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ASSESSMENT OF MOSQUITOES LARVAE AND THEIR PREDATORY AQUATIC INSECTS IN TAKSEBT DAM, ALGERIA

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Abstract. The present paper aims at evaluating the distribution pattern of Culicidae immature stages and the relationship with their potential insect predators in the Taksebt Dam, Algeria. The surveys in the temporary and permanent habitats of this dam using the dipping technique allowed us to identify 13 mosquito species belonging to four genera with a clear dominance of *Cx. pipiens* and *Cx. perexiguus*. Predators of mosquito larvae were abundant in permanent habitats with 11 families belonging to three orders: Hemiptera (57.7%), Odonata (28.5%) and Coleoptera (13.9%). Hemipteran predators were the most important and abundant with a dominance of the Naucoridae and Notonectidae families. All mosquito larvae abundance was negatively related with the Hemipteran species abundance, while the abundance of Odonata spp. showed a significant effect on *Anopheles* spp. abundance. The results are relevant for future research in conservation biological control of vector mosquitoes employing predatory insects.

INTRODUCTION

Vector-borne diseases are a major threat to the world, and more than 80% of its population is at risk of a vector-borne disease (WHO 2014). Vectors transmit some of the most important infectious diseases to humans (Godfray 2013), and their increasing negative impact is partly associated with the range expansions of their primary vectors (Bonizzoni et al. 2013). The spread of these vectors is influenced by the characteristics of their breeding sites (Lambin et al. 2010).

Mosquitoes are important vectors which can transmit protozoan, filarial and viral diseases (Cuervo-Parra 2016) and have different habitat preferences (Sithiprasasna et al. 2003). In fact, the ecological, spatial, and temporal distributions of many mosquito species are limited by habitat preference (Day 2016). For example, *Aedes* species have been shown to exploit different temporal larval habitats (Dom et al. 2013), while *Anopheles* species prefer shallow water bodies (Bashar et al. 2016), Culicine mosquitoes exploit a wide range of aquatic habitats with slight differences among individual species (Amini et al. 2020).

Wetlands are one of the characteristic breeding sites, and according to Rey et al. (2012), there is a need for mosquito control in wetlands as part of mosquito-borne disease management. Indeed, the use of chemical and microbial controls is constrained in wetlands in view of their potential impact on the biodiversity (Saha et al. 2014). On the other hand, biological control is an environment-friendly mosquito control promoted in order to avoid the damage caused by insecticides (Liu 2015). It is considered a sustainable means that could be included in an integrated vector management programme using natural organisms such as plants, bacteria, entomopathogenic fungi and especially predators (Benelli et al. 2016).

Predation on the late instar larval stages of the mosquitoes could reduce the population density dramatically. Predatory fish (Cohen and Silberbush 2021), amphibians (Sarwar 2015) and aquatic insects (Mansoreh et al. 2014) are known to inhibit mosquitoes breeding sites and reduce mosquito abundance. According to Schrama et al. (2018), insect predator presence decreased mosquito larval survival and adult emergence by 20–50%. Many aquatic insects of the orders Coleoptera (DeSiervo et al. 2020), Diptera (Moirangthem et al. 2018), Hemiptera (Allo and Mekhlif 2019) and Odonata (Córdoba-Aguilar et al. 2021) are known to be mosquito predators, commonly found with the mosquito immature in varied mosquito larval habitats including the wetland (Kumar 2009; Shaalan and Canyon 2009).

