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NECK-DROOPING POSTURE IN ORIENTAL WHITE-BACKED VULTURES (*GYPS BENGALENSIS*): AN UNSUCCESSFUL PREDICTOR OF MORTALITY AND ITS PROBABLE ROLE IN THERMOREGULATION

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ABSTRACT.—Populations of three *Gyps* vulture species in the Indian subcontinent have undergone recent rapid declines due to elevated mortality rates caused by diclofenac poisoning. Researchers have proposed that vultures adopt a previously-undescribed neck-drooping posture prior to death. Our study investigated two hypotheses: (1) neck drooping is temperature-dependent, (2) neck-drooping is indicative of poor health in Oriental White-backed Vultures (*Gyps bengalensis*) and is a prelude to death. Observations of neck-drooping were highly seasonal, with the majority of vultures observed neck-drooping in the hot season from April to October and no vultures observed neck-drooping during the cold months of December and January. Above a calculated threshold ambient temperature of 15.4°C (95% CI 9.9–22.5°C), there was a significant positive correlation between temperature and the proportion (expressed as an angular transformation) of vultures observed neck-drooping. Neck-drooping vultures were observed significantly more frequently with their back toward the sun with their head in their own shade than birds that were not neck-drooping, and vultures that were not neck-drooping were observed more frequently facing the sun than those neck-drooping. Together, these observations strongly suggest that neck-drooping posture has a role in thermoregulation. In contrast to the highly seasonal pattern of neck-drooping, mortality of vultures occurred in all months of the year. This finding indicates that neck-drooping has low specificity and sensitivity as an indicator of poor health and impending death in vultures.

KEY WORDS: *Oriental White-backed Vulture*; Gyps bengalensis; *diclofenac*; *mortality*; *neck-drooping*; *head-drooping*; *thermoregulation.*

POSTURA DE CUELLO CAÍDO EN *GYPS BENGALENSIS*: UN FACTOR QUE NO PREDICE EXITOSAMENTE LA MORTALIDAD Y SU POSIBLE ROL EN LA TERMORREGULACIÓN

RESUMEN.—Las poblaciones de tres especies de buitres del género *Gyps* han disminuido rápidamente en el subcontinente de India debido a incrementos en la mortalidad causados por envenenamiento con diclofenaco. Algunos investigadores han propuesto que los buitres adoptan una postura no descrita previamente en la que el cuello se muestra caído antes de la muerte. Nuestro estudio investigó dos hipótesis: (1) la postura de cuello caído es dependiente de la temperatura, (2) la postura de cuello caído indica una mala condición de salud en *Gyps bengalensis* y es un preludio a la muerte. Las observaciones de la postura de cuello caído fueron altamente estacionales; la mayoría de las observaciones de esta postura se registraron en la época cálida que va desde abril hasta octubre y ésta no se observó durante los meses fríos en diciembre y enero. Por encima de una temperatura de ambiente calculada de 15.4°C (95% IC 9.9–22.5°C), hubo una

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correlación positiva y significativa entre la temperatura y la proporción (expresada como una transformación angular) de buitres con el cuello caído. Los buitres con el cuello caído fueron observados significativamente con mayor frecuencia dando la espalda al sol y con sus cabezas en su propia sombra, que las aves que no presentaban la postura de cuello caído. Por otro lado, los buitres que no presentaban la postura de cuello caído se observaron con mayor frecuencia con las caras hacia el sol que aquellos que presentaban el cuello caído. En conjunto, estas observaciones sugieren fuertemente que la postura de cuello caído tiene un papel en la termorregulación. De manera contraria al patrón altamente estacional de la postura de cuello caído, la mortalidad de los buitres ocurrió en todos los meses del año. Este resultado indicó que la postura de cuello caído tiene poca especificidad como un indicador de una condición de salud pobre y que no es una señal de muerte inminente en los buitres.

