Original Article

First Comprehensive List of Phlebotomine Sand Fly Species (Diptera: Psychodidae) in a Leishmaniasis Focus (Djelfa, Algeria), Including the First Record of *Phlebotomus langeroni* Females in Algeria

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(Received 13 Nov 2023; accepted 13 May 2024)

Abstract

Background: The wide distribution of phlebotomine vectors complicates the leishmaniasis situation in the world, with the risk of spreading from rural to urban areas. Our study investigates for the first time the ecology and distribution of sand fly populations in leishmaniasis focus (Djelfa, Algeria).
Methods: Sampling is performed using light traps from August 2021 to July 2022 at ten sites with different biotopes: two peri-urban stations (Ain Oussera and Hassi Bahbah), one urban station (Djelfa), and three rural stations (Ain El-Bel, Haniet Ouled Salem and Mlaga).
Results: A total of 2,866 specimens with 12 species belonging to *Phlebotomus* and *Sergentomyia* were identified with the first record of *Phlebotomus langeroni* female nationally, revealing a good diversity of up to 1.812 bits. The greatest richness of species was found in rural sites at Ain El Bel station, while the lowest richness was observed in the first peri-urban site of Ain Oussera station. Interestingly, the surveyed species were evenly distributed across all the study sites, in particular *Phlebotomus papatasi* with 57.29%. Statistical treatment revealed a strong positive relationship between temperature and species ($p< 0.001$, $r= 0.91$). Considering the other factors, there was a moderate link between species richness and altitude ($p= 0.01$ $R^2= 0.519$). However, neither rainfall nor wind speed ($p> 0.05$) were correlated with sand fly fauna. Moreover, humidity showed a negative correlation ($p< 0.001$, $r= -0.96$).
Conclusion: The findings of this study on phlebotomine fauna in this endemic region for leishmaniasis provide valuable data to support the success of a vector control program.

Keywords: Sand fly; Environmental factors; *Leishmania*; Djelfa; Algeria

Introduction

Phlebotomine sand flies (Diptera: Psychodidae) are tiny haematophagous insects that take blood meals on a wide range of vertebrate hosts including humans (1–3). These arthropods can potentially transmit important pathogens such as the protozoan parasites of *Leishmania* spp. (4–6). Female sand flies infected with these parasites are known to carry pathogens that cause a range of diseases known as leishmaniasis, which affects humans and dogs causing public and veterinary health issues. The most notable forms including cutaneous leishmaniasis (CL) and visceral leishmaniasis (VL) (7, 8).

In Algeria, as in several Mediterranean and African countries, leishmaniasis is an endemic disease and the risk of important outbreaks is real. This disease has considerably increased over the last years (9). In Algeria, five species of *Phlebotomus* are proven vectors, while three

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Published Online: June 30, 2024

species of *Leishmania* are declared as causative agents of the disease (10). In the Sahara, *Phlebotomus papatasi* (Scopoli, 1780) is the known vector of *Leishmania major* (11). In the northern regions of Algeria, *Ph. perniciosus* (Newstead, 1911), *Ph. perfiliewi* (Parrot, 1930), and probably *Ph. longicuspis* (Nitzulescu, 1930) are vectors of *L. infantum*, causing two forms of the disease: CL and VL (12–15). In several regions of the country such as Ghardaïa in the South, Tipaza in the North-Center, and Annaba in the North-East, the vector *Ph. sergenti* is responsible for the transmission of *L. killicki*, which causes CL (16–19). The Djelfa region is endemic for leishmaniasis, primarily due to its agro-pastoral vocation, arid climate, significant population, and close geographical proximity to highly infected regions including M'sila to the East, Biskra to the South-East and Ghardaia to the South (20–24).

Entomological data specific to this unique geographical region are particularly scarce, highlighting a major lack of epidemiological information on phlebotomine sand flies in the Djelfa region. Therefore, the purpose of this research initiative is to respond to this lack by carrying out an in-depth survey for the first time, by establishing a rigorous and comprehensive taxonomic inventory of the sand fly species present in the area, with particular emphasis on their spatio-temporal dynamics. Consequently, this survey will make a significant contribution to advancing ecological and epidemiological knowledge of leishmaniasis transmission in the Djelfa region and, more widely, throughout central Algeria. Implications of our results for monitoring sand fly populations and, at the same time, the occurrence of leishmaniasis are discussed.

Materials and Methods

Study area

Djelfa region stretches over an area of 66,415 km^2, with a population of 1,538,476 (according to the updated census of 2018), As one of the largest steppe regions in Algeria, Djelfa is located on the high plateaus of central Algeria between the Tellian Atlas in the North and the Saharan Atlas in the South. with coordinates stretching from 2° and 5° East longitude to 33° and 35° North latitude. It comprises 36 communes and is recognized for its arid climate, marked by cold winters and hot summers. This climatic setting has propelled the region into eminence for its dominant pastoral vocation, serving a significant center for the sheep sector, as well as for arboriculture, cereal cultivation, market gardening, and fodder crops.

Sand fly collection and identification

The study sites were selected based on the most infected areas with leishmaniasis, according to epidemiological data from the Department of Health and Population (20). The surveys were carried out across 181 km transect linking the six study stations, in which ten sampling sites were selected (Fig. 1). The sampling took place from August 2021 to July 2022, over a year (i.e., 12 months) with one outing each month. The sites were chosen according to the type of area as follows: peri-urban (Ain Oussera and Hassi Bahbah), urban (center of Djelfa), rural (Ain El-Bel, Haniet Ouled Salem and Mlaga) (Table 1).

To ensure a high capture rate, CDC light traps were used. The selection of this trap was predicated on its effectiveness in attracting phototropic species and its ability to capture active sand flies (25, 26). Four light traps were set up from 7 p.m. to 7 a.m. for one night per month from August 2021 to July 2022. It was strategically placed within and in the vicinity of human and animal habitats, as well as in proximity to vegetation.

Specimens captured using light traps were stored in 70% ethanol. This was followed by dissection, in which we separated the head and genitalia from the body, and then these were clarified with 20% Potassium Hydroxide for 4 hours and Marc-André liquid for 2 hours and then fixed in chloral gum (25). Taxonomic identification was carried out individually with a focus on morphological characteristics of male genitalia and female structures such as spermatheca, pharynx, and cibarium, as described by Abonnenc (25), Dedet et al. (27), El Sawaf and Said (28) and Depaquit et al. (29). For some specimens that require further validations for identification, we used the Phleb Key Tool identification software (30).

