



Measuring diet during the breeding season & at other times



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MEASURING DIET DURING THE BREEDING SEASON & AT OTHER TIMES

HOW TO OBTAIN CONTEXTUAL DATA ON THE DIET OF RAPTORS

Quantifying the diet of raptor individuals or populations is relevant for most ecotoxicological studies considering that many contaminants accumulate in raptors via prey ingestion as the main exposure route.

Diet composition is highly relevant as contextual data when monitoring contaminants that bioaccumulate and biomagnify along the food webs, therefore reaching higher levels in top predators such as raptors. Diet composition is particularly important when a monitoring programme covers a broad geographic region, along which a raptor species is likely to have considerable variation in its main prey.

Dietary information should be comparable in order to be used to interpret the broad scale patterns of contaminant levels. The use of different methods to collect dietary data, its timing and the individuals sampled can result in undesired biases in interpretation due to a mismatch between the samples used for contaminant and dietary analysis.

Adequate planning should consider that some diet study methods are only possible for specific individuals (age, sex, status) or time of year, which may not agree with the ecotoxicology study targets.

Some diet description parameters are more relevant for ecotoxicological studies (e.g., proportion of diet composed of species from different trophic levels), however their importance can vary according to the objectives of each study. Therefore, it is relevant to be aware that not all diet study methods produce the same descriptive parameters.

The logistics, skills, permits, and equipment needed to each diet study method, from field work to laboratory analysis, is likely to be influential in their use. Some methods can have considerable costs associated (in personnel or equipment) or may be time consuming and extend beyond reasonable deadlines. Permits to handle raptors (cross-ref to advice hub section), previous experience on prey identification, and comprehensive reference collections are also pre-requisites for some diet study methods, making their quick implementation a hard task.

MAIN METHODS TO STUDY RAPTOR DIET

PELLET ANALYSIS

The visual analysis of the contents of regurgitated pellets is one of the most frequently used methods to study the diet of many raptor species. Regurgitated pellets consist mostly of hard, non-digested parts, such as bones, fur, feathers, arthropod chitinous parts, resulting from the digestive process of prey. The shape, colour, and consistency of pellets depend on the raptor species and its diet.

FIELD. Pellets are regularly expelled by raptors, being most frequently found in nests, hunting perches and roosting sites. Fresh pellets are generally compact, but they disaggregate with time, especially if left in exposed sites. This method can rely only on fresh pellets, but researchers often include older material accumulated in nests and roosting sites resulting from disaggregated pellets (most often loose bones). Pellets can be collected during breeding seasons in nests sites or nearby perches, but also during non-breeding seasons in territories of sedentary species or in communal wintering roosting sites. Pellets collected in nests will more closely reflect the diet of juveniles and incubating adults, while pellets collected in perching sites within a territory will better reflect the diet of adults.

LAB. Visual identification of the prey material contained in pellets is preferentially done in a lab. Prey parts most used for identification are bones, fur, feathers, arthropod chitinous parts, etc. Depending on the material and prey size, the identification of some prey parts may require the use of a microscope. Identification of prey may require good identification keys, detailed illustrative guides, and comprehensive reference collections.

PARAMETERS. Since it is possible to identify prey to the species level, or at least to a detailed enough taxonomic level, many descriptive parameters can be obtained from pellet analysis. Prey composition is generally described from a pool of pellets. In most cases, pellet analysis produces a list of prey with the number of individuals consumed in each species/prey group. This allows to determine diet composition and calculate most descriptive parameters. When quantification of individuals is difficult, or it is not possible to determine if different pellets contain remains of the same prey individual, it is possible to use the prey presence per pellet (frequency of occurrence).

