The Great Indian Vulture Crisis…

A mysterious and precipitous plunge in the number of vultures in South Asia, which has pushed three species to the brink of extinction, is probably a result of inadvertent poisoning by a drug used widely in livestock to relieve fever and lameness.

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"The decline of Asian vultures is one of the steepest declines experienced by any bird species." —Dr. Debbie Pain, RSPB

"Everyone knows about the plight of India's tigers, but in the race towards extinction the vultures will get there far sooner!" —Richard Cuthbert, RSPB

The oriental white-backed vulture, once thought to be the commonest bird of prey in the world is now the fastest declining wild bird in history.

The Asian vulture could be flapping its last. - Indian naturalists.

The vultures are disappearing faster than dodos did.

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Acknowledgement

We are very thankful to Prof. Shiv Vishvanathan for giving us an opportunity to work on this project.

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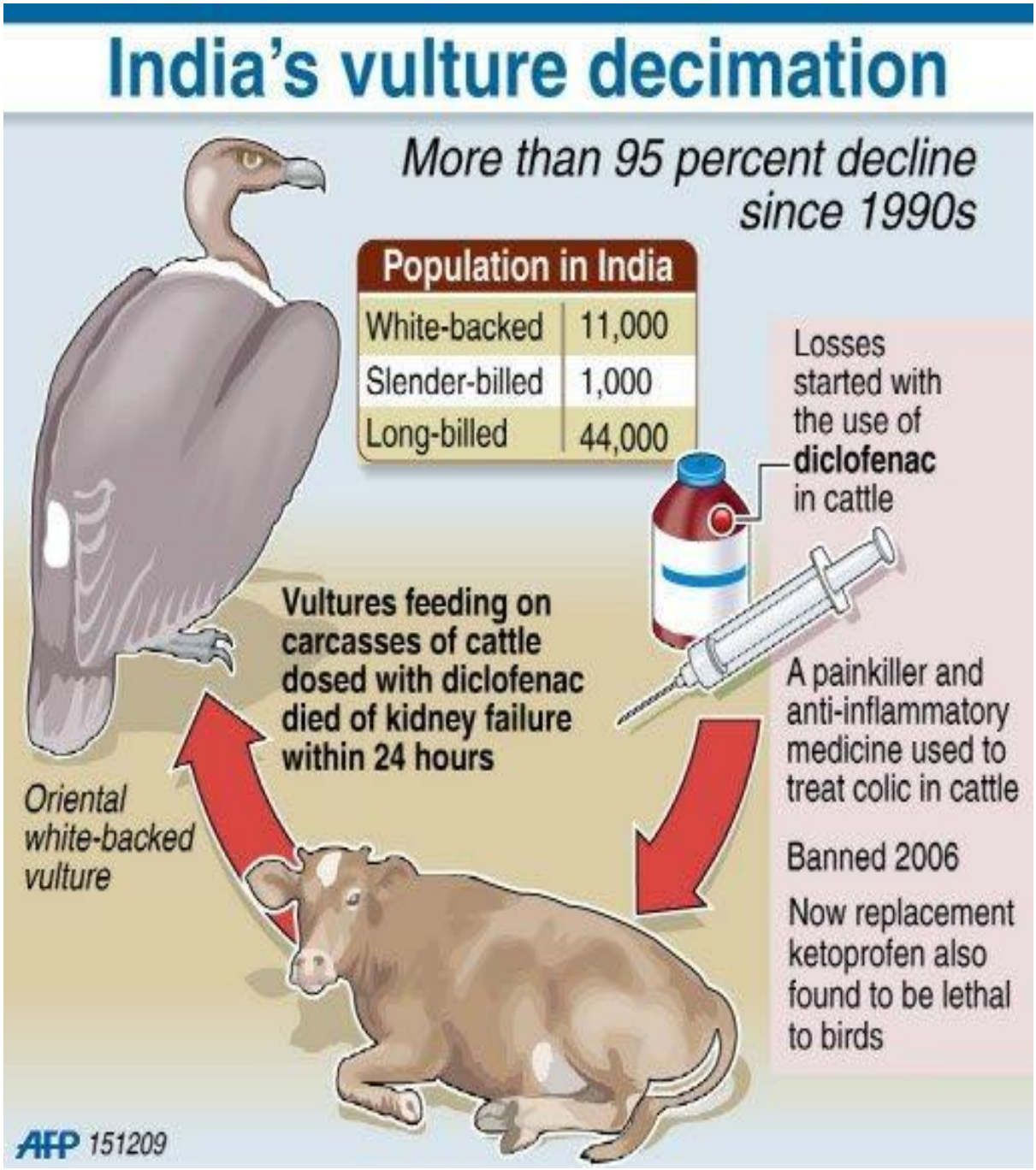

India's vulture decimation
More than 95 percent decline since 1990s
Population in India
White-backed 11,000
Slender-billed 1,000
Long-billed 44,000
Losses started with the use of diclofenac in cattle
A painkiller and anti-inflammatory medicine used to treat colic in cattle
Banned 2006
Now replacement ketoprofen also found to be lethal to birds
Vultures feeding on carcasses of cattle dosed with diclofenac died of kidney failure within 24 hours
Oriental white-backed vulture
AFP 151209

Problem definition

Gyps vulture populations across the Indian subcontinent collapsed in the 1990s and continue to decline. Repeated population surveys showed that the rate of decline was so rapid that elevated mortality of adult birds must be a key demographic mechanism. Post mortem examination showed that the majority of dead vultures had visceral gout, due to kidney damage. The realization that diclofenac, a non-steroidal anti-inflammatory drug potentially nephrotoxic to
birds, had become a widely used veterinary medicine led to the identification of diclofenac poisoning as the cause of the decline. Surveys of diclofenac contamination of domestic ungulate carcasses, combined with vulture population modeling, show that the level of contamination is sufficient for it to be the sole cause of the decline. Testing on vultures of meloxicam, an alternative NSAID for livestock treatment, showed that it did not harm them at concentrations likely to be encountered by wild birds and would be a safe replacement for diclofenac. The manufacture of diclofenac for veterinary use has been banned, but its sale has not. Consequently, it may be some years before diclofenac is removed from the vultures' food supply. In the meantime, captive populations of three vulture species have been established to provide sources of birds for future reintroduction programmes.

How it all started??

Nine species of vultures are recorded from Indian subcontinent.
The populations of three resident Gyps species:

1) Oriental White-Backed Vulture- Gyps bengalensis (OWBV)
2) Long-billed Vulture- Gyps indicus (LBV)
3) Slender-billed Vulture- Gyps tenuirostris(SBV)

crashed during the mid-nineties of the last century.
Vulture declines were first documented at Keoladeo National Park, Bharatpur, Rajasthan. Subsequently, the crash in populations was documented across the country. Surveys on identified tracks were done in 2007 to repeat surveys done previously in 1992, 2000, 2002 and 2003. This was done to determine the population trend in the three species of vultures and also to get a rough estimate of the surviving population of vultures in 2007.

Although Gyps vulture populations were probably declining slowly in many parts of the world during the 20th century, a very different situation existed in India, Nepal and Pakistan.
Here, large populations of OWBV and LBV remained until the 1990s. Large numbers of SBV, which was not distinguished as a separate species from LBV until recently , were also found in the northeastern parts of the subcontinent.

Indeed, during the 1980s OWBV was thought likely to be the commonest large bird of prey in the world. In India, Gyps vulture densities were so high in some areas that they were considered a hazard to aircraft. This abundance was undoubtedly due to a plentiful food supply, in the form of the carcasses of domesticated ungulates. The keeping of livestock for milk production and as
beasts of burden is common in rural areas across the Indian subcontinent and cattle are abundant in many towns and cities. Livestock numbers in India had exceeded 400 million since the 1980s,and reached 500 million in 2005
(ILC 2003, projection based on Animal Husbandry Statistics,Government of India).

In large parts of the subcontinent, Hindu beliefs prohibit the slaughter of
cows. When feral and domestic cows die a natural death they are left in the open in rural areas or disposed of in regulated carcass dumps around towns and cities. Skinners remove the hides from dead cattle for the leather industry, leaving vultures

to scavenge the remaining soft tissue. As Conservation of South Asian vultures S31 vulture populations benefited from the large amounts of food available, Indian society gained environmental health and other benefits from a free carcass disposal service. A flock of vultures can pick a cow carcass clean in a few hours, leaving little more than bones, that then dry rapidly in the sun, and are gathered by bone collectors for the fertilizer, gelatin and glue industries.
Whilst vultures feed primarily on large ungulates, they were also historically the key scavenger of the dead from the ancient Parsi religion, who lay their dead out in the open in enclosures or specially constructed 'Towers of Silence'. Vultures also have spiritual significance in Hindu mythology, as the vulture-king Jatayu died attempting to protect Sita, one of the principal characters of the Hindu epic 'Ramayana', from the demon king Ravana, while her husband Prince Rama was away hunting.

The era of abundant Gyps vultures in the Indian subcontinent came to a sudden end in the 1990s. By the mid-1990s, newspapers in north India started publishing reports of vultures rapidly disappearing from carcass dumps. This was also documented by the Bombay Natural History Society (BNHS) whilst monitoring raptor numbers in Keoladeo National Park, a World
Heritage Site at Bharatpur in eastern Rajasthan. In the mid-1980s, foraging vultures were numerous in the park. Several hundred pairs of OWBV nested within it and hundreds of pairs of LBV nested on cliffs at Bayana, not far outside. Between the late 1980s and mid to late 1990s, numbers of these two species found in the park had declined dramatically.
Numbers of OWBV nests declined from 244–353 in the 1980s to none by the 1999/2000 breeding season. There was also anecdotal evidence of a
general decline in vulture numbers throughout much of northern India during the late 1990s.

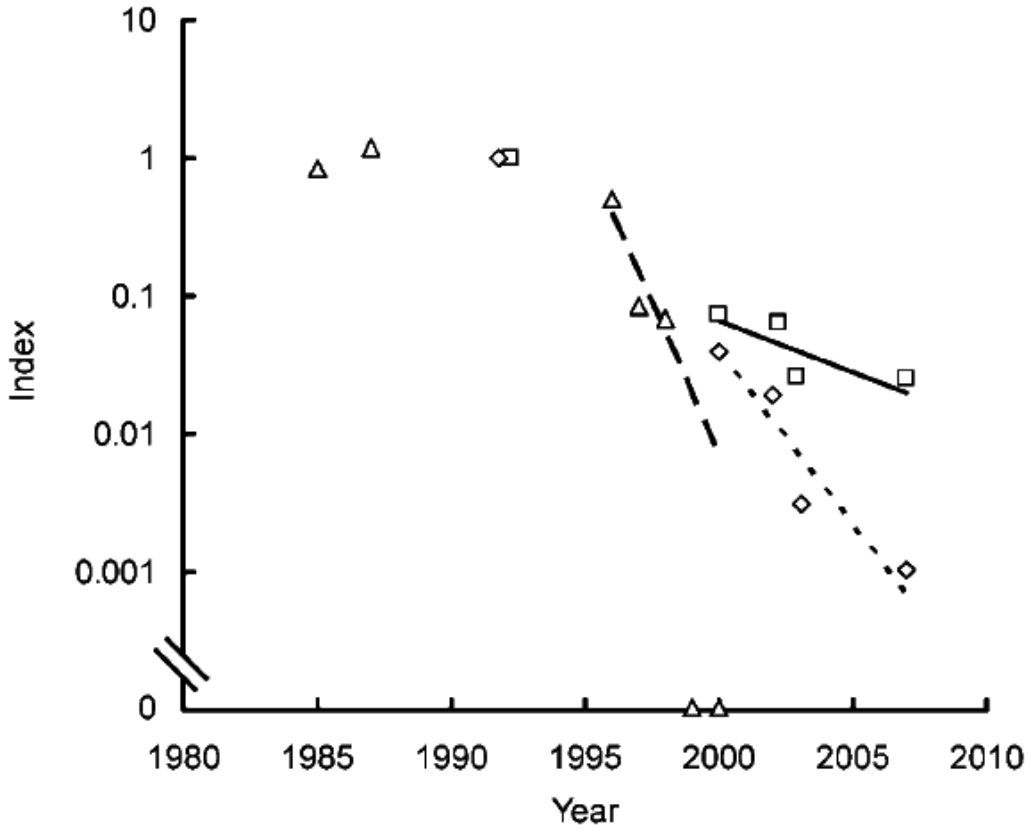
However, as there was little systematic bird monitoring, it was difficult to know whether reports reflected a truly nationwide decline, or isolated local changes. With support from the US Fish and Wildlife Service, BNHS had conducted nationwide raptor surveys in many parts of India between 1991 and 1993 using a repeatable road transect method. Surveys were carried out in, near, and along the routes travelled between protected areas. They covered large parts of north, west and eastern India. Unfortunately, not all Gyps vultures were counted
because they were considered too numerous for this to be practicable. However, the surveyors counted vultures in any groups of five or more birds. BNHS, with support from the RSPB, repeated the road transect surveys in 2000. The results were dramatic. **Both OWBV and LBV had almost disappeared from the areas surveyed**.

The population of OWBV across the surveyed range had declined by 96% between 1991–93 and 2000 and that of LBV by 92%. It should be noted that these were minimum declines because individual vultures and those seen in small groups were counted in 2000, but not in 1991–1993.
Subsequent counts on these and additional transects in 2002, 2003 and 2007 showed that OWBV and LBV continued to decline at an average rate of 44% (OWBV) and 16% (LBV) per year between 2000
and 2007. SBV was not distinguished from LBV until the 2002 count, when
it was found to comprise less than 2% of the combined total of the LBV and SBV count.
Comparison of the 2002, 2003 and 2007 counts indicated that the population of SBV was declining in India about as rapidly as LBV.
Following the results of the 2000 surveys, BNHS organized an international meeting in September 2000.
The meeting, held in New Delhi, was supported by the Ministry of
Environment and Forests (MoEF) of the Government of India and the RSPB, and was attended by national and international scientists, conservationists, and Indian government representatives. Among those represented was **The Peregrine Fund**, who joined forces with Washington State University and the **Ornithological Society of Pakistan (OSP)** to conduct vulture studies in
Pakistan. Subsequent counts of breeding pairs of OWBV in nesting colonies in Punjab province, Pakistan, revealed a population decline at a rate of 50% per year between 2000 and 2003.
This decline continued to extinction at several formerly large OWBV colonies in the province . This group also counted nesting LBV in Sind province, Pakistan , where numbers have declined by about two-thirds between 2002 and 2006; an average annual decline rate of 25% per year.
Hence, both in India and Pakistan, the rates of population decline of LBV, though rapid, are substantially slower (16% and 25% per year respectively) than the catastrophic decline rates for OWBV (44% and 50%). The similarity of the recent average decline rates in the two countries is striking for both species.
Backwards extrapolation of log-linear Poisson regression models of
counts of vultures across India, and of vulture nests at Keoladeo National Park, suggest that **the vulture declines probably started in the early to mid-1990s**.

Nest counts of OWBV by Bird Conservation Nepal (BCN) in eastern Nepal
suggested similar rates of decline there, with 65 active nests found at Koshi in 2000–01 falling to just 14 in 2002–03.
The work of two main research groups was to prove crucial in the search for the cause of declines and solutions.

BNHS led one group, initially comprising the Forest Department of the state government of Haryana, the RSPB, the Zoological Society of London (ZSL) and the National Birds of Prey Trust (NBPT), and later expanding to include a wide range of national and international organizations.

The second group comprised The Peregrine Fund (TPF),
Washington State University and the Ornithological Society of Pakistan (OSP). Whilst the BNHS consortium focused largely on India, the TPF/OSP group conducted a complementary research programme in Pakistan, and BCN worked in Nepal in collaboration with both groups.



Population declines of Gyps vultures in India.
Points show indices of population size from counts on a logarithmic scale, plotted against calendar year. This index represents the vulture population size as a proportion of the initial level. Triangles represent the number of active nests of Gyps bengalensis in Keoladeo National Park expressed as a proportion of the average in the 1980s.
Indices of population size, relative to that in 1992, of G. bengalensis (diamonds) and G. indicus and G. tenuirostris combined (squares) in northern India were calculated from road transect count data. Lines represent fitted log-linear regression models (dashed line 5 G. bengalensis at Keoladeo, dotted line 5 G. bengalensis on road transects, solid line 5 G. indicus/tenuirostris on road transects).

Uses and Significance of Vultures

1. In large parts of the subcontinent, Hindu beliefs prohibit the slaughter of cows. When feral and domestic cows die a natural death they are left in the open in rural areas or disposed of in regulated carcass dumps around towns and cities. Skinners remove the hides from dead cattle for the leather industry, leaving vultures to scavenge the remaining soft tissue. As Conservation of South Asian vultures S31 vulture populations benefited from the large amounts of food available, Indian society gained environmental health and other benefits from a free carcass disposal service. A flock of vultures can pick a cow carcass clean in a few hours, leaving little more than bones, that then dry rapidly in the sun, and are gathered by bone collectors for the fertilizer, gelatin and glue industries.

