LIFE Re-Vultures International Workshop on Vultures & Veterinary Drugs

Dadia, Greece, 19-20 February 2019

Notes
1. Introduction

- Knowledge exchange to inform best practice and identify priorities
- Identify widely used veterinary products
- Review current sampling and analysis protocols
- Inform LIFE Re-Vultures After-Life plan
2. Vulture conservation in Thrace, Greece

- Maximum 35 incubating pairs of cinereous vultures, 2014-2017
  - Poisoning incidents have slowed population increase
- Griffons: 0-1 pairs in Dadia; several pairs in other SPAs
  - Increasing numbers recorded at feeding site
- 3 pairs Egyptian vultures in Dadia; rapid decline since 2011
- Antipoisoning: detection dogs; networks of shepherd dogs
- Mitigation of pylons against electrocution
- Vulture data used to inform suitable placement of wind farms
- Supplementary feeding since 1987, weekly operation; 2018: small scale sites
- Support to extensive livestock farming is required
3. Vultures in Dadia & adjacent SPAs

- Management body: government body; functioning properly since 2015
- Many different activities with multiple stakeholders
- Dadia NP + 4 SPAs: >2000 km²
- Monitoring all species and habitats of conservation importance, especially black vultures: 30-35 breeding pairs, 10-20 fledglings; tagging of nestlings;
- Satellite tracking: identify feeding, roosting sites and movement corridors > informs placement of wind farms; tool to detect poisoning incidents
- Published in annual reports - available on request
- Supplementary feeding is funded by Regional Unit of Evros (public funding)
- Key point: coordinated, cross-border actions
4. Review on vet drugs and vultures

- Start.......
5. Diclofenac in Europe - update

- Small number of treated carcasses can cause high numbers of vulture deaths, even at low doses
- Authorised in Italy since 1993, and Estonia, Czech, Latvia since 2009
- 2013 - authorized in Spain (pending in Portugal)
- 9,460-27,700 animals estimated to have been treated with diclofenac in Spain
- Potential mortality of 683-4792 griffons/year (Green & Margalida (2014))
- 2014: FATRO refused voluntary withdrawal of diclofenac
- Campaign to ban diclofenac: >50,000 signatures
- 2014: European Medicine Agency confirm risk to vultures and other species
- EMA did not directly recommend banning diclofenac
- EU position: until evidence supplied, existing legislation is sufficient
5. Diclofenac in Europe - update

- Variation in status and action plans / measures between countries
- 2017-18: organisations researching evidence of exposure to diclofenac
- 2018: renewal for diclofenac in Spain and request for sale in Portugal
- 2017-2018: new campaign to remove diclofenac
- Still legally sold in Spain, Italy (and some others) and probably will be sold in Portugal
- In principal legislation prevents diclofenac exposure, but the risk remains
  - Continued vigilance and monitoring: increase testing for presence of diclofenac and other vet drugs
6. Session 1 discussion

- Greece - known cases of use of human diclofenac on livestock
- Israel - some cases of incorrect use of e.g. flunixin in sheep instead of horses
- Spain - concerns that farmers may intentionally use diclofenac as a poison to remove vultures
- Israel - price has gone down for meloxicam due to working with suppliers
7. France - overview

- Since early 2000s, different programs
- e.g. “Vigilance Poison” > all raptors in France with national action plan
- Monitoring protocol established
- Currently delay between analysis, results and report > difficult to respond
- 3 different laboratories with different specializations; + other partnerships
- Since 2005, 41/399 autopsied birds were poisoned (excl lead)
- No analysis possible in some cases e.g. degradation / lack of funding
- Euthanizing products: long persistence in carcasses
- No diclofenac used in France
- Systematic recording of drug use in breeding register
- Mandatory reporting of wildlife mortalities with evidence of veterinary drugs
7. France - overview

- Vultures mainly in extensive livestock farming areas
- Some supplementary feeding sites are managed by farmers, feeding small ruminants with limited vet med treatments
- Not all dead birds are collected and analysed
- Aim: to focus on raptor species covered by National Action Plans e.g. LIFE
  - Set up specialized analytical centre to detect illegal activity
  - Consolidate toxic threat surveillance network
- Toxic threat mainly from pesticides, not veterinary medicines
- Necessary to investigate sublethal effects
- 400 Euros to test each bird for all veterinary products > expensive
8. Spain - overview

- Supplementary feeding is an important conservation strategy, but no/very few regulations regarding control or monitoring of veterinary drugs in carcasses.
- 2013: diclofenac authorized in Spain for veterinary use.
- Many drugs, but limited evidence for toxicity to vultures.
- Flunixin case: Maria-Mojica et al. 2018, Alicante.
- Ketoprofen and meloxicam detected in infertile BV eggs in captive breeding centre - Zorilla et al. 2018, most likely through feeding.
- No national monitoring program in Spain > MAF grant to investigate NSAIDs in vultures in Spain:
  - 160 griffon vultures, 8 cinereous, 8 bearded, 7 Egyptian tested.
  - No diclofenac but 4.4% with other NSAID residues; acute flunixin in 1 case.
8. Spain - overview

