Preliminary investigations of the chironomid larvae fauna (Chironomidae, Diptera) from the Mavrovo reservoir – Republic of Macedonia

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Abstract Larval stages of the Chironomidae (Diptera) can be found in almost any aquatic habitat, including artificial lakes. They form an important fraction of the macrozoobenthos of most freshwater ecosystems. The Mavrovo reservoir as an artificial lake in the Republic of Macedonia, to date, has never been an object of hydrobiological research of this type. For that reason, the purpose of this study was to conduct research on the composition and structure of the chironomid larvae fauna that inhabits the Mavrovo reservoir. The quantitative samples for this research were collected during one year (2017/18), seasonally from five different localities (T1–T5) – T1-Ulazna, T2-Hotel Radika, T3-Middle of the reservoir, T4-Bunec, and T5-Old church (village Mavrovo). According to the results, Chironomidae was represented with five subfamilies, two tribes, and 25 taxa. The subfamilies are Chironominae with the two tribes Chironomini and Tanytarsini, Tanypodinae, Orthocladiinae, Diamesinae, and Prodiamesinae. The dominant taxon is Tanytarsus sp., and besides it, the following taxa: Procladius sp., Chironomus plumosus, Harnischia sp., Polypedilum nubeculosum, and Orthocladius saxicola are characterized by greater community participation. The highest density of chironomid larvae fauna was recorded during the summer, a total of 1013.69 ind/m², compared to the lowest registered in the spring season which is only 9.78 ind/m².

Badania wstępne nad fauną larw ochotkowatych (Chironomidae, Diptera) w zbiorniku Marvrovo – Republika Macedonii

Słowa kluczowe zbiornik, ochotkowate, struktura zgrupowań, dominacja, zagęszczenie sezonowe

Streszczenie Stadia larwalne Chironomidae (Diptera) są znajdowane w niemal wszystkich środowiskach wodnych, łącznie z jeziorami zaporowymi. Stanowią one bardzo ważną frakcję makrozoobentosu w większości słodkowodnych ekosystemów. Zbiornik Marvoro jest jeziorem zaporowym w Republice Macedonii i do tej pory nie było obiektem tego typu badań hydrobiologicznych. Z tego powodu celem było przeprowadzenie badań składu i struktury fauny larw ochotkowatych zamieszkujących zbiornik Marvovo. Badania prowadzono za pomocą prób ilościowych, pobieranych w ciągu jednego roku (2017/2018), w poszczególnych sezonach wegetacyjnych w następujących lokalizacjach (T1–T5) – T1 –Ulazna, T2 – Hotel Radika, T3 – środek zbiornika, T4 – Bunec i T5 – stary kościół (wieś Mavrovo). Fauna Chironomidae była reprezentowana przez pięć podrodzin, dwa plemiona i 25 taksonów. Podrodziny to Chironominae z dwoma plemionami Chironomini i Tanytarsini, Tanypodinae, Orthocladiinae, Diamesinae i Prodiamesinae. Dominującym taksonem był *Tanytarsus* sp., wysokimi udziałami w zbiorowisku charakteryzowały się również taksony: *Procladius* sp., *Chironomus plumosus, Harnischia* sp., *Polypedilum nubeculosum* i *Orthocladius saxicola*. Najwyższe zagęszczenie larw ochotkowatych odnotowano latem, łącznie 1013,69 ind/m², w porównaniu do najniższego zarejestrowanego w sezonie wiosennym, który wynosi zaledwie 9,78 ind/m².

