Iranian Journal of Fisheries Sciences 20(1) 109-128 2021
DOI: 10.22092/ijfs.2021.351016.0

Research Article

New report for *Pristinella jenkinae* Stephenson, 1931 (Annelida: Oligochaeta: Naididae) geographical distribution from Southern Caspian Sea basin, Mazandaran province - Iran

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Received: May 2020 Accepted: November 2020

Abstract

Pristinella jenkinae (Stephenson, 1931) is a freshwater cosmopolitan oligochaete. This species was found during limnological investigation in two rivers alongside Iranian coasts and has not been previously reported from Iran's freshwater fauna and Southern Caspian Sea basin. The specimens of *P.* jenkinae were collected bimonthly from Cheshmehkileh and Sardabrood rivers and estuaries (river mouth) using a Van Veen grab (0.03 m^2) and Surber (0.1 m^2, 0.2 mm-mesh size) samplers at three stations in each estuary (S1: riverine, S2: estuary and S3: marine ecosystem) with three replicates from November 2014 through September 2015. Results of temporal distribution showed that the highest and lowest density and biomass of this species were in January (102.3±68.3 ind m^{-2} and 0.075±0.034g m^{-2}) and in September (24.4±12.3 ind m^{-2} and 0.020±0.005g m^{-2}), respectively which were significantly different (N=57, t=0.99, p<0.05). Spatial distribution of *P. jenkinae* among sampling stations (S1, S2 and S3) showed significant differences (N=57, t=0.99, p<0.05). Freshwater stations (S1) within the river showed higher density and biomass (112.1±64.8 ind m^{-2} and 0.082±0.035g m^{-2}) than semi-brackish stations (S2) within the estuary (18.8±10.3 ind m^{-2} and 0.013±0.005g m^{-2}). Semi-brackish stations (S2) showed higher density and biomass than brackish stations (S3) within the sea (0±0 ind m^{-2} and 0±0g m^{-2}). Density and biomass of this species in Cheshmehkileh River and estuary was more than Sardabrood. A significant correlation (N=57, r=Pearson, p<0.05) between density and biomass of *P. jenkinae* with environmental variables was found.

Keywords: *Pristinella jenkinae*, Distribution, Riverine, Estuary, Caspian Sea

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[DOR: 20.1001.1.15622916.2021.20.1.18.7]

Introduction

Oligochaete fauna form a significant part of zoobenthos in freshwater and inland water bodies and are important food sources for benthophagous fish (Baturina, 2007). Aquatic oligochaetes are members of a main group of macroinvertebrates and include about 1,100 species of 13 families with worldwide distribution (Martin *et al.*, 2008). These species commonly inhabit within sediments of rivers, streams, lakes, marshes, ponds, springs and ground-waters (Alves and Lucca, 2000; Wetzel *et al.*, 2000; Collado and Schmelz, 2001; Montanholi-Martins and Takeda, 2001) showing that these species have been adapted to a wide variety of habitats and environments, such as freshwater, brackish or seawater. In addition, certain oligochaete species are abundant in organically polluted waters (Jabłońska and Pešić, 2006) and they have been used to monitor water pollution in rivers and streams (Lin and Yo, 2008). Oligochaetes are good indicators of environmental variation because of their easy sample collection and taxa identification, relatively long life cycle, limited migration ability, and different sensitivity to different environmental conditions (Liu *et al.*, 2004). Most species of Naidid worms are cosmopolitan, occurring throughout the world (Wetzel *et al.*, 2000) and due to their great ability to swim, they may have eyes, and are capable of exploring benthic habitats (Erséus and Gustarsson, 2002) such as aquatic macrophytes (Alves and Gorni, 2007), mosses and liverworts (Gorni and Alves, 2007), filamentous algae (Armendariz, 2000), sponges (Corbi *et al.*, 2005), odonate larvae (Corbi *et al.*, 2004) and gastropod mollusks (Gorni and Alves, 2006). Naididae is a large group of freshwater oligochaete (Milbrink 1987, Chapman, 2001, Smutná *et al.*, 2008), while 238 species are known worldwide (Martin *et al.*, 2008) of which approximately 36 species belong to the genus *Pristina* (Timm and Erséus, 2021). Cheshmehkileh of Tonekabon and Sardabrood of Chalus Rivers are the most important, mountainous and permanent rivers running from high elevation to the sea (southern waters of Caspian Sea). These rivers are important for reproductive migration of two indigenous valuable fisheries species *Salmo caspius* (Caspian trout) and *Rutilus frisii* (Caspian Kutum) as well as other migratory fish (Khara, 2016). For these reasons, estuary of these rivers is regarded as a 'Protected Area' and is conserved by the Department of Environment of Iran (DOE, 1996).

P. jenkinae is a cosmopolitan species, occurring throughout the world, such as Europe, South and North America, Africa, Asia, New Zealand and Australia (Pinder and Brinkhurst, 1994). Among studies on aquatic macroinvertebrate fauna of Iran, oligochaetes have been identified at family level, just a few studies identified at species level (Stephenson,

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1920; Egglishaw, 1980; Pourang, 1996; Aliyev and Ahmadi, 2010; Ahmadi *et al.*, 2011; 2012; Ardalan *et al.*, 2011; Basim *et al.*, 2012; Jabłońska and Pešić, 2014; Nazarhaghighi *et al.*, 2014; Tavol Koteri *et al.*, 2018, 2019). Based on these studies, currently 25 species of aquatic oligochaetes have been reported from inland waters of Iran of which 13 species belong to family Naididae and no species from the genus *Pristina*.

