**Oceanological and Hydrobiological Studies** 

International Journal of Oceanography and Hydrobiology

Volume 51, No. 2, June 2022 pages (212-223)

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ISSN 1730-413X eISSN 1897-3191

Evaluation of ecological risk analysis for benthic macroinvertebrates in paddy fields in the Meric-Ergene River Basin (Turkish Thrace)

by

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DOI: https://doi.org/10.26881/oahs-2022.2.09 Category: Original research paper Received: February 4, 2022 Accepted: May 10, 2022

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#### Abstract

Benthic macroinvertebrates are very important components of aquatic environments, and monitoring their population dynamics helps us understand the effects of environmental factors on ecosystems. This study aimed to determine the dynamics of benthic macroinvertebrate fauna in paddy fields in the Meric-Ergene River Basin (Turkish Thrace region) by investigating some physicochemical environmental parameters that may affect its distribution. For this purpose, water and sediment samples were collected from paddy fields in the study area during the cultivation season, including spring, summer and autumn of 2016, taking into account the water resources that supply the rice fields (artesian water, the Meric River, the Ergene River and Meric-Ergene mixed water). A total of 47 taxa (on average 8953 individuals per m<sup>2</sup>) were identified at the study sites. Water samples were analyzed to determine water temperature, pH, conductivity, salinity, total dissolved solids, calcium, magnesium, total hardness, nitrite nitrogen, nitrate nitrogen, phosphate, sulfate, dissolved oxygen and pesticides, and sediment samples were analyzed to determine the content of some heavy metals, including Cd, Ni, Cu, and Mn. The biological risk index (mERM-Q) and the potential ecological risk index (RI) were applied to the data and a hypothetical ecological risk analysis was conducted using our data and data available in the literature to assess the ecological risk profile of the ecosystem based on benthic macroinvertebrates. To this end, environmental factors were grouped based on the literature as heavy metals (S1), nutrients (S2), other physicochemical parameters (S3) and pesticides (S4), while organisms were grouped as Oligochaeta, Chironomidae, Insecta and others based on the dynamics of benthic macroinvertebrates to assess pressure factors. As a result, pesticides (S4) were found to exert the strongest ecological pressure on benthic macroinvertebrate fauna in paddy fields in the Meric-Ergene River Basin.

**Key words:** benthic macroinvertebrates, ecological risk index, hypothetical risk analysis, rice fields

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## **1. Introduction**

About 15% of the world's wetlands of various types comprise paddy fields where rice is cultivated (Zalom 1981; Lawler 2001; Katano et al. 2003; Leitao et al. 2007; Wilson et al. 2008; Islam et al. 2012). This type of ecosystem has a dynamic structure like other aquatic ecosystems, and its food chain begins with primary producers (Kim et al. 2011). Benthic macroinvertebrates are one of the essential elements necessary for the ecosystem continuity because they play an important role in the food chain, providing primary food for fish, frogs, birds, and other invertebrates. In addition, they are biological indicators that provide important clues about the ecological structure, biological efficiency and water quality of the system (Kenney et al. 2009; Rizo-Patrón et al. 2013). Like other aquatic ecosystems, paddy fields are also impacted by many environmental factors, such as daylight, wind, air temperature, rainfall, and are also exposed to anthropogenic effects. All of these affect the benthic macroinvertebrate dynamics (Molozzi et al. 2007).

The Meric-Ergene River Basin is located in the Thrace Region in the European part of Turkey and consists of the Meric and Ergene rivers and their tributaries. The basin is one of the 25 defined basins in Turkey. The Meric Delta, which is part of the basin, is listed as a Class A wetland on the list of Wetlands of International Importance, and can support more than 25,000 waterfowl (Tokatlı 2019a; Tokatlı, Islam 2022). Due to the abundance of freshwater resources, the basin features very large agricultural land. Approximately half of Turkey's rice crops are grown in the Meric-Ergene River Basin (Grain Report 2016). There are many industrial establishments in the Ergene River Basin. Therefore, a high pollution load on the resources of the basin is inevitable. Pesticides and fertilizers are commonly used in agriculture in this area and discharges of pollution from industrial activities in Turkish Thrace can reach surface waters. For this reason, assessment of ecological risk factors that may affect the ecosystem sustainability in paddy growing areas is also of great importance (USEPA 1998).

To date, a number of studies have been performed in aquatic environments in the Meric–Ergene River Basin (Çamur-Elipek et al. 2010; Taş et al. 2011; Öterler et al. 2015; Tokatlı 2019a,b; Tokatlı et al. 2020; Varol, Tokatlı 2021; Tokatlı, Islam 2022). In this study, both benthic macroinvertebrate dynamics in paddy fields of the Meric–Ergene River Basin and some physicochemical components of environmental pressure were evaluated. For this purpose, the biological risk index (mERM-Q) and the potential ecological risk index (R<sub>i</sub>) were applied to the obtained data, and a hypothetical ecological risk analysis based on analysis results and previous studies was used to determine the strongest environmental risk factor for benthic macroinvertebrates in the study area.

