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Long-term changes in the diet of Bonelli's eagle (Aquila fasciata) in Sicily, Italy, during the breeding period

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Complete List of Authors:	Di Vittorio, Massimiliano ; Ecologia Applicata Italia srl Lo Valvo, Mario; University of Palermo Di Trapani, Eduardo; Cooperativa Silene; Gruppo Tutela Rapaci Sicilia Sanguinetti, A.; Via E.Notabartolo 60 Ciaccio , Andrea; Gruppo Tutela Rapaci Sicilia Grenci, Salvatore; Gruppo Tutela Rapaci Sicilia Zafarana, M.; Gruppo Tutela Rapaci Sicilia Giacalone , G.; Cooperativa Silene; Gruppo Tutela Rapaci Sicilia Patti, Nino; Gruppo Tutela Rapaci Sicilia Cacopardi, S.; Gruppo Tutela Rapaci Sicilia Rannisi, P.; Gruppo Tutela Rapaci Sicilia Scuderi, A.; Gruppo Tutela Rapaci Sicilia Luiselli, Luca; Institute for Development, Ecology, Conservation and Cooperation La Grua, G.; Gruppo Tutela Rapaci Sicilia Cortone, Giuseppe; Ecologia Applicata Italia srl Merlino, Stefania; Gruppo Tutela Rapaci Sicilia Falci, Amedeo; Gruppo Tutela Rapaci Sicilia Spinella, G.; Gruppo Tutela Rapaci Sicilia López-López, Pascual; University of Valencia, Cavanilles Institute of Biodiversity and Evolutionary Biology
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We analysed the diet of Bonelli's eagle (*Aquila fasciata*) during the breeding season between 2011 and 2017 in Sicily (Italy) and compared to data collected between 1993 and 1998 in the same study area. Birds and mammals were the most important prey in terms of frequency and biomass, respectively. We found significant differences in diet composition between periods probably due to a generalized decrease in the availability of wild rabbits. Considering the critical status of Bonelli's eagle in Italy, measures aimed at increasing populations of its main prey should be promoted.

Long-term changes in the diet of Bonelli's eagle (*Aquila fasciata*) in Sicily, Italy, during the breeding period

Di Vittorio M.[1-5]*, Lo Valvo M.[2], Di Trapani, E.[3-5], Sanguinetti A.[4], Ciaccio A.[5], Grenci S.[5], Zafarana, M.[5], Giacalone, G.[3-5], Patti, N.[5], Cacopardi, S.[5], Rannisi, P.[5], Scuderi A.[5], Luiselli, L.[6], La Grua, G.[5], Cortone, G.[1], Merlino, S.[5], Falci A.[5], Spinella, G.[5]. López-López, P.[7]

Affiliations:

[1] Ecologia Applicata Italia, Via Jevolella 2, Termini Imerese, (PA). Italy

[2] Dipartimento di Scienze e Tecnologie Biologiche, Chimiche e Farmaceutiche. Laboratorio di Zoologia applicata. University of Palermo, Via Archirafi 18 - I 90123 Palermo.

[3] Cooperativa Silene, via Dondes Regio 8/ a 90127 Palermo, Sicily, Italy.

[4] Via E.Notabartolo 60. Palermo. Italy

[5] Gruppo Tutela Rapaci Sicilia, Pedara, CT, Italy

[6]IDECC – Institute for Development, Ecology, Conservation and Cooperation, via G. Tomasi di Lampedusa 33, I-00144 Rome, Italy and Niger Delta Ecology and Biodiversity Conservation Unit, Department of Applied and Environmental Biology, Rivers State University of Science and Technology, PMB 5080, Port Harcourt, Rivers State, Nigeria.

[7]Cavanilles Institute of Biodiversity and Evolutionary Biology, Terrestrial Vertebrates Group, University of Valencia, C/ Catedrático José Beltrán 2, E-46980 Paterna, Valencia, Spain.

