.1038/s41586-018-0010-9>

Cross-species transmission of viruses from wildlife animal reservoirs poses a marked threat to human and animal health. Bats have been recognized as one of the most important reservoirs for emerging viruses and the transmission of a coronavirus that originated in bats to humans via intermediate hosts was responsible for the high-impact emerging zoonosis, severe acute respiratory syndrome (SARS). Here we provide virological, epidemiological, evolutionary and experimental evidence that

a novel HKU2-related bat coronavirus, swine acute diarrhea syndrome coronavirus (SADS-CoV), is the etiological agent that was responsible for a large-scale outbreak of fatal disease in pigs in China that caused the death of 24,693 piglets across four farms. Notably, the outbreak began in Guangdong province in the vicinity of the origin of the SARS pandemic. Furthermore, we identified SADS-related CoVs with 96–98% sequence identity in 9.8% (58 out of 591) of anal swabs collected from bats in Guangdong province during 2013–2016, predominantly in horseshoe bats (*Rhinolophus* spp.) that are known reservoirs of SARS-related CoVs. We found that there were striking similarities between the SADS and SARS outbreaks in geographical, temporal, ecological and aetiological settings. This study highlights the importance of identifying coronavirus diversity and distribution in bats to mitigate future outbreaks that could threaten livestock, public health and economic growth.

Limb-use by foraging marine turtles, an evolutionary perspective

JA Fujii, D McLeish, AJ Brooks, J Gaskell, and KS Van Houtan. *PeerJ*. 2018;6(e4565). <https://doi.org/10.7717/peerj.4565>

The use of limbs for foraging is documented in both marine and terrestrial tetrapods. These behaviors were once believed to be less likely in marine tetrapods due to the physical constraints of body plans adapted to locomotion in a fluid environment. Despite these obstacles, ten distinct types of limb-use while foraging have been previously reported in nine marine tetrapod families. Here, we expand the types of limb-use documented in marine turtles and put it in context with the diversity of marine tetrapods currently known to use limbs for foraging. Additionally, we suggest that such behaviors could have occurred in ancestral turtles, and thus, possibly extend the evolutionary timeline of limb-use behavior in marine tetrapods back approximately 70 million years. Through direct observation in situ and crowd-sourcing, we document the range of behaviors across habitats and prey

types, suggesting its widespread occurrence. We argue the presence of these behaviors among marine tetrapods may be limited by limb mobility and evolutionary history, rather than foraging ecology or social learning. These behaviors may also be remnant of ancestral forelimb-use that have been maintained due to a semi-aquatic life history.

Nanostructure, osteopontin, and mechanical properties of calcitic avian eggshell

D Athanasiadou, W Jiang, D Goldbaum, A Saleem, K Basu, MS Pacella, CF Böhm, RR Chromik, MT Hincke, AB Rodríguez-Navarro, et al. *Science Advances*. 2018;4(3):eaar3219. <https://doi.org/10.1126/sciadv.aar3219>

Avian (and formerly dinosaur) eggshells form a hard, protective biomineralized chamber for embryonic growth—an evolutionary strategy that has existed for hundreds of millions of years. We show in the calcitic chicken eggshell how the mineral and organic phases organize hierarchically across different length scales and how variation in nanostructure across the shell thickness modifies its hardness, elastic modulus, and dissolution properties. We also show that the nanostructure changes during egg incubation, weakening the shell for chick hatching. Nanostructure and increased hardness were reproduced in synthetic calcite crystals grown in the presence of the prominent eggshell protein osteopontin. These results demonstrate the contribution of nanostructure to avian eggshell formation, mechanical properties, and dissolution.

Flexibility of foraging strategies of the great skua *Stercorarius skua* breeding in the largest colony in the Barents Sea region

D Jakubas, LM Iliszko, H Strøm, HH Helgason, and L Stempniewicz. *Frontiers in Zoology*. 2018;15(9):1. <https://doi.org/10.1186/s12983-018-0257-x>

Background Foraging strategies of seabird species often vary considerably between and within individuals. This variability is influenced by a multitude of factors including age, sex, stage of annual

life cycle, reproductive status, individual specialization and environmental conditions.