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## 2. Materials and methods

## 2.1. Study area

The Meriç-Ergene River Basin is located in the Thrace Region in the European part of Turkey. There are 53.4798 ha of paddy fields in the basin, and irrigation water is provided by the Meriç and Ergene rivers, their tributaries and reservoirs built on those rivers, as well as artesian water extracted from the ground (İstanbulluoğlu et al. 2006; TSPO 2016). The Meriç River originates in Bulgaria and merges with the Arda River in Edirne. After joining the Tunca River south of Edirne, it joins the Ergene River and flows into the Saros Gulf (Aegean Sea; Fig. 1).



#### Figure 1

Sampling locations according to irrigation water source (M1, M2 – Meriç River; A1, A2 – Artesian water; E1, E2 – Ergene River; ME1, ME2 – mixed water of the Meriç and Ergene rivers)

## 2.2. Sampling

During the cultivation season (May to September 2016), water and sediment samples were collected seasonally in paddy fields selected according to the source of water used for their irrigation (artesian water – A, Ergene River – E, Meriç River – M, and Meriç–Ergene mixed water – ME) to cover spring (first water phase), summer (aquatic phase) and autumn (semi-aquatic phase) periods (Fig. 1 and Table 1).

Sampling locations			
Sampling location	Location of paddy fields	Irrigation water source	Coordinates
A1	Havsa (Necatiye village)	Artesian water	41°30'20"N; 26°53'28"E
A2*	Tekirdağ (Hayrabolu)	Artesian water	41°13'05"N; 27°06'58"E
M1	Meriç (Küplü village)	Meriç River	41°02'10"N; 26°20'36"E
M2*	Edirne (Kapıkule)	Meriç River	41°42'37"N; 26°22'31"E
E1	Uzunköprü (Çiftlik village)	Ergene River	41°14'27"N; 26°37'02"E
E2*	Uzunköprü (Centrum)	Ergene River	41°15'46"N; 26°41'32"E
ME1	İpsala (Border Gate)	Meriç + Ergene River	40°55'47"N; 26°20'50"E
ME2*	İpsala (Yenikarpuzlu village)	Meriç + Ergene River	40°46'38"N; 26°17'08"E

\*locations sampled for heavy metals

To identify benthic macroinvertebrates, sediment samples were collected twice at the sampling locations using an Ekman grab ( $15 \times 15 \text{ cm}^2$ ). The collected sediment was washed using a series of mesh nets (1.5 mm, 0.7 mm, and 0.3) and the residual material was preserved in 250 cm<sup>3</sup> plastic bottles containing 70% ethanol. Samples of benthic macroinvertebrates were examined in the laboratory under a stereo binocular microscope and they were identified to the lowest possible taxonomic category. The identified benthic macroinvertebrate specimens were grouped as "Oligochaeta", "Chironomidae", "Other Insecta" and "Other Taxa", and the number of individuals per unit area was calculated in terms of both seasonal and location means for each group.

The pH, conductivity, salinity, TDS (total dissolved solids) and temperature of water were measured using a Consort<sup>TM</sup> multi-parameter analyzer C5020 at the same time when benthic material was sampled. Water samples were collected into dark glass bottles and transported to the laboratory for analysis of other parameters. Calcium, magnesium, total hardness, NO<sub>2</sub>-N, NO<sub>3</sub>-N, PO<sub>4</sub><sup>-3</sup>, sulfate and dissolved oxygen were measured using classical titrimetric and spectrophotometric methods (absorbances were read on a Cecil-CE 5502 brand spectrophotometer; Egemen, Sunlu 1996).

For the analysis of pesticides, water samples were transported to the laboratory of Trakya University Technology Research and Development Application and Research Center (TUTAGEM) and analyzed immediately. The QUECHERS (quick, easy, cheap, effective, rugged, safe) method, developed for the determination of multiple pesticide residues, was applied in the extraction of pesticides. Pesticide analyses were performed using Agilent 1260 infinity liquid chromatography, Agilent 6460 Triple Quadrupole MS/MS System (Jet Stream Electrospray ion source) and a total of 181 pesticide types were analyzed. For heavy metal analyses, sediment samples were collected into sterile polyethylene bottles (250 cm<sup>3</sup>) and were transported to the laboratory to analyze cadmium (Cd), copper (Cu), nickel (Ni) and manganese (Mn) concentrations by a Perkin Elmer Analyst 800 Flame Atomic Spectrophotometer.

