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Effect of Silver on Aquatic Ecosystems of Emet Stream Basin, Turkey

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Abstract.- Emet Stream is one of the most important branches of Uluabat Lake (Ramsar area) and also one of Turkey's most important river systems. In addition to the geologic structure of the basin, Eti Silver Mine is a significance source of silver for the basin. In this study, water, sediment and fish samples (*Squalius cii* (Richardson, 1857); *Capoeta tinca* (Heckel, 1843); *Barbus oligolepis* Battalgil, 1941) were collected from 8 stations (1 of them is on the Kınık Stream and 1 of them is on the Dursunbey Stream) from Emet Stream Basin seasonally between November 2010 – August 2011 to determine the Ag levels in biotic and abiotic components of Emet Stream Basin. Cluster Analysis was applied to the results to classify the stations according to the contents of silver by using the Past package program and also One Way Anova Test was applied to the results to determine the significant differences of silver accumulations by using the SPSS 17 package program. According to data, silver accumulations in abiotic components of Emet Stream Basin were determined as sediment > water and in biotic components (fish tissues) of Emet Stream were determined as kidney > liver > gill > muscle for *S. cii*, liver > gill > kidney > muscle for *C.tinca* and gill > liver > kidney > muscle for *B. oligolepis*, respectively.

Keywords: Emet Stream, Silver, *Squalius cii, Capoeta tinca, Barbus oligolepis*

INTRODUCTION

Silver, as ionic Ag^+, is one of the most toxic metals known to aquatic organisms. Water concentrations of 0.0012–0.0049 mg/L killed sensitive species, although large industrial losses to the aquatic environment are probably infrequent because of its economic value as a recoverable resource (EPA, 1980; Nebeker *et al*., 1983; Bulh and Hamilton, 1991; Bryan and Langston, 1992). However, silver is of concern in various aquatic ecosystems because of the severity of silver contamination in the water, sediment and biota (Rivera-Duarte and Flegal, 1993). Silver that occurs naturally in the earth's crust is a relatively rare metal and found in the environment combined with other elements such as sulfide, chloride, and nitrate. The natural wearing down of silver – bearing rocks and soil by the wind and rain releases large amounts of silver into the environment. It is also released to the environment from various industrial sources and may be carried long distances in water (ATSDR, 1990).

Emet Stream is one of the most important branches of Uluabat Lake and also one of Turkey's most important river system. Eti Silver Mine is the most important source of silver not only for the immediate operating area but also for all around the Kütahya region and also Emet Stream Basin. It is the only mine in Turkey that produces direct silver from ore and has 21,5 million ton reserves. Eti Silver Mine that covers 1% of world's and 45% of Turkey's silver production is located on the 33 km northwest of Kütahya and approximately 50 km far away from Emet Stream (www.etigumus.com.tr; www.mta.gov.tr)

The aim of this study is to determine the Ag levels in biotic (muscle, gill, liver and kidney) tissues of *Squalius cii* (Richardson, 1857), *Capoeta tinca* (Heckel, 1843) and *Barbus oligolepis* (Battalgil, 1941) and abiotic (water and sediment) components of Emet Stream Basin.

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MATERIALS AND METHODS

Study area and collection of samples

Sampling stations on the Emet Stream Basin were given in the map (Fig. 1). Sediment and water samples were collected seasonally between November 2010 – August 2011 from all stations by using sediment dipper and suitable containers. Fish samples were collected in winter season from four stations (E2, E4, E7, E8 stations) where were most suitable to get fishes by using Honda generator.

