



Effects of heavy metal pollution on population dynamics of another important pollinator insect group: Horseflies (Diptera: Tabanidae)

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Abstract

Horseflies represent important agents of pollination as well as other pollinator insects, like butterflies, bees and wasps. Horseflies belong to the genera *Atylotus*, *Dasyramphis*, *Glaucoptis*, *Pangonius*, *Philoliche*, *Chrysops*, *Hybomitra*, and *Tabanus* species are exclusive nectar feeders. Glucose, fructose and sucrose are major components of plant nectars which are used nourishing for developing eggs. Because of these features horseflies have a significant role in pollination of phanerogams. This study was conducted to investigate the effects of toxic heavy metals on the population dynamics of horsefly species. Monthly changes on the accumulation of heavy metals in the larval habitats of horsefly species were determined with Perkin Elmer Optical Spectrophotometer and also correlations between concentrations of heavy metals and population fluctuations of horse fly species were observed. The results demonstrated that lead, iron and cadmium have statistically important negative effects on the population dynamics of horsefly species.

Keywords: Diptera, heavy metal pollution, horseflies, population dynamics, Tabanidae

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Ağır metal kirliliğinin bir diğer önemli polinatör böcek grubu üzerindeki etkileri: Atsinekleri (Diptera: Tabanidae)

Özet

Atsinekleri, arılar ve kelebekler gibi diğer polinatör böcekler kadar önemli polenizasyon ajanları arasındadır. *Atylotus*, *Dasyramphis*, *Glaucoptis*, *Pangonius*, *Philoliche*, *Chrysops*, *Hybomitra* ve *Tabanus* cinslerine ait at sineği türleri özel olarak nektarla beslenirler. Glukoz, fruktoz ve sakkaroz, bitki nektarlarının başlıca bileşenleridir ve gelişmekte olan yumurtaların beslenmesinde kullanılırlar. Bu özellikleri nedeniyle at sinekleri fanerogamların tozlaşması için önemli bir role sahiptirler. Bu çalışma, toksik ağır metallerin at sineği türlerinin popülasyon dinamiği üzerindeki etkilerini araştırmak amacıyla yürütülmüştür. At sineği türlerinin larval yaşam alanlarında aylık ağırlık artışıdaki değişimler, Perkin Elmer Optik Spektrofotometre kullanılarak belirlenmiş ve aynı zamanda ağır metal konsantrasyonları ile at sineği türlerinin popülasyon dinamikleri arasındaki ilişkiler gözlemlenmiştir. Sonuçlar, kurşunun, demirin ve kadmiyumun at sineklerinin popülasyon dinamiği üzerinde istatistiksel olarak önemli negatif etkilere sahip olduğunu göstermiştir.

Anahtar kelimeler: Diptera, ağır metal kirliliği, At sinekleri, popülasyon dinamiği, Tabanidae

1. Introduction

Pollination is a significant issue for maintaining the ecological balance and ecosystem. Pollinator insects involve the majority of social and solitary bees, wasps, flies, beetles, butterflies, and moths. These pollinators are essential for the reproduction of fruits, vegetables, and grains for both animals and humans. Global changes, magnified

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land use, climate change, alien species, and the spread of pests, pathogens and intensive chemical use in agricultural lands have growing pressure on the insect pollinators [1, 2]. All these impacts not only change the status of pollinators but also effect their population dynamics, distribution, abundance, and dietary habits.