#### 2.3. Statistical Analysis

The data were evaluated using three statistical methods: Shannon–Wiener Diversity Index was used for statistical determination of species diversity; Bray–Curtis Similarity Index was used to compare the sampling locations and the seasons in terms of both physicochemical properties and benthic macroinvertebrate dynamics; and the Correspondence Analysis was used to support the results (Krebs 1999).

## 2.4. Ecological Risk Analysis

Ecological risks resulting from sediment contamination were determined using the potential ecological risk index (PER<sub>1</sub>) and the biological risk index (BRI) (Hakanson 1980; Long et al. 2005).

The potential ecological risk index was calculated using the following formula (Hakanson 1980):

$$R_{I} = \sum E_{r}^{i}$$
$$E_{r}^{i} = T_{r}^{i}C_{f}^{i}$$
$$C_{f}^{i} = \frac{C_{o}^{i}}{C_{n}^{i}}$$

where " $R_i$ " is the sum of all risk factors calculated separately for heavy metals in sediments; " $E_r^i$ " is the ecological risk factor for each heavy metal; " $T_r^i$ " is a toxic response factor (using reference values from

Table 2); " $C_{f}^{i}$ " is the contamination factor; " $C_{0}^{i}$ " is the concentration of a heavy metal in the sediment from the sampling locations; " $C_{n}^{i}$ " is a reference value for a metal (using reference values from Table 2). The scale used for " $R_{i}$ " is given in Table 2.

Table 2Reference values for the analyzed heavy metals  $(C_n^i)$ ,toxicity coefficient  $(T_r^i)$ , medium effect range (ERM)(Hakanson 1980; Xu et al. 2008)

Flomont	F	mERM-Q		
ciement	C <sup>i</sup> n	T <sup>i</sup> r	ERM	
Cd	0.5	30	9	
Cu	30	5	390	
Ni	50	5	50	
Mn	860	1	-	

The biological risk index was calculated using the following formula (Long et al. 2005):

$$mERM - Q = \frac{\left(\sum_{i=1}^{n} ERM - Q_i\right)}{n}$$
$$ERM - Q_i = \frac{C_i}{ERM_i}$$

where "mERM-Q" is the possible effect coefficient of multiple metal contamination; "C<sub>i</sub>" is the concentration of a heavy metal in the sediment from the sampling locations; "ERM<sub>i</sub>" is the ERM value of a determined heavy metal (Table 3); "n" is the number of selected heavy metals. The scale of "mERM–Q" is given in Table 3.

A hypothetical ecological risk analysis was performed to determine the potential ecological risk profile of the ecosystem. For this purpose, ecological pressure elements were identified based on the results of this study and previous studies performed in the basin (Çamur-Elipek et al. 2010; Taş et al. 2011; Öterler et al. 2015; Tokatlı 2019a,b; Tokatlı et al. 2020; Varol, Tokatlı 2021; Tokatlı, Islam 2022). These elements were grouped as heavy metals (S1), nutrients (S2), other physicochemical parameters (S3), and pesticides (S4). Benthic macroinvertebrates were also grouped based on the density ratio of specimens. Accordingly, benthic macroinvertebrate taxa were grouped as Oligochaeta, Chironomidae, Other Insecta, and Other Taxa to assess the effects of these pressure elements on each group. The effects of the pressure factors on organisms were scored on a relative scale of 0 to 3 to create the hypothesis effect matrix (0 - no effect; 1 - slight effect; 2 - considerable effect; 3 - severe effect; Table 4). Scoring was based on data available in the literature, including previous studies on the effect of these pressure elements on benthic macroinvertebrates (Kim et al. 2009; Rizo-Patron et al. 2013; Namwong et al. 2013; Prasetyo et al. 2016; Wandscheer et al. 2017). The results were evaluated by replacing the data in the formula (Harris et al. 1994):

$$D_k(i,j) = x_{ik} - x_{jk}$$

Matrix 
$$R = r_{ij} = \sum_{k=1}^{n} D_k(i, j); i, j = 1, 2, ..., m$$

Table 3

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The scale used to represent the fisk factors of E <sub>1</sub> , n <sub>1</sub> chin-Qr and merkin-Q (Long et al. 2003)										
	Potential Ecological Risk	Biologi	ical Risk Assessment							
E <sup>i</sup> r	Potential ecological risk for monomial factors R <sub>1</sub> Potential ecological ris for multinomial factor		Potential ecological risk for multinomial factors	ERM-Qi and mERM-Q	Biological toxicity risk for monomial and multinomial factors					
< 40	low ecological risk	< 95	low ecological risk	< 0.1	low priority side					
40–80	moderate ecological risk	95–190	moderate ecological risk	0.1-0.5	medium-low priority side					
80–160	considerable ecological risk	190–380	considerable ecological risk	0.5–1.5	high-medium priority side					
160–320	high ecological risk	> 380	very high ecological risk	> 1.5	high priority side					
> 320	very high ecological risk									

