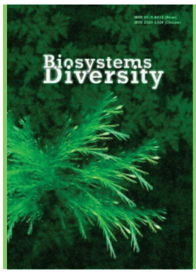

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Feeding ecology of *Tropidophoxinellus callensis* (Cyprinidae) in Algeria

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Understanding species diet is crucial for comprehending ecosystem functions and trophic dynamics within food webs. The Maghreb bleak *Tropidophoxinellus callensis* (Guichenot, 1850) (Cypriniformes, Cyprinidae) is an endemic fish species with populations isolated in Algeria and Tunisia. Unlike other species within the genus, ecological studies on the Maghreb bleak, particularly regarding its diet, are lacking. This study analyzed the diet composition of the Maghreb bleak over an annual cycle in northeastern Algeria. Diet analysis was conducted by examining the contents of 212 digestive tracts collected during seasonal sampling from February to September 2023. The diversity of the Maghreb bleak's diet was assessed based on the presence and abundance of each prey, differentiating between terrestrial and aquatic prey. Additionally, Costello's graphical method was utilized to evaluate potential prey and feeding strategies. Results indicate that insects (relative abundance = 86.1%) were more prevalent in the diet compared to plant material (relative abundance = 7.1%). The diet composition was more diverse in summer (Shannon-Weaver index H' = 4.43) and less diverse in spring (Shannon-Weaver index H' = 1.04). The Maghreb bleak exhibits a diverse diet with an omnivorous feeding strategy. The main prey and their abundance varied seasonally, with a consistent preference for dipteran larvae, namely, *Chironomus* sp. and Simuliidae sp. and plant material of *Alnus glutinosa* as predominant prey items throughout the year. The species also demonstrated opportunistic feeding behavior when these primary prey types were less abundant. These findings provide significant insights into the feeding ecology of the Maghreb bleak, with implications for understanding its role in the ecosystem and informing conservation efforts.

Keywords: Maghreb bleak; ecology; diet variation; digestive tract; Algeria.

Introduction

Basic information on life strategy is essential for studying and analyzing fish population performance, and therefore crucial for both theoretical ecology and fisheries management (Wootton, 1998). Moreover, the study of biological traits is an important aspect of species description, which is vital for understanding the role of biodiversity in ecosystem functioning (Boero, 2011). In the case of native fish, there is a general need to increase understanding of their biological characteristics as an essential tool for conservation programs (Cooke et al., 2012). In the absence of this information, which is very common in areas such as North Africa, it is very difficult to accurately assess the status of species and make appropriate management decisions. Biogeographically, the Maghreb ecoregion has a closer affinity with the Mediterranean part of the Palaearctic kingdom than with the rest of Africa (Lévêque et al., 2008). It is primarily characterized by a low richness of freshwater fish but is home to an interesting diversity of cyprinid species. Furthermore, cyprinids are the largest family with a significant number of species present in this ecoregion, including endemic species, with at least 50% known to have socio-economic value (Juffe-Bignoli & Darwall, 2012).

Feeding specialisations have been considered as important elements influencing the ecology and evolution of many different or-ganisms (Rodriguez et al., 2022). Diet analysis is important for understanding predator-prey interactions, food web dynamics, and ecological processes (Nielsen et al., 2018). In addition, the study of the feeding strategy and variations in the diet of sthe pecies would be relevant to understanding biodiversity and species' resource utilisation (Maran et al., 1998). Identifying species interactions and habitat requirements will grow more essential for effective wildlife conservation and management approaches (Maran et al., 1998). Understanding consumer resource utilisation further informs our understanding of niche ecology, both within and between species (Kratina et al., 2012). The analysis of species diets provides us with an extensive amount of information concerning ecosystem structure and function (Estes et al., 2011). Gut content analysis is a commonly used method for studying the feeding patterns of fish species, incorporating numerous ecological components such as behaviour, habitat use, and energy intake, as well as inter- and intra-specific relationships (Rodriguez et al., 2022). This is the case for the majority of cyprinids that inhabit freshwater and whose diets have been investigated using gut content analysis (Mimeche et al., 2018; Salhi et al., 2021; Vagenas et al., 2022).

Cyprinids are arguably the largest group of freshwater teleosts (Orban & Wu, 2008). Also, it is the largest freshwater fish family in the world (Endersby, 2007). Moreover, Cyprinids are the most diverse family of freshwater fishes in Algeria (Bacha & Amara, 2007). The Maghreb bleak *Tropidophoxinellus callensis* (Guichenot, 1850, Cypriniformes, Cyprinidae), previously known as *Pseudophoxinus callensis* or *Leuciscus callensis*, is part of the Cyprinidae and was recently transferred by Perea et al. (2010) to the genus *Tropidophoxinellus*. This cyprinid is endemic to Northern Algeria and Tunisia (Doadrio, 1994) in North Africa. In Algeria, this species has been described to have a wide endemic distribution in the central and the north-eastern continental hydro-systems (Gauthier, 1928; Pellegrin, 1933; Meddour & Bouderda, 2001; Bacha & Amara, 2007; Zouakh, 2009; Meddour, 2009). Despite being classified as "Least Concern" on the International Union for the Conservation of Nature's (IUCN)

Red List, and not considered a protected fish in Algeria, this species is on the decline due to the multiple threats it faces, including pollution, dam construction and invasive species (Freyhof & Ford, 2022). Studies on freshwater fish in Algeria, especially on the endemic species *T. callensis*, are currently very limited. One study by Salhi et al. (2021) investigated the diet of *Pseudophoxinus* spp. from the El Mellah ravine in the southern part of Algeria in general. In this study, the diet of *Pseudophoxinus* spp. was composed mainly of phytoplankton (algae), while the animal fraction was less represented.

This is the first attempt to examine the diet of *T. callensis* in North-East Algeria, providing insight into their food preferences and feeding strategy. We further investigated seasonal variations in their diet.

