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SEASONAL DIET OF THE MAGHREBIAN ENDEMIC BAT *MYOTIS PUNICUS* (VESPERTILIONIDAE, CHIROPTERA) IN THE ALGERIAN HIGHLANDS

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Abstract. This study investigated the diet of the vespertilionid bat *Myotis punicus* in the highland region of Bouira, Algeria, between March and November 2023. Arthropod remains recovered from guano samples in caves were microscopically examined. The analysis revealed that the species' diet comprises three main arthropod classes: Insecta, Chilopoda, and Arachnida. Based on a total of 2,684 identified insect fragments, Hymenoptera constituted the most frequently consumed prey (24.48%), followed by a diverse selection of other insect orders, including Lepidoptera (18.82%), Coleoptera (18.37%), and Diptera (12.15%). Given the proportional representation of these insect groups, *M. punicus* appears to exhibit a feeding strategy that combines opportunistic foraging with selective predation. Understanding how these bats interact with their environment in the Algerian highlands sheds light on their critical role in controlling insect populations, which helps maintain ecosystem balance and supports public health.

INTRODUCTION

Understanding the diet of bats is essential for conservation, as it provides crucial information regarding their ecological roles and guides habitat management efforts (Alberdi et al. 2012; Fenton and Simmons 2014; Arrizabalaga-Escudero et al. 2015; Cuff et al. 2022). Furthermore, recent developments in molecular approaches, such as metabarcoding, have provided precise data for bat conservation and management (Bourlat et al. 2023).

Myotis species demonstrate significant dietary variation, reflecting differences in habitat diversity, geographic distribution, and morphological adaptations (Dickman and Doncaster 1987; Brosset 2000; Dietz et al. 2007; Altringham and Altringham 2011; Weterings, Alberdi et al. 2012; Umponstira 2014; Loumassine et al. 2019; Massaad et al. 2022). According to recent studies, the coexistence of cryptic taxa is maintained by fine trophic partitioning and that a broad dietary niche is associated with ecological plasticity and spatial distribution (Bonnet-Garcia 2003; Alberdi et al. 2020; Bourlat et al. 2023).

The Maghrebian mouse-eared bat (Myotis punicus Felten, 1977), an endemic species to the Maghreb, is widely distributed in Morocco, Tunisia, and northern Algeria (Ahmim and Moali 2011; Riccucci and Lanza 2014). Its conservation status remains problematic due to habitat degradation and lack of specific ecological data (Wilson and Reeder 2005; Riccucci and Lanza 2014; Massaad et al. 2022; Simmons and Cirranello 2024). Existing dietary data are confined to a few studies, especially from Morocco and Tunisia (Ahmim and Moali 2011; Ahmim 2014), and indicate a diversified diet; however, a comprehensive comparative analysis across regional or altitudinal contexts is still lacking.

Consequently, little is known about the dietary ecology of *M. punicus* in Algeria, particularly in highland areas, which are essential to the conservation of Mediterranean bats (Massaad et al. 2022). Therefore, the purpose of this study is to characterize the diet of *M. punicus* in the understudied highland region of Algeria to better understand its ecological role. Specifically, this descriptive study aims, first, to identify and quantify the diverse arthropod prey groups consumed, thereby filling a significant data gap for this geographic context; second, to analyze seasonal variations in dietary composition across the bat's active period; and finally, to evaluate the species' trophic niche breadth to determine its foraging strategy (opportunistic generalist or a selective specialist) in this study area. By documenting these seasonal feeding patterns, this research provides essential baseline data that can inform future conservation efforts for Mediterranean bat populations.

MATERIALS AND METHODS

Study area and habitat mapping

The study area is located in the highlands region of Algeria, specifically within the Bouira province, situated between M'sila and Bordj Bou Arréridj (Figure 1(a) and (b)). The landscape is characterized by a rugged, sloping terrain that encompasses the southern highlands and the Biban mountain range, with elevations ranging from 700 to 1,400 meters above sea level. This topography frequently features gradients of up to 20% and offers diverse geographic orientations. The specific sampling site is located among the municipalities of Bordj Okhriss, Taguedit, and Mesdour (36°03' 43" N, 3°58'48" E), approximately 60 km north of the provincial capital of Bouira (Figure 1(c)). A mountainous ecosystem composed of natural Aleppo pine forests and cultivated olive groves surrounds the research site (Figure 1(d)). The dominant tree species is *Pinus halepensis*, while *Juniperus oxycedrus*, fig trees (*Ficus* spp.), and a few holm oaks (*Quercus ilex*) grow in the immediate vicinity.

Guano samples were collected from the primary roosting location utilized by the colony. As part of the analytical protocol, insect fragments extracted from the guano were identified using microscopic examination. The guano pellets of *M. punicus* are noticeably larger than those of other bat families present in the region, such as Rhinolophidae, Miniopteridae, and Rhinopomatidae, which facilitates their distinct identification in the field (Ahmim and Moali 2011). To enable comparisons with dietary studies conducted in other geographic regions, our results are expressed as a percentage of frequency of occurrence. A colony of approximately 280 *M. punicus* individuals (Figure 2) was observed in an underground cavity which, based on repeated site visits, serves as an