The scale used to represent the risk factors of  $E_{\mu}^{i}$ ,  $R_{\mu}$  ERM-Qi and mERM-Q (Long et al. 2005)

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#### Hypothesis effect matrix and relative values

Droccuro alamanta	Criteria						
	Oligochaeta	Chironomidae	Other Insecta	Other taxa			
Heavy metals (S1)	2	2	1	1			
Nutrients (S2)	2	1	1	1			
Other physicochemical parameters (S3)	1	1	1	1			
Pesticides (S4)	3	3	2	1			

0 - no effect; 1 - slight effect; 2 - considerable effect; 3 - severe effect

# 3. Results and Discussion

As a result of this study, 47 taxa of benthic macroinvertebrates were identified and an average of 8953 individuals per m<sup>2</sup> was found in the sampled paddy fields of the Meric-Ergene River Basin during the cultivation season. It was found that 12 taxa belonging to Oligochaeta were represented by 886 ind. m<sup>-2</sup>, 11 taxa belonging to Chironomidae were represented by 4199 ind. m<sup>-2</sup>, 14 taxa belonging to Other Insecta were represented by 2164 ind. m<sup>-2</sup>, and 10 taxa belonging to Other Taxa were represented by 1704 ind. m<sup>-2</sup> (Table 5). The group of chironomid larvae accounted for the largest percentage of the total abundance of all specimens with 46.9%, followed by the group of Other Insecta with 19%, and the group of Oligochaeta with 9.9% (Table 5).

In terms of the average number of individuals per m<sup>2</sup> in the sampling locations in relation to the irrigation water source, the largest number was found in fields irrigated with water from the Ergene River with 15,154 ind. m<sup>-2</sup>, followed by fields irrigated with artesian water – 11,146 ind. m<sup>-2</sup>, Meriç–Ergene water – 5146 ind. m<sup>-2</sup>, and Meriç water – 4364 ind. m<sup>-2</sup> (Table 5). During the cultivation season, the maximum number of individuals was found in summer with 14,290 ind. m<sup>-2</sup>, followed by autumn with 11,065 ind. m<sup>-2</sup> and spring with 1506 ind. m<sup>-2</sup> (Table 6).

During the rice growing season, it can be observed that the groups that first settle in the sediment in the spring season when water is first applied to the fields are those individuals whose eggs can survive during the dry phase, such as Annelida, some Insecta, Ostracoda, and Branchiopoda (Lawler 2001). We believe that the lower number of individuals found in this study in spring samples compared to samples from other seasons is due to the fact that the benthic fauna is just beginning to settle and temperatures are lower. In the summer season, there was an increase in the number of individuals and taxa presumably due to egg laying in the ecosystem, resulting in new taxa in the benthic fauna and individuals hatched from these eggs. The reason for the lower number of taxa identified and the number of individuals found in

autumn compared to summer may due to the relative decrease in temperature and depth, and the increase in the number of predatory species.

After evaluating the results of the research on benthic macroinvertebrates, it can be concluded that specimens of the identified taxa prefer shallow and stagnant waters and show wide tolerance to pollution and many environmental factors. Although the species identified in this study are generally adapted to stagnant water ecosystems, species that are often found in other habitats (e.g. *Stydorilus heringianus*) were also encountered in the study area, which may be due to river water recharge in these fields.

The highest abundance in the Chironomidae group was found for *P. nubifer* (40.6%) and the lowest for *K. tendipediformis* (0.1%), *C. mancus* (0.2%), *T. punctipennis* (0.4%), and *C. defectus* (0.8%). Also, the species observed with the highest abundance in this study (*P. nubifer, C. tentans* and *C. plumosus*) are known to be pollution indicators and they are species with high tolerance to environmental factors (Armitage et al. 1995; Epler 2001).

In the Oligochaeta group, *L. hoffmeisteri* (31.8%) proved to be the species with the highest abundance and is known to show high tolerance to pollution (Brinkhurst, Jamieson 1971). *H. perpusilla, L. variegatus,* and *E. tetraedra* are semi-aquatic forms of Oligochaeta (Schmelz, Collado 2010; Timm 2009). Therefore, the fact that these species were found in paddy fields with a maximum water depth of 10 cm and a semi-aquatic period proves that paddy fields are a very suitable ecosystem for their development.

In this study, *H. geminus*, belonging to Coleoptera in the Other Insecta Group, was found at all sampling locations and in all seasons, and was identified as the dominant species in the rice fields. *C. dipterum*, as the only species belonging to Ephemeroptera, was found at all sampling locations and in all seasons. These species prefer shallow water and stagnant ponds (Lee et al. 2013; Lupi et al. 2014).