Materials and methods

Study area and data collection. The study area is situated in North-Eastern Algeria. It is located in the Babors Kabylia biogeographical zone, in the Jijel and Bejaia provinces (Fig. 1). The climatic conditions in this area are characterized by average annual temperatures between 10 and 20 °C, an average annual precipitation of 900 mm, and a dry season between June and September (Ghorab et al., 2021). Our sampling was carried out in three perennial streams (Fig.1):

– Boulahdaid stream: this stream is located at an altitude of 274 m, in the village of El-Ancer, east from the town of Jijel. The vegetation covering this stream is characterized by a mixed forest consisting mainly of *Quercus canariensis* (Willd, 1809, Fegales, Fagaceae), *Quercus suber* (Linnaeus, 1753), (Fagales, Fagaceae), *Alnus glutinosa* (Linnaeus, 1790), Fagales, Betulaceae) and *Salix* sp.;

– Irjdana stream: this site is located in the village of Bordj T'har, south-east from the town of Jijel, at an altitude of 137 m. It flows under a canopy of *Alnus glutinosa* and *Celtis australis* (Linnaeus, 1753) (Urticales, Ulmaceae) trees.

– Oued Ghir stream: this stream is located in the village of Toudja, west of the town of Bejaia, where the sampling site is located at an altitude of 137 m. The vegetation of this stream is less developed, consisting mainly out of *Salix* sp., *Alnus glutinosa*, *Populus alba* (Linnaeus, 1753) (Salicales, Salicaceae) and *Fraxinus angustifolia* (Vahl, 1804) (Lamiales, Oleaceae).

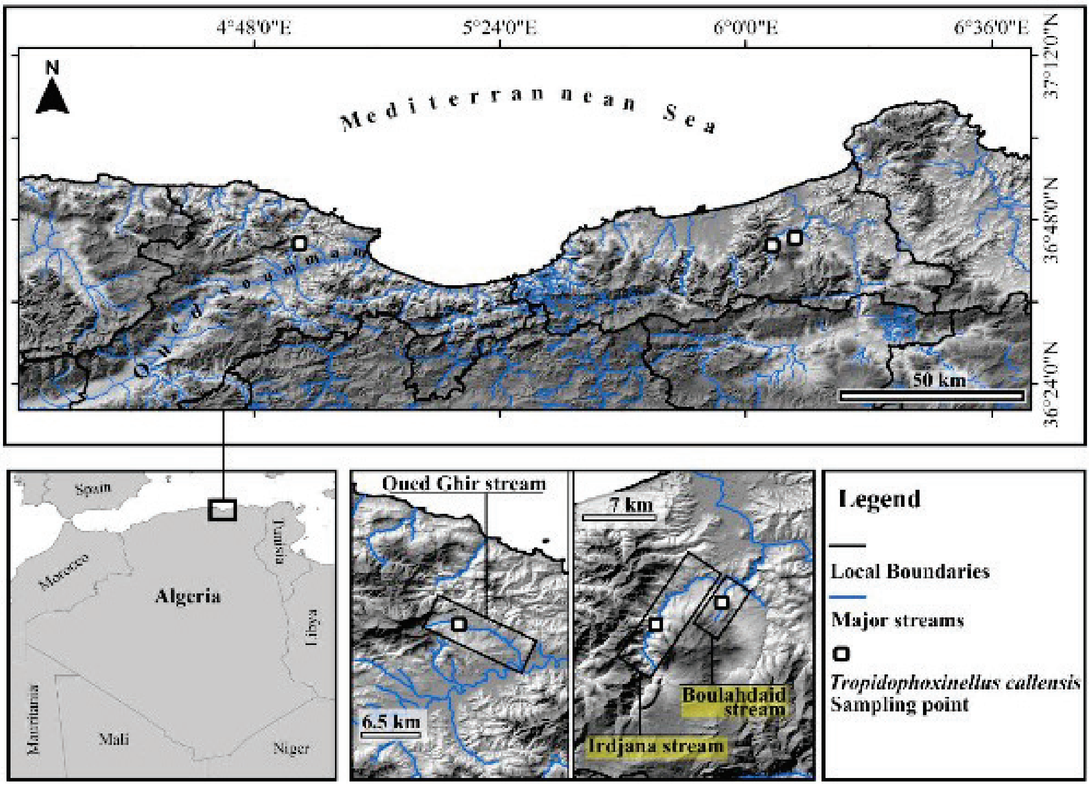

Fig. 1. Map of North-Eastern Algeria, with the collection sites of the Maghreb bleak *Tropidophoxinellus callensis* samples analysed in the study (using ArcMap 10.8, we performed the map creation process and utilized DEM data from opentopography.org to generate the reli base map)

Sample collection. The *T. callensis* specimens used in this study were collected in the field from February to September 2023, using a scoop net. Captured specimens were immediately cooled and brought to the laboratory. A total of 212 digestive tracts (esophagus to the anal opening) were removed, 23 in winter, 54 in spring, 86 in summer and 47 in autumn. These were preserved in 5 ml labeled Eppendorf tubes containing 70% ethanol. These samples were then placed in the fridge at 4 °C until further analysis.

Diet identification and analysis. Each digestive tract sample was investigated in a Petri dish containing water under an Olympus binocular stereo zoom Z410 (magnification range: 0.8–4.7 x 10). The sample was dissected using entomological forceps to separate and recover the fragments of the digested food items. The retrieved diet items were placed in another Petri dish lined with blotting paper. The digested, partly digested and complete food items, such as heads, elytra, mandibles, thoraxes, abdominal segments, and pronotums, were gathered and arranged by taxonomic groups (Boudrissa et al., 2023) to further identify and quantify the animal diet. The identification of each animal taxon was carried out using taxonomical keys (Tachet et al., 2000; Patsia, 2009). We used different fragment characteristics to identify spiders (Calver & Wooller, 1982; Ralph et al., 1985), insects (Tatner, 1983; Ralph et al., 1985; Chinery, 1986; Albouy & Richard, 2017) and molluscs (Tachet et al., 2000; Patsia, 2009). The taxonomic rank identification of ingested prey would depend on the degradation state of the recovered fragments. A complete set, including one head, one elytra, one mandible, one thorax, one pronotum and one chelicerae, was considered one prey item (Boudrissa et al., 2023). Fragments such as insect legs, dipteran wings and molluscs shells (Tatner, 1983) indicated at least the presence of one prey item. The plant diet items were compared and identified using a collection of seeds gathered from the area of sampling sites as a reference.