PREY REMAINS

The analysis of uneaten prey remains is among the most used methods to study the diet of raptors along with the analysis of pellets. It can be used for most raptors that regularly discard non-ingested prey items, sometimes using specific sites used for the purpose of feeding or plucking prey. These uneaten prey remains can consist of the entire animal or

part of it stored for consumption in subsequent days, but it can also be non-ingested parts, generally limbs with large bones difficult to break and ingest entire, heads, or hard skins (e.g., hedgehogs), or plucked feathers. Raptors that feed on large prey and birds and use easy to find sites to feed or pluck the prey are more likely for their diet to be studied using prey remains.

FIELD. Prey remains are generally collected in nests (non-ingested prey parts resulting from the feeding of nestlings and incubating adults), feeding perches (locations used specifically and regularly to feed), or plucking sites (locations used specifically and regularly for plucking the feathers of avian prey). Prey remains collected in these sites may yield differences in diet. The location of feeding perches and plucking sites in relation to the nest can also express differences related to food taken preferentially to nestlings or eaten by adults. Prey remains can be collected along the year in feeding perches and plucking sites, and in the nest during the breeding season.

LAB. Most prey remains are easy to identify (complete limbs, heads, skins). Visual identification of the prey remains might be easily done in the field but in most cases it is done in a lab. The identification of feathers requires a good reference collection or illustrated guide, benefiting from accumulated experience of the person carrying out the task.

PARAMETERS. Like in pellet analysis, most prey remains may be identified to the species level, or at least to a detailed enough taxonomic level. The analysis of prey remains will generally produce a list of consumed prey with the number of individuals in each species/prey group. This information can then be used to determine diet composition and calculate most descriptive parameters.

COMBINATION OF PELLETS & PREY REMAINS

A common approach to study the diet of raptors is combining the analysis of both pellets and prey remains by pooling the results together to avoid double-counting. It is suggested that pellets and prey remains can be complementary, reducing the opposite biases of the two methods towards prey types or sizes. There should be a relatively well-balanced collection of both materials, with effort made on several locations where these may be found.

FIELD. As described in the two methods above. Optimized by combining visits to sites that guarantee the representation of pellets and prey remains.

LAB. Similar to what was described for the two methods above.

PARAMETERS. The combination allows to determine diet composition and calculate most descriptive parameters.

FOOD STORES (AKA: PREY CACHING; FOOD HOARDING; LARDERS)

Some raptors species cache prey to deal with shortage periods. Food stores can consist of just a few items or be of considerable size. Prey caching may occur before and/or during breeding. Although food stores may be considered a type of prey remains, it has been used as a method per se to study the diet of some raptor species.

FIELD. Food stores are generally found in nest-boxes or nest-holes. Food may be accumulated prior to breeding, generally during late autumn and winter, or during breeding (particularly incubation and nestling period). Prey identification should be made in situ without removing prey. If repeated visits to the nest are foreseen, then prey should be marked. Prey identification is facilitated by being found whole. Identification of prey species requires identification skills of animals in hand (especially mammals and birds).

LAB. Not required

PARAMETERS. Most prey individuals may be identified to the species level, or at least to a very detailed enough taxonomic level. This method will produce a list of consumed prey with the number of individuals in each species/prey group per food store. This information can then be used to determine diet composition and calculate most relevant descriptive parameters.

STOMACH CONTENTS

One of the earliest methods used to study the diet of raptors has been the analysis of the contents of the digestive tract that are collected from dead individuals. Most commonly this consists of the contents found in the stomach, but in the case of diurnal raptors it can also include the contents found in the crop. This method implies that the individual is dead except in the case of the study of crop contents that are forced out from nestlings or fledglings. Ethically this method can only be applied to individuals found dead. To retrieve stomach contents, individuals should be recently dead, the best example being the road-killed raptors. This method has strong sample size limitations, it does not allow replicates for the individual/territory and depends on the non-desired mortality of individuals. It can be opportunistically used in the cases where the number of road-killing casualties is frequent, although this method has little feasibility as a long-term sampling method. This method can allow the differentiation of diet between age and sex groups.