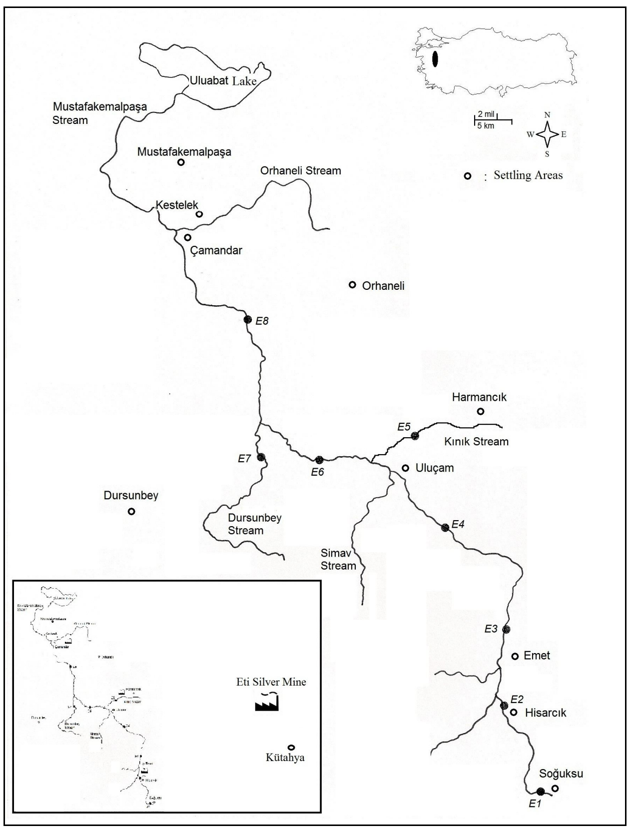

Fig. 1. Study area (Tokatlı *et al.*, 2012)

Chemical analysis

The procedure adopted for chemical analysis has been the same as reported earlier (Tokatli *et al.*, 2012).

Statistical analysis

Cluster Analysis according to Bray Curtis, Boxplot deviation and Matrixplot distribution diagrams were applied to the results by using the Past package program. One Way Test according to Tukey were applied to the results by using the SPSS 17 package program and the graphics of water, sediment and tissues of fishes were made by using the Microsoft Office 2007 package program.

RESULTS AND DISCUSSION

Seasonally Ag levels determined in water and sediment of Emet Stream Basin and seasonal variations were given in Figure 2. The highest Ag level in water was determined at station E4 (0.00141 mg/L) in spring season and the lowest Ag level in water was determined at station E1 (0.00035 mg/L) in autumn season. The highest Ag level in sediment was determined in station E4 (0.733 mg/kg) in autumn season and the lowest Ag level in sediment was determined at station E5 (0.273 mg/kg) in spring season.

It is known that, silver is usually found in extremely low concentrations in natural waters because of its low crustal abundance and low mobility in water (EPA, 1980). One of the highest silver concentrations recorded in fresh water, 0.038 mg/L, occurred in the Colorado River at Loma, Colorado, downstream of an abandoned gold – copper – silver mine, an oil shale extraction plant, a gasoline and coke refinery and an uranium processing facility (EPA, 1980). According to data

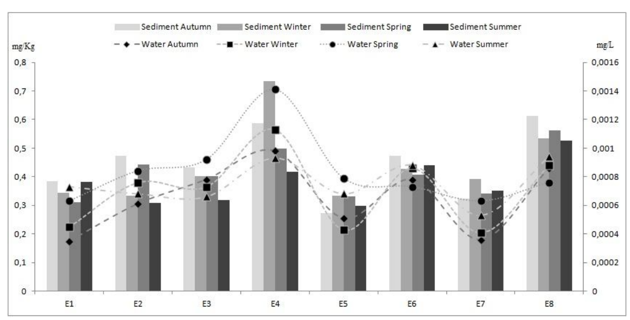

Fig. 2. Seasonal variations of Ag levels in water and sediment.

obtained in the present study, although an active silver mine is located in the Emet Stream Basin, Ag accumulations in water were not as much as Colorado River. So it can be thought to be that, Eti

Silver Mine may not affect the system directly and it may affect the system indirectly by groundwater line. The bottom and surface water samples of Alpine lake (Colorado) contained silver at concentrations ranging between 0.00022 and 0.00031 mg/L in 1973 and between 0.00044 and 0.00071 mg/L in 1974 where the silver iodide (43 kg), equivalent to 19,7 kg silver, released into system from local cloud-seeding practices between 1963 and 1973 (Freeman, 1979). If we compare the results of this study with Alpine Lake (known to be contaminated), Ag accumulations in Emet Stream Basin even the source regions with the lowest Ag accumulations of system were much higher than Alpine Lake. Environmental interactions with the geological structure of the Emet Stream Basin can cause a significant increasing of silver levels in water.

