REASONS FOR BREEDING FAILURES OF THE EGYPTIAN VULTURE *(NEOPHRON PERCNOPTERUS)* IN BULGARIA AND GREECE (2006-2015)

TECHNICAL REPORT UNDER ACTION A1

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ABOUT THE PROJECT

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Contents

1. SUMMARY

The present study aimed to investigate the reasons for breeding failures in the steeply declining population of Egyptian vulture in Bulgaria and Greece (holding ca. 50% of the Balkan population of the species), so that its results may be further used to inform a more effective and adequate conservation of the species. The study was conducted in the period 2006-2015 for Bulgaria and 2011-2015 for Greece based on the observations compiled through the implementation of the monitoring activities of Egyptian vulture breeding territories in both countries, the nest guarding programme and the use of camera devices (trail cameras) installed in selected nests.

The average percentage of unsuccessful pairs per year in Bulgaria and Greece was 38% (n = 366 breeding attempts), with 37% per year for Bulgaria (for a period of 10 years) and 48% for Greece (for a period of 5 years). In total, 54% of unsuccessful pairs (n = 132) did not initiate breeding attempt at all, while 46% initiated a breeding attempt but failed in different stages of the breeding period. In 54% (n = 61) reasons for breeding failure remained unknown, for 26 % the reason for breeding failure was evidenced, and for 20% the reason was suspected based on expert's opinion. For both evidenced (n = 16) and suspected (n = 12) causes for breeding failure, natural causes were much more frequent than human-induced causes. In the case of known causes of breeding failure (n = 16), natural drivers were represented by lack of experience in pairs (25%), predation (25%), diseases (12.5%) and weather conditions (6%), while in the case of human-induced causes they were represented by persecution (19%; registered only in Bulgaria and aiming at nest robbing and taxidermy) and poisoning (12.5%). In terms of the period, ca. 60% of the failures occur during incubation stage. In Bulgaria (2006-2015), the lack of experience in pairs was the most frequent natural cause (40%, n = 10), followed by diseases (20%) and bad weather conditions (10%), while persecution (30%) was the most frequent human-induced cause for breeding failure. In Greece (2011-2015), most of the evidence collected (67%; n = 6) was related to natural causes for breeding failure (predation of chicks), while only 33% referred to human-induced causes (mainly poisoning of adults).

Considering the high proportion of cases with unknown causes of breeding failures, further research with more robust methodology is needed. Appropriate measures to reduce the rate of breeding failures are suggested, such as securing the nests and supplementary feeding programmes.

2. INTRODUCTION

One of the main objectives of ecology is to reveal and understand the processes that shape population size and structure, a matter that is a subject of extensive study (Begon *et al.* 1996 , Levin *et al.*2009).The breeding performance parameters of raptors are crucial for understanding the health and condition of populations and the degree to which they are likely to persist in space and time (Newton 1979; Steenhoff and Newton 2007). Main factors influencing raptor's breeding parameters could be both natural and human induced such as the availability and quality of food and suitable breeding sites, the weather conditions, inter- and intraspecific competition, the age and experience of the partners and human pressure (Newton 1979; Newton 1994).

Many raptor species around the world are declining, and avian scavengers like vultures are among the most threatened raptor species (Thiollay 2006; Virani *et al.* 2011; Chaudhary *et al.* 2012; Ogada *et al.* 2012). Hence, in small and isolated populations of avian scavengers, understanding the factors affecting the breeding performance parameters such as productivity and breeding success and breeding failures on the other hand could play a key role in conservation of the species' populations (Liberatori & Penteriani 2001; Velevski *et al.* 2014).

The Egyptian vulture (*Neophron percnopterus*, Linnaeus 1758) is an opportunistic medium-sized scavenger distributed throughout southern Europe, North, West, Central and East Africa, the Middle East, Transcaucasia, Central Asia and the Indian subcontinent (Ferguson-Lees & Christie 2001). Because of consistent and steep decline throughout its range, in 2007 the species was up listed from Least Concern to Endangered in the IUCN Red List (Birdlife International 2008). The species is declining almost everywhere across its range with one of the steepest declines observed on the Balkans where the population is declining at a rate of ~7% per year for several decades; however the underlying causes of the observed rapid population decline are still poorly known (Velevski *et al.* 2015). Better understanding of the factors affecting the demographic parameters of the species on the Balkan peninsula is required for the implementation of successful conservation schemes. However, knowledge on the breeding performance and the breeding failures of the Egyptian vulture in the Balkans is scarce and insufficient (Baumgart 1991).