Collection of climatic data

The climatic parameters analysed in this study included the monthly data on maximum, average or minimum temperatures, rainfall, and wind speed obtained from the meteorological office of Djelfa province.

Data analysis

The data regarding the phlebotomine fauna was subjected to ecological analysis, employing various ecological indexes related to both composition and structure. The measures of composition that were included are relative abundance ($RA\% = ni/N \times 100$), where ("ni"= number of specimens of each sand fly species and "N"= number of total specimens in the surveyed station); Species richness (S), where ("S"= total number of species identified in each survey); and frequency of occurrence ($FO = Ri/R \times 100$), where ("Ri"= number of surveys containing the surveyed species and"R"= total number of surveys carried out). For the structure index, we used the Shannon-Weaver index ($H' = -\Sigma pi \, log_2 \, pi$), which is expressed in bit units ("pi"= the relative frequency of the species considered and "log_2"= the natural logarithm to base-2) and equipartition index ($E = H'/H'max$), where ("$H'max$"= maximum diversity that is represented as follows : $H'max = log_2 \, S$). Simpson's reciprocal index ($SRI = 1/D$) and dominance index ($SDI = \Sigma \, Ni \, (Ni-1)/N \, (N-1)$, where ("$Ni$"= number of species specimens considered and "N"= total number of species specimens) (31–34). Statistical analyses were conducted using the software R, v. 4.3.0. The graphs of multilines of ecological indexes, of points of species richness, and the altitude regression were performed using the packages Tidyverse, ggplot2 and reshape as well as species phenology (35–37). Correlation matrix between all of these ecological parameters was plotted using Corrplot (38). Furthermore, Metan package was applied to plot the Pearson correlation between all the environmental factors and surveyed species (39).

Results

Diversity of the sand fly fauna

Investigation of the sand fly population in Djelfa during the survey period from August 2021 to July 2022 in four different biotopes, urban, peri-urban, and rural, revealed a total of 2,866 specimens spread over 12 species belonging to two genera: *Phlebotomus* and *Sergentomyia* (Table 2).

The analysis of the indices of diversity and homogeneity have revealed variations from one site to another (Fig. 2). The highest richness was found in sites 7 and 8 of Ain El Bel station (S= 9) with an abundance of 315 in site 7 and 273 in site 8, while the lowest richness was observed in site 1 at Ain Oussera station. Shannon Weaver index values exhibited a range between 0.543 bits and 1.812 bits, indicative of a commendable diversity within the surveyed area. The equitability index revealed relatively even species distribution across most sites, with values spanning from $E = 0.572$ to $E = 0.825$. In contrast, the sites 1 (in the animal habitats) and 3 (surrounding human habitats) deviated from this pattern, displaying lower equitability values ($E = 0.494$ and $E = 0.404$, respectively). Examining Simpson's indices, these latter sites featured the highest values for Simpson's dominance index ($SDI = 0.721$ and $SDI = 0.713$, respectively) and the lowest values for Simpson's reciprocal index ($SRI = 1.386$ and $SRI = 1.402$, respectively), indicating a lower degree of species dominance. In contrast, site 8 (in the animal habitats) exhibited a substantially lowest Simpson's dominance index ($SDI = 0.215$) and a notably highest Simpson's reciprocal index ($SRI = 4.647$), reflecting a higher level of species dominance within this particular site.

Composition of the sand fly fauna

The comprehensive list of sand flies collected is presented in Table 2. The genus *Phlebotomus* is represented by seven species of which *Ph. papatasi* was the most abundant constituting 57.29% of the total specimens. This species exhibited a presence in various seasons, with 8.37% in the summer of 2021, 28.30% in autumn, 1.78% in spring, and 18.84% in the summer of 2022. It is followed by *Ph. alexandri* with 10.19% of the total population. Its distribution across the seasons was as follows: 2.38% in summer 2021, 13.44 % in summer 2022, 8.73% in autumn, and 0.24% in spring. The lowest reported species was *Ph. longiscuspis* that only appeared in summer 2022 with (2.09%). Concerning the second genus *Sergentomyia*, five species were collected. Among them, *Se. minuta* was particularly abundant, representing 9.42% of the overall population. Its distribution encompassed various seasons with 1.19% in summer 2021, 2.23% in autumn, 0.03% in spring, and 12.85% in summer 2022. It is followed by *Se. fallax* with a very low prevalence, constituting 3.52% of the total, respectively detailed in summer 2021 with (0.14%), in autumn with (1.05%) and in summer 2022 with (2.23%). The least represented species is *Se. dreyfussi*. Only one single specimen, with a rate of 0.03 %, represents this species. In addition, most of the surveyed species were captured with both sexes male and female. Exception for *Se. dreyfussi* and *Se. minuta* where only females were captured and for *Se. antennata* and *Se. schwetzi* where only males were collected.

Seasonal phenology and occurrence of phlebotomine species

The seasonal occurrence and phenology of phlebotomine sand flies in the Djelfa region from August 2021 to July 2022 revealed that *Ph. papatasi* is ubiquitous with the occurrence of 100% throughout summer 2021, autumn, and summer 2022 and occasional in spring with 33% at the study area. On the other hand, *Ph. alexandri* is presented only in autumn and summer 2022 in sites 1 and 2 and is considered as an accidental species (FO%≤25 %). Interestingly, the same species transitioned to the occasional ubiquitous category in sites 5, 6, and 10 in summer 2021 (100%), autumn (67%), spring (33%), and summer 2022 (100 %). A similar occurrence pattern was noted for *Ph. sergenti* at site 6 and *Ph. perniciosus* at sites 5, 6, and 7. Furthermore, *Se. schwetzi* displayed an occurrence of 67% in site 7 and 33% in site 8 during autumn, while reaching 100% in summer 2022 in both sites. During the two summers, this species was only observed in one site out of the ten surveyed. The occurrence of *Ph. ariasi* in site *7* was occasional in autumn and ubiquitous in the summer of 2022. However, at sites 2, and 8, this species exhibited occurrences ranging from 33% to 100% during both summer and autumn.

The accidental occurrence of *Se. antennata* during all seasons ($FO\% \leq 25$) was observed in some sites as follows: in site 3 during summer 2022 and site 5 during summer 2021, with the exception that this species was recorded during summer 2021, autumn, and summer 2022 in sites 4, 7 and 8. In addition, *Ph. longiscuspis* was only observed during summer 2022 in site 10 with 100%.