Aquatic system's sediments that receive metals, mining wastes or sewage usually have higher silver concentrations (> 0.1 mg/kg) than noncontaminated sediments (Bryan and Langston, 1992). In the present study, Ag accumulations in sediment of Emet Stream Basin (even source stations) have extremely (4 – 7 times) exceed the limit value specified by sediment by Bryan and Langston. So the sediments of Emet Stream Basin can be classified as contaminated sediment according to this classification.

According to the results of One Way Anova Test determined by using Ag accumulations of water and sediment, E4 station shows a statistically significance differences from E1, E2, E3, E5 and E7 stations according to water; E4 and E8 stations show a statistically significance differences from E1, E2, E3, E5 and E7 stations according to sediment ($p < 0.05$). It can be understood from the data that, system is being affected from environmental interactions and also Eti Silver Mine by especially groundwater line around E4 and E8 stations.

According to seasonal variations of silver levels in water and sediment, significant increases of Ag accumulations in water in spring season and in sediment in autumn season were determined. Rain washes silver compounds out of many soils so that it eventually moves into the surface and groundwater (ATSDR, 1990). In spring season, more rainfall in Emet Stream Basin may explains the sudden rise of silver in water and also the Ag content of water is transported directly into the Uluabat Lake, before accumulated on the sediment especially in this season. According to Arslan *et al*. (2009), the pressure of silver reduces the water quality of Uluabat Lake and they predicted that, water quality in Uluabat Lake will be reduced more in the future.

Ag levels of fish tissues and the results of One Way ANOVA Test that compares the Ag bioaccumulations of fishes are given in Table II. The Ag accumulations in tissues of *S. cii* follow as, kidney > liver > gill > muscle; in tissues of *C. tinca* follow as, liver > gill > kidney > muscle; in tissues of *B. oligolepis* follow as gill > liver > kidney > muscle respectively. The highest Ag level of tissues was determined in E4 station (0.765 mg/kg) in liver of *C. tinca* and the lowest Ag level of tissues was determined in E7 station (0.175 mg/kg) in muscle of *B. oligolepis*.

In general, the higher metal accumulations in fish tissues were determined in liver and kidney tissues and the lover accumulation was in muscle tissues. The metals (non – lethal concentrations) are accumulating in metabolically active tissues (Kargın and Erdem, 1992). In this study, the lowest Ag levels were determined in muscle tissues of all fish species and the highest Ag levels were determined in kidney tissues of *S. cii*, liver tissues of *C. tinca* and gill tissues of *B. oligolepis*. The higher accumulation in liver and kidney may alter the levels of various biochemical parameters in these tissues. This may also cause severe liver and kidney damage and a primary impact on the health of fish (Ferguson, 1989; Mayers and Hendricks, 1984).

If we evaluate the Ag bioaccumulations of fish tissues on human health, Ag contents of muscle tissues were not seen as dangerous. It can conclude that silver does not apparently pose a health threat in view of its concentration levels in the fish tissues investigated. But as it is known that, food chain biomagnification of silver in aquatic systems is unlikely at silver concentrations normally encountered in the environment (Connell *et al*., 1991), although regular ingestion of fish from contaminated waters may significantly affect dietary silver intake in humans (EPA, 1980).

According to the literature, freshwater fishes are the most sensitive vertebrates to dissolved silver,

Table I.- **Some metric characteristics of fishes.**

	E2		E4		E7		E8	
	Weight (g)	**Total length (mm)**	**Weight (g)**	**Total length (mm)**	**Weight (g)**	**Total length (mm)**	**Weight (g)**	**Total length (mm)**
S. cii (n=36)								
Minimum	16,00	121,00	14,00	113,00	14,00	114,00	34,00	149,00
Maximum	84,00	196,00	115,00	215,00	76,00	193,00	131,00	225,00
Mean	48,00	161,33	58,78	170,78	41,63	153,50	69,80	180,00
SD	19,68	22,02	31,18	32,31	23,47	29,09	28,63	21,41
C. tinca (n=35)								
Minimum	14,00	118,00	10,00	102,00	20,00	121,00	14,00	114,00
Maximum	114,00	223,00	168,00	245,00	72,00	193,00	132,00	229,00
Mean	51,56	165,44	62,67	170,00	37,89	155,56	52,00	166,88
SD	34,30	36,71	49,26	44,74	15,95	23,42	39,08	36,74
B. oligolepis (n=21)								
Minimum	9,00	94,00	9,00	98,00	8,00	95,00	8,00	103,00
Maximum	42,00	169,00	91,00	212,00	51,00	181,00	45,00	159,00
Mean	18,40	123,00	36,40	142,40	20,20	129,20	22,17	128,17
SD	13,72	28,13	33,22	45,11	18,27	34,80	14,19	22,66

