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Occurrence of veterinary pharmaceuticals in golden eagle nestlings: Unnoticed scavenging on livestock carcasses and other potential exposure routes

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HIGHLIGHTS

- Fluoroquinolones were found at high occurrence in nestling Golden eagles.
- Diet analysis suggests an apparently low dependence on livestock carcasses.
- Occurrence and concentration of fluoroquinolones was similar to those found in vultures.
- A greater than expected reliance on livestock carrion may explain exposure to fluoroquinolones

GRAPHICAL ABSTRACT

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ABSTRACT

Wildlife exposure to pharmaceuticals can occur through contaminated water, and through the excreta and carcasses of medicated livestock, with potential for bioaccumulation and transfer through food webs. We evaluated whether nestling exposure to pharmaceuticals can occur from food delivered to nests in the golden eagle (*Aquila chrysaetos*), a top predator and facultative scavenger. Despite the fact that diet analysis suggests an apparently low dependence on livestock carcasses reduced to two piglets remains (1.5% of food remains, $n = 134$), a high proportion of nestlings (71%, $n = 7$) showed fluoroquinolone residues in plasma, mostly enrofloxacin, which is exclusively used in veterinary treatments. The occurrence and concentration (54.5 ± 6.6 μg·L^{-1}) of fluoroquinolones in plasma was similar to those found in the nestlings of three vulture species largely dependent on livestock carcasses obtained at supplementary feeding stations, which are managed for the conservation of their populations. Although the number of analysed eaglets is comparatively small, the fact that enrofloxacin was found in all nests sampled in three breeding seasons suggest an exposure to the drugs similar to that of vultures. An underestimation of the role of carrion, especially from small piglets whose consumption may have gone unnoticed, and the predation of semi-domestic prey and generalist prey exploiting carcasses of medicated livestock, can contribute to explaining the unexpectedly high occurrence of these drugs in eaglets.

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1. Introduction

Wildlife exposure to human and veterinary pharmaceuticals can occur through contaminated water, as well as the excreta and carcasses of medicated livestock (Kummerer, 2010; Arnold et al., 2014; Gothwal and Shashidhar, 2015). Livestock carcasses from factory farming are often discarded at supplementary feeding stations (SFS), which are managed for the conservation of avian scavengers (Cortés-Avizanda et al., 2016). By foraging on these carcasses, vultures may be exposed to veterinary pharmaceuticals (Blanco et al., 2016, 2017a). Many other vertebrates, including canids, rats, corvids, storks, gulls and raptors such as kites and eagles, also forage at SFS and other carcass and rubbish dumps, to a variable extent depending on species, season and regions (e.g. Donázar, 1992; Blanco, 1996; Cortés-Avizanda et al., 2012). However, little information is available on exposure to pharmaceuticals in facultative scavengers and generalist omnivorous species habitually or occasionally feeding on livestock carcasses (Friend and Franson, 1999; Sharma et al., 2014).

Multiple veterinary pharmaceuticals, including antibiotics, antiparasitics, antifungals, analgesics, hormones, etc., and their mixtures, are extensively used in livestock farming (Sarmah et al., 2006; Woodward, 2009). Fluoroquinolones, a family of synthetic broad-spectrum antimicrobials, are widely used in food-producing animals (Brown, 1996; Martinez et al., 2006; Sharma et al., 2009). The presence of antimicrobial residues in animal meats destined to human consumption constitutes a potential risk for public health by their toxicity and effects in the development of bacterial resistance, allergic hypersensitivity and alteration of the normal microbiome (Dalhoff and Shalit, 2003; Andersson and Hughes, 2014) increasing the susceptibility to fungal and other infections by opportunistic pathogens (Levy, 2002; Keeney et al., 2014). Similar negative effects can be expected in wildlife exposed to fluoroquinolones through the consumption of medicated livestock carcasses (Blanco et al., 2016, 2017a). In addition, there is a growing concern on the role of fluoroquinolones as biologically active environmental contaminants even at low concentrations (Sukul and Spiteller, 2007; Zhou et al., 2008; Van Doorslaer et al., 2014; Li et al., 2016).