With the present study we aim to investigate the reasons for breeding failures in the Egyptian vulture population in Bulgaria and Greece (holding ca. 50% of the Balkan population of the species, Velevski *et al.* 2015), so that its results may be further used to inform a more effective and adequate conservation of the species.

3. METHODS

3.1. Egyptian vulture territory monitoring

To understand which Egyptian vulture pairs were successful and which not, we used the monitoring data (***Figure 1***) in the period 2006-2015 for Bulgaria and 2011-2015 for Greece (***Table 1***). Egyptian vulture nests were regularly monitored every year under the study with several visits to a breeding territory in each breeding season to confirm if the territory was occupied, to locate the exact nesting place and to record the breeding output or failure. A territory was considered occupied if a pair or a single bird was observed displaying, marking territory or building a nest in the beginning of the breeding season (end of March - beginning of April). All territories were visited again in May to confirm in which of the occupied territories the incubation had started, and in which territories the pairs had failed the breeding attempt and/or had abandoned the territory. All territories were checked at least once more in June- July to monitor the breeding success and the last visit was made during the post-fledgling period in August to confirm the number of successfully fledged juveniles.

Information on breeding failures was also regularly and timely provided in the frame of the nest-guarding programme implemented under the LIFE project in the period 2012-2015 (Dobrev *et al.* 2016c).

Table 1. *Number of monitored Egyptian vulture pairs in Bulgaria and Greece (2006-2015).*

Year	Bulgaria	Greece	Total
2006	40	0	40
2007	39	0	39
2008	34	0	34
2009	32	0	32
2010	33	0	33
2011	32	9*	41
2012	29	11	40
2013	26	12	38
2014	25	9	34
2015	27	8	35

** Data restricted to the territories in the wider area of Dadia.*

Figure 1. *Monitoring of Egyptian vulture breeding territories. Photo: V. Dobrev*

3.2. Investigation of breeding failures

The start of incubation of a pair was considered as a breeding attempt regardless of the outcome. During the monitoring the Egyptian vulture territories, breeding failures were also recorded and the following scenarios for unsuccessful pairs were considered:

- Pairs that had occupied a territory but didn't start a breeding attempt;
- Pairs that had initiated a breeding attempt in the beginning of the breeding season and that had failed at any given stage of the breeding period.

Two main approaches were used to determine the reason for the breeding failure:

1. *Collect evidence:*

- By entering the nest (***Figure 2***) – this was made in case of recent breeding failure (on the same day or up to 4-5 days after the event). Some of the evidence collected (eggs, dead chicks) were sent for analysis in specialized laboratories;
- By using trail camera installed in the nest (***Figure 3***) - in some of the accessible nests with appropriate niches, trail cameras were installed to study in detail the breeding of the Egyptian vultures. These devices helped to collect direct and indirect evidence for the reasons for breeding failures. Trail cameras were installed before the start of the breeding season, camouflaged with natural materials (such as small rocks and leaves or sometimes wool) and the infra-red sensor was switched off to avoid disturbance. Cameras were set to take a picture every 30 - 40 seconds. These settings allow the life span of a battery pack to continue up to two months when it was changed together with the memory card. This approach allowed to investigate breeding failures and also provided visual evidence of different moments during

the breeding cycle of the birds which is impossible to obtain in other way (copulation behaviour, nest building, food provision, and other aspects from the biology of the birds).

2. *Assumptions based on expert's opinion* – in those cases when the breeding failure was discovered at a later stage based on the visual observations during the monitoring programme. In such cases nests were not entered as it was considered unlikely to get reliable diagnosis from samples that are not fresh but also to avoid creating unnecessary disturbance on the vultures.

The following categories for breeding failures were used:

- Natural causes: (1) lack of experience in the pair (when there is a replacement of a bird in the pair, or the pair is newly formed, it is often that the pair failed in breeding due to lack of experience (Grande *et al.* 2009); (2) nest predation; (3) diseases and (4) bad weather conditions.
- Human-induced causes: adult mortality related to poisoning, persecution, electrocution, etc.

