## Diversity of Chironomidae (Insecta: Diptera) genera in Taleghan River, Alborz province, Iran

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#### Abstract

Chironomidae is one of the most important elements of the aquatic benthos all over the world, which has its own ecological and biological importance in different aspects. The present study has been conducted to investigate the chironomid diversity in Taleghan River in two seasons, spring and summer 2016 for six sampling points (Asfaran, Varkesh, Mir, Jostan, Barikan and Gooran). The sampling procedure was done by applying a simple dredge in five replications for each sampling point. The results have showed that there was a great diversity of Chironomidae in Taleghan River which composed of four sub-families and 21 different genera; Chironominae (Chironomus, Robackia, Stenochironomus, Omisus, Cryptochironomus, Saetheria, Cryptotendipes, Microtendipes, Paratendipes, Constempellina and Neozavrelia), Orthocladiinae (Bryophaenocladius, Comptocladius, Echinocladius, Smittia, Botryocladius, Symbiocladius, Psectrocladius and Gymnometriocnemus), Tanypodinae (unidentified genera in Pentaneurini tribe) and Diamesinae (Potthastia and Diamesa). Ten genera have been reported for the first time in Iran. Diversity on abundance of each genus in different months and sampling points might be the result of the environmental conditions and water pollution.

Keywords: Fauna, Chironomidae, Taleghan River, Alborz, Iran

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#### Introduction

Studying the biological and ecological aspects in aquatic resources make the basis of natural resource researches which is started by identification the biodiversity of the ecosystem (Mayer, 1963). Invertebrates are the most abundant animal taxa in different and terrestrial aquatic ecosystems (Barnes and Callow, 2001) which need more investigation in Iranian aquatic habitats, especially for ecological, biological and economical important family like Chironomidae. Bloodworm from the family of Chironomidae comprise a family of Dipteran flies which its larval stage is the main active developmental stage by bio-filtering and predation in aquatic habitats (Henriques-Oliveira et al., 2003). The family has been reported as the most abundant group of aquatic insect count and compromised more than 30% of its biomass (King and Wrubleski, 1998). Some of the species having considered as a very considerable food resource for fish and other aquatic animals (Hamid et al., 2015; Milakovic et al., 2001).

The Chironomidae typically have been shunned by many benthologists because of perceived difficulties in specimen preparation, identification, taxonomy, morphology and literature (Epler, 2001); this led to the point that most of the recent studies have ended to identification of genus. On the other hand, as Chironomidae resemble other insects, so the documentation of this fauna throughout the world is not complete (Armitage *et al.*, 1995). Chironomidae has been divided into 11 subfamilies; most of the species belong to Chironominae (Armitage et al., 1995). In Iran, most of the studies have been taxonomic inventories in different aquatic habitats like southern coastline of Caspian Sea (Ahmadi and Mousavi Nanekaran, 2002), Marbor River in Isfahan province (Ebrahimnezhad and Nikoo, 2004), Haji Abad River in Hormozgan province (Khosravani et al., 2014), Ghazal Ozan River in Zanjan province (Navan Maghsoodi, 2013), Zayandehrood River in Isfahan province (Ebrahimnezhad and Fakhri, 2005; Shayeghi et al., 2014). Golpaygan River (Ebrahimnezhad and Allahbakhshi. 2013: Allahbakhshi. 2005) and different rivers in Tehran province (Alvari, 1997; Arkia et al., 2016, 2017, 2019). Karami et al. (2014) provided a checklist and kev identification of Chironomidae Larvae in Marbor River (Isfahan, Iran) and has reported genera from four 39 subfamilies: Chironominae (15 genera), Diamesinae (2 genera), Orthocladiinae genera) and Tanypodinae (17)(5 genera). From these, 13 genera have reported for the first time in Iran. Sharifinia reviewed (2015)the macroinvertebrates of the Iranian running waters through 15 years of recent studies until 2015 and showed that Arthropoda comprised the most taxa with 34 described genera of Chironomidae as the dominant family. In a study on the macroninvertebrates of Karaj and Jajroud rivers. Chironomidae has been reported as one of the main ecological member of the aquatic systems in the area, but there is no genera identification (Taban et al., 2019).