n: number of samples; min: minimum; max; maximum; mean; average values; SD; standard deviation

Table II.- **Ag levels of fish tissues.**

Fish tissues	**Stations**				**Stational average* (mg/kg)**
	E2 (mg/kg±SE)	**E4 (mg/kg±SE)**	**E7 (mg/kg±SE)**	**E8 (mg/kg±SE)**	
S.cii					
Muscle	0,243±0,0533	0,255±0,045	0,242±0,0575	0,265±0,045	0,25125^{m}
Gill	0,551±0,1033	0,465±0,055	0,385±0,0975	0,396±0,0666	0,44925^{g}
Liver	0,747±0,13	0,742±0,115	0,635±0,0825	0,716±0,13	0,71^{L}
Kidney	0,746±0,0366	0,751±0,2	0,677±0,09	0,742±0,09	0,729^{K}
C. tinca					
Muscle	0,243±0,0313	0,243±0,04	0,213±0,045	0,225±0,035	0,231^{m}
Gill	0,592±0,135	0,565±0,125	0,435±0,075	0,513±0,12	0,52625^{g}
Liver	0,695±0,125	0,765±0,09	0,574±0,02	0,714±0,11	0,687^{L}
Kidney	0,514±0,16	0,533±0,08	0,434±0,1	0,542±0,05	0,50575^{k}
B. oligolepis					
Muscle	0,332±0,08	0,224±0,01	0,175±0,09	0,253±0,045	0,246^{m}
Gill	0,618±0,13	0,603±0,13	0,412±0,12	0,474±0,085	0,52675^{g}
Liver	0,526±0,05	0,531±0,03	0,454±0,08	0,538±0,06	0,51225^{l}
Kidney	0,485±0,09	0,512±0,1	0,425±0,08	0,547±0,12	0,49225^{k}

*: The values marked with uppercases and lowercases according to initial letters of tissues are statistically different ($p < 0,05$)

although relatively tolerant species exist. In fish toxicity tests with 22 metals and metalloids, silver was the most toxic tested element as judged by acute LC_{50} values (Birge and Zuiderveen, 1995; Ratte, 1999). In general, the detoxification mechanisms of fishes are the same and the metals are primarily linked to the metallothionein protein in the liver and the kidney is also the secondary gateway for heavy metal detoxification in body and reduces the effects of metals (Vinodhini and

Narayanan, 2008). But as reported in many studies, the types of fishes and different physiological structures may affect to accumulate metals in different organs in different levels (Canli and Kalay, 1998; Cid *et al*., 2001; Karadede *et al*., 2004; Mendil and Uluözlü, 2007; Al-Weher, 2008; Sen *et al*., 2011).

According to the results of One Way Anova Test determined by using Ag bioaccumulations of fish tissues (Table II), *S. cii* and *C. tinca* show a statistically significance differences from *B. oligolepis* according to liver tissues; *S. cii* shows a statistically significance differences from *C. tinca* and *B. oligolepis* according to kidney tissues ($p<0.05$). Metal accumulation in muscle and gill is not related with the physiology of fishes, but it is directly related with the accumulations of metals in water and the physiology of fishes impacts the activities of liver and kidney tissues more than accumulations of metals in water. So observing no significant differences between muscle and gill tissues of fishes and observing significant differences between liver and kidney tissues of fishes according to bioaccumulation of silver were expected results. If a fish species, which is exposed to a toxic metal at the same rate with other fish species, accumulates that metal in liver and kidney tissues more than other fishes, this may mean that, this species is more sensitive to this metal than the others. In a study performed on Ag accumulations of *Oncorhynchus clarki* (cutthroat trout), which is clearly known to be very sensitive to inorganic pollution (NRCS 2007), Ag accumulation in liver tissue of *O. clarki* (2,3 mg/kg) was reported extremely higher than the Ag content of muscle tissue (0.1 mg/kg) (Freeman, 1979). If we compare this study with the present study, although *O. clarki* was exposed to silver much less than the present study, the accumulation of Ag in liver tissue of cutthroat trout was extremely higher than the accumulations of Ag in liver tissues of *S. cii*, *C. tinca* and *B. oligolepis*.