Results Using GPS-loggers, we investigated factors affecting foraging flight characteristics (total duration, maximal range, total distance covered) of great skuas *Stercorarius skua* of known sex breeding on Bjørnøya, Svalbard, the largest colony in the Barents Sea region. We examined influence of sex (females are larger than males), phase of breeding (incubation, chick-rearing), reproductive status (breeders, failed breeders) and bird ID (they are known for individual foraging specialization). Our analyses revealed that only bird ID affected foraging flight characteristics significantly, indicating a high degree of plasticity regardless of sex, reproductive status or phase of breeding. We recognized three main groups of individuals: 1) those preying mainly on other seabirds in the breeding colonies (6%), 2) those foraging at sea (76%) and kleptoparasiting other seabirds and/or foraging on fish and/or offal discarded by fishing vessels, and 3) those alternating between preying on other seabirds in breeding colonies and foraging at sea (18%). Despite marked size sexual dimorphism, we found no apparent sex differences in flight characteristics. Birds after egg- or chick-loss and thus not constrained as central foragers did not modify their foraging flights.

Conclusions Great skuas breeding on Bjørnøya displayed a high degree of plasticity regardless of sex, reproductive status or phase of breeding. We recognized groups of individuals regularly preying in the seabird colonies, foraging at sea, and alternating between both strategies. This suggests foraging specialization of some individuals.

Recent extinctions disturb path to equilibrium diversity in Caribbean bats

L Valente, RS Etienne, and LM Dávalos. *Nature Ecology & Evolution*. 2017;1(0026). <https://doi.org/10.1038/s41559-016-0026>

Islands are ideal systems to model temporal changes in biodiversity and reveal the influence of humans on natural communi-

ties. Although theory predicts biodiversity on islands tends towards an equilibrium value, the recent extinction of large proportions of island biotas complicates testing this model. The well-preserved subfossil record of Caribbean bats—involving multiple insular radiations—provides a rare opportunity to model diversity dynamics in an insular community. Here, we reconstruct the diversity trajectory in noctilionoid bats of the Greater Antilles by applying a dynamic model of colonization, extinction and speciation to phylogenetic and palaeontological data including all known extinct and extant species. We show species richness in an asymptote to an equilibrium value, a demonstration of natural equilibrium dynamics across an entire community. However, recent extinctions—many caused by humans—have wiped out nearly a third of island lineages, dragging diversity away from equilibrium. Using a metric to measure island biodiversity loss, we estimate it will take at least eight million years to regain pre-human diversity levels. Our integrative approach reveals how anthropogenic extinctions can drastically alter the natural trajectory of biological communities, resulting in evolutionary disequilibrium.

Is captive breeding a priority for manatee conservation in Mexico?

A Ortega-Argueta and DN Castelblanco-Martínez. *Oryx*. 2018;1–8. <https://doi.org/10.1017/S0030605317001697>

The Endangered Antillean manatee *Trichechus manatus manatus* is one of the most threatened aquatic mammal species in Mexico and the wider Caribbean region. The decline of this subspecies is mainly a result of historical exploitation and the impact of current coastal development. The conservation strategies adopted for the Antillean manatee include habitat protection, reduction of the most severe threats, and the rescue of stranded, orphaned or injured individuals and their management in captivity. This latter strategy has produced positive outcomes in some countries but has been the subject of controversy in others, including Mexico. We analyse the benefits and challenges associated with

the management of captive manatees in Mexico, and the consequences of a lack of government policy and strategy for the post-rehabilitation release of individuals. We describe the evolution of this controversy from 1997–2017 in Mexico, analyze the consequences and implications for the conservation of the species, and propose an integrated management strategy that could address the issues raised. Although this strategy has been developed in the context of Mexico, it is applicable to management of this species across the Caribbean region.

