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NSAIDs and scavenging birds: potential impacts beyond Asia's critically endangered vultures

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Veterinary treatment of livestock with diclofenac, a non-steroidal anti-inflammatory drug (NSAID), has caused catastrophic declines of *Gyps* vultures in Asia. This has highlighted a lack of knowledge on the potential impacts of NSAIDs on scavenging birds. Surveys of veterinarians and zoos document the outcomes of the treatment of over 870 scavenging birds from 79 species. As well as diclofenac, carprofen and flunixin were associated with mortality, with deaths observed in 13 and 30% of cases, respectively. Mortality was also found following treatment with ibuprofen and phenylbutazone. NSAID toxicity was reported for raptors, storks, cranes and owls, suggesting that the potential conservation impact of NSAIDs may extend beyond *Gyps* vultures and could be significant for New World vultures. In contrast, there were no reported mortalities for the NSAID meloxicam, which was administered to over 700 birds from 60 species. The relative safety of meloxicam supports other studies indicating the suitability of this NSAID to replace diclofenac in Asia.

Keywords: *Gyps*; vultures; NSAIDs; diclofenac; meloxicam; toxicity

1. INTRODUCTION

Populations of three species of *Gyps* vulture in the Indian subcontinent have collapsed since the early 1990s and are now at high risk of extinction (IUCN 2004). Veterinary use of diclofenac, a non-steroidal anti-inflammatory drug (NSAID), is a major cause of the observed population declines (Green *et al.* 2004; Oaks *et al.* 2004). Vultures are exposed to diclofenac when they consume carcasses of livestock that were treated with the drug shortly before death. Experiments show that they die from kidney failure within days of exposure and have extensive visceral gout at post-mortem (Oaks *et al.* 2004; Swan *et al.* 2006*a*). Identical signs of toxicity have been found in carcasses of wild vultures (Oaks *et al.* 2004; Shultz *et al.* 2004).

The collapse in numbers of *Gyps* vultures across Asia means that other scavenging birds are increasingly exposed to contaminated carcasses. Whether diclofenac is affecting them is unknown, although Indian vultures from other genera are also in rapid decline (Cuthbert *et al.* 2006). Diclofenac and other veterinary NSAIDs are licensed and used in many areas of the world, including southern Africa and South America (Anderson *et al.* 2005; J. Parry-Jones 2006, unpublished information). Hence, the use for conservation of 'vulture restaurants' in southern Africa and the veterinary use of diclofenac in South America are a major cause for concern.

Steps are now being taken to control the veterinary use of diclofenac in India and the identification of the NSAID meloxicam as an alternative has facilitated this (Swan *et al.* 2006*b*). However, the safety of meloxicam to other species of scavenging birds and the potential toxicity and the safety of other NSAIDs have not been reported. In this study, we used questionnaire surveys on the clinical use of NSAIDs to make a preliminary assessment of their safety to vultures, raptors and other scavenging birds.

2. MATERIAL AND METHODS

Questionnaires were sent to zoos, wildlife rehabilitation centres and veterinarians worldwide. We requested detailed information on species and number of individuals treated, NSAID or other anti-inflammatory drug used, method of administration, number and frequency of doses, days of treatment, dose level, condition treated and the clinical outcome of treatment. Some survey information could not be completely quantified, particularly for the number of individuals treated. Where respondents replied with 'several', 'many' or 'more than 1', we recorded the number of birds treated as two. Consequently, final sample sizes are likely to be minima. Some birds were treated on multiple occasions. We considered the treatment of an individual (whether single or multiple treatments) with a specific NSAID as the unit of replication. Treatments of the same individual with separate courses of different NSAIDs ($n=4$) were recorded as separate cases.

3. RESULTS

A total of 31 veterinarians and institutions responded, providing information on over 870 cases of NSAID treatment for 79 species of birds including *Gyps* vultures, other raptors, storks, cranes, owls and crows. While owls and cranes are not scavenging birds, the survey provided comprehensive information for owls and one reported an instance of mortality for a crane: consequently the results are presented. Information was also provided on dexamethasone, a steroidal anti-inflammatory drug.