The aim of this study was to report *Pristinella jenkinae* from Iran for the first time together with its distribution pattern and density and its correlations with some abiotic factors of their environments.

Materials and methods

Study area

Cheshmehkileh is a mountainous and permanent river in north of Iran (Mazandaran Province). The river source is in Alamot region (northern central Alborz mountains) with total length of approximately 80 km, average annual discharge of 55 million m^3, average slop of 6.5% and basin area of 1200 km^2, entering the Caspian Sea in Tonekabon city. The Sardabrood is another mountainous and permanent river in north of Iran (Mazandaran Province), originating in the Kelardasht region (north central Alborz mountains) with total length of approximately 67 km, average annual discharge of 100 million m^3, average slop of 6.4% and basin area of 450 km^2, discharging into the Caspian Sea in Chalus city (Afshin, 1994).

Sampling

This study was carried out between November 2014 and September 2015 and random sampling was carried out at six stations with three replicates for each bimonthly sampling along each river (Fig. 1 and Table 1). Sampling was done using 0.03 m^2 Van Veen grab for soft sediments at the estuary area and for sampling at inner parts of the river with pebbles a 0.1 m^2 and 0.2 mm-mesh size Surber sampler was applied.

In total, 216 sediment samples (72 biotic and 144 abiotic) were collected. Samples were fixed in situ using a 5% formalin solution. In the laboratory, sediments were sieved through mesh sizes of 1, 0.5 and 0.25 mm and specimens were preserved in 70% ethanol and then sorted and counted under a stereomicroscope (Nikon SMZ800, Japan) and eventually the wet weight of worms was measured using a digital balance (0.0001 g, Mettler Toledo, AB204-N). For identification at species level, worm specimens were mounted on glass slides in Amman's lactophenol clearing agent (Smith, 2001) and covered by a coverslip and left for several hours to a day or two, and then for observation of setae and other details, a microscope was used (Nikon E200 and Nikon DIGTAL SIGHT DS Camera, Japan). The main identification keys used were: Brinkhurst (1971a and b, Brinkhurst and Wetzel (1984), Brinkhurst (1986), Pinder and Brinkhurst (1994), Smith (2001), Arslan and Sahin (2003), Krieger and Stearns (2010), and Pinder (2010).

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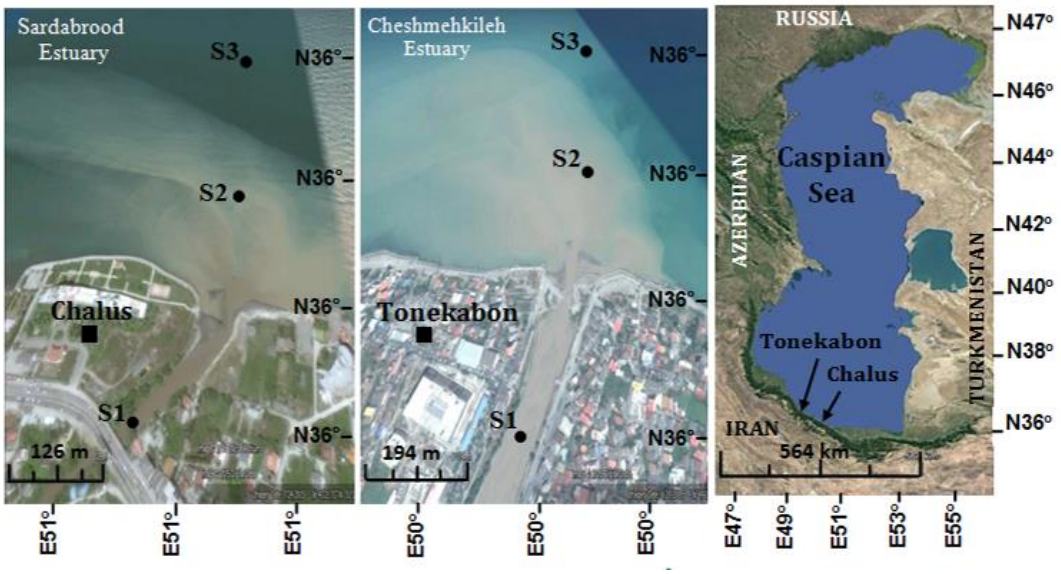


Figure 1: Study areas and sampling stations in Cheshmehkileh and Sardabrood estuaries, Caspian Sea basin (2014-15).

Table 1: Characteristics of sampling stations, Caspian Sea basin (2014-15).

Station	Latitude	Longitude	Sampling location	Water type	Substratum nature	Average depth (m)
S1	N 36˚ 49' 6.3"	E 50˚ 52' 52.3"	Cheshmehkileh River	Freshwater	Gravel, Sand, Silt, Clay, Vegetation	0.45
S2	N 36˚ 49' 20.0"	E 50˚ 53' 9.3"	Cheshmehkileh estuary	Semi-brackish	Gravel, Sand, Silt	0.86
S3	N 36˚ 49' 35.9"	E 50˚ 53' 24.6"	Marine	Brackish	Sand, Silt, Clay	7.08
S1	N 36˚ 41' 11.9"	E 51˚ 23' 55.4"	Sardabrood River	Freshwater	Gravel, Sand, Silt, Clay, Vegetation	0.5
S2	N 36˚ 41' 22.2"	E 51˚ 24' 8.7"	Sardabrood estuary	Semi-brackish	Gravel, Sand, Silt	0.88
S3	N 36˚ 41' 39.9"	E 51˚ 24' 26.3"	Marine	Brackish	Sand, Silt, Clay	8.08

In this study environmental variables, such as temperature and salinity in water and total organic matter (TOM) and grain size in sediments were measured. Alongside sampling biota, three sediment replicates were taken at each station for grain size and TOM analysis.