Similar researches around the globe, especially in Middle East countries have been done about diversity of Chironomidae in rivers and coastlines. Findik and Aras (2016) studied the Chironomid limnofauna of Kizilirmak River in Turkey and reported 11 genera in three subfamilies. Butakka et al. (2014) has studied the relationship between Chironomidae diversity and habitat features in Sepotuba River basin, Brazil and showed that feeding habit of Chironomidae larvae. collectors, filterers and predators, had direct relation to habitat condition. One study in Iraa considered the morphology of Chironomus species (Jabrial and Ahmad, 2012).

Arslan et al. (2010) studied the density and species composition of Chironomidae larvae fauna from twelve sampling sites of Lake Uluabat and showed that chironomid larvae were the third dominant group consisting 12.3% of the total zoobenthos density, as Chironomus tentans Fabricius was the most abundant species contributing with about 66.2% of the total chironomid populations. In a similar study in Yuvarlakcay stream in Turkey, 18 genera of Chironomidae were collected (Tasdemir et al., 2010).

Due to the lack of information about Chironomidae diversity in Iran, this study was conducted to investigate the family genera in Taleghan River, in Alborz Province.

#### Materials and methods

#### Sampling area

The study area is situated in the western part of Alborz Province, on the snowy foothills of Alborz Mountains, in Taleghan County. The county is surrounded by Alborz Mountains, Ramsar and Tonekabon cities to the north, Karaj County to the east, Hashtgerd and Savojbolagh to the south and Ghazvin to the west and western south. Taleghan River was selected as the main sampling point which passes the southern edge of Taleghan and ends to Shahrood River. Six sampling points have been considered according to availability, plant growth, the earth slope, branches and main river, and the river bed as Jostan, Gooran, Varkesh, Barikan, Mir and Esfaran (Table. 1). The features of sampling stations such as longitude, latitude and height above the sea level are given in Table 1.

Station	latitude	longitude	Height (m)	
Jostan	50.893°	36.187°	1992	
Gooran	50.869°	36.190°	1920	
Varkesh	50.783°	36.365°	2010	
Barikan	50.735°	36.156°	2050	
Mir	50.729°	36.190°	1800	
Esfaran	50.726°	36.180°	1720	

Table 1: geographical features of sampling stations through Taleghan River.

#### Sampling and preservation method

Samples of larvae were taken monthly through spring and summer 2016, resulting in five replicates for each station. Samples have been taken by dredge sampler and kept in 70% Ethanol until the laboratorial identification. For better systematic identification, permanent mounts were prepared. For this purposes, samples were soaked in a 10% solution of potassium hydroxide (KOH) on a hot plate at the temperature of 70°C. Ten minutes in acetic acid was followed by transfer to absolute alcohol. The dehydration process has performed to prepare the samples transferring to slide. Samples were glutted by a drop of Canada balsam.

#### Systematic identification

To identify the samples, valuable and available identification keys such as, Epler (2001), Ebrahimnezhad and Fakhri (2005),Madden (2010),Ebrahimnezhad and Allahbakhshi (2013) and Karami et al. (2014) have used to the generic level. The features of the head capsule have used for chironomid larvae. In order to identify the genera, head capsules and body characteristics of the larvae have studied under the microscope and according the collected genera, shorts keys have provided for each subfamily.

#### Statistical analysis

In order to determine whether any significant differences existed in larval abundance of the sites and seasons, mean and SD of larval abundance have calculated for each sample in each site

and season; then one way Kruskal-Wallis variance analysis and F test have applied to find any difference among treatments. For subfamilies with less than three genera, student T test has use to compare the mean frequency of the collected genera. After identification, the slides have deposited in the Entomology Laboratory Collection of Islamic Azad University, Varamin-Pishva branch.