Supplementary feeding of threatened avian scavengers is intended to provide benefits to their populations by enhancing individual survival and breeding success (Cortés-Avizanda et al., 2016). These practices, however, may also have unintended impacts on target and non-target species and communities when implemented without specific conservation goals supported by strict monitoring, scientific research and adaptive management (Blanco, 2006; Cortés-Avizanda et al., 2016; Blanco et al., 2017b). For instance, exposure to livestock pathogens and veterinary pharmaceuticals has been recorded in avian scavengers exploiting SFS in Spain (García-Fernández et al., 2013; Marín et al., 2014; Blanco 2015; Mateo et al., 2015; López-Rull et al., 2015; Blanco et al., 2016, 2017a). The chaotic, chronic and pulsed ingestion of fluoroquinolones throughout nestling development has been proposed as one of the most plausible explanations for the high occurrence and intensity of oral *Candida*-like lesions in nestling vultures (Blanco et al., 2016, 2017a). In addition to direct exposure from carcasses, the variable environmental persistence of pollutants facilitates their bioaccumulation/bioconcentration in prey species and biomagnification by transfer through food webs in top predators (Sergio et al., 2008).

In this study, we determined the diet of nestlings in the golden eagle (*Aquila chrysaetos*) to evaluate whether a potential exposure to pharmaceuticals can occur from food delivered at nests during the breeding season. We evaluated the occurrence of fluoroquinolone antibiotic residues in a sample of nestlings, as a preliminary assessment of exposure to veterinary pharmaceuticals in a top predator and facultative scavenger. We tested for the presence of fluoroquinolones because these antibiotics are often used in combination with other drugs in multiple treatments in livestock operations (Andriole, 2005; Sharma et al., 2009) and because they have been previously found in nestlings of three vulture species in the same area (Blanco et al., 2016, 2017a). We tested the hypothesis that the exposure risk to pharmaceuticals depends on the scavengers' reliance on medicated livestock carcass instead of wild prey (Blanco et al., 2017a). Because of the apparently low dependence of golden eagles on carcasses of livestock from factory farms available at SFS (Donázar, 1992; Cortés-Avizanda et al., 2012; Bautista et al., 2016; author's unpubl. data), we predict a low exposure to livestock pharmaceuticals in this species compared with obligate scavengers that are highly dependent on these carcasses (Blanco et al., 2016, 2017a). By analysing the composition of the nestling diet, we also considered whether exposure to fluoroquinolones could occur through the food web via eagle predation on semi-domestic medicated species, as well as on wild prey feeding on medicated livestock carcasses.

2. Material and methods

2.1. Study area and species

The study was conducted in central Spain (Segovia and northeastern Ávila provinces). This area holds a numerically important community of obligate scavengers that are highly dependent on livestock carrion at carcass dumps (Blanco, 2014). In addition, this region holds a rich community of predatory raptors often acting as facultative scavengers, including about 20 breeding pairs of golden eagles (authors' unpubl. data). This and the surrounding area have one of the highest concentrations and increasing numbers of farms devoted to fattening pigs reared under intensive conditions in Spain (Diputación de Segovia, 2006; Blanco, 2014).

The golden eagle is a large, highly territorial apex predator distributed across a variety of habitats in montane and open areas of the Northern Hemisphere, where it nests in cliffs and large trees. It is listed as 'Near Threatened' in Spain, where the largest European population resides (BirdLife International, 2015). This species suffered intense human persecution in the past, but current population trends show numerical stability in Spain, despite the fact that non-natural mortality persists due to electrocution and persecution by shooting and poisoning with baited carrion. Other threats include disturbance to nests by human activities and habitat alteration (Arroyo, 2004). In the Mediterranean region, this species predates on medium-sized vertebrates, especially lagomorphs, carnivore mammals, juvenile ungulates, corvids, pigeons and partridges, and often scavenges on carcasses of wild and domestic animals (Sánchez-Zapata et al., 2010; Bautista et al., 2016).

2.2. Nestling diet

During the breeding seasons of 2003–2016, food remains found in and below the nests were collected at the time of nestling ringing. The remains corresponded to the diet of 43 nestlings from 31 successful breeding attempts in 15 different territories. The remains were identified macroscopically and quantified assuming the smallest possible number of individual prey by considering the number, size and anatomical position of the remains of each species in each nest (Blanco, 1997; Seguin et al., 1998). Pellets found in the nests were not considered in the analyses to avoid including the diet of breeding adults. By using food remains, diet was likely biased in favor of the most durable prey remains and did not reflect the importance of smaller prey items, such as small animals and pieces of skinned carcasses and viscera of livestock and large game species (Seguin et al., 1998; Sánchez-Zapata et al., 2010). However, this may depend on pair specialization on particular prey and on whether each breeding pair cleans out their nest, especially the remains of larger prey items once exploited (Seguin et al., 1998; Moleón et al., 2002). The biomass of each food item was not estimated because of the difficulty in obtaining accurate measurements from scavenged food, particularly from livestock remains.

To assess broad differences between species in the consumption of livestock carrion, food remains were grouped as wild prey obtained by predation and scavenging (e.g. wild birds, mammals, reptiles) and