Organic matter content was measured by the weight lost during ashing; the sediment samples were oven-dried at 80°^{C} for 24h, weighed to the nearest 1 mg, ashed at 550°^{C} for 2h and reweighted, and the weight was expressed as the percentage of the total weight (Wildsmith *et al.*, 2011).

Grain size was analyzed by dry mechanical separation through a column of standard sieves of different mesh sizes, corresponding to classes described by Wentworth (1922), gravel (>2 mm), sand (2-0.063 mm), silt

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(0.063-0.004 mm) and clay (0.004-0.0002 mm). Particle sizes smaller than 0.063 mm (silt and clay) were measured by hydrometery (Densimetry) method. The relative content of different grain-size fractions was expressed as a percentage of total sample weight.

During sampling period, water temperature and salinity were measured in situ by multimeter portable HACH - HQ40d model.

The statistical analysis was performed by SPSS 22. Prior to the analysis, data were tested for normality using Kolmogorov-Smirnov test. If the data distribution were normal, three-way analysis of variance (ANOVA) test was used to determine the relationship between the environmental and biological variables. Duncan's test (p<0.05) was then used to assess the significant differences among the stations and months of sampling. Also relationships between density and biomass of *P. jenkinae* and environmental variables were estimated using a Pearson's correlation coefficient (p<0.05).

Results

The systematic account and description for the described species is as follows:

Kingdom: Animalia
Phylum: Annelida Lamarck 1802
Class: Clitellata
Subclass: Oligochaeta Grube 1850
Order: Haplotaxida Brinkhurst 1971
Family: Naididae Ehrenberg 1828
Subfamily: Pristininae Lastočkin 1921
Genus: *Pristinella* Brinkrust 1985
Species: *Pristinella jenkinae* (Stephenson 1931)

General description

Prostomium is without proboscis and most specimens have no eyes. Worms are olive gray or dark yellow with brown spots (Fig. 2A). Worms's body length was 1.6 to 2.6 mm and 0.2 to 0.5 mm in diameter with 14-33 segments. Dorsal chaetae beginning in segment II (Fig. 2B), hairs 1-2 per dorsal bundle (most worms have 1 hair per bundle), 120-250 µm long, needles 1-2 per dorsal bundle (most worms have 1 needle per bundle, Fig. 2E, F). The needles with two long parallel or slightly divergent teeth, distal (upper) tooth longer than proximal (lower) tooth, 38-85 µm long, with a distal nodulus (Fig. 2G). Ventral chaetae 4-5 per bundle in anterior segments, bifid crotchets, all teeth approximately equal in length, 30-75 µm long, with a median or slightly distal nodulus (Fig. 2D), Posterior Ventral chaetae 2-3 per bundle (Fig. 2C). No sexually active individuals were detected during study period.

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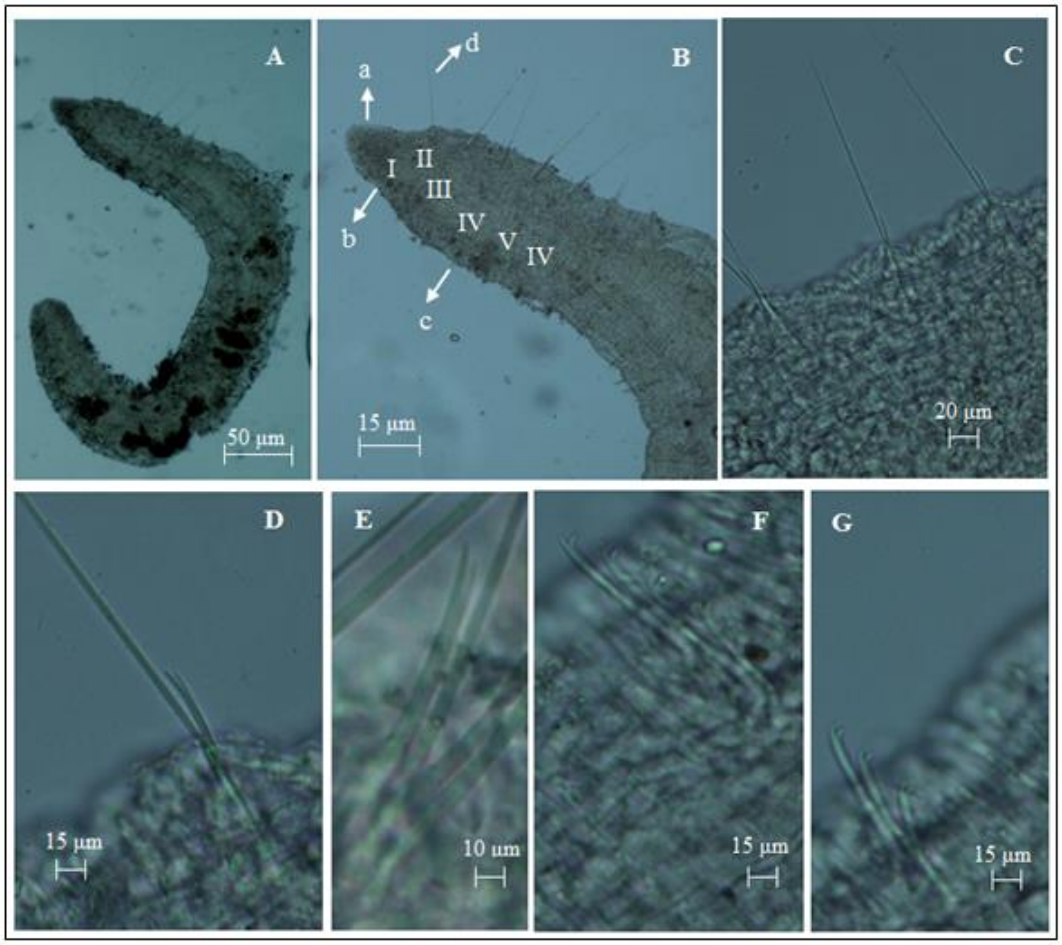