According to the Shannon–Weiner index, the species diversity of benthic macroinvertebrates of rice fields in the Meric–Ergene River Basin was low with an average value of H = 0.89. While the diversity at the

## Table 5

# Abundance of benthic macroinvertebrate species per m<sup>2</sup> at the sampling locations (Ave – average, Abd – abundance, TA – total abundance)

	Macroinvortobrator	Sampling locations						
	Macronivertebrates	A	М	E	ME	Ave	Abd	TA
	Henlea perpusilla	66	14	0	96	44	5	0.5
	Lumbriculus variegatus	0	0	0	236	59	6.7	0.7
	Stylodrilus heringianus	0	246	0	82	82	9.2	0.9
ΟD	Eiseniella tetraedra	44	0	0	14	15	1.6	0.2
ß	Aulophorus furcatus	36	0	0	0	9	1	0.1
ta G	Chaetogaster limnaei	134	0	0	0	34	3.8	0.4
Jae'	Dero digitata	38	0	0	0	9	1.1	0.1
gocl	Pristina longiseta	14	0	0	30	11	1.2	0.1
Oli	Limnodrilus hoffmeisteri	1104	22	0	0	281	31.8	3.1
	Limnodrilus sp.	378	0	0	0	94	10.7	1.1
	Tubifex tubifex	0	104	0	282	96	10.9	1.1
	Oligochaeta (unidentified)	548	0	0	58	152	17.1	1.7
	Tanypus punctipennis	0	0	74	0	19	0.4	0.2
	Halocladius fucicola	0	208	0	0	52	1.2	0.6
٩	Chironomus plumosus	60	408	296	2154	730	17.4	8.1
ROL	Chironomus tentans	0	0	5570	0	1393	33.2	15.6
в е	Cryptochironomus defectus	0	110	0	30	35	0.8	0.4
ida	Kiefferulus tendipediformis	0	6	0	0	2	0.1	0.1
шo	Polypedilum convictum	0	0	572	0	143	3.4	1.6
ron	Polypedilum nubifer	192	1926	4170	534	1705	40.6	19.1
Chi	Cladotanytarsus mancus	0	38	0	0	9	0.2	0.1
	Virgatanytarsus arduennensis	90	112	0	0	50	1.2	0.6
	Chironomidae (unidentified)	244	0	0	0	61	1.5	0.7
	Cloeon dipterum	14	58	266	14	88	4.1	1
	Ischnura elegans	0	8	28	8	11	0.5	0.1
	Orthetrum albistylum	0	8	38	0	11	0.5	0.1
	Sigara lateralis	itata         38         0         0         0         9         1.1         0.1           ongiseta         14         0         0         30         11         1.2         0.1           us hoffmeisteri         1104         22         0         0         281         31.8         3.1           lus sp.         378         0         0         0         94         10.7         1.1           ubifex         0         104         0         282         96         10.9         1.1           ubifex         0         0         74         0         19         0.4         0.2           ubifex         0         0         5570         0         1393         33.2         15.6           innonstefectus         0         10         0         30         35         0.8         0.4           istendipediformis         0         6         0         0         12         0.1         1.1           us tandipediformis         0         38         0         0         20         0.1         1.1         0.5         0.1           us tandipediformis         0         112         0         6	0.1					
DU	Sigara striata	0	0	112	0	28	1.3	0.3
GRC	Corixidae (nymph)	8	214	28	22	68	3.1	0.8
cta	Hydroglyphus geminus	4414	222	1978	22	1659	76.7	18.5
ıse	Peltodytes caesus	0	0	0	96	24	1.1	0.3
er Ir	Berosus spinosus	22	0	0	0	5	0.3	0.1
)the	Coleoptera (larvae)	30	52	112	140	84	3.9	0.9
Ŭ	Ceratopogonidae	8	0	8	8	6	0.3	0.1
	Culicidae	8	0	466	8	121	5.6	1.3
	Ephydridae	22	0	66	0	22	1	0.2
	Stratiomyidae	28	0	22	52	26	1.2	0.3
	Nematoda	1504	282	556	512	713	41.9	8
	Erpobdella octoculata	8	8	28	0	11	0.6	0.1
Ч	Physella acuta	206	52	186	282	182	10.7	2
ROI	Gyraulus piscinarum	0	0	14	38	13	0.8	0.1
а Ю	Planorbis carinatus	0	8	0	244	63	3.7	0.7
Тах	Planorbis sp.	44	88	0	148	70	4.1	0,8
her	Ostracoda	1874	126	0	0	500	29.3	5.6
đ	Hydrachnidae	0	44	8	36	22	1.3	0.2
	Diplostraca	0	0	512	0	128	7.5	1.4
	Gammarus komareki	8	0	0	0	2	0.1	0
	Oligochaeta Total	2362	386	0	798	886	100	9,9
	Chironomidae Total	586	2808	10682	2718	4199	100	46,9
	Other Insecta Total	4554	562	3168	370	2164	100	24,2
	Other Taxa Total	3644	608	1304	1260	1704	100	19
	Total	11146	4364	15154	5146	8953	100	100
	Number of taxa	28	24	23	25	47		