S. cii is only found on Lesbos and in the streams flowing into the northern Aegean in Turkey. The species is locally threatened by pollution, water abstraction and drought. The species only survives in a few narrow areas with limited populations due to heavy industrial pollution. Although, these threats are not thought to cause the species to qualify for a threatened or near threatened category, populations of *S. cii* are decreasing day by day because of especially inorganic pollution (IUCN, 2011). According to data, the accumulations of Ag in liver and kidney tissues of *S. cii* were statistically higher than the accumulations in *C. tinca* and *B. oligolepis*. So, it can be understood from the data that, *S. cii* can be more sensitive to silver pollution and adversely affected than the other species.

The matrixplot diagrams show the distribution of Ag levels in water and sediment of Emet Stream Basin according to stations and seasons, and average Ag levels in muscle, gill, liver and kidney tissues of three fish species according to stations (Fig. 3).

Boxplot deviation diagrams show the range and mean concentrations of Ag levels in water, sediment and three fish tissues of Emet Stream Basin (Fig. 4). According to the boxplot diagrams, the highest deviation of Ag concentrations was observed in autumn and winter seasons for water and in autumn season for sediment. Although the lowest deviation of Ag concentrations was observed in spring season for water, a high deviation between maximum value and upper quartile (Q3) of boxplot was also observed in this season. The highest deviation between maximum value and upper quartile (Q3) of boxplot was observed in winter season and a significant deviation between minimum value and lower quartile (Q1) of boxplot was observed in autumn season for sediment. According to the last boxplot diagram, the highest deviation of Ag accumulations was determined in kidney tissues and also a significant deviation was observed between median (Q2) and upper (Q3) quartile of boxplot. Although the highest deviations of maximum and minimum Ag accumulations from lower (Q1) and upper (Q3) quartile of boxplot were determined in muscle tissues, the lowest deviation was also determined in these tissues.

The diagram of Cluster Analysis calculated by using Ag levels of water and sediment is given in Figure 5. According to the Cluster similarity and distance analysis, E2 and E3 stations show the highest similarity (94,4%) and E5 and E8 stations show the lowest similarity (71,1%). While the stations of E1, E2, E3, E5, E6 and E7 with lower Ag