Introduction

The reservoirs are artificial ecosystems and their ecological functioning has characteristics falling between a river and a lake (Callisto et al., 2005). They are built to provide water reserves for a variety of purposes, including electricity, households, industrial materials, transportation, irrigation, and recreation (Prat et al., 1992; Petridis, Sinis, 1993; Holdren et al., 2001; Tundisi et al., 2008; Tskhai, 2015). The construction of a dam causes significant changes in the natural conditions of the river ecosystem and neighboring territories (Armitage, 1984; 1987; Petts, 1984; Penczak et al., 1998, Kornijów, 2009; Tskhai, 2015). This has a great impact on the entire aquatic life in that area. Chironomidae are a family of Diptera (Culicomorpha) with a wide distribution in every type of aquatic ecosystems (Giłka, 2009). In Macedonia, to date, it has been widely researched, especially in natural lakes (Ohrid, Prespa, and Dojran Lake) (Angelovski, 1980; Angelovski, Shapkarev, 1983;1991;1994), while in the reservoirs it's been poorly explored (Sidorovska, 1988, Smiljkov, 1996; Miljanovič et al., 2004; Slavevska-Stamenkovic, 2007; Rimceska, 2014). Mavrovo Lake as an artificial reservoir over the years has almost never been the subject of research of this type. In the context of hydrobiological data for this reservoir, Stojkovski (1960) published a paper on the composition of the benthic community several years after the construction of the dam, ie after the creation of the lake. Shortly afterward, Popovska-Stankovic (1963) conducted another study of the Mavrovo reservoir on zooplankton from the first years of the lake's existence. Regarding the study of the physicochemical characteristics and the ecological status of Mavrovo Lake, a master thesis was prepared by Latifi, Lj. (2011). Recently, there is almost no hydrobiological research of the Mavrovo reservoir as an artificial lake. Starting from that fact, and taking into account the specific characteristics of the selected group of animals – chironomids, originated the need, and also the goal to conduct research on the composition and structure of the chironomid larvae fauna that inhabits the Mavrovo reservoir.

Study area

One of the biggest artificial reservoirs on the territory of the Republic of Macedonia is Mavrovo Lake. It is located in the northwest region of the municipality of Mavrovo at the base of the mountains Bistra and Korab. In the south, it is limited by forested and grassland Vlainica and in the north by Shar Planina, at an altitude of 1223 m a.s.l. The lake started to form in 1953 when a high dam was built in the western part of the valley between Mavrovi Anovi and the village of Mavrovo. The Mavrovo reservoir belongs to the category of mountain lakes of alpine character, so it freezes during the winter. It is the highest mountain lake in Macedonia. The total length of the lakeshore is 24.25 km. The depth of the lake is relatively variable, but it is considered that the maximum depth is 40 m. Water temperature has significant seasonal and altitude changes. The average monthly temperature of the lake water in July and August is around 20°C, and the highest limit can exceed that limit.

The average monthly temperatures in the investigated area are negative in the three winter months and in January they are -2.2° C. The highest average temperatures are in July, 16.3°C, and August 16°C, so that the average annual temperature variation is 18.7°C. The samples for this research were collected seasonally in the period from July 2017 to June 2018 from 5 different points – Ulazna (T1), Hotel Radika (T2), Middle point of the Lake (T3), Bunec (T4), Old church (T5) (Figure 1). In continuation follows a brief description of the investigated localities:

- T1 (41°42'27.5"N 20°46'34.5"E) this point is located on the northern part of the lake, before the entrance to the settlement of Mavrovi Anovi. The shore of the lake is partly overgrown with grass, and the substrate of the littoral part is composed of coarser and finer stone. The substrate at the bottom is mainly represented by fine sand. Due to the variable level of the reservoir as well as the substrate of this locality, in the months of September, March, and June no samples were collected for quantitative analysis. The depth of this point is 3 m.
- **T2** (41°41'53.9"N 20°47'06.6"E) this site is located between the two villages on the east shore of the lake, Nikiforovo, and Leunovo. The bottom of the lake is composed of sandy substrate and sporadic with plant material residues, due to drifts. The depth of this point is 4.5 m.
- T3 (41°41'31.0"N 20°45'34.1"E) the middle point of the lake is located in the middle of the widest part of the lake, from where on one side you can see the Mavrovo dam, and on the other side is the village Nikiforovo. At the time the samples were collected, the depth was 20 m.
- T4 (41°42'03.7"N 20°47'31.3"E) this site is located on the north side of the lake, between T1 and T3. The lake bottom in this locality is represented by finer and coarser sand. The depth of this locality is 7.4 m.
- T5 (41°39'40.9"N 20°44'09.7"E) this locality is in the southern part of the lake, near the old church which in some periods of the year is submerged in the lake. It is important to note that due to large seasonal changes in the water level, the depth in this locality is not constant. The base of this locality is composed of fine sand and finer and coarser stone, mainly near the Mavrovska River, before the inflow into the lake, while around the lake the ground is muddy. The depth of this locality in the period of material collection was 5 m.