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## Number of benthic macroinvertebrate individuals per m<sup>2</sup> in different seasons

Table 6

	Benthic macroinvertebrates	Spring	Summer	Autumn	Average
	Henlea perpusilla	133	0	0	44
	Lumbriculus variegatus	0	0	178	59
	Stylodrilus heringianus	0	211	33	82
ط	Eiseniella tetraedra	6	11	28	15
SOL	Aulophorus furcatus	0	0	28	9
a GF	Chaetogaster limnaei	0	0	100	33
aet	Dero digitata	0	28	0	9
och	Pristina longiseta	0	11	22	11
Jlig	Limnodrilus hoffmeisteri	0	694	150	281
Ŭ	Limnodrilus sp.	0	0	283	95
	Tubifex tubifex	0	78	211	97
	Oligochaeta (unidentified)	56	0	400	152
	Tanypus punctipennis	0	0	56	18
	Halocladius fucicola	0	156	0	52
₫	Chironomus plumosus	161	1983	44	729
g	Chironomus tentans	0	4178	0	1393
e GF	Cladotanytarsus defectus	0	83	22	35
ida(	Kiefferulus tendipediformis	0	6	0	2
mo	Polypedilum convictum	0	0	428	143
iron	Polypedilum nubifer	44	2361	2711	1706
С	Cladotanytarsus mancus	28	0	0	9
	Virgatanytarsus arduennensis	0	150	0	50
	Chironomidae (unidentified)	0	183	0	61
	Cloeon dipterum	11	33	222	89
	Ischnura elegans	0	6	28	11
	Orthetrum albistylum	0	0	33	11
	Sigara lateralis	11	11	11	11
DUP	Sigara striata	0	0	83	27
GR(	Corixidae (nymph)	0	206	0	69
cta	Hydroglyphus geminus	0	267	4711	1659
nse	Peltodytes caesus	72	0	0	24
ler l	Berosus spinosus	17	0	0	6
Oth	Coleoptera ( larvae)	100	117	33	83
	Ceratopogonidae	6	0	11	6
		0	6	356	120
	Epnydridae	0	44	22	22
	Stratiomyldae	0	33	44	20
		567	1411	101	/13
		0	22	170	192
UD	Physella acuta	0	300	1/8	182
GRC	Planorhis carinatus	0	189	0	63
axa (	Planorhis so	111	33	67	70
er Ta	Ostracoda	122	1378	0	500
the	Hydrachaidae	0	61	6	22
0	Dinlostraca	0	0	383	128
	Gammarus komareki	0	6	0	2
	_Total Oligochaeta	195	1033	1433	887
	Total Chironomidae	233	9100	3261	4198
	Total Other Insecta	217	723	5554	2164
		861	3434	817	1704
	Total	1506	14290	11065	8953
	Number of taxa	16	32	33	47

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sampling locations ranked as follows: ME (H' = 0.95) > M (H' = 0.94) > A (H' = 0.86) > E (H' = 0.81), the diversity across the seasons was in the following order: summer season (H' = 0.98) > spring season (H' = 0.92) > autumn season (H' = 0.88). The values of the Shannon–Weiner index showed that although the highest number of individuals was found at sites fed by the Ergene River (15,154 ind. m<sup>-2</sup>), the species diversity at these sites was low due to large differences between the number of individuals of each taxon.

Values of the Bray-Curtis Similarity index showed that the most similar sampling locations in terms of the dynamics of benthic macroinvertebrates were M and ME (36.9% similarity), and the most similar seasons were summer and autumn (27.7% similarity; Figs 2a, 2b). Based on the values of both physicochemical properties and benthic macroinvertebrates, the Bray-Curtis Similarity index indicated that the spring season was different from the other seasons (Fig. 2b, Fig. 3). These results confirmed that during the rice growing season, when the benthic fauna had just settled in the habitat in the spring, its dynamics was different from the summer and autumn seasons. Further, in terms of the dynamics of the benthic fauna, location A differed from the three other locations (E, M, ME), as expected due to the quality of artesian water (Fig. 2). The results were also supported by the Correspondence Analysis (Fig. 4).