The horseflies belonging to the family Tabanidae are important regulators of the ecosystem. Although they are known as vector organisms, horseflies especially males need sucrose due to need large amounts of energy for flight, they obtain this energy from nectar and pollen. Despite bloodsucking behavior, long-proboscid Tabanidae species are significant nectar feedings and recognized as key species for pollination of plants of some regions [3]. Previous studies proved that long-proboscid Tabanidae co-evolved with flowering plants since the Late Jurassic and they are represented among the first pollinator insects of early angiosperms [4, 5]. Miller (1951) [6] confirmed that adult tabanids transport the pollens attached to their bodies. Horseflies belong to genus *Atylotus*, *Dasyramphis*, *Glaucops*, *Pangonius*, *Philoliche*, *Chrysops*, *Hybomitra*, and *Tabanus* are exclusive plant visitors for nectar, pollen, and plant exudates [7, 8, 9]. (Kniepert, 1980)[7] investigated the dietary preferences of adult male and female tabanids and reported that the 53% of female and 69% of male specimens carry nectar sugars. (Bosler & Hansens, 1974)[10] found that the greater than 80% of female *Tabanus nigrovittatus* had consumed nectars. Magnarelli & Anderson (1976) [11] reported that *T. nigrovittatus*, *Chrysops atlanticus* and *Chrysops fuliginosus* had used nectar sugars as normal diary diets. Furthermore, it is usual that most of the collected *C. celatus* had taken fructose, water-soluble sugar found in a variety of fruits for carbohydrates has been reported as a significant dietary elements for horseflies surely to renew decreasing energy reserves during flight [12, 13]. Magnarelli et al. (1979) [12] established that female *C. atlanticus* and *C. fuliginosus* visited yarrow and swamp rose many times, through the day and they also reported that 119 pollen grains, belonging to Compositae, Gramineae and Rosaceae, identified from digestive tracts of females. On the otherhand glucose, fructose and sucrose are the base components of plant nectars which are using for nourishing their developing eggs, were detected from adult digestive systems [11, 12, 13]. It can be easily argued that, horsefly species have important effect on the pollination of many phanerogam species of the Compositae, Gramineae, Rosaceae, Geraniaceae, Iridaceae, Orchidaceae, Amaryllidaceae, Scrophulariaceae, Apiaceae, Amaryllidaceae, Convolvulaceae, Poaceae, Umbelliferae, as well as other pollinator insects [14, 15]. Consequently, changes of population dynamics of horsefly species effect the life cycle of phanerogams.

On the other hand, all stages of tabanids are absolute food source for other ecosystem components. Thereby, decreasing of population density of horsefly species affects many other species. Natural habitats sustain the pollinators, providing a strong and reciprocal pollination service that increases yield capacity [16, 17]. However, intensive chemical pesticide usage in the agricultural areas affect to the pollinator insects with target pests and decrease the crop quality which is consumed by pollinators. With the pesticides some heavy metals accumulate in the soil and water sources and affect all ecosystem agents. Horseflies prefer different habitat conditions like aquatic, semiaquatic and terrestrial zones in accordance with their life cycle. These habitats may be affected because of some pollution ingredients and population dynamics of horseflies may be damaged. This damage may cause decreasing pollination by tabanids and may be destroyed food chain. Some recent studies indicate that toxic heavy metals such as cadmium, ferric, aluminum, mercury and lead inhibit or restrict larval growth rates of fly larvae [18, 19].

This study therefore set out to assess the effect of the heavy metal accumulation in the larval habits and the effects on the population dynamics of horsefly species.

2. Materials and methods

The study was conducted in the marsh area, which is center of oak and pine forest, in Yarımca village in Eskişehir Province, Türkiye (39° 53' 936" N, 30° 37' 747" E, 1171m).

In order to determine the presence of heavy metal concentration in larval habits, eight different streams and four different soil samples were collected each month during three years in active periods of horseflies. Collected samples were frosted by dry ice and transferred to the laboratory with thermoflasks. Concentrations of the heavy metals in water and soil were examined with Perkin Elmer Optical Emission Spectrometer Optima 4300 DV. Larval habitats of horsefly species show great variability, larvae of many species have cannibalistic behavior and larval development periods vary as per to species. Therefore, population dynamics were investigated on adult samples. Adult horse flies were collected with water and malaise traps, in the active period durations of species (May to October 2017-2019).

Adult specimens were collected every 20 minutes from 09:00 to 20:00 and they were placed in ethyl acetate containing jars and carried to the laboratory. Species identification was made according to the Chvala et al. [20].

Population dynamics of the collected horsefly species were analyzed and given in (Table 1). The statistical models were modified from Karpakakunjaram et al. [21]. SPSS was used and datas were evaluated with the nonparametric Kendall's correlation coefficient test.

The weekly population data of species were converted to monthly means for analysis and were investigated for correlation with heavy metal concentrations. All data (populations and concentrations) were converted to normal varieties; $Z = (X - \mu) / \sigma$; were Z is the standard normal deviation from the mean ($X - \mu$) of the data, measured in units of standard deviations. The conversions were done to import the values of heavy metals and the population input to a comparable scale for plotting the histogram and also further analyses. Totally 50 correlation coefficient values were

obtained from Kendall's correlation coefficient tests. In order to eliminate significant correlations, nonparametric sequential Bonferroni tests were performed.

3. Results

Tabanidae family represented with 176 species and 15 subspecies in Türkiye [22]. In this study, totally 7234 adult samples belonging to 52 species were collected and identified. The list of species and seasonal distributions are given in Table 1.