Likewise, *Ph. langeroni* occurred occasionally in sites 7 with 42% and site 8 with 33%, whereas this species occurred accidentally in site 3 with 8%. It is important to point out that this species is being reported for the first time in Algeria and that no studies are revealing its presence in endemic outbreaks of leishmaniasis. *Phlebotomus langeroni* females can be distinguished by several distinctive characteristics, including shorter wings (0.03 mm), a lengthier ascoid antennular segment IV (0.46

times the segment length), and a distinctive feature in the structure of the spermatheca. The spermatheca exhibits a reservoir composed of nine to ten annuli that extends into a duct culminating in a characteristic fusiform dilatation. This structural differentiation sets it apart from other *Larroussius* species (Fig. 4).

Statistical treatments

Correlation matrix based on ecological indices for phlebotomine species

The correlation matrix unveiled positive correlations among all the diversity parameters, except for the SDI parameter. Specifically, the latter parameter (SDI) exhibited a weak negative correlation with the number of species specimens but demonstrated strong negative correlations with the other indices. Notably, parameters J, H, SRI and S displayed strong positive correlations with each other. Conversely, the number of species specimens displayed weak positive correlations with the latter four parameters (Fig. 5).

Altitude distribution

The linear regression analysis revealed a moderate and average relationship between sand fly species richness and altitude $(P = 0.01\ ;\ R^2 =\ 0.519)$. It was observed that species richness increased moderately and significantly with higher altitude. However, there was no significant relationship between species abundance and altitude ($P = 0.731$; $R^2 = 0.016$), the results are shown in Fig. 6.

Pearson correlation between environmental factors and sand fly population

The correlation analyses carried out on the data obtained for environmental factors (maximum, mean and minimum temperatures, relative humidity, rainfall, and wind speed) and sand fly species are shown in Fig. 7.

The results unveiled robust and statistically significant positive correlations between maximum, mean, and minimum temperatures and all species belonging to the genus *Phlebotomus* (except for *Ph. longiscuspis*) and to the genus *Sergentomyia* (excluding *Se. dreyfussi*). The correlation values ranged from ($p < 0.05$, $r = 0.58$ to $p < 0.001$, $r = 0.91$). In contrast, concerning humidity, a pronounced and statistically significant negative correlation was observed with the sand fly population, except for two species, *Ph. longiscuspis*, and *Se. dreyfussi*. The correlation coefficients for humidity ranged from ($p < 0.05$, $r = 0.59$ to $p < 0.001$, $r = 0.89$). As for the correlations between rainfall and wind speed, no statistically significant relationships were detected with any of the sand fly species.

Table 1. Characteristics of sampling sites for sand flies in study region Djelfa, Algeria, 2021–2022

Station	Site	Coordinates	Altitude (m asl)	Biotope	Site description	Vegetation	Water source
Ain Oussera	1	35°28’18.815’’N	661	Periurban	Animal habitats	++	Irrigation+ Oued
	2	2°54’40.299’’E		Periurban	Surrounding human habitats	+	Irrigation+ Oued
Hassi Bahbah	3	35°3’4.63’’N 3°2’47.264’’E	850	Periurban	Surrounding human habitats	+	Irrigation
	4			Periurban	Animal habitats	++	Irrigation

Table 1. Continued ...

Djelfa	5	34°39’32.693’’N 3°13’12.636’’E	1186	Urban	Surrounding human habitats	+	Irrigation+ Oued
	6			Urban	Animal habitats	++	Irrigation+ Oued
Ain El-bel	7	34°20’47.347’’N 3°13’39.093’’E	1043	Rural	A junk room	++	Irrigation
	8			Rural	Animal habitats	+++	Irrigation
Mlaga	9	34°12’58.832’’N 3°23’49.377’E	811	Rural	Animal habitats	+++	Irrigation+ Oued
Haniet Ouled Salem	10	34°10’14.849’’N 3°27’22.755’’E	790	Rural	Animal habitats	+++	Irrigation

(+): low; (++) medium; (+++) dense, asl: above sea level

Table 2. Inventory and abundance of phlebotomine species sampled in Djelfa region, Algeria (August 2021−July 2022)

			Total		Summer 2021		Autumn		Winter		Spring 2022		Summer 2022	
	Sex	**N**	**N**	**RA%**	**N**	**RA%**	**N**	**RA%**	**N**	**RA%**	**N**	**RA%**	**N**	**RA%**
Phlebotomus papatasi **(Scopoli, 1780)**	M	484	1642	57.29	240	8.37	811	28.30	_	_	51	1.78	540	18.84
	F	1158												
Ph. langeroni **(Nitzulescu, 1930)**	M	23	89	3.10	6	0.21	27	0.94	_	_	0	0	56	1.95
	F	66												
Ph. sergenti **(Parrot, 1917)**	M	10	63	2.20	3	0.10	18	0.63	_	_	2	0.07	40	1.40
	F	53												
Ph. perniciosus **(Newstead, 1911)**	M	18	142	4.95	3	0.10	64	2.23	_	_	0	0	75	2.62
	F	124												
Ph. longiscuspis **(Nitzulescu, 1930)**	M	4	60	2.09	0	0	0	0	_	_	0	0	60	2.09
	F	56												
Ph. alexandri **(Sinton, 1928)**	M	16	292	10.19	8	0.28	104	3.63	_	_	7	0.24	173	6.04
	F	276												
Ph. ariasi **(Tonnoir, 1921)**	M	2	107	3.73	24	0.84	52	1.81	_	_	0	0	31	1.08
	F	105												
Sergentomyia schwetzi **(Theodor&Parrot, 1929)**	M	56	56	1.95	3	0.10	9	0.31	_	_	0	0	44	1.54
	F	0												
Se. fallax **(Parrot, 1921)**	M	12	101	3.52	4	0.14	30	1.05	_	_	3	0.10	64	2.23
	F	89												
Se. antennata **(Newstead, 1921)**	M	43	43	1.50	11	0.38	12	0.42	_	_	0	0	20	0.70
	F	0												
Se. dreyfussi **(Parrot, 1933)**	M	0	1	0.03	0	0	0	0	_	_	0	0	1	0.03
	F	1												
Se. minuta **(Rondani, 1843)**	M	0	270	9.42	34	1.19	64	2.23	_	_	1	0.03	171	5.97
	F	270												
Total			2866	100	336	11.71	1191	41.55	_	_	64	2.22	1275	44.49