FIELD. The collection of dead raptors is nowadays mostly associated to road-killings, and it may be carried out opportunistically or as a systematic survey in the case of projects dedicated to studying road mortality of wildlife. Dead individuals collected along

roads are stored in plastic bags and taken to the lab for analysis. Permits to hold dead raptors are often required.

LAB. The analysis of stomach contents requires skills and equipment for carrying out the necropsy of raptors. During the necropsy the digestive tract is extracted and the content of the stomach (and crop in the case of diurnal raptors) is separated to a container. Stomach contents can be in different stages of digestion, from recently ingested to fully digested, in the latter case, very similar to fresh pellets. Less digested contents may facilitate the identification of prey that have mostly soft parts and tend to be more heavily digested and thus be less frequently found in pellet analysis. Visual identification of the prey material is like the procedure used for pellet analysis. Prey parts mostly used for identification are bones, fur, feathers, and arthropod chitinous parts. Prey identification may require the use of a microscope, good identification keys, detailed illustrative guides, and comprehensive reference collections.

PARAMETERS. Similar to pellet analysis; it is possible to identify the prey to the species level, or at least a detailed enough taxonomic level. Prey composition may be given by the total prey identified from a pool of individuals or presence per individual (frequency of occurrence per individual). The analysis of stomach contents produces a list of prey with the number of individuals consumed in each species/prey group, which allows to determine diet composition and calculate most descriptive parameters.

DIRECT OBSERVATION

This method results from the direct observation of acts of prey catching and/or feeding by raptors. It is a relatively simple method, but it may require a large collection of occasional observations or long observation periods on fixed locations to obtain a sufficient number of prey species. This method has been used since soon to determine the diet of raptors and is still used nowadays. It may provide behavioural information about the feeding habits that is not possible by other methods (e.g., pellets, prey remains, stomach contents). It is more suitable to determine the diet of nestlings and fledglings as they are in a fixed and predicted site. This method is often dependent on the occurrence of breeding success to have results. Radio-tracking data has been used to identify feeding locations and subsequent confirmation of prey in situ, as for example in vultures.

FIELD. Direct observations may result from opportunistic observations in random locations or may be carried out systematically in places regularly used for feeding (feeding perches, artificial feeding stations) or in the nest (prey delivered to nestlings or incubating adults). Observations are made from a distance by an observer with the help of binoculars or telescope, often from a concealed location (natural or man-made hide). To obtain enough records it may be necessary (to devote/allocate?) a considerable total

time of observation, distributed along the period of the day when the predator is active. This method is not suitable to study the diet of most owl species, since observation during the night is highly limited. For systematic studies, this method depends on previous knowledge of nest sites or feeding perches/stations. Raptor nests may be in very concealed locations, preventing the direct observation of the food taken to the nest. This method is more suitable for species that feed on large prey, as smaller prey tends to be harder to identify from a distance. The use of the direct observation method to study diet outside the breeding season is possible if the species uses regular feeding perches or otherwise limited to opportunistic observations.

LAB. No laboratory work associated.

PARAMETERS. This method produces a list of prey items per nest or location, which allows to determine diet composition and calculate most descriptive parameters.

PHOTOS & VIDEO RECORDINGS

This method results from the indirect (passive) observation of acts of prey catching and/or feeding. It relies on the use of photographic or video equipment, placed in fixed locations, most frequently in the nest (near the nest?) of raptors. This method has advanced most recently with the technological development of cameras. It may provide complementary behavioural information about the feeding habits that is not possible by other methods (e.g., pellets, prey remains, stomach contents). It is more suitable to determine the diet of nestlings and fledglings. This method is often dependent on the occurrence of breeding success to have data. One of the main logistic limitations is the number of available equipment and their costs (cameras).

FIELD. This method depends on previous knowledge of nest sites or feeding perches/stations that are suitable for the installation of photographic or video cameras. The location of cameras must allow a good visibility of the nest or perch in order to identify the prey. There is a wide range of available equipment with different characteristics and prices, which can be selected according to the objectives and budget of the project. Equipment with infrared sensitivity allows capturing images in low light conditions.

