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Environmental Toxicology

Do Pharmaceuticals in the Environment Pose a Risk to Wildlife?

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Abstract: The vast majority of knowledge related to the question "To what extent do pharmaceuticals in the environment pose a risk to wildlife?" stems from the Asian vulture crisis (>99% decline of some species of Old World vultures on the Indian subcontinent related to the veterinary use of the nonsteroidal anti-inflammatory drug [NSAID] diclofenac). The hazard of diclofenac and other NSAIDs (carprofen, flunixin, ketoprofen, nimesulide, phenylbutazone) to vultures and other avian species has since been demonstrated; indeed, only meloxicam and tolfenamic acid have been found to be vulture-safe. Since diclofenac was approved for veterinary use in Spain and Italy in 2013 (home to ~95% of vultures in Europe), the risk of NSAIDs to vultures in these countries has become one of the principal concerns related to pharmaceuticals and wildlife. Many of the other bodies of work on pharmaceutical exposure, hazard and risk to wildlife also relate to adverse effects in birds (e.g., poisoning of scavenging birds in North America and Europe from animal carcasses containing pentobarbital, secondary and even tertiary poisoning of birds exposed to pesticides used in veterinary medicine as cattle dips, migratory birds as a vector for the transfer of antimicrobial and antifungal resistance). Although there is some research related to endocrine disruption in reptiles and potential exposure of aerial insectivores, there remain numerous knowledge gaps for risk posed by pharmaceuticals to amphibians, reptiles, and mammals. Developing noninvasive sampling techniques and new approach methodologies (e.g., genomic, in vitro, in silico, in ovo) is important if we are to bridge the current knowledge gaps without extensive vertebrate testing. *Environ Toxicol Chem* 2023;00:1–16. © 2022 The Authors. *Environmental Toxicology and Chemistry* published by Wiley Periodicals LLC on behalf of SETAC. This article has been contributed to by U.S. Government employees and their work is in the public domain in the USA.

Keywords: Amphibians; birds; pharmaceuticals; reptiles; wildlife toxicology

INTRODUCTION

With rising living standards increasing the demand for livestock production and the growing and aging human population reliant on healthcare systems, pharmaceutical contamination of the environment is increasingly of concern (see Boxall et al., 2012; Wilkinson et al., 2022). Although environmental contaminants such as pharmaceuticals are just one of the many stressors faced by free-ranging species (e.g., along with habitat destruction, climate change, and disease), with 41% of amphibian species, 21% of reptiles, 13% of birds, and 27% of mammals listed as threatened with extinction by the International Union for Conservation of Nature (IUCN, 2022), it is important that we understand the potential of pharmaceuticals to affect populations.

Pharmaceuticals encompass a broad range of substances of synthetic or biological origin used to diagnose, treat, mitigate, or prevent disease or to promote well-being (also see Supporting Information, 1). They are characterized as substances that have the ability to stimulate, depress, or replace physiological functioning in a biological system. This implies that the underlying mechanism by which the drug functions needs to be present for a physiological effect to occur. Some compounds that are primarily thought of and used as pesticides (e.g., organophosphorus insecticides [OPs]) are also registered and used in veterinary medicine as livestock dips to parasites, thus expanding the definition of what is traditionally thought of as a pharmaceutical. This definition of "pharmaceuticals" generally implies that most drugs have similar effects

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in whatever biological system they have been placed, based on their mode of action. However, for some species the effects may be exaggerated or unexpected as a result of interspecific physiological differences, including differences in pharmacokinetics and/or pharmacodynamics (Toutain et al., 2010).

The principal source of pharmaceuticals in the environment is believed to be usage in human patients, livestock, and companion animals, which results in excretion of active pharmaceutical ingredients (APIs) and metabolites (Daughton & Ternes, 1999). Manufacturing and inappropriate disposal of pharmaceuticals and the presence of veterinary drug residues in carcasses of livestock represent additional exposure pathways that can lead to exposure of wildlife (see Supporting Information, 2, and Figure S2a–d). To date, research on pharmaceuticals and wildlife has largely focused on effects of nonsteroidal anti-inflammatory drugs (NSAIDs) on avian scavengers, with some work also on exposure, hazard, and risk of euthanasia drugs, antidepressants, and synthetic hormones (Table 1), and the role of wildlife in the transfer of antimicrobial resistance (AMR; Arnold et al., 2014; Bean & Rattner, 2018; Shore et al., 2014). Geographically, the focus has been on areas of Europe, North America, South Asia, and South Africa (Kookana et al., 2014; see Supporting Information, 3, for further discussion).

Depending on the environmental fate and mechanism of action of pharmaceuticals, wildlife can be used as sentinels for environmental contamination, to monitor bioaccumulation processes and to act as bioindicators of potential adverse effects. Wildlife are an integral part of complex food webs including humans and livestock, and there is growing recognition for the One Health perspective, indicating a need to give greater consideration to pharmaceutical impacts on wildlife. Examples of direct relevance to human health include the spread of AMR by migratory birds (Blanco et al., 2020; Loucif et al., 2022; Navedo et al., 2021) and the negative impact of some veterinary drugs on avian scavengers (Cuthbert et al., 2014; Plaza et al., 2022) and coprophagous insects (Tonelli et al., 2020), all of which can have consequences on the health of humans and domestic animals, for example, transfer of AMR organisms.

Given the potential for pharmaceuticals to have therapeutic effects, side effects, or unexpected toxicity in nontarget wildlife, it is noteworthy just how many drugs are licensed for use and how little we know about hazard to wildlife. In the United States alone, the US Food and Drug Administration (USFDA) has approved over 1600 animal drug products and 20,000 prescription drug products for human use, which include one or more of the approximately 4000 different APIs (USFDA, 2021). A little over half (51%) of the drugs approved for use in veterinary medicine by the USFDA are also approved for use in humans (Scott et al., 2020). Notably there are no routine regulatory requirements for industry to perform tests in wildlife species for human (European Medicines Agency [EMA], 2006, 2016; USFDA, 1998a, 1998b) or veterinary (Veterinary International Conference on Harmonization, 2000, 2006) medicines. Typically, a phased approach (see EMA, 2006; USFDA, 1998a, 1998b) is used in risk assessment for pharmaceuticals (see Figure 1 for a simplified schematic of regulations and Supporting Information, 4, for a more detailed discussion), with the aim of the initial screening phase to approve those drugs used in very low volumes, while later phases use data from acute and chronic tests in aquatic vertebrates, aquatic invertebrates, algae, terrestrial invertebrates,

TABLE 1: Known and potential pharmaceutical classes and individual drugs of concern for wildlife from the perspective of exposure (high volume of use, persistence/pseudopersistence), hazard (highly potent), and risk (known exposure pathway combined with exposure at a sufficient level to cause adverse effects)

General class/description	Example	Exposure	Hazard	Risk
NSAIDs	Diclofenac			
Oral contraceptive	17α-ethinylestradiol			
Euthanasia drugs	Pentobarbitol			
Analgesics/Pain killers	Acetaminphen			
Antihypertensive/Blood pressure control	Diltiazem			
Antimicrobials	Amoxicillin, cotrimoxazole and tetracycline			
Antifungals	Triazole pesticides			
Organophosphorus insecticides	Diazinon			
Pyrethroid insecticides	Permethrin			
Endectocide	Ivermectin			
Stimulants/Lifestyle drugs	Caffeine, Nicotine			
Antihistamine/Allergy relief	Loratadine, Cetirizine			
Anti-convulsant	Carbamazepine, Gabapentin			
Antidiabetic medicines	Metformin			
Anticoagulants	Warfarin			
Illicit drugs				
Antidepressants or antipsychotics	Fluoxetine			
Antineoplastics	Tamoxifen			
Wildlife immobilization	Etorphine			

Yellow cells are theoretical exposure, hazard, or risk; gray cells are known exposure, hazard, or risk.
NSAIDs = nonsteroidal anti-inflammatory drugs.

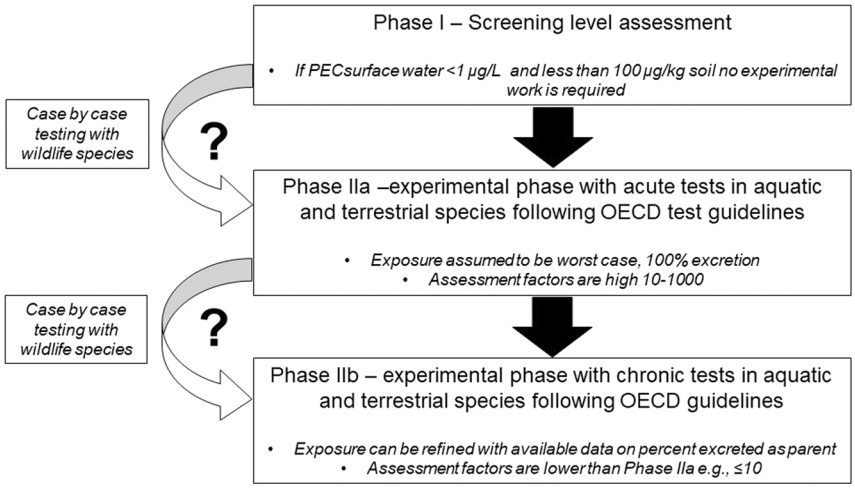

FIGURE 1: Simplified schematic of risk assessment for human and veterinary pharmaceuticals highlighting that specific testing with wildlife species is not routinely required but may be needed on a case-by-case basis. PEC = predicted environmental concentration; OECD = Organisation for Economic Co-operation and Development.

microbes, and plants. The need for tests in birds and mammals is considered on a case-by-case basis, based on considerations of potential exposure and acute toxicity. Registration of the NSAID flunixin by the USFDA as a transdermal formulation for cattle in the United States is one example of when testing in wildlife species was required because of concern around the toxicity of NSAIDs to avian scavengers (USFDA, 2017).

In the present review, we discuss current knowledge and research priorities involved with answering some of the key questions of the risk that pharmaceuticals in the environment pose to "wildlife." Although there are many varied definitions of "wildlife," we limited our scope to free-ranging amphibians, reptiles, birds, and mammals, as in Rattner (2009; although the definition varies among sources; see Supporting Information, 5). We focus on livestock dips, NSAIDs, euthanasia drugs, AMR, and antifungal resistance (AFR), drug effects on the microbiome of wildlife, the significance of exposure via wastewater, exposure hazard and risk for species other than birds, and 21st-century approaches for assessment of exposure and hazard.

PESTICIDES AND OTHER PHARMACEUTICALS USED AS DIPS FOR LIVESTOCK

What is our current understanding of the topic?

For over a century, a variety of chemicals applied in the form of a topical dip or pour-on have been used for control of parasites or disease treatment in livestock; in some instances, they have posed a significant secondary exposure hazard to wild birds. For example, in South Africa, arsenic-based compounds were introduced in the 1890s for livestock dipping, and frequent dipping of cattle often became compulsory in an effort to control the tick vector of the protozoan parasite that causes East Coast fever (Ramudzuli & Horn, 2014). South African ornithological records indicate that the yellow-billed oxpecker (*Buphagus africanus*), heavily dependent on tick prey, failed to breed in that country between 1907 and 1941 (Stutterheim & Brooke, 1981). This observation, in combination with lethality data from an experimental trial in which red-billed oxpeckers (*Buphagus erythrorhynchus*) were fed ticks dipped in arsenic trioxide (Bezuidenhout & Stutterheim, 1980), suggests that the collapse of the yellow-billed oxpecker population in this region was likely due to the reductions in tick prey coupled with consumption of arsenic-poisoned prey (Stutterheim & Brooke, 1981).