Figure 2. *Entering Egyptian vulture nest. Photo: V. Saravia*

Figure 3. *Installation of trail camera in a nest of Egyptian vulture. Photo: V. Dobrev*

4. RESULTS

4.1. Unsuccessful pairs

The average percentage of unsuccessful pairs per year (2006-2015) in Bulgaria and Greece was 38% (n = 366 breeding attempts), with 37% per year for Bulgaria (n = 317 breeding attempts for a period 10 years) and 48% for Greece (n = 49 breeding attempts for a period of 5 years). Over years, this percentage slightly decreased in Bulgaria, but increased in Greece (***Figure 4***).

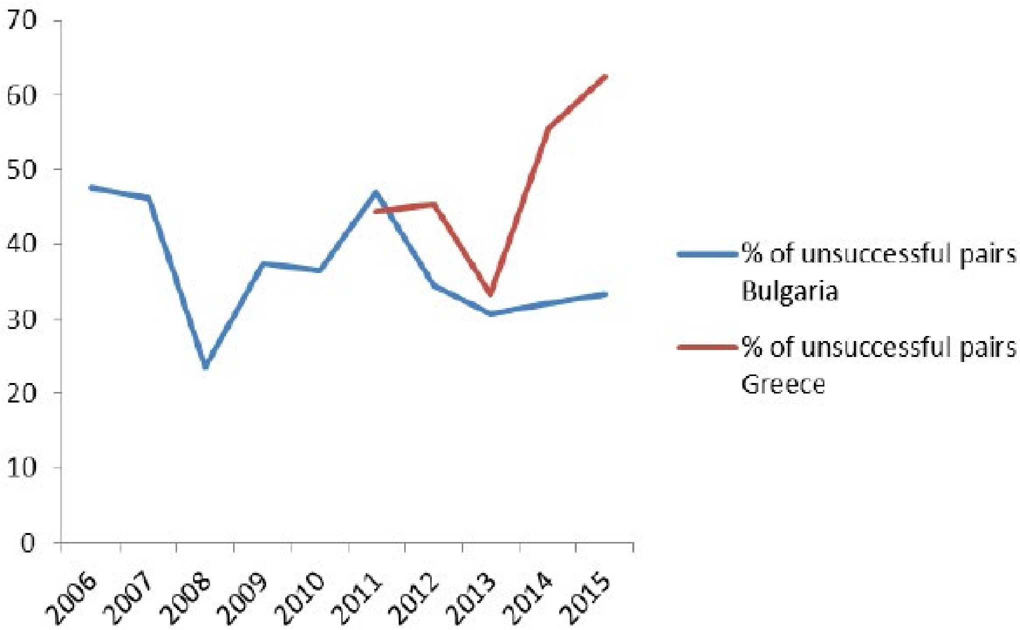

Figure 4. *Trend in the percentage of unsuccessful Egyptian vulture pairs in Bulgaria (2006-2015) and Greece (2011-2015).*

In total, 54% of unsuccessful pairs (n = 132) did not initiate a breeding attempt at all, while 46% initiated a breeding attempt but failed in different stages of the breeding period. The proportion of pairs that did not start breeding at all was higher in Bulgaria (58%) than in Greece (39%). Of all 36 breeding failures observed during the LIFE project (2012-2015), 53% were registered in Bulgaria and 47% in Greece. The percentage of pairs that failed breeding in Bulgaria ranged between 6% and 25% and between 11% and 63% in Greece throughout the study period.

4.2. Causes for breeding failure

4.2.1. Nature of the drivers

In 54% (n = 61) reasons for breeding failure remained unknown, for 26 % the reason for breeding failure was evidenced, and for 20% the reason was suspected based on expert's opinion (***Figure 5***).

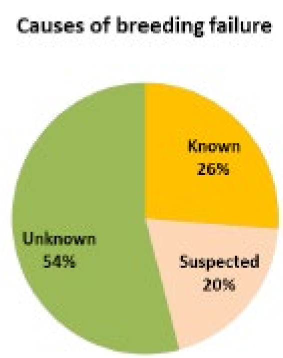

Figure 5. *Level of knowledge on breeding failures (n = 61) in Egyptian vultures in both Bulgaria and Greece (2006-2015).*

For both evidenced (n = 16) and suspected (n = 12) causes for breeding failure, natural causes were much more frequent than human-induced causes (***Figure 6***).