2. Spiritual significance - Vultures also have spiritual significance in Hindu mythology, as the vulture-king Jatayu died attempting to protect Sita, one of the principal characters of the Hindu epic 'Ramayana', from the demon king Ravana, while her husband Prince Rama was away hunting (Griffith 1870–1874).

3. German police are testing the use of vultures to seek out human corpses in a unique project aimed at dramatically speeding up criminal investigations.
A bird expert at a wildlife park in northern Germany is training Sherlock, a five-year old turkey vulture, to locate fabric containing the scent of dead people.
The scheme was commissioned by the Lower Saxony police force after a senior officer, Rainer Herrmann, watched a BBC wildlife documentary about the extraordinary sense of smell of turkey vultures, which are indigenous to the Americas. The programme showed the birds finding hidden meat with ease.

Trained sniffer dogs such as bloodhounds are highly effective in tracking and remembering scents, but they need to take frequent breaks and can only scour 100 square meters per day, or even less if the terrain is difficult.

"Vultures can fly over many square kilometers. They could make police work much more efficient," said Mr. Herrmann, an expert on forensic science and technology at the Lower Saxony criminal police force. It would take the birds just a few hours to cover areas that would take dogs days.

The birds are capable of detecting scents from the air, even through forest canopies. Under the plan, tracking devices would be attached to the vultures so that they could be traced by police.

4. Whilst vultures feed primarily on large ungulates, they were also historically the key
scavenger of the dead from the ancient Parsi religion, who lay their dead out in the open in
enclosures or specially constructed 'Towers of Silence' (Pain et al. 1993).

Why were the vultures getting affected?

Populations of the *Gyps* vultures of southern Asian countries have been declining precipitously during the recent past, especially in the western parts of its distributional range.

A linkage between the common non-steroidal anti-inflammatory drug (NSAID) 'Diclofenac' and the mortality of these vultures was established recently.Post-mortem examinations of vulture carcasses revealed visceral gout (deposition of uric acid crystals in the tissues) as the cause of mortality in most of the dead birds. The visceral gout condition could be induced by a variety of potential factors, mostly associated with environmental degradation and use of Diclofenac. It appears quite likely that many such factors together have made a cumulative contribution towards the observed population decline of *Gyps* vultures during the recent years, which has pushed these scavenging sky-lords to near extinction.

What Are NSAIDs

(Non-steroidal Anti-inflammatory Drugs)

Nonsteroidal anti-inflammatory drugs, usually abbreviated to NSAIDs or NAIDs, are drugs with analgesic, antipyretic (fever-reducing) and, in higher doses, with anti-inflammatory effects (reducing inflammation). The term "nonsteroidal" is used to distinguish these drugs from steroids, which (among a broad range of other effects) have a similar eicosanoid-depressing, anti-inflammatory action. As analgesics, NSAIDs are unusual in that they are non-narcotic.

NSAIDs are sometimes also referred to as nonsteroidal anti-inflammatory agents/analgesics (NSAIAs) or nonsteroidal anti-inflammatory medicines (NSAIMs). The most prominent members of this group of drugs are aspirin, ibuprofen, and naproxen partly because they are available over-the-counter in many areas.

Classification:

NSAIDs can be broadly classified based on their chemical structure.

Propionic acid derivatives

- Ibuprofen
- Naproxen
- Fenoprofen
- Ketoprofen
- Flurbiprofen
- Oxaprozin

Acetic acid derivatives

- Indomethacin
- Sulindac
- Etodolac
- Diclofenac

Enolic acid (Oxicam) derivatives

- Piroxicam
- Meloxicam
- Tenoxicam
- Droxicam
- Lornoxicam
- Isoxicam

Fenamic acid derivatives

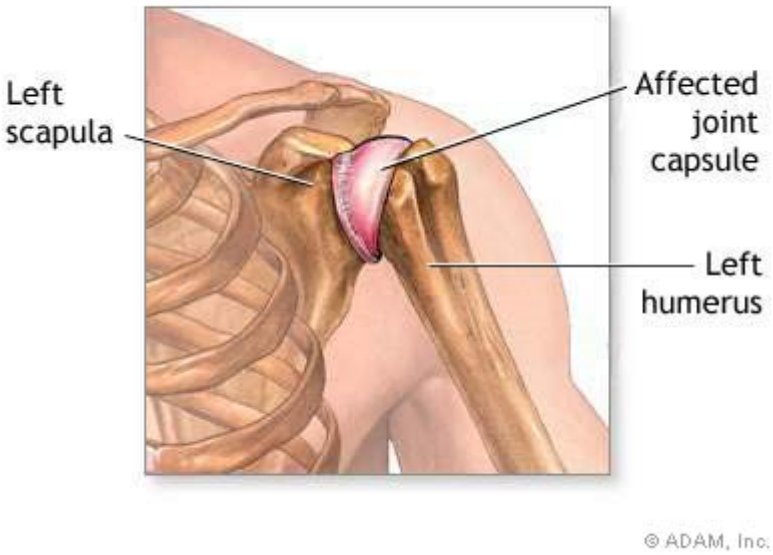
- Mefenamic acid
- Meclofenamic acid
- Flufenamic acid
- Tolfenamic acid

Selective COX-2 inhibitors (Coxibs)

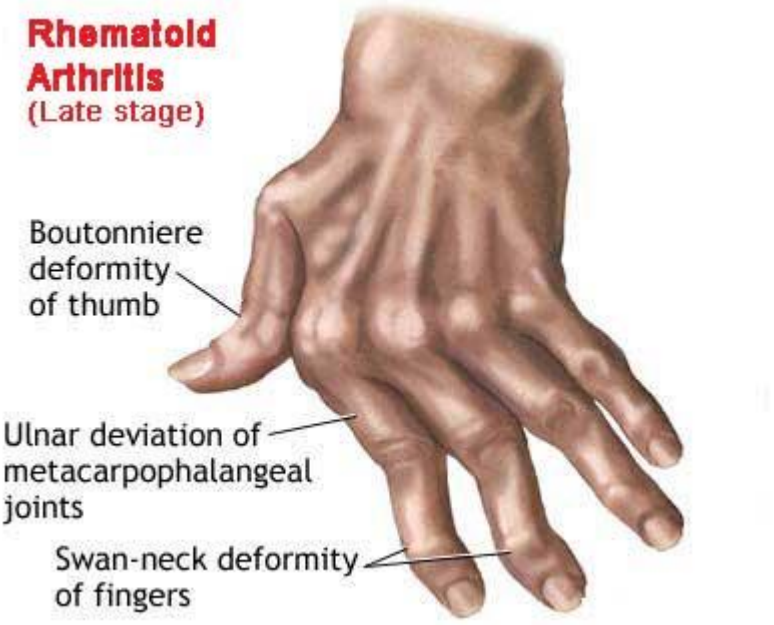
- Celecoxib
- Rofecoxib
- Valdecoxib
- Parecoxib
- Lumiracoxib
- Etoricoxib

Conditions for which NSAIDs are used:

NSAIDs are used primarily to treat inflammation, mild to moderate pain, and fever. Specific uses include the treatment of headaches, arthritis, sports injuries, and menstrual cramps. Ketorolac (Toradol) is only used for short-term treatment of moderately severe acute pain that otherwise would be treated with opioids. Aspirin (also an NSAID) is used to inhibit the clotting of blood and prevent strokes and heart attacks in individuals at high risk. NSAIDs also are included in many cold and allergy preparations.



An inflammation of the shoulder joint can cause pain and restricted joint movement.



Different types of NSAIDs:

NSAIDs vary in their potency, duration of action, how they are eliminated from the body, how strongly they inhibit COX-1 and their tendency to cause ulcers and promote bleeding. The more an NSAID blocks COX-1, the greater is its tendency to cause ulcers and promote bleeding. One NSAID, celecoxib (Celebrex), blocks COX-2 but has little effect on COX-1, and is therefore further classified as a selective COX-2 inhibitor. Selective COX-2 inhibitors cause less bleeding and fewer ulcers than other NSAIDs.

Aspirin is a unique NSAID, not only because of its many uses, but because it is the only NSAID that inhibits the clotting of blood for a prolonged period (4 to 7 days). This prolonged effect of aspirin makes it an ideal drug for preventing blood clots that cause heart attacks and strokes.

Most NSAIDs inhibit the clotting of blood for only a few hours. Ketorolac (Toradol) is a very potent NSAID and is used for moderately severe acute pain that usually requires narcotics. Ketorolac causes ulcers more frequently than other NSAID. Therefore, it is not used for more than five days. Although NSAIDs have a similar mechanism of action, individuals who do not respond to one NSAID may respond to another

Side effects of NSAIDs:

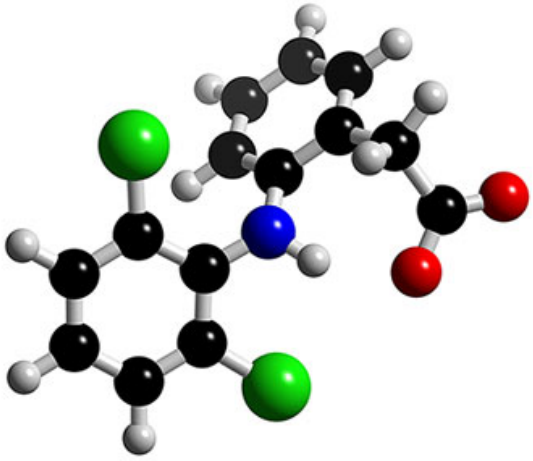
NSAIDs are associated with several side effects. The frequency of side effects varies among NSAIDs. The most common side effects are nausea, vomiting, diarrhea, constipation, decreased appetite, rash, dizziness, headache, and drowsiness. NSAIDs may also cause fluid retention, leading to edema. The most serious side effects are kidney failure, liver failure, ulcers and prolonged bleeding after an injury or surgery. Some individuals are allergic to NSAIDs and may develop shortness of breath when an NSAID is taken. People with asthma are at a higher risk for experiencing serious allergic reaction to NSAIDs. Individuals with a serious allergy to one NSAID are likely to experience a similar reaction to a different NSAID.

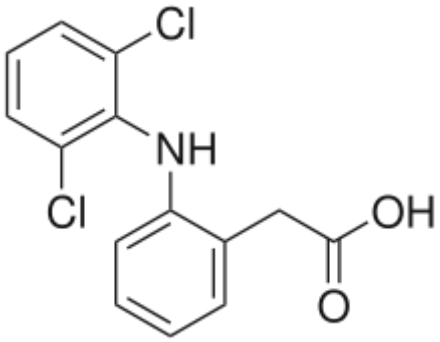
Use of aspirin in children and teenagers with chickenpox or influenza has been associated with the development of Reye's syndrome. Therefore, aspirin and non-aspirin salicylates [for example, salsalate (Amigesic)] should not be used in children and teenagers with suspected or confirmed chickenpox or influenza.

NSAIDs may increase the risk of potentially fatal, stomach and intestinal adverse reactions (for example, bleeding, ulcers, and perforation of the stomach or intestines).

These events can occur at any time during treatment and without warning symptoms. Elderly patients are at greater risk for these adverse events. NSAIDs (except low dose aspirin) may increase the risk of potentially fatal heart attacks, stroke, and related conditions. This risk may increase with duration of use and in patients who have underlying risk factors for heart and blood vessel disease. NSAIDs should not be used for the treatment of pain resulting from coronary artery bypass graft (CABG) surgery.

What is Diclofenac??





2-(2-(2,6-dichlorophenylamino)phenyl) acid

Chemical Data	
Formula	$C_{14}H_{11}Cl_2NO_2$
Mol. Mass	296.148 g/mol

Pharmacokinetic data	
Bioavailability	100%
Protein Binding	More than 99%
Metabolism	hepatic, no active metabolites exist

Half Life	1.2-2 hr
Excretion	biliary, only 1% in urine

Diclofenac (marketed as Voltaren and under a number of other trade names) is a non-steroidal anti-inflammatory drug (NSAID) taken to reduce inflammation and as an analgesic reducing pain in conditions such as arthritis or acute injury. The name is derived from its chemical name: 2-(2,6-dichloranilino)phenylacetic acid.

In the United Kingdom, India, and the United States, it may be supplied as either the sodium or potassium salt, in China most often as the sodium salt, while in some other countries only as the potassium salt. Diclofenac is available as a generic drug in a number of formulations. Over the counter (OTC) use is approved in some countries for minor aches and pains and fever associated with common infections.

***Mechanism Of Action:**

The exact mechanism of action is not entirely known, but it is thought that the primary mechanism responsible for its anti-inflammatory, antipyretic, and analgesic action is inhibition of prostaglandin synthesis by inhibition of cyclooxygenase (COX) and it appears to inhibit DNA synthesis.

Inhibition of COX also decreases prostaglandins in the epithelium of the stomach, making it more sensitive to corrosion by gastric acid. This is also the main side effect of diclofenac. Diclofenac has a low to moderate preference to block the COX2-isoenzyme (approximately 10-fold) and is said to have therefore a somewhat lower incidence of gastrointestinal complaints than noted with indomethacin and aspirin.

The action of one single dose is much longer (6 to 8 hours) than the very short half-life that the drug indicates. This could partly be due to a particular high concentration achieved in synovial fluids.

***Uses:**

Diclofenac is used for musculoskeletal complaints, especially arthritis, rheumatoid arthritis, Polymyositis, Dermatomyositis, osteoarthritis,dental pain, TMJ, spondylarthritis, ankylosing spondylitis, gout attacks, and pain management in cases of kidney stones and gallstones. An additional indication is the treatment of acute migraines. Diclofenac is used commonly to treat mild to moderate post-operative or post-traumatic pain, particularly when inflammation is also present, and is effective against menstrual pain and endometriosis.

As long-term use of diclofenac and similar NSAIDs predisposes for peptic ulcer, many patients at risk for this complication are prescribed a combination (Arthrotec) of diclofenac and misoprostol, a synthetic prostaglandin analogue, to protect the gastric mucosa.

An external, gel-based formulation containing 3% of diclofenac (Solaraze) is available for the treatment of facial actinic keratosis which is caused by over-exposure to sunlight. Some countries have also approved the external use of diclofenac 1% gel to treat musculoskeletal conditions.

***CLINICAL PHARMACOLOGY:**

Pharmacodynamics

Voltaren (diclofenac sodium enteric-coated tablets) is a nonsteroidal anti-inflammatory drug (NSAID) that exhibits anti-inflammatory, analgesic, and antipyretic activities in animal models. The mechanism of action of Voltaren, like that of other NSAIDs, is not completely understood but may be related to prostaglandin synthetase inhibition.

Pharmacokinetics

Absorption

Diclofenac is 100% absorbed after oral administration compared to IV administration as measured by urine recovery. However, due to first-pass metabolism, only about 50% of the absorbed dose is systemically available. Food has no significant effect on the extent of diclofenac absorption. However, there is usually a delay in the onset of absorption of 1 to 4.5 hours and a reduction in peak plasma levels of <20%.

Pharmacokinetic Parameters for Diclofenac:

PK Parameter	Normal Healthy Adults (20-48 yrs.)	
	Mean	Coefficient of Mean Variation (%)
Absolute Bioavailability (%) [N = 7]	55	40
T_{max} (hr) [N = 56]	2.3	69
Oral Clearance (CL/F; mL/min) [N = 56]	582	23
Renal Clearance (% unchanged drug in urine) [N = 7]	<1	—
Apparent Volume of Distribution (V/F; L/kg) [N = 56]	1.4	58
Terminal Half-life (hr) [N = 56]	2.3	48

Distribution

The apparent volume of distribution (V/F) of diclofenac sodium is 1.4 L/kg. Diclofenac is more than 99% bound to human serum proteins, primarily to albumin. Serum protein binding is constant over the concentration range (0.15-105 µg/mL) achieved with recommended doses.

Diclofenac diffuses into and out of the synovial fluid. Diffusion into the joint occurs when plasma levels are higher than those in the synovial fluid, after which the process reverses and synovial fluid levels are higher than plasma levels. It is not known whether diffusion into the joint plays a role in the effectiveness of diclofenac.

Metabolism
Five diclofenac metabolites have been identified in human plasma and urine. The metabolites include 4'-hydroxy-, 5-hydroxy-, 3'-hydroxy-, 4',5-dihydroxy- and 3'-hydroxy-4'-methoxy diclofenac. In patients with renal dysfunction, peak concentrations of metabolites 4'-hydroxy- and 5-hydroxy-diclofenac were approximately 50% and 4% of the parent compound after single oral dosing compared to 27% and 1% in normal healthy subjects. However, diclofenac metabolites undergo further glucuronidation and sulfation
followed by biliary excretion.
One diclofenac metabolite 4'-hydroxy- diclofenac has very weak pharmacologic activity.

Excretion
Diclofenac is eliminated through metabolism and subsequent urinary and biliary excretion of the glucuronide and the sulfate conjugates of the metabolites. Little or no free unchanged diclofenac is excreted in the urine. Approximately 65% of the dose is excreted in the urine and approximately 35% in the bile as conjugates of unchanged diclofenac plus metabolites. Because renal elimination is not a significant pathway of elimination for unchanged diclofenac, dosing adjustment in patients with mild to moderate renal dysfunction is not necessary. The terminal half-life of unchanged diclofenac is approximately 2 hours.