−: Absence**; M:** male**; F:** female; **RA**: relative abundance

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Published Online: June 30, 2024

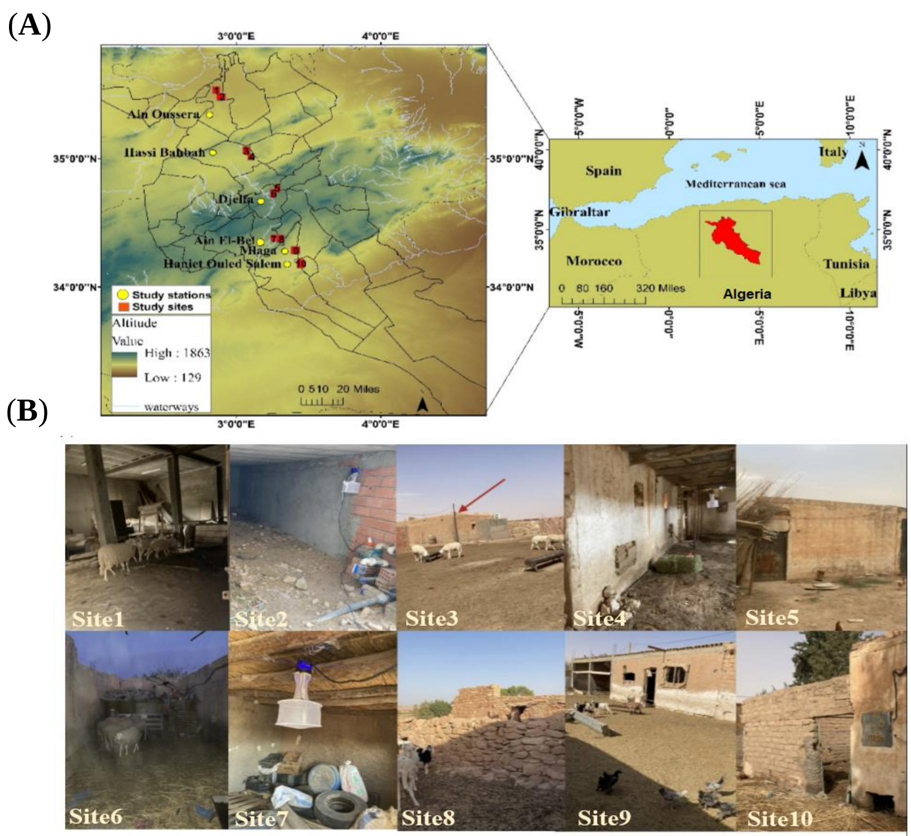

Fig. 1. (A) Map showing the geographical position of the Djelfa region, highlighting the study sites. (B) Photographs showing the habitats of the study sites: in the animal habitats (sites 1, 4, 6, 8, 9, and 10); Surrounding human habitats (sites 2, 3, and 5); and site 7, representing a junk room

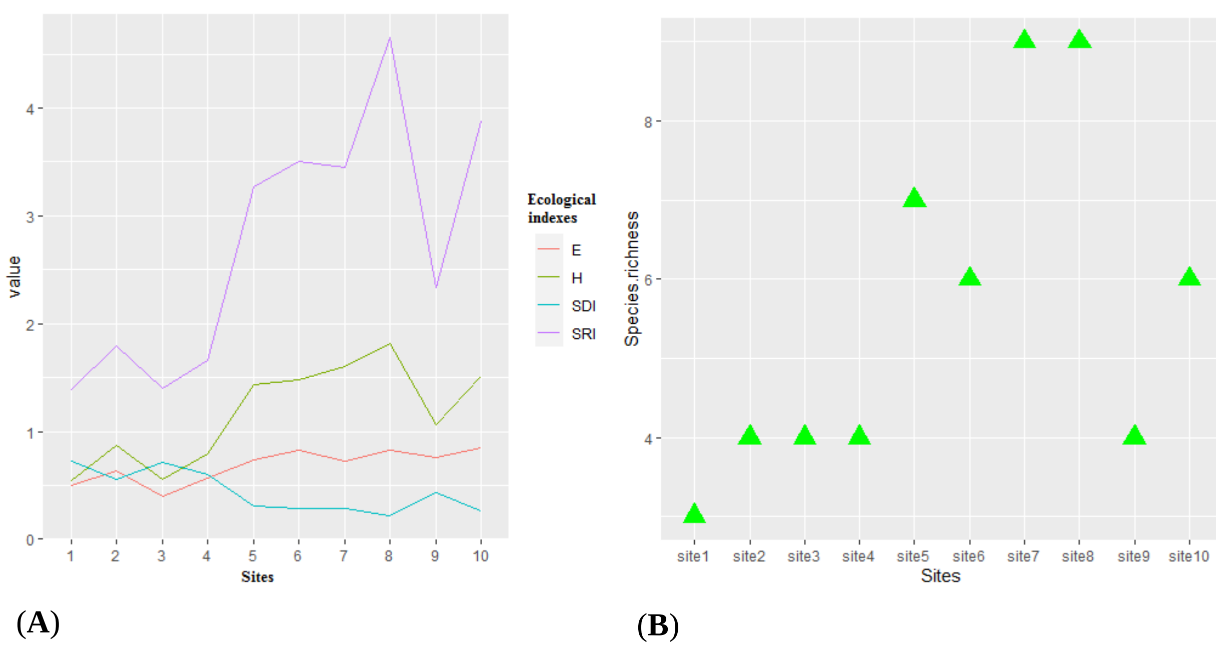

Fig. 2. Ecological analysis of the phlebotomine species in the study sites (A) Ecological indices: Shannon-Weaver index (H'), Equipartition (E), Simpson's reciprocal index (SRI) Simpson's dominance index (SDI) and (B) Species richness