Ecological analysis. Both the composition and diet diversity of *T. callensis* were considered in the following analyses. The richness (S) corresponded to the total number of diet taxa found in all digestive tracts analysed (Ramade, 1984), while the mean richness (s) represented the average number of diet taxa per digestive tract (Ramade, 1984). The relative abundance (RA%) of each diet taxon was calculated by the percentage of diet items per taxon over the total number of diet items (Hynes, 1950), while the frequency of occurrence (FO%) represented the percentage of digestive tracts where a diet taxa was found over the total number of digestive tracts examined (Hynes, 1950). Additionally, the Shannon-Weaver index (H') was calculated, which is an index of diet diversity (Clarke & Warwick, 2001), and the Pielou's equitability index (J), which is a measure of dietary evenness (Pielou, 1966b). To determine the importance of each diet taxon and the feeding strategy of *T. callensis* in general, the Costello's graphical method was utilised (Costello, 1990), where the relative abundance was assigned on the y-axis and the frequency of occurrence on the x-axis.

Measurement of prey sizes. For the complete animal prey items, the size was measured directly using a graph paper, while the size of digested or partly digested prey items were measured indirectly by measuring their fragments, e.g. heads, elytra, and thorax (Calver & Wooller, 1982). The number of size classes of the prey was determined using Sturge's (1926) formula. The intervals of the size classes were determined by dividing the difference between the largest and smallest size values by the number of identified classes (size class interval = largest size – smallest size/number of size classes).

Diet similarity. In order to compare the diet composition of *T. callensis* between seasons, we applied the Sörensen's similarity index (1948). This index ranges from zero to 100, with zero indicating no similarity and 100 indicating an absolute similarity in diet composition between the two seasons considered.

Aquatic and terrestrial diet items. The diet taxa were classified according to their original habitat, distinguishing between aquatic and terrestrial. Some taxa have a terrestrial or aquatic life cycle, while others have a strictly aquatic larval stage (Tachet et al., 2000; Patsia, 2009). For example, larvae of Diptera (Chironomidae and Simuliidae), Odonata, Ephemeroptera and Trichoptera were considered to originate from an aquatic habitat (Tachet et al., 2000; Zerguine, 2021), while winged adults of ants, adults of Coleoptera (e.g. Staphylinidae), caterpillars of Lepidoptera and plant seeds of *Alnus glutinosa* were classified as diet items originating from a terrestrial habitat (Bougaham et al., 2014).

Statistical analysis. We conducted two types of statistical testing, including a parametric Chi-square test (χ^2) and a non-parametric Kruskal-Wallis test (H), using the software R (version 4.1.2 (2021) by the The R Foundation for Statistical Computing, Vienna, Austria, 2021) and Xlstat (version 2016, Addinsoft, Paris, France, 2016). The average number of diet items per digestive tract and their sizes were analyzed using the non-parametric Kruskal-Wallis test, followed by Dunn's multiple comparisons test to assess the significance of seasonal differences. We opted for the parametric Chi-square test to verify whether there is a relationship between the relative abundances of the different diet groups consumed by *T. callensis* across different seasons. Also, we employed this test to investigate seasonal variations in the relative abundances of both terrestrial and aquatic prey consumed by the fish. Some observations were excluded from the comparison due to their theoretical values being less than five, which didn't correspond to the test's conditions. To highlight the relationship that might exist between each fish size and the size of their prey consumed, we used Pearson's correlation test (t).

Results

Diet composition. The overall diet of *T. callensis* was composed of arthropods (RA = 91.3%), plants (RA = 7.1%) and molluscs (RA = 1.7%). The main diet items were Insecta (RA = 86.1%), followed by Plantae (RA = 7.1%). Bivalvia, Malacostraca, Arachnida and Gastropoda were less prevalent in the overall diet (Fig. 2). The diet composition of *T. callensis* varied significantly across seasons (Chi-squared test, $\chi^2 = 402.32$, df = 6, P < 0.001). Insecta were the most consumed food item in both spring (4.8%) and summer (33.3%), while seeds of Plantae were more prevalent in winter (3.5%). Among the Insecta, Diptera were significantly more abundant in the diet, particularly in spring (38.7%) and summer (16.5%) (Chi-squared test, $\chi^2 = 452.45$, df = 9, P < 0.001), followed by Ephemeroptera (7.2%) and Hymenoptera (4.9%), especially in summer. Of the Diptera, *Chironomus* sp. was the dominant food item in both winter and summer (Table 1), while Simuliidae sp. was the most abundant food item in spring. In autumn, both the water mite Hydracarina sp. and the ant *Camponotus micans* (Nylander, 1856) were abundant. Of the Plantae, *Alnus glutinosa* (RA= 28.4%) and Plantae sp. 2 (RA = 20.8%) were more present in winter and autumn, respectively.

Table 1
Diet composition (relative abundance (RA, %) and frequency of occurrence (FO, %)) of *T. callensis* in Algeria (measured for diet item; prey size in mm)

