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Eco-biological Study of the Mosquitofish *Gambusia affinis* from Oubeira Lake

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Abstract

Gambusia was studied in Oubeira lake in spring of 2016. A total of 100 specimens were collected in the lake. The sex ratio was F:M = 5.12: 1. The length-weight relationship was estimated for the population, with $Wt= 0.0197Lt^{2.4451}$. The condition factor was calculated as C=1±0.2 (for population) and a significant difference was observed between males and females. For 100 examined stomachs, the diet mainly consists of algae (N=54.96%, Pi= 166%, O=91%), followed by the mosquito larvae (N=21.52%, Pi= 65%, O=49%), detritus (N=19.2%, Pi= 58%, O=46%) and arthropods (N=4.3%, Pi= 13%, O=13%). For mosquitoes, two genera were recorded: *Culiseta* and *Culex* with 4 species: *Culex pipiens*, *Culex theileri*, *Culex laticinctus*, and *Culex antennatus*. The culture experiments confirmed the important predation on mosquito larvae was observed with feeding intensity for females and males of 86 ± 12.28 % and 92 ± 8 % for the Culex larvae and 91 ± 7 and 93.66 ± 5.68 for the Culiseta larvae, respectively. No significant difference was observed between the sexes or for mosquito larvae. Our data support the idea that the introduction of mosquitofish has both negative and positive effects since this fish feeds on the most abundant food and not only on mosquitoes.

INTRODUCTION

In Algeria, introduced freshwater species continue to be restocked in many dams, and areas for economic, ecological, and aquaculture reasons (Kara, 2012). This introduction of alien species is considered among the major factors of global change and freshwater biodiversity loss (Gkenas *et al*., 2012; Ricciardi and MacIsaac, 2011). Currently, more than 50% of the total ichthyological diversity in Algeria represents introduced fish (Kara, 2012). One of the most widely introduced fish is mosquitofish *Gambusia affinis* (Baird and Girard, 1853).

The Mosquitofish, originally native to the coastal region of the southeastern United States, is found in Alabama, throughout the Mississippi River drainage,

into Texas and eastern Mexico (Rivas, 1963), this species has been widely introduced in the world in mosquito control programs. This species was introduced in Khemissa (Souk Ahras province, Algeria) around 1928, to biocontrol of Anopheles mosquitoes which transmit the Plasmodium parasites that cause malaria. In Algeria, *Culex pipiens* (Diptera: Culicidae) is the mosquito species of most interest because of its wide geographical distribution, its abundance, and its real nuisance, especially in urban areas (Rehimi and Soltani, 1999). The last biocontrol introduction made to date was carried out by the Directorate of Fisheries and Fishery Resources (DPRH) of Algiers in July 2021 by the introduction of 200 adults of *Gambusia* in Dounia park, Algiers province (CNRDPA, 2021).

However, apart from mosquitoes, *Gambusia* has a variability of items. In effect, dietary studies show that *Gambusia* feeds on a variety of diets, from detritus to organisms like insects, aquatic invertebrates, algae, fish, amphibian eggs, and larval stages (Macdonald and Tonkin, 2008; Singh and Gupta, 2010). Consequently, *Gambusia* is omnivores or even generalist predators, capable of changing the contents of their diet depending on food availability in their habitat (Macdonald and Tonkin, 2008). This can lead to a decrease in the abundance of prey species, especially during the breeding season, when food requirements are high by increasing the intraspecific intensity of competition for food (Hurlbert and Mulla, 1981).

To get a more complete picture of the influence of *Gambusia* introduction on the food web across a big Algerian lake we studied and aimed to assess: (a) biological characteristics and compare it with other studies on mosquitofish; (b) dietary habits; (c) feeding intensity (food per fish per unit time) under controlled conditions and to sex and mosquitoes. These data are very important for the assessment and management of fish stocks (Cai *et al*., 2019).

METHODOLOGY

Place and Time

Gambusia fishing was conducted at Oubeira lake using a beach seine in the two stations (36° 52.017'N 008°22.66'E and 36° 52.028'N 008° 22.614'E) in April 2016. The caught individuals were immediately dissected (fresh specimens). Mosquito sampling was collected in Reghaia lake, in April 2016. The study site is an endorheic permanent freshwater lake, covering an area of 3160 ha, with an average depth of 1.24 m (Direction generale des forets, 2003). The mosquito sampling took place in Reghaia lake on the Algerian Mediterranean coast (from 3°19’E to 3°21’E and 36°45’N to 36°48’N, Algiers). The wetland is fed by three rivers: Reghaia, El Biar, and Boureah River. The lake has a total surface area approximately of 100 ha.