Figure 6. *Nature of the drivers for known (n = 16) and suspected (n = 12) causes of breeding failures in both Bulgaria and Greece (2006-2015).*

In known causes of breeding failure (n = 16), natural drivers were represented by lack of experience in pairs (25%; ***Figure 7***), predation (25%; nest predation by an Eagle owl and foxes were evidenced but other potential predators such as wild cat and rock martin were also recorded close to the nests by the trails cameras - ***Figure 8***), diseases (12.5%; avian pox and sepsis) and weather conditions (6%; a cold weather spell during the incubation period and a summer storm during the fledgling period), while human-induced causes were represented mainly by persecution (19%; registered only in Bulgaria and aiming at nest robbing and taxidermy) and poisoning (12.5%) (***Figure 9***).

Amongst the suspected reasons for a breeding failure (n = 12), the lack of experience of pairs was the most frequent natural cause (50%), followed by bad weather conditions (8.3%) and diseases (8.3%), while human-induced causes were represented by poisoning only (33%) (***Figure 9***).

Alternatively, in terms of the stage of reproductive cycle, most (59%; n = 27 cases) of the breeding failures happened during incubation, 33% - during chick rearing, and 8% during fledgling stage (***Figure 10***). Moreover, about 32% of evidenced causes (n = 16) were related to adult mortality.

A ***B***

Figure 7. *Shots from the trail cameras installed in the nests:* ***A*** *- abandoned nest after a breeding failure;* ***B*** *– breeding failure due to unfertilized eggs.*

Wild Cat

Rock Martin

Fox

Eagle Owl

Figure 8. *Predators recorded around Egyptian vulture nests (photos taken with trail cameras).*

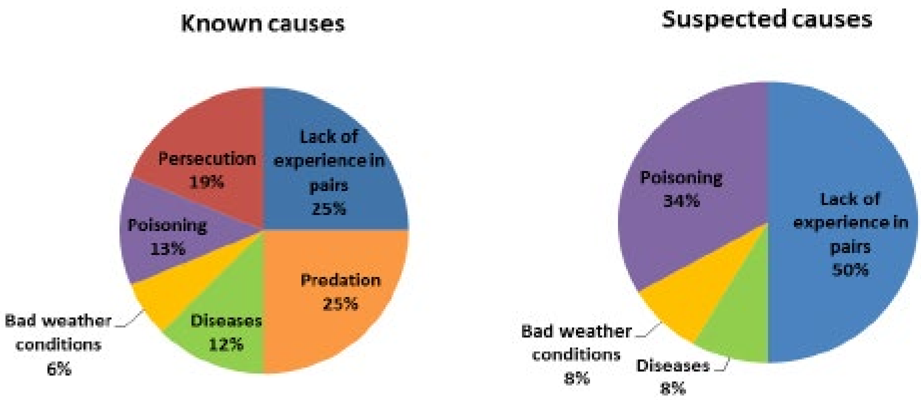

Figure 9. *Nature of known (n = 16) and suspected drivers (n = 12) for breeding failures in Egyptian vultures in both Bulgaria and Greece (2006-2015).*

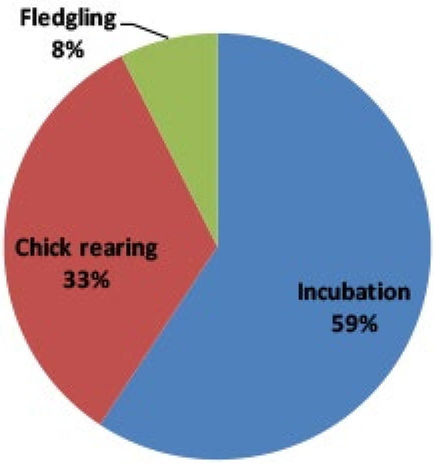

Figure 10. *Stages of the breeding cycle when failure was detected in both Bulgaria and Greece (2006-2015).*

4.2.2. Analysis per country

- **Bulgaria**

In 59% breeding failures in Bulgaria (n = 44) between 2006 and 2015, causes remained unknown. In 23% of the cases causes were evidenced, and in 18% causes were suspected.