[Traducción del equipo editorial]

The widespread mortality and population declines of *Gyps* vultures across India, Pakistan, and Nepal since the mid-1990s have been widely documented (Prakash 1999, Gilbert et al. 2002, Prakash et al. 2003, Gilbert et al. 2006). Because of the magnitude of these population declines, three species, Oriental White-backed Vulture (*Gyps bengalensis*), Long-billed Vulture (*G. indicus*) and Slender-billed Vulture (*G. tenuirostris*), have been listed as critically endangered by the IUCN (BirdLife International 2001). Findings in Pakistan (Oaks et al. 2004), and subsequently in India and Nepal (Shultz et al. 2004) showed that vulture mortality was primarily due to an acute toxicity to diclofenac, a pharmaceutical used across the Indian subcontinent for the treatment of livestock, the vultures' staple food source.

Initial reports of the vulture declines in Keoladeo National Park in the Indian state of Rajasthan indicated that, prior to death, vultures would adopt an unusual posture in which they would appear to "doze off with neck limp and hanging" (Prakash 1999). A sample of five vultures were reported to exhibit this posture for increasingly prolonged periods over 30 d prior to death (Prakash 1999), although measures taken to ensure continuity of individual observation were not described. In the absence of previous reports of this posture in Indian vultures (Prakash et al. 2003), it was assumed to be a novel behavior, a prelude to death, and was used as a primary indicator of ill health (Cunningham et al. 2003, Pain et al. 2003, Prakash et al. 2003). Although it was acknowledged that healthy birds may also adopt this posture, it was believed to be more frequently observed in populations experiencing elevated mortality (Cunningham et al. 2003, Prakash et al. 2003) and was used as an indicator for the presence of the causative agent responsible for vulture mortality and population decline (Cunningham et al. 2003).

Vultures have a wide thermoneutral zone and exhibit a variety of behavioral and physiological adaptations that allow them to maintain thermal homeostasis (Hatch 1970, Larochelle et al. 1982, Arad and Bernstein 1988, Arad et al. 1989, Bahat 1995). Postural changes have been shown to play an important part in dissipating heat at high ambient temperatures in Cathartid vultures (Hatch 1970, Larochelle et al. 1982, Arad and Bernstein 1988, Arad et al. 1989) and Old World vultures (Bahat 1995, Shobrak 2001). Birds extend their necks, and unfold or partly unfold their wings, in order to expose unfeathered areas. Vascular adaptations in Turkey Vultures (*Cathartes aura*) maximize heat dissipation (Arad et al. 1989), and Cathartid vultures and Wood Storks (*Mycteria americana*) also exhibit urohidrosis, increasing evaporative heat loss by excreting on their legs (Kahl 1963, Hatch 1970, Larochelle et al. 1982, Arad and Bernstein 1988, Arad et al. 1989).

It remains unclear whether there is any distinction between the postural thermoregulation described in Cathartid and Accipitrid vultures and the neck-drooping behavior observed in South Asian vultures. If this behavior were to prove a reliable indicator of illness prior to death, its utility as an early warning of impending vulture population declines could be significant to conservationists managing vulture populations. This study represents the first attempt to critically examine the neck-drooping posture described in South Asian vultures by correlating posture and ambient conditions, observed declines, and mortality rates at vulture colonies in Pakistan.

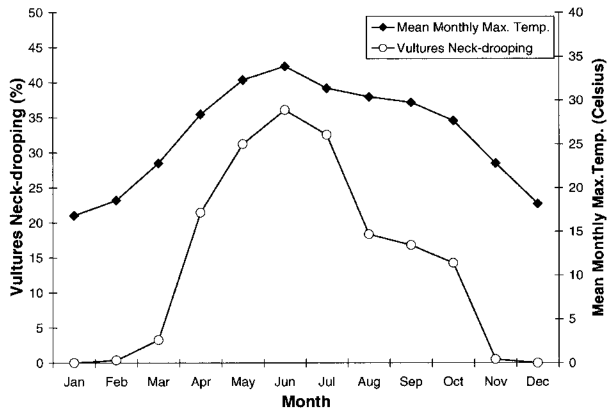
METHODS

This study was conducted in Punjab province, Pakistan, over 32 mo from January 2001 through August 2003, at three Oriental White-backed Vulture colonies, where vulture mortality and population size were simultaneously being measured as the focus of a larger research project. The colonies at Dholewala, Toawala, and Changa Manga were described in detail in Gilbert et al. (2002), and annual mortality rates and population declines in the same study sites and period were described in Gilbert et al. (2006).