The data for the physicochemical properties are presented in Table 7 as mean values from the sampling locations. According to the water quality



#### Figure 2

a – Dendrogram of similarity of the sampling locations for benthic macroinvertebrates; b – Dendrogram of similarity of the sampling seasons for benthic macroinvertebrates









assessment under the surface water quality regulation (Regulation on Surface Water Quality, 2016), the pH values were within the first quality level (Table 7). Conductivity, salinity, and TDS were at high levels in all seasons due to industrial wastewater discharge, as well as due to chemical fertilizers and pesticides used in agricultural fields (Tables 7 and 8). While the level of these parameters was lower in the spring season compared to other seasons due to the water used for irrigation of the fields, their values reached the highest level in the summer and autumn seasons due to high temperature. Although the temperature level was at the second quality level according to the water quality assessment under the regulation on surface water guality (Regulation on Surface Water Quality, 2016), it was suitable for rice growth. Total hardness FSº values indicated soft water in spring, and medium to hard water in summer and autumn. The values of nutrients NO<sub>2</sub>-N, NO<sub>3</sub>-N, and SO<sub>4</sub><sup>-2</sup> indicated the first quality level, while PO, -3 values indicated the second quality level (Table 7).

In this study, the presence of 21 types of pesticides out of 181 pesticides examined was determined in water samples collected from the sampling locations (Table 8). The results of heavy metal determination in the sediments collected at the sampling locations are presented in Table 9.

## Table 7

## Physicochemical parameters at the sampling locations in the study season

	Unit	Spring	Summer	Autumn	Average	Standard Deviation	Water quality
Temperature	°C	26.3	30.9	20.5	25.9	± 5.2	II
рН		8.8	7.6	5.8	7.4	± 1.5	I.
Conductivity	µS cm⁻¹	813.5	1707.5	2437.1	1652.7	± 813.2	III
Salinity	‰	0.5	0.8	1.3	0.9	± 0.4	-
TDS		500	1100	1400	1000	± 458.2	I.
Ca <sup>+2</sup>	mg l-1	53.3	33.2	57.9	48.1	± 13.1	-
Mg <sup>+2</sup>		6.1	4.2	0.0	3.4	± 3.1	-
T.H.	FS⁰	18.9	7.6	38.5	21.7	± 15.6	-
NO <sub>2</sub> -N		0.1	0.0	0.0	0.0	± 0.1	I
NO <sub>3</sub> -N		4.5	2.6	1.8	3	± 1.4	I. I.
PO <sub>4</sub> -3	mg l-1	0.3	0.3	0.2	0.3	± 0.1	II
SO <sub>4</sub> -2		3.6	3.1	2.7	3.1	± 0.5	I. I.
D.O.		4	5.6	3.5	4.4	± 1.1	III

## Table 8

Concentrations of the determined pesticides in water samples (ppm)								
Destiside	Station							
Pesticide	Artesian	Meriç	Ergene	Meriç–Ergen				
Acetamiprid	0.0227	0.0497	1.2092	0.672				
Azoxystrobin	0.0401	1.0789	13.6247	0.2291				
Carbendazim	0.374	1.8176	2.163	0.9806				
Carbofuran	0.0069	0.0105	*	*				
Cyproconazole	*	*	12.3969	*				
Dicrotophos	*	0.0122	*	0.0097				
Dimoxystrobin-688	0.0416	*	*	0.0488				
Ethiofencarb	*	0.0549	0.0734	0.0677				
Fluoxastrobin-698	0.0749	0.2242	0.1022	0.0803				
Flutriafol	0.1847	0.1777	0.2103	0.1871				
Forchlorfenuron-706	0.6974	1.3388	1.1613	2.4202				
Mandipropamid	*	*	*	0.0703				
Monocrotophos	0.0062	0.0102	*	0.0098				
Pirimicarb	0.0097	0.01	0.0052	0.0066				
Prometryn	0.021	*	*	*				
Spiroxamine	*	0.0546	*	0.0721				
Tebuthiuron	0.0204	0.0113	0.0228	0.0206				
Thiabendazole	0.0402	0.0353	0.0276	0.0228				
Thiacloprid	0.0229	0.025	0.026	0.0273				
Trifloxystrobin	0.1061	0.1048	0.1055	0.0507				
Vamidathion	0.0051	0.0087	0.0036	0.0077				
Number of pesticides	16 kinds	17 kinds	15 kinds	18 kinds				

No pesticide detected

Table 9

Concentration of heavy metals in sediment samples (ppm)

Water recourses	Heavy metals					
water resources	Cd	Ni	Cu	Mn		
Artesian	0.441	1.388	3.659	13.722		
Meriç River	0.818	0.78	7.397	13.169		
Ergene River	0.42	1.217	2.748	14.405		
Meriç–Ergene rivers (mixed)	0.538	0.858	2.855	13.485		

All the observed data and the literature data were used to assess the ecological risk profile in the study area.