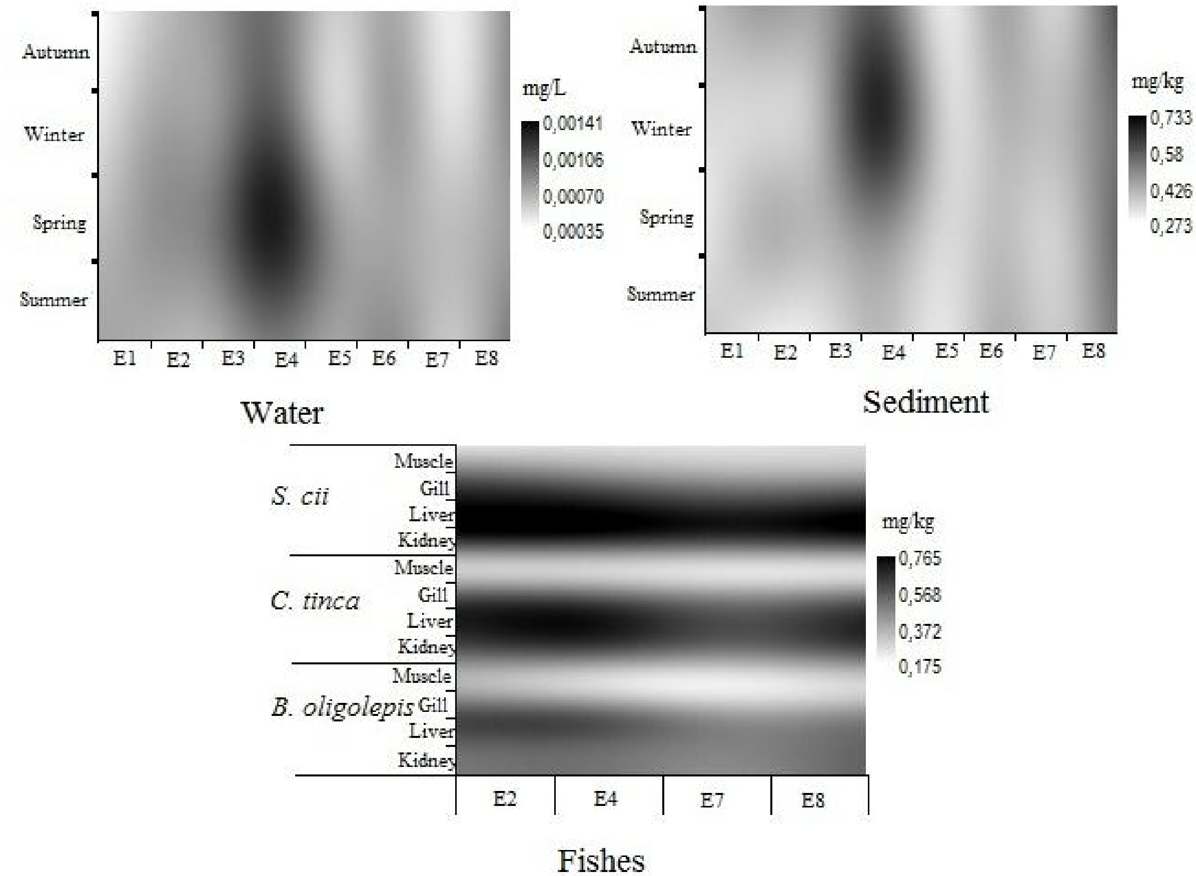

Fig. 3. Matrixplot diagrams for water, sediment and fishes

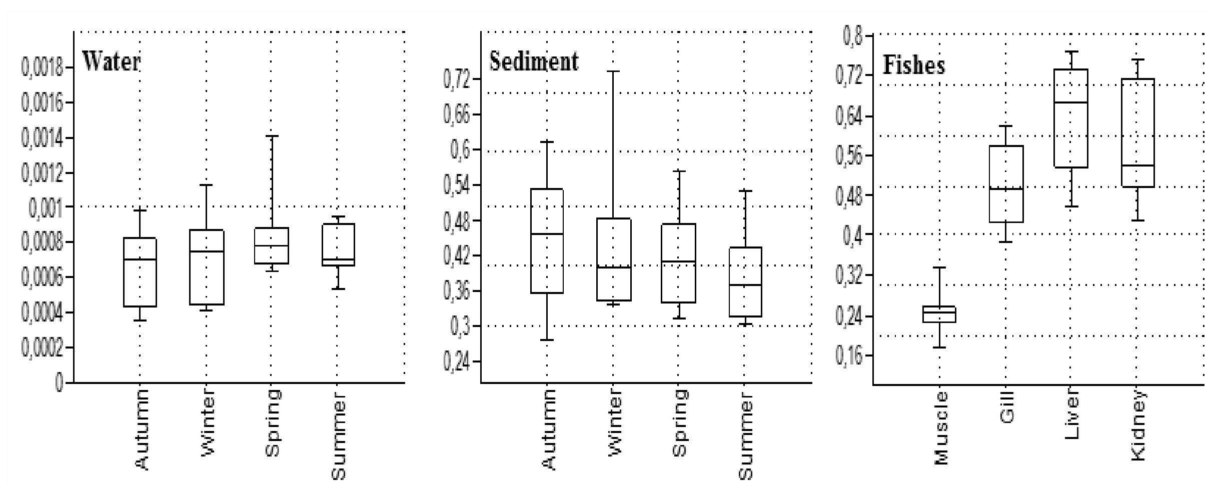

Fig. 4. Boxplots for water, sediment and fish tissues

levels are forming a group with each other, the stations of E4 and E8 with higher Ag levels are forming a group with each other.