Figure 2: ***Pristinella jenkinae*****, Caspian Sea basin (2014-15). A, General body form; B, Anterior end of the body (a: Prostomium; b: Mouth; c: Ventral bundle; d: Dorsal hair chaetae beginning in segment II); C and D, Dorsal chaeta bundle (1 hair and 1 needle); E, Dorsal chaeta bundle (2 hairs and 2 needles); F, Ventral chaeta bundle in anterior part of body; G, Ventral chaeta bundle in posterior part of body.**

Cocoon is a cover or bag containing worm eggs and embryos that is a result of sexual reproduction in the worms (Fig. 3). The cocoons usually contain several eggs and embryos, in most cases, only one or two embryos survive and come out of the cocoon as young worms. Cocoons are considered as a protective layer for eggs and embryos (Smith, 2001).

In total, 303 individuals of *P. jenkinae* were examined. During the present study, this species occurred in stations 1 (river) and 2 (estuary) but was absent in station 3 (marine) in both sampling areas. Density and biomass of this species among sampling months and stations were significantly different (N=57, t=0.99, p<0.05), as the highest average density and biomass were observed in Cheshmehkileh (station 1) in November (231.6±94.3 ind m^{-2} and 0.170±0.05g m^{-2}), respectively.

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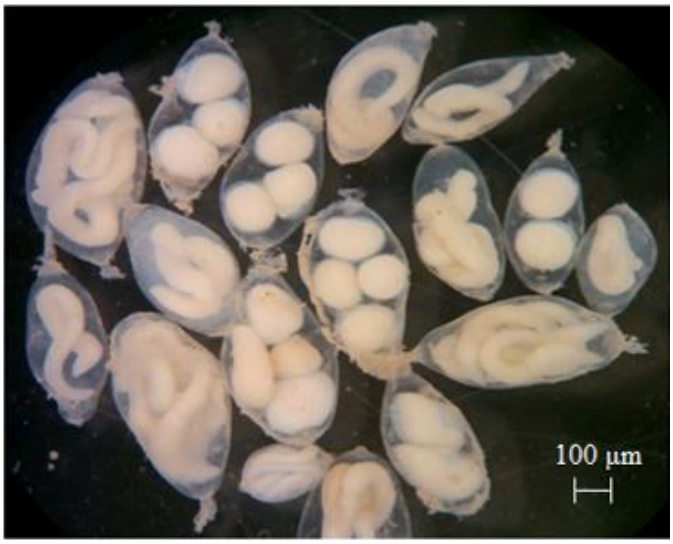


Figure 3: Oligochaeta cocoons with eggs and embryos, Caspian Sea basin (2014-15).

While the lowest of those values were observed in all sampling months at stations 3 in both sampling areas (0±0 ind m^{-2} and 0±0 g m^{-2}, Table 2).

Table 2: Density (ind m^{-2}) and biomass (g m^{-2}) of *Pristinella jenkinae* (average±SE), Caspian Sea basin (2014-15).

		Cheshmehkileh			Sardabrood		
		S1	**S2**	**S3**	**S1**	**S2**	**S3**
November	Density	a 231.6±94.3 A	a 17.6±5.3 C	-	b 125.4±32.1 B	a 23.5±5.8 C	-
	Biomass	a 0.170±0.05 A	a 0.013±0.003 C	-	ab 0.092±0.023 B	a 0.021±0.002 C	-
January	Density	b 184.8±69.3 A	a 23.5±8.8 B	-	a 170.3±70.1 A	a 30.7±9.4 B	-
	Biomass	b 0.140±0.034 A	a 0.018±0.005 B	-	a 0.115±0.034 A	a 0.027±0.004 B	-
March	Density	d 100.1±33.2 A	-	-	c 87.3±20.3 A	a 16.6±5.5 B	-
	Biomass	c 0.081±0.017 A	-	-	c 0.066±0.011 A	a 0.011±0.002 B	-
May	Density	c 154±44.9 A	a 12.8±4.4 C	-	b 118.6±32.5 B	-	-
	Biomass	c 0.101±0.034 A	a 0.008±0.001 B	-	bc 0.087±0.017 A	-	-
July	Density	d 81±26.2 A	-	-	d 27.1±6.6 B	-	-
	Biomass	d0.055±0.011A	-	-	d 0.025±0.003 B	-	-
September	Density	e 31.7±10.6 A	-	-	d 33.6±8.7 A	a 8±2.02 A	-
	Biomass	e 0.029±0.004A	-	-	d 0.027±0.003 A	a 0.005±0.001 B	-
Annual average	Density	130.53±32.8 A	17.96±6.2 C	-	93.71±20.1 B	19.7±4.95 C	-
	Biomass	0.096±0.017 A	0.012±0.003 B	-	0.068±0.011 A	0.015±0.002 B	-

Different letters indicate significant differences among averages (p<0.05). Capital letters indicate variation among stations (horizontal), small letters indicate variation among months (vertical).