LAB. Photos and records are analyzed in the computer to identify prey items. Better quality images in general will allow more accurate prey identification.

PARAMETERS. This method produces a list of prey items per nest or location, which allows to determine diet composition and calculate most descriptive parameters.

STABLE ISOTOPE ANALYSIS

The use of stable isotopes analysis (SIA) to study the diet of raptors is a relatively recent method, which started mostly during the 1990's. Stable carbon, nitrogen and sulphur isotopes are the most frequently used. Stable isotope analysis may be carried out on several tissues, including feathers, blood plasma, muscle, liver, claws. Some tissues require access to dead individuals and may be carried out in collection specimens. SIA can be used to assess the diet of juveniles and adults. Often potential prey items are collected to establish their isotopic signature. Complementary, SIA can be used to determine the migratory origin of an individual, tracing back the growth of feathers to a certain latitude.

FIELD. Sample tissue collection can be done directly on live or dead individuals, or indirectly on shed feathers. If prey samples are collected, specific methods may be needed. Prey samples may be taken from prey remains if available.

LAB. This method requires laboratory protocols to process the tissues before isotopic analysis. The samples are subsequently analysed in specific equipment (isotope ratio mass spectrometer).

PARAMETERS. Stable isotope analysis results are often analysed together with another method of diet analysis such as pellet analysis. Isotopic values can be compared between territories, regions or countries to determine spatial variations in diet composition. This method does not produce a quantified list of prey species.

DNA-BASED APPROACHES

Conventional methods of diet analysis rely mostly on visual observation of direct feeding events and prey deliveries or of the detailed contents of pellets, prey remains and stomach contents. These visual methods often fail to detect small prey or soft items that are easily digested completely. The development of molecular techniques, such as next generation sequencing (high-throughput sequencing), has created new approaches to study the diet of animal species, including raptors.

Recent DNA-based approaches have been applied to dietary studies especially in the last decade. Specific PCR primers can be used to amplify DNA in order to detect predation on a few specific prey or prey groups. DNA amplification may also be done using general or group-specific primers (barcoding approach).

In the case of raptors, DNA samples can be extracted from several sources. Buccal and talon swabs can be a source of prey DNA by retaining prey blood, skin, feathers, or fur. Cloacal swabs can also provide dietary DNA samples. Pellets is another potential source of prey DNA, being particularly useful when the visual identification of prey contents is difficult.

Although the number of dietary studies of raptors using DNA-based approaches is still quite reduced, this method has a great margin to progress in the next years.

FIELD. Buccal and talon swabs can be taken from nestlings and adult birds. It requires accessing the nest in the case of nestlings and capturing raptor individuals in the case of adults. Handling and capturing skills and permits are necessary.

LAB. DNA extraction and amplification needs skilled personnel and specific techniques and equipment. Prey identification from DNA sequences requires adequate computational competences.

PARAMETERS. This method typically produces a list of prey with the corresponding frequency of presence per sample analysed. It does not allow the quantification of the number of individuals of the same prey species per sample in those cases that more than one could be present, as for example in pellets.

This section of the advice hub does not attempt to come up with the best single method to study the diet of European raptors for the purpose of ecotoxicological studies. Instead, it intends to be a first guide highlighting the pros and cons of the several methods and exploring the extent to which these methods may be employed and comparable as contextual data in contaminant monitoring schemes (Table 4). Ultimately, the most suitable method to obtain contextual diet data may be determined by the study aims combined with its logistic capacities and the avian order or bird species involved (Table 5, Table 6).