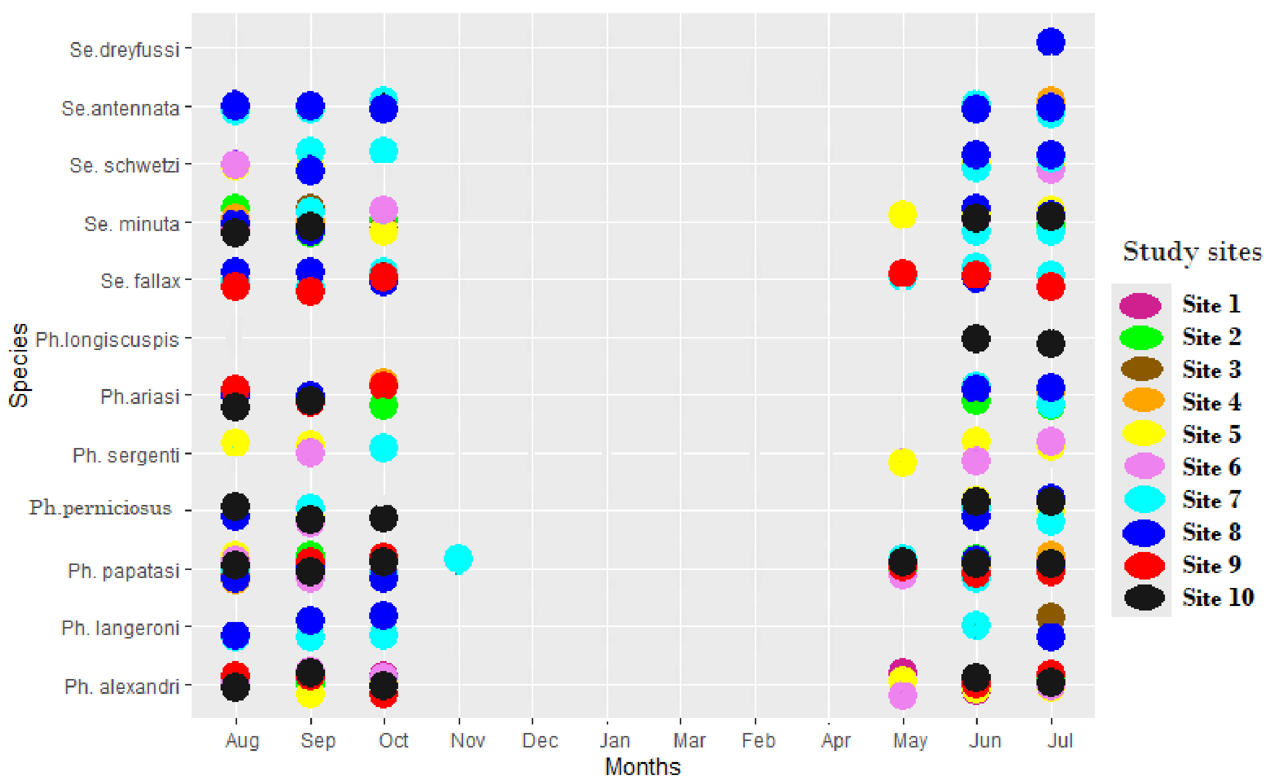

Fig. 3. Phenology of phlebotomine species surveyed in Djelfa region, Algeria (August 2021−July 2022)

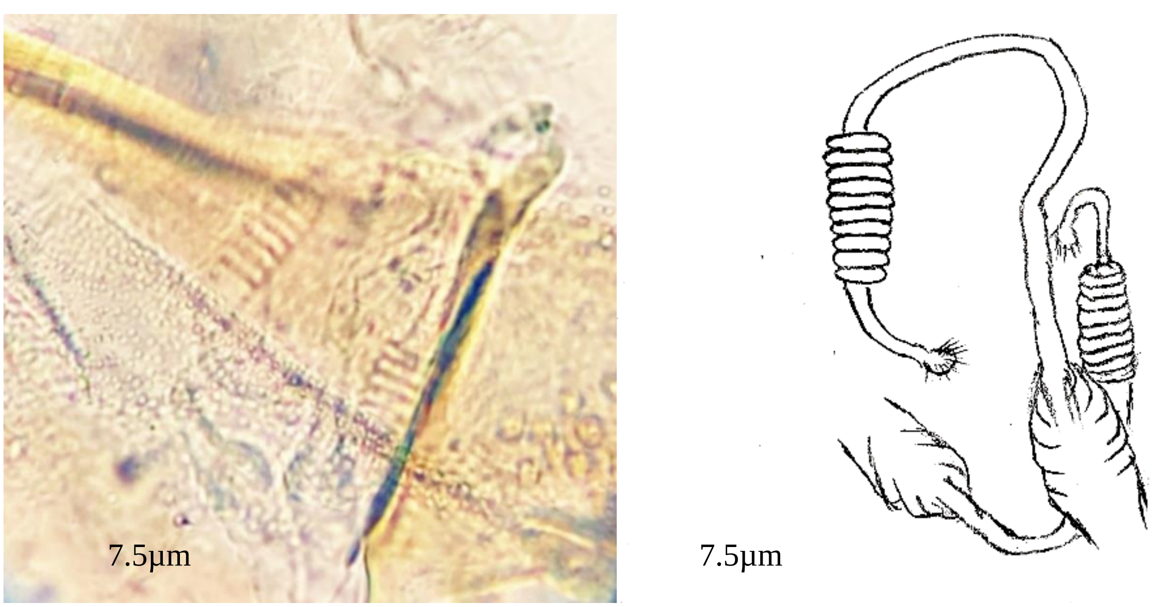

Fig. 4. Spermatheca of *Phlebotomus langeroni* female captured in Djelfa region (Algeria), 2021–2022