In the United States, avian poisoning events related to topically applied OPs have been documented and studied in far greater detail. A 1982 field study demonstrated that black-billed magpies (*Pica pica*) and red-tailed hawks (*Buteo jamaicensis*) were killed by famphur used as a pour-on to control warbles (*Hypoderm* sp.) on cattle in the states of Washington and Oregon (Henny et al., 1985). Gizzard contents of many dead magpies included cattle hair containing famphur or its activated metabolite, famphur oxon, and brain acetylcholinesterase (AChE) activity of the dead magpies was markedly depressed. During peak magpie mortality, some 10 days after cattle had been treated with famphur, a dead red-tailed hawk was found nearby with brain AChE activity depressed by 87% and magpie remains with 21 µg/g famphur detected in its crop. Subsequently, five cases involving bald eagles (*Haliaeetus leucocephalus*), red-tailed hawks, and a great horned owl (*Bubo virginianus*) collected in Oregon, California, Iowa, and Colorado were also linked to topically applied OPs, with remains of dead livestock found in proximity to dead birds (Henny et al., 1987). Stomach or crop contents of birds contained hair, flesh, and hide of livestock that tested positive for famphur and, in one case, fenthion; brain AChE activity was markedly depressed in the raptors. The exposure route was complex, including exposure via cow hair to magpies as an intermediary (sometimes cowbirds [*Molothrus ater*] or starlings [*Sturnus vulgaris*]) and finally to raptors.

In a large-scale study of avian scavenger poisoning between 2004 and 2013 in Spain, four cases involving bearded vultures (*Gypaetus barbatus*) tested positive for antiparasitics (three with diazinon, one with permethrin; Mateo et al., 2015), likely

representing legal use of these veterinary pharmaceuticals. A large number of lamb feet collected from abattoirs and supplemental vulture feeding stations had detectable quantities of OP or pyrethroid insecticides. A relatively simple washing procedure greatly reduced residues and was proposed as a risk-mitigation measure to protect birds. There are other groups of pharmaceuticals used as pour-on formulations including some NSAIDs, such as flunixin (e.g., Banamine® transdermal) used for control of pain and pyrexia associated with bovine respiratory disease (USFDA, 2017). Differential sensitivity among avian species is the hallmark of some NSAIDs (e.g., diclofenac [Rattner et al., 2008]), and flunixin may cause renal lesions in some avian species at a therapeutic dose extrapolated from that used in mammals (i.e., 1 mg/kg/day; Klein et al., 1994). However, a detailed environmental assessment of Banamine transdermal examining potential exposure pathways with calculation of risk quotients for nontarget wildlife (red-tailed hawk, coyote [*Canis latrans*]) concluded no significant impact from its proposed use (USFDA, 2017).

What are the future research priorities related to livestock dips and wildlife?

1. Evaluate the extent to which topical use of flunixin (and other NSAIDs if registered for that use) are hazardous to insectivorous birds.
2. Determine the extent to which active ingredients, in some cases thought of as plant protection products, that are also used as antiparasitics in veterinary medicine are a risk to wildlife.

NSAIDS AND SCAVENGERS

What is our current understanding of the topic?

One of the most recent instances of population-level effects in wildlife due to an environmental contaminant occurred in Old World vultures on the Asian subcontinent following their exposure to residues of diclofenac in the carcasses they fed on (see Figure 2). Oaks et al. (2004) was the first published study to make the link between diclofenac residues in liver and kidney of dead and dying vultures in Pakistan, with visceral gout, tubular necrosis, and renal failure which led to death and the observed population-level declines. Although diclofenac is now recognized as the cause, it took many years to reach this conclusion (Green et al., 2004; Oaks et al., 2004; Shultz et al., 2004; Swan, Cuthbert, et al., 2006). The exposure scenario was unpredictable and linked to the large number of cattle in the area due to the religious significance of cattle in Hinduism. Cattle are not slaughtered as in typical production systems and when ill were treated palliatively with NSAIDs, commonly diclofenac (a cyclooxygenase-2 and prostaglandin synthetase inhibitor). The toxicity of diclofenac to *Gyps* vultures was also unexpected, with it being extremely nephrotoxic (Meteyer et al., 2005). This resulted in estimated population declines for some species of 99% over a 15-year period from levels in the early to mid-1990s. In India, according to the IUCN (2022), white-rumped vulture (*Gyps bengalensis*), slender-billed vulture (*Gyps tenuirostris*), Indian vulture (*Gyps indicus*), and red-headed vulture (*Sarcogyps calvus*) are critically endangered, while Egyptian vulture (*Neophron percnopterus*) is listed as endangered, cinereous vulture (*Aegypius monachus*) is classed as near threatened, and bearded vulture (*Gypaetus barbatus*), griffon vulture (*Gyps fulvus*), and Himalayan vulture (*Gyps himalayensis*) are listed as of least concern.

Following the elucidation of the toxicity of diclofenac, meloxicam was introduced as a safe alternative, with countries on the subcontinent banning the manufacture and importation of diclofenac for use in veterinary medicine but not its use (Swan, Naidoo, et al., 2006). As a result, initial usage of stockpiles caused ongoing losses of birds (Taggart et al., 2009). As meloxicam use became more prominent and diclofenac was phased out (see Cuthbert et al., 2014), certain areas on the Asian subcontinent have seen a degree of vulture population recovery, albeit not a return to their previous numbers (see Supporting Information, 6.1, for more detail on vulture population status and the Indian government's response). In areas where signs of total recovery were evident, the change has been attributed to migration and not true recovery (Galligan et al., 2014, 2020; Paudel et al., 2016). Indeed, captive breeding programs successes have mainly been restricted to providing a sanctuary to prevent extinctions; only relatively small numbers of endangered vultures have been bred, and habituation of captive bred chicks limits success of releases (V. Naidoo, personal communication, June 21, 2021). Nonetheless, despite the positive benefits achieved with the removal of diclofenac, the safety of vultures was not fully protected; and other NSAIDs have since been identified as being toxic to birds, such as aceclofenac and nimesulide (Galligan et al., 2016, 2022). In areas where these drugs are used, toxicity is being reported with continued loss of birds. A summary of the toxicity of diclofenac and other NSAIDs to other avian species is provided in Table 2, with further discussion of differential metabolism in Supporting Information, 6.2. There are a couple of notable instances of insensitive species; for example, New World vultures are apparently tolerant of diclofenac (Rattner et al., 2008; Table 2). Perhaps the most significant development on this topic in the last decade came in 2013, when diclofenac was approved for veterinary use in Spain and Italy, countries holding approximately 95% of all European vultures. In this instance, the potential for exposure (Herrero-Villar et al., 2020) combined with the known hazard could translate into risk to individuals and even populations. In fact, the first case of diclofenac poisoning in a wild vulture in Europe was detected in 2020 in a cinereous vulture in Spain (Herrero-Villar, Delepoulle, et al., 2021).

What are the future research priorities related to NSAIDs and hazard to birds?

1. Continue to monitor the use of diclofenac and other NSAIDs for treatment of livestock in the Old World and monitor the status of critically endangered vultures.
2. Gain further understanding of exposure, hazard, and risk of NSAIDs for other avian species.
3. Determine exposure (e.g., carcass surveys) and risk posed to vultures in Europe and Africa as a result of NSAID use in veterinary medicine.

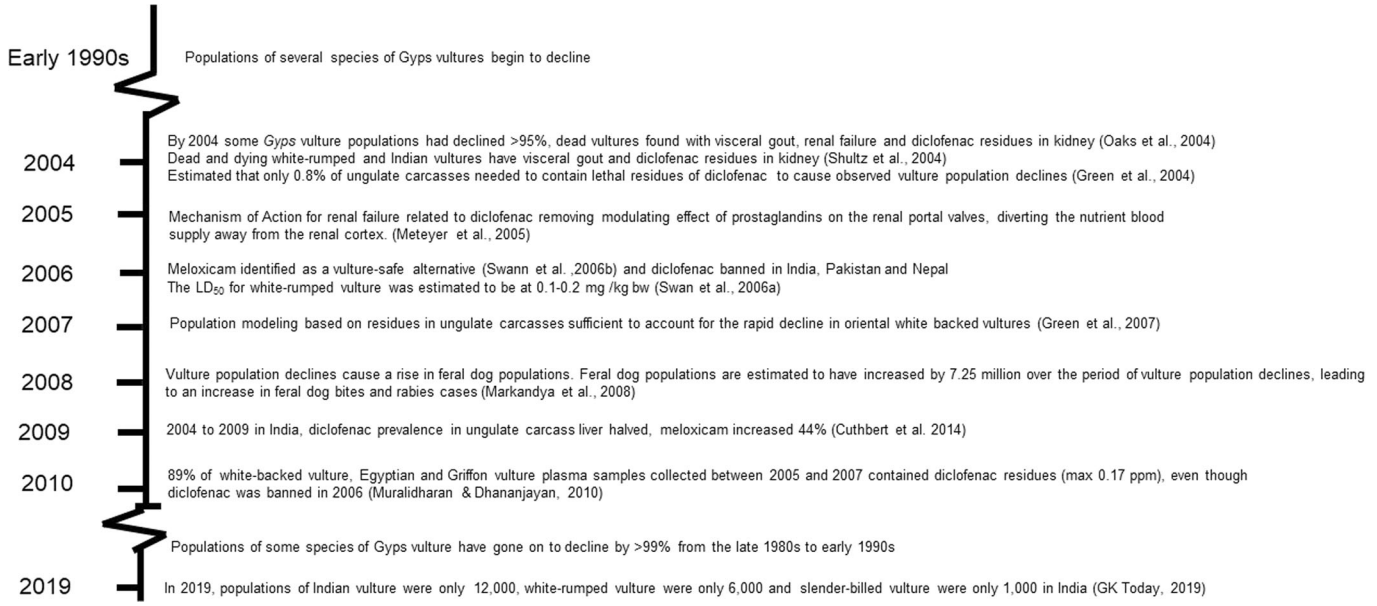

FIGURE 2: Timeline of Asian vulture crisis and research in the years that followed identification of diclofenac as the cause. LD50 = median lethal dose; bw = body weight.

EUTHANASIA DRUGS AND SCAVENGERS

What is our current understanding of the topic?