Amongst the evidenced causes (n = 10), natural causes (70%) dominated over human-induced causes (30%). The lack of experience in pairs was the most frequent natural cause (40%), followed by diseases (20%) and bad weather conditions (10%). Human-induced causes were represented by persecution (30%) (***Figure 11***).

Amongst the suspected reasons for a breeding failure (n = 8), natural causes (62%) predominated over human-induced causes (38%). The lack of experience in pairs was the most frequent natural cause (50%), followed by bad weather conditions (12%). Human-induced causes were represented by poisoning (38%) (***Figure 11***).

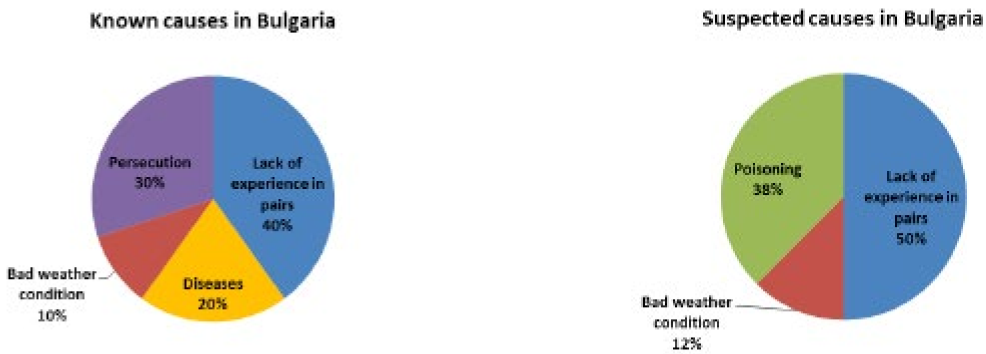

Figure 11. *Known and suspected causes for breeding failures in Bulgaria (2006-2015)*

In 41% of breeding failures in Greece (n = 17) between 2011 and 2015 causes are unknown, in 35% of the cases causes were evidenced and in 24% causes were speculated.

Most (67%) of the evidence collected (n = 6) was related to natural causes for breeding failure (predation of chicks), and 33% referred to human-induced causes (poisoning of adults). Similar were the results based on suspected causes of breeding failure (n = 4): natural causes were suspected in 75% of the cases (50% related to lack of experience in pairs and 25% due to diseases), and human-induced causes suspected in 25% (poisoning) (***Figure 12***).

Figure 12. *Known and suspected causes for breeding failures in Greece (2011-2015).*

4.2.3. Analysis over time

There were fewer cases (n = 25) of breeding failures before the LIFE project (2006-2011) compared to the number of cases (n = 36) identified during the period of the LIFE project implementation (2012-2015). This difference probably resulted from the increase of the scale and intensity of investigation on the causes for breeding failure during the LIFE+ project, and thus the proportion of the cases of breeding failures with unknown causes decreased from 76% to 39% between the two periods. The number of evidenced causes for breeding failures also increased from 24% before the LIFE project to 61% during the LIFE project.

5. DISCUSSION

5.1. Unsuccessful pairs

We found that almost half of the unsuccessful pairs in the overall studied area (Bulgaria and Greece) failed in the breeding (46%), while the other half (54%) did not initiate breeding attempt at all. Analysis per country showed that the percentage of unsuccessful Egyptian vulture pairs is lower in Bulgaria and higher in Greece compared to other countries such as Italy (Liberatori & Penteriani 2001), France (Kobierzycki 2012) and Spain (Del Moral 2009). However it is known that Egyptian vulture breeding parameters in Greece (Vlachos *et al.* 1998, Kret 2013a,b) are similar to the mean values for the European population (Iñigo *et al.* 2008).

Demographic parameters depend on different factors which have a complex impact on the populations (Newton 1979). The population in the Balkans is undergoing a steep decline and now remains gathered in several clusters (Velevski *et al.* 2015). The population is decreasing mainly due to increased adult mortality, leading to shortage of experienced adult breeders. High quality territories are known to persist longer over time as they offer better conditions for reproduction and thus are more attractive. They attract the "surplus" non-breeding adults which are recruited into the breeding population by replacing dead partners in the pairs and breed themselves. Such newly formed pairs may initiate breeding attempt but due to the low experience of the partners they often fail. This results in a lower overall breeding performance and increases ratio of breeding failures (Grande *et al.* 2009).