The climate in Punjab province is seasonal and can be summarized as cool and dry from October to March, hot and dry from April to June, and hot and wet from July to September (Federal Bureau of Statistics 2005). Mean monthly temperatures at Multan, the meteorological station closest to our study site, range from a mean minimum of 4.5°C in January to a mean maximum of 42.3°C in June, and mean annual rainfall is 195 mm (Federal Bureau of Statistics 2005). Vultures were present at the colonies throughout the year, with highest numbers during the breeding season. The majority of eggs were laid in October/November, with fledging in April/May.

Surveys to record the posture of perched vultures were conducted on a weekly or monthly basis beginning in January 2001 and ending in August 2003. Surveys were suspended in Changa Manga in July 2001, after vulture numbers had become severely depleted at this site due to mortality from diclofenac poisoning (Gilbert et al. 2002, Gilbert et al. 2006). Surveys were conducted during two sessions (sunrise to noon and noon to sunset) on consecutive days, with equal effort expended during all hours of daylight. Counts of vultures were made by observers who walked through the colonies on established, frequently-used, foot trails, and care was taken to avoid disturbing and flushing the birds. The posture of all perched vultures when first seen was recorded. A vulture was considered to be in neck-drooping posture when its neck was hanging vertically down with its head inverted and beak pointing toward its feet for a period of at least 15 sec. The orientation of each bird relative to the sun (side, back or breast directed toward the sun) was also recorded. Ambient temperature was recorded every 30 min during observations using HOBO® automated data loggers (Onset Computer Corporation, Bourne, MA, U.S.A.). Loggers were placed in sheltered positions out of direct sunlight at each colony.

Vultures found dead with evidence of avian visceral gout were assumed to have died from diclofenac poisoning (Gilbert et al. 2006), based on diagnostic results published by Oaks et al. (2004) and Schultz et al. (2004). The mean number of vultures dying of diclofenac poisoning in each month of the study period was calculated from dead vultures found with gout during intensive transect surveys located opportunistically within the study area and conducted at least twice weekly (Gilbert et al. 2006).



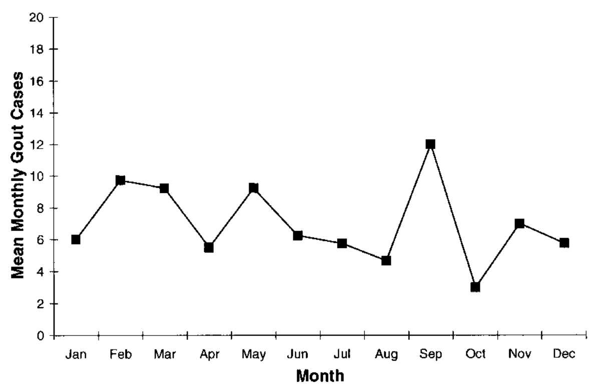


Figure 1. (a) Monthly proportion of vultures observed neck-drooping (open circles, expressed as a percentage of the total number of vultures for which posture was observed each month) at three Oriental White-backed Vulture colonies in Punjab province, Pakistan, and mean monthly maximum temperature at Multan (closed diamonds, expressed in degrees Celsius, Federal Bureau of Statistics 2005). (b) Mean monthly number of deaths due to diclofenac poisoning (closed squares) identified by the presence of visceral gout during necropsy examination of vultures at the three colonies between November 2000 and July 2004.

RESULTS

Colony sizes at the beginning of the study in January 2001 were 758 breeding pairs at Changa Manga, 421 pairs at Dholewala, and 445 pairs at Toawala. An unknown number of nonbreeding vultures including adults, subadults, juveniles, and fledglings were also observed perched in the colonies and included in posture surveys. Additional variability in colony sizes resulted from reproduction, mortality, and movement of individuals between these and other colonies. Thus, we could not estimate the total number of vultures potentially available for observation, but it exceeded at least 3248 (maximum breeding pairs × 2) by an unknown and varying number during the study period. During the 32 mo study period, we made 31 683 observations of individual vulture posture. Although the chance of counting individual birds more than once at any given ambient temperature was probably small, we took the added precaution against making Type I errors in statistical tests from possible pseudo-replication by adopting a significance level of alpha = 0.01.