There are many instances of barbiturates, and specifically those used as euthanizing agents, causing intoxication and mortality in wildlife, mainly in North America and Europe in avian scavengers, although exposure of some mammalian scavengers has also been reported in Spain (Herrero-Villar, Sánchez-Barbudo, et al., 2021). Exposure to barbiturates occurred when scavengers ingested carcasses of domestic animals that had been euthanized with pentobarbital but had not been properly disposed of (i.e., appropriate disposal is via a means that prevents potential scavenger exposure: burial, incineration; Russell & Franson, 2014; Wells et al., 2020). Between 1975 and 2013 mortality due to pentobarbital intoxication in bald eagles submitted to the US Geological Survey, National Wildlife Health Center (Madison, WI), represented 1.1% (33/2980) of all bald eagle mortalities and 4.3% of poisonings (33/762). For golden eagles (*Aquila chrysaetos*) the equivalent mortality and poisoning numbers were 0.2% (3/1427) and 2.6% (3/117), respectively (Russell & Franson, 2014). More recently, a study in Spain found barbiturates in 5.9% (28 of 473) of intoxicated griffon vultures between 2004 and 2020 (Herrero-Villar, Sánchez-Barbudo, et al., 2021). Other raptors have also been affected by exposure to barbiturates throughout Europe, including cinereous vulture (*Aegypius monachus*), Egyptian vulture (*Neophron percnopterus*), Spanish imperial eagle (*Aquila adalberti*), red kite (*Milvus milvus*), and Eurasian buzzard (*Buteo buteo*; Herrero-Villar, Sánchez-Barbudo, et al., 2021; Moriceau et al., 2022; Wells et al., 2020). Pentobarbital concentrations in avian scavengers found dead ranged between 0.12 and 344 mg/kg in gastric contents and between 0.20 and 164 mg/kg in liver (Herrero-Villar, Sánchez-Barbudo, et al., 2021). Some of these mortality events have been clearly linked to the consumption of contaminated livestock carcasses (Herrero-Villar, Sánchez-Barbudo, et al., 2021).

What are the future research priorities related to the risk posed by euthanasia drugs in the environment to wildlife?

1. Understand the extent to which accidental and intentional barbiturate intoxication occurs in geographic regions beyond Spain and North America using existing incident reporting databases, and if such schemes are not currently monitoring for barbiturates, start including them in the list of analytes.

AMR, AFR, AND THE MICROBIOME

What is our current understanding of the topic?

AMR. The increased use of antibiotics and antifungals in human and veterinary medicine has led to frequent detection of antimicrobials (and, to a lesser extent, antifungals) in the environment (Wilkinson et al., 2022). One of the most disconcerting effects of their presence is the development of resistant (AMR and AFR) organisms. In this context, scavenging birds have been identified as potential dispersants of AMR because of their likely exposure via domestic animal carcasses potentially treated with veterinary drugs and their capacity to travel long distances (Blanco et al., 2020). Some of the AMR organisms are zoonotic pathogens (i.e., *Salmonella*) that are a particular concern from a human health and food safety standpoint, and some of these organisms have been reported in vultures and wild ungulates (Blanco, 2018; Marin et al., 2018; Ramos et al., 2022). Other avian species have been described as being potential dispersants and reservoirs of AMR, such as shorebirds, with compound exposure linked to use in aquaculture (Carroll et al., 2014; Navedo et al., 2021).

Antimicrobial resistance has also been reported in wild ungulates, where the pathway of transfer is via direct contact with domestic livestock as a consequence of human disturbance

TABLE 2: Collated evidence from the literature of exposure, hazard, and risk of nonsteroidal anti-inflammatory drugs to birds

Drug	Species	Geographic region or country	Evidence/effect	Citation
Aceclofenac	Potential exposure via cattle	Pharmacokinetic laboratory experiment	Rapidly (2 h postadministration) metabolized into diclofenac once it is administered to domestic animals (cattle) and therefore poses the same toxicity risk as diclofenac for *Gyps* vultures and other avian species.	Galligan et al. (2016)
Carprofen	Cape griffon vulture *Gyps coprotheres*	South Africa (laboratory experiments)	Acute toxicity test with two birds dosed at 11.5 mg/kg body weight, no mortality or lesions observed, lethargy and depression noted in one of the two birds. However, increases in alanine transferase activity and inhibition of uric acid excretion with subsequent increase in plasma uric acid concentration were observed, so further investigation of the safety of these NSAIDs for birds is warranted.	Fourie et al. (2015)
	African white-backed vulture *Gyps africanus*	South Africa (laboratory experiments)	Carprofen has been reported to cause mortality and visceral gout in white-backed vultures that consume concentrations equivalent to those at the injection site in muscle tissue of cattle (64 mg/kg body wt).	Naidoo et al. (2018)
Diclofenac	Steppe eagle *Aquila nipalensis*	India	Found dead at cattle carcass dumps with diclofenac residues and visceral gout.	Sharma et al. (2014)
	Turkey vulture *Cathartes aura*	United States (laboratory study)	LD50 estimated to be >25 mg/kg body weight, that is, >100 times less sensitive than *Gyps* vultures.	Rattner et al. (2008)
	Pied crow *Corvus albus*	South Africa (laboratory study)	No toxicity following a single oral dose ranging from 0.8 to 10 mg/kg body weight.	Naidoo et al. (2011)
	African white-backed vulture *Gyps africanus* Eurasian griffon vulture *Gyps fulvus*	South Africa (laboratory study)	LD50 found to be 0.8 mg/kg body weight, which is approximately 4 times that of the white-rumped vulture.	Swan, Cuthbert, et al. (2006)
	Cinereous vulture *Aegypius monachus*	Spain	First case of diclofenac poisoning in a wild vulture in Europe.	Herrero-Villar, Delepoulle, et al. (2021)
	Osprey *Pandion haliaetus*	United States	Plasma from 2/29 nestlings from Delaware Bay contained diclofenac residues of 2.33 and 3.73 ng/ml without evidence of adverse effects.	Bean et al, (2018)
Diclofenac, carprofen, flunixin, ibuprofen, and phenylbutazone	Owls, cranes, storks, raptors	NA	A survey of veterinary drug usage in zoos with the outcome from treatment of over 870 scavenging birds covering 79 species reported. Cuthbert et al. (2007) found that acute toxicity was associated with diclofenac, carprofen, flunixin, ibuprofen, and phenylbutazone, with mortalities reported for owls, cranes, storks, and raptors.	Cuthbert et al. (2007)
Flunixin	Eurasian griffon vulture *Gyps fulvus*	Spain	Necropsy of an individual found dead revealed flunixin residues of 2.7 and 6.5 mg/kg in liver and kidney, respectively; and the bird had severe visceral gout and renal failure.	Zorrilla et al. (2015)
	Eurasian griffon vulture *Gyps fulvus* Cape griffon vulture	Spain	Flunixin residues were associated with visceral gout in 3/306 Eurasian griffon vulture carcasses collected.	Herrero-Villar et al. (2020) Fourie et al (2015)

(*Continued*)

TABLE 2: (*Continued*)

Drug	Species	Geographic region or country	Evidence/effect	Citation
	Gyps coprotheres	South Africa (laboratory experiments)	Acute toxicity test with two birds dosed at 1.0 mg/kg body weight, no mortality or lesions observed, lethargy and depression noted in one of the two birds. However, increases in alanine transferase activity and inhibition of uric acid excretion with subsequent increase in plasma uric acid concentration were observed, so further investigation of the safety of these NSAID for birds is warranted.	
Ketoprofen	Cape griffon vulture *Gyps coprotheres* African white-backed vulture *Gyps africanus*	South Africa (laboratory experiments)	The hazard of ketoprofen to Cape griffon vultures and African white-backed vultures was evaluated at doses ranging 0.5–5 mg/kg body weight. Mortality occurred in the range of 1.5–5 mg/kg body weight with an environmentally relevant dose estimated to be 1.54 mg/kg body weight. At 5 mg/kg body weight, 7/11 birds died within 40 h, and it was concluded that it appeared that elimination of ketoprofen was via zero-order kinetics.	Naidoo, Wolter, et al. (2010); Naidoo, Venter, et al. (2010)
Meloxicam	Owls, cranes, storks, raptors	NA	A survey of veterinary drug usage in zoos with the outcome from treatment of over 870 scavenging birds covering 79 species reported. No mortalities were associated with meloxicam, providing further evidence of its safety to birds.	Cuthbert et al. (2007)
	White-rumped vulture *Gyps bengalensis* Indian vulture *Gyps indicus* African white-backed vulture *Gyps africanus*	Captive birds in India, Namibia, and South Africa	Experiments to evaluate toxicity of meloxicam up to and above the maximum likely environmental exposure; all vultures survived, with no toxicity observed.	Swan, Naidoo, et al. (2006)
Nimesulide	White-rumped vulture *Gyps bengalensis*	India	In 2019 tissues collected from four white-rumped vulture carcasses were screened for pesticides and 13 common NSAIDs, and only nimesulide was detected in all tissues, with concentrations ranging from 17 to 1400 ng/g; and the birds also had visceral gout.	Nambirajan et al. (2021)
	Cape griffon vulture *Gyps coprotheres*	South Africa (laboratory experiment)	Acute toxicity demonstrated for nimesulide to Cape griffon vultures at a predicted environmentally realistic dose of 17.6 mg/kg body weight. Both birds died within 30 h and displayed signs of visceral gout.	Galligan et al. (2020)
Phenylbutazone	Cape griffon vulture *Gyps coprotheres*	South Africa (laboratory experiments)	Acute toxicity test with two birds dosed at 1.7 mg/kg body weight, no mortality or lesions observed, lethargy and depression noted in one of the two birds. However, increases in alanine transferase activity and inhibition of uric acid excretion with subsequent increase in plasma uric acid concentration were observed, so further investigation of the safety of these NSAID for birds is warranted.	Fourie et al. (2015)

(*Continued*)

TABLE 2: (*Continued*)

Drug	Species	Geographic region or country	Evidence/effect	Citation
Tolfenamic acid	Himalayan griffon vultures *Gyps himalayensis*	India (laboratory experiment with captive birds)	Tolfenamic acid has been reported as a safe veterinary drug for vultures feeding on carcasses of treated livestock at the estimated maximum level of exposure, although the uric acid in blood plasma increased after exposure and 2/40 died with signs of visceral gout; the other 38 survived without any adverse clinical or biochemical signs; it was suggested, based on the absence of effects in critically endangered species, that the two deaths in Himalayan griffon vultures may have been anomalous because of the high doses used and the high environmental temperatures during testing.	Chandramohan et al. (2022)
	White-rumped vulture *Gyps bengalensis* India vulture *Gyps indicus*	India (laboratory experiment with captive birds)	No mortality or any clinical effects when dosed at the maximum likely environmental exposure.	Chandramohan et al. (2022)

NSAID = nonsteroidal anti-inflammatory drug; LD50 = median lethal dose.

and habitat conversion to agricultural systems (Espunyes et al., 2021; Ramos et al., 2022). Some of these wild ungulates play an important role in transmission between environmental compartments, such as wild boar, with interactions between urban, agricultural, and natural environments (Torres et al., 2020). This has also been reported in wild rodents with close contact with farms (Arnold et al., 2016). This may indicate a potential for the spread of AMR organisms globally, with subsequent implications from a "One Health" perspective.

What is our current understanding of the topic?