Triticum vulgare, *Brassica juncea* and *Zea mays* are commonly harvested crops at study area (about 5 km²). These wetland agricultural areas are well productive but insecticides are used concentratedly. According to the results of analysis, the accumulation of lead, iron, cadmium, mangan and aluminum were determined in the samples of soil and stream. Results of correlations between these heavy metals and population dynamics of selected specimens indicate that lead, iron and cadmium have significantly negative effects on the populations. Meanwhile the lead pollution is commonly originated from the presence of highway and dye; lead from processing fabrics; iron and cadmium pollution is generally based on insecticides, pesticides and other chemicals which are used for agricultural purpose.

Table 1. The list of the collected species and seasonal distributions

Species	2017					2018					2019				
	May	June	July	Aug	Sept	May	June	July	Aug	Sept	May	June	July	Aug	Sept
<i>Tabanus bromius</i> L.	4	141	528	75	19	2	47	86	43	5	2	133	367	51	12
<i>Tabanus briani</i> Lec.	-	-	2	-	-	-	-	-	1	-	-	-	-	-	-
<i>Tabanus bifarius</i> Loew.	-	47	26	9	-	-	11	5	1	-	-	268	26	2	-
<i>Tabanus autumnalis</i> L.	-	33	11	-	-	-	-	5	-	-	-	6	5	-	-
<i>Tabanus armeniacus</i> Kröb.	-	-	1	-	-	-	-	-	-	-	-	-	3	-	-
<i>Tabanus cordiger</i> Meig.	-	14	-	1	-	-	11	4	-	-	-	6	4	-	-
<i>Tabanus cuculus</i> Szi.	-	-	2	1	-	-	-	-	1	-	-	-	1	1	-
<i>Tabanus eggeri</i> Sch.	-	-	2	-	-	-	-	-	-	-	-	-	1	-	-
<i>Tabanus exclusus</i> Pand.	-	4	27	9	-	-	1	7	8	-	-	6	11	1	-
<i>Tabanus glaucopsis</i> Meig.	-	-	10	17	1	-	1	12	11	2	-	-	7	13	3
<i>Tabanus golovi</i> Ols.	-	-	-	1	-	-	-	1	-	-	-	-	2	-	-
<i>Tabanus indrae</i> Haus.	-	-	1	1	-	-	-	2	3	-	-	-	3	-	-
<i>Tabanus laetinctus</i> Bec.	-	-	2	-	-	-	1	-	-	-	-	-	1	-	-
<i>Tabanus leleani</i> Aust.	-	2	-	-	-	-	5	7	-	-	-	4	1	-	-
<i>Tabanus lunatus</i> Fabr.	-	4	37	3	-	-	1	22	14	2	-	10	138	11	1
<i>Tabanus maculicornis</i> Zet.	-	3	2	-	-	-	-	1	-	-	-	-	4	1	-
<i>Tabanus martini</i> Kröb.	-	-	1	8	1	-	-	-	3	2	-	-	-	7	1
<i>Tabanus miki</i> Brau.	-	-	18	4	-	-	1	5	2	-	-	8	11	8	-
<i>Tabanus portschinskii</i> Ols.	-	-	28	13	-	-	-	14	9	-	-	-	39	6	-
<i>Tabanus prometheus</i> Szi.	-	-	7	-	-	-	-	3	-	-	-	-	16	-	-
<i>Tabanus quatuorotatus</i> Meig.	9	379	9	-	-	7	118	3	-	-	3	77	5	-	-
<i>Tabanus regularis</i> Jaen.	-	-	-	1	-	-	-	-	4	-	-	-	-	7	-
<i>Tabanus rupium</i> Brau.	-	38	7	-	-	-	11	13	-	-	-	28	3	-	-
<i>Tabanus spodopteroides</i> O.M.C.	-	-	2	-	-	-	-	3	-	-	-	-	1	-	-
<i>Tabanus spodopterus</i> Meig.	-	-	27	3	-	-	-	4	3	-	-	-	11	2	-
<i>Tabanus sudeticus</i> Zel.	-	3	4	1	-	-	2	-	-	-	-	-	-	2	-
<i>Tabanus spectabilis</i> Loew	-	-	1	-	-	-	-	2	-	-	-	-	-	-	-
<i>Tabanus tergestinus</i> Egg.	-	-	7	-	-	-	-	2	-	-	-	-	4	-	-
<i>Tabanus terterjani</i> Dol. & And.	-	-	-	-	-	-	-	-	-	-	-	-	-	3	-
<i>Tabanus tinctus</i> Wal.	-	-	4	-	-	-	-	1	-	-	-	-	2	-	-
<i>Tabanus unifasciatus</i> Loew.	4	141	37	7	3	3	14	29	13	4	3	67	-	4	11
<i>Theriopectes tricolor</i> Zel.	-	13	-	-	-	-	-	9	-	-	-	4	-	-	-
<i>Philipomyia zizaniae</i> Leg.	-	-	-	-	-	-	-	-	-	-	-	1	7	-	-
<i>Philipomyia aprica</i> Meig.	-	13	201	26	4	-	13	38	12	2	5	259	483	11	-
<i>Atylotus fulvus</i> Meig.	-	-	24	9	-	-	-	-	-	-	-	13	7	-	-
<i>Chrysops caecutiens</i> L.	-	2	13	2	-	-	-	4	1	-	-	17	15	2	-
<i>Chrysops viduatus</i> Fab.	-	2	3	1	-	-	-	1	-	-	-	3	4	1	-
<i>Dasyrhamphis carbonarius</i> Meig.	-	1	-	-	-	-	3	-	-	-	-	1	-	-	-
<i>Dasyrhamphis umbrinus</i> Meig.	11	155	21	-	-	8	21	-	-	-	8	119	-	-	-
<i>Haematopota bigoti</i> Gob.	-	3	1	-	-	-	-	-	-	-	-	-	2	-	-