Observations of neck-drooping were highly seasonal, with the majority of neck-drooping vultures recorded in the hot season between April and October (Fig. 1). By contrast, no vultures were observed neck-drooping during the cold months of

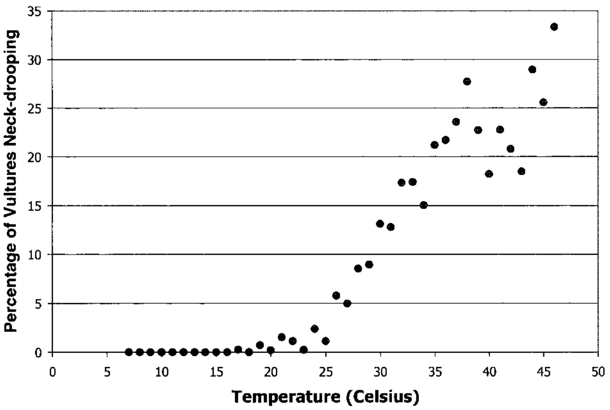


Figure 2. Proportion of vultures observed neck-drooping in each 1°C ambient temperature interval from 7–46°C at three Oriental White-backed Vulture colonies in Punjab province, Pakistan.

December (N = 594 vulture observations) and January (N = 1459). Monthly proportions of neck-drooping vultures were significantly correlated with mean maximum monthly temperature at Multan meteorological station (Fig. 1, $r^2 = 0.95$, $F = 195.4$, $P < 0.001$).

Vulture deaths due to diclofenac poisoning occurred in all months of the year with no seasonal pattern evident (Fig. 1), and no significant variation in the bimonthly rate of occurrence of gout (Gilbert et al. 2006). There was no correlation ($r^2 = 0.01$, $F = 0.07$, $P = 0.79$) between mean monthly deaths due to diclofenac and the monthly proportion of neck-drooping vultures (Fig. 1).

To determine whether the observed seasonal correlation in neck-drooping was correlated with ambient temperature, we assigned every vulture observation to the ambient temperature at the nearest 30-min interval using data from the HOBO® loggers. For each 1°C ambient temperature interval from 7–46°C (N = 40) we calculated the number of vultures neck-drooping as a proportion of the total number of vultures observed in each temperature interval and plotted them against ambient temperature (Fig. 2). No neck-drooping vultures were observed when ambient temperature was <17°C. Above a calculated threshold temperature of 15.4°C (95% CI: 9.9–22.5°C), there was a significant positive correlation between temperature and the proportion (expressed as an angular transformation) of vultures observed neck-drooping ($N = 30$, $r^2 = 0.90$, $F = 253.1$, $P < 0.001$) described by the equation $Y = 1.168\ X - 17.931$ where Y is the proportion (angular transformed) of neck-drooping vultures and X is ambient temperature.

Vulture posture and orientation to the sun was recorded in 30 439 cases of the total of 31 683 vulture observations, of which 4397 (14.4%) were vultures in the neck-drooping posture (Table 1). Neck-drooping vultures were more frequently observed with their back directed toward the sun (69.7%) than birds that were in a typical posture (i.e., not neck-drooping, 49.2%). Likewise, vultures that were not neck-drooping were more frequently observed facing the sun (17.6%) than those that were neck-drooping (7.3%). These differences in orientation of vultures in each posture category (neck-drooping and typical posture) were significant ($\chi^2 = 676.26$, df = 2, $P < 0.001$).

DISCUSSION

Our study represents the first attempt to critically examine the function of neck-drooping behavior in Oriental White-backed Vultures. We examined two hypotheses: (1) Neck-drooping is temperature-correlated because it may have a thermoregulatory function, and (2) Neck-drooping is indicative of poor health in a population and is a prelude to vulture death due to diclofenac poisoning.

The strong positive correlation between the proportion of vultures observed neck-drooping and ambient temperature (both inferred by seasonal trends, Fig. 1, and ambient temperature measured at the time of observation, Fig. 2), and the tendency for drooping vultures to orientate their backs toward the sun and thus droop their heads into their own shade, support the hypothesis that neck-drooping in Oriental White-backed Vultures is a behavior with a thermoregulatory function. This conclusion is consistent with behavioral and postural responses to thermoregulation recorded in other vulture species (Hatch 1970, Larochelle et al. 1982, Arad and Bernstein 1988, Arad et al. 1989, Bahat 1995, Shobrak 2001). It is probable that neck-drooping is used by vultures in high temperatures to protect the unfeathered head and neck from direct solar radiation, thus facilitating cooling. These correlations do not, by themselves, prove that thermoregulation is occurring in response to posture and orientation. Such proof would require controlled experimental testing that was beyond the scope of this study.