AFR. Antifungal resistance needs to be studied further before the role of wildlife in the dispersal of resistant organisms can be understood. There are only a few reports that describe resistant fungi isolated from European mammals such as wild boar and hedgehog that are closely linked to urbanized habitats (Gnat et al., 2021; Rhimi et al., 2022). Triazoles are also used as pesticides, and, as such, development of cross-resistance to triazoles used in the treatment of human fungal diseases (Bowyer & Denning, 2014; Snelders et al., 2012) should be considered.

What is our current understanding of the topic?

Microbiome. The role of the microbiome in human health is an emerging topic (Valles-Colomer et al., 2019; Zheng et al., 2020) that may also be an important aspect of wildlife ecotoxicology. Studies in laboratory animals and humans have shown changes in the enteric microbiome that are caused by antibiotics (Cho et al., 2012; Dethlefsen & Relman, 2011), and there is evidence that the enteric microbiome can also be affected by nonantibiotic pharmaceuticals (Maier et al., 2018). In captivity, the treatment of koalas for chlamydia with antibiotics caused changes in their intestinal microbiome which affected plant tannin degradation (Dahlhausen et al., 2018). In a hypothesis-driven research study, Thomason et al. (2017) demonstrated that ocular microbiomes of house finches were altered after antibiotic treatment; the subjects exhibited more severe Mycoplasma-induced conjunctival inflammation than untreated finches. To date, however, although the presence of AMR bacteria has been widely described across taxa throughout the world (see previous discussion), few studies have explored the effects of pharmaceuticals on the microbiome of wildlife. Exceptions include impacts of antimicrobials on resistant bacteria in the intestinal microbiota of mallard ducks (*Anas platyrhynchos*; Atterby et al., 2021), and Pitarch et al. (2017) observed oral mycoses in avian scavengers exposed to antibiotics that may alter the host's normal microbiota composition and that can facilitate opportunistic pathogenic yeast growth to cause disease.

What are the future research priorities related to AMR/AFR and the microbiome?

1. Gain a greater understanding of the role of wildlife in transferring AMR organisms.
2. Develop an understanding of the significance of wildlife in transferring AFR.

3. Conduct controlled experiments to understand the enteric microbiome in different wildlife species and their implications for immune function.

IMPORTANCE OF WASTEWATER AS A PHARMACEUTICAL EXPOSURE PATHWAY FOR WILDLIFE

What is our current understanding of the topic?

Foraging directly on wastewater-treatment plants. Controlled studies designed to simulate exposure of European starlings foraging on invertebrates at wastewater-treatment plant (WWTP) trickling filter beds to the antidepressant fluoxetine have been conducted in the United Kingdom (Bean et al., 2014, 2017; Whitlock et al., 2018, 2019). These studies found indications that environmentally realistic concentrations administered via invertebrates injected with fluoxetine for approximately 6 months may cause subtle effects on foraging (e.g., timing and frequency of foraging bouts [Bean et al., 2014]) and courtship behavior (Whitlock et al., 2019). At present, the importance of this exposure pathway (i.e., birds eating fluoxetine-contaminated invertebrates from WWTPs) and the biological significance of the effects of fluoxetine remains a knowledge gaps (i.e., do they translate from the laboratory to the field, behavior as a relevant apical endpoint; see Supporting Information, 7). Only the experiment of Whitlock et al. (2019) detected fluoxetine residues in free-ranging starlings. However, the residues were detected in feathers of starlings that had been grown in the wild (21 of 25 birds, up to 27 ng/g dry wt) but sampled after the birds had been brought into the laboratory and dosed with fluoxetine. As there was also fluoxetine contamination of feathers and the liver of one control bird, it was not clear whether the fluoxetine in these feathers was transferred during captivity (e.g., via contact with excreta or during preening) or was the result of exposure prior to capture. The importance of this exposure route and the risk posed remain to be determined.

Water–fish–osprey food webs. To date, two studies have examined exposure, potential effects, and trophic transfer of APIs and metabolites in the osprey food web. Between 2012 and 2015, water samples, blood samples from various species of fish commonly consumed by osprey, and blood samples from 40- to 45-day-old osprey nestlings were collected in Chesapeake and Delaware Bays and associated tributaries in the United States (Bean et al., 2018; Lazarus et al., 2015). Water and blood plasma samples were analyzed for more than 20 APIs or metabolites by liquid chromatography–tandem mass spectrometry. The antihypertensive diltiazem consistently exceeded detection limits in osprey nestling plasma samples from the Chesapeake region. In the Delaware region, the analgesic acetaminophen was detected in 75% of the osprey nestling plasma samples, and the NSAID diclofenac was detected in only 7% of the nestling plasma samples. Although the effect thresholds of these three APIs are unknown for ospreys, observed concentrations were well below the human plasma therapeutic concentration (HTC; 28% of the HTC for diltiazem and two to three orders of magnitude lower for acetaminophen and diclofenac). Overall, these data and predictions may indicate that the risk of therapeutic or toxicological effects associated with trophic transfer of APIs and metabolites to osprey nestlings in the Chesapeake and Delaware regions is low (see Supporting Information, 8, for further discussion of kinetics).

What are the future research priorities related to evaluating the significance of exposure to pharmaceuticals from wastewater?

1. Fill knowledge gaps around the significance of exposure to pharmaceuticals at or near WWTPs in regions other than Europe and North America.
2. Understand the exposure and hazard for aerial insectivores foraging on insects that emerge from WWTP filter beds.
3. Periodic monitoring/decadal reevaluation of pharmac eutical contamination of the environment using noninvasive methods in sentinel species.

EFFECTS ON WILDLIFE OTHER THAN BIRDS

Research related to pharmaceuticals in the environment has focused on exposure and effects in birds, leaving knowledge gaps on the risk of pharmaceuticals to amphibians, reptiles, and mammals.

What is our current understanding of the topic?

Amphibians and reptiles. The detection of many classes of pharmaceuticals in wastewater effluent, groundwater, untreated drinking water, and runoff from concentrated animal feed operations (see Barnes et al., 2008; Bartelt-Hunt et al., 2011; Focazio et al., 2008; Roberts & Thomas, 2006) has led to extensive laboratory exposure studies, field monitoring, and modeling efforts to assess the risk to aquatic species, including reptiles and amphibians. Notably, direct evidence of pharmaceutical exposure through detection of parent compound or metabolites is generally lacking for free-ranging amphibians and reptiles. However, in laboratory studies the pharmaceutical 17α-ethinylestradiol (EE2) and several progestogens have been shown to evoke reproductive toxicity in *Xenopus tropicalis* and *Xenopus laevis* at environmentally relevant concentrations (Orlando & Ellestad, 2014; Safholm et al., 2014). The highly publicized findings of feminization and endocrine disruption in American alligators (*Alligator mississippiensis*) at Lake Apopka, Florida, USA, were principally attributed to chlorinated hydrocarbon pesticides and not pharmaceuticals (see Guillette et al., 2000); however, this triggered many other such investigations in reptiles and other wildlife, with the potential for such effects from pharmaceuticals like EE2 and diethylstilbesterol often mentioned (see Guillette & Edwards, 2008). Nile crocodiles (*Crocodylus niloticus*) were studied at a commercial crocodile farm downstream from a sewage-treatment plant in Brits, South Africa (Arukwe et al., 2015, 2016). Carbamazepine, EE2, galaxolide, and tonalide were detected in water at various locations in proximity to the farm

using passive samplers, and more commonly studied contaminants (aliphatic hydrocarbons, aromatic hydrocarbons, metals, halogenated pesticides) were present in the liver of crocodiles, with correlative evidence potentially indicating effects on biotransformation and oxidative stress endpoints, and reproductive and endocrine pathways.

Aerial insectivores. Bats can also be exposed to pharmaceuticals when they forage on insects that emerge from sewage filter beds (Park & Cristinacce, 2006). Park et al. (2009) determined that concentrations of the synthetic estrogen EE2 were greater in insects collected around trickling filers than at sites over 2 km away but never actually demonstrated exposure of bats to pharmaceuticals. In high-income countries at least, activated sludge has been replacing trickling filter beds in recent years; and thus, the relevance and importance of this exposure route for aerial insectivores remains unknown.

Marine mammals. A recent review of pharmaceuticals and personal care products and their toxicity to aquatic organisms (Srain et al., 2021) describes effects on a range of aquatic invertebrates and fish but reports evidence from only one nonfish vertebrate, citing an in vitro study which suggests potential impacts on immune function in harbor seals (*Phoca vitulina*; Kleinert et al., 2018).

Otters feed almost exclusively in aquatic systems, which may be contaminated by both effluent discharge and runoff. Fish are the predominant prey of most of the 13 otter species worldwide, although for some species aquatic crustaceans are commonly consumed (Kruuk, 2006). Trophic transfer of pharmaceuticals to otters seems likely in view of evidence of contamination of freshwater invertebrates and fish (see Cerveny et al., 2021; Miller et al., 2019). As yet, there is no published evidence of internal pharmaceutical exposure of otters, but recent data from the LIFE Apex Project (Gkotsis et al., 2022) indicates that a wide range of antidepressants, drugs of abuse, and stimulants are present in apex predators, and their prey, including a metabolite of the analgesic metamizole in an otter from Germany, and screening of hair samples identifies NSAIDS (ibuprofen and diclofenac) in Eurasian otter (*Lutra lutra*) from the United Kingdom (Richards et al., 2011).

What are the future research priorities related to studying exposure and effects of pharmaceuticals in the environment to reptiles, amphibians, and mammals?

1. Despite a clear potential for exposure via trophic transfer from fish and invertebrates, research has yet to document the occurrence, accumulation, or risk from pharmaceuticals in aquatic and, particularly, marine mammals (Kleinert et al., 2018).
2. There are still many knowledge gaps around the exposure and hazard of pharmaceuticals to reptiles and amphibians. Evaluating the ability to use read-across data from fish and endocrine disruption screening studies (e.g., amphibian metamorphosis assay) would be helpful for risk assessment.

NONINVASIVE METHODS FOR EVALUATING PHARMACEUTICAL EXPOSURE OF WILDLIFE

What is our current understanding of the topic?

Opportunistic sampling of animals found dead or carcasses provided by hunters or animal control programs represents one method for evaluating exposure without invasive sampling. As previously documented, wildlife poisoning by pharmaceuticals has focused on avian scavengers exposed to highly toxic compounds such as NSAIDs and barbiturates (Pain et al., 2008; Russell & Franson, 2014; Wells et al., 2020). The catastrophic situation in Asia with diclofenac could potentially have been ameliorated sooner, preventing the vulture population crash. For example, the WILDCOMS network (www.wildcoms.org.uk) in the United Kingdom brings together a number of surveillance schemes which monitor disease and contaminants in vertebrates found dead, including predatory birds, otters, and cetaceans. Collection is necessarily ad hoc, and careful consideration must be given to potential biases (e.g., unequal probability of sampling certain demographics or locations) and variables which might confound interpretation (e.g., spatial distributions shift over time).

Other noninvasive sampling matrices such as feathers or hair and sampling food items such as invertebrates or ungulate carcasses represent options that adhere to the principles of the 3Rs (reduce, refine, replace). Supporting Information, Table S9, contains examples of some of the studies and schemes in place for contaminant exposure assessment in wildlife.