Table 1. Continued

<i>Haematopota crassicornis</i> Wah	-	2	3	-	-	-	-	1	-	-	-	-	2	-	-
<i>Haematopota grandis</i> Meig.	-	-	1	1	-	-	-	1	-	-	-	-	1	-	-
<i>Haematopota longantennata</i> Ols.	-	-	1	-	-	-	-	1	-	-	-	-	1	-	-
<i>Haematopota ocelligera</i> Kröb.	-	6	-	-	-	-	5	-	-	-	-	1	-	-	-
<i>Haematopota pluvialis</i> L.	-	19	11	-	-	-	15	9	-	-	-	4	-	-	-
<i>Haematopota scutellata</i> O.M.C.	-	7	-	-	-	-	5	-	-	-	-	2	-	-	-
<i>Haematopota subcylindrica</i> Pand.	11	433	681	57	10	7	66	63	31	8	14	118	211	5	-
<i>Hybomitra caucasica</i> End.	6	4	-	-	-	7	3	-	-	-	9	2	-	-	-
<i>Hybomitra ciureai</i> Seg.	-	-	3	-	-	-	-	-	-	-	-	-	2	-	-
<i>Hybomitra pilosa</i> Loew.	-	4	-	-	-	2	-	-	-	-	-	3	-	-	-
Total			3572					934					2771		

3.1. Lead

Correlation coefficient data show that the lead concentration had significant effects on the populations of *T. bifarius* ($T_1 = -0.482$ and $p = 0.012$) (Fig. 1.a), *T. bromius* ($T_1 = -0.310$ $p = 0.0008$) (Fig. 1.b), *T. unifasciatus* ($T_1 = -0.411$ $p = 0.0008$) (Fig. 1.c), *T. quatuornotatus* ($T_1 = -0.505$ $p = 0.012$) (Fig. 1.d) and *T. rupium* ($T_1 = -0.392$ $p = 0.012$) (Fig. 1.e). These significance levels were lower than the α/k value (0.013) of the sequential Bonferroni test and hence the lead concentrations have negative effect on the population dynamics of these species. On the other hand, correlation coefficient data show that lead concentrations had no effects on the population dynamics of *T. portschinskii*, *D. umbrinus*, *P. aprica*, *H. pluvialis* and *H. subcylindrica*. These differences caused by the habitat preferences of species. Larvae of *T. bifarius*, *T. bromius*, *T. unifasciatus*, *T. quatuornotatus* and *T. rupium* are hemihydrobiont and concentration of lead in streams are higher than soil in the study area, hence lead pollution has important effect on the population dynamics of these species. But larvae of *T. portschinskii*, *D. umbrinus*, *P. aprica*, *H. pluvialis* and *H. subcylindrica* edaphebiont, and larval duration occurs terrestrial or semiaquatic places, hence lead pollution has negative effects on the population dynamics of these species, but the effects are not statistically important.