Order	Prey groups	Prey size, mm	Winter		Spring		Summer		Autumn	
			RA, %	FO, %	RA, %	FO, %	RA, %	FO, %	RA, %	FO, %
Coleoptera	*Apion* sp.*	4.0	0.6	4.3	0.0	0.0	0.0	0.0	0.0	0.0
	Apionidae sp.*	3.0	0.0	0.0	0.0	0.0	0.2	1.2	0.8	2.1
	Cerambycidae sp. 1*	6.0	0.0	0.0	0.2	1.9	0.0	0.0	0.0	0.0
	Cerambycidae sp. 2*	3.0	0.0	0.0	0.0	0.0	0.4	2.3	0.0	0.0
	Cerambycidae sp. 3	—	0.0	0.0	0.0	0.0	0.4	2.3	0.0	0.0
	Cerambycidae sp. 4*	3.0	0.0	0.0	0.0	0.0	0.0	0.0	0.8	2.1
	Chrysomelidae sp.	—	0.0	0.0	0.2	1.9	0.0	0.0	0.0	0.0
	Coleoptera sp. 1	—	0.0	0.0	0.2	1.9	0.0	0.0	0.0	0.0
	Coleoptera sp. 2	—	0.0	0.0	0.2	1.9	0.2	1.2	0.0	0.0
	Coleoptera sp. 3*	3.0	0.0	0.0	0.0	0.0	0.4	2.3	0.0	0.0
	Coleoptera sp. 4	—	0.0	0.0	0.0	0.0	0.5	3.5	0.0	0.0
	Coleoptera sp. 5	—	0.0	0.0	0.0	0.0	0.0	0.0	0.8	2.1
	Curculionidae sp. 1*	1.5	0.0	0.0	0.2	1.9	0.4	1.2	0.0	0.0
	Curculionidae sp. 2*	10.5	0.0	0.0	0.0	0.0	0.4	2.3	0.0	0.0
	Curculionidae sp. 3	—	0.0	0.0	0.0	0.0	0.2	1.2	0.0	0.0
	Curculionidae sp. 4*	6.5	0.0	0.0	0.0	0.0	0.0	0.0	0.8	2.1
	Dytiscidae sp. 1*	4.0	4.3	21.7	0.0	0.0	0.4	2.3	0.0	0.0
	Dytiscidae sp. 2*	2.5	0.0	0.0	0.2	1.9	0.0	0.0	1.7	4.3
	Dytiscidae sp. 3*	3.0	0.0	0.0	0.2	1.9	0.0	0.0	0.0	0.0
	Dytiscidae sp. 4*	2.4	0.0	0.0	0.0	0.0	0.2	1.2	3.3	8.5
	Dytiscidae sp. 5*	8.0	0.0	0.0	0.0	0.0	0.2	1.2	0.0	0.0
	Elmidae sp. *	3.5	0.0	0.0	0.0	0.0	0.9	5.8	0.0	0.0
	Gyrinidae sp.*	3.0	0.6	4.3	0.0	0.0	0.0	0.0	0.0	0.0
	Haliplidae sp.*	5.0	0.0	0.0	0.0	0.0	0.4	2.3	0.0	0.0
	Hydrophilidae sp. 1*	5.0	0.6	4.3	0.0	0.0	0.4	2.3	0.0	0.0
	Hydrophilidae sp. 2	—	0.0	0.0	0.0	0.0	0.0	0.0	0.8	2.1
	Microlestes sp.*	3.0	1.9	13.0	0.0	0.0	0.0	0.0	0.0	0.0
	*Oryzaephylus surinameusis**	3.0	0.0	0.0	0.0	0.0	0.7	3.5	0.0	0.0
	Rhynchitidae sp.*	6.0	0.0	0.0	0.0	0.0	0.2	1.2	0.0	0.0
	*Scymnus mimulus**	6.0	1.2	8.7	0.0	0.0	0.0	0.0	0.0	0.0
	Staphylinidae sp. 1	—	0.0	0.0	0.0	0.0	0.4	2.3	0.0	0.0