Special Populations
Pediatric: The pharmacokinetics of Voltaren has not been investigated in pediatric patients.
Race: Pharmacokinetic differences due to race have not been identified.
Hepatic Insufficiency: Hepatic metabolism accounts for almost 100% of Voltaren elimination, so patients with hepatic disease may require reduced doses of Voltaren compared to patients with normal hepatic function.
Renal Insufficiency: Diclofenac pharmacokinetics has been investigated in subjects with renal insufficiency. No differences in the pharmacokinetics of diclofenac have been detected in studies of patients with renal impairment. In patients with renal impairment (inulin clearance 60-90, 30-60, and <30 mL/min; N=6 in each group), AUC values and elimination rate were comparable to those in healthy subjects.

***Side Effects:**

- Diclofenac is among the better tolerated NSAIDs. Though 20% of patients on long-term treatment experience side effects, only 2% have to discontinue the drug, mostly due to gastrointestinal complaints.

Cardiovascular Effects

Cardiovascular Thrombotic Events

Clinical trials of several COX-2 selective and nonselective NSAIDs of up to three years duration have shown an increased risk of serious cardiovascular (CV) thrombotic events, myocardial infarction, and stroke, which can be fatal. All NSAIDs, both COX-2 selective and nonselective, may have a similar risk. Patients with known CV disease or risk factors for CV disease may be at greater risk. To minimize the potential risk for an adverse CV event in patients treated with an NSAID, the lowest effective dose should be used for the shortest duration possible. Physicians and patients should remain alert for the
development of such events, even in the absence of previous CV symptoms. Patients should be informed about the signs and/or symptoms of serious CV events and the steps to take if they occur.

There is no consistent evidence that concurrent use of aspirin mitigates the increased risk of serious CV thrombotic events associated with NSAID use. The concurrent use of aspirin and an NSAID does increase the risk of serious GI events.

Two large, controlled, clinical trials of a COX-2 selective NSAID for the treatment of pain in the first 10-14 days following CABG surgery found an increased incidence of myocardial infarction and stroke.

Hypertension

NSAIDs can lead to onset of new hypertension or worsening of preexisting hypertension, either of which may contribute to the increased incidence of CV events. Patients taking thiazides or loop diuretics may have impaired response to these therapies when taking
NSAIDs. NSAIDs, including Voltaren (diclofenac sodium enteric-coated tablets), should be used with caution in patients with hypertension. Blood pressure (BP) should be monitored closely during the initiation of NSAID treatment and throughout the course of therapy.

Congestive Heart Failure and Edema

Fluid retention and edema have been observed in some patients taking NSAIDs. Voltaren should be used with caution in patients with fluid retention or heart failure.

Gastrointestinal (GI) Effects: Risk of GI Ulceration, Bleeding, and Perforation

NSAIDs, including Voltaren, can cause serious gastrointestinal (GI) adverse events including inflammation, bleeding, ulceration, and perforation of the stomach, small intestine, or large intestine, which can be fatal. These serious adverse events can occur at any time, with or without warning symptoms, in patients treated with NSAIDs. Only one in five patients, who develop a serious upper GI adverse event on NSAID therapy, is symptomatic. Upper GI ulcers, gross bleeding, or perforation caused by NSAIDs occur in approximately 1% of patients treated for 3-6 months, and in about 2%-4% of patients
treated for one year. These trends continue with longer duration of use, increasing the likelihood of developing a serious GI event at some time during the course of therapy. However, even short-term therapy is not without risk.

NSAIDs should be prescribed with extreme caution in those with a prior history of ulcer disease or gastrointestinal bleeding. Patients with a *prior history of peptic ulcer disease and/or gastrointestinal bleeding* who use NSAIDs have a greater than 10-fold increased risk for developing a GI bleed compared to patients with neither of these risk factors.

Other factors that increase the risk for GI bleeding in patients treated with NSAIDs include concomitant use of oral corticosteroids or anticoagulants, longer duration of NSAID therapy, smoking, use of alcohol, older age, and poor general health status. Most spontaneous reports of fatal GI events are in elderly or debilitated patients and therefore, special care should be taken in treating this population.

To minimize the potential risk for an adverse GI event in patients treated with an NSAID, the lowest effective dose should be used for the shortest possible duration. Patients and physicians should remain alert for signs and symptoms of GI ulceration and bleeding during NSAID therapy and promptly initiate additional evaluation and treatment if a serious GI adverse event is suspected. This should include discontinuation of the NSAID until a serious GI adverse event is ruled out. For high risk patients, alternate therapies
that do not involve NSAIDs should be considered.

Renal Effects

Caution should be used when initiating treatment with Voltaren in patients with

considerable dehydration. Long-term administration of NSAIDs has resulted in renal papillary necrosis and other renal injury. Renal toxicity has also been seen in patients in whom renal prostaglandins have a compensatory role in the maintenance of renal perfusion. In these patients, administration of a nonsteroidal anti-inflammatory drug may cause a dose-dependent
reduction in prostaglandin formation and, secondarily, in renal blood flow, which may precipitate overt renal decompensation. Patients at greatest risk of this reaction are those with impaired renal function, heart failure, liver dysfunction, those taking diuretics and ACE inhibitors, and the elderly. Discontinuation of NSAID therapy is usually followed by recovery to the pretreatment state.

Advanced Renal Disease
No information is available from controlled clinical studies regarding the use of Voltaren in patients with advanced renal disease. Therefore, treatment with Voltaren is not recommended in these patients with advanced renal disease. If Voltaren therapy must be initiated, close monitoring of the patient's renal function is advisable.

Hepatic Effects
Elevations of one or more liver tests may occur during therapy with Voltaren. These laboratory abnormalities may progress, may remain unchanged, or may be transient with continued therapy. Borderline elevations (i.e., less than 3 times the ULN [ULN = the upper limit of the normal range]) or greater elevations of transaminases occurred in about 15% of diclofenac-treated patients. Of the markers of hepatic function, ALT (SGPT) is recommended for the monitoring of liver injury.
In clinical trials, meaningful elevations (i.e., more than 3 times the ULN) of AST (GOT) (ALT was not measured in all studies) occurred in about 2% of approximately 5,700 patients at some time during diclofenac treatment. In a large, open-label, controlled trial of 3,700 patients treated for 2-6 months, patients were monitored first at 8 weeks and 1,200 patients were monitored again at 24 weeks. Meaningful elevations of ALT and/or AST occurred in about 4% of patients and included marked elevations (i.e., more than 8
times the ULN) in about 1% of the 3,700 patients. In that open-label study, a higher incidence of borderline (less than 3 times the ULN), moderate (3-8 times the ULN), and marked (>8 times the ULN) elevations of ALT or AST was observed in patients receiving diclofenac when compared to other NSAIDs. Elevations in transaminases were seen more frequently in patients with osteoarthritis than in those with rheumatoid arthritis.
Almost all meaningful elevations in transaminases were detected before patients became symptomatic. Abnormal tests occurred during the first 2 months of therapy with diclofenac in 42 of the 51 patients in all trials who developed marked transaminase elevations.

In postmarketing reports, cases of drug-induced hepatotoxicity have been reported in the first month, and in some cases, the first 2 months of therapy, but can occur at any time
during treatment with diclofenac. Postmarketing surveillance has reported cases of severe hepatic reactions, including liver necrosis, jaundice, fulminant hepatitis with and without jaundice, and liver failure. Some of these reported cases resulted in fatalities or liver transplantation.
Physicians should measure transaminases periodically in patients receiving long-term therapy with diclofenac, because severe hepatotoxicity may develop without a prodrome of distinguishing symptoms. The optimum times for making the first and subsequent transaminase measurements are not known. Based on clinical trial data and postmarketing experiences, transaminases should be monitored within 4 to 8 weeks after initiating treatment with diclofenac. However, severe hepatic reactions can occur at any time during treatment with diclofenac.
If abnormal liver tests persist or worsen, if clinical signs and/or symptoms consistent with liver disease develop, or if systemic manifestations occur (e.g., eosinophilia, rash, abdominal pain, diarrhea, dark urine, etc.), Voltaren should be discontinued immediately.
To minimize the possibility that hepatic injury will become severe between transaminase measurements, physicians should inform patients of the warning signs and symptoms of hepatotoxicity (e.g., nausea, fatigue, lethargy, diarrhea, pruritus, jaundice, right upper quadrant tenderness, and "flu-like" symptoms), and the appropriate action patients should take if these signs and symptoms appear.
To minimize the potential risk for an adverse liver related event in patients treated with Voltaren, the lowest effective dose should be used for the shortest duration possible. Caution should be exercised in prescribing Voltaren with concomitant drugs that are known to be potentially hepatotoxic (e.g., antibiotics, anti-epileptics).

Anaphylactoid Reactions
As with other NSAIDs, anaphylactoid reactions may occur in patients without known prior exposure to Voltaren. Voltaren should not be given to patients with the aspirin triad. This symptom complex typically occurs in asthmatic patients who experience rhinitis with or without nasal polyps, or who exhibit severe, potentially fatal bronchospasm after taking aspirin or other NSAIDs. (See CONTRAINDICATIONS and PRECAUTIONS, Preexisting Asthma.) Emergency help should be sought in cases where an anaphylactoid reaction occurs.

Skin Reactions
NSAIDs, including Voltaren, can cause serious skin adverse events such as exfoliative dermatitis, Stevens-Johnson Syndrome (SJS), and toxic epidermal necrolysis (TEN), which can be fatal. These serious events may occur without warning. Patients should be

informed about the signs and symptoms of serious skin manifestations and use of the drug should be discontinued at the first appearance of skin rash or any other sign of hypersensitivity.

Pregnancy
In late pregnancy, as with other NSAIDs, Voltaren should be avoided because it may cause premature closure of the ductus arteriosus.

***DOSAGE AND ADMINISTRATION (Humans):**

For the relief of osteoarthritis, the recommended dosage is 100-150 mg/day in divided doses (50 mg b.i.d. or t.i.d., or 75 mg b.i.d.).

For the relief of rheumatoid arthritis, the recommended dosage is 150-200 mg/day in divided doses (50 mg t.i.d. or q.i.d., or 75 mg b.i.d.).

For the relief of ankylosing spondylitis, the recommended dosage is 100-125 mg/day, administered as 25 mg q.i.d., with an extra 25-mg dose at bedtime if necessary.

Different formulations of diclofenac are not necessarily bioequivalent even if the milligram strength is the same.

*Trade Names:

- Anuva
- Abitren
- Arthrotec
- Betaren
- Cataflam
- Clofast
- Dedolor
- Deflamat
- Deflox
- Diclac
- Diclofenacum
- Dicloflex
- Diclogem
- Diclohexal
- Diclomax
- Diclon
- Motifene
- Naklofen
- Olfen
- Panamor
- Pennsaid
- Rhumalgan
- Sandoz
- Solaraze
- Topac
- Uno
- Vetagesic
- Voltaren
- Voltarol
- Voltfast
- Votral (Pakistan)
- Votrex

- Diclopar (Tanzania)
- Difen
- Difene
- Dyloject
- Feloran
- Flector patch
- Modifenac
- Morbidic
- Vostar
- Votalin (China)
- Voveran
- Zipsor
- Zolterol

Effect Of Diclofenac On Vultures:

Toxic side effects of Diclofenac have been demonstrated in various animals. While minor upper gastrointestinal problems such as dyspepsia are common during Diclofenac administration, severe pathologic conditions such as peptic ulceration, gastrointestinal bleeding, hepatotoxicity (nausea, fatigue, lethargy, pruritus (intense itching sensation), jaundice, 'flu-like' symptoms, etc.), renal papillary necrosis, and renal failure are reported in patients subjected to long-term administration of the drug. Cardio-protective effect is a positive side effect revealed recently. Hepato-toxicity and even acute immune haemolytic anaemia, renal dysfunction and acute renal failure are reported in mammals. However, in humans it is generally considered that the benefits are sufficient enough to offset the potential risks.

Gout is a disorder of metabolism that allows uric acid to accumulate in the blood and tissues. Uric acid is the end-product of purine (nucleic acid component of DNA) metabolism and is produced normally by the body during tissue remodelling and breakdown. Gout in birds is caused by renal failure and is analogous to hyperuricemia (accumulation of urea) in humans. When kidneys fail to remove the uric acid efficiently from the blood, tissues become supersaturated with uric acid, resulting in urate salt precipitation as crystals. A cellular reaction to uric-acid crystal deposition causes gout. These crystals are less soluble under acid conditions and any condition predisposing to acidosis also precipitates urate crystals. These crystals stimulate phagocytosis by neutrophils and initiate the inflammatory cascade. Interleukin-1 and tumour necrosis factor-*a* are known to be involved in the inflammatory cascade.

Visceral gout had been the prominent abnormality reported in most of the post-mortem reports of vultures. Hence it is normally assumed that whatever the responsible factor might be, it is in some way predisposing the vultures to a gouty condition. Gout has been reported from various animal taxa such as snakes, lizards, crocodiles, birds and mammals. Presence of red meat in the diet is considered to be a major contributing factor for susceptibility to gout. Interestingly, among the dinosaurs, evidence for gout has been found in the fossils of the carnivorous Tyrannosaurus, a red meat-eater. This, along with high levels of uric acid in raptors, might have contributed to predisposing vultures to increased risk of gout formation compared to other birds. No treatment for visceral gout is immediately known. Hence it is felt essential that immediate attention be given to explore the treatment of visceral gout in birds and to identify the possible causative factors that are in operation. It appears that along with Diclofenac, a host of other factors might also

have contributed to the development of gout in vultures that ultimately led to OWBV mortality in such great magnitude.

Causes of gout can be divided into two major categories: increased synthesis of uric acid and decreased clearance of uric acid by the kidneys. High intake of purine-rich foods such as red meat and poultry (especially the offal foods like liver, kidney, heart, tripe, etc.) is an important causative factor that enhances uric acid synthesis in the body. Apart from the Diclofenac residues, which is only a latest addition on the list, many other factors are already known to cause the physiological condition of gout. Major among them are renal impairment, dietary excess of purines, fasting-induced ketosis, inborn errors of metabolism, lead poisoning hyperproliferative skin disorders (e.g.- psoriasis), haemolytic and pernicious anaemia, radiation exposure, gut sterilization by antibiotics and various medications, such as caffeine, corticosteroids, cytotoxic drugs dopa-mine, nicotinic acid, salicylates, low dose sulfinpyrazone and vitamins B-12 and C.

Another factor that was probably overlooked in the study of vultures from Pakistan is the effect of Diclofenac metabolites. Although *in vitro* studies in rat hepatocytes have shown that the reactive Diclofenac metabolite such as acyl glucuronide is less cytotoxic than the drug itself in mammals, it need not necessarily be true in vultures. Hence the toxicity of mammalian metabolites of Diclofenac and other NSAIDs also needs to be screened.

Apart from the above-mentioned possible contributory factors, deteriorating health, affecting the general immunity of the birds, might have also assisted in ushering them into their gouty graves. Vultures had wide home ranges of more than 100 km radius, and before finding the waste dumps of urban areas as convenient 'fast food restaurants', the OWBVs had to forage over vast areas of land for finding carcasses. It is thus quite likely that the vultures too have become far more sedentary in their habits compared to earlier times. Mass congregations in places such as solid-waste dump yards have exposed them to various noxious chemicals and pollutants, which would have taken a heavy toll on their health and immunity levels and rendered them increasingly susceptible to environmental stress and contagious diseases. The solid wastes in the country are not segregated and contain a variety of insidious and recalcitrant polluting chemicals. It may be noted that lesions reported from the carcasses of Indian OWBVs have indicated the role of an undiagnosed disease factor as well. Such ecological factors along with other inconclusive hypotheses would have added on to the overall population decline.

The lacunae existing in the ecological and behavioural information on our common birds is a major lesson to be learned during the recent episode of vulture population decline. For instance, the normal behaviour of 'head drooping' during the hot summer days was often mistakenly identified as a disease symptom. (It is actually a natural behaviour of these birds possibly to avoid direct sunlight into their eyes). Unfortunately, the

situation is not different with most other urban birds such as house crows and sparrows. Serious ecological studies and long-term population monitoring programmes need to be targeted on the urban bird populations. It would serve as an early warning system to facilitate effective precautionary, mitigatory and remedial measures to be taken up well in advance, before the populations cross critical limits as happened with *Gyps* vultures.

There are multiple factors, which possibly have played a role in the drastic population decline of the Oriental white-backed vultures of southern Asia. Prudent management actions duly considering all such potential factors only can save them from the risk of extinction. In the absence of a detailed diagnostic investigation and management strategy specific to the Indian conditions, activities such as captive breeding and release may not yield the expected results. Although the role of Diclofenac residues in the OWBV mortality from the Indian region is yet to be confirmed experimentally, a substitution of Diclofenac with other NSAID compounds may be advocated as an immediate (and desperate) measure to put breaks on the decline of the vulture population. However, care should be taken while replacing Diclofenac with other NSAIDs, because the replacement drugs may not be environmentally safe either, and their impacts are unknown. Research into the ecological impact of all major therapeutics, especially the common and widely used ones, should be undertaken urgently to facilitate the policy makers to regulate these compounds with potentially hazardous environmental impacts. The exact physiological mechanism through which Diclofenac acts in three species of only a single genus (*Gyps*), appears mysterious and highly intriguing. The metabolic pathways through which Diclofenac poisoning causes renal complications, gout and consequent mortality in only *Gyps* vultures is yet to be elucidated. As of now, it appears premature to conclude that the Diclofenac residue is the universal causative agent behind the decline of vulture population.