* Contact: Pascual.Lopez@uv.es

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Abstract

Context: Dietary analyses are essential to achieve a better understanding of animals' ecology. In the case of endangered species, assessing dietary requirements is crucial to improving their management and conservation. The Bonelli's eagle (*Aquila fasciata*) has experienced a severe decline throughout its breeding range in Europe and, in Italy, less than 50 pairs still remain in Sicily. This species is subject of major threats including changes in landscape composition and consequently, prey availability.

Aims: To provide current data on the diet of the Bonelli's eagle in Sicily during the breeding period and to examine dietary shifts with regards to previous studies. To discuss possible implications for conservation of the Italian population of this endangered species.

Methods: We used a combination of three methods including pellet analysis, collection of prey remains, and camera-traps imagery installed on nests, to examine the diet of 12 breeding pairs of Bonelli's eagle from 2011 – 2017. We compared this information with data collected between 1993 and 1998 in the same study area.

Key results: In number, birds were the most frequently preyed items (61.6%), followed by mammals (36.88%) and reptiles (1.52%). However, in terms of biomass, mammals were the main prey (65.71%), followed by birds (34.12%) and reptiles (0.17%). There was a decrease over the course of the current decade in the consumption of European wild rabbit (*Oryctolagus cuniculus*) which was compensated for with an increase in both dietary diversity and breadth in bird consumption; a trend not observed in the earlier study in the same region.

Conclusions: We found significant differences in terms of frequency of occurrence, percentage of biomass, dietary diversity and dietary breadth in a species at risk.

Implications: Our results indicate that changes in prey abundance are linked to shifts in diet, which may be contributing to population declines in the Bonelli's eagle population in Sicily. Measures aimed at increasing main dietary prey should be promoted.

Key-words: camera-trap; conservation; dietary composition; food; pellets.

Detailed knowledge of animals' diet is a crucial step in understanding their ecology (Newton 1979) and consequently this information can help to improve their management and conservation. Despite their general importance, diet studies of threatened species are constrained by conflicting evidence on robustness of the methodology. Direct observations of hunting behaviour, analysis of pellets, and collection of prey remains are methods usually employed to study avian diet (e.g., Mersmann et al. 1992; Lewis et al. 2004; López-López et al. 2009). However, these methods may give conflicting results. For example, the exclusive use of pellets tends to overestimate small prey (Real 1996), whereas the use of prey remains tends to overestimate large prey (Rosenberg and Cooper 1990). For this reason, some authors recommend the combined use of different methods (i.e., analysis of pellets and prey remains; Oro and Tella 1995) as well as trail camera imagery if possible (e.g., López-López and Urios 2010; García-Salgado et al. 2015) in order to provide the most accurate picture of animals' diet.

The Bonelli's eagle (*Aquila fasciata*) is a resident threatened raptor with western Palaearctic populations ranging across the circum-Mediterranean area and southern Europe (Ferguson-Lees & Christie, 2001). Starting by the second half of the 20th century, this species has decreased sharply throughout its European range due to habitat modification due to rural abandonment and changes in agricultural practices, which have caused changes in prey availability. In parallel, mortality also increased, mainly due to direct persecution (i.e., poaching, illegal harvest) and electrocution on electric pylons (Ontiveros et al. 2004; Birdlife International, 2016; Di Vittorio et al. 2018). The European population is estimated at 1100-1200 breeding pairs with 80-90% of them located in the Iberian Peninsula (BirdLife International 2016). In Italy, the Bonelli's eagle was historically present in Sardinia and Sicily, and irregularly in the southern Apennines (Cortone & Mirabelli 1987). Currently, it breeds only in Sicily (Di Vittorio et al. 2012; López-López et al. 2012), with an estimated population of 44 breeding pairs (Di Vittorio et al. 2018) and, because of low population size and reduced distribution, is currently listed as critically endangered in Italy (Rondinini et al. 2013).