#### Results

#### Systematic identification:

Twenty-one genera belonged to four subfamilies Chironominae, (eleven genera), Orthocladiinae (seven genera), Tanypodinae (unidentified genera) and Diamesinae (two genera) have been identified in this study. Subfamilies, genera and tribes have shown in Table 2 in which ten genera have reported for the first time from Iranian fauna (marked by star).

#### Statisctical analysis

Mean and standard deviation of chironomid genera abundance from six sites in two seasons are shown in Table 3. Mean frequency difference by student T analysis method showed that there was no significant difference total frequency the between of Diamesine genera (df=1, F.=1.216, Sig.= 0.299). For Orthocladinae, totally 41 samples in seven genera were collected and the result of mean difference analysis was similar (df=6, F=1.088, Sig.=0.389).

Family	Subfamily	Tribe	Genera
			Chironomus Meigen
			Robackia Saether*
			Stenochironomus Kieffer*
		Chironomini	Omisus Townes*
			Cryptochironomus Kieffer
	Chironomidae		Saetheria Jackson
			Cryptotendipes Lenz
			Microtendipes Kieffer
			Paratendipes Kieffer
		Tanytarsini	Constempellina Brundin*
hironomidae	Orthocladiinae		Neozavrelia Goetghebuer
linononnuae			Bryophaenocladius Thienemann
			Comptocladius Goetghebuer*
			Echinocladius Cranston*
		Orthocladini	Smittia Holmgren
		Ofulociauliii	Botryocladius Cranston and Edwards *
			Symbiocladius Kieffer*
			Psectrocladius Kieffer
			Gymnometriocnemus Goetghebuer *
	Tanypodinae	Pentaneurini	unidentified genera
	Diamesinae	Diamesini	Potthastia Kieffer*
	Diamesinae	Diamesini	Diamesa Meigen

# Table 2: Taxonomic diagram of chironomid larvae identified in five sites from Taleghan River,Iran, 2015-2016.

Eighteen samples of Tanypodinae have found which was unidentified and statistical analysis was not applied for. Chironominae had the most abundant samples among the collected genera and *Chironomus spp*. was considered as the most frequent genera of the subfamily in Taleghan River which showed significant difference with other genera of Chironominae (df=10, F.=6.352, Sig.=0.000). There was not any difference among other member of the same subfamily.

subfamily	Genera	Mean	Std. deviation	SE Mean
	Bryophaenocladius	0.833	0.983	0.401
Orthocladiinae	Comptocladius	0.333	0.516	0.210
	Echinocladius	0.500	0.547	0.223
	Smittia	0.333	0.816	0.333
	Botryocladius	0.333	0.516	0.210
	Symbiocladius	0.166	0.408	0.166
	Gymnometriocnemus	0.333	0.594	0.0859
Chironominae	Chironomus	3.550	2.588	2.056
	Robackia	0.333	0.514	0.210
	Stenochironomus	0.166	0.408	0.166
	Omisus	0.166	0.408	0.166
	Cryptochironomus	0.166	0.408	0.166
	Saetheria	0.166	0.408	0.166
	Cryptotendipes	0.166	0.408	0.166
	Microtendipes	0.166	0.408	0.166
	Paratendipes	1.166	0.752	0.175
	Constempellina	0.500	0.836	0.341
	Neozavrelia	0.667	1.032	0.421
Diamaginaa	Diamesa	0.333	0.816	0.333
Diamesinae	Pottastia	0.834	0.757	0.307

Table 3: Mean number ±SE of different genera of Chironomidae larvae in Taleghan River

Mean frequency difference for Diamesinae samples in different months showed no significant difference in the frequency of the genera (df =3, Chi Sq. =4, Sig. =0.261), most of this subfamily samples were collected in August. However, no statistical analysis has done for Tanypodinae, most of the samples were collected through June. Time of sampling had significant effect on the frequency of Orthocladiinae genera (df=3, Chi Sq. =2.493, Sig. =0.477); the least number of the samples have been collected in August and most in May. Chironominae samples mostly have collected through June, however, there was significant difference among different sampling months on the frequency of the subfamily (df.=3, Chi Sq.=0.379, Sig.=0.379), the lowest number of samples of Chironominae have been collected in July.