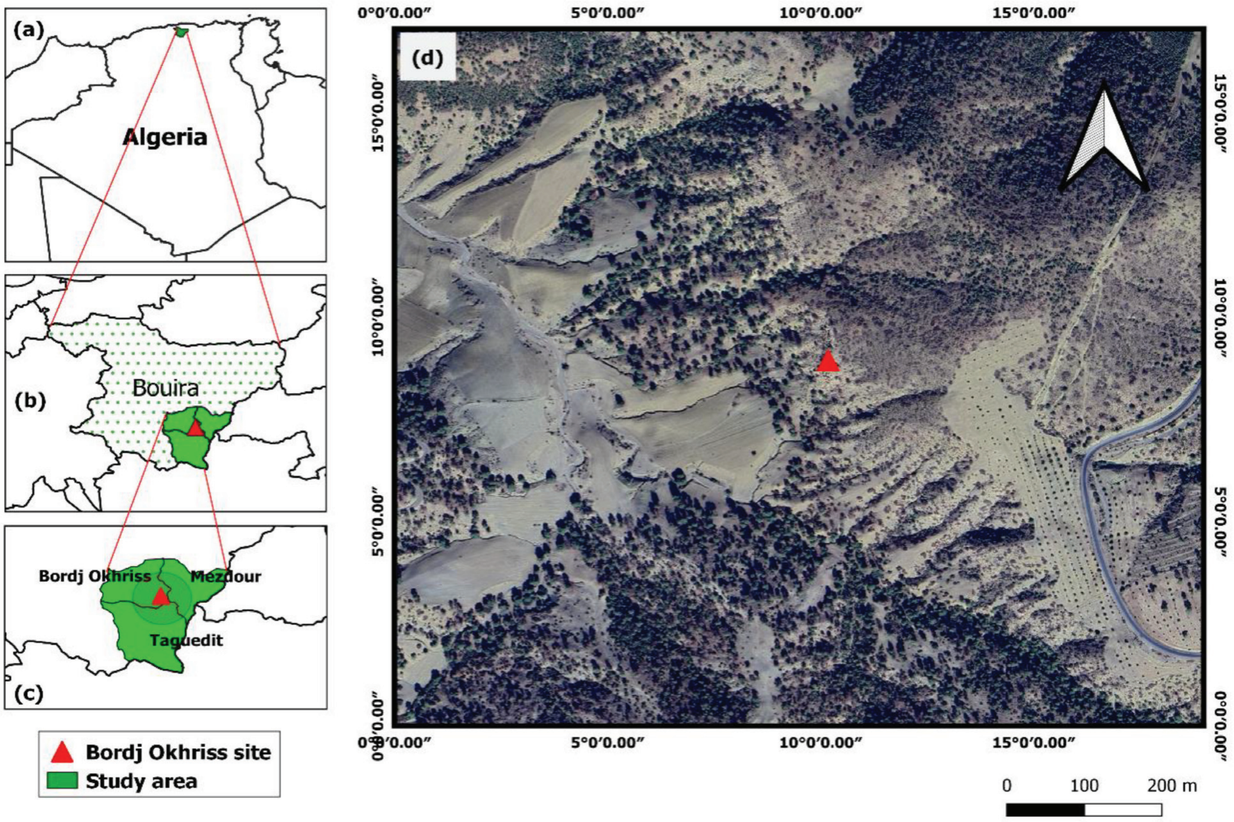

Figure 1. Geographic presentation of the study region: (a) Map of Algeria indicating the location of the Bouira province. (b) Map of the Bouira province detailing the study region. (c) Detailed map of the study area between the municipalities. (d) Satellite image showing the local topography and habitat where a large red triangle marks the exact location of the underground cave for the *M. punicus* colony.

annual refuge. This multi-specific natural cave hosts several nocturnal species and is characterized by a rocky texture. The cave entrance is highly camouflaged among the rocks, with a diameter not exceeding 50 cm (Figure 2). Internally, the cavity extends up to 250 m in length, with a ceiling height varying between 0.5 and 4 m. The average internal temperature is 17°C, and the relative humidity – a critical factor for bat survival – remains above 65% throughout the site.

Sample collection, analysis and identification

Samples were collected on a weekly or biweekly basis between March 2023 and November 2023. This timeframe was specifically selected to capture the bats' active period and intentionally avoid their hibernation phase. During winter, the scarcity of insects induces a prolonged hibernation in insectivorous bats, which is physiologically characterized by significant weight loss and a decrease in internal body temperature, though drastic drops in ambient temperature can sometimes trigger brief awakenings (Dietz et al. 2009).

To collect the samples, a sheet of paper was placed beneath the roost and retrieved every fifteen days to gather fresh droppings. The collected guano was initially placed in tubes and subsequently dried in small paper cones, which were meticulously labelled with the collection date and location.

Over the course of the study, 31 field visits were conducted. During each visit, a sample comprising more than 20 guano pellets was collected, resulting in a total of 620 pellets for analysis (Table 1). According to Kervyn (1998), analyzing 100 pellets annually is sufficient to definitively identify the prey items consumed, although larger samples are necessary to precisely quantify dietary composition and track variations across years. Therefore, our sample size of 620 pellets provides a highly robust dataset for evaluating the species' diet.

Based on Beck (1995), researchers can determine the diet and preferred prey groups by examining the quality and quantity of bat faecal pellets. This method has been confirmed as an ideal approach to obtain comprehensive data on food components, preferred prey, seasonal and regional variations, and indirect inferences regarding hunting areas and strategies. The diet of *M. punicus* was studied using the collected faecal material. Because the colony usually stayed in the same place in the cave during our survey, a sheet of paper was placed underneath to collect all the guano.

Figure 2. Presentation of the study site and the focal species. Left, the natural habitat in the Bouira highland region. Right, an adult *M. punicus* captured at the study site.

Table 1. Sampling dates and number of samples collected per month.

Months	MAR	APR	MAY	JUN	JUL	AUG	SEP	OCT	NOV	Total
Sampling date	01/3/2023 14/3/2023 29/3/2023	6/4/2023 22/4/2023	3/5/2023 21/5/2023	1/6/2023 12/6/2023 29/6/2023	2/7/2023 11/7/2023 21/7/2023 31/7/2023	1/8/2023 25/8/2023	1/9/2023 15/9/2023 28/9/2023	1/10/2023 14/10/2023 20/10/2023 29/10/2023	1/11/2023 13/11/2023 27/11/2023	
Number of treats	3	2	2	3	4	2	3	4	3	31
Number of pellets collected	60	60	72	80	67	73	65	75	68	620

In the laboratory, the guano was soaked for at least one hour in 70% ethanol to sterilize it and eliminate all pathological germs, before being dissected using forceps under a binocular microscope. The contents of each pellet were spread into a Petri dish and dried. Chitinous remains were then identified and counted under a microscope, with each distinct fragment recorded as a separate item.

No attempt was made to reconstruct whole individuals. To identify the prey items to the lowest possible taxonomic resolution, we used the identification keys provided by Shiel (1991) and García et al. (2021). Following the methodology of Vaughan (1997) and David et al. (2011), the percentage frequency of occurrence was used to express the results.

Data analysis

The data analysis included in this research focused on alpha diversity metrics, specifically: the abundance of the species (*n*), the richness of the species (*S*), and the relative abundance of the species (RA %), which is the ratio of the number of individuals of each species to the total number of individuals of all the species in a particular community.

Additionally, species diversity (*H'*) was determined using the Shannon-Weaver diversity index, calculated as follows: $H' = \sum_{i=1}^{S} Pi \times \ln P_i$, where $pi = ni/N$, ni is the number of individuals of species *i*, *N* is the total number of individuals of all species, pi represents the relative abundance of the species, and *S* is the total number of species; Finally, species evenness (*E*) was calculated using the formula: $E = \frac{H'}{H_{max}}$, where *H'* is the Shannon diversity index, H_{max} = Ln*S*, and *S* is the total number of species.

To investigate the patterns of prey selection and visualize associations between sampling periods and prey taxa, a factorial correspondence analysis (FCA) was performed on a contingency table of fragment counts categorized by taxon and month.