The potential ecological risk index showed that all sampling locations were within the low ecological risk limits in terms of the content of heavy metals (Table 10). However, it was determined that the locations irrigated with water from the Meriç River were within the medium ecological risk limits for cadmium

Evaluation of ecological risk analysis for benthic macroinvertebrates in paddy fields in the Meric–Ergene River Basin (Turkish Thrace)

#### Table 10

Results of the potential ecological risk index associated with the presence of heavy metals measured in sediment at sampling sites

Mator recourses			:i 			
water resources	Cd	Cu	Ni	Mn	R <sub>I</sub>	
Artesian	26.826	0.610	0.139	0.016	27.591	
Meriç River	49.125	1.233	0.078	0.015	50.451	
Ergene River	25.215	0.458	0.122	0.017	25.811	
Meriç-Ergene mixed	32.280	0.476	0.086	0.016	32.857	

(Fig. 5). According to the biological risk index (BRI) data, the heavy metal was found at low priority limits for all sampled locations (Table 11, Figure 6). Evaluation of the results relating to the indices indicates the existence of an ecological risk, especially for cadmium in the Meric-Ergene River Basin.



## Figure 5

Potential ecological risk index graph of the sampling locations (PERI)

There are many studies related to ecological risk analysis based on heavy metal contamination of sediments in the basin and their results support the results of this study for cadmium risk (Tokatlı 2017; Tokatlı 2019a; Varol, Tokatlı 2021; Tokatlı, Islam 2022). Although these high concentrations are believed to be caused by industry in the basin, it can be concluded

Biological	risk	index	results	for	sediments	at	the
sampling l	ocati	ons					

Motor recourses	ERM-Q						
water resources	Cd	Cu	Ni	mERM-Q <sub>i</sub>			
Artesian	0.049	0.009	0.02	0.026			
Meriç River	0.09	0.018	0.01	0.039			
Ergene River	0.046	0.007	0.02	0.024			
Meriç-Ergene mixed	0.059	0.007	0.01	0.025			





Multinomial biological risk index graph for the sampling locations (BR,)

that agricultural activities also have a major impact. The study by Köleli and Kantar (2005) reported that high levels of cadmium were detected in the content of many fertilizers derived from different fertilizer factories in Turkey and used in agriculture (Köleli, Kantar 2005). Thus, it is inevitable that the content of heavy metals in the paddy fields is of anthropogenic origin and poses an ecological threat to the invertebrate fauna in the ecosystem. According to Tokatlı et al. (2020), carbendazim dominated in the Meric River Basin and pesticide concentrations, especially in the Ergene River, were at quite high levels and the system has water guality Class III-IV in terms of total pesticide accumulation (Tokatlı et al. 2020). According to the results of this study, carbendazim was found at all sampling sites and 21 types of pesticides were found in water of paddy fields (Table 6). These contaminants, introduced into the ecosystem through anthropogenic practices, pose a threat to benthic macroinvertebrates and consequently to the biodiversity of the ecosystem.

As a result of the evaluation of hypothetical ecological risk factors that were identified based on the results of statistical analyses applied to the obtained data, it can be concluded that pesticides (S4) have a strong effect on benthic macroinvertebrates in paddy fields in the Meric-Ergene Basin (Table 12), followed by the effects of heavy metals and nutrients, while the remaining physicochemical parameters can be interpreted as having the least effect (Table 12).

#### Table 12

Results of the hypothetical impact matrix

Stressors assessment	S1	S2	S3	S4	Row sum
					(impact assessment)
S1	0	1	2	-3	0
S2	-1	0	1	-4	-4
S3	-2	-1	0	-5	-8
S4	3	4	5	0	12

Table 11

The literature reports that species diversity is higher in organically farmed paddy fields where pesticides are not used (Kim et al. 2009; Rizo-Patron et al. 2013; Namwong et al. 2013; Prasetyo et al. 2016; Wandscheer et al. 2017). Extending the hypothetical risk analysis further, the water quality and accumulation status of each risk factor and the degree of impact on aquatic birds and fish that feed on invertebrates can be assessed. Consequently, it can be concluded that biodiversity provided by benthic macroinvertebrates will increase in these special wetland ecosystems by eliminating or at least significantly controlling the identified high-impact ecological risks.

# 4. Conclusions

In this study, the benthic macroinvertebrate fauna of paddy fields in the Meriç–Ergene River Basin was qualitatively and quantitatively assessed for the first time, and it was emphasized that paddy fields are an important aquatic ecosystem with significant biodiversity. However, these areas are under ecological risk due to climate change caused by global warming and especially anthropogenic effects such as excessive use of pesticides and chemical pesticides. For the sake of biodiversity of these areas, the use of pesticides and fertilizers to control all kinds of pests that damage paddy plants should be reduced, and instead biological control should be carried out and both farmers and the public should be made aware of this issue.

# **Acknowledgements**

This study is part of the corresponding author's Ph.D. thesis.

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