Subfamilies have found in different sampling stations is given in Table 4. The result for Diamesinae genera showed that sampling station had significant effect on the number of collected samples; two genera of this subfamilies have been collected from Barikan, Mir, Jostan and Esfaran, which the most number collected from Jostan (df=3. Chi Sq.=3.20, Sig.=0.362). Tanypodinae have been collected from Barikan, Mir, Varkesh and Esfaran and Kruskal Wallis one-way variance analysis showed significant difference among the sampling points (df=3, Chi Sq.=2.50, Sig.=0.475) which Varkesh had the lowest number of collected Tanypodinae. Orthocladiinae have been collected from all the six sampling points with significant difference among the mean number of samples collected in each sampling station (df=5, Chi Sq.=5.362, Sig.=0.373) which Jostan had the most Orthocladiinae samples. Significant difference has observed among collected Chironominae samples from six sampling stations (df =5, Chi sq. =6.454, Sig. =0.265) which Esfaran had the most and Jostan had the least number of Chironominae.

 Table 4: The effect of sampling stations of Taleghan River on the presence of different subfamilies of Chironomidae.

month	Jostan	Mir	Varkesh	Esfaran	Barikan	Gooran
Chironominae	+	+	+	+	+	+
Orthocladiinae	+	+	+	+	+	+
Tanypodinae		+	+	+	+	
Diamesinae	+	+		+	+	

#### Discussion

Among the genera belonged to Orthocladiinae, three genera have been reported previously for the Iranian fauna such as *Bryophaenocladius* and *Smittia* from Talesh Mountains (Aubert *et al.*, 2017), *Psectrocladius* from Talesh Mountains (Aubert *et al.*, 2017) and Marbor River (Karami *et al.*, 2014); and five genera are new to Iranian Chironomidae checklist which have been reported from other parts of the world. *Comptocladius* was at first considered to be a Nearctic region genus (Tucker, 1906), but has reported from Portugal (Coro et al., 2001). *Echinocladius* has been reported previously from Australia (Krosch, 2011) and Korea (Kim et al., 2012). Botryocladius has been reported from Australia (Krosch, 2011), Argentina (Donato et al., 2008) and New Zealand (Boothroyd, 2004). Different species of Symbiocladius have been reported as an ectoparasite of Ephemeroptera taxa from Peru (Prat et al., 2013), Ukraine (Gilka et al., 2007) and Australia (Krosch, 2001). Gymnometriocnemus has been reported from Holarctic region and is considered to have worldwide distribution as Norwey (Stur and Ekrem, 2015) and Australia (Krosch, 2001).

Two genera from Diamesinae subfamily have been collected which Diamesa has been previously reported from Marbor River (Karami et al., 2014) Golpayegan River and (Ebrahimnezhad and Allahbakhshi, 2013); the other genus, Potthestia is new to Iranian fauna as has been reported from Russia as the closest area in Palearctic region to Iran (Ermolaeva, 2009).