Research Materials

Temperature, pH, salinity, and dissolved oxygen (DO) were measured immediately in the field at both lakes using a multi-parameter analyzer (WTW 340i, France). Regarding the mosquito larvae collected from Reghaia lake, the identification was carried out using the Mediterranean Africa software (Brunhes *et al*., 1999) with a microscope (Optika, Italy) at 10x and 40x magnification after following a special mounting protocol (Rioux, 1958).

Research Design

The study took place in aquariums with 50 L capacity. The experiments were carried out on three females (5.6 ±0.26 cm) and three males (3.7 ±0.1cm) at a temperature of 25 °C (an individual /aquarium), feed every 24h with *Culex* and *Culiseta* larvae (n=100) collected from Reghaia lake, after a 24-hour fast.

Work Procedure

Study of *Gambusia* Population

For each fish, the total length and weight were measured and the sex was determined. The condition factor was

calculated according to the formula of Fulton's coefficient of condition factors as follows (Ricker, 1975):

$$C = \frac{W \times 100}{L^3}$$

Where:
C = condition factors
W = weight (g)
L = length (cm)

Diet

The stomach contents of fished *Gambusia* were stored in 4% formalin. The identification of the prey was conducted in a generic state and counting was done on the easily identifiable body parts. The preys were determined to be the possible lowest taxon. Some indices have been estimated to quantify the importance of different prey items in the diet of *Gambusia*. The stomach vacuity index (VI), percentage frequency of occurrence (O), percentage numerical abundance (N), and predation intensity (PI) were calculated as follows (Hynes, 1950):

$$VI = \frac{ES}{TES} \times 100$$

$$O = \frac{SP}{TES} \times 100$$

$$N = \frac{NP}{TP} \times 100$$

$$PI = \frac{TPC}{TES} \times 100$$

Where:
ES = empty stomach
TES = total examined stomach
SP = number of stomach containing prey
NP = number of each prey item
TP = total number of prey
TPC = total prey category

Data Analysis

Linear regression for *Gambusia* population was determined to estimate the length-weight relationship (Ricker, 1975), and the constants a (initial growth coefficient) and b (relative growth rate of size parameters) were estimated, while t-tests (H0=3) at a confidence level of 95% were applied to determine if relative growth were allometric.

For the condition factor (C), the differences between females and males in groups were tested by t-test.

RESULTS AND DISCUSSION

The Oubeira lake is one of the most important hydro systems in Algeria. This natural system is situated in the extreme northeast of Algeria in the National Park of El Kala (36°50' N and 08°23' E) classified as a biosphere reserve by UNESCO in 1990.

The mean values and standard deviations of the physicochemical parameters for both lakes are given in Table 1. The temperature and the pH were significantly higher in Reghaia lake than in Oubeira lake.

Table 1. Value of physical and chemical parameters in lakes.

Physical Parameters	Lakes	
	Oubeira	Reghaia
Temperature (°C)	12.41 ± 0.46	26 ± 0.07
pH	7.14 ± 0.15	8.75 ± 0.01
Salinity (PSU)	0.81 ± 0.02	0.8 ± 0.007
DO (mg/L)	2.75 ± 0.3	2.68 ± 0.39

Several authors reported that temperature, pH, and other environmental, can influence the feeding rates (i.e., the number of prey items caught) and diet diversity of *Gambusia* directly or indirectly on aquatic biodiversity (Blanco *et al*., 2004; Cabral *et al*., 1998). However, Alcaraz and García Berthou (2007) noted that *Gambusia* can tolerant to a wide range of water quality parameters with very low oxygen (Cech *et al*., 1985), and high temperatures (Pyke, 2005). The results in the present study of physicochemical parameters can explain the wide adaptation and proliferation of Gambusia in Oubeira lake.

The presence of mosquito species in Reghaia lake is the result of several factors

including physicochemical parameters. The temperature, pH, salinity, and dissolved oxygen recorded during the present study are favorable for the development of several species of mosquitoes as mentioned by several authors (El Ouali Lalami *et al.*, 2010; Ibrahim *et al.*, 2011; Marc *et al.*, 2016; Iro *et al.*, 2020).

The overall sex ratio for *Gambusia* was 5.12 : 1 female to male. The total length and weight of Gambusia collected from Oubiera lake ranged between 2.5 cm and 6.4 cm (Mean ± SE: 3.63 ± 0.6 cm) and between 0.1 g and 2.2 g (Mean ± SE: 0.5 ± 0.26g), respectively. Males total length and weight ranged from 2.7 cm to 6.4 cm (Mean ± SE: 0.5± 0.26 cm) and 0.1 g to 2.2 g (Mean ± SE: 0.56 ± 0.55g) and females from 2.5 cm to 4.8 cm (Mean ± SE: 3.59±0.37 cm) and from 0.2 g to 1 g (Mean ± SE: 0.5±0.14 g), respectively. Length-frequency distribution is shown in Figure 1. Length frequency distribution indicates that the [3.5- 4.5[length classes had the highest number of females, however, the [2.5-3.5[length classes had the highest number of males. In regards to the weight frequency distribution, the [0.1-0.6[weight classes had the highest number of both females and males (Figure 2).