Spatial distribution of *P. jenkinae* among sampling stations (S1, S2 and S3) showed significant differences (N=57, t=0.99, p<0.05). In both sampling areas, freshwater stations (S1) within the river (112.1±64.8 ind m^{-2} and 0.082±0.035g m^{-2}) showed higher average density and biomass than the semi-brackish station (S2) within the estuary (18.8±10.3 ind m^{-2} and

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0.013±0.005g m^{-2}). Semi-brackish station (S2) showed higher average density and biomass than the brackish stations (S3) within the sea (0±0 ind m^{-2} and 0±0g m^{-2}, Fig. 4). In all sampling months stations S1 had higher density and biomass than stations S2 and S3. No significant difference was observed between the annual average density and biomass of this species for the two study areas (Table 2), density and biomass of this species in Cheshmehkileh area (74.24±19.5 ind m^{-2} and 0.054±0.01g m^{-2}) was higher than that in Sardabrood (56.7±12.5 ind m^{-2} and 0.041±0.006g m^{-2}).

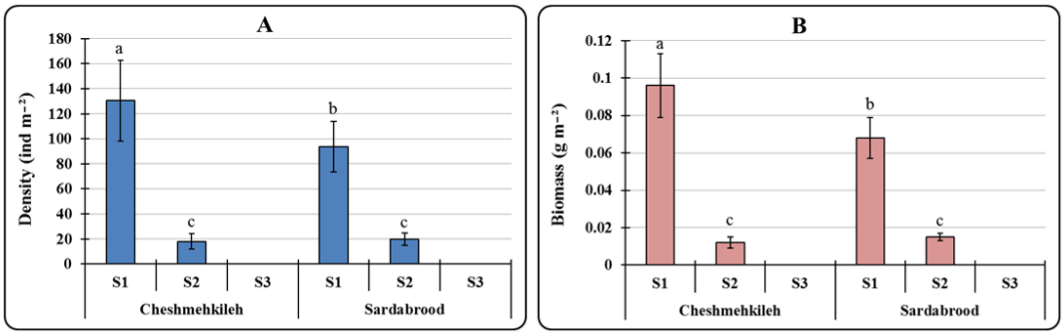


Figure 4: Annual average (±SE) Density (A) and biomass (B) of *Pristinella jenkinae* in sampling stations, Caspian Sea basin (2014-15). Different letters indicate significant differences among averages (p<0.05).

Temporal distribution of this worm was significantly different among sampling months (N=57, t=0.99, p<0.05), as the highest average density and biomass of this worm were observed in January as 102.3±68.3 ind m^{-2} and 0.075±0.034g m^{-2}, respectively. While, the lowest of those values were in September as 24.4±12.3 ind m^{-2} and 0.020±0.005g m^{-2} (Fig. 5). The highest average density and biomass of this species in Cheshmehkileh area were in November and January in Sardabrood area and the lowest values in both areas were observed in September (Table 2). A significant correlation (N=57, r=Pearson, p<0.05) between density and biomass of *P. jenkinae* with environmental variables was found (Table 3). A positive correlation was found between this species and gravel, silt and clay, while its correlations with temperature, salinity, TOM and sand were negative. The lowest and highest average water temperature were recorded as 10.2±0.1ºC and 30±0.1ºC in Cheshmehkileh (station 1) in March and Sardabrood (station 3) in July, respectively (Fig. 6).

The lowest average salinity (0±0 ppt) was observed in all sampling months in stations 1 (fresh water) in both rivers and the highest average value (11.61±0.01 ppt) was recorded in Sardabrood in station 3 (brackish) in July (Fig. 7). According to the annual average, salinity level of Sardabrood area (5.77±4.71 ppt) was slightly higher than that in the Cheshmehkileh area (5.64±4.39 ppt).

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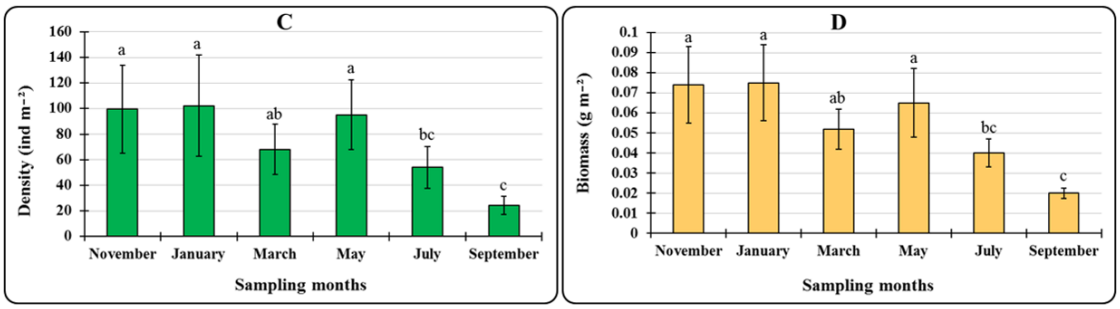


Figure 5: Density (C) and biomass (D) average (±SE) of *Pristinella jenkinae* among sampling months, Caspian Sea basin (2014-15). Different letters indicate significant differences among averages (p<0.05).

Table 3: Pearson's correlation coefficient between density and biomass of *Pristinella jenkinae* and environmental variables, Caspian Sea basin (2014-15)

	Temperature	Salinity	TOM	Gravel	Sand	Silt	Clay
Density	- 0.455 *	- 0.615 *	- 0.078	0.410 *	- 0.522 *	0.303	0.381
Biomass	- 0.413 *	- 0.660 *	- 0.094	0.400 *	- 0.509 *	0.299	0.333

* Correlation is significant at 0.05 level.