To visualize seasonal shifts in prey consumption, a Cluster Hierarchical Analysis (CHA) was generated using PAST software (Version 5.0; Hammer et al. 2001). Furthermore, to statistically evaluate the overall effect

Figure 3. Samples parts of insect preys found in the diet of *M. punicus*: A – Coleoptera, B – Trichoptera, C – Diptera, D – Hymenoptera, E – Orthoptera, F – Hemiptera.

of seasonality on diet composition, a Permutational Multivariate Analysis of Variance (PERMANOVA) was conducted. This was followed by a test for Homogeneity of Multivariate Dispersions to assess whether the variability in prey consumption differed significantly among seasons. These multivariate statistical analyses were based on a Bray-Curtis distance matrix and performed using R software (Version 4.5.2; R Core Team 2024), utilizing the 'vegan' package.

RESULTS

M. punicus consumed prey from three different classes of arthropods in our study area: the first class was Insecta, representing a frequency of 94%, followed by Arachnida (4.99%) and Chilopoda (1.01%) (Table 2).

A total of 2,684 identifiable prey fragments were recovered from the 620 guano pellets, representing 11 insect orders, two arachnid orders, and one chilopod order. Insects dominated the faeces, while the 27 items from the class Chilopoda made up only 1.01% of the total (Figure 3).

The most abundant order was Hymenoptera (24.48%), comprising Ichneumonidae (16.84%), Chalcidoidea (7.41%), and Formicidae (0.15%); moths (Lepidoptera) represented the second most numerous group (18.82%). Additionally, Coleoptera constituted a major portion of the diet (18.37%), including Scarabaeidae (9.54%) and its subfamily Cetoniinae (0.34%), Carabidae (4.28%), Curculionidae (2.09%), Cerambycidae (1.30%), and smaller proportions of Staphylinidae (0.41%). The order Diptera was also significant (12.15%), represented by nine families across three suborders, with a predomi-

Table 2. Number of identifiable prey fragments of arthropods found in the guano of *M. punicus*.

Class	Order	S. Order	Family	Super family	Number	Percentage
Insecta	Lepidoptera		/		505	18.82
	Siphonaptera		/		41	1.53
	Hymenoptera	Apocrita	/	Chalcidoidea	199	7.41
			Ichneumonidae		454	16.90
		/	Formicidae		4	0.15
	Diptera	Cyclorrhapha	Calliphoridae		52	1.94
			Sphaeroceridae		55	2.05
			Syrphidae		2	0.07
			Phoridae		3	0.11
		Nematocera	Tipulidae		88	3.28
			Anisopodidae		95	3.54
			Chironomidae		40	1.49
		/	Mycetophilidae		1	0.04
		Brachycera	Empididae		3	0.11
	Hemiptera	Homoptera	Cercopidae		123	4.58
		Heteroptera	Cimicidae		41	1.53
			Corixidae		18	0.67
			Acanthosomatidae		8	0.30
	Coleoptera	Polyphaga	Scarabaeidae		256	9.54
			Scarabaeidae (Cetoniinae)		9	0.34
			Staphylinidae		25	0.41
			Curculionidae		56	2.09
			Cerambycidae	Chrysomeloidea	35	1.30
		Adephaga	Carabidae		115	4.29
	Dermaptera		Forficulidae		154	5.74
	Neuroptera		Chrysopidae		10	0.37
			Hemerobiidae		10	0.37
	Trichoptera		Limnephilidae		76	2.72
			Polycentropidae		4	0.15
			Hydropsychidae		4	0.15
	Ephemeroptera		Ephemeridae		23	0.86
	Orthoptera		Gryllidae		30	1.12
Arachnida	Araneae		/		102	3.80
	Opiliones		/		32	1.19
Chilopoda	Chilopoda		/		27	1.01
			Total		2684	100

nance of the families Anisopodidae (3.54%), Tipulidae (3.28%), and Sphaeroceridae (2.05%). Other dipteran families were present at lower frequencies (<2%), including Calliphoridae (1.94%), Chironomidae (1.60%), Phoridae (0.11%), Empididae (0.11%), Syrphidae (0.07%), and Mycetophilidae (0.03%). The order Hemiptera was represented by four families (Cercopidae, Cimicidae, Corixidae, and Acanthosomatidae), ranging from 0.30% to 4.58% in frequency. Dermaptera was represented by the family Forficulidae (5.74%), while arachnids accounted for 4.99% of the diet, including Araneae (3.80%) and Opiliones (1.19%). The order Trichoptera constituted a smaller proportion of the diet, represented by three families: Limnephilidae (2.72%), Polycentropidae (0.30%), and Hydropsychidae (0.30%). Other taxa were found in trace quantities; specifically, Orthoptera, Neuroptera, Ephemeroptera, and Siphonaptera collectively constituted 4.25% of the prey.

Regarding the alpha diversity of the diet, the calculated ecological indices reflected a high diversity of consumed prey, with a Shannon-Weaver index (*H'*) of 2.10, a Simpson index of 0.15, and an evenness (*E*) of 0.79.

Finally, there were statistically highly significant variations in the frequency of different prey taxa (G-test, G = 2889.6, $p < 0.0001$).

A Cluster Hierarchical Analysis (CHA), merging a heatmap with a dendrogram, was used to visualize shifts in prey consumption (Figure 4). *M. punicus* consumed prey from 14 arthropod orders between March and November. Throughout all study seasons, Hymenoptera, Lepidoptera, and Coleoptera constituted the main part of the diet, displaying the highest counts (represented by the red-yellow cells). Conversely, orders such as Neuroptera, Ephemeroptera, Siphonaptera, Chilopoda, and Opiliones were consistently rare (dark blue cells) across all seasons. During spring, the heatmap exhibits moderate consumption of the main orders (Hymenoptera, Lepidoptera, Coleoptera, Diptera, and Hemiptera). In summer, the counts of Hymenoptera and Lepidoptera reached their peak, while Coleoptera and Diptera also persisted in significant numbers. During autumn, the occurrence of most orders decreased; however, Hymenoptera, Coleoptera, Araneae, and Trichoptera continued to be present in the diet.

The dendrogram clusters the taxonomic orders into distinct branches based on similarity in their seasonal consumption patterns. Lepidoptera, Hymenoptera, and Coleoptera form the primary cluster, exhibiting similar seasonal trends. Diptera and Hemiptera are grouped into a second cluster, following a comparable seasonal pattern. Furthermore, Araneae, Dermaptera, and Trichoptera create a third branch. The seasonal tree shows a closer connection between spring and summer compared to autumn, indicating that diet composition remains relatively consistent between these two seasons but undergoes changes later in the year.