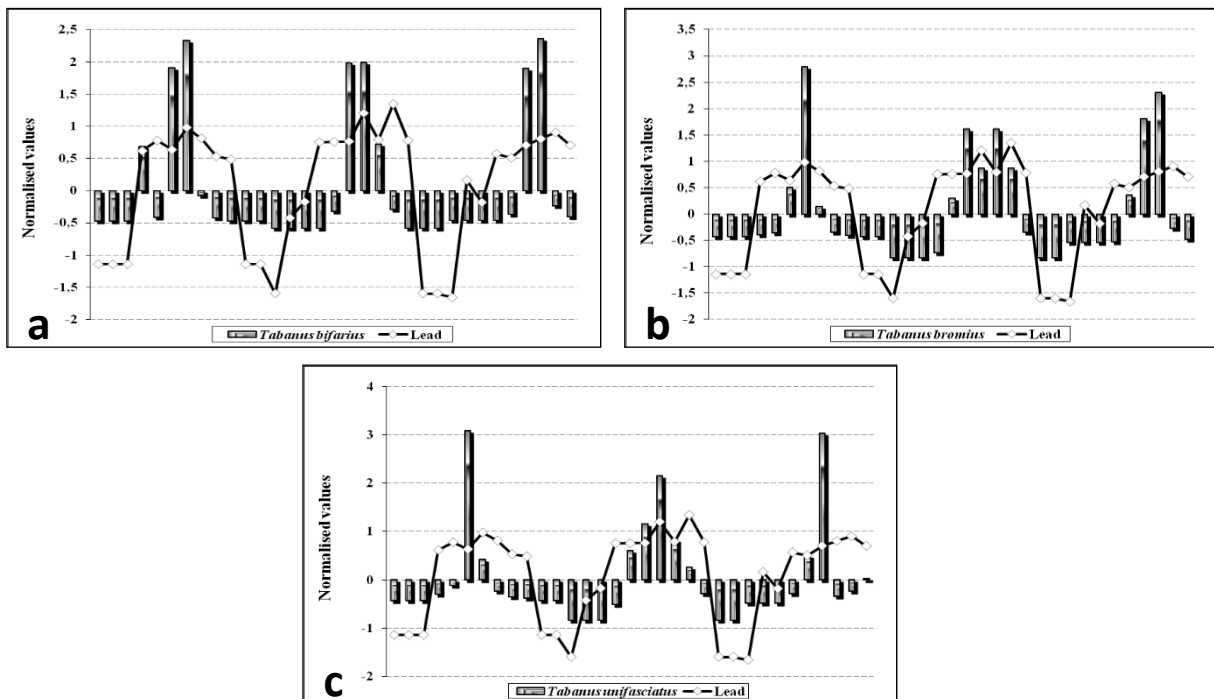


Figure 1. Effects of the lead on the population dynamics of *Tabanus bifarius* (a); *Tabanus bromius* (b); *Tabanus unifasciatus* (c)

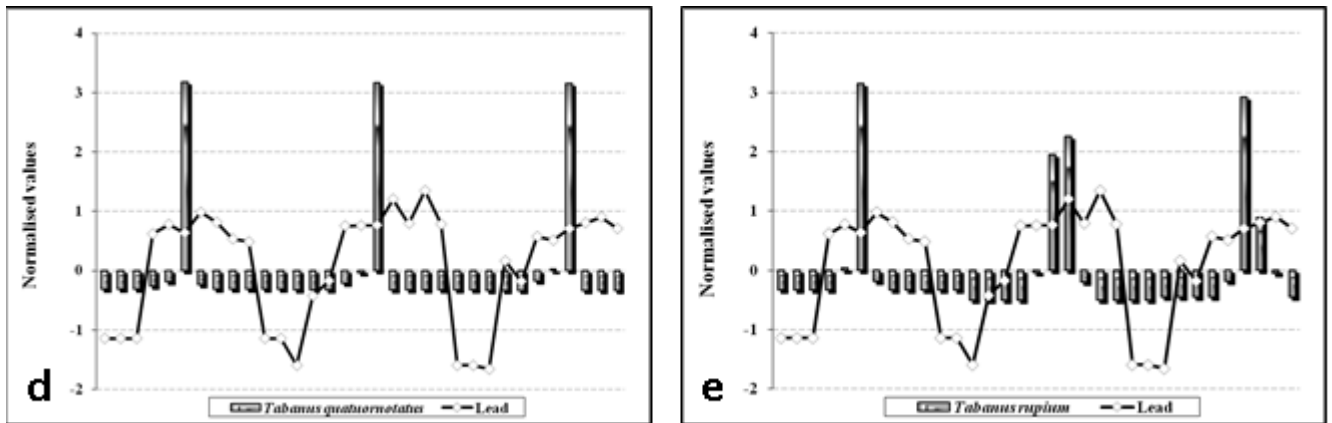


Figure 1. Effects of the lead on the population dynamics of *Tabanus quatuornotatus* (d); *Tabanus rupium* (e)