As well as the known diclofenac mortalities, there were 16 instances of mortality with renal disease and gout for a number of NSAIDs across a range of species (figure 1; table 1). Carprofen and flunixin meglumine were associated with mortality of *Gyps* vultures and other species, with a reported mortality of 13% (5/40 cases) and 30% (7/23), respectively. These figures do not include a *Gyps africanus* that died after treatment with both carprofen and ketoprofen, and another that died after receiving either flunixin or ketoprofen. There is no indication that the birds which died received a particularly high dose of carprofen (1–3, 4 and 5 mg kg^{-1}, cf. 1.5–7.6 mg kg^{-1} for all birds treated) or flunixin (1–4.5 mg kg^{-1}, cf. 0.5–12 mg kg^{-1}). Two instances of mortality with renal disease and gout are reported for ibuprofen and phenylbutazone.

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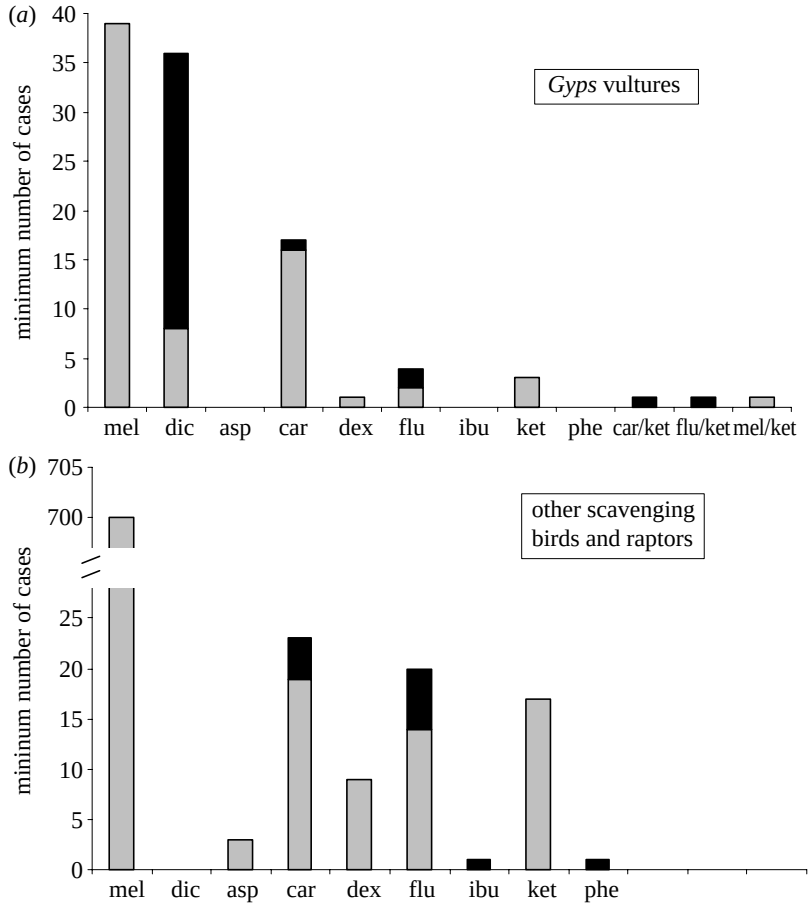

Figure 1. Number of cases of (*a*) *Gyps* vultures ($n=6$ species) and (*b*) other scavenging birds ($n=54$ species) treated with NSAIDs that did not die with gout or renal failure (grey shading) and those treated that died with visceral gout and/or renal failure (black shading). Diclofenac data is taken from Oaks *et al.* (2004) and Swan *et al.* (2006*a*). mel, meloxicam; dic, diclofenac; asp, aspirin; car, carprofen; dex, dexamethasone; flu, flunixin; ibu, ibuprofen; ket, ketoprofen and phe, phenylbutazone. Where two drugs are indicated both were administered simultaneously or there is uncertainty about which drug was used (table 1).