Several works have studied the species composition of mosquitoes (Robert et al. 2019; Merabti et al. 2021), the abiotic characteristics of mosquito breeding sites (Merabti et al. 2017; Oussad et al. 2021), and the larvicidal effect of herbal preparation (Kharoubi et al. 2020; Djeddar et al. 2021); however, few studies have focused on the insect predation efficiency (Culler and Lamp 2009; Ohba and Takagi 2010; Saha et al. 2010, 2012) and the relationship between mosquito and predator density (Das et al. 2006; Leitão et al. 2007; Lytra and Emmanouel 2014; Sunish and Reuben 2002). The wetlands are one of the prominent mosquito breeding sites (Dale and Knight 2008) featured by the presence of a wide range of freshwater predatory insects like the odonate larvae, water bugs and beetles (Zuharah and Lester 2010; Watanabe et al. 2013). As a consequence, the conservation biological control can be achieved provided the exploration on the species specific abundance of the predatory insects and the vector mosquitoes are available thorough assessment. Thus the present study was devoted to the structural composition and systematic determination of the species in the Taksebt Dam, as well as the evaluation of the prey-predator relationship. The results of this study could be useful in promoting the implications of biological control of mosquitoes in the area. In this study, we evaluate the mosquito and predatory insect fauna existing in the Taksebt Dam in order to highlight if it is a suitable breeding site for mosquito vectors and whether the predatory insects like odonate larvae, water bugs and beetles in this biotope are more abundant and diversified.

MATERIALS AND METHODS

Study area

This study was conducted on the Taksebt Dam, central north Algeria, 10 km southeast of Tizi-Ouzou town and 100 km east of Algiers. Our study area is characterized by the Mediterranean climate, with mild and humid winters alternating with hot and dry summers. There is a rainy season from October to May, and a dry season from June to September. The average maximum and minimum temperatures are about 36°C and 6.6°C. The study area is subject to high water conditions due to heavy, often irregular, rainfall varying annually between 600 mm and 800 mm. This hydraulic infrastructure is the biggest and largest wetland in the region. It is fed by rainfall, snowmelt from the Djurdjura mountains and wastewater. This reservoir is characterised by clay soil and a moisture content of over 60%.

The study site includes three permanent and three temporary water stations (Figure 1). In fact, certain stations dry up, which leads to a different number of samples from one station to another, ranging from 6 to 10 times in the temporary stations and 22 to 24 times in the so-called permanent stations. The temporary ones are located upstream of the dam (36º40'35.375"N, 4º7'17.7880"E), approximately 800 m to the south, these ponds are formed by the lowering of the water level; whereas the permanent ones are located downstream of the dam (36º40'46.978"N, 4º7'8.76"E), characterized by very dense vegetation and fed by the overflow of the dam.

Aquatic entomofauna sampling

For the collection of mosquito larvae and other insects, we used the dipping method (Silver 2008) which consisted of performing ten sweeps at each sampling site, using a 500 ml dipper. Selected aquatic insect habitats were sampled once every two weeks for one year in the permanent sites and until drying in the temporary sites. The predatory species were preserved in 96% ethanol, and the mosquito larvae were raised in the laboratory. The obtained nymphs were sorted and placed in plastic cups containing water, then in emergence cages covered with a mosquito net. The obtained adults were preserved for morphological identification. The fourth instar larvae were cleared and mounted using the protocol of Matile (1993) for a better observation under a microscope. Mosquito larvae and adults were identified using the dichotomous keys (Himmi et al. 1995; Becker et al. 2010) and the software of African mosquitoes (Brunhes et al. 2000). Insect predators were classified to order level (Odonata, Hemiptera, Coleoptera). Odonate larvae were identified and confirmed by observation and recognition of their imagoes around the sites, Hemiptera and Coleoptera were identified as adults; however, the identification was confirmed by the presence of their larval stages and using the appropriate keys (Guignot 1947, 1959; Poisson 1957; Heidemann and Seidenbusch 2002; Doucet 2011).