White deposits on internal body organs of dead vultures are indictive of visceral gout and renal failure.

Why Livestock were treated with Diclofenac???

Prostaglandins are a family of chemicals that are produced by the cells of the body and have several important functions.

They promote inflammation, pain, and fever; support the blood clotting function of platelets; and protect the lining of the stomach from the damaging effects of acid.

Prostaglandins are produced within the body's cells by the enzyme cyclooxygenase (COX). There are two COX enzymes, COX-1 and COX-2. Both enzymes produce prostaglandins that promote inflammation, pain, and fever. However, only COX-1 produces prostaglandins that support platelets and protect the stomach. **Non-steroidal anti-inflammatory drugs (NSAIDs) block the COX enzymes and reduce prostaglandins throughout the body.**

As a consequence, ongoing inflammation, pain, and fever are reduced. Since the prostaglandins that protect the stomach and support platelets and blood clotting also are reduced, NSAIDs can cause ulcers in the stomach and promote bleeding.

Diclofenac was not covered by patent at that time and more than 50 companies in India manufactured veterinary formulations. Across the subcontinent, it had been the welfare drug of choice for veterinarians treating livestock for a range of conditions. It is generally administered as an intramuscular injection, although an ingestible bolus form also exists. It is likely to be useful in a range of situations, including in rural communities, where families frequently keep water buffalo and cattle for working the land and for milking.
As a potent anti-inflammatory drug, diclofenac helped to temporarily alleviate the effects of a range of veterinary problems (e.g. muscle inflammation in the limbs, and mastitis) . This was the main reason for it being so popular among the poor as they needed an **effective,cheap and safe** measure to treat their cattle.

How Diclofenac content was detected in Vultures?

GC/MS

Gas chromatography ("GC") and mass spectrometry ("MS") make an effective combination for chemical analysis. The GC-MS has been widely heralded as a gold standard for forensic substance identification because it is used to perform a *specific test*. A specific test positively identifies the actual presence of a particular substance in a given sample. GC analysis is a common confirmation test. Among its uses are drug testing and environmental contaminant identification. GC analysis separates all of the components in a sample and provides a representative spectral output. A mixture of chemicals present in a specimen can be separated in the GC column. Some chemical and physical characteristics of the molecules cause them to travel through the column at different speeds. If the molecule has low mass it may travel more swiftly. Also, the molecule's shape may affect the time needed to exit the column. How the different substances relate to each other may cause the time needed to travel the column to increase or decrease. Interactions between the sample's molecule and the column surface may cause the molecule to be retained inside the column for a different amount of time than similar molecules that interact with the column differently.

PROCEDURE TO PERFORM GC/MS:

The technician injects the sample into the injection port of the GC device. The GC instrument vaporizes the sample and then separates and analyzes the various components. Each component ideally produces a specific spectral peak that may be recorded on a paper chart or electronically. The time elapsed between injection and elution is called the "retention time." The retention time can help to differentiate between some compounds. The size of the peaks is proportional to the quantity of the corresponding substances in the specimen analyzed. The peak is measured from the baseline to the tip of the peak.

The Peregrine Fund:

Aim:

The Asian Vulture Population Project is a community effort hosted by The Peregrine Fund to record and monitor changing vulture populations over time using the internet to gather and disseminate up-to-date, verified data provided by volunteers across Asia.

How it Works:

The Asian Vulture Population Project provides up-to-date locations and summaries of the known breeding populations of three Asian vulture species. It is a factual data resource for the use of all, compiled and verified from information received from observers across the range of these species.

Information is represented geographically, with a range-wide map indicating the positions of breeding colonies reported so far.

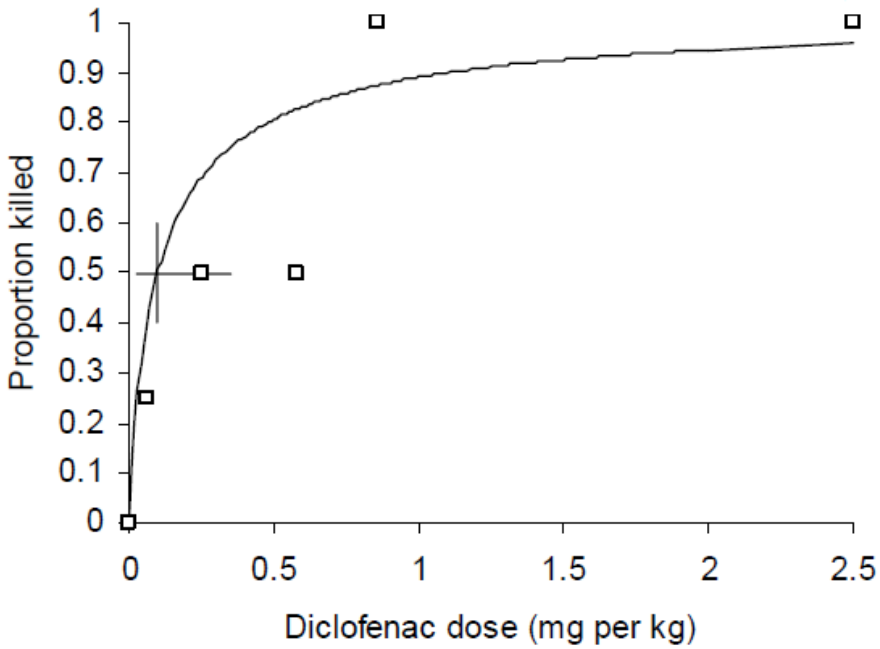
RESULTS THAT CAME FROM THE *TESTS* & EXPERIMENTS UNDERTAKEN BY THE PEREGRINE FUND AND THEIR CO-WORKERS!!!

Data on the toxicity of diclofenac to *Gyps bengalensis* were obtained from experiments
undertaken by the Peregrine Fund and their co-workers (text and Table 2 of Oaks *et al.* 2004). Vultures were fed over a period of a few days on tissues from goats or buffaloes that had been treated with a standard veterinary course of single daily doses of diclofenac, the last being administered a few hours before slaughter. Diclofenac concentrations were determined from samples of ungulate tissues, and the dosage (mg kg-1 vulture body weight) was estimated from this concentration and the weight of tissue that each vulture ingested. There were four experimental groups fed in this way and one group of controls (Oaks *et al.* Table 2). In addition, data were also included on two vultures given oral doses of 2.5 mg kg-1 vulture body weight and two vultures given oral doses of 0.25 mg kg-1 vulture body weight. The estimate of m obtained from birds given tissue from treated livestock was reasonably similar to the estimate from the birds receiving the oral dose (m = -1.056 cf. m = -0.6021 respectively: back transformed LD50 0.088 mg kg-1 cf. 0.250 mg kg-1 respectively). Observed and expected mortality rates were compared graphically (Figure A1) and by calculating expected mortality rates for each experimental group under the fitted model (Table A1). The data were too sparse for a meaningful goodness-of-fit test, but

the model does not appear to fit the grouped data particularly well. Inspection of individual doses revealed that this is primarily because of an outlier (Vulture 11 in Table 2 of Oaks *et al.* (2004)); a bird that received the lowest dose of diclofenac (0.007
mg kg-1), but died of gout. Although the death of this bird from such an apparently low dose and the high concentration of diclofenac in kidney tissue taken from it at post-mortem (Figure A3) might suggest that it actually ingested more diclofenac than was estimated, the low uric acid concentration in plasma taken 24 h after treatment is consistent with a low dose. Hence, it is not clear whether this low dose estimated for this individual is correct or not. For this reason, we have estimated LD50
with and without this datum.

Experimental group	Range of doses mg kg^{-1}	Mean dose	Number of birds	Number died	Percent died	Modelled percent died (1)	Modelled percent died (2)
Untreated control	0	0	6	0	0	0	0
Fed tissue of treated ungulate	0.005 to 0.3	0.066	8	2	25	33	11
Fed tissue of treated ungulate	0.5 to 0.6	0.575	2	1	50	83	86
Fed tissue of treated ungulate	0.8 to 1.0	0.863	10	10	100	87	94
Dosed orally	0.25	0.25	2	1	50	69	55
Dosed orally	2.5	2.5	2	2	100	100	100

Outcome of experiments on *Gyps bengalensis* in which diclofenac was administered to vultures orally or by feeding them on diclofenac-treated ungulate tissue. The modelled percentage mortality was calculated as the mean for each experimental group of the expected individual probabilities of death from the fitted
probit model. Two versions of this analysis are shown, with (1) and without (2) the
inclusion of an outlier



Fitted models of the median lethal dose from experiments on *Gyps bengalensis* in which diclofenac was administered to vultures orally or by feeding them on tissues from ungulates treated with the drug shortly before slaughter (Oaks *et al.* 2004). Squares represent the proportion of vultures that died in each experimental group in relation to the mean dose, in mg of diclofenac ingested per kg of vulture body weight, for individuals in that group. The curve shows the probability of death in relation to dose for the fitted probit model. The median lethal dose is indicated by the vertical line and its 95% confidence interval by the horizontal line. Fitted models are shown separately for data for all birds.

The group of J. Lindsay Oaks, Martin Gilbert, Munir Z. Virani, Richard T. Watson, Carol U. Meteyer, Bruce A. Rideout, H. L. Shivaprasad, Shakeel Ahmed, Muhammad Jamshed Iqbal Chaudhry, Muhammad Arshad, Shahid Mahmood, Ahmad Ali & Aleem Ahmed Khan performed different experiments and analyzed how vultures responded to different amounts of diclofenac content on intake. The results and conclusion what came out of this were published later in 'Nature'.

Procedure For Analysis

Kidney samples (0.5 g) were homogenized in 4ml of acetonitrile and centrifuged to pellet debris. Three millilitres of the supernatants were then passed through solid-phase extraction cartridges (Waters Sep-Pak Plus t-C18) at 1 ml min21. The cartridges were then washed with an additional 3ml of acetonitrile, the eluates pooled, and concentrated to final volumes of 1 ml. For samples that weighed less than 0.5 g, the protocol was adjusted proportionally for the smaller weights.

Diclofenac was detected and quantified by high-performance liquid chromatography and mass spectroscopy (Agilent 1100 series equipped with a Water's Xterra MS C18 (3.9mm £ 150 mm, 5 mm) and guard column (3.9mm £ 20 mm, 5 mm)). Diclofenac standard (Sigma, D6899) was dissolved into 1:1 (v/v) acetonitrile:water at 1,040 mgml21, diluted to a final concentration of 10.4 mgml21, and this stock was used to prepare calibration standards of 0.104, 0.052, 0.026, 0.0104 and 0.0052 mgml21 in acetonitrile, which generated a linear calibration curve with R2 values equal to 0.999. Samples and standards (20 ml) were subjected to a binary gradient elution profile, which consisted of 0.1% acetic acid in water (solution A) and 100% acetonitrile (solution B) as follows:

starting conditions 75% A/25% B for 0.1 min, a 15-min linear gradient from 75% A/25% B to 5% A/95% B, followed by a 5-min column-wash step in 5% A/95% B, and a 10-min re-equilibration step with 75% A/25% B before the next injection. Flow rate was 0.7 ml min21 and the column temperature was 40 8C. The mass spectrometer (Agilent 1946D) was equipped with an electrospray ionization inlet andmass spectra were acquired in the negative ion mode. The mass spectrometer was set to selectively monitor for mass ions 294 and 296 m/z of which mass 294 m/z, the deprotonated molecular mass, was used to quantify the diclofenac residues. The mean recovery from spiked avian kidney tissue was approximately 70%, and this value was used to calculate the final sample concentration of diclofenac.

Vulture no	Exposure (mg kg^{-1})	Result	Diclofenac residues (μg g^{-1})
11	0.007	Gout	0.38
12	0.025	Survived	ND
8	0.027	Survived	ND
10	0.028	Survived	ND
7	0.029	Survived	ND
9	0.029	Survived	ND
D	0.140	Gout	0.07
F	0.240	Survived	BDL (8 days)
H	0.550	Gout	0.25
B	0.600	Survived	BDL (8 days)
64	0.820	Gout	0.54
65	0.820	Gout	ND
62	0.840	Gout	0.22
63	0.840	Gout	0.12
70	0.860	Gout	0.74
67	0.860	Gout	ND
68	0.860	Gout	ND
66	0.880	Gout	0.26
73	0.910	Gout	0.91
72	0.940	Gout	ND
1	None (control)	Survived	ND
2	None (control)	Survived	ND
3	None (control)	Survived	ND
4	None (control)	Survived	ND
cc1	None (control)	Survived	BDL (8 days)
cc2	None (control)	Survived	BDL (8 days)

Values in parentheses in the last column indicate time after exposure. OWBVs that died and displayed gross characteristics of visceral gout on necropsy are labelled as 'Gout' in the third column. BDL, below detection limit of diclofenac assay (0.005–0.01 mgg21); ND, analysis not done.

Another group of scientists namely Richard Cuthbert, Jemima Parry-Jones, Rhys E. Green and Deborah J. Pain had also analysed the effect of diclofenac content to the deaths of vultures and came up with the article named ***NSAIDs and scavenging birds: potential impacts beyond Asia's critically endangered vultures.*** *In this article they had mentioned the effect of diclofenac on vultures among many other things.*

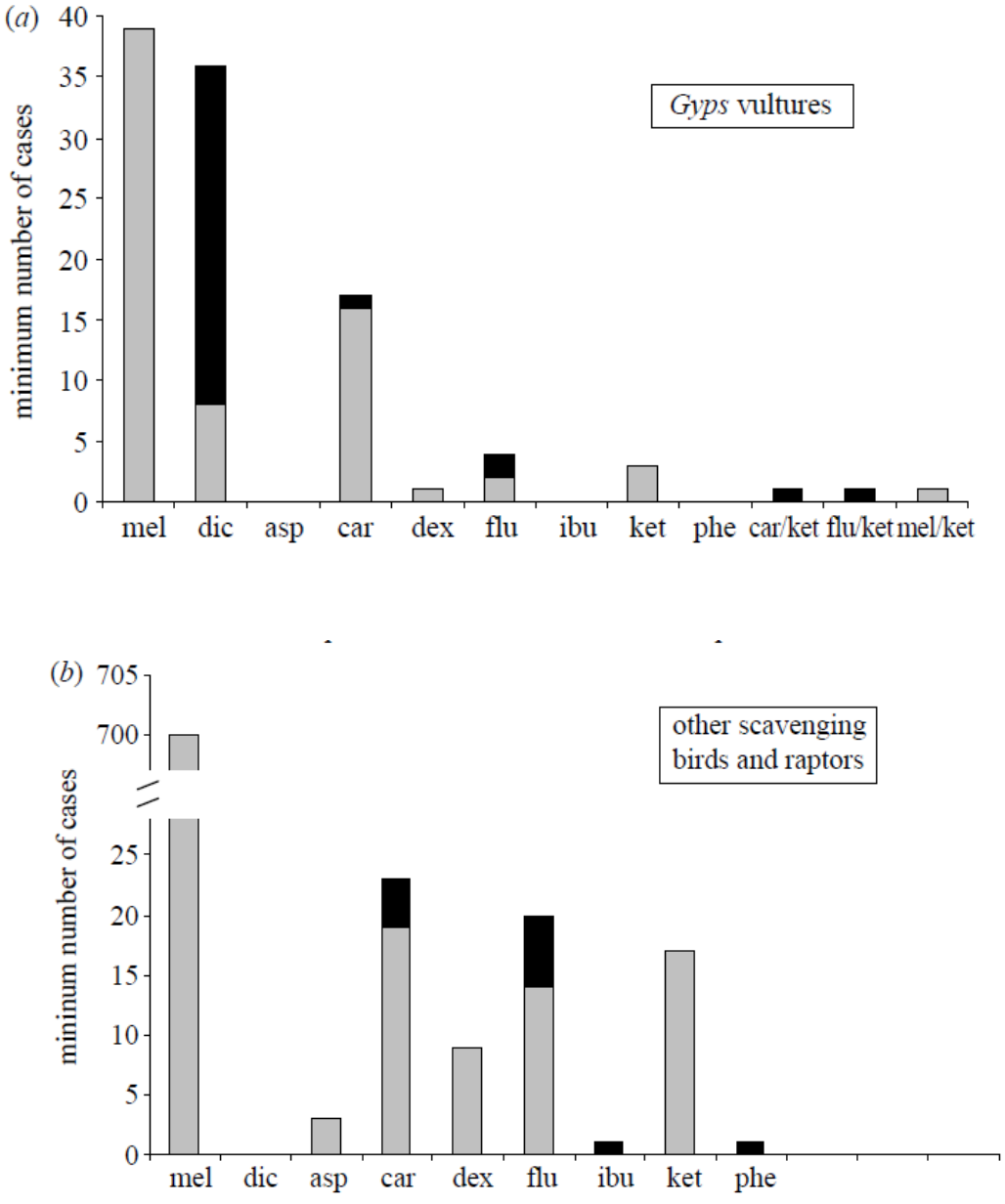
Method of Working

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 (nZ4) were recorded as separate cases.