What are the future research priorities for exposure assessment of pharmaceuticals and wildlife?

1. Further investigate the link between internal exposure and residues in feathers and hair as a noninvasive matrices.
2. Routinely monitor livestock carcasses left for scavengers and determine the awareness of stakeholders responsible for the disposal of medicated carcasses (i.e., veterinarians and farmers) about the impacts of drug residues for wildlife.
3. There is a need to implement a global wildlife monitoring system that enables the correlation of nontarget wildlife intoxications with residues of emerging contaminants such as pharmaceuticals.

3Rs APPROACHES THAT COULD BE USED FOR EFFECTS ASSESSMENT

As outlined in Figure 1, industry is not routinely required to conduct Organisation for Economic Co-operation and Development guideline tests to assess the safety of pharmaceuticals

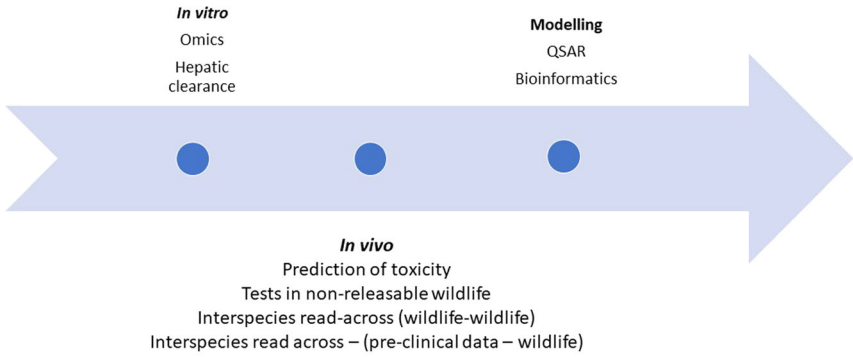

FIGURE 3: In vitro, in vivo, and modeling approaches for filling data gaps for pharmaceutical effects assessment in wildlife. QSAR = quantitative structure–activity relationship.

for wildlife species. With thousands of active ingredients, conducting in vivo safety tests would be extremely costly and entail use of large numbers of vertebrates. For effects assessment, there are options with minimal vertebrate usage (e.g., Swan, Cuthbert, et al. [2006] demonstrated that treating only two birds provides sufficient replication to determine whether the toxicity of diclofenac was similar to that of the white-rumped vulture) of nonreleasable animals at wildlife rehabilitation centers (e.g., Galligan et al., 2020; Rattner et al., 2008). Figure 3 outlines some of the approaches that could be used to fill these data gaps (e.g., quantitative structure–activity relationships [QSARs], approaches looking at effects on the genome or transcriptome, indirect effects, in ovo and in vitro methods, evaluation of sublethal effects, identification of biomarkers that could be used in pathway-based approaches to assist read-across from preclinical mammalian data), while further discussion of priorities related to effects assessment is provided in Supporting Information, 10.

What are the future research priorities for effects assessment and wildlife?

1. Validation and eventual utilization of new approach methodologies to complement or replace safety data in mammals generated in preclinical trials.

CONCLUSIONS

Pharmaceuticals in the environment have been found to cause individual lethality and even population-level effects in wildlife as a result of unique exposure pathways and unexpected sensitivity. Much of our knowledge on the topic has focused on NSAIDs and birds as a result of the Asian vulture crisis. With thousands of drugs licensed for use and no regulatory requirement to conduct in vivo safety tests on a routine basis, it is important to employ noninvasive methods in the field and laboratory to prevent another such crisis involving wildlife. The key questions related to the topic are identified as follows (Textboxes 1–5).

TEXTBOX 1: NSAIDs and wildlife, diclofenac and vultures, current status of population recovery, exposure and risk

Almost two decades after the work of Lindsay Oaks and coworkers (2004), the risk posed by NSAIDs to avian scavengers remains a key research priority. Specifically, 1) documenting the current status of vulture populations in Asia, 2) determining the extent of illegal use of diclofenac in cattle on the Indian subcontinent (Galligan et al., 2021), 3) documenting mortality incidents in avian scavengers associated with diclofenac and other NSAIDs in other geographic regions, 4) determining the relevance of human use of NSAIDs as an exposure route for wildlife, 5) characterizing the hazard of other NSAIDs to other avian species, and 6) elucidating the mechanism of toxicity of NSAIDs in Old World vultures.

TEXTBOX 2: Antimicrobials and effects on microbiota

Antimicrobial resistance is a major challenge in human medicine but may be less relevant for wildlife because they are not intentionally treated with antimicrobials unless admitted to a wildlife sanctuary, which is a rare circumstance. Nonetheless, gaining a greater understanding of the role of wildlife in transferring AMR and AFR organisms is a data gap and research need. In addition, further research is needed to determine whether exposure to antimicrobials and antifungals at much lower doses (i.e., environmental concentrations or residues in treated food rather than therapeutic doses), together with the presence of resistant microorganisms, can affect the microbiome in wildlife.

TEXTBOX 3: The importance of trophic transfer from WWTPs

Insectivorous and omnivorous wildlife (e.g., birds, bats) could theoretically be exposed to pharmaceuticals through direct dietary or dermal routes at or near urban WWTPs (particularly in developing countries with poor secondary or tertiary treatment works) and in rural settings where wastewater or biosolids are used as fertilizer to amend soils, fields, and woodlands (reviewed in Bean & Rattner, 2018). Although modeling efforts have identified physiochemical and pharmacokinetic properties (e.g., environmental persistence, log octanol–water partition coefficient, leachability, half-life) for which pharmaceutical exposure of wildlife has greatest likelihood, robust data (i.e., parent compound or metabolite detected in tissue or excreta) documenting such exposure are lacking.

TEXTBOX 4: To what extent are pharmaceuticals in the environment affecting populations and the diversity of reptiles, amphibians, and mammals?

At present, much of the research on wildlife and pharmaceuticals has been on birds. Global biodiversity is changing rapidly, and initiatives to reduce or halt losses have thus far had limited success (Jetz et al., 2019). Causative agents are likely to be multiple, simultaneous, and potentially synergistic, making direct links between population-level change and specific toxicological threats difficult to decipher. Amphibians are likely to be particularly at risk because of their permeable skin and their reliance on both aquatic and terrestrial habitats at different life stages. Reptiles are the least studied group of vertebrates with regard to environmental contaminant exposure (Hopkins, 2000). Pharmaceutical risk to this vertebrate class remains largely unexplored (although see Mesak et al., 2019). Despite potential trophic transfer of pharmaceuticals by fish and invertebrates, very little research has yet focused on the occurrence, accumulation, or risk from pharmaceuticals in marine mammals (Kleinert et al., 2018).

TEXTBOX 5: Development of specific noninvasive tools for exposure and effects assessment

Development of tools that enable exposure assessment (e.g., feather and hair, expansion of wildlife incident reporting schemes) and the use of new approach methodologies for effects assessment (e.g., genomic and transcriptome analysis, development of wildlife-specific QSARs) would be highly beneficial to minimize the likelihood of another Asian vulture crisis. Ideally, such approaches would involve noninvasive or alternative (to vertebrate) methods.

Supporting information—The Supporting Informationis available on the Wiley Online Library at https://doi.org/10.1002/etc.5528.

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Data Availability Statement—The present study is a review, we have extensively referenced our sources. Data, associated metadata, and calculation tools are available from the corresponding author (thomas.bean@fmc.com).

REFERENCES

Arnold, K. E., Brown, A. R., Ankley, G. T., & Sumpter, J. P. (2014). Medicating the environment: Assessing risks of pharmaceuticals to wildlife and ecosystems. *Philosophical Transactions of the Royal Society B*, *369*(1656), Article 20130569. https://doi.org/10.1098/rstb.2013.0569

Arnold, K. E., Williams, N. J., & Bennet, M. (2016). "Disperse abroad in the land": The role of wildlife in the dissemination of antimicrobial resistance. *Biology Letters*, *12*, Article 20160137. https://doi.org/10.1098/rsbl.2016.0137

Arukwe, A., Myburgh, J., Langberg, H. A., Adeogun, A. O., Braa, I. G., Moeder, M., Schlenk, D., Crago, J. P., Regoli, F., & Botha, C. (2016). Developmental alternations and endocrine-disruptive responses in farmed Nile crocodiles (*Crocodylus niloticus*) exposed to contaminants from the Crocodile River, South Africa. *Aquatic Toxicology*, *173*, 83–93.

Arukwe, A., Rosbak, R., Adeogun, A. O., Langberg, H. A., Venter, A., Myburgh, J., Botha, C., Benedetti, M., & Regoli, F. (2015). Biotransformation and oxidative stress responses in captive Nile crocodile (*Crocodylus niloticus*) exposed to organic contaminants from the natural environment in South Africa. *PLOS ONE*, *10*, Article e0130002.

Atterby, C., Nykvist, M., Lustig, U., Andersson, D. I., Järhult, J. D., & Sandegren, L. (2021). Selection of resistant bacteria in mallards exposed to subinhibitory concentrations of ciprofloxacin in their water environment. *Antimicrobial Agents and Chemotherapy*, *65*(3), Article e01858-20.

Barnes, K. K., Kolpin, D. W., Furlong, E. T., Zaugg, S. D., Meyer, M. T., & Barber, L. B. (2008). A national reconnaissance of pharmaceuticals and other organic wastewater contaminants in the United States—I) groundwater. *Science of the Total Environment*, *402*, 192–200.

Bartelt-Hunt, S., Snow, D. D., Damon-Powell, T., & Miesbach, D. (2011). Occurrence of steroid hormones and antibiotics in shallow groundwater impacted by livestock waste control facilities. *Journal of Contaminant Hydrology*, *123*, 93–103.

Bean, T. G., Arnold, K. E., Lane, J., Bergström, E., Thomas-Oates, J., Rattner, B. A., & Boxall, A. B. A. (2017). Predictive framework for estimating exposure of birds to pharmaceuticals. *Environmental Toxicology and Chemistry*, *36*(9), 2335–2344. https://doi.org/10.1002/etc.3771

Bean, T. G., Boxall, A. B. A., Lane, J., Herborn, K. A., Pietravalle, S., & Arnold, K. E. (2014). Behavioural and physiological responses of birds to environmentally relevant concentrations of an antidepressant. *Philosophical Transactions of the Royal Society B*, *369*(1656), Article 20130575. https://doi.org/10.1098/rstb.2013.0575

Bean, T. G., & Rattner, B. A. (2018). Environmental contaminants of health-care origin: Exposure and potential effects in wildlife. In A. B. A. Boxall & R. S. Kookana (Eds.), *Health care and environmental contamination* (pp. 87–122). Elsevier.

Bean, T. G., Rattner, B. A., Lazarus, R. S., Day, D. D., Burket, S. R., Brooks, B. W., Haddad, S. P., & Bowerman, W. W. (2018). Pharmaceuticals in water, fish and osprey nestlings in Delaware River and Bay. *Environmental Pollution*, *232*, 533–545. https://doi.org/10.1016/j.envpol.2017.09.083

Bezuidenhout, J. D., & Stutterheim, C. J. (1980). A critical evaluation of the role played by the red-billed oxpecker *Buphagus erythrorhynchus* in the biological control of ticks. *Onderstepoort Journal of Veterinary Research*, *47*(2), 51–75.