The Factorial Correspondence Analysis (FCA) of the diet of *M. punicus* across nine consecutive months projects the structural relationships between prey orders and sampling periods (Figure 5). The first two dimensions contain the fundamental gradients of variation in prey consumption; months positioned in close proximity share similar prey groups, while months that are distant from each other signify changes in dietary constituents. Central months, approximately from March to August, are associated with a diverse range of arthropod orders,

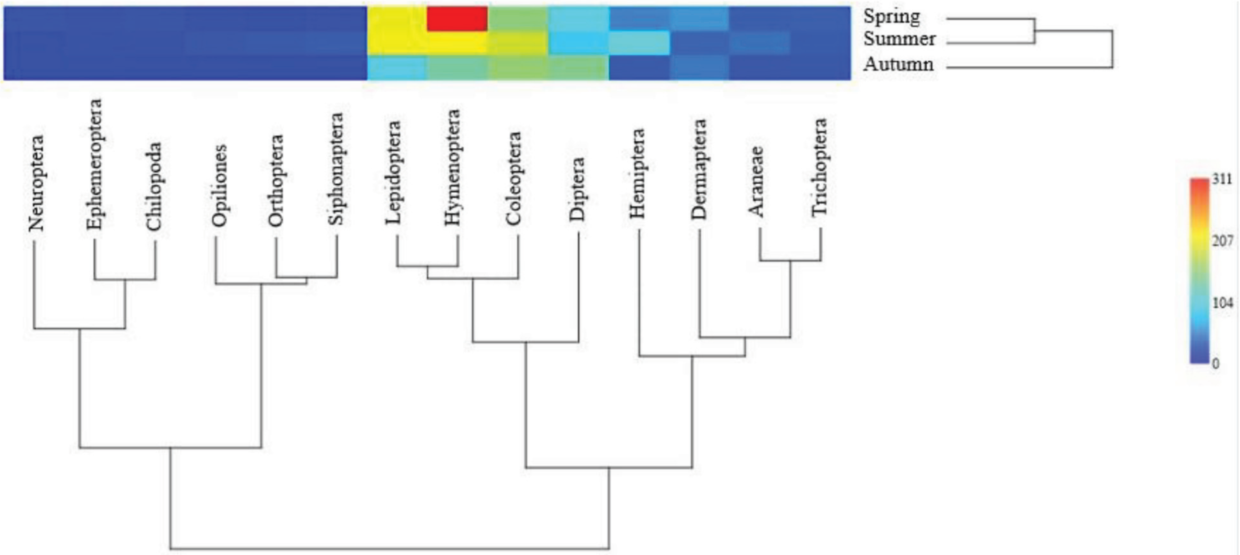

Figure 4. Cluster Hierarchical Analysis (CHA) illustrating seasonal shifts in the diet of *M. punicus.* The colour gradient indicates the absolute number of identified prey fragments, ranging from 0 (dark blue, representing the absence or rare occurrence) to 311 (red, representing the highest abundance). The bottom dendrogram groups arthropod orders based on the similarity of their seasonal consumption patterns.

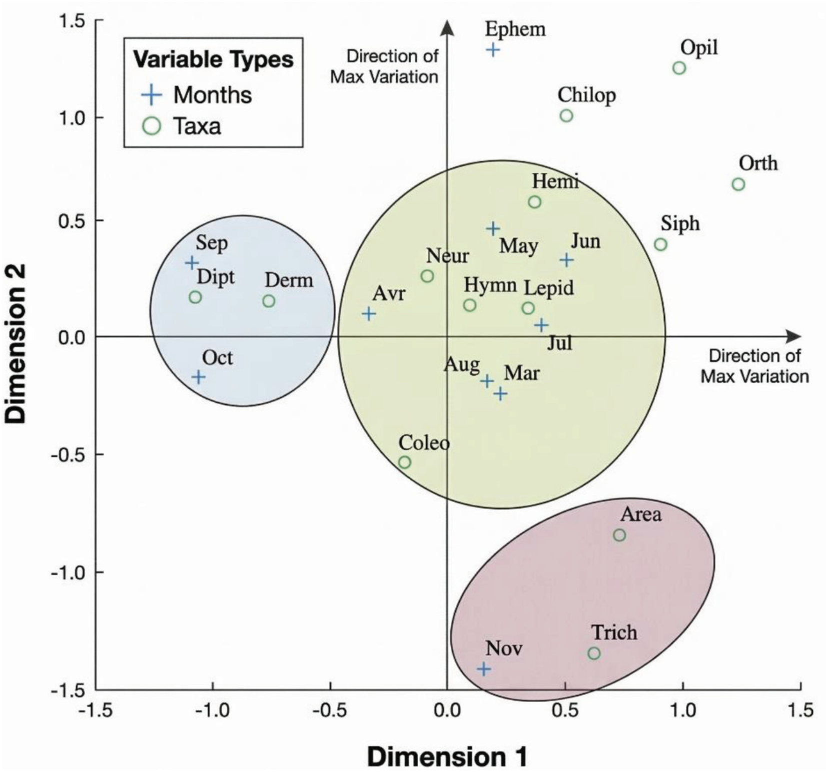

Figure 5. Factorial Correspondence Analysis (FCA) projecting prey orders of *Myotis punicus* at Bouira (Abbreviations: Area: Areanae, Dipt: Diptera, Ephem: Ephemeroptera, Neur: Neuroptera, Chilop: Chilopoda, Hymn: Hymenoptera, Trich: Trichoptera, Coleo: Coleoptera, Hemi: Hemiptera, Opil: Opiliones, Orth: Orthoptera, Siph: Siphonaptera, Derm: Dermaptera, Lepid: Lepidoptera, Mar: March, Avr: April, May: May, Jun: June, Jul: July, Aug: August, Sep: September, Oct: October, Nov: November).

Table 3. Results of the Permutational Multivariate Analysis of Variance (PERMANOVA) and test for Homogeneity of Multivariate Dispersions assessing the effect of season on the diet composition of *M. punicus.*

Analysis	Source of variation	df	Sum of squares	F-value	R^2	*p*-value	Significance
Permanova	Season (model)	2	0.251	3.100	0.608	0.016	Significant
	Residual	4	0.162		0.392		
	Total	6	0.412		1.000		
Homogeneity	Season (groups)	2	0.026	28.036	–	0.001	Significant
	Residual	4	0.002		–		

including Hemiptera, Coleoptera, Neuroptera, and Hymenoptera. In contrast, prey groups such as Ephemeroptera, Chilopoda, Orthoptera, and Opiliones are distinctly positioned toward the upper right of the plot.