The final results and findings:

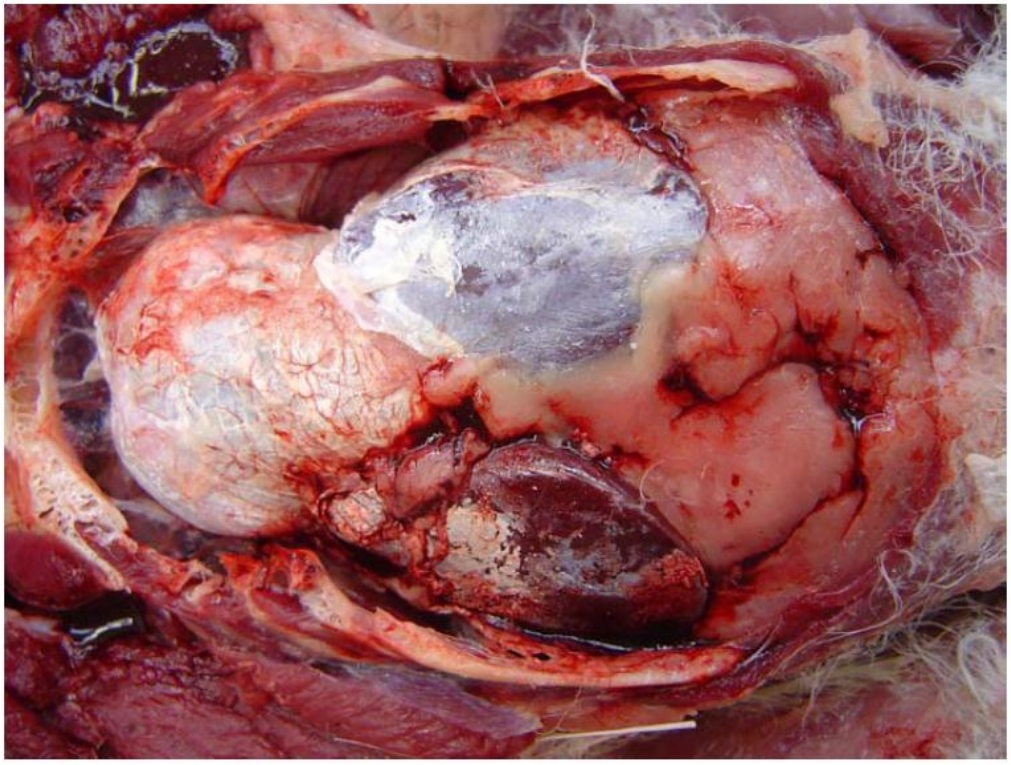
A total of 31 veterinarians and institutions responded, providing information on over 870 cases of NSAID
treatment for 79 species of birds including Gyps vultures, other raptors, storks, cranes, owls and crows. While owls and cranes are not scavenging birds, the survey provided comprehensive information for owls and one reported an instance of mortality for a crane: consequently the results are presented. Information was also provided on dexamethasone, asteroidal 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. 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 articularly high dose of carprofen (1–3, 4 and 5 mg kgK1,cf. 1.5–7.6 mg kgK1 for all birds treated) or flunixin (1–4.5 mg kgK1, cf. 0.5–12 mg kgK1). Two instances of mortality with renal disease and gout are reported for ibuprofen and phenylbutazone. 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 kgK1 bw, with a median dose of 0.5 mg kgK1. 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 antiinflammatory) and 20 instances where ketoprofen (when this drug was administered on its own) have been administered with no reported mortalities.



Number of cases of (a) Gyps vultures (nZ6 species) and (b) other scavenging birds (nZ54 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. (2006a).

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



Abdominal cavity of vulture AW4 (*G. africanus*) at necropsy showing visceral gout on the liver surface following experimental dosing with diclofenac

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Number of Active Nests of Gyps tenuirostris

Location	1985/86	1987/88	1991/92	1992/93	1993/94	1994/95	1995/96	1996/97	1997/98	1998/99	1999/00	2000/01	2001/02	2002/03	2003/04	2004/05	2005/06	2006/07	2007/08	2008/09	Source	Last Known Update	Notes	Map
India																								
Bandhavgargh NP																			x		TPF	Feb 07	Notes	Map
Bandhavgarh NP (New Sites)																			x		TPF	Feb 07	Notes	Map
Chambal Ravine																	x	x			TPF / R. Singh / H. Vardhan	Feb 07	Notes	Map
Ghilabari															2						Rishad Naoroji	Feb 04	Notes	Map
Katernaighat Wildlife Sanct.												13 to 15									Suchitra Ghosh	October 2000	Notes	Map
Kaziranga															1						Rishad Naoroji	Feb 04	Notes	Map
Ranthambhor NP																			x		TPF	Feb 07	Notes	Map
Sadiya															1						Rishad Naoroji	Feb 04	Notes	Map
Nepal																								
Koshi Tappu												2	0	0	0	0					TPF/BCN	Feb 07	Notes	Map
Nawalparasi												1									BCN		Notes	Map
Royal Bardia Wildlife Reserve													2								BCN/OBC	May 02	Notes	Map
Royal Suklaphanta WR													7	0							BCN/OBC		Notes	Map

Facts and figures:

Vulture species observed in India

S. No.	Cmmon Name	Scientific name	Size	Sighting	Status
1.	Long-billed vulture	*Gyps indicus*	80-95 cm	Rare	Resident, breeding
2.	Slende billed vulture	*Gyps tenirostris*	80-90 cm	Rare	Resident, breeding
3.	White-backed vulture	*Gyps bengalensis*	75-85 cm	Rare	Resident, breeding
4.	King vulture	*Sarcogyps calvus*	80-85cm	Common	Resident, breeding
5.	Egyptian vulture	*Neophron percnopterus*	60-70 cm	Very common	Resident, breeding
6.	Himalayan griffon	*Gyps hymalayensis*	115-125 cm	Common in winters	Resident, breeding & Migrant
7.	Eurasian griffon	*Gyps fulvus*	95-105 cm	Very common in winter	Resident, breeding & Migrant
8.	Cinereous vulture	*Aegypius monachus*	100-110 cm	Rare in winters	Resident, breeding & Migrant
9.	Lanmergeier vulture	*Gypaetus barbatus*	75-85 cm	Rare, found only in Himalayas	Resident, breeding

Gyps bengalensis

Gyps indicus

Gyps tenirostris

Amongst the 9 species of vultures found in India, 3 of them are included in IUCN (International Union for the Conservation of Nature and Natural Resources) Red List of Threatened Species.

1.Gyps bengalensis

Taxonomy

Kingdom	Phylum	Class	Order	Family
ANIMALIA	CHORDATA	AVES	Falconiformes	Accipitridae

Scientific Name: Gyps bengalensis

Species Authority: (Gmelin, 1788)

Common Name/s:

English - White-rumped Vulture, Asian White-backed Vulture, Oriental White-backed Vulture, White-backed Vulture

Spanish - Buitre Dorsiblanco Bengalí, Buitre Leonado Bengalés

Assessment Information

Red List Category & Criteria: Critically Endangered A2bce+4bce ver 3.1

Year Assessed: 2009

Assessor/s BirdLife International

Evaluator/s: Bird, J., Butchart, S., Symes, A.(BirdLife International)

Justification:

This species qualifies as Critically Endangered because it has suffered an extremely rapid population decline primarily as a result of feeding on carcasses of animals treated with the veterinary drug diclofenac.

	2008 – Critically Endangered
	2004 – Critically Endangered
History:	2000 – Critically Endangered
	1994 – Lower Risk/near threatened
	1988 – Lower Risk/least concern

Geographic Range

Range Description:

Gyps bengalensis occurs in **Pakistan**, **India**, **Bangladesh**, **Nepal**, **Bhutan**, **Myanmar**, **Thailand**, **Laos**, **Cambodia** and southern **Vietnam,** and may be extinct in southern China and Malaysia. It has been recorded from south-east Afganistan and Iran where its status is currently unknown. As recently as 1985 the species was described as "possibly the most abundant large bird of prey in the world". However, it disappeared from most of South-East Asia in the early 20th century and the only viable populations in the region are found in Cambodia and Myanmar (both probably in the low hundreds of individuals). Given the lack of intensive agriculture and associated chemical use in South-East Asia and the continued presence of large areas of suitable habitat for the species, the primary reason behind its decline in the region is thought to be the demise of large ungulate populations and improvements in animal husbandry resulting in a lack of available carcasses for vultures. Since the mid-1990s, it has suffered a catastrophic decline (over 99%) across the Indian Subcontinent (the majority of its historic range), first noticed in Keoladeo National Park, India, but mirrored in Pakistan and Nepal, to the point that the species is highly threatened with extinction. Extensive research has identified the non-steroidal anti-inflammatory drug (NSAID), diclofenac, to be the cause behind this rapid population collapse[9,10,11,12]. This drug, used to treat domestic livestock, is ingested by vultures feeding on their carcasses leading to renal failure causing visceral gout[10,12,14,15]. Declines in India between 2000-2007 averaged 43.9% per year[22], and ranged between 11%-61% in Punjab province, Pakistan over the same period[23], while surveys of 23 known colonies in Punjab province in 2006 found a total of only 37 breeding pairs[23]. Diclofenac is apparently entirely absent in Cambodia, adding greater emphasis to that remaining small population (171 counted at vulture restaurants in 2008)[28].

Countries:

Native:

Bhutan; Cambodia; India; Iran, Islamic Republic of; Lao People's Democratic Republic; Myanmar; Nepal; Pakistan; Thailand; Viet Nam

Regionally extinct:

Bangladesh; China; Malaysia

Vagrant:

Afghanistan; Brunei Darussalam; Russian Federation

Range Map:

Population

Population:

Formerly described as possibly the most abundant large bird of prey in the world, this species global population almost certainly numbered several million individuals. However, following dramatic declines through the 1990's across its range its global population is now estimated to fall within the band 2,500-9,999 individuals.

Population Trend: ↓ Decreasing

Habitat and Ecology

Habitat and Ecology: It occurs mostly in plains and less frequently in hilly regions where it utilises light woodland, villages, cities, and open areas. It feeds on carrion, both putrid and fresh. While feeding considerable aggregations can form, and regular communal roost sites are used. It is social and usually found in conspecific flocks. It breeds in colonies in tall trees, often near human habitation. Movements are poorly known, although satellite-tagged birds have shown that they will forage over a vast range. The degree of connectivity of apparently separate populations is not known. Vultures also play a key role in the wider landscape as providers of ecosystem services, and were previously heavily relied upon to help dispose of animal and human remains in India.

Systems: Terrestrial

Threats

Major Threat(s): By mid-2000, *Gyps* vultures were being found dead and dying in Nepal, Pakistan, and throughout India, and major declines and local extirpations were being reported. The anti-inflammatory veterinary drug diclofenac, used to treat domestic livestock, has been identified as the cause of mortality from renal failure resulting from visceral gout in the vast majority of examined vultures. Modelling has shown that to cause the observed rate of decline in *Gyps* vultures, just one in 760 livestock carcasses need contain diclofenac residues[9]. Other likely contributory factors are changes in human consumption and processing of dead livestock, and poison and pesticide use, but these are probably of minor significance. In South-East Asia, the near-total disappearance of the species pre-dated the present crisis, and probably resulted from the collapse of large wild ungulate populations and improved management of deceased livestock[7].

Conservation Actions

Conservation actions underway:

Conservation Actions: CITES Appendix II. CMS Appendix II. It has been reported from many protected areas across its range. There has been a proposal to add the species to the protected species list of the 1973 National Parks and Wildlife Conservation Act in Nepal. The Indian government has now passed a bill banning the manufacture of the veterinary drug diclofenac

that has caused the rapid population decline across the Indian Subcontinent; their aim was to phase out its use by late 2005, although its sale has not been banned and it is likely to remain in widespread use for several years. Similar laws banning import and manufacture of diclofenac are now in place in Nepal and Pakistan. Efforts to replace diclofenac with a suitable alternative are ongoing; drug companies have now developed meloxicam, an alternative to diclofenac which has been tested on a number of species including *Gyps* vultures with no apparent ill-effects[25]. In 2008 the Indian government ordered a crackdown on companies selling diclofenac. A letter from the Drug Controller General of India warned more than 70 drugs firms not to sell the veterinary form of diclofenac, and to mark human diclofenac containers 'not for veterinary use'[27]. A study of 11 administrative districts in Nepal found diclofenac use dropped by 90% since 2006 following the introduction of measures to reduce its use[24]. Vulture restaurants are increasingly used as ecotourism attractions in parts of the species's range, particularly Cambodia, to raise awareness and fund supplementary feeding programmes and research. The exchange of diclofenac with meloxican near breeding colonies is taking place in Nepal in combination with diversionary feeding with diclofenac-free carcasses. Diversionary feeding has been shown to reduce but not eliminate vulture mortality from diclofenac poisoning, and uncertainty over the movements of Asian *Gyps* vultures makes the effectiveness of measures such as these uncertain[25]. Birds have been satellite tagged in various parts of their range to improve understanding of their movements, foraging range, site fidelity etc., to aid development of suitable conservation strategies for the species[4]. Socioeconomic surveys in Nepal have shown that local people are strongly in favour of vulture conservation because of the associated ecological services that vultures provide[16]. The Report of the International South Asian Vulture Recovery Plan Workshop in 2004 gave a comprehensive list of recommendations including establishing a minimum of three captive breeding centres each capable of holding 25 pairs[20]. Captive breeding efforts are ongoing and met with success when two chicks hatched in early 2007 at a breeding centre in Pinjore, Haryana[21]. The centre is part of a captive breeding programme established by the RSPB and Bombay Natural History Society. A website has been set up to allow researchers to contribute data on known colonies to identify founder individuals for captive flocks that will ensure the full geographical spread of the species is represented in captive breeding efforts[18]. By April 2008 there were 88 in captivity at three breeding centres in India, as well as 11 at a centre established by

WWF-Pakistan and 14 in captivity in Nepal[25].

Conservation actions proposed:

Identify the location and number of remaining individuals and identify action required to prevent extinction. Continue to measure the frequency of diclofenac-treated carcasses available to vultures. Support the ban on the veterinary use of diclofenac, and support species management or restoration, as needed. Initiate public awareness and public support programmes. Monitor remaining populations, in particular replicate conservation and research activities that have been implemented in Cambodia in Myanmar and survey southern India where it is hoped vulture populations may not have crashed to the same extent that they have in the rest of the Subcontinent. Provide supplementary food sources where necessary for food-limited populations in South-East Asia. Support captive breeding efforts at a number of separate centres with the aim of holding at least 150 pairs of each species in captivity[25,26]. Promote the immediate adoption of meloxicam as an alternatives to diclofenac. Test other non-steroidal anti-inflammatory drugs (NSAIDs) to identify additional safe alternative drugs to diclofenac and also other toxic ones.

Citation: BirdLife International 2009. *Gyps bengalensis*. In: IUCN 2009. IUCN Red List of Threatened Species. Version 2009.2. <www.iucnredlist.org>.

2.Gyps Indicus

Taxonomy

Kingdom	Phylum	Class	Order	Family
ANIMALIA	CHORDATA	AVES	Falconiformes	Accipitridae

Scientific Name: *Gyps indicus*

Species Authority:(Scopoli, 1786)

Common Name/s:

English - Indian Vulture

Assessment Information

Red List Category & Criteria: Critically Endangered A2bce+4bce ver 3.1

Year Assessed: 2009

Assessor/s BirdLife International

Evaluator/s: Bird, J., Butchart, S., Symes, A.(BirdLife International)

Justification:

This species is classified as Critically Endangered because it has suffered an extremely rapid population decline as a result of feeding on carcasses of animals treated with the veterinary drug diclofenac.

History:

2008 – Critically Endangered

2004 – Critically Endangered

2002 – Critically Endangered

2000 – Not Recognized

1994 – Not Recognized

1988 – Not Recognized

Geographic Range

Range Description:

Gyps indicus breeds in south-east **Pakistan** (where it is rare although a 200-250 pair colony was discovered in 2003 in Sindh Province, Pakistan[3]) and peninsular **India** south of the Gangetic plain, north to Delhi, east through Madhya Pradesh, south to the Nilgiris, and occasionally further south. It was common until very recently, but since the mid 1990s, it has suffered a catastrophic decline (over 97%) throughout its range, first noticed in Keoladeo National Park, India[2] where the population fell from 816 birds in 1985-1986 to just 25 in 1998-1999. Just one tiny population in the Ramanagaram Hills of Karnataka remains in inland southern India, and it is rare elsewhere within its former range. Extensive research has identified the non-steroidal anti-inflammatory drug (NSAID), diclofenac, to be the cause behind this rapid population collapse[5,6,7,8]. This drug, used to treat domestic livestock, is ingested by vultures feeding on their carcasses leading to renal failure causing visceral gout[6,8,10,11]. In 2007 the total Indian population, based on extrapolations from road transects, was estimated at 45,000 individuals, with a combined average annual decline for this species and *G. tenuirostris* of over 16% between 2000-2007[19].