Bonelli's eagle diet has been studied in Western Europe since the 1980s (see e.g. Cheylan 1977; Simeon & Wilhelm 1988; Real 1996; Di Vittorio et al. 2001; Moleón et al. 2009, 2012). This

raptor preys upon a wide range of species including mammals, birds and reptiles, usually in relation to habitat suitability and prey availability (Cheylan 1977; Simeon and Wilhelm, 1988). In southern Europe, European wild rabbits (*Oryctolagus cuniculus*) and pigeons (*Columba* spp.) represent its main prey, particularly during the breeding season (Gil-Sánchez et al. 1998; Resano-Mayor et al. 2016; Rollan et al. 2016). However, wild rabbits, a keystone species in Mediterranean landscapes (Villafuerte et al. 1995), have substantially declined in several parts of its current range in Europe. Consequently, it has been classified as a near-threatened species on the Red List of Italian Vertebrates (Rondinini et al. 2013; http://www.iucn.it/) even though it is also considered an agricultural pest species in many areas (Lo Valvo et al. 2017). This could have major impact on its main predators, including the scarce Bonelli's eagle (López-López et al., 2012).

In this paper, we aimed to examine potential shifts the diet of the endangered Bonelli's eagle in Sicily by comparing data from two different decades (1990's vs 2010's), across a variety of diet estimation methodologies, to determine if prey availability might be impacting population trends for this region. We also discuss possible implications for conservation of the Italian population of this endangered species.

Methods

Data was collected across 12 breeding pairs from February-May each year (corresponding with the breeding season) from 2011 – 2014 and 2016 – 2017. We collected pellet, prey remains in the nests and below usual perches, and photographs from camera-traps installed on nests but ensured that we avoided multiple counts of the same prey by only using one method at any given time at a particular nest site.

Regurgitated pellets were collected from nest sites and below usually used perches for five breeding pairs. Pellets and prey remains searches were conducted throughout the breeding period, but different amounts of material were collected from each nesting area due to varying nest accessibility. Pellets were stored individually in plastic bags and dried prior to laboratory analysis (Marti 1987). For the identification of prey species contained in each pellet, we applied a

comparison with feathers, hairs and bones collections at the Department of Animal Biology of the University of Palermo (Italy), applying a standard methodology (see Litvaitis 2000; Milchev et al. 2012) as well as the use of specialized guides (Desse et al. 1986, Cohen & Serjeantson 1986). Prey remains were reconstructed to estimate the minimal number of individuals of each species to avoid over-representation biases (Real 1996; Milchev et al. 2012). In addition, we also analysed images obtained by six camera-traps and one webcam which collected data from mid-April-June, located at the other seven nest sites.

To facilitate comparison of our results with other studies, we calculated prey diversity and dietary breadth. Prey diversity was calculated using the Gini index of diversity (Gotelli and Ellison 2004). This index is valuable for comparison as, unlike other commonly used indexes of diversity (e.g., the Shannon-Weiner index), it does not confound species richness and evenness and it does not depend on sample size (Gotelli and Graves 1996; review in Magurran 2003). The index was computed at species level and similarly to other indexes, the higher the index value, the higher the diversity measure (Gotelli and Graves 1996). Dietary breadth was calculated using the methods in Steenhof and Kochert (1985). This value is similar to the Gini prey diversity index, but in this case prey items are grouped by taxa higher than the species level (family level in our case). Values for this index range from 1 to ∞.

To test for statistical differences in prey composition among years we used a Kruskal-Wallis test and Monte Carlo randomizations (9999 simulations) (Gotelli and Graves 1996). To test for differences in the frequency and percentage of biomass contributions in the dietary habits of Bonelli's eagles between the current time (i.e., hereafter referred to as "current" data) and the 1990s (data from 1993 – 1998; Di Vittorio et al. 2001; hereafter referred to as "previous"), we used a Mann-Whitney U test with 9999 random permutations of the original raw data implemented in Ecosim software (Gotelli and Ellison, 2013). Both the data from Di Vittorio et al. (2001) and current data came from the same study area and were collected using the same methodology. Statistical tests were considered significant if p-value < 0.05 and marginally significant if p-value < 0.10.