Late-season months, particularly September and October, exhibit a distinct shift toward Diptera and Dermaptera. Furthermore, the FCA reveals an extremely isolated distribution for Trichoptera and Araneae, positioning them far from the central core of the active season and closer to the month of November.

The statistical validation of seasonal variations confirmed a significant overall effect of seasonality on diet composition (Permanova: $F = 3.10$, $R^2 = 0.608$, $p = 0.016$; Table 3). The season factor explained approximately 60.8% of the dietary variation. Furthermore, the test for homogeneity of dispersions revealed significant differences in the variability of prey consumption among the seasons ($F = 28.03$, $p = 0.001$). This statistical pattern aligns with our visual CHA and FCA models, confirming that temporal dynamics strongly influence prey selection throughout the bat's active period.

DISCUSSION

In order to understand the adaptive significance of insectivorous bat families, it is imperative to obtain a comprehensive understanding of their feeding habits and foraging behaviour (Neuweiler 2000; Solari and Baker 2007). The diets of bats as primary nocturnal insect consumers (Kunz 1974) encompass a variety of insect groups (Kunz and Kunz 1982), and a single bat's consumption can account for up to half of its body weight. Consistent with previous research, our findings confirm that *M. punicus* primarily consumes Diptera, Lepidoptera, and Coleoptera, alongside other arthropods (Puechmaille et al. 2023). In our study area, the diet was predominantly composed of Hymenoptera (24.48%, primarily Ichneumonidae), followed by Lepidoptera (18.82%) and Coleoptera (18.37%).

M. punicus notably corresponds to the "wide-niche" category when compared to other bats; its dietary breadth qualitatively aligns with the behaviour of known generalists. For instance, studies on *Myotis lucifugus* indicate a similarly diverse order-level diet across several environments, which is considered indicative of significant foraging flexibility (Agosta et al. 2006; Wray et al. 2017). Furthermore, multi-species analyses of insectivorous bats in Mediterranean and Asian habitats reveal that species identified as wide-niche generalists, such as *Rhinolophus ferrumequinum* and its congeners, maintain broad diets (Deng et al. 2023). Conversely, narrow specialists exhibit highly restricted diets essentially consisting of a single order, as represented by *Myotis rufus* in southern Brazil (Silva et al. 2023). Relatively lower prey taxon diversity is also usual in more specialized or resource-limited habitats, as observed for *Myotis myotis* in urbanized western Poland (Zipperer et al. 2000; Graclik and Wasielewski 2012) and for serotine bats *(Eptesicus serotinus)* in temperate Europe (Tiede et al. 2020). The high Shannon and Simpson indices for ordinal-level data, together with significant evenness, position *M. punicus* closer to generalists with broad niches and distinctly above specialists, suggesting that it utilizes its highland ecosystem to exploit various prey taxa rather than concentrating on a singular insect order.

These dietary proportions reflect a flexible generalist foraging strategy. During spring and summer, the bats concentrate on a broad spectrum of prey rather than specializing in a single taxon. The presence of flying insects (such as Hymenoptera and Diptera) proves that *M. punicus* frequently forages along forest edges, using an aerial-hawking strategy similar to other *Myotis* species adapted to semi-open habitats (Norberg and Rayner 1987; Aldridge and Rautenbach 1987; Ahmim and Moali 2011). Simultaneously, the consumption of non-flying or surface-dwelling prey (such as Coleoptera, Arachnida, and Chilopoda) indicates concurrent gleaning behaviour. Ichneumonid parasitoids, in particular, are slow-flying insects that patrol or linger near plants while hunting (Quicke 2015). This behaviour makes them particularly susceptible to bats combining aerial hawking with short-range gleaning along forest borders and beneath the highland Aleppo pine canopy.

Comparing our findings with existing literature reveals significant geographical variation in the diet of *M. punicus*, demonstrating its capacity to opportunistically consume locally available resources (Kowalski et al. 1986; Kowalski and Rzebik-Kowalska 1991; Beuneux 2004). An analysis of prey composition across different Mediterranean regions highlights distinct ecological adaptations.

In island Mediterranean habitats, such as Corsica and Malta, the diet is heavily dominated by Orthoptera (36% in Corsica), alongside significant proportions of Coleoptera and Lepidoptera (Borg 1998; Beuneux 2004). This reflects a more open, scrub-dominated landscape where ground-dwelling orthopterans constitute the main prey resource. Conversely, in the lowland regions of northern Algeria, Diptera emerges as the primary prey, accounting for over half of the consumed taxa (Ahmim and Moali 2011; Ahmim 2014). This shift is facilitated by milder winters and wetter environments closer to coastal influences, which support prolonged activity periods for aquatic and semi-aquatic dipterans.

In contrast, our study area in the Algerian highlands presents a unique dietary profile dominated by Hymenoptera. This semi-arid to sub-humid bioclimatic zone (Fischer et al. 2022) supports a forest massif dominated by *Pinus halepensis* and *Juniperus oxycedrus*, which hosts a complex arthropod food chain (Korenko et al. 2022). The dense populations of parasitoid wasps (Ichneumonidae and Chalcidoidea), which exhibit an elevated adaptation to habitat structure (Kruidhof et al. 2015), provide an abundant resource that *M. punicus* heavily exploits in this high-elevation ecosystem. Additionally, the significant presence of Coleoptera aligns with patterns observed in many insectivorous bats across diverse habitats (Graclik and Wasielewski 2012; Smirnov and Vekhnik 2014).

The seasonal diet of *M. punicus* in the study area precisely tracks the phenological variations of the highland arthropod population. Research indicates that bats opportunistically consume the resources that are accessible to them, meaning their dietary variety directly reflects prey availability (Tiede et al. 2020; Divoll et al. 2022). During spring (March–May), parasitic wasps and early-emerging Lepidoptera dominate. As summer progresses (June–August), the bats exploit an expanding assemblage of ground- and foliage-associated beetles and flies. However, a distinct shift in prey selection occurs in autumn as overall insect availability decreases. During this period (September–November), parasitoid wasps and Lepidoptera decline, prompting the bats to forage opportunistically on late-season taxa, such as specific Diptera and Dermaptera. This adaptive shift is consistent with observations of other bat species expanding their dietary breadth during periods of reduced resource availability (Vallejo et al. 2023). Because

M. punicus relies considerably on these dynamic insect populations, environmental stressors such as climate warming, drought, and habitat fragmentation (Dikman and Doncaster 1987) could disrupt these delicate food webs, forcing a shift toward less profitable prey or necessitating wider foraging ranges.