There were no mortalities following treatment with meloxicam. For *Gyps* vultures, 39 individuals from six species (*G. africanus*, *Gyps bengalensis*, *Gyps coprotheres*, *Gyps fulvus*, *Gyps himalayensis* and *Gyps rueppellii*) have been treated and a minimum of 700 birds from 54 other raptors and scavenging species were given meloxicam (see electronic supplementary material). Meloxicam doses ranged from 0.1 to 0.75 mg kg^{-1} bw, with a median dose of 0.5 mg kg^{-1}. Meloxicam was administered by intramuscular injection (57% of treatments), orally (32%) or through a combination of one intramuscular injection followed by oral dosing (11%). Treatment ranged from 1 to 120 days (median 5 days). Less information is available on the safety of other NSAIDs, although the survey results indicate ten cases where dexamethasone (a steroidal anti-inflammatory) and 20 instances where ketoprofen (when this drug was administered on its own) have been administered with no reported mortalities.

4. DISCUSSION

Our results show that certain NSAIDs are toxic to raptors, storks, cranes and owls and suggest that the conservation impact of diclofenac and other NSAIDs may not be restricted to *Gyps* vultures. Of particular significance is the mortality of a Marabou stork (*Leptoptilus crumeniferus*) following treatment with flunixin. Storks and New World vultures are phylogenetically closely related (Sibley *et al.* 1988), and the veterinary use of NSAIDs within South America is consequently of potential conservation concern; testing the toxicity of NSAIDs to New World vultures is a priority. The survey also highlights the relative safety of meloxicam to a wide range of bird species, with over 739 individuals from 60 species treated with no mortalities. More information is required to assess the safety of ketoprofen and dexamethasone since the number of birds treated is small, and there are reported concerns on the safety of ketoprofen in ducks (Mulcahy *et al.* 2003).

Carprofen and flunixin appear to carry a high risk of renal damage in birds, which supports earlier findings concerning the safety of flunixin (Klein *et al.* 1994; Clyde & Murphy 1999). Carprofen and flunixin are used for the treatment of livestock within Europe, although they are not yet available in South Asia. The published information on carprofen and flunixin in livestock tissue residues indicate that a vulture consuming a 1 kg meal from an animal that died shortly after a veterinary course of these drugs, could be exposed to doses close to, or within, the range of doses (1–5 mg kg^{-1}) that caused mortality of birds after clinical treatment

Table 1. Questionnaire results indicating drug used, toxicity, number of cases, range of doses and species treated. (Detailed results on the 60 species treated and dose of meloxicam are available from the electronic supplementary material.)

drug	toxicity	*N* cases	dose (mg kg^{-1})	species treated ($n>1$)
aspirin	no	3	5.4–6.4	*Aegypius monachus*, *Ciconia ciconia*, *Corvus corax*
dexamethasone[a]	no	10	0.2–5.0	*Gyps himalayensis*, *Leptoptilos crumeniferus*, *Bubulcus ibis*, *Vultur gryphus* (2), *Ciconia ciconia* (4), *Tyto alba*
ketoprofen	no	20	1.0–7.7	*Gyps fulvus* (2), *Gyps rueppellii*, *Aegypius monachus*, *Necrosyrtes monachus*, *Buteo jamiacensis* (2), *Geranoaetus melanoleucus* (2), *Vultur gryphus* (2), *Leptoptilos crumeniferus* (2), *Corvus ossifragus*, *Asio flammeus flammeus* (2), *Bubo virginianus* (2), *Otus asio* (2)
meloxicam	no	739	0.1–0.75	60 species treated (see electronic supplementary material for details on species and dose rates)
ketoprofen and meloxicam	no	1	ket 1.0, mel 0.2	*Gyps africanus*
carprofen	yes	5	1.0–5.0	*Gyps fulvus*, *Parabuteo unicinctus* (2), *Aegolius acadicus* (2)
carprofen	no	35	1.5–7.6	*Gyps africanus* (3), *Gyps bengalensis* (2), *Gyps fulvus* (5), *Gyps himalayensis* (3), *Gyps rueppellii* (2), *Gyps rueppellii x africanus*, *Aegypius monachus* (3), *Necrosyrtes monachus*, *Torgus tracheliotus*, *Buteo jamiacensis Haliaeetus leucocephalus* (7), *Ciconia ciconia*, *Ephippiorhynchus senegalensis*, *Bugeranus carunculatus*, *Grus vipio*, *Ardeotis kori* (2)
diclofenac	yes	28	0.1–2.5	*Gyps bengalensis* (23), *Gyps africanus* (2), *Gyps fulvus* (3)
diclofenac	no	8	0.25–0.6	*Gyps bengalensis* (8)
flunixin	yes	7	1.0–4.5	*Gyps rueppellii*, *Cariana cristata*, *Leptoptilos crumeniferus*, *Platalea alba*, *Aegypius monachus* (3)
flunixin	no	16	0.5–12.0	*Gyps fulvus*, *Gyps rueppellii*, *Haliaeetus leucocephalus*, *Terathopius ecaudatus*, *Parabuteo unicinctus*, *Leptoptilos crumeniferus*, *Aegypius monachus*, *Vultur gryphus* (2), *Ciconia ciconia* (2), *Buteo jamiacensis* (5)
ibuprofen	yes	1	—	*Aegypius monachus*
phenylbutazone	yes	1	—	*Torgus tracheliotus*
flunixin or ketoprofen	yes	1	—	*Gyps africanus*
carprofen and ketoprofen	yes	1	car 7.2, ket 4.3	*Gyps africanus*