Results

A total of 98 pellets and 13960 pictures taken from camera-traps were analysed. Overall, 263 prey items were identified: 103 from pellets, 105 from prey remains and 55 from camera-traps. Prey remains were taxonomically classified, and included mammals, birds and reptiles, across 22 different species (Table 1). The mean number of prey identified per year was 49.83 ± 19.91 and there were marginal differences in prey frequency among years (Kruskal-Wallis test: χ^2= 7.75, Montecarlo p = 0.087).

Birds were the most frequent prey item, followed by mammals and reptiles (Table 1). However, mammals constituted the majority of consumed biomass, followed by birds and reptiles (Table 1). While the European wild rabbit (*Oryctolagus cuniculus*) and pigeons (*Columba spp.*) were the main proponents of the Bonelli's eagle diet, rabbits were the most important consumed prey in terms of biomass (Table 1). Interestingly, the contribution of the reptiles was generally very low (Table 1). Throughout the current study period, diet frequency of wild rabbit declined by 66.67 % (beta = -0.87; R^2 = 0.76; p = 0.022) while there was no observed change in pigeon consumption (beta = 0.20; R^2 = 0.04; p = 0.710; Figure 1).

There were major differences between previous and current data in both frequency (z= -2.224; Montecarlo p = 0.023, n = 22) and percentage of biomass (z = -2.430; Montecarlo p = 0.013, n = 22) with the main differences being attributed to changes in the consumption of lagomorphs and birds. Additionally, in the current study there was a greater diet diversity (Gini Index: previous data = 0.734; current data = 0.820) and dietary breadth (previous data = 3.119; current data = 3.607).

Discussion

Diet composition

Wild rabbits and pigeon species were the main prey of Bonelli's eagles in both this study and previous studies for this species across its breeding range (e.g., Ontiveros and Pleguezuelos 2000;

Moleón et al., 2009; Resano-Mayor et al. 2014). However, diet diversity was lower than that reported in other literature (e.g., Moleón et al., 2009; Caro et al. 2011; Resano-Mayor et al. 2016) likely due to regional differences in prey richness (Gasc, 1997; Hagemeijer and Blair, 1997). Our results also confirm that Bonelli's eagle concentrates its predation effort mainly on birds in Sicily (Massa 1981, Salvo 1988; Di Vittorio 2001) and, in accordance with previous work in the area, that the European wild rabbit represented the main source of dietary biomass. Of particular concern however, is that wild rabbit consumption was reduced over the course of the study period which was likely compensated for by increases in the amount of birds in the diet. Similar trends in compensation for reductions in rabbit in the diet of Bonelli's eagles have been reported elsewhere (Ontiveros and Pleguezuelos 2000; Moleón et al. 2009, 2012). Our results seem to be consistent with this pattern, which could suggest a decrease in the availability of wild rabbit in Sicily is occurring (Lo Valvo et al. 2014, 2017). This is likely, given that in recent decades the wild rabbit has undergone a progressive decline in abundance in Italy (Lo Valvo et al. 2014) due to new viral diseases (e.g., MEV/RHDV2; Camarda et al., 2014) and loss of suitable habitat (Lo Valvo et al. 2017). Similar reductions in wild rabbit availability are likely affecting Bonelli's eagle in other European regions such as Iberian Peninsula as well (Villafuerte et al. 1995; Moleón et al., 2007; Caro et al. 2011; Resano-Mayor et al. 2014). In particular, this raptor may select alternative prey species (pigeons and other birds) particularly in areas where rabbit haemorrhagic disease has drastically depleted rabbit abundances (Moleón et al. 2009, 2012; Caro et al. 2011; Resano-Mayor et al. 2014).