Statistical tests confirm that seasonality significantly drives overall dietary variation. Rather than abruptly switching prey bases between seasons, *M. punicus* maintains a continuous reliance on its primary insect orders (Hymenoptera, Lepidoptera, and Coleoptera), making gradual, proportional adjustments to its foraging focus based on emerging ecological factors and localized prey availability.

While guano analysis remains the most ethical and practical technique for examining insectivorous bat diets, this strategy is exposed to multiple biases that may systematically influence dietary assessments. Specifically, differential digestibility of arthropod chitin, size-dependent fragment detection, and taxonomic identification limitations form a bias profile that frequently favours the recovery of hard-bodied, larger insects over soft-bodied, smaller insects. Soft-bodied arthropods, notably those in the Diptera and Hemiptera orders, are digested more completely by the acidic mammalian chitinase found in bat stomachs (Gianfranceschi et al. 2013), leaving minimal recognizable chitinous residues. Research combining conventional microscopic examination with DNA metabarcoding frequently finds significant differences, with metabarcoding discovering much larger proportions of aquatic and semi-aquatic soft-bodied organisms (Ling et al. 2023). This suggests that our estimated Diptera frequency (12.15%) may greatly underestimate their real dietary implications. Conversely, hard-bodied arthropods with tough chitinous structures, such as Coleoptera and Orthoptera, are undigested and leave more recognizable remnants (Mason et al. 2016). Therefore, while the 18.37% frequency of Coleoptera reflects realistic food selection, it may also be artificially inflated by the specific conservation of hard elytra relative to softer body areas.

Furthermore, our protocol recorded each distinct chitinous fragment as a separate item. While this is a standard approach in dietary studies, it introduces a specific counting bias: insects with numerous hard parts (beetles with robust elytra, mandibles, and heavily sclerotized legs) inevitably produce more countable fragments per individual than soft-bodied insects like moths or flies. Consequently, the relative abundance of heavily sclerotized taxa such as Coleoptera may be further overestimated in our frequency calculations. Calculating the Minimum Number of Individuals (MNI) is a recognized method to mitigate this bias; however, as our methodology did not involve reconstructing whole individuals or pairing bilateral appendages during the initial sorting, MNI could not be calculated for this dataset. We acknowledge this as a limitation of our study, and we recommend that future dietary assessments of *M. punicus* incorporate MNI alongside fragment counts to provide a more conservative estimate of prey consumption.

Finally, the recovery of Siphonaptera fragments from the guano samples warrants specific clarification. As wingless ectoparasites, fleas are highly unlikely to be targeted as actual prey during the bat's foraging flights. Instead, their presence in the faecal material is most likely the result of two non-foraging mechanisms. First, bats frequently engage in self-grooming and allogrooming, during which they may incidentally ingest their own ectoparasites. These ingested fleas would then pass through the digestive tract and appear in the guano alongside true dietary items. Second, direct environmental contamination of the samples is highly probable; fleas dislodged from the roosting bats may simply fall directly onto the collection sheets beneath the colony. Consequently, while Siphonaptera fragments were recorded in our analysis, they should be interpreted as an artifact of grooming behaviour and roost contamination rather than a deliberate component of the bat's selected trophic niche.

CONCLUSION

The diet of *M. punicus* in the Algerian highlands consists of three main arthropod classes: Insecta, Chilopoda, and Arachnida. Consistent with its classification as an opportunistic insectivore (Beuneux 2004), insects constitute the primary component of its diet (94%), with Hymenoptera, Lepidoptera, and Coleoptera being the most abundant prey, reflecting local insect availability in this habitat. This dietary flexibility underscores the bat's capacity to adjust to seasonal and geographical fluctuations in prey availability when compared to other study areas, establishing it as an important factor in regulating local arthropod dynamics. Moreover, while it should be acknowledged that morphological guano examination may be slightly biased, it remains the most ethical methodological alternative. Certainly, the application of genetic techniques to determine the diet of bats offers a higher level of taxonomic precision for prey identity than is possible via direct microscopic examination of faecal material (Lino et al. 2014). The observed foraging strategies (edge space aerial hawking, narrow space flutter detecting, and narrow space aerial hawking/gleaning) align with this diverse diet, further emphasizing the importance of these bats in insect pest control (Alviola et al. 2023) and highlighting their significant ecological value to local communities.

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Ethics Approval

This study involved the collection of fecal samples (guano) from the Maghrebian mouse-eared bat (*Myotis punicus*). No direct interaction or harm was caused to the individual bats during the sample collection process. All applicable international, national, and institutional guidelines and protocols for the ethical treatment and conservation of wildlife were strictly followed. This article does not involve any studies with human participants conducted by the authors.

Data Availability Statement

All data supporting the findings of this study are available within the paper. The guano samples collected from the Maghrebian mouse-eared bat (*Myotis punicus*) and the analyzed arthropod fragments (Insecta, Chilopoda, and Arachnida) are stored in the first author's collection at the Ecology and Environment Research Laboratory, University of Bejaia, Bejaia, Algeria. These materials are available for access upon request to the curator.

REFERENCES

Agosta, S.J., Morton, D., Marsh, B.D., & Kuhn, K.M. 2006. The influences of habitat on the diet of little brown bats (*Myotis lucifugus*). *Journal of Mammalogy* 87(1), 139–145. https://doi.org/10.1644/05-MAMM-A-277R2.1

Ahmim, M. 2014. Ecology and conservation biology of bats in the Babors region (Algeria). Doctoral dissertation, University of Bejaia, 183 pp.

Ahmim, M., & Moali, A. 2011. The diet of the Maghrebian mouse-eared bat *Myotis punicus* (Mammalia, Chiroptera) in Kabylia, Northern Algeria. *Ecologia Mediterranea* 37(1), 45–51. https://doi.org/10.3406/ecmed.2011.1346

Alberdi, A. Garin, I., Aizpurua, O., & Aihartza, J. 2012. The Foraging Ecology of the Mountain Long-Eared Bat *Plecotus macrobullaris* Revealed with DNA Mini-Barcodes. *Plos one* 7(4), e35692. https://doi.org/10.1371/journal.pone.0035692

Alberdi, A., Razgour, O., Aizpurua, O., Novella-Fernandez, R., Aihartza, J., Budinski, I., Garin, I., Ibáñez, C., Izagirre, E., Rebelo, H., Russo, D., Vella, E., Zukal, J., & Gilbert, M.T.P. 2020. DNA metabarcoding and spatial modelling link diet diversification with distribution homogeneity in European bats. *Nature Communications* 11(1), 1154. https://doi.org/10.1038/s41467-020-14961-2

Aldridge, H.D.J.N., & Rautenbach, I.L. 1987. Morphology, echolocation and resource partitioning in insectivorous bats. *Journal of Animal Ecology* 56(3), 763–778. https://doi.org/10.2307/4947

Altringham, J.D., & Altringham, J.D. 2011. *Bats: From Evolution to Conservation* (Second Edition). Oxford University Press.