Blanco, G. (2018). Supplementary feeding as a source of multiresistant *Salmonella* in endangered Egyptian vultures. *Transboundary and Emerging Disease*, *65*, 806–816. https://doi.org/10.1111/tbed.12806

Blanco, G., López-Hernández, I., Morinha, F., & López-Cerero, L. (2020). Intensive farming as a source of bacterial resistance to antimicrobial agents in sedentary and migratory vultures: Implications for local and transboundary spread. *Science of the Total Environment*, *739*, Article 140356.

Bowyer, P., & Denning, D. W. (2014). Environmental fungicides and triazole resistance in *Aspergillus*. *Pest Management Science*, *70*, 173–178.

Boxall, A. B. A., Rudd, M. A., Brooks, B. W., Caldwell, D. J., Choi, K., Hickmann, S., Innes, E., Ostapyk, K., Staveley, J. P., Verslycke, T., Ankley, G. T., Beazley, K. F., Belanger, S. E., Berninger, J. P., Carriquiriborde, P., Coors, A., DeLeo, P. C., Dyer, S. D., Ericson, J. F., … Van Der Kraak, G. (2012). Pharmaceuticals and personal care products in the environment: What are the big questions? *Environmental Health Perspectives*, *120*(9), Article 12211229.

Carroll, D., Wang, J., Fanning, S., & McMahon, B. J. (2014). Antimicrobial resistance in wildlife: Implications for public health. *Zoonoses and Public Health*, *62*, 534–542. https://doi.org/10.1111/zph.12182

Cerveny, D., Grabic, R., Grabicova, K., Randak, T., Joakim Larsson, G. J., Johnson, A. C., Jurgens, M. D., Tysklind, M., Lindberg, R. H., & Fick, J. (2021). Neuroactive drugs and other pharmaceuticals found in blood plasma of wild European fish. *Environment International*, *146*, Article 106188.

Chandramohan, S., Mallord, J. W., Mathesh, K., Sharma, A. K., Mahendran, K., Kesavan, M., Gupta, R., Chutia, K., Pawde, A., Prakash, N. V., Ravichandran, P., Saikia, D., Shringarpure, R., Timung, A., Galligan, T. H., Green, R. E., & Prakash, V. M. (2022). Experimental safety testing shows that the NSAID tolfenamic acid is not toxic to *Gyps* vultures in India at concentrations likely to be encountered in cattle carcasses. *Science of the Total Environment*, *809*, Article 152088. https://doi.org/10.1016/j.scitotenv.2021.152088

Cho, I., Yamanishi, S., Cox, L., Methé, B. A., Zavadil, J., Li, K., Gao, Z., Mahana, D., Raju, K., Teitler, I., Li, H., Alekseyenko, A. V., & Blaser, M. J. (2012). Antibiotics in early life alter the murine colonic microbiome and adiposity. *Nature*, *488*(7413), 621–626. https://doi.org/10.1038/nature11400

Cuthbert, R., Parry-Jones, J., Green, R. E., & Pain, D. J. (2007). NSAIDs and scavenging birds: Potential impacts beyond Asia's critically endangered vultures. *Biology Letters*, *3*, 91–94. https://doi.org/10.1098/rsbl.2006.0554

Cuthbert, R. J., Taggart, M. A., Prakash, V., Chakraborty, S. S., Deori, P., Galligan, T., Kulkarni, M., Ranade, S., Saini, M., Sharma, A. K., Shringarpure, R., & Green, R. E. (2014). Avian scavengers and the threat from veterinary pharmaceuticals. *Philosophical Transactions of the Royal Society, B: Biological Sciences*, *369*(1656), Article 20130574.

Dahlhausen, K. E., Doroud, L., Firl, A. J., Polkinghorne, A., & Eisen, J. A. (2018). Characterization of shifts of koala (*Phascolarctos cinereus*) intestinal microbial communities associated with antibiotic treatment. *PeerJ*, *6*, Article e4452.

Daughton, C. G., & Ternes, T. A. (1999). Pharmaceuticals and personal care products in the environment: Agents of subtle change? *Environmental Health Perspectives*, *107*, 907–937.

Dethlefsen, L., & Relman, D. A. (2011). Incomplete recovery and individualized responses of the human distal gut microbiota to repeated antibiotic perturbation. *Proceedings of the National Academy of Sciences of the United States of America*, *108*(Suppl. 1), 4554–4561.

Espunyes, J., Cabezón, O., Días-Alves, A., Miralles, P., Ayats, T., & Cerdà-Cuéllar, M. (2021). Assessing the role of livestock and sympatric wild ruminants in spreading antimicrobial resistant *Campylobacter* and *Salmonella* in alpine ecosystems. *BMC Veterinary Research*, *17*, Article 79. https://doi.org/10.1186/s12917-021-02784-2

European Medicines Agency. (2006). *Guideline on the environmental risk assessment of medicinal products for human use*. https://www.ema.europa.eu/en/documents/scientific-guideline/guideline-environmental-risk-assessment-medicinal-products-human-use-first-version_en.pdf

European Medicines Agency. (2016). *Questions and answers on "Guideline on the environmental risk assessment of medicinal products for human use."* https://www.ema.europa.eu/en/documents/scientific-guideline/questions-answers-guideline-environmental-risk-assessment-medicinal-products-human-use-revision-1_en.pdf

Focazio, M. J., Kolpin, D. W., Barnes, K. K., Furlong, E. T., Meyer, M. T., Zaugg, S. D., Barber, L. B., & Thurman, M. E. (2008). A national reconnaissance of pharmaceuticals and other organic wastewater contaminants in the United States—II) Untreated drinking water sources. *Science of the Total Environment*, *402*, 201–216.

Fourie, T., Cromarty, D., Duncan, N., Wolter, K., & Naidoo, V. (2015). The safety and pharmacokinetics of carprofen, flunixin and phenylbutazone in the Cape vulture (*Gyps coprotheres*) following oral exposure. *PLOS ONE*, *10*(10), Article e0141419. https://doi.org/10.1371/journal.pone.0141419

Galligan, T. H., Amano, T., Prakash, V. M., Kulkarni, M., Shringarpure, R., Prakash, N., Ranade, S., Green, R. E., & Cuthbert, R. J. (2014). Have population declines in Egyptian vulture and red-headed vulture in India slowed since the 2006 ban on veterinary diclofenac? *Bird Conservation International*, *24*(3), 272–281.

Galligan, T. H., Bhusal, K. P., Paudel, K., Chapagain, D., Joshi, A. B., Chaudhary, I. P., Chaudhary, A., Baral, H. S., Cuthbert, R. J., & Green, R. E. (2020). Partial recovery of critically endangered *Gyps* vulture populations in Nepal. *Bird Conservation International*, *30*(1), 87–102.

Galligan, T. H., Green, R. E., Wolter, K., Taggart, M. A., Duncan, N., Mallord, J. W., Alderson, D., Li, Y., & Naidoo, V. (2022). The non-steroidal anti-inflammatory drug nimesulide kills *Gyps* vultures at concentrations found in the muscle of treated cattle. *Science of the Total Environment*, *807*, Article 150788.

Galligan, T. H., Mallord, J. W., Prakash, V. M., Bhusal, K. P., Sarowar Alam, A. B. M., Anthony, F. M., Dave, R., Dube, A., Shastri, K., Kumar, Y., Prakash, N., Ranade, S., Shringarpure, R., Chapagain, D., Chaudhary, I. P., Jashi, A. B., Paudel, K., Kabir, T., Ahmed, S., … Green, R. E. (2021). Trends in the availability of the vulture-toxic drug, diclofenac, and other NSAIDs in South Asia, as revealed by covert pharmacy surveys. *Bird Conservation International*, *31*(3), 337–353.

Galligan, T. H., Taggart, M. A., Cuthbert, R. J., Svobodova, D., Chipangura, J., Alderson, D., Prakash, V. M., & Naidoo, V. (2016). Metabolism of aceclofenac in cattle to vulture-killing diclofenac. *Conservation Biology*, *30*(5), 1122–1127.

Gkotsis, G., Nika, M.-C., Nikolopoulou, V., Alygizakis, N., Bizani, E., Aalizadeh, R., Badry, A., Chadwick, E., Cincinelli, A., Claßen, D., Danielsson, S., Dekker, R., Duke, G., Drost, W., Glowacka, N., Göckener, B., Jansman, H. A. H., Juergens, M., Knopf, B., … Thomaidis, N. S. (2022). Assessment of contaminants of emerging concern in European apex predators and their prey by LC-QToF MS wide-scope target analysis. *Environment International*, *170*, 107623. https://doi.org/10.1016/j.envint.2022.107623

Gnat, S., Lagowski, D., Dylag, M., & Nowaklewicz, A. (2021). European hedgehogs (*Erinaceus europaeus L.*) as a reservoir of dermatophytes in Poland. *Microbial Ecology*, *84*(2), 363–375. https://doi.org/10.1007/s00248-021-01866-w

Green, R. E., Newton, I., Shultz, S., Cunningham, A. A., Gilbert, M., Pain, D. J., & Prakash, V. (2004). Diclofenac poisoning as a cause of vulture

population declines across the Indian subcontinent. *Journal of Applied Ecology*, *41*(5), 793–800.

Green, R. E., Taggart, M. A., Senacha, K. R., Raghavan, B., Pain, D. J., Jhala, Y., & Cuthbert, R. (2007). Rate of decline of the oriental white-backed vulture population in India estimated from a survey of diclofenac residues in carcasses of ungulates. *PLOS ONE*, *8*, Article e686. https://doi.org/10.1371/journal.pone.0000686

Guillette, L. J., Jr., Crain, D. A., Gunderson, M. P., Kools, S. A., Milnes, M. R., Orlando, E. F., Rooney, A. A., & Woodward, A. R. (2000). Alligators and endocrine disrupting contaminants: A current perspective. *American Zoologist*, *40*, 438–452.

Guillette, L. J., Jr., & Edwards, T. M. (2008). Environmental influences on fertility: Can we learn lessons from studies of wildlife? *Fertility and Sterility*, *89*, e21–e24.

Henny, C. H., Blus, L. J., Kolbe, E. J., & Fitzner, R. E. (1985). Organophosphate insecticide (famphur) topically applied to cattle kills magpies and hawks. *Journal of Wildlife Management*, *49*, 648–658.

Henny, C. H., Kolbe, E. J., Hill, E. F., & Blus, L. J. (1987). Case histories of bald eagles and other raptors killed by organophosphorus insecticides topically applied to livestock. *Journal of Wildlife Diseases*, *23*, 292–295.