Regarding the woodpigeon, we found a higher frequency of occurrence in the diet of this raptor comparing to previous studies in Sicily (Massa 1981; Salvo 1988; Di Vittorio et al. 2001), possibly due to the fact that, in comparison with several decades years ago (Lo Valvo et al. 1993), the distribution of woodpigeons in Sicily has increased by 39% (AA.VV. 2008). Considering the eclectic diet of this raptor, it is unlikely that the abundance of the main prey could limit its distribution (Caro et al. 2011). However, several studies indicate that abundance of rabbit in the diet may affect the productivity and mortality of this species, especially of young and immature

190 birds (see e.g. Carrete et al. 2002; Balbontín et al. 2003). High consumption of optimal prey (i.e.,
191 rabbits) or moderate consumption of these species, complemented by alternative items (e.g.
192 pigeons), could improve productivity, adult survival and nestling body condition (Resano-Mayor
193 et al. 2014, 2016), whereas an increase in diet diversity has the opposite effect (Moleón et al.
194 2012; Resano-Mayot et al. 2016; Rollan et al. 2016).

195

196 *Management implications*

197 Some authors have proposed measures to increase prey availability to enhance Bonelli's eagle
198 conservation (Resano-Mayor et al. 2014), including management guidelines to maintain high-
199 density populations of rabbits, and enhance populations where they are scarce (Caro et al. 2011).
200 Increasing prey availability in low quality territories could be an adequate management measure
201 for the recovery and conservation of Bonelli's eagle populations where prey scarcity affects
202 breeding success (Ontiveros et al. 2004; Ferrer et al. 2018). In addition, actions to improve prey
203 populations, particularly rabbits, could also be an important conservation strategy in dispersal
204 areas (Rollan et al. 2016) and eventually to promote the establishment of new breeding pairs.

205 In general, main management actions to recover and increase prey populations in the long term
206 could include habitat restoration and implementation of sustainable hunting programmes (Rollan
207 et al. 2016). However, when local populations of Mediterranean raptors, such as the Sicilian
208 population of Bonelli's eagle, are subjected to other factors which impact population persistence
209 such as severe habitat degradation (Di Vittorio et al. 2012), which is further compounded by
210 severe and sharp reduction in prey species, especially wild rabbit (Lo Valvo et al. 2014, 2017), it
211 could be advisable to provide supplementary feeding (Rollan et al. 2016) in order to increase
212 productivity in occupied territories until habitats are restored and prey population stability has
213 returned. For example, the ongoing LIFE ConRaSi project (Conservation of Raptors in Sicily)
214 funded by the European Union, supplemented food availability via the construction of several
215 strategically located rabbit farms across eagles' territories. This management action has already
216 had benefits on other threatened species such as the Spanish Imperial eagle (*Aquila adalberti*;

Blanco 2006; González et al. 2006; Ferrer et al. 2013, 2018) and the Eastern Imperial eagle (*Aquila heliacal*; Demerdzhiev et al. 2011). These structures, considered as temporary and maintained for a medium timespan (e.g., five years) provide safe places where rabbits can breed and find refuge from predators, increasing their survival and therefore their population size (Fernandez-Olalla et al. 2010; Guil et al. 2014). This strategy could favour occupation of new territories and enhance demographic performance (i.e., breeding success and survival rate) of the Sicilian population of Bonelli's eagle (Di Vittorio et al. 2018) over the short term until key factors attributed to population declines can be addressed.