Data analysis

The obtained results were treated by an ecological index. The specific richness (S = number of species), relative abundance ($F = N_i \times 100/ N$), Shannon's index ($H' = -\sum P_i \ln (P_i)$) and Simpson's index ($D_1 = 1 - \sum P_i^2$) were calculated (Magurran 2004). One-way ANOVA (with Tukey's test) was then estimated to compare predator insect and mosquitoes abundances among

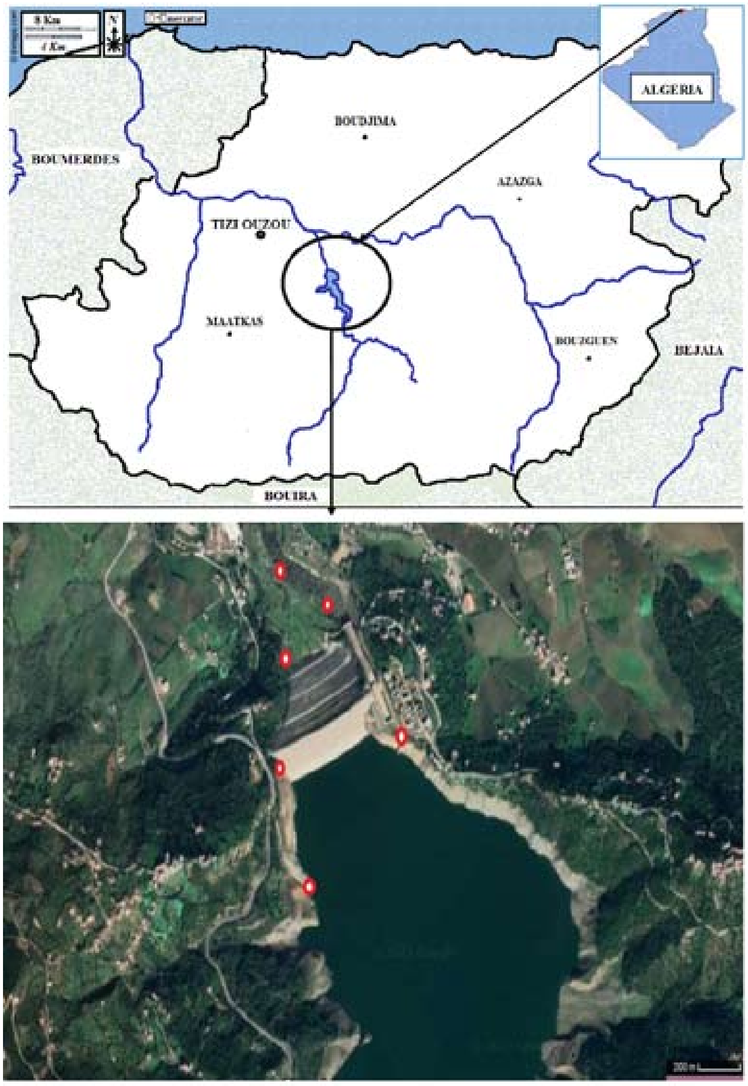

Figure 1. Study site location (Taksebt Dam) from Tizi-Ouzou area in Algeria.

temporary and permanent sites. In order to analyse the relationship between predator insect and mosquitoes, the generalized linear model (GLM) with negative binomial (glm.nb package) in R statistical package (version 3.0.2) was used, with a significance threshold of $p < 0.01$.

RESULTS

Aquatic entomofauna diversity

Total species diversity ranged from 2 to 18, and the number of specimens varied from 92 to 428 (Table 1). Shannon's diversity index was the highest (1.71, 1.84, 1.91) at permanent sites and the lowest (0.19, 0.58, 0.59) at temporary ones. Sampling time differed due to the drying of the temporary sites, and predatory species were collected only from the permanent habitats. Simpson's index was the highest at permanent sites (0.76, 0.77, 0.78) and the lowest (0.08, 0.29, 0.40) at the temporary ones, indicating that populations were rather homogeneous at temporary sites and heterogeneous at permanent sites.