- Most causes of death are trauma, electrocution etc
- Kidney lesions do not always indicate visceral gout caused by vet drugs
- 183 livestock carcasses analysed, only 5 with NSAID residues
  - Flunixin (2), diclofenac, meloxicam, ketoprofen, all only low levels
  - Diclofenac only found at the injection site in one pig
- Antibiotics detected but at low levels. Exposure confirmed, unknown effects
- Nestlings potentially good sentinels of exposure
- Euthanasia drugs (pentobarbital) in Spain > Aldeguar et al, 2009; Mojica et al, 2017 > 5 griffons poisoned by pentobarbital from goat at feeding site
- Very limited legislation about veterinary drugs and reducing exposure for vultures / scavengers
9. Bulgaria - overview

- Procedures for authorisation of vet medicines:
  - National > Mutual Recognition > Decentralized > EU

- Four part registration dossier, including safety data, at national level > takes up to 210 days, once granted it lasts 5 years

- Wholesale of veterinary drugs must be authorized by veterinarian

- Retail only available at veterinary pharmacies, under prescription by a registered veterinary practitioner (kept in duplicate at farm and pharmacy)

- BFSA implements pharmacovigilance system and cooperates in EU program > mainly for information exchange e.g. safety data

- National laboratories have difficulties to analyse pesticides, antibiotics etc. Do not have accredited matrices for animal products. Private laboratories exist
9. Bulgaria - recommendations

- Increase public awareness, including veterinary practitioners > many unaware of the problems discussed previously i.e. sensitivity of vultures
- Establish maximum residue levels, withdrawal periods etc to inform proper legislation
- Currently would need to send a dead vulture to a private laboratory for analysis of dead animals
10. Greece - overview

● Several national laws related to veterinary medicines

● Total number of vet med packages sold (includes pets):
  ○ 2016 = 1,203,235 packages
  ○ 2017 = 993,850 packages [n.b. measured in packages not weight]

● Surveillance and control plan drawn up and implemented annually
  ○ Group A (anabolic) and Group B (antibiotics and other drugs)

● Network of state laboratories to analyse NSAID etc residues in livestock

● Very low number of non-compliance samples e.g. 12/3860 (0.3%) in 2017

● Diclofenac is not authorized but was detected in milk in 2015 > due to use of human diclofenac on a single animal

● All VPs under veterinary prescription
11. Comparison and discussion

- Is the data / information passed on to the pharmacovigilance network?
  - Varies between nations

- Are dead wild animals accepted for testing in laboratories that test for veterinary drugs in human food products?

- What are peoples’ experiences with prosecuting misuse of veterinary drugs?
  - Some cases in Spain; very difficult to prosecute
12. LIFE Re-Vultures

- Study health status of large vultures
- Blood samples from 33 *G fulvus*, 2016-17 [compared to tissue samples]
- Dadia - 45 cinereous vultures sampled
- Samples sent to Spanish laboratory (Rafa Mateo and colleagues)
- Liquid chromatography “time-of-flight” mass spectrometry
- Heavy metals, antibiotics and anti-inflammatory drugs and other substances
- No presence of antibiotics or anti-inflammatory drugs
- Organochlorine compounds present in low levels, accumulate with age
- Lead levels present but lower than reference value for lethal dose; higher in *G fulvus* than cinereous
- [VPs remain for less time in blood than tissues in live birds]
12. Vultures Back to LIFE

- 22 samples from *G fulvus* and 16 from other species: need to send for full analysis, but currently no registered cases of vet meds intoxication
- Since 2003 toxicological analysis introduced by national poison working group
- No current evidence for poisoning by veterinary drugs in Bulgaria
- But other types of poisoning (carbamates, organophosphates, rodenticides) continue e.g. Kresna Gorge, 2017, 18 vultures poisoned
- Illegal dumping of livestock carcasses is quite common > potential source of exposure to contaminants (including euthanized dogs from shelters)
13. BSPB - Egyptian vulture projects

- 36 EVs sampled from Bulgaria and Greece, 2012-13; blood samples
- Samples analysed in Spain
- Negative for pesticides, antibiotics; no sig lead intoxication
- Aspirin was detected in one sample from Greece > possibly from turkeys/chickens. No toxicity documented for wild birds
- NEW LIFE: survey on use of VPs and agriculture chemicals throughout flyway
- Harmful NSAIDS such as ketoprofen and flunixin are registered for use
- 44 farmers, 22 vets, 5 farm managers interviewed: 9/34 licensed NSAIDS frequently used (Analgin 30% solution most frequent), then ketoprofen
- Diclofenac reported used in 20 settlements, likely human diclofenac
- Treatment without vet consultation is common
13. Greece - Egyptian vulture projects