Countries:

Native:

India; Pakistan

Vagrant:

Malaysia

Present - origin uncertain:

Afghanistan

Range Map:

Population

Population: Population estimate extrapolated from 2007 survey results by Prakash *et al.* (2007) who recorded 337 individuals along >18,000 km road transects.

Population Trend: Decreasing

Habitat and Ecology

Habitat and Ecology: It is found in cities, towns and villages near cultivated areas, and in open and wooded areas. This species feeds almost entirely on carrion, and often associates with the White-rumped Vulture *G. bengalensis* when scavenging at rubbish dumps and slaughterhouses. It nests almost exclusively in small colonies on cliffs and ruins, although in one area, where cliffs are absent, it has been reported nesting in trees. Vultures also play a key role in the wider landscape as providers of ecosystem services, and were previously heavily relied upon to help dispose of animal and human remains in India.

Systems: Terrestrial

Threats

Major Threat(s): By mid-2000, *Gyps* vultures were being found dead and dying in Pakistan and throughout India, and major declines and local extirpations were being reported. The anti-inflammatory drug diclofenac, used to treat domestic livestock, has been

identified as the cause of mortality from renal failure resulting from visceral gout in the vast majority of examined vultures[6,7,8,10]. Modelling has shown that to cause the observed rate of decline in the species just one in 760 livestock carcasses need contain diclofenac residues[5]. Other likely contributory factors are changes in human consumption and processing of dead livestock, and poison and pesticide use, but these are probably of minor significance.

Conservation Actions

Conservation actions underway:

Conservation Actions:

CITES Appendix II. CMS Appendix II. It has been reported from many protected areas across its range. The Indian government has now passed a bill banning the manufacture of the veterinary drug diclofenac that has caused the rapid population decline across the Indian Subcontinent; their aim was to phase out its use by late 2005[9,10], although its sale has not been banned and it is likely to remain in widespread use for several years. Similar laws banning import and manufacture of diclofenac are now in place in Nepal and Pakistan. A letter from the Drug Controller General of India in 2008 warned more than 70 drugs firms not to sell the veterinary form of diclofenac, and to mark human diclofenac containers 'not for veterinary use'[17]. Efforts to replace diclofenac with a suitable alternative are ongoing; drug companies have now developed meloxicam, an alternative to diclofenac, which has been tested on *Gyps* vultures with no ill-effects. The Report of the International South Asian Vulture Recovery Plan Workshop in 2004 gave a comprehensive list of recommendations including establishing a minimum of three captive breeding centres each capable of holding 25 pairs[15] - ultimately at least 150 pairs of the three species should be held in captivity to ensure sufficient birds are available to re-establish wild colonies in the future[18]. Captive breeding efforts are ongoing and as of April 2008 there were 71 individuals in captivity at three conservation breeding centres in India[20].

Conservation actions proposed:

Identify the location and number of remaining individuals and identify action required to prevent extinction. Measure the frequency of diclofenac treated carcasses available to vultures. Support the ban on the veterinary use of diclofenac, and support species management or restoration, as needed. Initiate public awareness and public support programmes. Monitor remaining populations, in particular,

survey southern India where it is hoped vulture populations may not have crashed to the same extent that they have in the rest of the Subcontinent. Support captive breeding efforts at a number of separate centres. Promote the immediate adoption of meloxicam as an alternatives to diclofenac. Test other non-steroidal anti-inflammatory drugs (NSAIDs) to identify additional safe alternative drugs to diclofenac and also other toxic ones.

Citation: BirdLife International 2009. *Gyps indicus*. In: IUCN 2009. IUCN Red List of Threatened Species. Version 2009.2. <www.iucnredlist.org>

3. Gyps Tenuirostris

Taxonomy

Kingdom	Phylum	Class	Order	Family
ANIMALIA	CHORDATA	AVES	Falconiformes	Accipitridae

Scientific Name: Gyps tenuirostris

Species Authority: Gray, 1844

Common Name/s:

English - Slender-billed Vulture

Assessment Information

Red List Category & Criteria: Critically Endangered A2ce+4ce ver 3.1

Year Assessed: 2009

Assessor/s BirdLife International

Evaluator/s: Bird, J., Butchart, S., Symes, A.(BirdLife International)

Justification:

This species is classified as Critically Endangered because it has suffered an extremely rapid population decline, particularly across the Indian subcontinent, largely as a result of feeding on carcasses of animals treated with the veterinary drug diclofenac, perhaps in combination with other causes.

History:

2008 – Critically Endangered

2004 – Critically Endangered

2002 – Critically Endangered

2000 – Not Recognized

1994 – Not Recognized

1988 – Not Recognized

Geographic Range

Range Description:

Gyps tenuirostris is found in **India** north of, and including, the Gangetic plain, west to at least Himachal Pradesh and Haryana, south to southern West Bengal (and possibly northern Orissa), east through the plains of Assam, and through southern **Nepal**, and north and central **Bangladesh**. It formerly occurred more widely in South-East Asia, but it is now thought to be extinct in Thailand and Malaysia, and the only recent records are from **Cambodia**, southern **Laos** and **Myanmar**. Considerable confusion over the taxonomy and identification of *Gyps* vultures has occurred, making it difficult to be sure of claims for this species. However, it appears to be allopatric or parapatric with Indian Vulture *G. indicus* where their ranges abut (or potentially do so) in northern India. It was once common, but in South-East Asia populations declined through the latter half of the nineteenth century and the first half of the twentieth century, and are now probably very small and restricted in distribution and limited mainly to Cambodia (where the first nests recorded in the country were recently found and surveys in 2008 recorded a total of 51 individuals at vulture 'restaurants'[19]) and Myanmar (small numbers seen recently on surveys in Shan State[3]). Given the lack of intensive agriculture and associated chemical use in South-East Asia and the continued presence of large areas of suitable habitat for the species, the primary reason behind its decline in the region is thought to be the demise of large ungulate populations and improvements in animal husbandry resulting in a lack of available carcasses for vultures[7,8]. In India and Nepal, the species was common until very recently, but since the mid 1990s, it has suffered a catastrophic decline up to 99%, with a combined average decline in India of this species and *G. indicus* of over 16% annually between 2000-2007[20]. Extensive research has identified the non-steroidal anti-inflammatory drug (NSAID), diclofenac, to be

the cause behind this rapid population collapse[9,10,11,12]. This drug, used to treat domestic livestock, is ingested by vultures feeding on their carcasses leading to renal failure causing visceral gout[10,12,14,15]. Probably owing to the effects of diclofenac breeding success in parts of its Indian range is reportedly low; of 14 nests found in Assam just four had chicks[17]. Diclofenac is apparently entirely absent in Cambodia, adding greater importance to that remaining small population.

Countries:

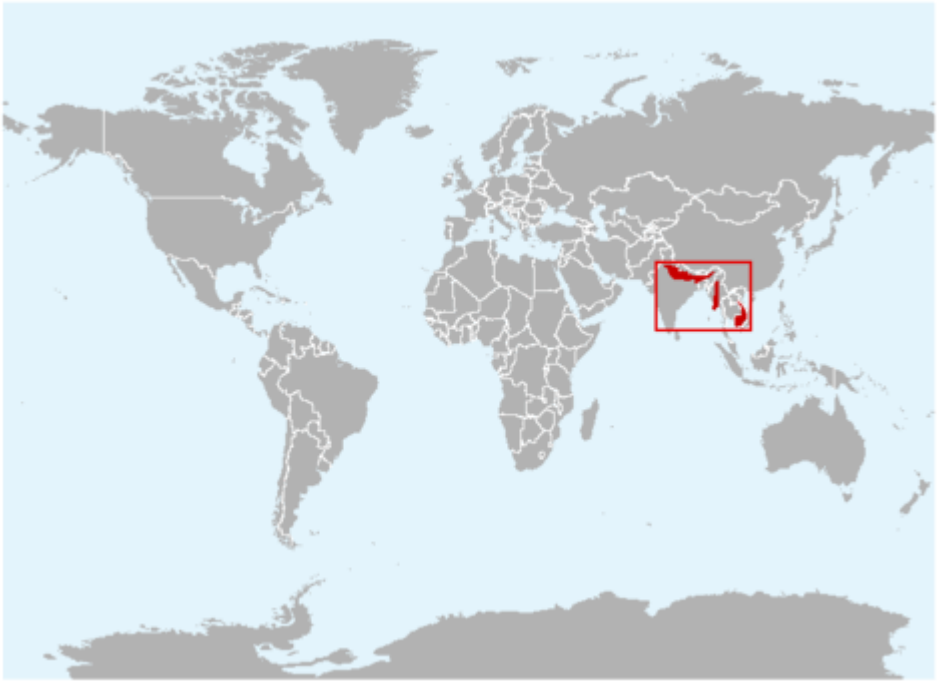
Native:

Bangladesh; Cambodia; India; Lao People's Democratic Republic; Myanmar; Nepal

Regionally extinct:

Malaysia; Thailand; Viet Nam

Range Map:



Population

Population Trend: Decreasing

Habitat and Ecology

Habitat and Ecology:

It inhabits dry open country in the vicinity of human habitation, but also breeds in open country far from villages. In South-East Asia it was found in open and partly wooded country, generally in the lowlands. This species feeds almost entirely on carrion, scavenging at rubbish dumps and slaughterhouses. It has only been recorded nesting in trees, usually large ones, usually at a height of 7-14 m, often

near villages. While feeding considerable aggregations can form, and regular communal roost sites are used. It is social and usually found in conspecific flocks, interacting with other vultures at carcasses. Movements are poorly known, and the degree of connectivity of apparently separate populations is not known. Vultures also play a key role in the wider landscape as providers of ecosystem services. They were previously heavily relied upon to help dispose of animal and human remains in India.

Systems: Terrestrial

Threats

Major Threat(s): By mid-2000, *Gyps* vultures were being found dead and dying in Nepal, Pakistan, and throughout India, and major declines and local extirpations were being reported. The anti-inflammatory drug diclofenac, used to treat domestic livestock, has been identified as the cause of mortality from renal failure resulting from visceral gout in the vast majority of examined vultures[10,11,12,14]. Modelling has shown that to cause the observed rate of decline in *Gyps* vultures, just one in 760 livestock carcasses need contain diclofenac residues[9]. Other likely contributory factors are changes in human consumption and processing of dead livestock, and poison and pesticide use, but these are probably of minor significance. In South-East Asia, the near-total disappearance of the species pre-dated the present crisis, and probably results from collapse of large wild mammal populations and improved management of deceased livestock[7], but persecution is also thought to be a problem.

Conservation Actions

Conservation Actions: CITES Appendix II. CMS Appendix II. It has been reported from many protected areas across its range. The Indian government has now passed a bill banning the manufacture of the veterinary drug diclofenac that has caused the rapid population decline across the Indian Subcontinent; their aim was to phase out its use by late 2005[13,14], although its sale has not been banned and it is likely to remain in widespread use for several years. Similar laws banning import and maufacture of diclofenac are now in place in Nepal and Pakistan. Efforts to replace diclofenac with a suitable alternative are ongoing; drug companies have now developed meloxicam, an alternative to diclofenac which has been tested on a number of species including *Gyps* vultures with no ill-effects[18]. Vulture restaurants are used as ecotourism attractions in parts of the species's range to raise awareness and fund supplementary feeding programmes

and research - in Cambodia these are run by The Cambodia Vulture Conservation Project, a partnership between the Royal Cambodian Government and NGOs, led by the Wildlife Conservation Society (WCS) and also including BirdLife International, WWF, RSPB and Angkor Center for the Conservation of Biodiversity[19]. Birds have been satellite tagged in various parts of their range to improve understanding of their movements, foraging range, site fidelity etc., in order to develop suitable conservation strategies for the species[4]. Socioeconomic surveys in Nepal have shown that local people are strongly in favour of vulture conservation because of the associated ecological services that they provide[16]. The Report of the International South Asian Vulture Recovery Plan Workshop in 2004 gave a comprehensive list of recommendations including establishing a minimum of three captive breeding centres each capable of holding 25 pairs[5]. Captive breeding efforts began in 2006 when 18 Slender-billed Vultures were captured for the captive-breeding facility in Pinjore, India. The centre is part of a captive breeding programme established by the RSPB and Bombay Natural History Society. In April 2008 there were 28 birds at the three Indian breeding centres[18]. A website has been set up to allow researchers to contribute data on known colonies to identify founder individuals for captive breeding efforts that represent the full geographical spread of the species[1].

Conservation actions proposed:

Identify the location and number of remaining individuals and identify action required to prevent extinction. Measure the frequency of diclofenac treated carcasses available to vultures. Support the ban on the veterinary use of diclofenac, and support species management or restoration, as needed. Initiate public awareness and public support programmes. Monitor remaining populations, in particular replicate conservation and research activities that have been implemented in Cambodia in Myanmar. Provide supplementary food sources where necessary for food-limited populations in South-East Asia. Support captive breeding efforts at a number of separate centres. Promote the immediate adoption of meloxicam as an alternatives to diclofenac. Test other NSAIDs to identify additional safe alternative drugs to diclofenac and also other toxic ones.

Actions taken to curb this decimation

In October 2000, BNHS jointly applied for a grant from the U.K. Government under the Darwin Initiative for the survival of species to fund the urgent vulture investigations. BNHS was successful in its bid and from 1 April 2001, the Darwin Initiative provided.

Vulture Conservation Breeding Centre, Pinjore The Vulture Conservation Breeding Centre (VCBC) is a joint project of the Haryana Forest Department and the Bombay Natural History Society (BNHS). It is a collaborative initiative to save the three species of vultures, the White-backed, Long-billed and Slender-billed, from looming extinction.
The VCBC, earlier known as Vulture Care Centre (VCC), was established in September 2001 with the UK Government's 'Darwin Initiative for the Survival of Species' fund, to investigate the dramatic declines in India's Gyps species of vultures.
Subsequent to the release of the South Asia Vulture Recovery Plan in February 2004, the VCC was adapted and upgraded to being the first VCBC, in line with a key recommendation of the Recovery Plan to set up a conservation breeding programme for the three critically endangered species of vultures. The centre sprawls over 5 acres of Haryana Forest Department's land at village Jodhpur.
Objectives

- **To establish a founder population of 25 pairs each of 3 species of vultures**
- **To produce a population of at least 200 birds of each species in 15 years to be reintroduced in the wild.**

Mission

- **To release 100 pairs each, of the three species of vultures, in the next fifteen years, to establish at least one viable population of resident Gyps, in an environment free of diclofenac and other poisons.**

1.Vulture Conservation Breeding Programme

The conservation breeding of vultures became a major objective of the vulture project after the release of Vulture Recovery Plan in February 2004. The major recommendation of the plan was to set up at least three conservation breeding facilities in India, immediately and ultimately six across south Asia.

A simple deterministic model of a captive vulture population and the wild population eventually derived from it indicated that a breeding centre with 25 pairs would be capable of producing a derived wild population of 100 pairs about 10 years after the beginning of releases. To allow for mortality in captivity and unequal number of the sexes taken from the wild, it would be necessary to take about 60 birds of each species to establish 25 pairs of each species at each breeding centre which would eventually lead to the restoration of a single wild population of 100 pairs 15 or more years later. Releases would not begin until a minimum of 6 years had elapsed, since the capture of the founding stocks (assuming that most of the founders are taken as nestlings or juveniles) and provided diclofenac is completely out of the system. The suggested age-structure of the founder population is 70-85% of known-age nestlings and 10-15% sub-adults and adults, so that most of the captive population is of known-age and are most likely to breed.

*Infrastructure & Facilities at Vulture Conservation Breeding Centre

There are various types of aviaries for keeping vultures of different age and health conditions.

Housing:

The salient features and specifications of different aviaries for housing the vultures are given below.

The Quarantine aviaries (20x20x12') are located 5 km south of the centre on

Forest Department land. There are three temporary aviaries with a capacity to hold 20 birds at a time. The aviaries are made up of iron poles and netlon. The facility provides total isolation to birds to recover from any stress or diseases. Any bird brought to the centre is first kept in these aviaries and their health is monitored for 45 days. Blood and fecal samples are analysed every fifteen days to make sure they are free of diseases.

The Hospital aviaries (12x10x8') house any bird found injured or sick with in the centre, for treatment and care. The centre has four hospital aviaries with capacity to hold a bird each.

The Colony aviaries (100x40x20') hou house sub-adults and adults of a single species after they have been quarantined for at least 45 days. These aviaries are large enough for the birds to do wing exercises by flying from one end to another and feed socially on carcasses, exactly as they do in the wild. There are three such aviaries with capacity to hold 40 birds. The colony aviaries are equipped with CCTV cameras.

The Nursery aviaries(12x10x8')are designed to provide natural nest like environment to the nestlings. The centre has ten nursery aviaries with a total capacity of rearing at least 50 nestlings at a time.