TABLE 1 – Pros and cons of the main diet analysis methods for obtaining contextual data in contaminant biomonitoring schemes

METHOD	ADVANTAGES	DISADVANTAGES/LIMITATIONS
PELLETS	<p>Familiar and widely used, suitable for comparison studies.</p> <p>Suitable for most raptor species.</p> <p>Identification keys and reference collections are relatively common and accessible for most frequent prey species (particularly mammals).</p> <p>Pellets can be collected all year round.</p> <p>It generally provides a large sample size (number of prey items per site)</p>	<p>Depends on finding a nest site, roost site or regular perch.</p> <p>It may underestimate the importance of soft-bodied prey, large prey, and prey that are regularly decapitated.</p> <p>It may not be suitable for raptors that do not ingest hard parts of prey (only ingest soft parts or little bones), or those with strong digestive capacities.</p> <p>Prey identification requires experience and specific knowledge</p> <p>Prey identification is not always possible at the species level</p>
PREY REMAINS	<p>Can be found in various places: feeding perches; under and inside nests; under roosting sites.</p> <p>Particularly useful for species that use plucking perches (e.g., sparrowhawk, goshawk).</p> <p>Prey identification is generally easy.</p> <p>Samples collected from prey remains may be analysed for contaminants to better estimate biomagnification.</p>	<p>It may underestimate the importance of small prey while overestimating the importance of large prey, and especially birds.</p> <p>Sample size based on the method may be of limited size for some species.</p> <p>Results may be biased if adults clean the nest from larger prey remains, or if prey remains are removed by scavengers</p>
COMBINATION OF PELLETS AND PREY REMAINS	<p>Reduces the potential biases of using only pellets or prey remains.</p> <p>Frequently pellets and prey remains can be found in the same place, so it does not require extra searching effort.</p> <p>Suitable for most raptor species.</p> <p>Very common in literature, suitable for comparison studies.</p>	<p>Possible double counting of individuals in pellets and prey remains should be accounted for.</p> <p>Requires balancing the contribution of both methods.</p> <p>Easily digested prey items may not be detected in both pellets and prey remains.</p>
FOOD STORES	<p>Can allow the collection of a considerable sample size when the raptor accumulates large food stores.</p> <p>Prey species is generally easy to identify from whole individuals.</p> <p>Can allow detailed analysis of prey, namely contamination levels and body condition.</p>	<p>Method restricted to raptor species that accumulate food stores.</p> <p>Depends on relatively good accessibility to the food store (easier if raptor uses a nest-box).</p>

METHOD	ADVANTAGES	DISADVANTAGES/LIMITATIONS
STOMACH CONTENTS	<p>Provides diet information for individuals found dead and simultaneously sampled for ecotoxicology, and for which the nest location is not known.</p> <p>Allows determining exactly what the individual has eaten and can facilitate identification of prey with soft parts if undigested.</p> <p>Can provide information about poisoning events.</p> <p>Provides easy comparison of diet between age classes and sexes.</p>	<p>Depends on encountering dead individuals (most suitable for common species, or those which are frequent victims of road-killing or collision with human-made structures).</p> <p>Only single samples per unit (i.e., territory or breeding pair), resulting in limited representativeness due to small sample sizes.</p> <p>Requires many individuals (and often long periods) to obtain moderate sample sizes.</p>
DIRECT OBSERVATION	<p>Generally feasible even when the nest is not accessible.</p> <p>It can provide behavioural information on feeding.</p> <p>Relatively simple method.</p> <p>Generally, results are less biased by prey type compared to pellets and prey remains.</p>	<p>Observation of prey deliveries to nestlings only possible on active breeding pairs.</p> <p>It requires much observation time (human resources) to obtain a good sample size.</p> <p>Generally, it has an unfavourable cost-effective relation.</p> <p>Smaller prey may be difficult to identify in observations from considerable distances.</p> <p>Visibility depends much on weather conditions.</p> <p>Not suitable for most owl species.</p>
CAMERAS (PHOTOS OR VIDEOS)	<p>Generally, results are less biased by prey type compared to pellets and prey remains.</p> <p>Existing technology allows 24h monitoring (IR cameras).</p> <p>It can provide behavioural information on feeding.</p> <p>Solar panels allow large autonomy for long-term monitoring.</p> <p>Camera technology is improving fast (e.g., image quality, storing capacity).</p> <p>It reduces disturbance from human presence and visits to the nest.</p>	<p>It requires a considerable logistic (equipment and battery supply and maintenance).</p> <p>It requires many cameras (high cost) to obtain samples from a good number of sites.</p> <p>Several items may not be identifiable in images.</p> <p>Video analysis to extract the information can be very time-consuming.</p> <p>Some species may be sensitive to disturbance by the equipment or its installation.</p> <p>Some nests may not have conditions (structure and position) for installing cameras.</p>