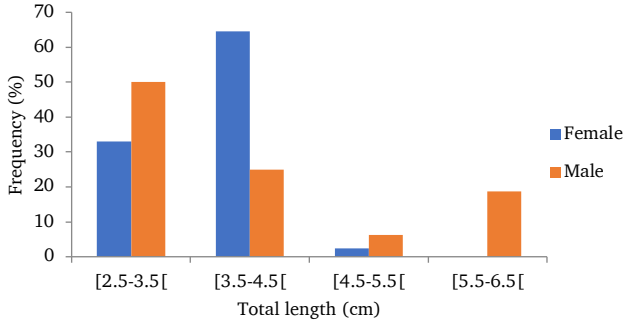

Figure 1. Length-frequency distribution of *Gambusia* (n=100).

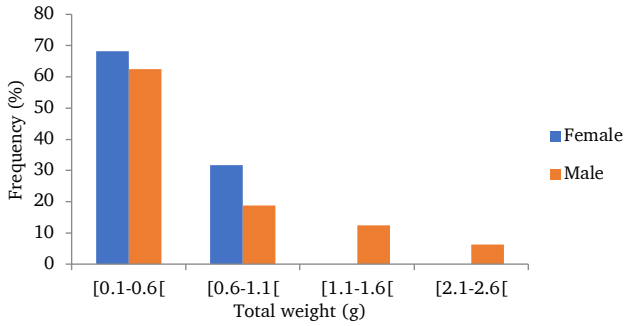

Figure 2. Weight-frequency distribution of *Gambusia* (n = 100, SD = 0.37).

The condition factor was estimated for the entire population (C=1±0.2) and sexes. The average condition factor value estimated for females (C=1.06±0.13) was significantly important than for males (C=0.8±0.31).

The relationships between parameters are given in Figure 3. Negative allometry was detected in the relationships of total weight against total length (b=2.44, r= 0.71). This means that the size of the Gambusia grows faster than its total weight.

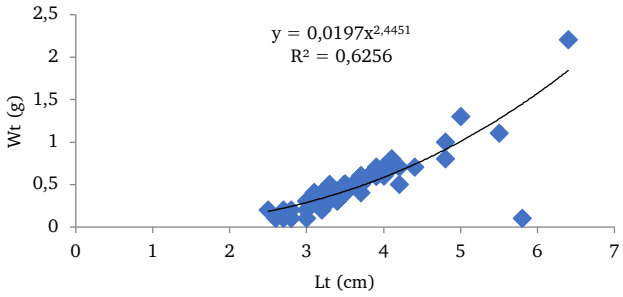

Figure 3. Relationships between size and weight parameters of *Gambusia* from Oubiera lake (b=slope, r= correlation coefficient, n= 100).

For the feeding intensity, among 100 stomachs of Gambusia examined, 4 were empty (VI = 4%). Four major groups, algae and mosquito larvae, were identified in the stomach contents (N=100) of Gambusia. For all the studied indices, algae constitute the most frequently observed group (91%, followed by mosquito larvae. The other groups, i.e., detritus (46%) and arthropods (13%) were of a secondary group (Table 2).

Table 2. Diet composition of *Gambusia* (N%: Numerical composition, PI %: Predation intensity, O%: Frequency of occurrence, n=100).

Diet	N (%)	PI (%)	O (%)
Algae	54.96	166	91
Mosquito larvae	21.52	65	49
Detritus	19.2	58	46
Arthropods	4.3	13	13

In the present study, two genera of mosquitoes were identified: *Culiseta* and *Culex* with 4 species: *Culex pipiens* (Linné, 1758), *Culex theileri* (Theobald, 1903), *Culex laticinctus* (Edwards, 1913) and *Culex antennatus* (Becker, 1903).

In both culture experiments, important predation on mosquito larvae was observed with feeding intensity for females and males of 86 ± 12.28 % and 92 ± 8 % for the *Culex* larvae and 91 ± 7 and 93.66 ± 5.68 for the *Culiseta* larvae, respectively. No significant difference was found between the sexes or by mosquito larvae.