E1 and E2 stations were closed to the source of Emet Stream and were not exposed to Ag discharge as much as the other stations. Although abiotic components of E2 station were not contaminated by silver, biotic components of E2 station were significantly contaminated. Fishes are active organisms so they can be affected by contaminations from large areas. Fishes of E2 station were affected from the high contaminated areas on the Emet Stream.

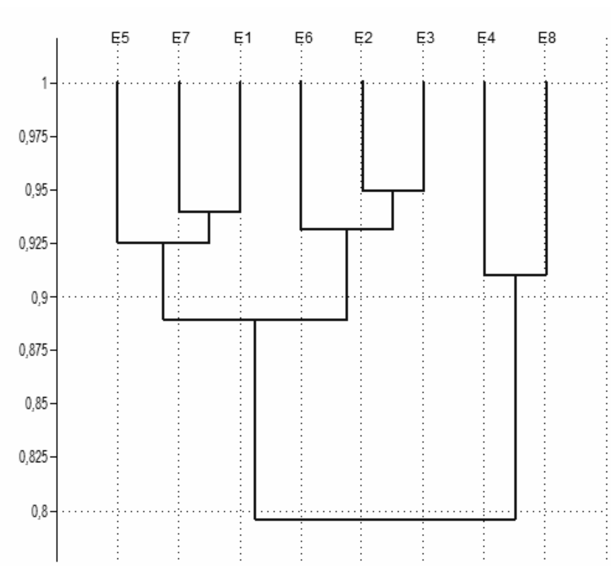

Fig. 5. Diagram of Cluster Analysis

E3 station was located closed to the Emet District and exposed to domestic wastes. Although it is contaminated by organic pollution and located closed to the Eti Silver Mine, it was not contaminated by Ag.

E4 station was located on the Emet Stream and it was the closest station to the Eti Silver Mine. A significant increasing of Ag levels in both biotic and abiotic components of Emet Stream was determined after especially from this station. So it can be thought that, Eti Silver Mine can primarily affect the system around E4 station by groundwater line.

E5 station was located on the Kınık Stream and far away from domestic and agricultural activities. But this stream is under pressure of Harmancık Chromium Mine and it is exposed to an important inorganic pollution. Ag content of this station in water and sediment shows similarity with E1 and E7 stations. So it can be understood that, Kınık Stream does not constitute any risk for Emet Stream Basin in terms of silver, and chromium mines around the basin are ineffective for Ag contaminations to environment.

Although E6 station was located on the down side of Emet Stream and exposed to organic and inorganic pollutions, E5 station has been caused to diluting the silver content of water and sediment in this station.

E7 station was located on the Dursunbey Stream which is an important branch of Emet Stream. Ag levels in water, sediment and fish tissues of E7 station reflect that, there is no significant Ag source around the region. Although this station was located on the down side of Dursunbey Stream, it has a low silver content. So it can be understood that, Dursunbey Stream is not exposed to Ag discharge and does not constitute any risk for Emet Stream Basin in terms of silver.

E8 station was located on the down side of Emet Stream and the Ag accumulations on biotic and abiotic components of E4 and E8 stations show similarity. Eti Silver Mine and environmental interactions can affect the system around E4 and E8 stations by groundwater line and rain wash.

CONCLUSIONS

As a result, the study clearly indicates that, Ag accumulations in abiotic and biotic components of Emet Stream Basin are under affect of environmental interactions directly and Eti Silver Mine indirectly and carries this pollution to the Uluabat Lake. The silver plant that is located in the basin and the environmental interactions affect E4 and E8 stations significantly higher than the other stations and Ag levels of water in the entire river are rising in parallel of rainfall. It was also determined that, the higher metal accumulations in fish tissues were seen in liver and kidney tissues and the lover accumulations were seen in muscle tissues in general. The fishes of Emet Stream Basin accumulate the silver more than their environments, they can affect from changes of Ag content of water and sediment, and also *S. cii* is more sensitive to silver pollution and adversely affected than the other species. These all adverse situations pose an important risk factor for the aquatic system and also for public health around the Emet Stream Basin.

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