Order	Prey groups	Prey size, mm	Winter		Spring		Summer		Autumn	
			RA, %	FO, %	RA, %	FO, %	RA, %	FO, %	RA, %	FO, %
	Staphylinidae sp. 2*	6.0	0.0	0.0	0.0	0.0	0.2	1.2	0.0	0.0
	Staphylinidae sp. 3	—	0.0	0.0	0.0	0.0	0.2	1.2	0.0	0.0
	Staphylinidae sp. 4	—	0.0	0.0	0.0	0.0	0.0	0.0	0.8	2.1
	Tachinidae sp.	—	0.0	0.0	0.0	0.0	0.0	0.0	0.8	2.1
	Tenebrionidae sp.*	3.6	0.0	0.0	0.0	0.0	0.2	1.2	0.0	0.0
Diptera	Brachycera sp.	—	4.9	30.4	0.2	1.9	1.1	7.0	0.0	0.0
	Chironomus sp.	—	35.8	34.8	25.1	66.7	18.8	36.0	5.0	12.8
	Diptera sp. 1	—	0.0	0.0	0.3	3.7	0.0	0.0	0.0	0.0
	Diptera sp. 2	—	0.0	0.0	0.2	1.9	0.9	5.8	0.0	0.0
	Diptera sp. 3	—	0.0	0.0	0.0	0.0	0.2	1.2	0.0	0.0
	Caloparyphus sp.*	5.0	0.6	4.3	0.2	1.9	0.2	1.2	0.0	0.0
	Diptera sp. 4	—	0.0	0.0	0.0	0.0	0.2	1.2	0.0	0.0
	Diptera sp. 5	—	0.0	0.0	0.0	0.0	1.2	3.5	0.0	0.0
	Culicidae sp.	—	0.0	0.0	6.2	37.0	0.9	5.8	0.0	0.0
	Hippoboscidae sp.	—	0.0	0.0	0.0	0.0	0.2	1.2	0.0	0.0
	Muscidae sp. 1*	2.0	0.0	0.0	0.2	1.9	0.2	1.2	0.0	0.0
	Muscidae sp. 2	—	0.0	0.0	0.0	0.0	0.2	1.2	0.0	0.0
	Muscidae sp. 3	—	0.0	0.0	0.0	0.0	0.4	1.2	0.0	0.0
	Simuliidae sp.	—	0.6	4.3	56.5	81.5	18.6	39.5	3.3	8.5
	Simulium sp.	—	0.0	0.0	0.2	1.9	0.0	0.0	0.0	0.0
	Stratiomydae sp. *	2.5	0.0	0.0	0.2	1.9	0.0	0.0	0.0	0.0
	Tipulidae sp. 1*	15.0	0.0	0.0	0.2	1.9	0.4	2.3	0.0	0.0
	Tipulidae sp. 2*	13.0	0.0	0.0	0.0	0.0	0.5	2.3	0.0	0.0
Ephemeroptera	Baetidae sp.	—	0.0	0.0	0.0	0.0	15.0	59.3	3.3	8.5
	Ephemeroptera sp.	—	0.0	0.0	0.0	0.0	1.4	4.7	1.7	4.3
	Oligoneuriidae sp.	—	0.0	0.0	0.6	5.6	0.0	0.0	0.0	0.0
	Potamanthidae sp.	—	0.0	0.0	0.3	3.7	2.5	11.6	0.0	0.0
Hemiptera	Corixidae sp. 1	—	0.0	0.0	0.0	0.0	0.2	1.2	0.0	0.0
	Corixidae sp. 2	—	0.0	0.0	0.0	0.0	0.2	1.2	0.0	0.0
	Hemiptera sp.	—	0.6	4.3	0.0	0.0	0.2	1.2	0.0	0.0
	Veliidae sp.	—	0.0	0.0	1.1	9.3	0.5	3.5	0.8	2.1
Heteroptera	Gerridae sp. 1	—	0.0	0.0	0.2	1.9	0.2	1.2	0.0	0.0
	Gerridae sp. 2	—	0.0	0.0	0.0	0.0	0.7	2.3	0.8	2.1
	Hydrometra sp.	—	0.0	0.0	0.0	0.0	0.5	3.5	0.0	0.0
Hymenoptera	Braconidae sp.	—	0.0	0.0	0.2	1.9	0.0	0.0	0.0	0.0
	*Pheidole pallidula**	3.0	0.0	0.0	0.0	0.0	0.4	2.3	1.7	4.3
	Aphaenogaster sp.	—	0.0	0.0	0.0	0.0	0.9	5.8	0.0	0.0
	*Tapinoma negerrimum**	5.0	0.0	0.0	0.2	1.9	1.6	8.1	0.0	0.0
	*Camponotus micans**	12.0	0.0	0.0	0.0	0.0	0.4	1.2	16.7	21.3
	Camponotus sp.	—	0.0	0.0	0.2	1.9	0.0	0.0	0.0	0.0
	Plagiolepis sp.	—	0.0	0.0	0.0	0.0	0.5	3.5	0.0	0.0
	*Crematogaster scutellaris**	5.0	0.6	4.3	0.0	0.0	6.9	24.4	0.8	2.1
	Formicidae sp.	—	0.0	0.0	0.0	0.0	0.2	1.2	0.0	0.0
	Hymenoptera sp. 1	—	0.0	0.0	0.0	0.0	0.4	2.3	0.0	0.0
	Hymenoptera sp. 2	—	0.0	0.0	0.0	0.0	0.0	0.0	0.8	2.1
	Ichneumonidae sp. 1	—	0.0	0.0	0.2	1.9	0.2	1.2	0.0	0.0
	Ichneumonidae sp. 2	—	0.0	0.0	0.6	5.6	0.0	0.0	0.0	0.0
	Ichneumonidae sp. 3	—	0.0	0.0	0.2	1.9	0.9	5.8	0.0	0.0
	Ichneumonidae sp. 4	—	0.0	0.0	0.0	0.0	0.2	1.2	0.0	0.0
	Ichneumonidae sp. 5	—	0.0	0.0	0.0	0.0	0.4	2.3	0.8	2.1
	Apoidae sp.	—	0.0	0.0	0.0	0.0	0.2	1.2	0.0	0.0
Lepidoptera	Lepidoptera sp.*	4.0	0.0	0.0	0.2	1.9	0.0	0.0	0.0	0.0
Mantodea	Mantodea sp.	—	0.0	0.0	0.0	0.0	0.2	1.2	0.8	2.1
Odonata	Gomphidae sp.	—	0.0	0.0	0.0	0.0	0.2	1.2	0.0	0.0
	Ishnura sp.	—	0.6	4.3	0.0	0.0	0.0	0.0	0.0	0.0
	Odonata sp. 1	—	0.6	4.3	0.0	0.0	0.0	0.0	0.0	0.0
	Odonata sp. 2	—	0.0	0.0	0.2	1.9	0.0	0.0	0.0	0.0
	Odonata sp. 3	—	0.0	0.0	0.0	0.0	0.2	1.2	0.0	0.0
	Odonata sp. 4	—	0.0	0.0	0.0	0.0	0.2	1.2	0.0	0.0
Plecoptera	Plecoptera sp.	—	0.0	0.0	0.0	0.0	0.2	1.2	0.0	0.0
Trichoptera	Beraeidae sp.*	3.5	0.6	4.3	0.0	0.0	0.0	0.0	0.0	0.0
	Hydropsychidae sp.	—	0.0	0.0	0.6	5.6	0.7	4.7	0.0	0.0
	Philopotamidae sp.	—	3.1	21.7	1.2	13.0	1.4	9.3	0.0	0.0
	Trichoptera sp. 1	—	7.4	8.7	0.0	0.0	0.0	0.0	0.0	0.0
	Trichoptera sp. 2	—	0.6	4.3	0.0	0.0	0.0	0.0	0.0	0.0
	Trichoptera sp. 3	—	0.6	4.3	0.0	0.0	0.2	1.2	0.0	0.0
	Trichoptera sp. 4	—	0.0	0.0	0.0	0.0	0.0	0.0	0.8	2.1
	Polycentropodidae sp.	—	0.0	0.0	0.3	1.9	0.0	0.0	0.0	0.0
Bivalvia	Bivalvia sp. 1	—	0.6	4.3	0.0	0.0	1.2	5.8	0.0	0.0
	Bivalvia sp. 2	—	0.0	0.0	0.0	0.0	0.7	4.7	0.0	0.0
	Bivalvia sp. 3	—	0.0	0.0	0.0	0.0	0.5	1.2	0.0	0.0
Gastropoda	Gastropoda sp. 1	—	0.0	0.0	0.2	1.9	0.0	0.0	0.0	0.0
	Gastropoda sp. 2	—	0.0	0.0	0.3	1.9	0.0	0.0	0.0	0.0
	Gastropoda sp. 3	—	0.0	0.0	0.5	1.9	0.0	0.0	0.0	0.0
	Gastropoda sp. 4	—	0.0	0.0	0.2	1.9	0.0	0.0	0.0	0.0
	Gastropoda sp. 5	—	0.0	0.0	0.0	0.0	0.2	1.2	0.0	0.0
	Gastropoda sp. 6	—	0.0	0.0	0.0	0.0	0.4	2.3	0.0	0.0
Aranea	Aranea sp. 1	—	0.0	0.0	0.2	1.9	1.2	8.1	0.0	0.0
	Aranea sp. 2	—	0.0	0.0	0.5	5.6	2.8	16.3	0.8	2.1