The potential utility of neck-drooping as an indicator of poor health and death (Prakash 1999, Cunningham et al. 2003) is of interest to vulture re-

Table 1. Orientation of neck-drooping and non-neck-drooping vultures with respect to the sun: number of individual vulture observations made and percentage of the total number of observations in each posture category.

POSTURE	BACK TO SUN (%)	FACING SUN (%)	SIDE TO SUN (%)	TOTAL
Not neck-drooping	12 808 (49.2)	4587 (17.6)	8647 (33.2)	26 042
Neck-drooping	3065 (69.7)	320 (7.3)	1012 (23.0)	4397
Total	15 873	4907	9659	30 439

searchers and conservationists worldwide. The value of any test in determining the presence of a disease or disorder is defined in terms of its specificity and sensitivity. A test with high specificity will correctly exclude those individuals that do not have the disorder (i.e., it results in few false positives), and a highly sensitive test will correctly identify those individuals that do have a condition (i.e., it results in few false negatives).

The specificity of neck-drooping as an indicator of diclofenac exposure would depend on the proportion of neck-drooping vultures that do not subsequently die (i.e., false positives). Annual mortality rates attributed to diclofenac (MD_{DV} in Table 2, Gilbert et al. 2006) ranged from 0.01 to 0.31 at the same study sites and periods as presented here, and bimonthly rates of diclofenac exposure did not vary significantly throughout the year (Gilbert et al. 2006). It has been shown experimentally that death occurs within 6 d of consuming a toxic dose of diclofenac (Oaks et al. 2004). Therefore, if we conservatively assume that all vultures that ingested a lethal dose of diclofenac survived for 6 d, and exhibited neck-drooping posture throughout that period, the daily chance of observing a diclofenac-contaminated vulture ranged from $(0.01 \times 6)/365 = 0.000164$ to $(0.31 \times 6)/365 = 0.005096$ depending on the location and period of the study. In contrast, mean monthly proportions of neck-drooping ranged from 0.0 in winter to a high value of 0.289 in the hottest month of June when neck-drooping behavior was 57 times more likely to be seen than a diclofenac contaminated vulture at even the highest measured rate of diclofenac mortality. It was therefore likely that most, if not all, of the neck-drooping vultures observed were clinically healthy, so neck-drooping predicts a high number of false positives and is of low specificity. Likewise, vultures continued to die in months when neck-drooping was not observed at all (Fig. 1), indicating that neck-drooping has a low sensitivity in detecting diclofenac exposure, and predicts a high proportion of false negatives.

We concluded that neck-drooping posture in the Oriental White-backed Vulture is not specific or sensitive enough to be used as an indicator of diclofenac poisoning, the agent responsible for the vast majority of vulture mortality and recent population decline among vultures in South Asia (Green et al. 2004, Oaks et al. 2004, Shultz et al. 2004, Gilbert et al. 2006). Given the close genetic relationship between *Gyps* species and the phylogenetic position of *Gyps bengalensis* relative to congenerics (Johnson et al. 2006), it may be reasonable to predict that neck-drooping behavior will also be insufficiently specific or sensitive as an indicator of diclofenac poisoning in all *Gyps* species. Likewise, neck-drooping may have a similar thermoregulatory function in other members of the *Gyps* genus, such as the Eurasian Griffon (*Gyps fulvus*; Camiña 2001) and the Cape Griffon (*Gyps coprotheres*; P. Benson pers. comm.).

The only known test for assessing exposure to diclofenac in vulture populations where availability of the drug is unknown is the occurrence of death with visceral gout. Testing for diclofenac poisoning should be included as a differential diagnosis whenever avian scavengers are found to have visceral gout at necropsy. Conservationists, veterinarians, and policy makers outside South Asia should be made aware of the risks posed by diclofenac to scavenging birds and remain vigilant to the introduction of diclofenac products that may have intended or unintended applications in livestock that may become available as food to avian scavengers.

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