Conservation and the 4 Rs, which are rescue, rehabilitation, release, and research

GH Pyke and JK Szabo. *Conservation Biology*. 2018;32(1):50–59. <https://doi.org/10.1111/cobi.12937>

Vertebrate animals can be injured or threatened with injury through human activities, thus warranting their “rescue.” Details of wildlife rescue, rehabilitation, release, and associated research (our 4 Rs) are often recorded in large databases, resulting in a wealth of available information. This information has huge research potential and can contribute to understanding of animal biology, anthropogenic impacts on wildlife, and species conservation. However, such databases have been little used, few studies have evaluated factors influencing success of rehabilitation and/or release, recommended actions to conserve threatened species have rarely arisen, and direct benefits for species conservation are yet to be demonstrated. We therefore recommend that additional research be based on data from rescue, rehabilitation, and release of animals that is broader in scope than previous research and would have community support.

Exploring wellness of wildlife carers in New Zealand: A descriptive study

P Yeung, B White, and BL Chilvers. *Anthrozoös*. 2017;30(4):549–63. <https://doi.org/10.1080/08927936.2017.1370213>

The rescue and care of vulnerable wildlife is rewarding. Most people involved in animal rescue have a strong commitment

to service but the work can be profoundly challenging. The ability to know and respond appropriately to an animal's needs depends on the professional skills and knowledge of wildlife carers. In the face of unrelenting suffering and countless numbers of animals in need, there are multiple stressors, vulnerabilities, and barriers that can undermine carers' well-being and put them at risk of compassion fatigue. The balance between compassion satisfaction and compassion fatigue is considered professional quality of life. The aim of this study was to provide a preliminary understanding of the potential for compassion satisfaction and the risk of compassion fatigue among wildlife carers within New Zealand. Thirty wildlife carers voluntarily completed a self-report survey, which included questions on socio-demographics, self-perceived quality of life (EUROHIS-QOL), professional quality of life (ProQOL), job satisfaction, motivation for ongoing work in wildlife rehabilitation, and coping mechanisms. We found that there were significant differences in compassion satisfaction and compassion fatigue on the basis of age, gender, financial capability, and years of experience. Overall, this sample showed high levels of compassion satisfaction and low levels of compassion fatigue. Understanding the elements of professional quality of life can have a positive effect on work environment. These results may provide clues to help identify wildlife carers' strengths for compassion satisfaction and vulnerabilities to compassion fatigue, and to help develop strategies to improve their professional quality of life.

Life and death: How should we respond to oiled wildlife?

LA Henkel and MH Ziccardi. *Journal of Fish and Wildlife Management*. In-Press. December 2017 (online). <https://doi.org/10.3996/062017-JFWM-054>

There is ongoing public debate about the best course of action to take when wildlife are affected by oil spills. Critics of wildlife rehabilitation suggest that the cleaning and release of oiled animals is a waste of resources focused on individual animals (not populations), thus the most

responsible course of action is to immediately euthanize affected animals. These critics claim that survival of rehabilitated animals is poor, and that the funds spent on rehabilitation would benefit wildlife more if spent on other conservation efforts. In this opinion piece, with a focus on birds, we review reasons for engaging in a coordinated response to oiled wildlife that includes cleaning and rehabilitation. The reasons for responding to oiled wildlife in any capacity include ethical, human safety, and legal aspects. Our rationale for proposing that responders attempt to rehabilitate wildlife, rather than planning on immediate euthanasia, includes clarification of financial, scientific, and additional ethical issues. Financially, costs for wildlife rehabilitation are typically a very small portion of overall oil spill response costs, and are typically independent of post-spill enforcement and funds used to restore damaged natural resources. Scientifically, we review recent studies that have shown that animals cleaned and rehabilitated after oil spills can often survive as well as non-oiled control animals. Ethically, some people would consider individual animals to have intrinsic value and that we, as consumers of petroleum products, have an obligation to reduce suffering and mitigate damage associated with such accidents. For these reasons, we suggest that, although humane euthanasia should always be considered as an option for animals unlikely to return to normal function after rehabilitation, response to oil spills should include a coordinated effort to attempt wildlife rehabilitation.

Inland oiled wildlife response: It's a different animal.