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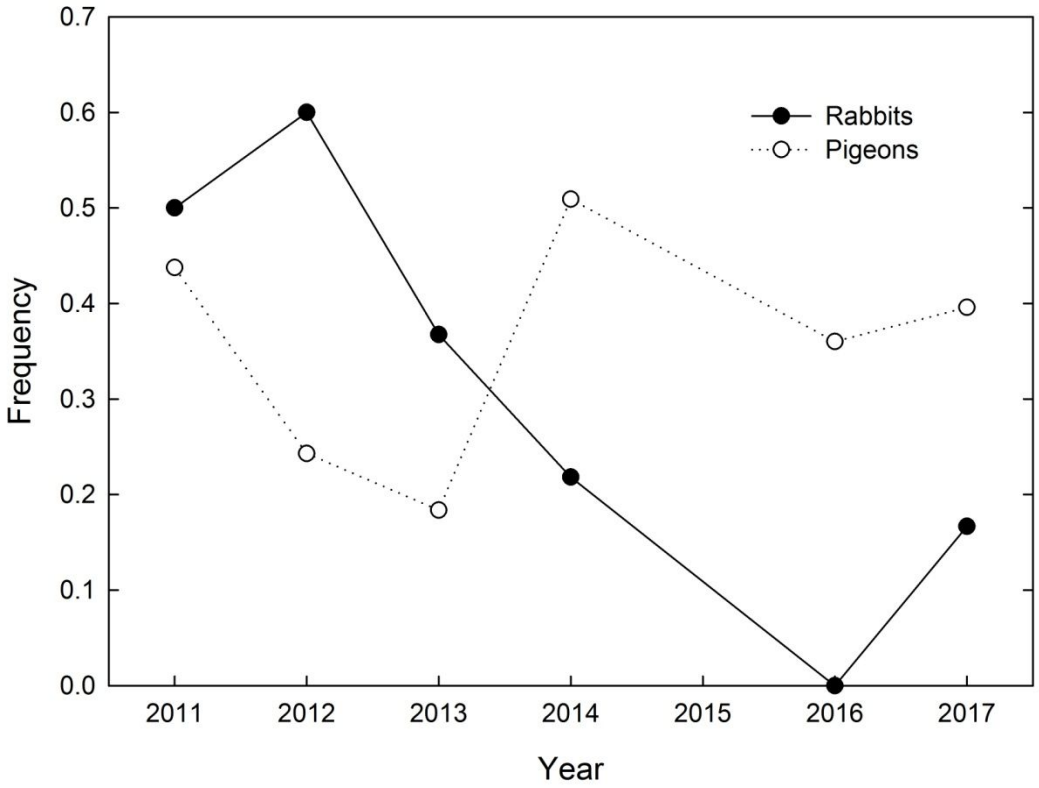

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380 Fig. 1. Frequency of wild rabbit (*Oryctolagus cuniculus*) and pigeons (*Columba* spp.) in the diet

381 of Bonelli's eagle in Sicily (Italy) during the study period.

382 Table 1.- Dietary composition of Bonelli's eagle in Sicily during the breeding period. N =
383 number of prey items recorded. Data from Di Vittorio et al. (2001) span from 1993 to 1998;
384 current data were obtained from 2011 to 2017.

		Di Vittorio et al. (2001)			present study		
Group	Taxon	N	Number (%)	Biomass (%)	N	Number (%)	Biomass (%)
Mammals			39.07	69.84		36.88	65.71
	LEPORIDAE		37.09	69.43		36.50	65.64
	European rabbit *(Oryctolagus cuniculus)*	56	37.09	69.43	88	33.46	56.08
	Italian hare *(Lepus corsicanus)*		0.00	0.00	8	3.04	9.56
	MURIDAE		1.99	0.41		0.38	0.07
	Rattus sp	3	1.99	0.41	1	0.38	0.07
Birds			57.62	29.55		61.60	34.12
	CORVIDAE		18.54	7.16		15.59	5.80
	Hooded crow *(Corvus cornix)*	2	1.32	0.90	10	3.80	2.31
	Jackdaw *(Coloeus monedula)*	22	14.57	5.50	15	5.70	1.93
	Eurasian Magpie *(Pica pica)*	4	2.65	0.76	16	6.08	1.56
	COLUMBIDAE		38.41	22.18		34.22	19.54
	Common Wood Pigeon *(Columba palumbus)*	10	6.62	4.90	45	17.11	11.33
	Rock Dove *(Columba livia)*	48	31.79	17.28	44	16.73	8.14
	European turtle dove *(Streptopelia turtur)*		0.00	0.00	1	0.38	0.07
	TURDIDAE		0.00	0.00		1.14	0.15
	Common Blackbird *(Turdus merula)*		0.00	0.00	3	1.14	0.15
	STURNIDAE		0.00	0.00		2.28	0.26
	Spotless Starling *(Sturnus unicolor)*		0.00	0.00	6	2.28	0.26
	PHASIANIDAE		0.00	0.00		3.04	3.34
	Rock Partridge *(Alectoris graeca)*		0.00	0.00	5	1.90	1.41
	Chicken *(Gallus gallus domesticus)*		0.00	0.00	3	1.14	1.93
	FALCONIDAE		0.66	0.21		2.66	0.75
	Common Kestrel *(Falco tinnunculus)*	1	0.66	0.21	5	1.90	0.54
	Cesser Kestrel *(Falco naumanni)*		0.00	0.00	2	0.76	0.21
	ACCIPITRIDAE		0.00	0.00		0.38	0.49
	Common Buzzard *(Buteo buteo)*		0.00	0.00	1	0.38	0.49
	BURHINIDAE		0.00	0.00		0.38	0.40
	Eurasian Stone Curlew *(Burhinus oedicnemus)*		0.00	0.00	1	0.38	0.40
	PHALACROCORACIDAE		0.00	0.00		0.38	1.54
	Great Cormorant *(Phalacrocorax carbo)*		0.00	0.00	1	0.38	1.54
	ARDEIDAE		0.00	0.00		0.76	1.54
	Grey Heron *(Ardea cinerea)*		0.00	0.00	2	0.76	1.54
	LARIDAE		0.00	0.00		0.76	0.31
	Black-headed Gull *(Chroicocephalus ridibundus)*		0.00	0.00	2	0.76	0.31
Reptiles			3.31	0.62		1.52	0.17
	COLUBRIDAE		2.65	0.60		0.76	0.15
	Green Whip Snake *(Hierophis viridiflavus)*	4	2.65	0.60	2	0.76	0.15
	LACERTIDAE		0.66	0.02		0.76	0.02
	Western Green Lizard *(Lacerta bilineata)*	1	0.66	0.02	2	0.76	0.02
		151			263		