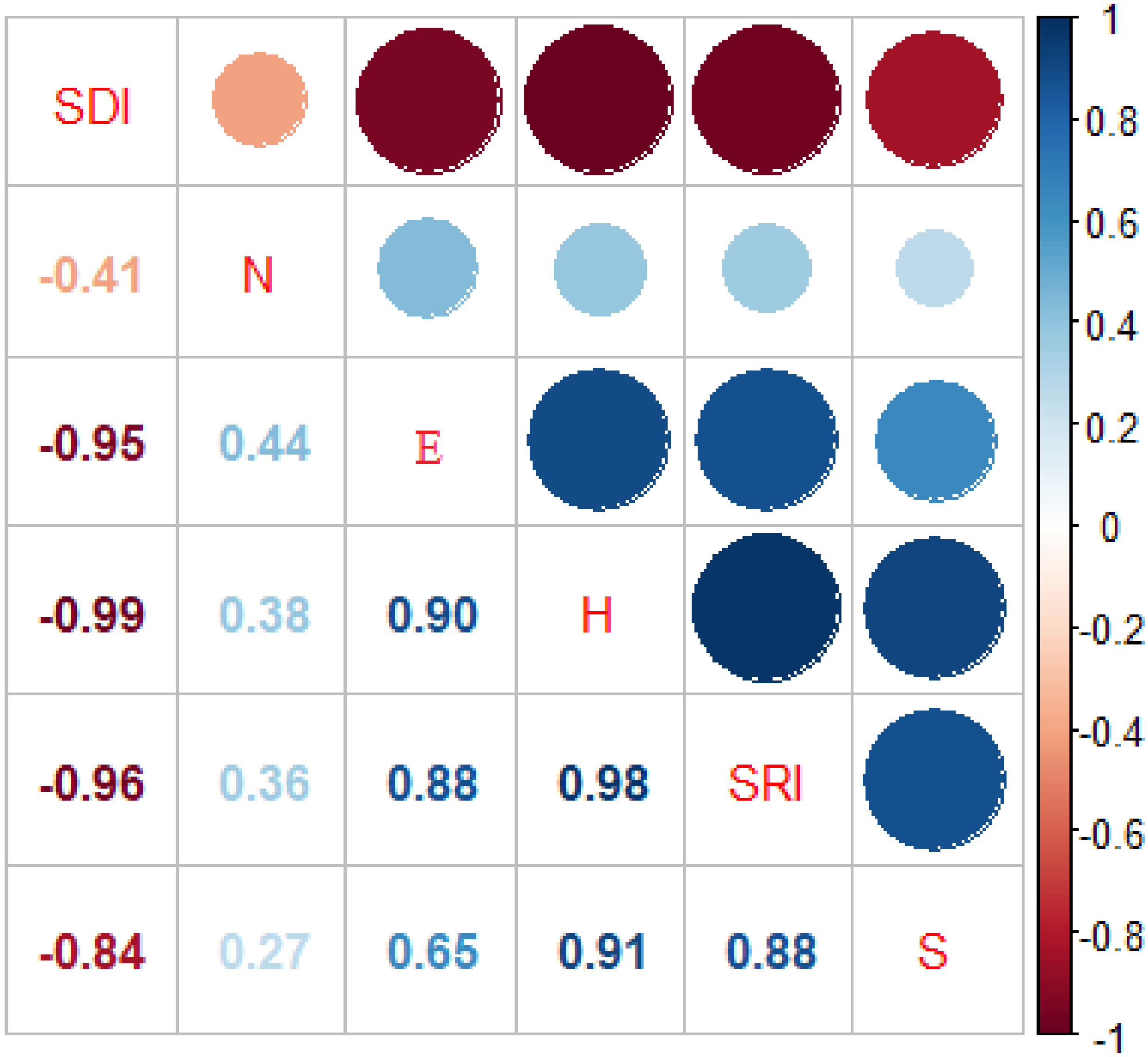

Fig. 5. Corrplot of the correlation matrix between ecological parameters of phlebotomine species from August 2021 to July 2022 in the Djelfa region. The parameters are SDI (Simpson's Dominance Index), E (Equipartition Index), N (Total Number of Species Specimens), H (Shannon-Weaver Index), SRI (Simpson's Reciprocal Index), and S (Species Richness); blue circles represent positive correlations, while red circles represent negative correlations and color degradations represent the intense of correlation