METHOD	ADVANTAGES	DISADVANTAGES/LIMITATIONS
STABLE ISOTOPES ANALYSIS (SIA)	<p>More likely to reflect individual long-term diet and biomass ingestion and particularly assimilation.</p> <p>Can be used to obtain retrospective information on diet composition of specimens from collections or samples collected previously.</p> <p>SIA can provide information on migratory origin coupled with dietary information.</p>	<p>Requires the availability of expensive analytical equipment and complex sample processing.</p> <p>SIA performs best combined with other diet study methods: it will not produce qualitative information of prey composition if used by itself.</p> <p>Dietary studies using SIA require information on isotope signatures of main prey groups.</p> <p>Similar stable isotope values do not necessarily mean similar diets.</p> <p>May have low sensitivity to assess local or regional differences in diet.</p>
DNA-BASED APPROACHES	<p>Provides dietary information for migratory raptors or individuals captured once (with no information on nest site or perches). In pellets, apart from the identification of prey, it allows the identification of the predator when this is unknown.</p> <p>Prey list may increase by detection of species missed by most other methods.</p> <p>Samples can be collected during other activities involving bird handling and associated with breeding performance parameters.</p>	<p>Currently it is still an unfavourable cost-effective method.</p> <p>Requires the availability of expensive analytical equipment and complex sample processing.</p> <p>The method does not quantify prey number of the same species, in particular when analysing pellet or faeces matrices.</p> <p>Prey identification depends on the use of adequate primers for DNA amplification.</p> <p>Correct identification of prey depends on a comprehensive database of DNA sequences for the several prey species.</p>

TABLE 2–. Number of entries per raptor species and per diet study method (1931-2021).

Each entry is a unique combination of a raptor species/diet study method obtained from the review of 434 published studies on diet of raptors in Europe.

RAPTOR SPECIES	PELLETS	PREY REMAINS	COMBINATION OF PELLETS AND PREY REMAINS	FOOD STORES	STOMACH CONTENTS	DIRECT OBSERVATION	CAMERAS PHOTO/VIDEO	STABLE ISOTOPES	DNA-BASED APPROACHES	TOTAL
Eagle owl <i>Bubo bubo</i>	12		39		2					53
Goshawk <i>Accipiter gentilis</i>	4	23	11			5	5			48
Bonelli's eagle <i>Aquila fasciata</i>	11	6	9			9	1	2		38
Golden eagle <i>Aquila chrysaetos</i>	8	10	12			6				36
Little owl <i>Athene noctua</i>	26		3		1					30
Common buzzard <i>Buteo buteo</i>	5	8	9		3	2	3			30
Common kestrel <i>Falco tinnunculus</i>	11	3	8		1	3	2			28
Barn owl <i>Tyto alba</i>	23	1	2		1					27
Long-eared owl <i>Asio otus</i>	22		4							26
Tawny owl <i>Strix aluco</i>	18	2	3	1	2					26