C Clumpner and M Ziccardi. *International Oil Spill Conference Proceedings: May 2017*. 2017;1:1795–1805. <https://doi.org/10.7901/2169-3358-2017.1.1795>

The Oiled Wildlife Care Network (OWCN) was established in 1994 to address the need for timely, consistent, and professional science-based preparedness and response for wildlife at risk from oil spills occurring in the marine waters of California. Its mission focused on

providing a high level of professional care based on the best available technology and science combined with the experience of many organizations that were pioneers in the field of wildlife rehabilitation. Since that time, the OWCN and its now more than 35 members have responded to over 100 spills while caring for more than 8,200 impacted animals. In 2014, in response to the increased risk due to changing sources and transport patterns in oil coming to refineries in California, the State legislature expanded the OWCN's responsibility to include responding to oiled wildlife impacted during oil spills in all surface waters of California. Since then, the OWCN has worked to expand its plan and resources to ensure readiness to provide best achievable capture and care to a host of new species in the myriad of habitats and locations found in a large and ecologically diverse state. The biggest challenges to this expansion are the increased diversity of species and their habitats (California has 233 species and subspecies of reptiles and amphibians), and the increase in geographical scope. Working with the California Department of Fish and Wildlife (specifically the Office of Spill Prevention and Response, or OSPR), the OWCN staff have identified species at risk, the challenges of response unique to an inland environment and terrestrial species, and the appropriate resources to meet those challenges and fill current gaps. We have incorporated lessons learned by colleagues during wildlife responses to inland spills including CNR Lake Wabamun (2005), Enbridge Kalamazoo River (2010), Silvertip Pipeline Yellowstone River (2011), and CNRL Cold Lake (2013). We have repurposed and redesigned existing equipment as well as acquiring additional mobile equipment to increase capacity and decrease response time. We have identified and trained first responders over a wide geographical area focusing on regions with increased risk of incident and impacts while leveraging our current primary care facilities with field stabilization and wildlife transportation plans to achieve maximum flexibility and cost effectiveness. We detail both the process that was used

to develop this expansion and the resulting additions to the wildlife plan aimed to provide best achievable care to all wildlife species impacted by an inland oil spill in California.

Movement patterns of California brown pelicans (*Pelecanus occidentalis californicus*) following oiling and rehabilitation

JS Lamba, CV Fiorello, YG Satgé, K Mills, M Ziccardi, and PGR Jodice. *Marine Pollution Bulletin*. June 2018;131(A):22–31. <https://doi.org/10.1016/j.marpolbul.2018.03.043>

Direct mortality of wildlife is generally used to quantify the damage caused by pollution events. However, free-ranging wildlife that survive initial exposure to pollutants may also experience long-term consequences. Individuals that are rehabilitated following oil exposure have a known history of oiling and provide a useful study population for understanding behavior following pollution events. We GPS-tracked 12 rehabilitated brown pelicans and compared their movements to those of eight non-oiled, non-rehabilitated controls over 87–707 (mean = 271) days. Rehabilitated pelicans traveled farther, spent more time in long-distance movements, and occupied more productive waters than controls. These differences were more apparent among females than males. Rehabilitated pelicans also visited breeding colonies and nest sites at lower rates than controls. Our results indicate that, although rehabilitated pelicans undertake long-distance movements, they may display increased dispersion and reduced breeding investment, particularly among females. Such behavioral changes could have long-term effects on populations.

First quantification of plastic ingestion by short-tailed albatross *Phoebastria albatrus*

E Donnelly-Greenan, D Hyrenbach, J Beck, S Fitzgerald, H Nevins, and M Hester. *Marine Ornithology*. 2018;46:79–84.