The centre has three Holding aviaries, one of dimensions (60x40x20') and two of dimensions 20x20x20' with capacity to hold 10 pairs in the big aviary and 2 pairs each

in the smaller ones. The birds are kept in these aviaries after they fledge in nursery aviaries. These aviaries are large enough for the birds to do wing exercise and flap fly from one end to another.

The centre has two Display aviaries (25x17x14'). Presently being used as holding aviaries, they will be eventually used as display aviaries for visitors. Vultures which are not fit for the breeding programme will be kept in these aviaries.

Laboratory and Veterinary Care Facilities :

The centre has a well-equipped laboratory with the following facilities.

The Clinical Room(12x10x10') is equipped with gas anaesthesia machine and other equipment required for basic surgery and disease diagnostics. The birds with any sickness or injury are treated here.

The Critical Care Room(12x10x10') is next to the clinical room. It is thermo-controlled and has critical care boxes for keeping birds while recovering from serious illness. The wooden boxes are of dimensions 3x3'.

The birds are released in the Recovery aviary(12x10x8') after recovering in the critical care room. The aviary is open to sky and has a layer of netlon on the top. The aviary has a similar design as the hospital aviary. The perching in this aviary is done in accordance to the requirement of the bird.

The Brooder Room(12x10x10') is utilised for keeping newly hatched nestlings. The nestlings would be hand reared and fed by the staff of the centre with the help of vulture puppets. This will be done to make sure that the nestlings do not get imprinted on humans. The room shares one of its walls with an aviary where handicapped adult birds would be kept. The intervening wall has a one way glass which will make sure that the adult vultures are always in sight of the nestlings.

The Incubator Room (12x10x10') is thermo-controlled and has two state-of-the-art incubators with the latest contact incubation technology. The incubator works by having a balloon filled with warm air pushing gently down on the eggs, so only the

top half of the egg receives the heat - much like a natural bird sitting on eggs. On a programmed timer - with times of our choice, the balloon lifts off the eggs, the eggs are turned on a roller and the balloon returns down on the eggs, thus warming the other side of the eggs.

Incubator Room

The Molecular Room(15x10x10') has a PCR (Polymerase Chain Reaction) machine with accessories. The equipment is utilised for sexing birds by using DNA. There is a fully automated blood biochemistry machine which helps in determining the levels of uric acid, albumin, total protein and creatine kinase in blood serum of vultures. The presence of diclofenac in the vulture tissue as well as cattle carcass tissue is detected with the help of ELISA reader also present in the lab.

The Haematology Room(12x12x10') has all facilities for carrying out routine hematology on vulture blood. The lab has a powerful Leica microscope, centrifuge machine, a Haemacue, a blood mixer and other relevant instruments and accessories to carry out haematology.

The Closed-Circuit Television Camera Monitor Room (10x10x10') is utilized for carrying out observations on the birds. All three colony aviaries are equipped with CCTV cameras to study vulture behaviour. The camera pans and tilts up to 180°. It has a zoom of 20-27X. Every corner of the colony aviary can be monitored with the help of these cameras.

The Freezer Room (12x10x10') has three -20°C freezers for storing important tissue samples of vultures.

Methodology for Collection of Vulture:

There are a total of 127 birds at the centre of which 55 are White-backed vultures, 55 Long-billed vultures, 15 Slender-billed vultures and 2 Himalayan Griffons. Of the 55 White-backed vultures, two are juveniles which were hatched at the centre during the breeding season of 2007-08. and 3 are of 2008-09 breeding season. Of the 15 Slender-billed Vultures, one is a juvenile which hatched at the centre.

Vultures have been brought to the centre from various states. The maximum number of birds (N=35) have been brought from Gujarat followed by Rajasthan (N=22), Maharashtra (N=21), Haryana (N=20), Madhya Pradesh (N=17), Assam (N=14) and Delhi (N=1). Two Himalayan Griffons have been rescued from Haryana and are not part of the breeding programme.

So far, we have lost 9 White-backed vultures and 3 Long-billed vultures at the centre.

Nestling collection

Nestlings are collected from the wild after they are 45 days old as by then their thermoregulation is well developed and they do not require beak to beak feeding. A very high percentage of mortalities in the wild occur at the nestling stage, hence, bringing nestlings into captivity saves individuals. The White-backed vultures and the Slender-billed vultures are tree nesters and nest on tall trees, sometimes as tall as 80 metres. The Long-billed vultures are cliff nesters and generally nest on inaccessible cliffs and rocky outcrops. Quite often, they nest in deep caves on a vertical face of the cliff. Nestling collection has been possible only due to the help from expert climbers trained in handling vultures. Nestlings are collected in specially designed duffel bags and lowered with the help of a rope to the team waiting on the ground. Once the nestlings are brought down, they are transported in specially designed wooden boxes.

A total of 7 White-backed vulture nestlings and 35 Long-billed vulture nestlings have been collected, so far, from the wild.

Trapping of vultures

The sub-adult and adult vultures have been successfully trapped from the wild with the help of the snake trap method. This method is very effective, inexpensive, totally non-invasive and quick. A fresh cattle or goat carcass is used as bait and the trapper sits in a grass hide some 10-15m from the carcass, waiting for vultures to feed. He uses a long bamboo pole which is about 60ft long and collapsible. The terminal end of the bamboo, of approx. 3' long, is very supple, thin and bifid. It is coated with very sticky glue, which is a mixture of the latex of peepal Ficus religiosa and mustard oil. When the vultures start feeding on the carcass, the bamboo pole is gradually slithered on the ground towards the foraging vultures by the trapper, sitting in the grass hide. When the bamboo is approx. 5 feet away, the trapper swiftly thrushes it on the body of the feeding vulture. The vulture is then unable to fly. The trapper then rushes and grabs the bird. The glue does not damage the plumage and comes of easily with any vegetable oil. A total of 14 White-backed and 14, most endangered, Slender-billed vultures have been trapped with the help of this method.

Vultures rescued:

In all, 38 White-backed vultures have been rescued and brought to the centre. Of these, 33 birds have been rescued during kite flying, which is part of the festival "Uttrayan" in Gujarat. A good number of birds get serious injuries due to kite strings. Grounded glass is used to coat the string so that in a duel with other flying kites, the ground glass coated string cuts through the strings of other kites. The flying birds get entangled in these strings and suffer serious injuries. Many birds suffer serious wing damage and are maimed for life, while many die of excessive bleeding. Vulture relief camps organized by the Gujarat Forest Department, Animal Health foundation, Bird Conservation Society of Gujarat and VCBC, Pinjore, have provided first aid care and surgery, as necessary, to these birds. The birds were then transported to Pinjore and are part of the breeding programme.

A total of 4 White-backed vultures and 1 Long-billed vulture have been rescued by individuals, NGO's and Forest Departments of various states and sent to the centre.

Husbandry and Care of Vulture:

Husbandry and care is possibly the most important component of any conservation breeding programme. The correct procedures followed ensure a healthy population in captivity.

The protocols for husbandry and care followed at the centre have been developed in consultation with international organisations with expertise in captive management of birds. A number of modifications have been made based on our experience and local circumstances. The following parameters are covered in routine husbandry and care of vultures.

Identification :

When a vulture is brought to the centre, a strong plastic ring etched with a number is put on one of the legs and a microchip is implanted in its breast muscle for identification.

Health Check :

Different examinations are carried out for providing veterinary care to vultures. A physical examination of vultures in quarantine is carried out three times during the quarantine period of 45 days. The initial check up is carried out for any signs of injury or fracture. Once out of quarantine period, vultures at the centre are physically examined once a year. Daily visual examination is carried out four times in a day. The examination is done from a distance usually through binoculars or through CCTV monitors. It includes the evaluation of the general appearance, attitude and activity of individual bird. These are the main parameters that are likely to change in response to a disease condition.

Laboratory examination involves haematology, parasitology, and basic microbiology on

live birds and necropsy on dead vultures which gives a clue of the health condition of the vultures. The centre has a lab which is equipped to carry out all this. Post mortem examination is carried out by using a standard protocol, which includes systematic visual examination of external and internal organs.

Blood Sampling

Feeding the vultures

Vultures are fed on freshly slaughtered skinned goats. Each vulture is provided with 3-4 kg of meat per week depending on the body weight; spread over two days, to maintain its daily food requirement of 5% of its body weight. To ensure that there is no diclofenac in the tissues of the goat carcasses, a herd of goats is kept in the care of the centre for at least ten days before slaughter.


Food through Food Hatch

Aviary maintenance

Aviary maintenance is critically important for ensuring the good health of the captive vultures. Routine and annual maintenance of aviaries are regularly carried out at the centre.

Perches

Special attention is given to the perch management in the aviary. Perches at different heights are provided with uneven surfaces. Coconut rope is wound around the perches for giving it a rough surface. This prevents foot problems which are common in captive raptors.

Water

Water is provided in cemented troughs inside the aviaries. Four water troughs are provided. The troughs are cleaned from outside once a week and water is topped up every day.

Breeding Success

at Vulture Conservation Breeding Centre, Pinjore

The centre has successfully bred five nestling of White-backed vultures and one nestling of Slender-billed vulture at the centre, during the breeding seasons of 2007-08 & 2008-09.

White-Backed vultures with nestling on nest

Slender-Billed Vulture with nestling

Breeding season of vultures at the centre commences from the month of September when established pairs in the colony aviaries begin defending their nest ledges and sit together most of the times. They copulate on the nest ledges, collect nest material and build nests. This coincides with the on set of breeding season in wild.

The White-backed vultures were the first ones to initiate breeding at the centre in the year 2005-06. The Long-billed's and the Slender-billed's attempted breeding for the first time in 2006-07.

Research and training

Safety testing of the non-steroidal anti-inflammatory drug Meloxicam was carried out on the White-backed and Long-billed vultures at the centre during 2005. The testing was jointly conducted by BNHS, IVRI and Haryana Forest Department. It was done in three phases: In Phase I, Meloxicam was administered to vultures orally; in Phase II, it was administered to other scavenging birds and in Phase III, the vultures were fed on Meloxicam treated buffalo meat. Blood samples were collected 48 hours after feeding. There was no adverse reaction to the drug. No significant changes were noticed in the haematological and biochemical parameters either. This experiment gave strong evidence that Meloxicam is totally safe for vultures and other scavenging birds. It is known to be as effective to cattle as diclofenac is and has hardly any side effects.

Vulture Monitoring and Surveillance

One of the main activities of the vulture centre has been nation-wide surveillance and monitoring of vulture populations and colonies. Annual nation wide surveys have been an integral part of the project since its inception, as without the data provided by these observations, it is impossible to know the extent or spatio-temporal variations of the declines in India. The annual surveys conducted have provided valuable data on the degree and geographical extent of the declines. The latest survey carried out in 2007 has showed that populations of three species of Gyps vulture continue to decline precipitously, with White-backed vulture declining at the catastrophic rate of approximately 42.3% per year and the Long-billed and Slender-billed vultures at the rate of over 17% per year.

Workshop on tissue extraction and ELISA based analysis of Diclofenac

The workshop was held at the centre from 28th April to 6th May. The main resource persons were Dr. Mark Anthony Taggart from University of Aberdeen, UK, and Dr. Mohini Saini from IVRI. The participants were the researchers from BNHS and IVRI. The main objective of the workshop was to impart training on the ELISA (Enzyme Linked Immuno-Sorbant Assay) based analysis of diclofenac which will help in determining whether an animal carcass has diclofenac in the tissue or not. This will help in making sure that the food offered to vultures is free of diclofenac. The workshop gave attendees a practical, highly interactive, hands-on experience of the laboratory based techniques that could be used to both extract and analyse diclofenac in animal tissues. Such work is relevant to the ongoing long term conservation and protection of critically endangered Gyps sp. of vultures within India.

Workshop for establishment of ex-situ

conservation centres for vultures in Indian Zoos

The WII organised a three days workshop for 'Establishment of ex-situ conservation Centres for Vultures in Indian Zoos' at Pinjore from 1-3 November 2006 in collaboration with the BNHS and the Haryana Forest Department. The CZA, the apex regulatory body of Government of India for keeping animals and birds in captivity, sponsored this workshop.

The major objective of the workshop was to train the participants in developing a project proposal for the proposed conservation breeding centres in the four zoos identified by Government of India.

These zoos are:

1. Van Vihar, Bhopal, Madhya Pradesh
2. Nehru Zoological Garden, Hyderabad, Andhra Pradesh
3. Nandan Kanan Zoo, Bhubaneshwar, Orissa,
4. Sakkarbaug Zoo, Junagadh, Gujarat.

The main resource persons for the workshop were drawn from Haryana Forest Department, VCBC, Pinjore, WII and IVRI.

Training in captive care and management of vultures imparted by the centre

The centre now has gained more than 8 years experience in the captive management and care of resident Gyps species of vultures; it is recognised by the CZA as the coordinating institution for the conservation breeding of vultures in the country. It has been imparting training to various stakeholders in conservation breeding programme.

A team of six staff of Vulture Conservation Breeding Centre, Nepal, including the Project Manager, vulture keepers and an Assistant Wildlife Conservation Officer, Chitwan National Park, came for training at the centre in August 2008.

Training of VCBC NepalTeam

They were showed the centre and were explained the functioning of the centre in detail. The birds and aviaries were shown through CCTV monitors and identification of the three species of vultures was explained. Data collection and recording was demonstrated.

Catching of birds at the centre was demonstrated and their processing like ringing, microchipping, morpho-metrics, moulting patterns was explained to them.

Zoo Officials from Sakkarbaug Zoo, Gujarat, Van Vihar, Madhya Pradesh, Nandan Kanan, Orissa and Nehru Zoological Park, Andhra Pradesh were imparted training in captive breeding, vulture husbandry care and captive management.

A visit to the centre by In-service Officers and Officer Trainees has become part of the various technical courses conducted by the country's premier forest training colleges - Indira Gandhi National Forest Academy, Dehradun, Uttranchal and forest training school.

Batches of officers and field staff regularly visit the centre. Students of Master's course as well as Forest Officer's refreshers course conducted by the WII, visit the centre as part of their ex-situ training module.

The Future Plans

1. The ultimate aim of the conservation breeding programme is to reintroduce vultures in the wild. The centre, in the coming years, will endeavour to create facilities and breed vultures in good numbers for the success of the release programme.

2. Construction of a new colony aviary, eight breeding aviaries and a CCTV monitor cum interpretation room is proposed.

3. Monitoring of identified release sites will be taken up to make sure there is enough food and habitat available for the released vultures. It would be totally ensured that there is no diclofenac used for treating livestock in at least 10 km radius of the released sites, prior to release.

The release programme will commence after the year 2012. The birds born at the centre will be kept for two years and then will be released in the pre-monitored sites. The birds will be released in flocks of 20 with a few wild caught adults which will act as guide birds. The released birds will be fitted with satellite tags and will be constantly monitored.

There are nine species of vultures found in .This includes the Long-billed vulture (*Gyps indicus*), Slender billed vulture (*Gyps tenirostris*), White-rumped vulture (*Gyps bengalensis*), Red-headed vulture (*Sarcogyps calvus*), Cinereous vulture (*Aegypius monachus*), Lanmergeier vulture (*Gypaetus barbatus*), Egyptian vulture (*Neophron percnopterus*), Eurasian griffon (*Gyps fulvus*) and Himalayan griffon (*Gyps himalayensis*). Long-billed vulture, Slender billed vulture and white-backed vultures listed in Indian Wildlife Act Schedule I and IUCN list as "Critically Endangered" (Birdlife International, 2000).

Climatic conditions, socio-economic condition, traditional livestock rearing and concept of village institutions, *Orans* (village forests) and *Gauchars* (village pasture) seems to have played important role in the conservation of vulture population in different parts of India. A variety of explanations and hypotheses have been proposed, including a reduction in food availability, poisoning, habitat loss, pesticide intoxication, calcium deficiency, infectious diseases or a-viral disease The cause of mortality remains unidentified but is suspected to be an infectious disease. Recently diclofenac residues have been identified as a cause for declining population of oriental white-backed vulture (*Gyps bengalensis*) in Pakistan, but no such study has been conducted in India on any Gyps species. The Union Ministry of Environment and Forests has, with assistance from the Bombay Natural History Society, the Royal Society for the protection of birds and the Zoological Society London, started captive

breeding centres at Pinjore in Haryana and Buxa in west Bengal. However, no Indian organization has expertise in captive breeding and there are already problems because of a lack of skilled hands. Temperature fluctuations led to the death of two nestling in Pinjore in January 2007 (Chhangani, 2007b).Captive breeding should not be the only way to conserve all the vultures with the little effort and low cost by involving local community and forest officials. My team has saved more than 30 long-billed and white-romped vultures in the last two years in Rajasthan and Gujarat. I am quite certain that a countrywide rescue programme can save over 100 threatened vultures every year. This is something which an expensive captive breeding programme will not be able to match in the next 10-15 years.

Vulture Conservation Action Plan

1. There is an urgent need of state wise regular monitoring and survey of important vulture population sites (feeding, roosting and nesting) of all vulture species, so that the population trend and ecology could be understood and more importantly any sick, weak and nest fallen vulture of threatened species can be rescued before it dies or becomes victim of predators.