There is scarce data on the *Gambusia* population structure in Algeria. The maximum length of females (4.8 cm) observed in the present study was similar or close to the length of 4.91 cm reported by Sellaoui and Bounaceur (2020) for *Gambusia* of Abadla dam and inferior to the other *Gambusia* populations of Oued Sebaine (6.02 cm), Gazelle fountain dam (6.402 cm) and Timimoune's foggara (5.9 cm). However, the maximum length obtained of males (6.4 cm) in the present study was slightly more than the length of *Gambusia* of Abadla dam (3.805 cm), Oued Sebaine (4.307 cm), Gazelle fountain dam (4.507 cm) and Timimoune's foggara (4.3 cm) reported by Sellaoui and Bounaceur (2020). The proportion of males increased in the large length classes oppositely to females. This result was different and opposite from the values found by Sellaoui and Bounaceur (2020). Vargas and de Sostoa (1996) noted that females take more time to mature and continue to grow throughout their life. However, Krumholz (1948) reported that males stop growing or grow very soon after the gonopodium formation.

The sex ratio of *Gambusia affinis* from the Oubeira lake was 5.12 : 1 (females : males). This result is close to or similar to other results found by Öztürk

(2004) in Akgol (4.38:1), Dalaman (2.51:1), and Ortaca (2.38:1), and Sellaoui and Bounacer (2020) in Timimoune (2.14:1), Bechar (2.19:1), Biskra (2.61:1) and Tiaret (2.35:1) for *Gambusia affinis*.

In the present study, the condition factor value for the entire population was 1±0.2. Erguden and Goksu (2009) noted a similar condition factor value (1.28±0.04) for *Gambusia affinis*. Also, Erguden (2013) worked in Seyhan Dam lake (Turkey) and reported a condition factor value of 1.38±0.26 for the entire population of *Gambusia holbrooki* (Girard, 1859).

Dietary studies of *Gambusia affinis* show a low proportion of empty stomachs and a lot of prey items per stomach. The results obtained in the Oubeira lake are very similar to those reported in the literature (Crivelli and Boy, 1987; Blanco *et al*., 2004).

The present finding showed that *Gambusia affinis* is an omnivore, that feeds on the most abundant food in the lake. Sokolov and Chvaliova (1936) studied *Gambusia affinis* from the rice field of Turkestan and reported that Anopheles, Cladocera, Coleoptera, Rotatoria, and Acarina are the major food items. Etnier and Starnes (1993) noted that the stomach content of *Gambusia affinis* was determined by terrestrial and aquatic insects, crustaceans, and other invertebrates. Ghrab and Bouattour (1999) confirmed the high efficacy of *Gambusia affinis* against mosquito larvae, under laboratory conditions and the trophic test shows that this fish has a clear preference for Culicidae larvae. However, García-Berthou (1999) reported that mosquitofish ingest a wide range of algae, terrestrial insects, and crustaceans rather than mosquitoes in Banyoles lake (Spain). In addition, Al Hafedh (2007) found that green algae were the dominant food item in *Gambusia* food followed by the mosquito larvae, benthic algae, other insects and crustaceans. Erguden (2013) mentioned that *Gambusia holbrooki* is a carnivore in Seyhan Dam lake that feeds on Diptera, Crustacea, Fish, and Fish eggs. For the mosquitoes, the situation of Reghaia provides a suitable habitat for mosquitoes to breed and grow. Brunhes *et al*. (2000) noted that the Culicidian fauna of Algeria includes 48 species. In a previous study on Culicidae biosystematics at four stations in the eastern part of Algiers, Lounaci (2003) recorded 13 species belonging to two sub-families: *Aedes caspius*, *Anopheles labranchiae*, *Culex mimeticus*, *Culex perexiguus*, *Culex pipiens*, *Culex theileri*, *Culex impudicus*, *Culex territans*, *Culex hortensis*, *Culex modestus*, *Culiseta longiareolata*, *Culiseta subochrea* and *Uranotaenia unguiculata*. Benhissen *et al*. (2018) identified *Culiseta longiareolata*, *Culiseta subochrea, Culex brumpti*, *Culex déserticola*, *Culex laticinctus, Culex perexiguus, Culex pipiens*, *Culex theileri.* Matoug *et al*. (2018) recorded *Culex pipiens*, *Culex impidicus*, *Culex hortensis*, *Culiseta longireolata*, *Culiseta annulata*. Some of them were collected in our study.

CONCLUSION

In the present study, the biological and ecological characteristics have provided novel and complementary data for the Gambusia population in East Algeria. The sex ratio was much more in favor of females and the average condition factor value calculated for females was significantly higher than for males. Remarks are that the present study confirms the field data showing a greater impact of mosquitofish introduction on the food web, where major groups were identified in the stomach contents which are the algae (the most frequent prey found with 91%), mosquito larvae (49%), detritus (46%), and arthropods (13%). Despite its feeding intensity towards mosquitoes, predation of mosquitofish is one of the most abundant prey in the biotope.

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