Mosquito composition

The analyzed results of the studied sites in the Taksebt Dam during a period of 12 months showed that the culicidian fauna was represented by 13 species belonging to the following subfamilies (Table 2): Culicinae represented by *Culex laticinctus*, *Cx. perexiguus*, *Cx. hortensis*, *Cx. pipiens* complex, *Cx. theileri*, *Cx. mimeticus*, and *Cx. impudicus*, Anophelinae (*Anopheles maculipennis*, *An. claviger,* and *An. Algeriensis),* Aedinae (*Aedes caspius* and *Ae. vexans)*, and Uranotaeniini *(Uranotaenia unguculata*). The *Culex pipiens* complex was the most frequent with a rate of 83.6%, 72.8%, and 96.1% in the temporary sites (T1, T2, and T3, respectively). In the permanent sites (P1, P2, and P3), *Cx. perexiguus* seemed to be the most abundant with a rate of 50.0%, 42.6%, and 44.4%, respectively.

Table 1. Species diversity indices for the study zones (total abundance, richness, Shannon index, H max, Evenness (H), Simpson index (D_1)).

	P1	P2	P3	T1	T2	T3
Samples	24 (13)	24 (20)	22 (16)	10 (6)	8 (6)	6 (4)
Total	201	428	121	383	92	77
Species richness (S)	13	18	10	6	2	3
Shannon index (H’)	1.84	1.91	1.71	0.59	0.58	0.19
H max	3.7	4.17	3.32	2.58	1	0.51
Evenness (H)	0.5	0.46	0.51	0.23	0.58	0.37
Simpson index (D_1)	0.77	0.78	0.76	0.29	0.40	0.08

Table 2. Species composition, abundance and mean ± SE of mosquitoes by station.

Genus	Mosquito species	Temporary stations			Permanent stations		
		T1	T2	T3	P1	P2	P3
Culex 89.42%	*Cx. pipiens* Linnaeus 1758	83.6% (53.3 ± 35.2)	72.8% (11.17 ± 4.7)	96.1% (18.5 ± 8.5)	–	1.1% (0.2 ± 0.1)	–
	Cx. hortensis Ficalbi 1889	12.5% (8 ± 2.4)	27.2% (4.17 ± 1.7)	1.3% (0.25 ± 0.3)	28.5% (3.5 ± 0.7)	32.8% (5.8 ± 2)	17.6% (1.2 ± 0.4)
	Cx. perexiguus Theobald 1903	–	–	–	50% (6.1 ± 1.7)	42.6% (7.6 ± 2.6)	44.4% (3 ± 1.4)
	Cx. impudicus Ficalbi 1890	–	–	–	14.6% (0.5 ± 0.3)	5.1% (0.9 ± 0.3)	9.3% (0.6 ± 0.3)
	Cx. laticinctus Edwards 1913	0.5% (0.3 ± 0.3)	–	–	–	–	–
	Cx. theleiri Theobald 1903	1.6% (1 ± 0.5)	–	–	–	1.1% (0.2 ± 0.1)	–
	Cx. mimeticus Noé 1899	–	–	–	–	–	3.7% (0.3 ± 0.2)
Aedes 0.51%	*Ae. vexans* Meigen 1830	–	–	2.6% (0.5 ± 0.5)	–	–	–
	Ae. caspius Pallas 1771	–	–	–	2.5% (0.3 ± 0.2)	–	–
Anopheles 7.51%	*An. labranchiae* Falleroni 1926	1% (0.7 ± 0.4)	–	–	4.4% (1.8 ± 0.6)	8.8% (1.6 ± 0.9)	25% (1.7 ± 0.6)
	An. algeriensis Theobald 1903	0.8% (0.5 ± 0.4)	–	–	–	–	–
	An. claviger Meigen 1804	–	–	–	–	7.9% (1.6 ± 0.6)	–
Uranotania 0.17%	*Ur. unguculata* Edwards 1913	–	–	–	–	0.6% (0.1 ± 0.1)	–
Total genera	Total species	Total individuals	Total individuals	Total individuals	Total individuals	Total individuals	Total individuals
4	13	383	92	77	201	428	121