Herrero-Villar, M., Delepoulle, É., Suárez-Regalado, L., Solano-Manrique, C., Juan- Sallés, C., Iglesias-Lebrija, J. J., Camarero, P. R., González, F., Álvarez, E., & Mateo, R. (2021). First diclofenac intoxication in a wild avian scavenger in Europe. *Science of the Total Environment*, *782*, Article 146890. https://doi.org/10.1016/j.scitotenv.2021.146890

Herrero-Villar, M., Sánchez-Barbudo, I. S., Camarero, P. R., Taggart, M. A., & Mateo, R. (2021). Increasing incidence of barbiturate intoxication in avian scavengers and mammals in Spain. *Environmental Pollution*, *284*, Article 117452. https://doi.org/10.1016/j.envpol.2021.117452

Herrero-Villar, M., Velarde, R., Camarero, P. R., Taggart, M., Bandeira, V., Fonseca, C., Marco, I., & Mateo, R. (2020). NSAIDs detected in Iberian avian scavengers and carrion after diclofenac registration for veterinary use in Spain. *Environmental Pollution*, *266*, Article 115157. https://doi.org/10.1016/j.envpol.2020.115157

Hopkins, W. A. (2000). Reptile toxicology: Challenges and opportunities on the last frontier in vertebrate ecotoxicology. *Environmental Toxicology and Chemistry*, *19*(10), 2391–2393.

International Union for Conservation of Nature. (2022). *The IUCN red list of threatened species* (Version 2021-3). https://www.iucnredlist.org

Jetz, W., McGeoch, M. A., Guralnick, R., Ferrier, S., Beck, J., Costello, M. J., Fernandez, M., Geller, G. N., Keil, P., Merow, C., Meyer, C., Muller-Karger, F. E., Pereira, H. M., Regan, E. C., Schmeller, D. S., & Turak, E. (2019). Essential biodiversity variables for mapping and monitoring species populations. *Nature Ecology & Evolution*, *3*, 539–551. https://doi.org/10.1038/s41559-019-0826-1

Klein, P. N., Charmatz, K., & Langenberg, J. (1994). The effects of flunixin meglumine (Banamine®) on the renal function in northern bobwhite (*Colinus virginianus*): An avian model. In R. E. Junge (Ed.), *American Association of Zoo Veterinarians and Association of Reptilian and Amphibian Veterinarians, Annual Conference*, 1994, 128–131.

Kleinert, C., Lacaze, E., Fortier, M., Hammill, M., De Guise, S., & Fournier, M. (2018). T lymphocyte-proliferative responses of harbor seal (*Phoca vitulina*) peripheral blood mononuclear cells (PBMCs) exposed to pharmaceuticals in vitro. *Marine Pollution Bulletin*, *127*, 225–234.

Kookana, R. S., Williams, M., Boxall, A. B. A., Larsson, D. G. J., Gaw, S., Choi, K., Yamamoto, H., Thatikonda, S., Zhu, Y.-G., & Carriquiriborde, P. (2014). Potential ecological footprints of active pharmaceutical ingredients: An examination of risk factors in low-, middle- and high-income countries. *Philosophical Transactions of the Royal Society B*, *369*(1656), Article 20130586. https://doi.org/10.1098/rstb.2013.0586

Kruuk, H. (2006). *Otters—Ecology, behaviour and conservation*. Oxford University Press.

Lazarus, R. S., Rattner, B. A., Brooks, B. W., Du, B., McGowan, P. C., Blazer, V. S., & Ottinger, M. A. (2015). Exposure and food web transfer of pharmaceuticals in ospreys (*Pandion haliaetus*): Predictive model and empirical data. *Integrated Environmental Assessment & Management*, *11*, 118–129.

Loucif, L., Chelaghma, W., Cherak, Z., Bendjama, E., Beroual, F., & Rolain, J.-M. (2022). Detection of NDM-5 and MCR-1 antibiotic resistance encoding genes in Enterobacterales in long-distance migratory bird species *Ciconia ciconia*, Algeria. *Science of the Total Environment*, *814*, Article 152861.

Maier, L., Pruteanu, M., Kuhn, M., Zeller, G., Telzerow, A., Anderson, E. E., Brochado, A. R., Fernandez, K. C., Dose, H., Mori, H., Patil, K. R., Bork, P., & Typas, A. (2018). Extensive impact of non-antibiotic drugs on human gut bacteria. *Nature*, *555*(7698), 623–628.

Marin, C., Torres, C., Marco-Jiménez, F., Cerdà-Cuéllar, M., Sevilla, S., Ayats, T., & Vega, S. (2018). Supplementary feeding stations for conservation of vultures could be an important source of monophasic *Salmonella typhimurium* 1,4,[5],12:i:. *Science of the Total Environment*, *636*, 449–455.

Markandya, A., Taylor, T., Longo, A., Murty, M. N., Murty, S., & Dhavalad, K. (2008). Counting the cost of vulture decline—An appraisal of the human health and other benefits of vultures in India. *Ecological Economics*, *67*, 194–204. https://doi.org/10.1016/j.ecolecon.2008.04.020

Mateo, R., Sanchez-Barbudo, I. S., Camarero, P. R., & Martinez, J. M. (2015). Risk assessment of headed vulture (*Gypaetus barbatus*) exposure to topical antiparasitics used in livestock within an ecotoxicovigilance framework. *Science of the Total Environment*, *536*, 704–712.

Mesak, C., Montalvão, M. F., Paixão, C. F. C., de Oliveira Mendes, B., Pereira da Costa Araújo, A., Chagas Quintão, T., & Malafaia, G. (2019). Do Amazon turtles exposed to environmental concentrations of the antineoplastic drug cyclophosphamide present mutagenic damages? If so, would such damages be reversible? *Environmental Science and Pollution Research*, *26*, 6234–6243.

Meteyer, C. U., Rideout, B. A., Gilbert, M., Shivaprasad, H. L., & Oaks, J. L. (2005). Pathology and proposed pathophysiology of diclofenac poisoning in free-living and experimentally exposed oriental white-backed vultures (*Gyps bengalensis*). *Journal of Wildlife Diseases*, *41*(4), 707–716. https://doi.org/10.7589/0090-3558-41.4.707

Miller, T. H., Ng, K. T., Bury, S. T., Bury, S. E., Bury, N. R., & Barron, L. P. (2019). Biomonitoring of pesticides, pharmaceuticals and illicit drugs in a freshwater invertebrate to estimate toxic or effect pressure. *Environment International*, *129*, 595–606.

Moriceau, M. A., Lefebvre, S., Fourel, I., Benoit, E., Buronfosse-Roque, F., Orabi, P., Rattner, B. A., & Lattard, V. (2022). Exposure of predatory and scavenging birds to anticoagulant rodenticides in France: Exploration of data from French surveillance programs. *Science of the Total Environment*, *810*, Article 151291.

Muralidharan, S., & Dhananjayan, V. (2010). Diclofenac residues in blood plasma and tissues of vultures collected from Ahmedabad, India. *Bulletin of Environmental Contamination and Toxicology*, *85*(4), 377–380. https://doi.org/10.1007/s00128-010-0109-7

Naidoo, V., Mompati, K. F., Duncan, N., & Taggart, M. A. (2011). The pied crow (*Corvus albus*) is insensitive to diclofenac at concentrations present in carrion. *Journal of Wildlife Diseases*, *47*(4), 936–944. https://doi.org/10.7589/0090-3558-47.4.936

Naidoo, V., Taggart, M. A., Duncan, N., Wolter, K., Chipangura, J., Green, R. E., & Galligan, T. H. (2018). The use of toxicokinetics and exposure studies to show that carprofen in cattle tissue could lead to secondary toxicity and death in wild vultures. *Chemosphere*, *190*, 80–89. https://doi.org/10.1016/j.chemosphere.2017.08.167

Naidoo, V., Venter, L., Wolter, K., Taggart, M., & Cuthbert, R. (2010). The toxicokinetics of ketoprofen in *Gyps coprotheres*: Toxicity due to zero-order metabolism. *Archives of Toxicology*, *84*, 761–766.

Naidoo, V., Wolter, K., Cromarty, D., Diekmann, M., Duncan, N., Meharg, A. A., Taggart, M. A., Venter, L., & Cuthbert, R. (2010). Toxicity of non-steroidal anti-inflammatory drugs to *Gyps* vultures: A new threat from ketoprofen. *Biology Letters*, *6*(3), 339–341.

Nambirajan, K., Muralidharan, S., Ashimkumar, A. R., & Jadhav, S. (2021). Nimesulide poisoning in white-rumped vulture *Gyps bengalensis* in Gujarat, India. *Environmental Science and Pollution Research International*, *28*(41), 57818–57824. https://doi.org/10.1007/s11356-021-14702-y

Navedo, J. G., Araya, V., & Verdugo, C. (2021). Upraising a silent pollution: Antibiotic resistance at coastal environments and transference to long-distance migratory shorebirds. *Science of the Total Environment*, *777*, Article 146004. https://doi.org/10.1016/j.scitotenv.2021.146004

Oaks, J. L., Gilbert, M., Virani, M. Z., Watson, R. T., Meteyer, C. U., Rideout, B. A., Shivaprasad, H. L., Ahmed, S., Chaudhry, M. J. I., Arshad, M., Mahmood, S., Ali, A., & Khan, A. A. (2004). Diclofenac residues as the cause of vulture population decline in Pakistan. *Nature*, *427*, 630–633.

Orlando, E. F., & Ellestad, L. E. (2014). Sources, concentrations, and exposure effects of environmental gestagens on fish and other aquatic wildlife, with emphasis on reproduction. *General and Comparative Endocrinology*, *203*, 241–249.

Pain, D. J., Bowden, C. G. R., Cunningham, A. A., Cuthbert, R., Das, D., Gilbert, M., Jakati, R. D., Jhala, Y., Khan, A. A., Naidoo, V., Oaks, L., Parry-Jones, P., Prakash, V., Rahmani, A., Ranade, S. P., Baral, H. S., Senacha, K. R., Saravanan, S., Shah, N., … Green, R. E. (2008). The race to prevent the extinction of South Asian vultures. *Bird Conservation International*, *18*, S30–S48. https://doi.org/10.1017/S0959270908000324

Park, K., & Cristinacce, A. (2006). Use of sewage treatment works as foraging sites by insectivorous bats. *Animal Conservation*, *9*, 259–268.

Park, K., Muller, C. T., Markman, S., Swinscow-Hall, O., Pascoe, D., & Buchanan, K. L. (2009). Detection of endocrine disrupting chemicals in aerial invertebrates at sewage treatment works. *Chemosphere*, *77*(11), 1459–1464. https://doi.org/10.1016/j.chemosphere.2009.08.063

Paudel, K., Amano, T., Acharya, R., Chaudhary, A., Baral, H. S., Bhusal, K. P., Chaudhary, I. P., Green, R. E., Cuthbert, R. J., & Galligan, T. H. (2016). Population trends in Himalayan griffon in Upper Mustang, Nepal, before and after the ban on diclofenac. *Bird Conservation International*, *26*(3), 286–292.

Pitarch, A., Gil, C., & Blanco, G. (2017). Oral mycoses in avian scavengers exposed to antibiotics from livestock farming. *Science of the Total Environment*, *605–606*, 139–146. https://doi.org/10.1016/j.scitotenv.2017.06.144

Plaza, P. I., Wiemeyer, G. M., & Lambertucci, S. A. (2022). Veterinary pharmaceuticals as a threat to endangered taxa: Mitigation action for vulture conservation. *Science of the Total Environment*, *817*, Article 152884.