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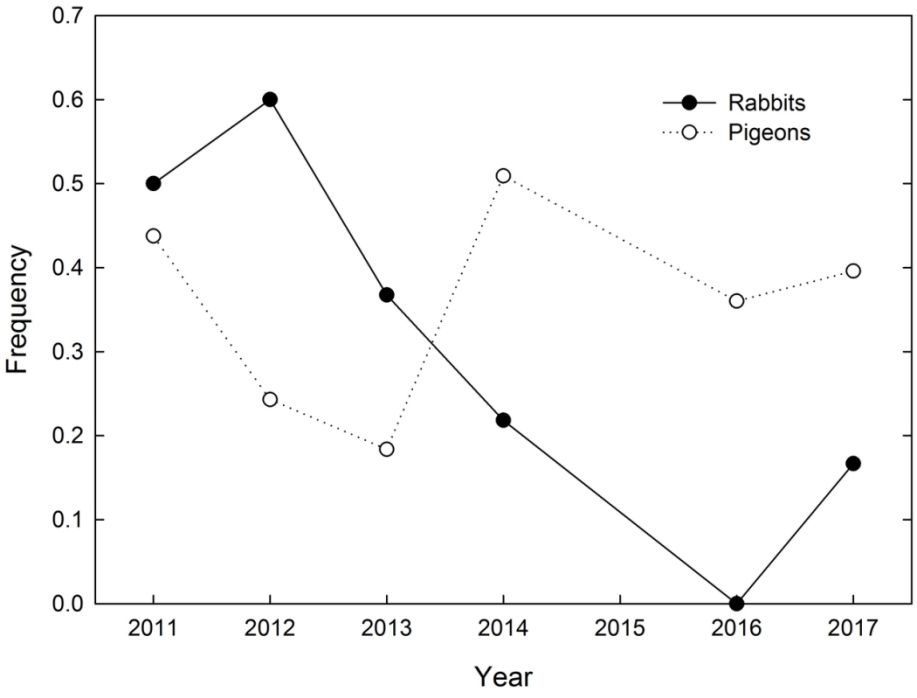

Frequency of wild rabbit (Oryctolagus cuniculus) and pigeons (Columba spp.) in the diet of Bonelli's eagle in Sicily (Italy) during the study period

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497x332mm (96 x 96 DPI)