Predator composition

A total of 130 macroinvertebrate predators belonging to 3 orders: Hemiptera (57.7%), Odonata (28.5%) and Coleoptera (13.9%) were collected from the permanent mosquito habitat. Twenty species representing eleven families and 18 genera were identified. Six families (Naucoridae, Nepidae, Corixidae, Hydrometridae, Notonectidae, and Pleidae) were recorded within the order Hemiptera, with members of family Naucoridae and genus *Naucoris* (28.46%) dominating. Three families (Libellulidae, Aeshnidae, and Coenagrionidae) were registered within Odonata, dominated by the Libellulidae family, while the order Coleoptera was represented by two families (Dytiscidae, Noteridae) dominated by *Laccophilus* (10%) (Table 3).

Abundance comparison

One-way Tukey test comparisons in Figure 2 showed significant differences between the abundance of mosquitoes and predators in the Taksebt Dam sites. In fact, there was a significant difference between the abun-

Table 3. Abundance and mean ± SE and distribution of predator species sampled.

Order	Family	Species	T1	T2	T3	Abundance
Hemiptera 57.69%	Naucoridae	*Naucoris maculatus* Fabricius 1798	–	1.9 ± 0.5	–	28.46%
	Nepidae	*Nepa cinerea* Linnaeus 1758	–	–	1 ± 0.1	1.54%
	Corixidae	*Corixaaffinis* Leach 1817	0.3 ± 0.2	0.3 ± 0.2	–	7.69%
	Hydrometridae	*Hydrometra stagnorum* Linnaeus 1758	–	–	0.2 ± 0.1	2.31%
	Notonectidae	*Notonecta glauca* Linnaeus 1758	0.8 ± 0.7	0.4 ± 0.1	–	13.85%
	Pleidae	*Plea minutissima* Leach 1817	–	0.3 ± 0.2	–	3.85%
Odonata 28.46%	Libellulidae	*Orthetrum cancellatum* Linnaeus 1758	–	0.2 ± 0.1	–	2.31%
		Crocothemis erythrea Brullé 1832	0.8 ± 0.4	–	–	7.69%
		Sympetrum striolatum Charpentier 1840	–	–	0.1 ± 0.1	1.54%
		Sympetrum fonscolombii Sélys 1840	0.2 ± 0.1	–	–	1.54%
	Aeshnidae	*Aeshna mixta* Latreille 1805	0.1 ± 0.1	0.5 ± 0.2	–	8.46%
		Anax imperator Leach 1815	–	0.1 ± 0.1	–	0.77%
		Anax parthenope Selys 1839	0.1 ± 0.1	–	–	0.77%
	Coenagrionidae	*Ischnura graellsii* Rambur 1842	–	–	0.3 ± 0.2	3.85%
		Coenagrion scitulum Rambur 1842	–	0.1 ± 0.1	–	1.54%
Coleoptera 13.85%	Dytiscidae	*Laccophilus minutus* Linnaeus 1758	1 ± 0.5	–	–	10%
		Hyphydrus aubei Ganglbauer 189	–	0.05 ± 0.05	–	0.77%
		Ilybius sp. Erichson 1832	–	0.05 ± 0.05	–	0.77%
		Agabus didymus Olivier 1795	–	–	1 ± 0.1	0.77%
	Noteridae	*Noterus laevis* Sturm 1834	0.2 ± 0.2	–	–	1.54%
Total orders	Total families	Total species	Total individuals	Total individuals	Total individuals	Total individuals
3	11	20	43	74	13	130