Alviola, P.A., Alviola, M.S., Taray, K.J., Lucañas, C.C., De Guia, A.P.O., Dupo, A.L.B., Cuevas, V.C., Pampolina, N.M., & Lit, I.L. 2023. Dietary analysis of eight insectivorous bats (Chiroptera) from Puting Bato Cave Complex, Burdeos, Polillo Island, Philippines. *Journal of Asia-Pacific Biodiversity* 16(3), 291–299. https://doi.org/10.1016/j.japb.2023.05.003

Arrizabalaga-Escudero, A., Garin, I., García-Mudarra, J.L., Alberdi, A., Aihartza, J., & Goiti, U. 2015. Trophic requirements beyond foraging habitats: The importance of prey source habitats in bat conservation. *Biological Conservation* 191, 512–519. https://doi.org/10.1016/j.biocon.2015.07.043

Beck, A. 1995. Fecal analyses of European bat species. *Myotis* 32, 109–119.

Beuneux, G. 2004. Morphometrics and ecology of Myotis cf. Punicus (Chiroptera, Vespertilionidae) in Corsica. *Mammalia* 68(4), 269–273. https://doi.org/10.1515/mamm.2004.025

Bonnet-Garcia, N. 2003. *Bat protection: Its ecological and health issues*. National Institute of Agricultural Medicine, 123 pp.

Borg, J.J. 1998. Observations on the diet of the Maghrebian mouse-eared bat *Myotis punicus* Felten, 1977 from the Maltese Islands. *The Central Mediterranean Naturalist* 2(4), 169–174.

Bourlat, S.J., Zizka, V.M.A., Scherges, A., Sickel, W., Dieker, P., & Pfenninger, M. 2023. Metabarcoding dietary analysis in the insectivorous bat Nyctalusleisleri and implications for conservation. *Biodiversity Data Journal* 11, e111146. https://doi.org/10.3897/BDJ.11.e111146

Brosset, A. 2000. *Revue d'Écologie (La Terre et La Vie)* 55(3), 303–304.

Cuff, J.P., Windsor, F.M., Tercel, M.P.T.G., Kitson, J.J.N., & Evans, D.M. 2022. Overcoming the pitfalls of merging dietary metabarcoding into ecological networks. *Methods in Ecology and Evolution* 13(3), 545–559. https://doi.org/10.1111/2041-210X.13796

David, M. A., Hadjisterkotis, E., Bosso, L., & Russo, D. 2011. Feeding habits of the Egyptian fruit bat *Rousettus aegyptiacus* on Cyprus Island: A first assessment. *Hystrix, the Italian Journal of Mammalogy* 22(2), 281–289. https://doi.org/10.4404/Hystrix-22.2-4587

Deng, X., Chen, Y., Zuo, Y., Liu, H., Wang, Y., Sun, B., et al. 2023. Diet composition, niche overlap and partitioning of five sympatric rhinolophid bats in Southwestern China during summer. *Frontiers in Ecology and Evolution* 11, 1108514. https://doi.org/10.3389/fevo.2023.1108514

Dickman, C.R., & Doncaster, C.P. 1987. The Ecology of Small Mammals in Urban Habitats. I. Populations in a Patchy Environment. *The Journal of Animal Ecology* 56(2), 629. https://doi.org/10.2307/507

Dietz, C., von Helversen, O., & Nill, D. 2007. *Handbook of the Bats of Europe and Northwest Africa: Biology, Characteristics, Conservation*. Stuttgart: Franckh-Kosmos.

Dietz, C., von Helversen, O., & Nill, D. 2009. Bats of Britain, Europe & Northwest Africa. London: A&C Black Publishers Ltd, 2009. *Revue d'Écologie (La Terre et La Vie)* 65(2), 393.

Divoll, T.J., Brown, V.A., McCracken, G.F., & O'Keefe, J.M. 2022. Prey size is more representative than prey taxa when measuring dietary overlap in sympatric forest bats. *Environmental DNA* 4(6), 1407–1419. https://doi.org/10.1002/edn3.354

Fenton, M.B., & Simmons, N.B. 2014. *Bats: A World of Science and Mystery*. University of Chicago Press.

Fischer, C., et al. 2022. Seasonal and temporal patterns of rainfall shape arthropod communities in arid and semi-arid ecosystems. *Scientific Reports* 12, 4253.

García, R.M., Díez, I.F., Agudo, S.M., Garay, J.A., Cuevas, Á.G., Cifrián, S.I., Fernández, M.Ó.G., & Sánchez, M. 2021. Bat Research & Conservation www. Secemu. Org. *Journal of Bat Research &Conservation* 14(1), 152.

Gianfranceschi, L., Strobel, S., Roswag, A., Becker, N.I., Trenczek, T., & Encarnação, J.A. 2013. Insectivorous bats digest chitin in the stomach using acidic mammalian chitinase. *PLOS ONE* 8(9), e72770. https://doi.org/10.1371/journal.pone.0072770

Graclik, A., & Wasielewski, O. 2012. Diet composition of *Myotis myotis* (Chiroptera, Vespertilionidae) in western Poland : Results of fecal analyses. *Turkish Journal of Zoology* 36(2), 209–213. https://doi.org/10.3906/zoo-1007-35

Hammer, Ø., Harper, D.A.T., & Ryan, P.D. 2001. PAST: Paleontological statistics software package for education and data analysis. *Palaeontologia Electronica* 4(1), 4. https://palaeo-electronica.org/2001_1/past/issue1_01.html

Kervyn, T. 1998. Methods for determining the diet of insectivorous bats. *Arvicola, Proceedings "Amiens 97*", 53–56.

Korenko, S., Sýkora, P., Sadílek, D., Řezáč, M., Korenková, M., & Nachman, R. 2022. Elevation gradient affects the distribution and host utilisation of Zatypotaanomala (Hymenoptera: Ichneumonidae) associated with mesh-web weaving spiders (Araneae: Dictynidae). *Journal of Hymenoptera Research* 93, 81–103. DOI:10.3897/jhr.93.91513

Kowalski, K., & Rzebik-Kowalska, B. 1991. (with Institute of Systematics and Evolution of Animals (Polish Academy of Sciences)). *Mammals of Algeria*. Ossoliński National Institute, Publishing House of the Polish Academy of Sciences.