2. Monitoring of selected breeding colonies of long-billed and white-backed vultures for 2-3 years to understand the breeding success and breeding ecology. This is also important during the breeding season to locate the nest fallen chicks rescue and care which is going to increase the breeding success and conservation of the vulture.

3. The long-billed vulture breeding populations were observed in and around the protected areas and unprotected areas. These are however facing many biotic and aboitic threats. Therefore, rigorous efforts for *in-situ* conservation are needed. The areas with significant vulture population should be declared as vulture protected areas or vulture sanctuaries.

4. Most of the white-backed vulture populations were observed out side protected areas in association with human habitation, farmlands, agriculture and horticulture areas, gardens, etc. We consider it important to take up education and awareness programmes in the communities and ensuring their participation in vulture conservation.

5. Habitat loss should be controlled by stopping mining and quarrying in and around the nesting and roosting sites. Tree cutting and lopping of branches should be stopped in the nesting and roosting areas.

6. Improvement in the ecology of different vulture sites as a long-term strategy is called for. This includes regular supply of food and water, eradication of exotic plant needs from nesting sites and protection of nesting sites including ban of hunting.

7. A Special Cell be constituted at State Level, which can coordinate the vulture conservation and monitoring, sample collection, permission for care and rescue, veterinary facilities, etc.

8. All existing Zoo's in Rajasthan may play important role in rescue and care. They be given backup support to initiate with small facility and veterinary care.

2. Ban on the veterinary use of Diclofenac in India

The centre played a crucial role in getting the veterinary use of diclofenac banned in the country. This became possible because of the efforts taken by the Hon'ble Forest Minister, Ms. Kiran Choudhry. She met the Prime Minister and the Chairperson of the ruling United Progressive Alliance and effectively convinced them to ban the veterinary use of diclofenac as it was responsible for the crash in vulture populations. The Drug Controller General of India instructed vide his letter dated 11 May 2006, to all the state drug controllers to withdraw the licences granted to manufacture diclofenac formulations for veterinary use. The final gazette notification was issued in August 2008. The BNHS advocacy programme worked tirelessly behind the scene to make this happen.

F.No. 18-03/2006-DC

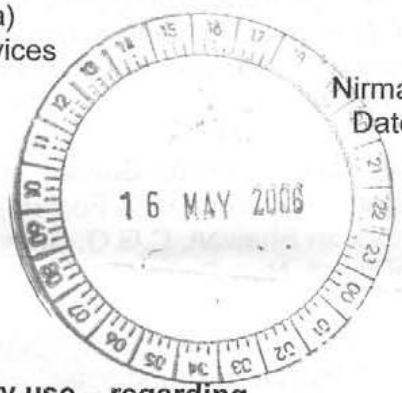
From:

The Drug Controller General (India)
Directorate General of Health Services

Nirman Bhawan, New Delhi
Dated the 11th May, 2006

To

All State Drugs Controllers



Subject: Diclofenac for veterinary use – regarding.

Sir,

Serious concern has been expressed at different fora over the decline in the population of Vultures in Indian subcontinent. Extensive studies have indicated that use of diclofenac in livestock's is the major cause of Vultures decline. Vultures are exposed to diclofenac when they consume carcasses of livestock's treated with diclofenac before death. This results the poisoning of Vultures leading to their death because of renal failure etc. It has therefore been felt that diclofenac for veterinary use should be phased out and alternate safer and effective drug like Meloxicam etc. should be permitted to be used for the treatment of animals in veterinary healthcare. This would help in saving the vulture population and ecological balance in animal world.

It is understood that, Dept. of Animal Husbandry and Dairying, Ministry of Agriculture has also issued direction to State Veterinary Departments not to purchase diclofenac for further veterinary use.

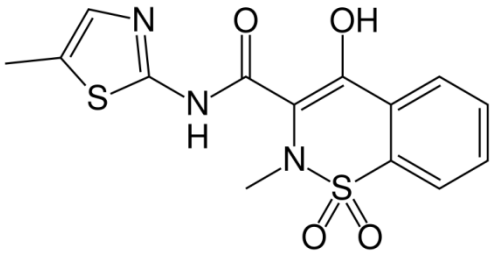
It has therefore been decided with the approval of Health Ministry that Licences granted to manufacture diclofenac formulations for veterinary use should be withdrawn and the marketing of such formulations to be phased out within a period of three months. You are therefore requested to implement the decision to withdraw the veterinary formulations of diclofenac and to ensure its phasing out within three months.

Yours faithfully,

(ASHWINI KUMAR)
DRUGS CONTROLLER GENERAL (I)

Order of The Director Controller General of India

3. Finding Alternative Drugs



Meloxicam, a non-steroidal anti-inflammatory drug, has been shown to be a safe and effective replacement for diclofenac, a widely used veterinary NSAID that has inadvertently killed tens of millions of Asian vultures on the Indian subcontinent. An international team identified meloxicam as a potential replacement drug because it has similar efficacy to diclofenac and is already approved for human and veterinary use in many countries. The researchers then gave vultures oral doses of meloxicam and meat from cattle treated with the drug and found that the birds suffered no ill effects.

A series of experiments were conducted on the administration of diclofenac and Meloxicam.

Combining the results of this study with those from the questionnaire to zoo veterinarians, a total of at least 88 individual birds from seven *Gyps* species are known to have received meloxicam at various doses with no recognized adverse effects. Hence, with this total of treated birds there is a 95% chance that the per trial probability of mortality caused by meloxicam is no higher than 3.5%. The observation that serum concentrations of uric acid remain within the normal range for all meloxicam dose rates adds substantially to the evidence that meloxicam has low toxicity to*G. africanus,* given that uric acid concentrations in this and two other *Gyps* species were markedly elevated by lethal treatment with diclofenac. Preliminary results from the NSAID questionnaires indicate the safety of meloxicam to a wide range of other vultures, raptors, and scavenging bird species, and to date we know of more than 700 individuals from more than 30 species that have been treated with no apparent adverse effects (unpublished data). This demonstrates that, at recommended clinical dose levels, meloxicam is not toxic to a wide range of avian species.

Tests conducted by Richard Cuthbert from the Royal Society for the Protection of Birds (RSPB) led the study, which involved researchers from academic institutes and conservation organisations in Europe and Africa.
These included the Bombay Natural History Society, Namibia's Rare and Endangered Species Trust and the University of Pretoria in South Africa] showed that meat from animals that had been treated with ketoprofen could be lethal for the birds.
"It's fair to say that ketoprofen is less toxic than diclofenac," said Dr Cuthbert, "and if it's used properly, there probably would be a very low risk to vultures.

"But we know that these drugs are often not used properly, and two or three times the dose is often administered to cattle."
Rhys Green, a zoologist from the University of Cambridge concluded: "There are also other drugs of the same family in use, which have not been tested for their effects on vultures.
"Testing is expensive and pharmaceutical companies aren't required to do it."

Gyps Species	NSAID	Phase	Dose (mg kg^{-1})	Route	*N* Dosed	*N* Died	% Mortality	*N* Control	Status and Source of Birds
G. bengalensis	Diclofenac	—	0.007 to 0.940	Fed treated tissue	20	13	65	—	Captive birds (Pakistan)[a]
G. bengalensis	Diclofenac	—	0.25 and 2.5	Gavage	4	3	75	2	Captive birds (Pakistan)[a]
G. africanus	Diclofenac	—	0.8	Gavage	2	2	100	2	Captive birds (South Africa)[b]
G. africanus	Meloxicam	I	0.5	Gavage	5	0	0	3	Captive birds (South Africa)
G. africanus	Meloxicam	II	1.0	Gavage	5	0	0	3	Captive birds (South Africa)
G. africanus	Meloxicam	III	2.0	Gavage	5	0	0	3	Captive birds (South Africa)
G. africanus	Meloxicam	IV.1	2.0	Gavage	14[c]	0	0	—	Captive birds (South Africa)
G. africanus	Meloxicam	IV.2	2.0	Gavage	21	0	0	4	Wild-caught birds (Namibia)
G. africanus	Meloxicam	V	0.03 to 1.98	Fed treated tissue	6[d]	0	0	—	Captive birds (South Africa)
G. africanus	Meloxicam	V	1.18 to 2.45	Gavage	6[d]	0	0	—	Captive birds (South Africa)
G. bengalensis	Meloxicam	VI	0.5	Gavage	3	0	0	1	Captive birds (India)
G. bengalensis	Meloxicam	VI	2.0	Gavage	3	0	0	1	Captive birds (India)
G. indicus	Meloxicam	VI	0.5	Gavage	2	0	0	2	Captive birds (India)
G. indicus	Meloxicam	VI	2.0	Gavage	2	0	0	1	Captive birds (India)

There was no mortality in any of the control birds.

[a]Experimental results from reference [1].

[b]Experimental results from reference [7].

[c]Experimental and control birds from phases I to III (including three control birds not previously dosed with meloxicam).

[d]Five of the six birds were experimental birds from Phase III and IV.1. The same birds were used for feeding tissue and oral gavage, with a 2-wk washout period between treatments (see Materials and Methods).

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Experimental Results

Conferences in which the topic of Vulture conservation was raised

5th World conference on Birds of prey and owls had resolved the following regarding the vulture conservation :

Resolution 4 :

RECOGNISING that recent studies clearly show that the European Griffon Vulture *Gyps fulvus* makes very long movements in their Mediterranean area and that there is a high exchange of individuals between different and distant colonies, and

RECOGNISING that the European Griffon Vulture is particularly threatened by poisoning, electrocution, direct killing and disturbance at the nesting colonies

URGES the Mediterranean countries involved to protect carefully the existing colonies and suitable buffer zones in order to prevent disturbance and habitat deterioration, to enforce legislation forbidding poisoning and to re-introduce Griffon Vultures where appropriate.

Resolution 6 :

DEEPLY CONCERNED by the recent massive poisoning of Griffon Vultures *Gyps fulvus* in Israel,

SUPPORTS the very important decision of the Israeli Minister of the Environment to nominate a committee to investigate the incident and

STRONGLY URGES the Israeli Minister of the Environment to implement all recommendations of the committee at the earliest.

6th World conference on Birds of prey and owls had resolved the following regarding the vulture conservation :

Resolution 4 :

WHEREAS windfarms can cause considerable mortality of raptors such as vultures and eagles, and threaten endangered or highly vulnerable migratory and resident species of raptor and other birds;

RECOGNISING the importance of renewable energy sources;

URGES all governments to assess the positioning of windfarms for impacts on raptors and other aspects of the environment; and

FURTHER URGES governments to ban their use in areas which possess high densities of vulnerable bird species or are located on major bird migration routes; and

SPECIALLY URGES the Israeli Government to cancel the plan to build windfarms along the Great Rift Valley, a bird migration route of global importance.

Resolution 5 :

WHEREAS there is growing evidence that pesticides of high toxicity to birds such as Monocrotophos as well as other organophosphorus and carbonate pesticides in particular are being used to poison birds and other vertebrates;

WHEREAS raptor populations, such as Vultures in southern Europe, the Balkans, Middle East and Africa are placed at risk by this practice;

RECOGNISING that insecticides of high toxicity to birds are often not essential to farming practices;

URGES the competent authorities to restrice the availability of such insecticides where these products are being abused and to carry out the necessary education and enforcement to ensure swift ceasation of these practices.

Resolution 15 :

WHEREAS populations of three species of vulture, numerous in the Indian Subcontinent as recently as six years ago, are rapidly declining as a result of a high rate of mortalities and may even be approaching extinction;

THANKS The Peregrine Fund of the USA, the Royal Society for the Protection of Birds in the United Kingdom and their partner organisations for their efforts to find the cause of these mortalities in programmes they have developed with colleagues in Pakistan, Nepal and India.

ENCOURAGES their ongoing cooperation towards the recovery of vulture populations.

Interview: Viral Prajapati-Nature Club,Surat

Nature Club, Surat is a nature education and conservation, non-government organization, located in Surat, Gujarat state, India.

We : Whether the NCS knows about the Indian Vulture Crisis?
Mr. Prajapati : Ofcourse, we know.

We : What have you done in this regard?
Mr. Prajapati : We have formed a "Vulture team" comprising of 22 members, which keep an eye on the flying and resting place of the vultures.

We : What regions do you cover?
Mr. Prajapati : Our NCS is confined to Surat district, Dang district and the areas nearby.

We : What are the major programs you have conducted so far?
Mr. Prajapati : It was decided to conduct Vulture counts in whole of Gujarat on 26th and 27th of May 2007. To keep an eye on them or to count them was not a big issue but what was important was doing the counts on the said dates in the whole of Gujarat at the same time.
So, the work of counting Vultures in Surat as well as Dang district was undertaken by Nature Club Surat and Vulture Cell of Bird Conservation Society of Gujarat.

We : How was this counting procedure carried out?
Mr. Prajapati :
SURAT DISTRICT :

There are only two places in Surat district where Vultures(Gyps Bengalensis) are seen.

1. Hazira region where Vultures built their nests and
2. Akhakhol where dead cattle's are disposed off due to Panjarapole, which is a good feeding site for the Vultures.

Apart from Surat they were also seen in Mahuva Taluka of Surat District.

Hazira has a vast area which is in Olpad Taluka near Surat and so considering the area six places were selected to keep an eye on the flying and resting place of the Vultures. Two volunteers each were keeping an eye on all the six points. White-rumped Vultures (*Gyps bengalensis*) recorded in the Hazira area are as follows:

Places	**26th May '07**		**27th May '07**	
	In flight	**Perching**	**In flight**	**Perching**
Hazira (3 points)	26	09	25	15
Suvali (1 point)	25	05	05	19
Junagam (1 point)	27	04	03	28
Vasva (1point)	24	08	27	13
Total	102	26	60	75
		128		**135**

Hazira is one of the favorable places for the White-rumped Vultures (*Gyps bengalensis*) to breed. And if the trees are being saved from cutting where the vultures roost or built their nests and if food is easily available to them nearer than we can save the White-rumped Vultures (*Gyps bengalensis*) from being extinct or may even find an increase in their number.

DANG DISTRICT :

There are two places in Dang district where Vultures(Gyps Indicus) are seen.

1. Gadad and Chinchli
2. Sanghvad (Nakatiya Hanvat)

Places	**26th May '07**	**27th May '07**
	In Flight	**In Flight**
Gadad and Chinchli	26	36
Sanghvad (Nakatiya Hanvat)	-	05
Total	**26**	**41**

During this year survey was undertaken only at the above said places. Apart from here Vultures have been sighted near Bhintbari and Moriyaghat also. And so it can be concluded that we could find Vultures in good number here and also their nests.

We : What do you do to create awareness among the local folk?
Mr. Prajapati :

1. Documentary "Vanishing Vultures" in Gujarati, given to us by Mr. Chris Bowden of RSPB and Mr.Vibhuprakash of BNHS is being used to create awareness among the villagers where the vultures are breeds.
2. We visit local colleges and schools to spread awareness regarding the same.

We : What do you think is the root cause for this decline?
Mr. Prajapati : There are 3 major reasons :

1. Use of Diclofenac and ketoprofen(veterinary drugs) in livestock.
2. Low availability of food.
3. Loss of nesting habitat due to deforestation and industrialization in those regions.

We : What steps have you taken so far to prevent this?
Mr. Prajapati :

1. For reducing the usage of Diclofenac and ketoprofen, we sell the alternative safer drug Meloxicam at subsidized rates to the veterinary doctors.
2. We have set up feeding centres at the periphery (7 kms) of the nesting sites. Because of this we could breed the Vultures naturally rather than breeding them in captivity. For feeding we bring dead cattle, for which we are constantly in touch with the shepherds and butchers.

Making arrangements of food for vultures

Conclusion

The investigations described here have some interesting features. Quantification of the scale of the declines and estimation of minimum mortality rates by carcass searching were important in establishing elevated adult mortality as the main demographic mechanism of the declines. However, because of the absence of widespread bird population monitoring, it proved difficult to measure the declines and identify when and where they began. The mobilization of scientific effort was rapid, once the declines had been recognized, but wildlife protection legislation, though essential in other contexts, was an obstacle to identifying the cause of elevated mortality. The engagement of a diversity of researchers from several countries and scientific disciplines and from academic institutions, NGOs and government agencies was vital. Their organization into separate research groups, which were conducting complementary research, in competition, but also in communication with one another, was a stimulus to progress and rigorous evaluation of hypotheses.

Once the cause was identified, the international research and conservation community, and the Indian Ministry of the Environment and Forests, closely followed by the authorities in Nepal and Pakistan, pulled together with remarkable rapidity and determination to find a solution to the problem. This international collaborative effort was exceptional, with academics setting aside their research agendas to give priority to this work alongside conservation scientists, advocates, civil servants and politicians. Conservation NGOs played a central role in arguing for action and in funding and designing relevant research. Considerable progress has already been made, but saving Asian vultures remains a daunting challenge, requiring effort and vigilance for decades to come. Establishing viable captive populations, removing diclofenac from the vulture food supply in the Indian subcontinent and preventing its replacement by other toxic NSAIDs and urging them to use safer NSAIDs (viz. Meloxicam) are the main short-term priorities. Maintaining and breeding vultures in captivity for reintroduction, restoring wild populations and preventing future adverse impacts of NSAIDs and other veterinary drugs are tasks for the longer term.

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