3.2. Iron (Ferric)

Iron has also negative effect on the population dynamics of species, and correlations between iron and *T. bifarius* ($T1 = -0.294$ $p = 0.012$) (Fig. 2.a); *T. bromius* ($T1 = -0.412$ $p = 0.005$) (Fig. 2.b); *T. quatuornotatus* ($T1 = -0.365$ $p = 0.0008$) (Fig. 2.c); *T. rupium* ($T1 = -0.488$ $p = 0.005$) (Fig. 2.d); *T. unifasciatus* ($T1 = -0.206$ $p = 0.0008$) (Fig. 2.e), and *H. subcylindrica* ($T1 = -0.266$ $p = 0.005$) (Fig. 2.f) are statistically important.

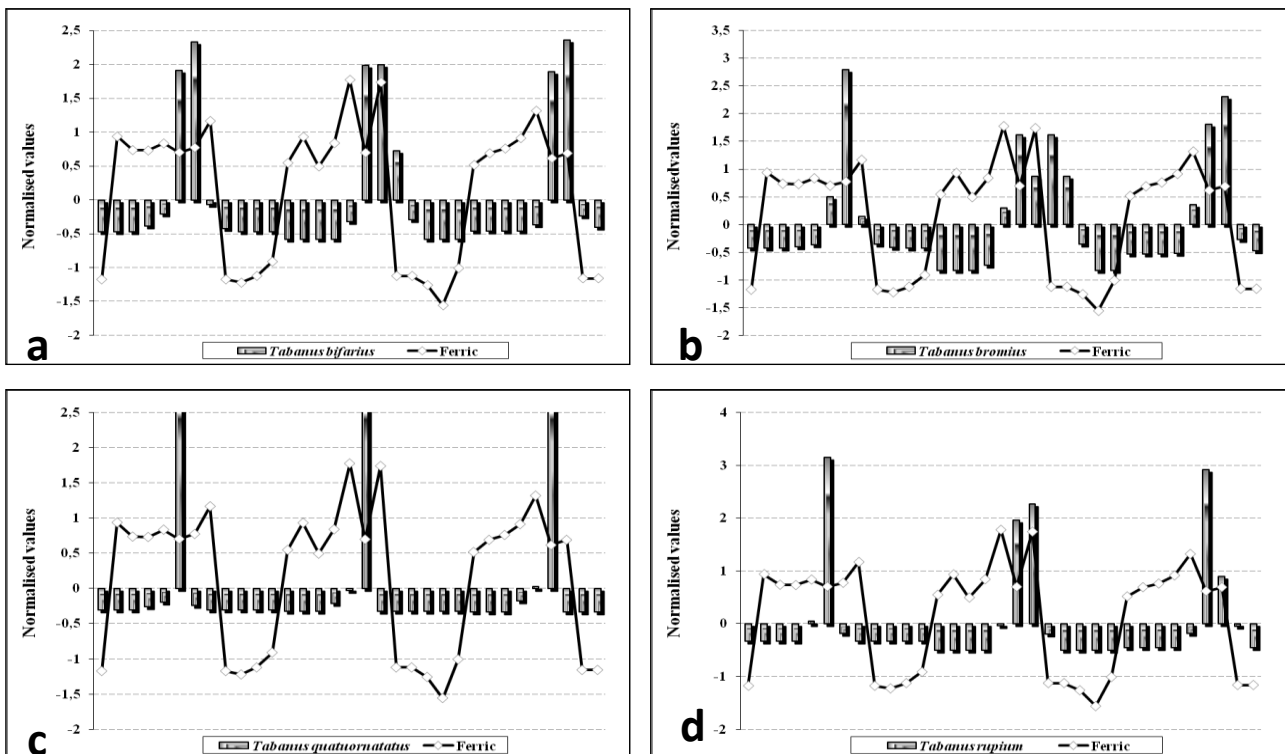


Figure 2. Effects of the iron on the population dynamics of *Tabanus bifarius* (a); *Tabanus bromius* (b); *Tabanus quatuornotatus* (c); *Tabanus rupium* (d)