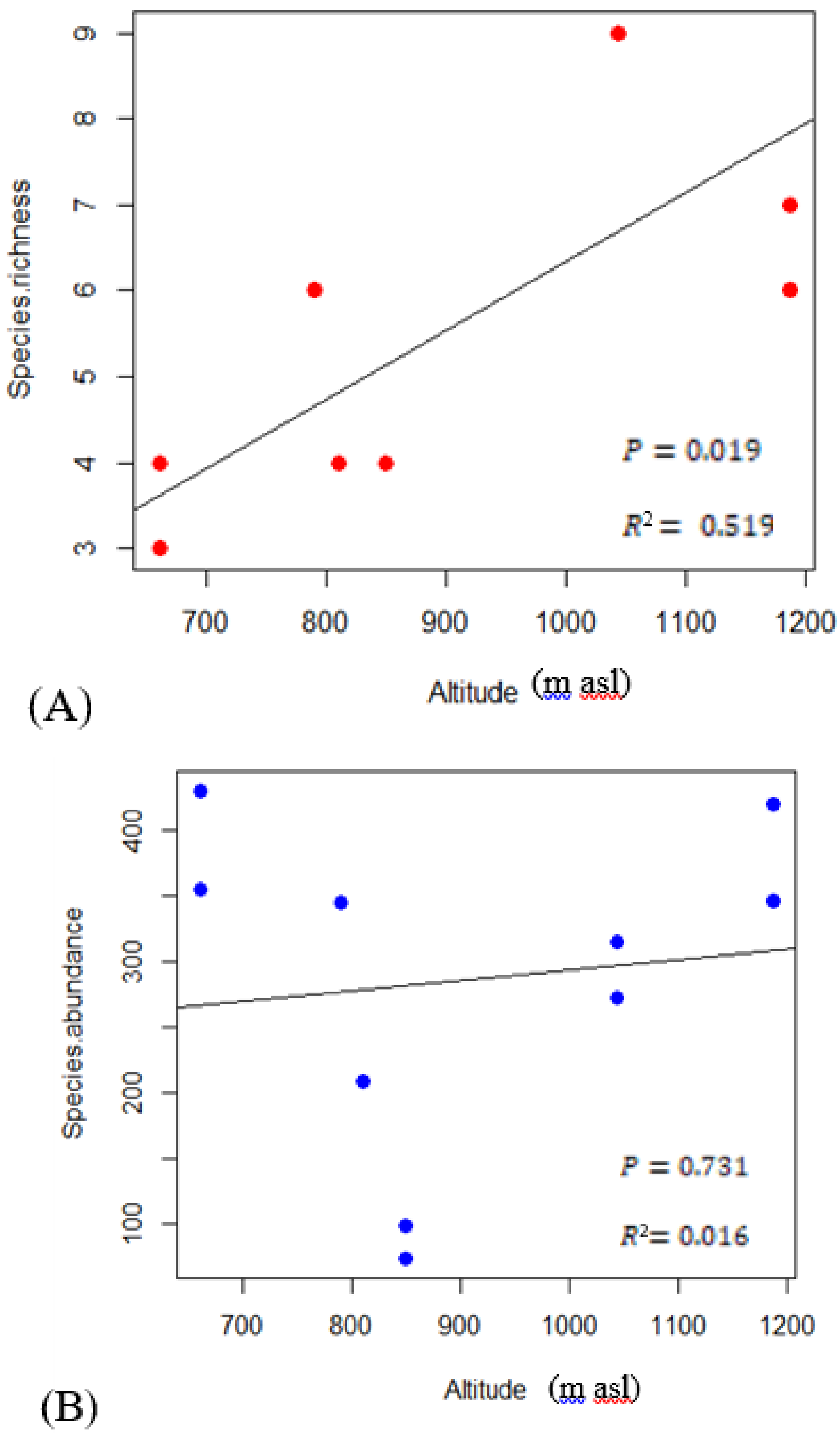

Fig. 6. Plot showing the relationship between altitude (m asl) and sand fly species richness (A); abundance (B) in Djelfa region, Algeria, 2021–2022

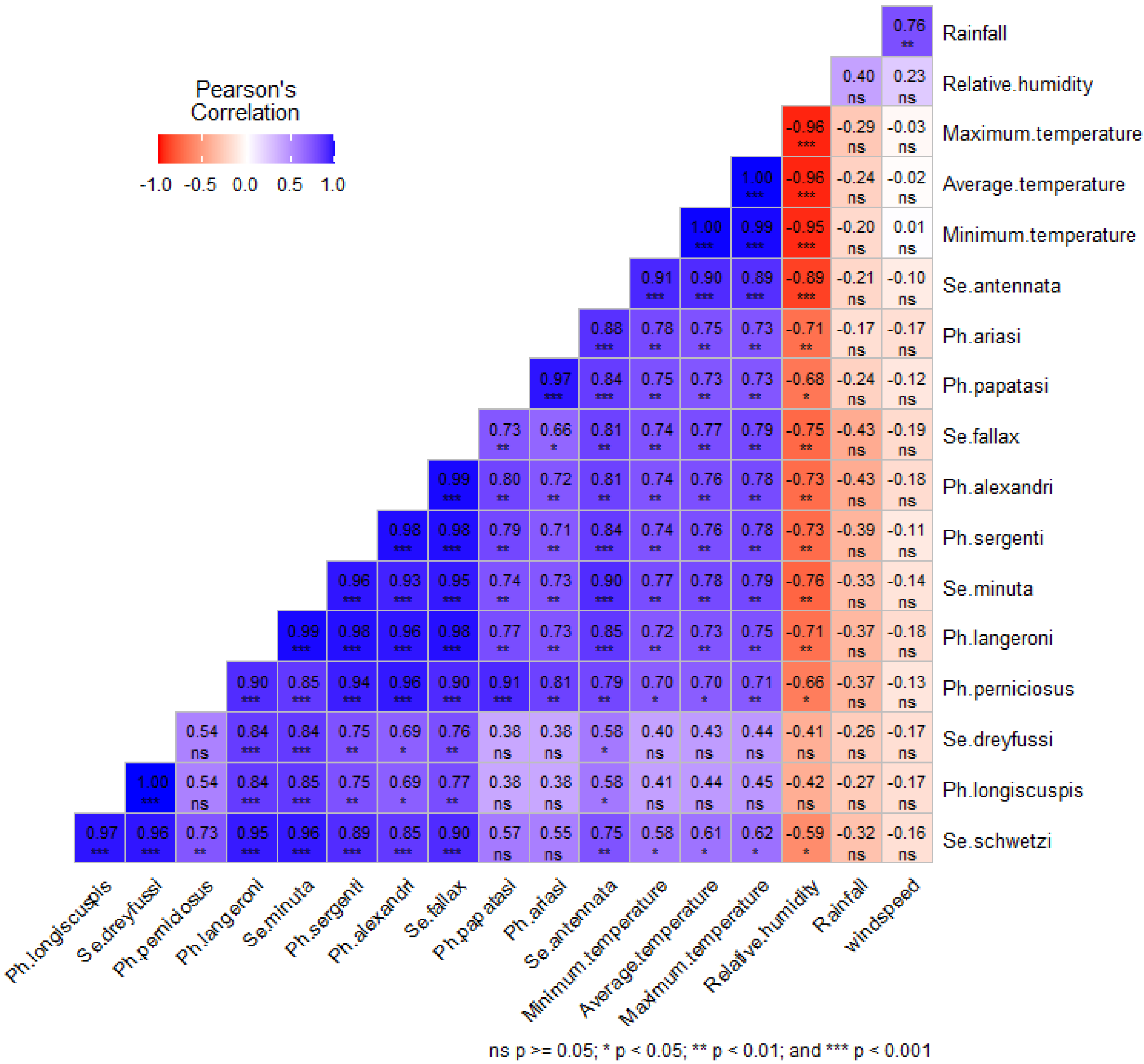

Fig. 7. Corrplot of the Pearson correlation between all the environmental factors and the phlebotomine species in the Djelfa region, Algeria, 2021–2022

Discussion

In Algeria, the risk of leishmaniasis has increased significantly in recent years, according to the Algerian National Institute of Health and Population. In the current study, the first entomological survey was carried out in Djelfa, one of the regions mostly affected by this disease (20). Our investigations led to the identification of 12 species of both genera *Phlebotomus* and *Sergentomyia*, out of the 26 species known in Algeria (41). All species that are proven or suspected vectors of *Leishmania* have been found in the current survey. Starting with the genus *Phlebotomus*, *Ph. papatasi* emerges as the most prevalent species, constituting 57.29% of the observed specimens across all surveyed biotopes. The known distribution of this species is in steppe areas and the northern Sahara, and it is a proven vector of *L. ma-*

jor in Algeria (42). Although it can be found in the Tell region and the Highlands (27). For *Ph. alexandri* which represents 10.19% of collected sand flies in the peri-urban and urban biotopes. This species is known for its ability to transmit several *Leishmania* species such as *L. major*, *L. infantum*, *L. donovani*, and *L. tropica*, and it inhabits a wide range of environments, extending from natural habitats to peri-urban and urban areas (43–47). In China, this species is present in restricted areas such as the stony desert (48, 49). *Phlebotomus perniciosus*, found in rural and urban areas at a rate of 4.95%, is a proven vector of *L. infantum* MON 1, which causes visceral leishmaniasis in rural villages (50). This species is mostly distributed in the northern part of Algeria (27). *Phlebotomus ariasi*, collected in rural and peri-urban settings with a prevalence of 3.73%, has been proved as a vector of *L. infantum* in Morocco and Tunisia, causing VL (51, 52).