Among the genera collected from Chironominae subfamily, seven genera have been reported previously from Iran and the rest are introduced for the first time for Iranian fauna. Chironomus has reported from Talesh been Mountains (Aubert et al., 2017), Lar river (Arkia et al., 2019), Hablehrood River (Arkia et al., 2017), Jajrood River (Arkia et al., 2016) in Tehran province, Marbor River (Karami et al., 2014) and Zayandehrood River in Isfahan

province (Ebrahimnezhad and Fakhri, 2005; Shayeghi et al., 2014), Ghazal Ozan River in Zanjan Province (Navan 2013), Maghsoodi, ponds around Tehran province (Alvary, 1997) and Caspian Sea shore (Mousavi, 1995) and it is reported for the first time for Taleghan River. Cryptochironomus has been reported from Marbor River (Karami et al., 2014), Golpayegan River (Ebrahimnezhad and Allahbakhshi, 2013). Zayandehrood River (Ebrahimnezhad and Fakhri, 2005) in Isfahan province and it is reported for the first time for Taleghan River fauna. Saetheria and Neozavrelia have been previously reported from Marbor River (Karami et al., 2014). Neozavrelia has been recently reported from Talesh Mountains (Aubert et al., 2017). Cryptotendipes has been reported from Golpayegan River (Ebrahimnezhad and Allahbakhshi, 2013). Paratendipes has been reported from Talesh Mountain (Aubert et al., Golpayegan 2017), River (Ebrahimnezhad and Allahbakhshi. 2013), Marbor River (Karami et al., Zayandehrood 2014) and River (Ebrahimnezhad and Fakhri, 2005). The four remained genera reported in table 2 from Chironominae have been reported once for the Iranian fauna which have been reported from other parts of the world also; Robackia has been reported as a European genus from Netherlands (Balzer, 1997) which then its different species were reported from Turkey (Ozkan, 2002). Stenochironomus has been reported from Brazil (Parise and de Pinho, 2016), China (Qi et al., 2015) and Russia (Zarina, 2001). Omisus has

been reported in many species from many rivers and pools of Europe and North America (Mousavi, 2002) and Finland (Passivirta, 2012). *Constempellina* has been reported from Russia (Zorina, 2013), the United States (Rufer and Ferrington, 2007) and Finland (Grimas and Wiederholm, 1979)

According to the geographical features of the sampling stations through Taleghan River, especially the height, it could be assumed that there would be a great difference between some stations such as Barikan as the highest point and Esfaran as the lowest (Table 1). As it shown in Table 2, these two stations on the two limits of height had all the Chironomid subfamilies, so it could be concluded that not only height above the sea level could not be an effective factor on the abundance and distribution of Chironomidae, but also there would other factors such as rural pollution, plant growth along the river, mean temperature of water, etc. As the result showed some subfamilies such Orthocladiinae as and Chironomonae have distributed in all the sampling points, which would be considered more resistant than other subfamilies to abiotic factors of the environment. As the result of the effect of the month on the genera frequency showed, Orthocladiinae genera were affected by the month, which would be the result of the environment temperature and water pollution. The area in May is cooler and less polluted where would be more suitable for Orthocladiinae, on the other hand, high temperature in August would lead

Orthocladiinae to growth and adult removing from the water. In addition, egg-laying period in August would limit the sampling procedure (Garbary et al., 2009). In a similar study, seasonal changes of Chironomid communities at three subfamilies level, Orthocladiinae Chironominae. and Tanypodinae were studied and it was shown that Chironominae community increased by decreasing temperature through August to October, on the other hand, Orthocladiinae were more abundant in April-July which environmental temperature increased monthly which admitted the effect of temperature on Chironomidae populations (Hirabayashi et al., 2004). Arslan (2010)et al. reported Chironomus. Cryptochironomus and Microchironmus as the positive indices. environmental which are tolerant to pollutants.

The result of the abundance of the collected genera through different stations (Table 3) showed different effect of the sampling points on the collected genera. As it has seen, sampling points had no significant effect on the genera of Diamesinae, Tanypodinae. Orthocladiinae and Although, Chironominae had similar distribution among different stations except for *Chironomus* spp. which was significantly different in number and distribution.

According to the obtained results, more investigation about Chironomidae diversity in other branches of Taleghan River and other aquatic ecosystems of Iran is suggested. Further study to identify the species would be so helpful for deep knowledge of Iranian Chironomidae.

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