[a] Dexamethasone is a steroidal anti-inflammatory drug, not an NSAID.

Table 2. Evidence for NSAID toxicity on vultures, raptors and other scavenging birds indicating the number of birds that died with gout and/or renal failure and total number of birds treated, the ratio of COX-1/COX-2 inhibition in human, equine and canine blood, and the presence of either an –NH, –COOH or both –NH and –COOH groups in the molecular structure. (Data on COX-1/COX-2 ratios come from Brideau *et al.* (2001) and Lees *et al.* (2004).)

			COX-1/COX-2 inhibition			molecular structure		
drug	toxicity	died/total	(human)	(equine)	(canine)	–NH	–COOH	–NH and –COOH
aspirin	no	0/3	0.14	—	—	no	yes	no
ketoprofen	no	0/20	0.02	—	0.6	no	yes	no
meloxicam	no	0/739	2.71	—	10	yes	no	no
carprofen	yes	5/40	0.02	1.6	6.5	yes	yes	yes
diclofenac	yes	28/36	1.97	—	1	yes	yes	yes
flunixin	yes	7/24	—	0.3	—	yes	yes	yes
ibuprofen	yes	1/1	1.06	—	—	no	yes	no
phenylbutazone	yes	1/1	—	1.6	0.6	no	no	no

(see electronic supplementary material). Consequently, the veterinary use of flunixin and carprofen in South Asia could result in similar problems to those caused by diclofenac. Currently, a range of NSAIDs are recommended for veterinary use in India (Anonymous 2002), including drugs this study has found associated with mortality. This highlights the need for robust safety testing before recommending any NSAID as a safe replacement for diclofenac (Swan *et al.* 2006*b*).

Knowledge of the mechanism of NSAID toxicity in vultures is currently lacking, although Meteyer *et al.*

(2005) propose that diclofenac toxicity of *G. bengalensis* is a consequence of renal ischemia through activation of renal portal valves. The same clinical signs at post-mortem (renal disease and visceral gout) are found for diclofenac, carprofen and flunixin; suggesting that the mechanism of toxicity may be similar. NSAIDs operate through the inhibition of the cyclo-oxygenase enzymes, COX-1 and COX-2, and the relative inhibition of these two enzymes is thought to alter the risk of adverse effects on renal function (Brater 2002). The hepatotoxicity of different NSAIDs has also been linked to chemical structure, with evidence for toxicity where there is a carboxylic acid group (–COOH) in combination with a nearby linking –NH group (Sussman & Kelly 2003). Consideration of the eight NSAIDs reported in this study, suggest that there is no simple relationship between NSAID toxicity and COX-1/COX-2 inhibition (table 2). However, there is some support that the presence of both –COOH and –NH groups is associated with toxicity, as these structures are present in the NSAIDs most associated with mortality and are absent from those NSAIDs that exhibited no signs of toxicity (table 2). However, ibuprofen and phenylbutazone do not conform to this pattern and this hypothesis requires further investigation.

In conclusion, our survey suggests that widespread use of NSAIDs may be having impacts on bird populations in addition to the known effect of diclofenac on *Gyps* vultures. At least two NSAIDs, in addition to diclofenac, show evidence of toxicity to scavenging birds. However, the conclusion that meloxicam is not toxic to scavenging birds at concentrations likely to be encountered is supported by the survey and supports the use of this drug as an alternative for diclofenac.

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