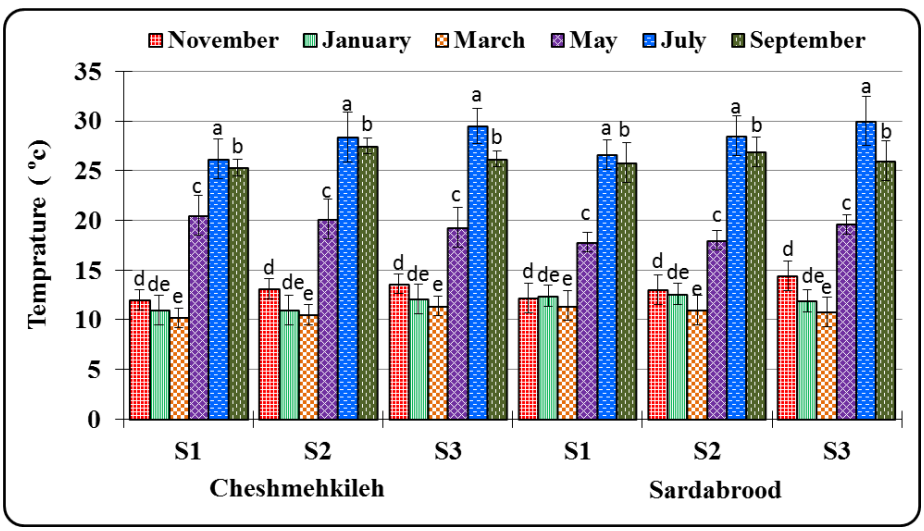


Figure 6: Annual average water temperature in sampling stations, Caspian Sea basin (2014-15). Different letters indicate significant differences among averages (p<0.05).

Also the highest average percentage of total organic matter (TOM, 3.97±0.005 %) was measured in May in Sardabrood (station 3) and the lowest average value (1±0.008 %) was in January in Cheshmehkileh (station 2, Fig. 8). According to the annual average, TOM percentage of Sardabrood area (2.20±0.87 %) was higher than that in Cheshmehkileh area (1.94±0.89 %).

According to the results of sediment grain size in both estuaries, two types of coarse grain sediment (gravel) and fine grain (sand, silt and clay) were observed (Table 4). Sediment texture in river (S1) and river mouth (S2) sations were gravely sand and in marine sations (S3) was silty sand.

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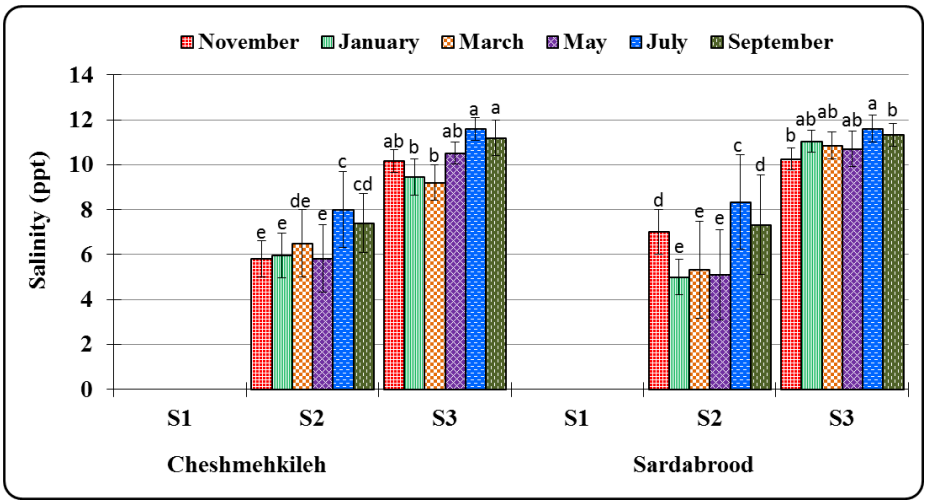


Figure 7: Annual average water salinity in sampling stations, Caspian Sea basin (2014-15). Different letters indicate significant differences among averages (p<0.05).

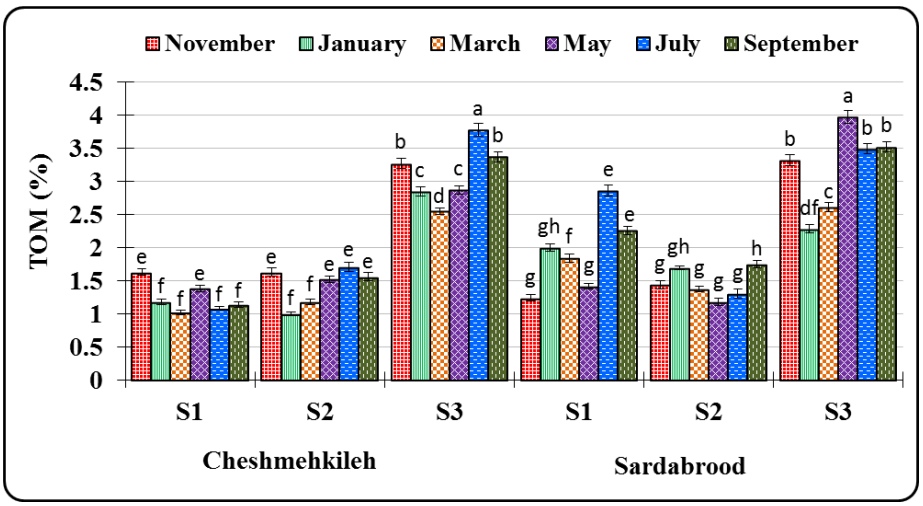


Figure 8: Annual average of TOM percentage in sampling stations, Caspian Sea basin (2014-15). Different letters indicate significant differences among averages (p<0.05).

Table 4: Average sediment grain size among sampling months and stations in the studied estuaries, Caspian Sea basin (2014-15).