We investigated the sex, age, body condition, and ingested plastics in six short-tailed albatross *Phoebastria albatrus*, bycaught or opportunistically salvaged

in US North Pacific groundfish fisheries. Necropsies revealed a 1:1 sex ratio, and a 2:1 juvenile (≤ 4 years of age) to adult (≥ 5 years of age) ratio, with five birds in healthy body condition and four in active molt. Of the six birds examined, two females (one adult, one juvenile) and two males (both juvenile), contained ingested plastics. Of the four birds with plastic, the number and mass of total plastic per bird was variable (number: mean 4.75, SD 2.1; mass: mean 0.2921 g, SD 0.3250 g). Plastics were categorized as fragments ($n = 11$), sheets ($n = 4$), foam ($n = 2$), and rubber ($n = 2$). Fragments were the most numerous type, occurring in all four birds that had ingested plastic and accounting for 57.9% of the plastic items and 90.5% of the plastic mass (dry weight). We documented greater incidence of ingested plastic in the ventriculus (75.0%) than in the proventriculus (16.7%). The overall plastic incidence was 75.0% in juveniles and 50.0% in adults. While this research provides quantitative evidence that short-tailed albatross juveniles and adults ingest plastics, additional analyses are needed to fully quantify the prevalence of plastic ingestion and to investigate potential persistent organic pollutants and plasticizers in short-tailed albatross.

A review of roadkill rescue: who cares for the mental, physical and financial welfare of Australian wildlife carers?

B Englefield, M Starling, and P McGreevy. *Wildlife Research*. 2018;45(2):103–18. <https://doi.org/10.1071/WR17099>

The non-human animal deaths and injuries that result from collisions with motor vehicles are known colloquially as roadkill, and often lead to individuals from various taxa being orphaned. The complexities of multiple spatial and temporal variables in the available data on Australian roadkill and the scale of orphaning and injury make statistical analysis difficult. However, data that offer proxy measures of the roadkill problem suggest a conservative estimate of 4 million Australian mammalian roadkill per year. Also, Australian native mammals are mainly marsupial,

so female casualties can have surviving young in their pouches, producing an estimated 560 000 orphans per year. A conservative estimate is that up to 50 000 of these are rescued, rehabilitated and released by volunteer wildlife carers. These roadkill-associated orphans are in addition to those produced by other anthropogenic and natural events and the injured adult animals in the care of volunteers. In accepting total responsibility for rescued animals, wildlife carers face many demands. Their knowledge base can require days of initial instruction with the need for continual updates, and their physical abilities and personal health can be tested by sleepless nights, demanding manual tasks and zoonoses. This review article explores the impact of this commitment and conservatively estimates carers' financial input to raise one joey at approximately \$2000 a year, and their time input at 1000 h, equating to \$31 000 per year, applying a dollar value of \$31 per hour. It categorises relevant types of grief associated with hand-rearing orphans and rehabilitating injured animals, and suggests that wildlife carers most likely experience many types of grief but are also susceptible to burn-out through compassion fatigue. A perceived lack of understanding, empathy and appreciation for their work by government can add to the stressors they face. Volunteering is declining in Australia at 1% per year, social capital is eroding and the human population is aging, while the number of injured and orphaned animals is increasing. Wildlife carers are a strategic national asset, and they need to be acknowledged and supported if their health and the public service they provide is not to be compromised. ■

TAIL END



And all I wanted was a little soul patch.

**A Mary River turtle (*Elusor macrurus*),
underwater, with algal growth.**

Endemic to the Mary River in southeast Queensland, the species is one of Australia's largest turtles. It is sometimes called the green-haired turtle in honor of its tendency to harbor hair-like algal growth. It is ICUN red-listed as Endangered.

PHOTO © CHRIS VAN WYCK. USED WITH PERMISSION.

INSTRUCTIONS FOR AUTHORS

POLICY Original manuscripts on a variety of wildlife rehabilitation topics (e.g., husbandry and veterinary medicine) are welcomed. Manuscripts that address related topics such as facility administration, public relations, law, and education are invited as well.

Associate editors and anonymous reviewers, appropriate to the subject matter, evaluate each submitted manuscript. Concurrent submission to other peer-reviewed journals will preclude publication in the *Journal of Wildlife Rehabilitation* (JWR). The International Wildlife Rehabilitation Council (IWRC) retains copyright on all original articles published in the JWR but, upon request, will grant permission to reprint articles with credit given to the IWRC–JWR.

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Include an abstract that does not exceed 175 words and choose several (up to 14) key words.

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Endangered Antillean manatee (*Trichechus manatus manatus*), being examined by US Fish & Wildlife. See Abstracts, page 31.



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