Materials and methods

The materials used for this research were collected from five points during one year (2017/18), seasonally. Three samples were taken from each point, and the results are shown as mean values. The collection of the samples was carried out with the Ekman dredge with a volume of 225 cm² which is used for collecting from a substrate with small particle diameter (fine sand, silt) from the lake. Immediately after taking the samples, they were preserved with 70% ethanol. After the preservation they were transported to the laboratory were permanent microscopic slides were made using a mounting medium of Canada-balsam and then determined to the species level. The identification was done using proper taxonomic keys: D.R. Oliver et al. (1978), B. Rossaro (1982), J.H. Epler (2001), Orendt et al., (2012), Andersen et al., (2013), B. Rossaro and V. Lencioni



Figure 1. Map of the investigated points

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(2015). All identified taxa are in a larval stage, and for their identification were used keys from authors that identified Chironomidae larvae, not pupae or adults. The abundance is shown as the number of individuals of unit meter square (ind/m²) and dominance is calculated with the formula of Balogh (1958). According to this author the values for dominance are divided into four groups: for dominance greater than 10% are considered dominant (D), between 5–10% subdominant (SD), between 1–5% recendent (R), and lower than 1% subrecendent (SR).

Results

During the research conducted on the chironomid larvae fauna of the Mavrovo reservoir in the period 2017/18, the presence od 25 species was established. They belong to five subfamilies and two tribes: subfamilies Tanypodinae, Orthocladiinae, Diamesinae, Prodiamesinae, and Chironominae with the two tribes Chironomini and Tanytarsini (Table 1). As can be seen from the table, most species belong to the subfamily Chironominae, 13 in total.

Таха						
Subfam. Tanypodinae						
Procladius choreus Meigen, 1804						
Procladius sp. Skuse, 1889						
Thiennemannymia sp. Fittkau, 1957						
Subfam. Chironominae – tribe Chironomini						
Harnischia sp. Kieffer, 1921						
H. fuscimania Kieffer, 1921						
H. curtilamelata Malloch, 1915						
Cryptochironomus deffectus Kieffer, 1913						
Polypedilum nubeculosum Meigen, 1804						
P. scalaneum Schrank, 1803						
Chironomus plumosus Linnaeus, 1758						
C. thummi (riparius) Kieffer, 1911						
Microtendipes pedellus De Geer, 1776						
Cladopelma viridula Linnaeus, 1767						
Subfam. Chironominae – tribe Tanytarsini						
Cladotanytarsus mancus Walker, 1856						
Tanytarsus sp. Van der Wulp, 1874						
Micropsectra sp. Kieffer, 1908						
Subfam. Orthocladiinae						
Orthocladius saxicola Kieffer, 1911						
Thienemanniella clavicornis Kieffer, 1911						
Brilia bifida (modesta) Kieffer, 1990						
Cricotopus sylvestris Fabricius, 1794						
Parakiefferiela batophila Kieffer, 1912						
Paracricotopus niger Kieffer, 1913						

Table 1. List of taxa of Chironomidae larvae fauna from the Mavrovo reservvir

Continuation of Table 1

Psectrocladius sordidelus Zetterstedt, 1838					
Subfam. Prodiamesinae					
Prodiamesa olivacea Meigen, 1818					
Subfam. Diamesinae					
Diamesa insignipes Kieffer, 1908					
Total	25				

According to the results shown in Table 2, the average annual density of the chironomid larvae fauna of the Mavrovo reservoir is 539.13 ind/m². The average annual density ranges from 6.67 ind/m^2 in the spring, which is the lowest value, to 2225.9 ind/m², which is the highest density in the summer season.