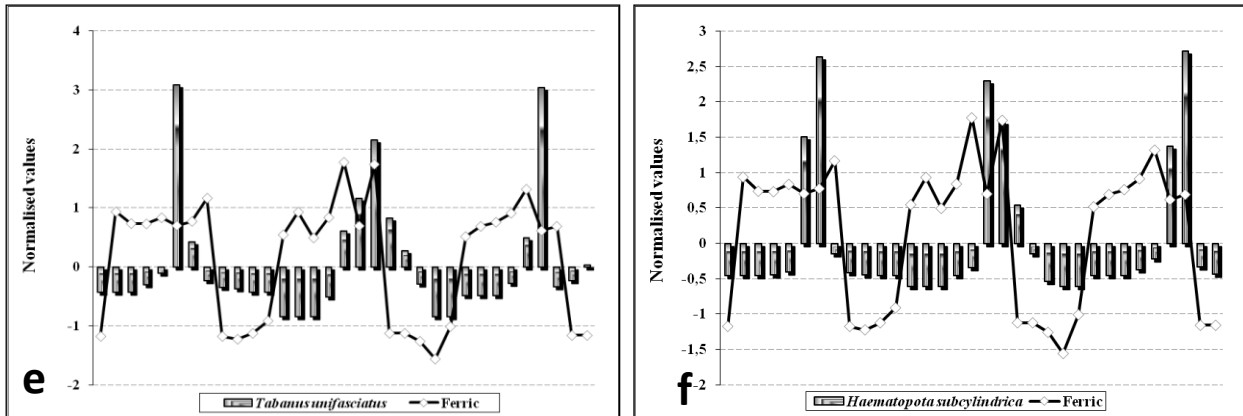


Figure 2. Effects of the iron on the population dynamics of *Tabanus unifasciatus* (e); *Haematopota subcylindrica* (f)

3.3. Cadmium

Comparing with iron and lead, less amounts of cadmium accumulation were detected. Due to the toxicity level of cadmium, it has further negative effects on the population dynamics of all species. However, correlations between cadmium and *T. bifarius* ($T1 = -0.311$ $p = 0.0012$) (Fig. 3.a), *T. bromius* ($T1 = -0.304$ $p = 0.0012$) (Fig. 3.b) and *T. unifasciatus* ($T1 = -0.403$ $p = 0.0005$) (Fig. 3.c) were statistically important.