Order	Prey groups	Prey size, mm	Winter		Spring		Summer		Autumn	
			RA, %	FO, %	RA, %	FO, %	RA, %	FO, %	RA, %	FO, %
	Aranea sp. 3	—	0.0	0.0	0.0	0.0	0.2	1.2	0.0	0.0
	Aranea sp. 4	—	0.0	0.0	0.0	0.0	0.2	1.2	1.7	4.3
	Aranea sp. 5	—	0.0	0.0	0.0	0.0	0.0	0.0	0.8	2.1
Acari	Hydracarina sp.	—	0.0	0.0	0.0	0.0	1.9	7.0	25.0	2.1
Ixodida	Ixodidae sp. 1	—	0.0	0.0	0.3	3.7	0.0	0.0	0.0	0.0
	Ixodidae sp. 2	—	0.0	0.0	0.2	1.9	0.0	0.0	0.0	0.0
Isopoda	Isopoda sp.	—	0.6	4.3	0.0	0.0	0.0	0.0	0.0	0.0
Malpighiales	*Salix* sp.	—	4.3	17.4	0.0	0.0	0.0	0.0	0.0	0.0
Fagales	*Alnus glutinosa**	2.0	28.4	60.9	1.5	18.5	0.5	2.3	0.0	0.0
Plantae	Plantae sp. 1	—	0.0	0.0	0.2	1.9	0.0	0.0	0.0	0.0
	Plantae sp. 2	—	0.0	0.0	0.0	0.0	1.6	9.3	20.8	53.2
	Plantae sp. 3	—	0.0	0.0	0.0	0.0	0.0	0.0	1.7	4.3
	Plantae sp. 4	—	0.0	0.0	0.0	0.0	0.0	0.0	0.8	2.1
	Bryophyta sp.	—	0.0	0.0	0.0	0.0	0.4	1.2	0.0	0.0
	124 prey species	—	100.0	—	100.0	—	100.0	—	100.0	—

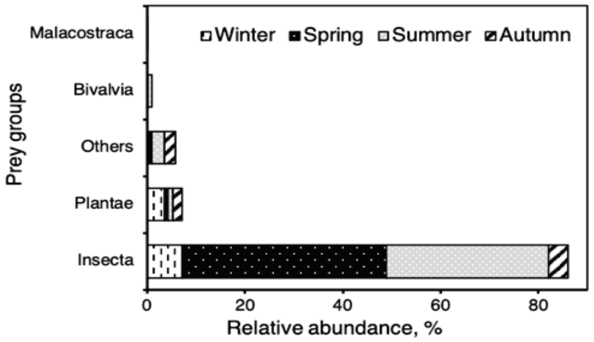

Fig. 2. Barplot representing the overall relative abundance (RA, %) of the taxa found in the diet of *T. callensis* in Algeria over four different seasons

Diet diversity. The analysis of 212 digestive tracts showed the presence of 124 diet taxa (Item). The number of diet taxa varied between 23 in winter and 80 in summer. The mean number of taxa per digestive tract varied between 2.83 (winter) and 3.93 (summer), with an average richness of 3.29. There was a significant difference between seasons in the number of diet taxa per digestive tract (Kruskal-Wallis test, H = 50.353, df = 3, $P < 0.0001$). The results of Dunn's multiple comparisons test indicated that autumn significantly differed from the other seasons ($P < 0.01$). The most pronounced differences ($P < 0.0001$) were observed between autumn and both spring and summer. The diet of the species was more diverse in summer (H' = 4.43), while less diverse in spring (H'= 1.04). There were a few diet taxa that dominated the diet composition of *T. callensis* in spring, while the Pielou's evenness index indicated that a higher number of taxa dominated the diet in the other seasons (Table 2).

Table 2
Diet composition and diversity of *T. callensis*
in Algeria over different season

Characteristics	Winter	Spring	Summer	Autumn	Total
Number of digestive tracts	23	54	86	47	212
Number of measured items	9	10	24	9	52
Total richness (S)	23	43	80	29	124
Mean richness (s)	2.83 ± 1.56	3.77 ± 1.85	3.93 ± 2.28	1.79 ± 0.91	3.29 ± 2.04
Diversity (H')	2.9	1.04	4.43	3.58	4.04
Pielou's evenness index (j)	0.64	0.19	0.7	0.74	0.58

Primary diet items. We identified four primary diet taxa for the *T. callensis*, which included two Diptera (*Chironomus* sp. and Simuliidae sp.), one Ephemeroptera (Baetidae sp.) and one Plantae (Plantae sp. 2). These diet taxa were found in every season, while this fish also ate more of seeds of *Alnus glutinosa* (RA = 60.9%, FO = 28.4%) and Brachycera sp. (RA = 30.4%, FO = 4.9%) in winter (Fig. 3a), Culicidae sp. (RA = 37.0%, FO = 6.2%) in spring, *Camponotus micans* (RA = 42.6%, FO = 16.7%) and Hydracarina sp. (RA = 63.8%, FO = 25.0%) in autumn (Fig. 3d).