The Tanytarsini tribe from the subfamily Chironominae has by far the largest representation in this reservoir, 2484.39 ind/m², or 73.18% of the total chironomid larvae (Figure 2). Next is the subfamily Tanypodinae with a presence of 457.29 ind/m² (13.47%), then the tribe Chironomini from the subfamily Chironominae with 330.78 ind/m² (9.74%). Diamesinae and Orthocladiinae with 64.45 ind/m² (1.90%) and 51.28 ind/m² (1.51%) respectively have a smaller share in relation to the overall chironomid larvae fauna. Finally, the subfamily Prodiamesinae with a presence of 6.67 ind/m² or only 0.2% accounts for the smallest and almost insignificant share of the chironomid community in this reservoir.

Table 2. Annual dynamics of the density (ind/m²) of the Chironomidae larvae broken down by seasons and points

Seasons/Points	T1	T2	Т3	T4	Т5
Summer	2,225.9	93.35	909.01	1,840.30	/
Autumn	/	1,251.30	215.58	291.15	1,397.95
Winter	/	688.98	526.73	/	1,293.50
Spring	/	42.23	/	/	6.67
Average annual density (ind/m ²)			539.13		



Figure 2. Percentage representation (%) of the subfamilies/tribes in the investgated period

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In terms of the individual species of the benthos of the reservoir, the dominant taxon is *Tanytarsus* sp., with a share of 67.78% (7318.7 ind/m²) in this community (Table 3). The taxa *Procladius* sp., *Chironomus plumosus*, and *Harnischia* sp. are subdominant with a share of 8,32% (896,8 ind/m²), 8, 03% (865,66 ind/m²) and 5,68% (612,31 ind/m²), respectively.

Species	ind/m ²	%	Dominance	Species	ind/m ²	%	Dominance
<i>Tanytarsus</i> sp. Van der Wulp, 1874	7318.70	67.88	D	Micropsectra sp. Kieffer, 1908	50.01	0.46	SR
Procladius sp. Skuse 1889	896.8	8.32	SD	Cryptochironomus deffectus Kieffer, 1913	45.56	0.42	SR
Chironomus plumosus Linnaeus, 1758	865.66	8.03	SD	Parakiefferiela batophila Kieffer, 1912	20.00	0.19	SR
Harnischia sp. Kieffer. 1921	612.31	5.68	SD	<i>Thiennemannymia</i> sp. Fittkau, 1957	17.78	0.16	SR
Polypedilum nubeculosum Meige, 1804	398.94	3.70	R	<i>Cladopelma viridula</i> Linnaeus, 1767	11.11	0.10	SR
Brilia bifida (modesta) Kieffer, 1990	195.58	1.81	R	Prodiamesa olivacea Meigen, 1818	6.67	0.06	SR
Orthocladius saxicola Kieffer, 1911	131.13	1.22	R	Cricotopus sylvestris Fabricius, 1794	4.45	0.04	SR
Cladotanytarsus mancus Walker, 1856	84.46	0.78	SR	Thienemanniella clavicornis Kieffer, 1911	3.34	0.03	SR
Diamesa insignipes Kieffer, 1908	64.45	0.60	SR	Paracricotopus niger Kieffer, 1913	2.22	0.02	SR
Microtendipes pedellus De Geer, 1776	51.12	0.47	SR	Psectrocladius sordidelus Zetterstedt, 1838	2.22	0.02	SR

Table 3. Average annual density (ind/m²) and percentage representation (%) of the species from the Mavrovo reservoir in the investigated period

At all points (depths) they occur with different density (Figure 3). It ranges from a minimum of 412.83 ind/m² at point T3, which is also the greatest depth at which the samples were collected, up to a maximum of 674. 53 ind/m² at T5. At the other three points, the density is 556.46 ind/m² at T1, 518.95 ind/m² at T2, and 532.85 ind/m² at T4.