We also found that about 60% of breeding failures occured during incubation which is consistent with other studies (Liberatori & Penteriani 2001). The incubation period is a crucial part of the reproductive cycle in birds of prey and it is also the most hazardous because during incubation birds and clutch are mostly vulnerable to disturbance and bad weather conditions which may cause cooling down the clutch and a breeding failure (Newton 1979, Newton 1994, Liberatori & Penteriani 2001, Carrete *et al.* 2007).

Even though robust studies on the Egyptian vulture population on the Balkans were done (e.g. Velevski *et al.* 2015, Dobrev *et al.* 2016a), it is still challenging to understand some aspects of the breeding performance such as the reasons leading to a breeding failure. Hence, only in 26% of the breeding failures during the last 10 years the causes were evidenced.

5.2. Causes for breeding failures

Larger scale and more intensive systmetic monitoring in the last decade and the use of trail cameras in the nests allowed to substantially improve the knowledge on the reasons for a breeding failures (Velevski *et al.* 2015, this study). The nest-guarding programme implemented under the LIFE project (Dobrev *et al.* 2016c) had also a key role for timely identifying problems in the nests and collecting evidence.

Natural causes predominated in both evidenced and suspected causes of breeding failures in Bulgaria and Greece. The level of experience of the pair was the most frequent natural cause for a breeding failure in our study. Assessing the level of experience of the breeders and the consequences for the breeding parameters is quite challenging and subjective. Nevertheless, it is known that the level of experience in newly formed pairs where the breeders are still not well experienced can affect the breeding parameters and increase the ratio of breeding failures (Newton 1979, Grande *et al.* 2009).

Nest predation was evidenced as another frequent natural factor causing breeding failure. The Egyptian vulture breeds in cracks in cliffs, or on cliff edges, very often occupying nest places accessible to predators or raptors (Fergusson-Lees & Christie 2001). This is especially valid for places where Egyptian vultures share the same breeding habitat and many competitors are available, and thus the risk of predation is higher. Identified predation on the Egyptian vulture by eagle owl and foxes is already known for other areas (Donazar & Ceballos 1988, Stoyanova & Stefanov 1993) and is more likely to appear in relatively accessible nesting cliffs (Tella & Manosa 1993, Nikolov *et al.* 2013).

The Egyptian vulture is a medium sized opportunistic scavenger and it feeds on carcasses and excrement of vertebrates, human refuse, eggs, invertebrates and small vertebrates that are captured alive (Cramp & Simmons, 1980, Ceballos & Donázar 1990, Nergo *et al.* 2002). Thus, breeding failures due to bad health status associated with antibiotic residues, primarily quinolones, and severe disease due to bacterial, virulent or fungal pathogens could be speculated. Indeed evidence exists for diverse pathogens found in Egyptian vultures in Bulgaria and Greece, such as gumboro disease, salmonellosis, trichomoniasis (BSPB unpubl. data), as well as low concentrations of Newcastle, Avian adenovirus and Avian circovirus (Andevski & Delgado 2015). Intoxication with antibiotics (oxytetraciclines and amoxiline) has been also recorded (BSPB unpubl. data) as well as a very small insignificant amount of Aspirin (Acetylsalicylic acid) (Andevski & Delgado 2015). Andevski and Delgado (2015) conclude that studied individuals (mainly juvenile birds) had only a contact with these pathogens (probably very common), but for sure were not suffering disease, and wildlife disease and intoxication with heavy metals, and with toxic compounds from agriculture or veterinarian practices may not be a significant threat to Egyptian vultures in Bulgaria and Greece. However based on the results from the present study we recommend conducting a research on the health status of adult birds with a larger sample size in both Bulgaria and Greece.

The reason for breeding failure due to unfavourable weather conditions was detected in only two cases. Unfavourable weather conditions during the incubation or the chick rearing period can cause breeding failure due to fluctuations in the temperature which can affect the eggs or the young chicks, or to create conditions that prevent regular food provision for the chick, especially in the first days after the eggs are hatched (Zuberogoitia *et al.* 2014).