Ramos, B., Rosalino, L. M., Palmeira, J. D., Torres, R. T., & Cunha, M. V. (2022). Antimicrobial resistance in commensal *Staphylococcus aureus* from wild ungulates is driven by agricultural land cover and livestock farming. *Environmental Pollution*, *303*, Article 119116.

Ramudzuli, M. R., & Horn, A. C. (2014). Arsenic residues in soil at cattle dip tanks in the Vhembe district, Limpopo Province, South Africa. *South African Journal of Science*, *110*(7/8), Article 2013-0393.

Rattner, B. A. (2009). History of wildlife toxicology. *Ecotoxicology*, *18*, 773–783. https://doi.org/10.1007/s10646-009-0354-x

Rattner, B. A., Whitehead, M. A., Gasper, G., Meteyer, C. U., Link, W. A., Taggart, M. A., Meharg, A. A., Pattee, O. H., & Pain, D. J. (2008). Apparent tolerance of turkey vultures (*Cathartes Aura*) to the non-steroidal anti-inflammatory drug diclofenac. *Environmental Toxicology and Chemistry*, *27*, 2341–2345.

Rhimi, W., Sgroi, G., Aneke, C. I., Annoscia, G., Latrofa, M. S., Mosca, A., Veneziano, V., Otranto, D., Alastruey-Izquierdo, A., & Cafarchia, C. (2022). Wild boar (*Sus scrofa*) as reservoir of zoonotic yeasts: Bioindicator of environmental quality. *Mycopathologia*, *187*, 235–248. https://doi.org/10.1007/s11046-021-00613-6

Richards, N. L., Cook, G., Simpson, V., Hall, S., Harrison, N., & Scott, K. S. (2011). Qualitative detection of the NSAIDs diclofenac and ibuprofen in the hair of Eurasian otters (*Lutra lutra*) occupying UK waterways with GC-MS. *European Journal of Wildlife Research*, *57*(5), 1107–1114.

Roberts, P. H., & Thomas, K. V. (2006). The occurrence of selected pharmaceuticals in wastewater effluent and surface water of the lower Tyne catchment. *Science of the Total Environment*, *356*, 143–154.

Russell, R. E., & Franson, J. C. (2014). Causes of mortality in eagles submitted to the national wildlife health center 1975–2013. *Wildlife Society Bulletin*, *38*, 697–704. https://doi.org/10.1002/wsb.469

Safholm, M., Ribbenstedt, A., Fick, J., & Berg, C. (2014). Risks of hormonally active pharmaceuticals to amphibians: A growing concern regarding progestogens. *Philosophical Transactions of the Royal Society B*, *369*(1656), Article 20130577. https://doi.org/10.1098/rstb.2013.0577

Scott, K. A., Qureshi, M. H., Cox, P. B., Marshall, C. M., Bellaire, B. C., Wilcox, M., Stuart, B. A., & Njardarson, J. T. (2020). A structural analysis of the FDA Green Book–approved veterinary drugs and roles in human medicine. *Journal of Medicinal Chemistry*, *63*(24), 15449–15482.

Sharma, A. K., Saini, M., Singh, S. D., Prakash, V., Das, A., Dasan, R. B., Pandey, S., Bohara, D., Galligan, T. H., Green, R. E., Knopp, D., & Cuthbert, R. J. (2014). Diclofenac is toxic to the steppe eagle *Aquila nipalensis*: Widening the diversity of raptors threatened by NSAID misuse in South Asia. *Bird Conservation International*, *24*(3), 282–286. https://doi.org/10.1017/S0959270913000609

Shore, R. F., Taggart, M. A., Smits, J., Mateo, R., Richards, N. L., & Fryday, S. (2014). Detection and drivers of exposure and effects of pharmaceuticals in higher vertebrates. *Philosophical Transactions of the Royal Society B*, *369*(1656), Article 20130570. https://doi.org/10.1098/rstb.2013.0570

Shultz, S., Baral, H. S., Charman, S., Cunningham, A. A., Das, D., Ghalsasi, G. R., Goudar, M. S., Green, R. E., Jones, A., Nighot, P., & Pain, D. J. (2004). Diclofenac poisoning is widespread in declining vulture populations across the Indian subcontinent. *Proceedings of the Royal Society of London. Series B: Biological Sciences*, *271*(S6), S458–S460.

Snelders, E., Camps, S. M. T., Karawajczyk, A., Schaftenaar, G., Kema, G. H. J., van der Lee, H. A., Klaassen, C. H., Melchers, W. J. G., & Verweij, P. E. (2012). Triazole fungicides can induce cross-resistance to medical triazoles in *Aspergillus* fumigatus. *PLOS ONE*, *7*, Article e31801.

Srain, H. S., Beazley, K. F., & Walker, T. R. (2021). Pharmaceuticals and personal care products and their sublethal and lethal effects in aquatic organisms. *Environmental Reviews*, *29*, 142–181. https://doi.org/10.1139/er-2020-0054

Stutterheim, C. J., & Brooke, R. K. (1981). Past and present ecological distribution of the yellow billed oxpecker in South Africa. *South African Journal of Zoology*, *16*, 44–49.

Swan, G. E., Cuthbert, R., Quevedo, M., Green, R. E., Pain, D. J., Bartels, P., Cunningham, A. A., Duncan, N., Meharg, A. A., Oaks, L. J., Parry-Jones, J., Shultz, S., Taggart, M. A., Verdoorn, G., & Wolter, K. (2006). Toxicity of diclofenac to *Gyps* vultures. *Biology Letters*, *2*, 279–282. https://doi.org/10.1098/rsbl.2005.0425

Swan, G., Naidoo, V., Cuthbert, R., Green, R. E., Pain, D. J., Swarup, D., Prakash, V., Taggart, M., Bekker, L., Das, D., Diekmann, J., Diekmann, M., Killian, E., Meharg, A., Patra, R. C., Saini, M., & Wolter, K. (2006). Removing the threat of diclofenac to critically endangered Asian vultures. *PLOS Biology*, *4*(3), Article e66. https://doi.org/10.1371/journal.pbio.0040066

Taggart, M. A., Senacha, K. R., Green, R. E., Cuthbert, R., Jhala, Y. V., Meharg, A. A., Mateo, R., & Pain, D. J. (2009). Analysis of nine NSAIDs in ungulate tissues available to critically endangered vultures in India. *Environmental Science & Technology*, *43*, 4561–4566.

Thomason, C. A., Mullen, N., Belden, L. K., May, M., & Hawley, D. M. (2017). Resident microbiome disruption with antibiotics enhances virulence of a colonizing pathogen. *Scientific Reports*, *7*(1), Article 16177.

Tonelli, M., Verdu, J. R., Morelli, F., & Zunino, M. (2020). Dung beetles: Functional identity, not functional diversity, accounts for ecological process disruption caused by the use of veterinary medical products. *Journal of Insect Conservation*, *24*(4), 643–654.

Torres, R. T., Fernandes, J., Carvalho, J., Cunha, M. V., Caetano, T., Mendo, S., Serrano, E., & Fonseca, C. (2020). Wild boar reservoir of antimicrobial resistance. *Science of the Total Environment*, *717*, Article 135001. https://doi.org/10.1016/j.scitotenv.2019.135001

Toutain, P. L., Ferran, A., & Bousquet-Mélou, A. (2010). Species differences in pharmacokinetics and pharmacodynamics. *Comparative and Veterinary Pharmacology*, *199*, 19–48.

US Food and Drug Administration. (1998a). *Guidance for industry: Environmental assessment of human drug and biologics applications*. https://www.fda.gov/media/70809/download

US Food and Drug Administration. (1998b). *Environmental assessment: Questions and answers regarding drugs with estrogenic, androgenic or thyroid activity: Guidance for industry*. https://www.fda.gov/media/91941/download

US Food and Drug Administration. (2017). *Finding of no significant impact (FONSI) for Banaminetransdermal (flunixin transdermal solution) pour-on for beef and dairy cattle®*. https://animaldrugsatfda.fda.gov/adafda/app/search/public/document/downloadFonsi/321

US Food and Drug Administration. (2021). *FDA at a glance: FDA-regulated products and facilities*. https://cacmap.fda.gov/media/154548/download

Valles-Colomer, M., Falony, G., Darzi, Y., Tigchelaar, E. F., Wang, J., Tito, R. Y., Schiweck, C., Kurilshikov, A., Joossens, M., Wijmenga, C., Claes, S., Van Oudenhove, L., Zhernakova, A., Vieira-Silva, S., & Raes, J. (2019). The neuroactive potential of the human gut microbiota in quality of life and depression. *Nature Microbiology*, *4*(4), 623–632. https://doi.org/10.1038/s41564-018-0337-x

Veterinary International Conference on Harmonization. (2000). *Environmental impact assessments (EIA's) for veterinary medicinal products (VMP's)—Phase I, VICH GL6 final guidance* (Guidance for Industry 89). https://cacmap.fda.gov/media/70340/download

Veterinary International Conference on Harmonization. (2006). *Environmental impact assessments (EIA's) for veterinary medicinal products (VMP's)—Phase II, VICH GL38 final guidance* (Guidance for Industry 166). https://www.fda.gov/media/69927/download

Vulture population in India. (2019, July 19). *GK Today*. Retrieved May 22, 2022, from: https://www.gktoday.in/topic/vulture-population-in-india

Wells, K., Butterworth, A., & Richards, N. (2020). A review of secondary pentobarbital poisoning in scavenging wildlife, companion animals and captive carnivores. *Journal of Veterinary Forensic Sciences*, *1*(1), 1–15.

Whitlock, S. E., Pereira, M. G., Lane, J., Sleep, D., Shore, R. F., & Arnold, K. E. (2019). Detecting fluoxetine and norfluoxetine in wild bird tissues and feathers. *Environment International*, *126*, 193–201.

Whitlock, S. E., Pereira, M. G., Shore, R. F., Lane, J., & Arnold, K. E. (2018). Environmentally relevant exposure to an antidepressant alters courtship behaviours in a songbird. *Chemosphere*, *211*, 17–24.

Wilkinson, J. L., Boxall, A. B. A., Kolpin, D. W., Leung, K. M. Y., Lai, R. W. S., Galbán-Malagón, C., Adell, A. D., Mondon, J., Metian, M., Marchant, R. A., Bouzas-Monroy, A., Cuni-Sanchez, A., Coors, A., Carriquiriborde, P., Rojo, M., Gordon, C., Cara, M., Moermond, M., Luarte, T.,… Teta, C. (2022). Pharmaceutical pollution of the world's rivers. *Proceedings of the National Academy of Sciences of the United States of America*, *119*(8), Article e2113947119. https://doi.org/10.1073/pnas.2113947119

Zheng, D., Liwinski, T., & Elinav, E. (2020). Interaction between microbiota and immunity in health and disease. *Cell Research*, *30*(6), 492–506. https://doi.org/10.1038/s41422-020-0332-7

Zorrilla, I., Martinez, R., Taggart, M. A., & Richards, N. (2015). Suspected flunixin poisoning of a wild Eurasian griffon vulture from Spain. *Conservation Biology*, *29*, 587–592.