As for the percentage representation of subfamilies and tribes, the dominance of the Tanytarsini tribe above all others, along with the vertical distribution, is due to the large representation of the *Tanytarsus* sp. (Figure 4). The highest percentage of this taxon (82.78%) is represented at point T1 and the lowest percentage (53.79%) is represented at T5 regarding the overall chironomid community. On the other hand, the point at which all subfamilies/tribes are represented is T5 (5 m). Here alongside *Tanytarsus* sp., *Chironomus plumosus* is also characterized by great representation, i.e. the 23.97% share in the communitycomposition.

The seasonal dynamics of the chironomid larvae fauna in the investigated period 2017/18, ranges from 1013,69 ind/m², which is the highest density recorded in the summer period, to 9,78 ind/m², which is the lowest density recorded in the spring period (Figure 5).



Figure 3. Dynamics of the average density (ind/m²) of the chironomid community from the Mavrovo reservoir



Figure 4. Average percentage annual representation of the chironomid species broken down by points

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Figure 5. Seasonal dynamics of the average density (ind/m²) of the chironomid larvae fauna in the investigated period

As we can see from Figure 6, in all four seasons *Tanytarsus* sp. is the dominant taxon. Beside it, *Chironomus plumosus* is also characterized by a high percentage in autumn (13.52%), winter (14.70%) and spring (22.72%).



Figure 6. Average percentage annual representation of the chironomid species followed by seasons

Discussion

Mavrovo Lake is an artificial reservoir in the western part of Macedonia. This research is the first study of the species composition of the chironomidae larvae in this reservoir. The results showed the presence of a total of 25 species in the lake. Most species (13) belong to the subfamily Chironominae with the two tribes Chironomini and Tanytarsini, followed by the subfamily Orthocladiinae (6) and Tanypodinae (4). Of the subfamilies Diamesinae and Prodiamesinae, 1 species was found. Each subfamily has its own predispositions to certain environmental conditions. The subfamily Chironominae and certain species of the subfamily Tanypodinae prefer finer sand and silt sediments (Pinder, 1977; Carew, 2007). The general conclusion is that there is a strong correlation between the organic composition of the sediment and the density of most species of these two subfamilies (Moore, 1979; Vos et al., 2004; Carew, 2007). However, interactions between environmental factors can greatly contribute to hide this correlation (Vodopich, Cowell, 1984). Also, many species belonging to the subfamily Chironominae can tolerate conditions with low oxygen concentrations, as well as heavy metal contamination with certain exceptions (Pinder, 1986; Tomilina et al., 2016; Zelalem, Prokin, 2017). Given that in some of the points from which the materials were collected (T2, T5), the conditions coincide with the above mentioned, more precisely it has an anthropogenic impact, the number of taxa with which the subfamily Chironominae is represented is not unexpected.