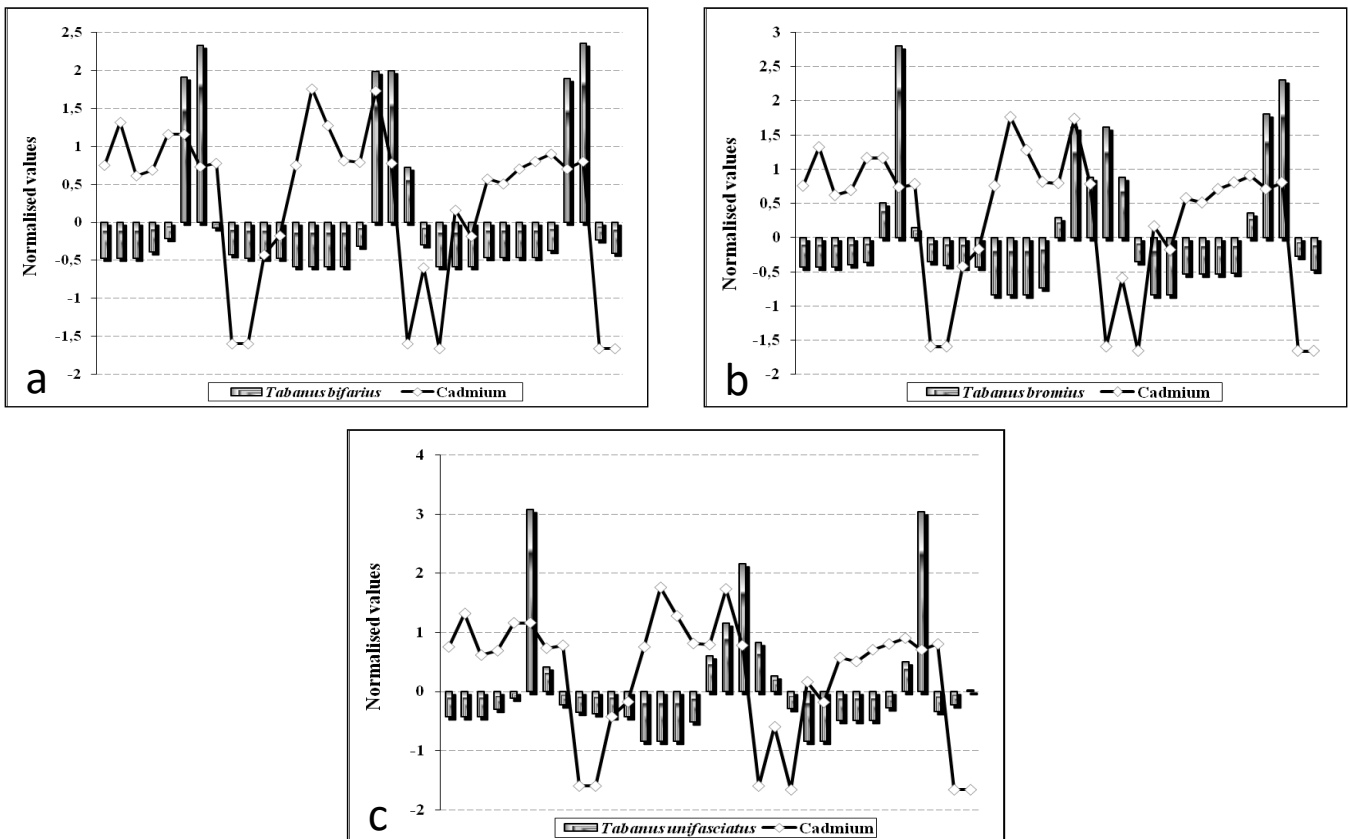


Figure 3. Effects of the cadmium on the population dynamics of *Tabanus bifarius* (a); *Tabanus bromius* (b); *Tabanus unifasciatus* (c)

4. Conclusions and discussion

Toxic heavy metal presence in water, air and soil is known as a serious problem that increasingly threatens the environment. There are many sources of heavy metal pollution, consisting of the chlor-alkali industries, natural gas, coal, paper, and dye industries [23]. Nevertheless, if heavy metal pollution exponentially grows over the years, the life quality of many species are affected irreversibly.

Heavy metal pollution can affect population dynamics of Tabanidae species as well as other groups. In this study, annual increase on accumulation of Lead (Pb), Iron (Fe), Cadmium (Cd), Mangan (Mn) and Aluminium (Al) were investigated in the samples of soil and stream. All toxicants have negative impacts on the population dynamics of Tabanidae species. Due to the accelerating concentrations and higher toxic effect, however, correlations between only lead, iron and cadmium were statistically important.

The many authors have investigated the effects of Zn, Pb, Cd and Cu on the population dynamics and diversity of different species and reported the negative impressions of stated heavy metals on the life cycles of studied populations [24, 25]. Hladun et al. [19] investigated the effect of soil-borne pollutants like selenium, methionine and elenomethionine on the honey bee and concluded that contaminated soil and plants have negative impact on the insects. Azam et al. [26] stated that Cr, Cu, Cd, Zn, and Ni in the water, soil and air can affect the species. They also determined that the various insect groups are potential indicators of heavy metal pollution and they may be used as bioindicator species. they conducted a study on heavy metal pollution in *Salicornia europaea* L. growing in wetlands and found that there was an increase in the accumulation of Pb and Zn. These heavy metals accumulated in water, soil and plants negatively affect both the horseflies, whose larvae are aquatic, and the ecosystem. Ay et al. [27] conducted a study on heavy metal pollution in *Salicornia europaea* L. growing in wetlands and found that increasing in the accumulation of Pb and Zn. These heavy metals accumulated in water, soil and plants and negatively affect both the horseflies, whose larvae are aquatic, and the ecosystem. Many stresses that are connected with all biological processes on the ecosystems and the living organisms are a hazard for pollinator welfare, diversity, and abundance. In addition to pollinators these pressures affect all species step by step, decreasing pollinators cause to decrease in plant species and decreasing the plant species also causes ecosystem degradation. It is urgent to restrict the negative compulsions of ongoing pollinator decreases for ecological services, human health and agricultural production. The present study which is the first attempt for Tabanidae in Türkiye, clearly emphasizes the requirement of adequate studies about the impacts of heavy metals on population dynamics in the field of observations on population ecology of Tabanidae species. Moreover, in the everchanging global world, the pollinator insects which are necessary for the ecosystem and their habitats should be protected from any human activities.

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