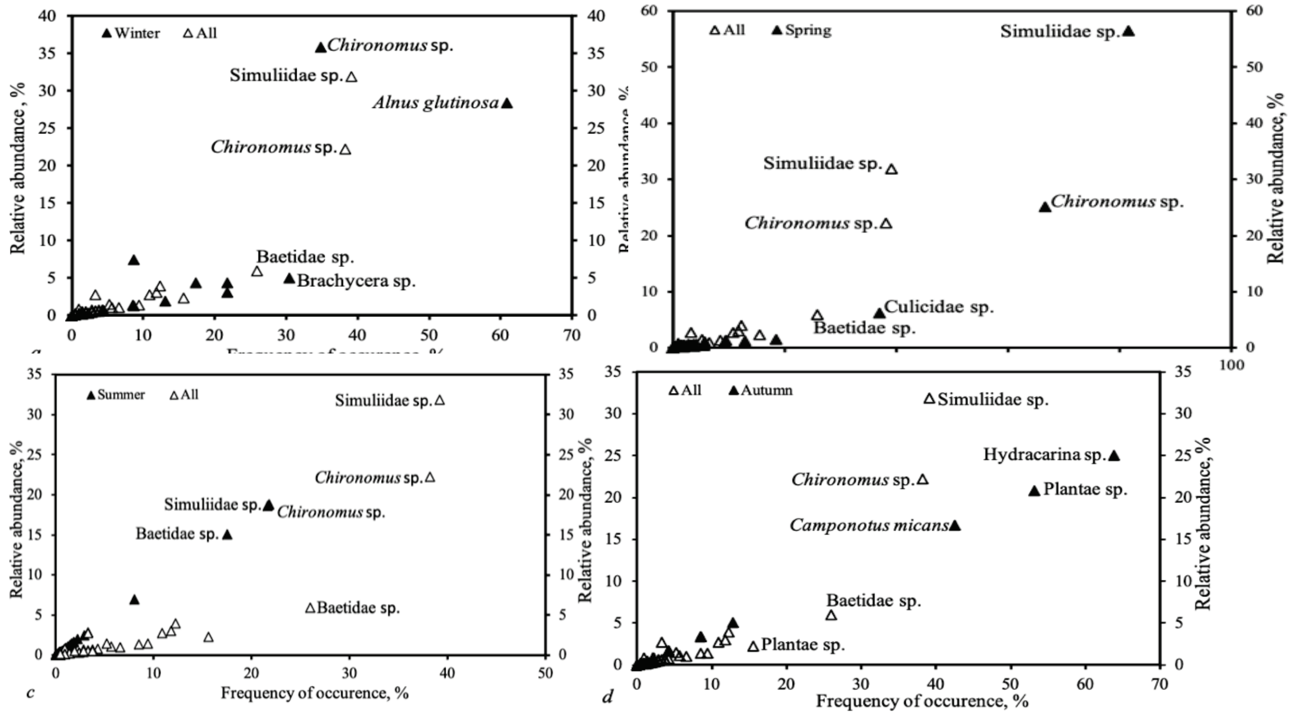

Fig. 3. Costello's graphical representation of the diet composition of *T. callensis* in Algeria, with one graph per season: *a* – winter, *b* – spring, *c* – summer, *d* – autumn

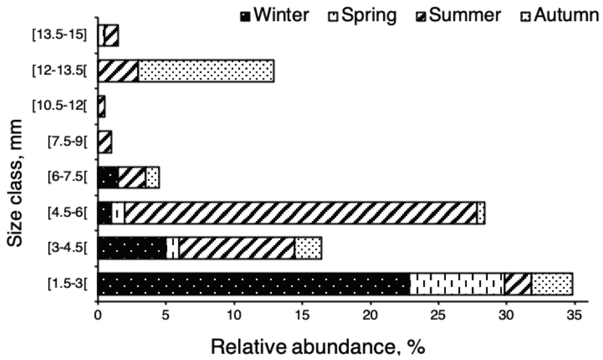

Fig. 4. Barplot representing the relative abundance (RA, %) of each item's size class found in the diet of *T. callensis* in Algeria over all seasons winter, spring, summer and autumn

Class sizes of diet items. The size of the food items of *T. callensis* varied between 1.5 mm (Curculionidae sp. 1) and 15 mm (Tipulidae sp. 1). The highest average size was observed in autumn (7.16, SD = 4.49), while the lowest was measured in winter (3.17, SD = 1.43). A significant difference in the mean food sizes per digestive tract was found between seasons (Kruskal-Wallis test, H = 37.37, df = 3, $P < 0.0001$). Most diet items had a size ranging between 1.5 and 6 mm in winter and summer (Fig. 4), and between 12 and 13.5 mm in autumn (Fig. 4). A positive correlation was only observed between fish size sampled in autumn and those of consumed food items, which was 0.63 (Pearson's correlation test, t = 3.58, df = 19, P = 0.001), while no correlation was detected in the other three seasons (winter, spring, summer).

Seasonal diet variation. There are seasonal dissimilarities in the diet composition of *T. callensis*, whose calculated Sorensen index values are below 35.0%. A weak similarity of 11.5% was noted between winter and autumn. Nevertheless, a value of 34.1% is ob-served between spring and summer.

Aquatic and terrestrial diet items. The abundances of the aquatic and terrestrial diet taxa varied significantly by season (Chi-squared test, χ^2 = 203.21, df = 3, $P < 0.001$). Aquatic taxa were more abundant in winter, spring and summer (RA > 57.0%), while both the aquatic and terrestrial taxa were more abundant in autumn (RA = 50.0%), although the latter occurred more frequently than the former (FO = 106.4% and 66.0%, respectively, Table 3).

Table 3
Abundance (RA, %) and occurrence (FO, %) of taxa grouped by their original habitat, in the diet of *T. callensis* in Algeria over different seasons winter, spring, summer and autumn

Seasons	Winter		Spring		Summer		Autumn	
Habitats	RA, %	FO, %	RA, %	FO, %	RA, %	FO, %	RA, %	FO, %
Terrestrial	42.6	143.5	11.7	101.7	26.0	134.9	50.0	106.4
Aquatic	57.4	134.8	88.3	213.0	74.0	251.2	50.0	66.0

Discussion

This study provides a first insight into the dietary composition of *T. callensis*. We also note that we have no comparative data on the availability of arthropod and aquatic invertebrate studies in our sampling sites. Our findings differ from the study conducted by Salhi et al. (2021) on the diet of *Pseudophoxinus* spp. (now *Tropidophoxinellus* spp.) in general from the El Mellah ravine, where the insects were overall less abundant (RA < 3.5%) and absent in summer. These variations can be explained by differences in habitat and sampling locality, since the El Mellah ravine is located in a semi-arid region in the south of Algeria, while the present study was conducted in a forested area in the north-east of Algeria. Additionally, while Salhi et al. (2021) investigated the diet of the *Tropidophoxinellus* genus in general, our study focussed on the diet of only one specific species, which might further explain the varying observations. Moreover, the environmental and watercourses' characteristics affect the diet of fish where the prey availability in forested streams is more important than the streams located in the agricultural surroundings (Effert-Fanta et al., 2022).

On the other hand, our findings align with those of Vagenas et al. (2022), who observed a significant proportion of insects (32.2%) in the diet of another *Tropidophoxinellus* species, namely *T. hellenicus* (Stephanidis, 1971) (Cypriniformes, Cyprinidae). When focussing on non-animal food taxa, the seeds of *Alnus glutinosa* were more abundant in the diet of *T. callensis* in winter; whereas Salhi et al. (2021) reported that phytoplankton such as *Spirogyra* sp. and *Melosira* sp. represent the main diet components of *Pseudophoxinus* spp. in the El Mellah ravine. This variation could be explained by differences in vegetation along the different water bodies, with *Alnus glutinosa* being more present along the streams sampled in our study and having a higher abundance of mature seeds before and during winter (Gosling et al., 2009). On the contrary, algae show their peak development in the spring and summer due to favourable environmental conditions in climate, water physio-chemistry, and oxygen levels (Fqih Berrada et al., 2000). Eutrophication caused by agricultural activities in the El Mellah ravine might further enhance algae blooms and which could explain elevated levels of algae found in the diet of *Pseudophoxinus* spp. (Salhi et al., 2022). The diet of *T. hellenicus* differs from those of *T. callensis*, particularly in their consumption patterns of certain diet taxa (Vagenas et al., 2022). Unlike *T. callensis*, *T. hellenicus* does not consume Ephemeroptera and Hymenoptera. Additionally, Diptera are less prevalent in the diet of *T. hellenicus* (1.9%) in Greece compared to *T. callensis* in Algeria, where Diptera are the main food item. *Chironomus* sp. (Diptera), *Austrosimulium* sp. (Diptera), and *Ecdyonorus* sp. (Ephemeroptera) were found to be the most abundant Insecta in the digestive tracts of *Pseudophoxinus* spp. in the El Mellah ravine (Salhi et al., 2021), while *T. callensis* contained more *Chironomus* sp. (Diptera), Simuliidae sp. (Diptera) and *Camponotus micans* (Hymenoptera).