Station	Month	Gravel			Sand			Silt			Clay		
		S1	S2	S3	S1	S2	S3	S1	S2	S3	S1	S2	S3
Cheshmehkileh	November	36.5	30.8	0	46.6	63.4	84	11.4	4	9.7	5.2	1.6	6.2
	January	35.5	35.4	0	53.5	57.1	84	7.2	4.6	10.7	3.7	2.8	5.2
	March	33.8	28.7	0	47.7	60.6	75.7	11.1	7.3	16.4	7.4	3.2	7.6
	May	21.7	18.5	0	60.4	75	77.3	11.6	4.2	14.2	6.3	2.2	8.4
	July	0.03	0.06	0	73.2	86.2	42.8	18.4	9	38.4	8.3	4.7	18.7
	September	13.8	9.8	0	60.2	75	79.7	16.5	11.3	12.8	9.4	3.9	7.4
Sardabrood	November	7.1	32.4	0	66.4	61.8	79.7	17.3	3.4	14.2	9.2	1.8	5.8
	January	21.8	14.8	0	56.2	77.5	85.2	15	5	9.3	6.8	2.7	5.3
	March	17.3	28.9	0	53	59.5	73.2	18.1	7.6	19.1	11.5	3.8	7.6
	May	14.6	23.1	0	70.2	69.2	76.6	10.1	5.2	14	5	2.4	9.5
	July	4	21.6	0	63.1	73.3	86.4	22.6	3.7	9.3	2.1	1.3	4.2
	September	6.4	18.8	0	59.6	72.4	79	21.7	5.7	13.6	12.1	2.9	7.3

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Discussion

During one year sampling in Cheshmehkileh and Sardabrood estuary, 303 individuals of *Pristinella jenkinae* were discovered. This paper updated a short checklist of Iranian aquatic oligochaetes to 26 species (Naididae to 13 species plus 1 species for *Pristinella* genus). In Table 5, the identified species of Naididae from Iran until now are listed including *P. jenkinae* which is new record for Iran. In this study, *P. jenkinae* was permanent inhabitant during all sampling periods.

Approximately 14 species belong to the genus *Pristinella* are known worldwide, so far. *P. jenkinae* is clearly separated from other species of *Pristinella* by the dorsal needle setae characteristic (type, number, size and form). Dorsal bifid needles in *P. jenkinae* have long parallel teeth while in other species (*P. amphibiota*, *P. rosea* and *P. sima*) have short and not parallel teeth (Arslan and Sahin, 2003 and 2004).

Table 5: List of identified Naididae species from Iran.

	Species	Reference
1	*Aulophorus furcatus* Oken 1815	Ahmadi *et al*. 2012
2	*Chaetogaster diastrophus* Gruithuisen 1828	Stephenson, 1920
3	*Chaetogaster limnaei* Baer 1827	Stephenson, 1920, Jabłońska and Pešić, 2014
4	*Dero dorsalis* Ferroniere 1899	Jabłońska and Pešić, 2014
5	*Dero digitata* Müller, 1773	Nazarhaghighi *et al*., 2014
6	*Nais communis* Piguet 1906	Stephenson, 1920
7	*Nais pardalis* Piguet 1906	Nazarhaghighi *et al*., 2014
8	*Ophidonais serpentina* Müller 1774	Ardalan *et al*., 2011, Nazarhaghighi *et al*., 2014
9	*Pristina breviseta* Bourne 1891	Jabłońska and Pešić 2014
10	*Stylaria lacustris* Linnaeus, 1767	Stephenson, 1920, Aliyev and Ahmadi, 2010, Ahmadi *et al*., 2012, Nazarhaghighi *et al*., 2014
11	*Slavina appendiculata* dUdekem 1855	Nazarhaghighi *et al*., 2014
12	*Nais variabilis* Piguet 1906	Tavol Koteri *et al*., 2018
13	*Nais elinguis* Müller 1773	Tavol Koteri *et al.*, 2019
14	*Pristinella jenkinae* Stephenson 1931	Current study

According to the results of this study, *P. jenkinae* strongly peaked at initial cold period (from November to May) and dropped at the time of summer solstice, whereas density and biomass of this species show a significant negative correlation (N=57, r= Pearson, $p<0.05$) with water temperature (Table 3); also the peak (higher mean numbers) appeared to be linked to the rather instable river flow regime (the first spate sampled) and the time of

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strong discharge fluctuations. This temporal pattern was similar to observation on the Kodungaiyur and Pandinellur Swamps in cold period from October to February (Naveed, 2012).

However higher density of *P. jenkinae* from November to March were initially sustained by asexual reproduction (accelerated paratomy), similar to other Naididae species (Loden, 1981). The shifts of asexual to sexual reproduction require cocoon production that allows survival of the eggs and embryos during adverse environmental conditions and remain throughout the year until December (Learner *et al.*, 1978; Loden, 1981). Coincidence of active mature individuals of *P. jenkinae* in winter and spring population release might also be explained as dispersal strategy. Therefore *P. jenkinae* might be classified as species intolerant of "late spring-summer-early autumn" conditions in Cheshmehkileh and Sardabrood rivers. According to Learner *et al.* (1978), Paoletti and Sambugar (1984), Juget and Lafont (1994) and Martinez-Ansemil and Collado (1996) spring is a key period in the life cycle of naidid in running waters. In the present study, dispersal strategy was observed among sampling months from November to May.