In terms of the human-induced causes of breeding failure, poisoning and direct persecution were the most frequently detected. Poisoning is supposed to be the main cause for decline in vulture populations accross the world (Hernández and Margalida 2008; 2009; Mateo-Tomás *et al.* 2012; Galligan *et al.* 2014). Hence it can also be a major reason for a breeding failure when it comes to intentional poisoning which seems to be a major threat for the Egyptian vulture in Bulgaria and Greece (Skartsi *et al.* 2014; Saravia *et al.* 2016; Kret *et al.* 2016). Intentional poisoning with poison baits affects mainly adult birds but may also affect juveniles which consume the food brought to the nest (Kret *et al.* 2015). Poisoning is also the main driver for population decline in the Egyptian vulture in its wintering grounds and other vulture species accross Africa (Ogada *et al.* 2015).

Direct persecution is most probably underestimated in the Balkan countries due to the lack of data and cases discovered. All three observed and revealed cases happened in the north of Bulgaria, and were linked either with direct persecution by poachers and taxidermists or with nest robbers. Direct persecution is a serious threat not only in the breeding grounds where it can cause a breeding failure or territory abandonment but also in wintering areas of the species (Thiollay 2006; Margalida *et al.* 2008, Nikolov 2014, Oppel *et al.* 2015).

5.3. Implications for conservation

Productivity has been predicted to have a small effect on the Egyptian population viability in the Balkans (Velevski *et al.* 2014), but increasing this demographic parameter along with increasing adult survival may play an important role for improving the status of the population. Most of the known reasons for breeding failures could be partly mitigated with appropriate conservation measures and thus increase the productivity:

- *Securing the nests* – breeding failures during the incubation or fledgling period could be potentially decreased by applying nest-guarding schemes (Dobrev *et al.* 2016b), at least in terms of failures due to nest robbing, direct persecution around the nest, disturbance or chicks fallen from nests (due to unexperienced fist flights or bad weather conditions). Nest predation by terrestrial carnivores (foxes, rock martins, etc.) could be prevented by use of electric fences or other devices that would keep away carnivores from the vulnerable nests. Communication programmes aiming to raise the awareness of local communities towards the conservation of the species could also play a positive role to reduce the risk of disturbance during the breeding period.

- *Supplementary feeding schemes* – providing Egyptian vulture pairs with safe food (Dobrev & Stoychev 2013, Dobrev *et al.* 2016b) could potentially reduce the risk of consuming a poison bait and contaminated food, and thus decrease the rate of breeding failures. In some extent this measure is also related to the health status of the species (i.e. diseases and contaminants in the body) as with the supplementary food the level of contaminants and pathogens in the food could be reduced to some extent.

Although these measures should be applied in large scale and long term in the breeding grounds to have any significant effect on the population, sometimes they are more feasible for implementation and control than securing the survival of birds along the flyway or in the wintering grounds (Arkumarev *et al.* 2015, Oppel *et al.* 2015).

5.4. Lessons learned

As for most of breeding failures causes remain unknown, further investigation is needed. In this line, we would like to share several recommendations for efficient detection and identification of breeding failures that could be considered in future research in this field:

- *Monitoring schemes* – regular monitoring of each Egyptian vulture pair is crucial for the timely detection of a problem (missing adult bird or two, missing or dead eggs and chicks, etc.) and thus timely and adequate intervention could be conducted (e.g. collect evidence for analysis – eggs or egg shells, dead chicks or parents). Setting a scheme for individual identification of vultures could improve the knowledge for the rate of partner replacements in the pairs.

- *Photo and video devices for nest observation* – video-observation in the nest provides opportunity to observe vultures distantly in real time within the nest and observe even small details of its behavior or potential problem related to breeding failure. A disadvantage is that this method requires expensive equipment and installation, energy provision and transmission of signal, and which should also be very carefully considered to avoid disturbance on vultures (Dobrev *et al.* 2016c). Trail cameras are also effective in collecting evidence on breeding failure and they are less expensive, but the disadvantage is that they provide the evidences for problems in the nest post-factum and do not provide the opportunity to take timely adequate actions to prevent breeding failure.
- *Collection of samples (evidence)* – dead eggs, chicks, adults or their remains should be collected timely and in a standardized way, along with photographs taken in situ, samples should be stored appropriately and send promptly to a reliable laboratory for analysis and diagnosis.

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