In our study, *T. callensis* from Babors' Kabylia consumed overall more food items than *Pseudophoxinus* spp. in El Mellah ravine (Salhi et al., 2022) and *T. hellenicus* in Lake Trichonis in Greece (Vagenas et al., 2022), which each consumes only seven and 13 prey, respectively. This dissimilarity could be linked to the differing structure of the sites investigated in Greece and in Algeria; given that forest streams are more diversified in prey and fish's trophic position is more distinguished by consuming both aquatic and terrestrial prey (Effert-Fanta et al., 2022). At the end of summer, high air temperatures might cause an increase in water temperature and a decrease in water level, resulting in fewer food items and more fish movements in autumn (personal observation). This trend can reduce the amount of prey observed in the fish's digestive tract in autumn compared to other seasons (Salhi et al., 2022). Seasonal variations in the abundance of food taxa in the digestive tract of *T. callensis* can explain the seasonal variations of the diversity index (H'). In fact, the diet of *T. callensis* in spring was less diverse compared to other seasons. This difference might be driven by the high abundance of some dipteran taxa, such as Simuliidae sp. and *Chironomus* sp., in the digestive tracts examined in spring (J = 0.19), possibly driven by a higher presence of these taxa in the environment during this season (Doney et al., 2012; De Carvalho et al., 2019). Another possible explanation is that the foraging behaviour of ectotherm fish might vary seasonally due to fluctuations in the surrounding temperature, with fish becoming less active at lower temperatures in winter and at the beginning of spring (Volkoff & Rønnestad, 2020).

The dipterans Simuliidae sp. and *Chironomus* sp. were the most abundant diet taxa of *T. callensis*. Indeed, these prey items are considered an important trophic resource for fish in general (Rieradevall et al., 1995). The fact that dipterans are widely dispersed throughout the various humid ecosystems of northern Algeria (Cherairia & Samraoui, 2014; Oussad et al., 2021) may explain its high prevalence in the diet of *T. callensis*. Certain groups of dipterans can reproduce several times a year and lay their eggs on the water surface where the larvae further develop (Tachet et al., 2000). The yearly developmental cycle of certain dipteran taxa might explain their high prevalence in the diet during winter, spring, summer. During sampling between spring and late summer, we noted the emergence of large numbers of adult mosquitoes at the study sites (personal observation). Thus, we supposed that the high consumption of *Chironomus* sp. in winter is due to their high caloric content, which is necessary for the breeding period (Encina, 1999; Santos et al., 2013), as well as their low level of movement, which facilitates their capture (Easton & Orth, 1992). However, Salhi et al. (2022) do not mention these Diptera as potentially ingested by fish and they are absent in the summer. On the other hand, dipterans are less prevalent in the diet of *T. callensis* in the autumn. In autumn, dipterans are partly replaced by other freshwater invertebrates such as water mites, and terrestrial food items including ants and plants. A decrease in the availability of preferred food items, might drive fish towards the consumption of other available food items and as such diversifying its diet (Effert-Fanta et al., 2022). Furthermore, the autumn period coincides with the swarming event of ants (Dehina, 2009), with winged individuals falling into the streams, which might explain the higher prevalence of this group in the diet of *T. callensis* during this season.

As previously stated, we have no elements of comparison for the prey sizes consumed by fish of the genus *Tropidophoxinellus* spp. The sizes of the diet items of *T. callensis* ranged from 1.5 to 6.0 mm. Smaller food items included the most prevalent food items, Simuliidae sp. (3 mm) and *Chironomus* sp. (2 mm). The largest food items were present in autumn, which included mainly the ant *Camponotus micans* (12 mm), where large fish consume large food items. While in other seasons, the fish would consume prey of all size categories, allowing them perhaps to meet certain imperative reproductive needs and adapt to changing environmental conditions (personal observation).

The diet of *T. callensis* in Babors Kabylia was more diversified in terms of prey groups compared to *Pseudophoxinus* spp. (Salhi et al., 2021) and *T. hellenicus* (Vagenas et al., 2022).This result might be due to a difference in food availability, where fish from forested streams consume a greater variety of food taxa in general, and more terrestrial and aquatic invertebrate (Effert-Fanta et al., 2022). According to previous studies conducted in the United States (Jackson et al., 2012), terrestrial invertebrates account for up to 44.0% of cyprinid and other small stream fish diets. In our study, aquatic food items always dominate the diet of *T. callensis* throughout the year, while both kinds of food item are consumed equally in the autumn. The higher presence of aquatic invertebrates in the diet during the wet season, might be due to their higher abundance in the environment (Rodriguez-Silva et al., 2022). Thus, it could be explained by a decrease in the availability of certain diet items that are abundant and consumed during other seasons, which is induced by changes in the hydrobiological structure of watercourses (Bougaham et al., 2019). In addition, invertebrates of terrestrial origin could constitute a more energy-rich food source for the fish than invertebrates of aquatic origin (Effert-¬Fanta et al., 2022), particularly during the pre-breeding period.

Conclusion

Unlike other Cyprinidae species, *T. callensis* in Algeria presents a large range of taxa in its diet. This fish consumes arthropods, plant material and molluscs, revealing a tendency towards an omnivorous feeding strategy. Further, seasonal fluctuations in the diet of *T. callensis* are observed, specialising on one diet item in each season. The consumption of the main prey groups does not fluctuate from winter to summer, while alternative prey items are consumed in autumn. Indeed, the diet is mainly dominated by dipterans larvae and plant material, while in autumn, a more opportunistic feeding behaviour is observed when the main prey becomes less abundant and available. Falling back to diet items from terrestrial origin in autumn indicates some degree of generalised feeding strategy and a response to certain requirements for the reproduction of this fish in forest watercourses.

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