RAPTOR SPECIES	PELLETS	PREY REMAINS	COMBINATION OF PELLETS AND PREY REMAINS	FOOD STORES	STOMACH CONTENTS	DIRECT OBSERVATION	CAMERAS PHOTO/VIDEO	STABLE ISOTOPES	DNA-BASED APPROACHES	TOTAL
Montagu's harrier <i>Circus pygargus</i>	6	1	8			2				17
Sparrowhawk <i>Accipiter nisus</i>		13	2			1				16
Tengmalm's owl <i>Aegolius funereus</i>	2		4	5		1	4			16
Egyptian vulture <i>Neophron percnopterus</i>	2	9	2					1		14
Peregrine falcon <i>Falco peregrinus</i>	2	3	4			2	1			12
Hen harrier <i>Circus cyaneus</i>	3	1	1			4	1		1	11
Marsh harrier <i>Circus aeruginosus</i>	4	1	1			3				9
Lanner falcon <i>Falco biarmicus</i>	3	3				3				9
Red kite <i>Milvus milvus</i>	6	2	1							9
Short-eared owl <i>Asio flammeus</i>	8									8

RAPTOR SPECIES	PELLETS	PREY REMAINS	COMBINATION OF PELLETS AND PREY REMAINS	FOOD STORES	STOMACH CONTENTS	DIRECT OBSERVATION	CAMERAS PHOTO/VIDEO	STABLE ISOTOPES	DNA-BASED APPROACHES	TOTAL
Pygmy owl <i>Glaucidium passerinum</i>	3		2	3						8
White-tailed eagle <i>Haliaeetus albicilla</i>	2	3	1					2		8
Booted eagle <i>Hieraaetus pennatus</i>			5			3				8
Ural owl <i>Strix uralensis</i>	3		5							8
Cinereous vulture <i>Aegypius monachus</i>	4	2	1							7
Greater spotted eagle <i>Clanga clanga</i>	3	1	1			2				7
Black kite <i>Milvus migrans</i>		6	1							7
Spanish imperial eagle <i>Aquila adalberti</i>	2	2	1			1				6
Lesser spotted eagle <i>Clanga pomarina</i>	1	2	1			1	1			6

RAPTOR SPECIES	PELLETS	PREY REMAINS	COMBINATION OF PELLETS AND PREY REMAINS	FOOD STORES	STOMACH CONTENTS	DIRECT OBSERVATION	CAMERAS PHOTO/VIDEO	STABLE ISOTOPES	DNA-BASED APPROACHES	TOTAL
Lesser kestrel <i>Falco naumanni</i>	3					2		1		6
Bearded vulture <i>Gypaetus barbatus</i>		3				2	1			6
Eastern imperial eagle <i>Aquila heliaca</i>		1	3			1				5
Rough-legged buzzard <i>Buteo lagopus</i>	1	1	1				1	1		5
Long-legged buzzard <i>Buteo rufinus</i>	1	1	2			1				5
Short-toed snake-eagle <i>Circaetus gallicus</i>	2	1	1			1				5
Osprey <i>Pandion haliaetus</i>		2				3				5
Black-shouldered kite <i>Elanus caeruleus</i>	4									4
Merlin <i>Falco columbarius</i>		4								4

RAPTOR SPECIES	PELLETS	PREY REMAINS	COMBINATION OF PELLETS AND PREY REMAINS	FOOD STORES	STOMACH CONTENTS	DIRECT OBSERVATION	CAMERAS PHOTO/VIDEO	STABLE ISOTOPES	DNA-BASED APPROACHES	TOTAL
Gyrfalcon Falco rusticolus			4							4
Hobby Falcon subbuteo		1	2			1				4
Red-footed falcon Falco vespertinus		3				1				4
Griffon vulture Gyps fulvus	1	1				2				4
Scops owl Otus scops	1	1	1			1				4
Eleonora's falcon Falco eleonora	1		1		1					3
Honey buzzard Pernis apivorus		2			1					3
Hawk owl Surnia ulula	1		1		1					3
Saker falcon Falco cherrug			2							2
Great grey owl Strix nebulosa	1		1							2
Snowy owl Nyctea scandiaca			1							1

RAPTOR SPECIES	PELLETS	PREY REMAINS	COMBINATION OF PELLETS AND PREY REMAINS	FOOD STORES	STOMACH CONTENTS	DIRECT OBSERVATION	CAMERAS PHOTO/VIDEO	STABLE ISOTOPES	DNA-BASED APPROACHES	TOTAL
Levant sparrowhawk Accipiter brevipes										0
Number of entries	210	123	161	9	13	63	20	7	1	607

DIET DESCRIPTION PARAMETERS

Several parameters can be used to describe the diet of raptors. Here we focus on the most frequently used and at the same time the most relevant to be used as contextual data for interpretation of contamination levels in ecotoxicological studies.