The dominant taxon in this study is *Tanytarsus* sp. It was present in all seasons during our research. According to Ekrem (2003) and Epler (2014), their adaptability allows them to survive in waters with low quality and clean waters as well. This finding was confirmed by Brundin (1958), who indicates that the dominant representation of the tribe Tanytarsini is associated with a moderate-oligotrophic character of water. According to H.R. Ingvason et al. (2004), in terms of nutrition, Tanytarsini are considered larvae that feed non-selectively on detritus and debris from the surface parts of the sediment, which is another proof and confirmation that they can survive in different types of habitats and sediments. Apart from Tanytarsus sp., Chironomus plumosus is another taxon that covers most of the chironomid larvae fauna of the Mavrovo reservoir. It is important to note that the highest density of this species at point T5 is consistent with the fact that in that part, i.e. in the vicinity of this point, there are a number of catering facilities that discharge their wastewater directly into the lake. The fact that this species inhabits the littoral zone of mesotrophic and eutrophic lakes is confirmed by a number of authors, including Saether (1979). It is characteristic that the density trend of this species shows somewhat similar values in the autumn and winter season at T5. As we said before, along *Tanytarsus* sp. and *Chironomus plumosus*, also Procladius sp., Harnischia sp., Polypedilum nubeculosum, and Orthocladius saxicola are characterized by greater community participation in the Mavrovo reservoir. Procladius choreus and Ch. plumosus are also dominant in the reservoirs Matka (Smiljkov, 1996), Mantovo reservoir (Slavevska-Stamenkovic, 2006), and Prilepsko Lake (Rimcheska, 2014). Regarding Tanytarsus sp., it is dominant in Prilepsko Lake, subrecendent in the Mantovo reservoir and it's not found in the Matka reservoir.

The vertical distribution of the chironomids shows oscillations at different points, with the lowest density established at T3, which is also the highest depth (20 m) at which the samples were collected, while the highest density is at T5 (5 m). According to Salmoiraghi et al. (2001), for most lakes and reservoirs, density, diversity, and the number of species have the highest values in the littoral zone (less than 10 m), and as the depth increases, the number of species decreases. Since point T3 has the greatest depth, it is not an exception to the rule that the more the conditions

deviate from the normal, or rather, the normal optimum of the most species, the smaller the number of inhabited species (Odum, 1959). Also, according to Smiljkov and Slavevska-Stamenkovic (2006), low oxygen concentration, in the deepest parts of the lake, acts as a limiting factor in the development of benthic organisms.

Regarding the seasonal density of the chironomid larvae fauna, in this study, the lowest density was recorded in the spring season. According to various authors, spring is the period when the lowest density of the chironomid larvae fauna is recorded, which coincides with the finding of this research, because in this season there is an emergence of adults. Considering the altitude at which the reservoir is located, as well as the lower temperatures that are characteristic of this mountainous area, the presence of only one generation per year of these larvae can be ascertained. This coincides with the research of certain authors (Tokeshi, 1995, Lencioni, 2004), according to which, voltinism of less than 1 generation is a characteristic of species that inhabit predominantly cold regions. This is confirmed by the research of T. Stojkoski (1960), where the composition of the benthos in Mavrovo Lake was quantitatively examined immediately after the dam was constructed and where it was established that in the spring season (May) there is the lowest density of the chironomids while in the summer-autumn season (September-October) the density of this dipterian family reached its maximum.

Conclusions

Based on the above data, the following conclusions can be drawn:

- 1. Generally, the dominant taxon in this study is *Tanytarsus* sp., which indicates that the water of this reservoir most likely is of moderate-oligotrophic character.
- 2. The dominance of *Chironomus plumosus* at T5 is consistent with the fact that at this locality we have a larger amount of waste material which is discharged directly in the lake by the nearby catering facilities.
- 3. In terms of vertical distribution, the highest density is recorded at point T5 (5 m), while the lowest density at T3 (20 m). It is evident that the depth is a limiting factor that affects the density, ie the deepest parts of the reservoir, which are also the poorest in oxygen, are characterized by lower community density.
- 4. The seasonal dynamics of the chironomid larval fauna showed the highest community density in the summer season and the lowest density in the spring season. This indicates that most likely the chironomids are univoltine, ie they have one generation per year in the Mavrovo reservoir.

However, this research has a preliminary character. In order to get a complete picture of the structure and composition of the Chironomidae larvae, it is necessary to collect more samples and to do longer, wider, comprehensive studies, including physicochemical analysis of the water.

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