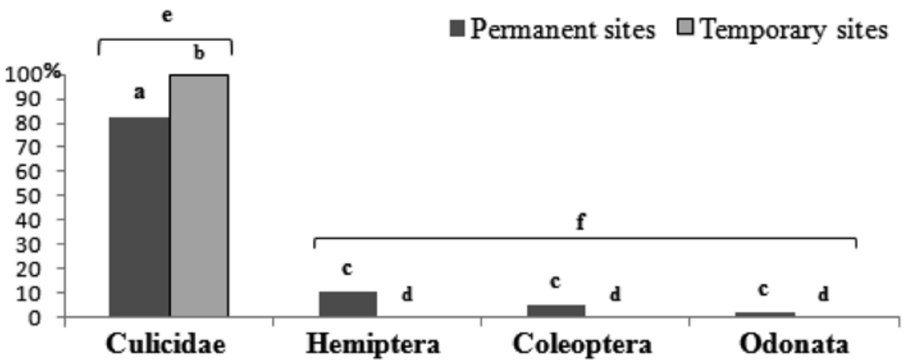

Figure 2. Comparison between aquatic insect abundances in permanent and temporary sites. $p < 0.05$ (one-way ANOVA for taxa comparison: (e–f); Sites-predators (c–d); Sites – Culicidae: (a–b).

dance of predators and mosquitoes (df = 3; f = 340.44; $p < 0.001$), but also a difference between the abundance of mosquitoes in the temporary and permanent sites (df = 1; f = 28.23; $p < 0.05$), as well as a significant difference in the abundance of predators in the temporary and permanent sites (df = 1; f = 18.42; $p < 0.001$).

Relationship between mosquito larvae and predators

The first GLM model (with negative binomial) considered all mosquito larvae, while the second model considered *Anopheles* spp. larvae only, and the third model used *Culex* spp. larvae only (Table 4). The first model revealed that all mosquito larvae were affected only by the abundance of species of the order Hemiptera, while the second model estimated that the abundance of *Anopheles* species was significantly affected by abundances of Odonata and Hemiptera spp. and not by the abundance of Coleoptera. While for the third model, the predation effect between *Culex* larvae and predators was not significant.

Table 4. Generalized linear model results for the mosquito and predator abundance in Taksebt Dam.

Response variable	Estimated coefficient		SE	Z	p
Total species abundance	(Intercept)	4.66334	0.31421	14.841	<2e^{-16} *
	Odonata	–0.04731	0.09044	–0.523	0.60092
	Hemiptera	–0.13313	0.05151	–2.584	0.00976 *
	Coleoptera	–0.04495	-0.04495	–0.340	0.73383
Anopheles abundance	(Intercept)	3.02093	0.14494	20.843	<2e^{-16} *
	Odonata	–0.40112	0.09937	–4.037	5.42e^{-05} *
	Hemiptera	–0.05944	0.02681	–2.217	0.0266 *
	Coleoptera	0.17635	0.14106	1.250	0.2112
Culex abundance	(Intercept)	4.44394	0.58659	7.576	3.57e^{-14} *
	Odonata	0.02724	0.16543	0.165	0.8692
	Hemiptera	–0.16912	0.09523	–1.776	0.0758
	Coleoptera	–0.04882	0.24023	–0.203	0.8390

SE: Standard error. Z: (Estimate / SE), *p* (>|z|) significant at < 0.05.

DISCUSSION

We collected 1172 mosquito larvae among six breeding sites of the Taksebt Dam, with a total of 94 samples. Larvae of the four mosquito genera *Culex*, *Anopheles*, *Aedes* and *Uranotaenia* were found and a total of 13 species were identified based on morphological identification of the fourth larvae stage as well as the examination of the male genitalia, which carries the most discriminating and distinguishing characters. *Culex* was the most species-rich genus, with *Culex pipiens* being the dominant species in the temporary sites and *Culex perexiguus* in the permanent sites (Table 2). A high plasticity of the *Culex pipiens* complex to a wide range of ecological conditions in Algeria was demonstrated (Amara Korba et al. 2016), and according to Lafri et al. (2019), these two species are potential vectors of West Nile Virus in the area. The main groups of potential aquatic insect predators of mosquitoes in Taksebt habitats are shown in Table 3. A total of twenty species from three orders (Odonata, Coleoptera, and Hemiptera) were recorded belonging to 18 genera (*Naucoris, Nepa, Corixa, Hydrometra, Notonecta, Plea, Orthetrum, Crocothemis, Sympetrum, Aeshna, Anax, Ischnura, Coenagrion, Laccophilus, Hyphydrus, Ilybius, Agabus,* and *Noterus).* Species richness, diversity, and aquatic insect numbers showed a significant difference between permanent and temporary sites (Table 1). A higher species diversity (S = 18) as well as a high Shannon diversity index (1.91) were recorded upstream from the dam, which can be attributed to a high spatial heterogeneity and vegetation cover of this habitat.