Kowalski, K., Gaisler, J., Bessam, H., Issaad, C., & Ksantini, H. 1986. Annual life cycle of cave bats in northern Algeria. *Acta Theriologica 31*, 185–206, plate 7. https://doi.org/10.5281/zenodo.13454480

Kruidhof, H.M., Gols, R., Vet, L.E.M., & van Lenteren, J.J.A. 2015. Habitat complexity reduces parasitoid foraging efficiency, but does not prevent orientation towards learned host plant odours. *Oecologia* 179(2), 353–361. https://doi.org/10.1007/s00442-015-3349-9

Kunz, T.H. 1974. Feeding Ecology of a Temperate Insectivorous Bat (*Myotis Velifer*). *Ecology*, 55(4), 693–711. https://doi.org/10.2307/1934408

Kunz, T.H., & Kunz, T.H. 1982. Roosting Ecology of Bats. *Ecology of Bats,* 1–55.

Ling, N., Tempero, G.W., & Schamhart, T. 2023. Using faecal DNA metabarcoding to determine the diet of the long-tailed bat, *Chalinolobus tuberculatus*. *New Zealand Journal of Zoology* 50(4), 303–314. https://doi.org/10.1080/03014223.2023.2240711

Lino, A., Fonseca, C., Goiti, U., & Pereira, M.J.R. 2014. Prey Selection by *Rhinolophus hipposideros* (Chiroptera, Rhinolophidae) in a Modified Forest in Southwest Europe. *Acta Chiropterologica* 16(1), 75–83. https://doi.org/10.3161/150811014X683282

Loumassine, H.E., Marniche, F., Bounaceur, F., & Aulagnier, S. 2019. Seasonal diet of *Asellia tridens* (Chiroptera : Hipposideridae) in North-Western Africa. *The European Zoological Journal* 86(1), 354–362. https://doi.org/10.1080/24750263.2019.1648574

Mason, C., Stevenson, M., Kuhlmann, M., & Hill, D. 2016. Guano morphology has the potential to inform conservation management of British bats. *Mammal Research* 61(3), 213–223. https://doi.org/10.1007/s13364-016-0271-3

Massaad, M., da Silveira Bueno, R., & La Mantia, T. 2022. Priorities and gaps in Mediterranean bat research: evidence from a systematic review for the early twenty-first century. *Hystrix, the Italian Journal of Mammalogy* 33(2), 151–162. https://doi.org/10.4404/hystrix-00534-2022

Neuweiler, G. 2000. *The biology of Bats*. Oxford University Press, Oxford & New York.

Norberg, U.M., & Rayner, J.M.V. 1987. Ecological morphology and flight in bats (Mammalia; Chiroptera): wing adaptations, flight performance, foraging strategy and echolocation. *Philosophical Transactions of the Royal Society B*, 316, 335–427. https://doi.org/10.1098/rstb.1987.0050

Puechmaille, S.J., Dool, S., Beuneux, G., & Ruedi, M. 2023. Newly described and already endangered : A new mammal species endemic to Corsica. *Revue suisse de Zoologie* 130(2), 335–351. https://doi.org/10.35929/RSZ.0108

Quicke, D.L.J. 2015. *The Braconid and Ichneumonid Parasitoid Wasps: Biology, Systematics, Evolution and Ecology*. Wiley-Blackwell.

R Core Team. 2024. R: A language and environment for statistical computing. *R Foundation for Statistical Computing*. Vienna, Austria. https://www.R-project.org/

Riccucci, M., & Lanza, B. 2014. Bats and insect pest control : A review. *Vespertillio* 17, 161–169.

Shiel, C.B., C.M. Mcaney, and J.S. Fairley. 1991. Analysis of the diet of Natterer's bat *Myotis nattereri* and the common long-eared bat *Plecotus auritus* in the West of Ireland. *Journal of Zoology (London)* 223, 299–305.

Silva, C.R., Esbérard, C.E.L., Moratelli, R., Bernard, E., & Garbino, G.S.T. 2023. Seasonal analysis of the diet and trophic niche breadth of insectivorous bats in a Neotropical landscape. *Anais da Academia Brasileira de Ciências* 95: e20220177. https://doi.org/10.1590/0001-3765202320210905

Simmons, N.B., & Cirranello, A.L. 2024. *Bat Species of the World* : *A taxonomic and geographic database*. Version 1.4.

Smirnov, D.G., & Vekhnik, V.P. 2014. Ecology of nutrition and differentiation of the trophic niches of bats (Chiroptera :Vespertilionidae) in floodplain ecosystems of the Samara Bend. *Biology Bulletin* 41(1), 60–70. https://doi.org/10.1134/S1062359014010105

Solari, S., & Baker, R. 2007. Mammal species of the world : A taxonomic and geographic reference. *Journal of Mammalogy* 88, 824–830. https://doi.org/10.1644/06-MAMM-R-422.1

Tiede, J., Diepenbruck, M., Gadau, J., Wemheuer, B., Daniel, R., & Scherber, C. 2020. Seasonal variation in the diet of the serotine bat (*Eptesicus serotinus*) : A high-resolution analysis using DNA metabarcoding. *Basic and Applied Ecology* 49, 1–12. https://doi.org/10.1016/j.baae.2020.09.004

Vallejo, N., Aihartza, J., Olasagasti, L., Aldasoro, M., Goiti, U., & Garin, I. 2023. Seasonal shift in the diet of the notched-eared bat (*Myotis emarginatus*) in the Basque Country: From flies to spiders. *Mammalian Biology* 103(4), 419–431. https://doi.org/10.1007/s42991-023-00353-8

Vaughan, N. 1997. The diets of British bats (Chiroptera). *Mammal Review* 27(2), 77–94. https://doi.org/10.1111/j.1365-2907.1997.tb00373.x

Weterings, R., & Umponstira, C. 2014. Bodyweight-forearm ratio, cranial morphology and call frequency relate to prey selection in insectivorous bats. *Bioscience Journal* 30(4), 1215–1224.

Wilson, D.E., & Reeder, D.M. 2005. *Mammal Species of the World : A Taxonomic and Geographic Reference*. JHU Press.

Wray, A.K., Reilly, ziP.F., Wiggins, E.L., O'Sullivan, K.M., & Francl, K.E. 2017. Increased diet breadth of little brown bats (*Myotis lucifugus*) at their northern range boundary reveals winter foraging and insectivory in a cold-climate bat. *Canadian Journal of Zoology* 95, 723–734. https://doi.org/10.1139/cjz-2017-0017

Zipperer, W., Wu, J., Pouyat, R., & Pickett, S.T.A. 2000. The Application of Ecological Principles to Urban and Urbanizing Landscapes. *Ecological Applications – ECOL APPL* 10, 685–688. https://doi.org/10.2307/2641038