In the present study, *P. jenkinae* was observed in both freshwater river (salinity 0 ppt) and semi-brackish (salinity 5-8 ppt) ecosystems indicating that this species is to some extent euryhaline. But it was not observed in marine brackish ecosystem (salinity 9-11.5 ppt). The limiting factor for distribution of this species in marine environment may be salinity intolerance of more than 8 ppt or unwillingness to live in fine grained silty sand substrates.

Most naidid species have clearly adapted to a wide range of environmental conditions (Brinkhurst and Jamieson, 1971). The naidid oligochaete *P. jenkinae* was only observed in stations 1 (freshwater river) and 2 (semi-brackish water), higher in freshwater than semi-brackish water ecosystems ($N=57$, $t=0.99$, $p<0.05$), in both Cheshmehkileh and Sardabrood rivers. While commonly found around the world in freshwater river, tidal freshwater and sometimes in brackish environments (Arslan and Sahin, 2003; Arslan *et al.*, 2014; Cui *et al.*, 2015). However, *P. jenkinae* has been found in river mouth-brackish water habitat (Yildiz *et al.*, 2007). *P. jenkinae* is a cosmopolitan species, occurring throughout the world, such as Europe, South and North America, Africa, Asia, New Zealand and Australia (Pinder and Brinkhurst, 1994).

Martinovic-Vitanovic *et al.* (2007) concluded that the main factors determining Naididae distribution and abundance are nature of substrate and presence and type of vegetation, as periphyton offering shelter for their populations. Pascar-Gluzman and Dimentman (1984) reported that *P. jenkinae* as a euryoeic and eurythermal species and it was collected from a wide range of current velocities and within a wide temperature range. Arslan

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and Sahin (2003) also reported that *P. jenkinae* was collected from muddy, sandy, gravel substratum and among thick vegetation.

Most naidid species are grazers, positively correlated with periphyton (Schenkova and Helesic, 2006). Therefore, abundance of the Naididae depends on the amount of periphyton (Baturina *et al.*, 2014). Naidids occur mainly among the filamentous algae and aquatic vegetation (Naidu 2005), and the absence of naidids at Kodungaiyur swamp may therefore be due to poor vegetation (Naveed, 2012). Maciorowski *et al.* (1977) and Harman *et al.* (1979) reported that *P. jenkinae* is tolerant to severe pollution and have been found in a stream degraded by industrial effluents. Likewise, other studies found that this species was abundant in stony bed, muddy substratum, organically enriched streams and lakes in Turkey (Arslan and Sahin, 2006; Camur-Elipek *et al.*, 2006; Akbulut *et al.*, 2009). Arslan *et al.* (2014) reported high naidid abundance in Catoren Dam Lake, maybe partly due to their rich littoral vegetation.

Substrates in both rivers at Freshwater sampling stations (S1) were covered with vegetation in littoral and some central zone, but in brackish stations substrate (S2 and S3) vegetation cover was absent. Density and biomass of *P. jenkinae* among sampling stations was significantly deferent (N=57, t=0.99, $p<0.05$), as density and biomass of this species in freshwater stations (S1) was higher than brackish stations (S2 and S3, Table 2 and Fig. 4). According to the results of above mentioned studies about dependency of naidid species to vegetation habitats (Naidu, 2005; Naveed, 2012; Arslan *et al.*, 2014), the main reason of density and biomass increase of this species in freshwater sampling stations was presence of vegetation habitats and its decrease in brackish water stations was due to absence of this habitat. Possibly, for this is reason, *P. jenkinae* showed significant negative correlation (N=57, r=Pearson, $p<0.05$) with salinity (Table 3).

Naidid are eurytolerant oligochaetes and are numerically dominant and common oligochaetes in rivers and small estuaries. Beyond being the most abundant in Cheshmehkileh and Sardabrood rivers, *P. jenkinae* was also the most responsive organism to the environmental differences between the two rivers and throughout the year. The lower abundance of *P. jenkinae* in lower reaches of both estuaries was indicative of reduced environmental stressors in the lower reaches. Moreover, *P. jenkinae* as a swimming naidid occurred in superficial gravely sand sediments of Cheshmehkileh and Sardabrood estuaries. As was expected, the silt-clay composition of the sediments of Cheshmehkileh and Sardabrood estuaries were noticeably changed during the study period especially in July and May and silt-clay percentages of Cheshmehkileh were

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approximately twice as the amount found in Sardabrood. The trend of more silts and clays in the lower reaches was also noted by Lerberg (1997) and is thought to be a function of particle settling rate. Absence of detectable patterns in temporal and spatial measures of TOM of Cheshmehkileh and Sardabrood estuaries, similar to Sanger (1998), likely indicated a relatively variable input of refractory organic material from the uplands to the sediments of the estuaries.

While there was no strong estuary-to-estuary difference in water composition, there were significant differences in the sediment silt-clay and TOM range, and flushing rates between Cheshmehkileh and Sardabrood estuaries. These differences were a function of land cover and orientation between the two estuaries. Compared to Sardabrood estuary, Cheshmehkileh had a greater flushing rate, greater salinity fluctuation over annual cycle, which was correlated to a more stressful environment in Cheshmehkileh estuary with less particulate organic matter (Fig. 8) as a food source for the benthos. These differences are most clearly reflected in the greater abundance of *P. jenkinae* in Cheshmehkileh estuary compared to Sardabrood estuary. Differential predation pressure could also be invoked to explain the differences in the abundance of *P. jenkinae* between the estuaries, but the abundance of fishes was greater in Cheshmehkileh estuary than in Sardabrood during autumn and winter (unpublished data). The greater macrobenthic abundance in Cheshmehkileh estuary in spite of the marked difference in predator abundance, strongly suggests that food supply, periphyton, and sediment quality had a greater effect on *P. jenkinae* in these rivers than predation pressure.

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