According to Pintar and Resetarits (2020), the distribution between permanent and temporary habitats is one of the most dominant abiotic features influencing community structure in freshwater systems. Indeed, mosquito larvae abundance in temporary and permanent sites was significantly different; it was higher in temporary sites (Figure 2), and it was obvious that there would be fewer mosquitoes in habitats with predators. The aquatic entomofauna was less diverse in temporary sites, and predatory insects were present only in permanent habitats, in agreement with the result of Fischer and Schweigmann (2008), which indicates that predation in temporary habitats is less important than in permanent habitats. In fact, the breeding site water must persist long enough to allow colonization by predatory species.

According to Vonesh and Blaustein (2010), the density and diversity of mosquito predators can directly affect the abundance of mosquito larvae through predation and indirectly by preventing the oviposition of female adult mosquitoes, and a negative effect of predation could be detected after five months of sampling (Ohba et al. 2013). The abundance of mosquito larvae in the Taksebt Dam was negatively affected by predator abundance (Table 4). A negative relationship between mosquito larvae and Hemiptera spp. abundances was observed. Nattawut et al. (2019) reported that members of Hemiptera significantly suppress *Culex* spp. density. In this study, Hemiptera were the most abundant group (57%), Notonectidae and *Naucoris* were the most abundant Hemipteran predators. According to Silberbush et al. (2014), the presence of Notonectidae predators in water bodies can reduce adult mosquito oviposition. With a daily mean predation of 71.5 larvae, the backswimmer is described as the most aggressive predator on *Anopheles* mosquito larvae (Eba et al. 2021), while Buxton et al. (2020) states that Notonectidae colonize a wide range of aquatic habitats where they are often top predators. Moreover, nine species of Odonata were identified and showed a negative effect on the abundance of *Anopheles* spp. Various species from the order of Odonata are known to be voracious predators of mosquito larvae (Weterings et al. 2015). Dragonfies and damselfies are among the main predators associated with *Anopheles larvae* (Roux and Robert 2019); based on laboratory test results, Libellulidae was found to feed on larvae at a higher rate than Coenagrionidae (Dasrat and Maharaj 2021). In Swedish wetlands, beetles were found suitable for biological control of mosquitoes (Vinnersten et al. 2009). Species from *Laccophilus, Agabus, Noterus, Ilybus,* and *Hyphydrus* genera have already been selected to reduce mosquito larvae (Ohba and Takagi 2010), and experimental studies (Vinnersten et al. 2015) have detected *Aedes* larvae from the diving beetles gut (Dytiscidae). Nonetheless, there was no predation effect of the beetle species on mosquito larvae in our study and predatory efficiency of the identified beetle members remains unknown. According to Pintar et al. (2021), differences in biotic and abiotic habitat characteristics are also involved in the geographic variation of species interactions, and understanding which factors favour mosquitoe and macroinveterbrate co-occurrence may contribute to the control of vector-born disease (Dida et al. 2015).

CONCLUSION

To conclude, permanent habitats of the Taksebt Dam are of major importance for many species of mosquitoes and predators. This study demonstrated the presence of high interaction between mosquito larvae and predators in wetlands, hence the need to communicate and cooperate between mosquito and wetland management regarding the possibility of integrating natural enemies into overall management plans as a new method that will reduce labour costs and other inputs in order to preserve human health and wetlands.

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