QUANTIFICATION: Apart from a simple list giving the detailed prey composition of the diet of a certain raptor in a certain location, it is extremely relevant to be able to quantify the relative contribution of each prey species or group to the diet of a raptor. This is achieved by quantification measures, which may be determined depending on the diet study method. The use of the same quantification measure allows the comparison of results, even if the diet study method is different, although it should always be considered the time of the year and part of the population the samples represent.

PERCENTAGE OF OCCURRENCE BY NUMBER (PON): This parameter is possible to determine when the study method provides the number of individuals consumed for all the prey species or groups found in the diet. It is calculated as the percentage of the number of individuals from a certain prey species/group divided by the total number of prey individuals consumed. It can be calculated for individuals, territories, or populations depending on the unit of analysis.

PERCENTAGE OF OCCURRENCE BY BIOMASS (POB): This parameter requires information of the number of individuals consumed for all the prey species/groups found in the diet but also of the weight of the individuals consumed. The weight of prey individuals can be estimated by measurements of bones and other hard parts or alternatively by using a mean weight of prey taken from literature. To reduce potential biases in POB it is important to attribute the most accurate prey weight as possible, especially in the case of species with large weight variation. POB is calculated as the percentage of the biomass of the individuals from a certain prey species/group divided by the total biomass of the prey individuals consumed.

FREQUENCY OF OCCURRENCE (FO): In some cases, it is not possible to determine the exact number of individuals in a sampling unit (e.g., sample contents, pellets). This is for example the case when the prey items found are only fur or feathers, with no specific structure to count individuals being present. In these cases, quantification can be made by determining the frequency in which each prey species/groups appears in the sampling units (i.e., stomach contents, pellets, prey remains from a nest). Although less informative than the two previously mentioned parameters, it can be an alternative to determine the relative importance of prey species/groups.

DIET DIVERSITY PARAMETERS

The parameters that estimate the degree of diversity of the diet of raptors provide insight about the position of the species in a gradient between more specialist and more generalist diets. The relative position of specialist versus generalist diet can play a role in the susceptibility of a raptor individual/population to the accumulation of contaminants. Among the diet diversity parameters most used are Simpson's diversity index, Shannon's diversity index, food niche breadth, and evenness.

BASIC CONSIDERATIONS FOR DIET CONTEXTUAL DATA USED IN RAPTOR ECOTOXICOLOGY STUDIES

- It is important to have as much as possible a good temporal match between the period represented by the samples analyzed for contaminants and the diet. This match should consider both year and seasonal variations that may occur in the diet of raptors (e.g., dietary shifts due to low prey abundance may have noticeable consequences on contaminant exposure).

- Another match that should be achieved as much as possible is between the population fraction that is sampled for ecotoxicology and for diet. Diet can vary within short distances (between neighbouring territories), but also between age classes (adults vs. nestlings), sexes, and social status (territorial vs. floaters).
- If more than one method has been used to study the diet of a raptor species in order to obtain contextual data for an ecotoxicology analysis, it is important to ensure as much as possible the adequacy to compare the dietary parameters obtained from different methods.
- Ability to assign prey numeric percentage (or preferentially biomass percentage) to trophic level.

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FIGURES AND CHARTS

TABLE 1 – Pros and cons of the main diet analysis methods for obtaining contextual data in contaminant biomonitoring schemes 12

TABLE 2–. Number of entries per raptor species and per diet study method (1931-2021). 15



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