Phlebotomus langeroni, comprises 3.1% of the specimens in peri-urban and rural areas. We achieved the interesting presence of female specimens of this species in two rural sites at the Ain El Bel station, with a total of 66 specimens, although this is a significant number, it is certainly an underestimate of reality. Furthermore, a unique specimen was collected in peri-urban site 3. This species was found infected with *L. infantum,* responsible for CL in northern Tunisia (53) as well as *L. donovani* which causes VL in Egypt (54, 55). This sand fly occurs particularly in dry, rocky biotopes, where rodent and reptile breeding grounds are found (27). In Algeria, only male specimens of the *Ph. langeroni* species have been documented (10, 13, 27) and this is the first survey that highlights the presence of females of this species.

Phlebotomus sergenti is distributed in rural and urban (2.2%) and is the proven vector of *L. tropica* in Morocco (56) and Iran (57) and *L. killicki* in Algeria (16) and in Tunisia (58), where it is mostly found in the South of Algeria in the Saharan Atlas mountains, the pre-Saharan steppes and the Tell areas (16, 27, 59). *Phlebotomus longicuspis*, representing 2.09% in rural areas, has epidemiologically been linked to *L. infantum* transmission in bioclimatic arid zones of northern Africa (14, 60, 61).

With regard to sand flies belonging to the genus *Sergentomyia*, several studies have raised the potential involvement of species belonging to this genus as vectors of *Leishmania* (62, 63). In the present survey, six species were identified in a variety of biotopes. Previous studies in northern Africa prove that all these species were found to be infected by *L. major* in rocky areas, the natural biotope of the rodents (64, 65). This is in line with the features of the study region Djelfa, which is characterized by its great diversity of rodents, in particular *Meriones shawii*, which is considered the secondary *L. major* reservoir after the sand rodent *Psammomys obesus* and plays an important role in the transmission of zoonotic cutaneous leishmaniasis in pastoral areas (66, 67).

Investigations on the potential of *Sergentomyia* species as vectors of the parasite to mammal hosts are actively pursued in several countries such as Tunisia, Kenya, Iran, and Portugal (65, 68, 69, 47). In Algeria, there exists a scarcity of experimental research focusing on the genus *Sergentomyia*, emphasizing the need for greater attention in future studies (41).

To understand the epidemiology and ecology of parasite activity, environmental and demographic factors must be taken into account jointly (70). This study marks the first investigation into the ecology and spatio-temporal distribution of sand fly populations within the Djelfa region. Initially, the effect of climatic and environmental factors on the dynamics of these species, which are distinguished by various ecological and biological traits, was assessed. Next, the findings were compared with other data to discern alterations in the seasonal dynamics and abundance of sand flies.

http://jad.tums.ac.ir
Published Online: June 30, 2024

Throughout the winter months, all sand fly species were absent due to their diapause response, an adaptation to survive the cold season (71,72). However, during the summer of 2021 and subsequently, in the autumn, spring, and summer of 2022, most species reappeared, primarily due to the increasing temperature. It is noteworthy that the increase in temperature, associated with the socio-ecological changes transpiring in the region (humidity, rainfall, and altitude), could potentially influence the dynamics of *Leishmania* infection. Additionally, it must be highlighted that in this study, the CDC light trap was used exclusively for the faunistic study of sand flies. This trap choice imposes a considerable limitation on capturing sand fly species as effectively as other trapping techniques, such as sticky traps.

The correlation of Pearson has shown a positive link ranging from ($p < 0.05, r = 0.58$ to $p < 0.001, r = 0.91$). As a result of the opposite relationship between relative humidity and temperature, a negative correlation has been noted with sand flies to humidity ranging between ($p < 0.05, r = 0.59$ to $p < 0.001\ r = 0.96$). The findings of this correlation are in line with those reported in central Morocco (73). Although there was no correlation between relative humidity and temperature between the two species *Se. dreyfussi* and *Ph. longiscuspis*, which could be related to the disappearance of these two species during most sampling months and their appearance only during the summer of 2022. Neither rainfall nor wind speed ($p > 0.05$) were correlated with phlebotomine fauna, which is consistent with the previous results from central and southern Morocco, where both factors were determined not to be decisive for sand fly abundance (73, 74). The knowledge that bio-ecological factors play a significant role in the geographical and seasonal distribution of disease vectors (75, 76).

The relationship between altitude and the incidence of leishmaniasis is contingent on a myriad of environmental factors, most notably temperature, which plays a pivotal role in the development of parasites within sand flies (77). In the current study, the linear regression analysis indicated a moderate and intermediate relationship between sand fly species richness and altitude. This aligns with observations in Morocco, Libya, and Turkey, where the distribution of sand fly species is notably influenced by altitude (78–81). In Golestan Province, Iran, an increase in the Shannon-Wiener and Evenness indices at higher altitudes, coupled with a decrease in the Simpson index, suggests that the sand fly fauna is more stable in areas with higher altitude compared to lower altitude areas (82). Inversely, regarding sand fly abundance, there is no discernible relationship with altitude, consistent with research conducted in plains of North-Eastern Italy (83) but contradictory to findings in South-Central Turkey, where altitude was found to significantly impact population abundance (84).

Conclusion

The widespread distribution of sand flies in the endemic focus of Djelfa along with the extended activity of 12 sand fly species highlights the potentially high risk of the spread of leishmaniasis throughout the Algerian steppes. These findings provide the foundation for subsequent research in the region. Furthermore, it is highly recommended that continuous monitoring efforts be implemented to mitigate the risk of spreading leishmaniasis disease. Additionally, the risk of spreading needs to be elucidated in terms of the climatic conditions that favour sand fly proliferation and to understand host-blood feeding preferences. These precious findings are essential for the development of effective control strategies, the optimization of operational measures, and the evaluation of their effectiveness.

Acknowledgment

No funding was received for conducting this study.

Ethical consideration

The ethics committee of the Laboratory of Exploration and Valorization of Steppe Ecosystems at the University of Djelfa (Algeria) has approved this entomological investigation.

Conflict of interest statement

The authors declare there is no conflict of interest.

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