- Interviews with 22 vets and 41 livestock keepers, covering 10 SPAs
- Anti-inflammatory drugs rarely used; antibiotics when applicable; antiparasitics preventative once or twice per year
- NSAIDs: carprofen, flunixin, ketoprofen, meloxicam
13. Sardinia - under griffon wings

● Food shortage is a critical factor > activation of two centralized feeding stations and 40 farm feeding stations
  ○ ~35 tonnes livestock biomass provided in 2.5 years

● Livestock carcasses were tested and analysed for multiple substances
  ○ 9% of (54?) samples contained VP residues
  ○ Oxytetracycline and ivermectin detected

● Informal questioning of vets suggest diclofenac is rarely used (despite being legal in Italy)
14. LIFE Rupis - Douro region

- Produced protocols for taking biological samples from live + dead birds
- Live birds:
  - blood, serum, feathers etc
  - how to conserve and store the samples
- Dead birds: forensic collection of evidence
- Human safety is prioritized
- Necropsy must be completed fully to inform toxicological analysis
- Five EVs captured for tracking were tested, no residues detected (but does not mean no exposure)
- Forensic investigations processes rely on national regulations
15. European Raptor Biomonitoring Facility - protocols

- All environmental contaminants monitored in raptors as indicators of threats to human health > early warning system
- 3 key elements........
- 27 national partners across Europe
- Raptors as sentinel species e.g. detect emerging contaminant problems
- https://erbfacility.eu/
- Jovan involved in establishing sampling protocols (live and dead animals)
- Also see Eurapmon protocols
16. Baseline methodology and sampling protocols

1. Survey of livestock vets an vulture feeding station managers to estimate diclofenac and NSAIDs use, and evaluate knowledge levels of risk to scavengers
   - Most vets unaware of risk
   - Feeding station managers are aware
   - Low use of diclofenac by veterinarians

2. Evaluate diclofenac and other NSAID residues in dead vultures
   - Liver/kidney/brain/stomach content from dead vultures + necropsy + multi-organ histopathological evaluation
   - Necropsy and histology can inform toxicological analysis

3. Evaluate residues in livestock carcasses at vulture feeding stations
   - Kidney, liver and skeletal muscle samples
16. Baseline methodology and sampling protocols

- Investigating detection of drug residues in blood samples from vultures
- New project proposal: Integration of livestock farming practices into conservation of avian scavengers........
  - Compare high and low intensity farming areas with drug residues and diet
  - Evaluate prevalence of pathogens in scavengers and effects on condition and disease
  - Analysis of vet drugs in carrion and scavengers, esp NSAIDs
  - Study existing gaps in mechanisms of toxicity and metabolism of drugs in vivo and in vitro e.g. using partridges as models

- Monitoring drug residues, overall approach:
  - Assess farming practices and feeding sites
  - Analysis of carrion from different areas
  - Exposure of scavengers: dead, live etc
17. Discussion

- Florence: can we send you samples for histopathology and toxicology? Not many labs do both. Ignasi: yes, we do it.
  - Histology needs to be a fresh sample (unfrozen); then formalin; then frozen.
  - Only small pieces required for histology
- Volen: lab in SA can analyse for lead after fixing in formalin
  + lab in USA that can analyse paper after applying acetone

- What gaps, concerns, priorities etc., esp. in Bulgaria and Greece?
  - Need good connection with vets at local level > mutual benefit
  - Need to improve capacity for good necropsy and good toxicology labs
  - Funding: who pays? State or NGOs? Depends on who requests analysis?
  - Need improved protocols for sample collection, storage and analysis.
  - Dora: in Greece the necropsies are rarely completed properly, so causes remain unknown. Due to lack of capacity, funding etc.
  - Dora: We have identified the same gaps over the years, how do we move forward?
  - Can we use dogs to detect and prevent poisoning before the fact?
18. Day 2 - Recap and discussion

- Good necropsies are key > develop local capacity
- Insert recap slides here..................
- Diseases, WNV, *Toxoplasma gondii* etc
- Distinguish between exposure to contaminants and negative effects of contaminants
- Euthanasia drugs need to be included in the analysis in the Balkans
- Find different laboratories for the different analyses: necropsy, toxicology, histopathology, microbial pathogens etc.
- When handling vultures monitor and record presence/absence of oral lesions
19. Future action points

- Lists of approved drugs / substances from each country > inform analysis
- Use existing project activities to do additional monitoring e.g. oral lesions
- Need to process large numbers of samples in the Balkans. Possibility for analysis to be done in Bulgaria or for Spanish colleagues to travel there to do analysis
- Need to develop official collaborations between state labs and/or local veterinary labs to conduct the analysis
- Need to collect information from all labs in each country: which labs do which analysis
- Full analysis needs to be done to reduce number of “unknown” mortalities
- It would be useful to know the withdrawal period for certain drugs to inform deposition of carcasses at feeding sites
- Must communicate about treatments with farmers and vets before carcasses are provided at feeding sites
Thanks for listening
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Together for Vultures

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