Journal of Environmental Studies
Vol. 48, No. 1, Spring 2022
Journal Homepage: www.Jes.ut.ac.ir
Print ISSN: 1025-8620 Online ISSN 2345-6922

The Effect of Heavy Metals and Petroleum Hydrocarbons on the Collembolan Fauna in Soil of Some Industrial Regions of Isfahan

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DOI: 10.22059/JES.2022.328017.1008209

Document Type
Research Paper

Received
November 26, 2021

Accepted
April 21, 2022

Abstract
This research was aimed to investigate the effects of heavy metals and petroleum hydrocarbons contaminants in Isfahan Refinery and Mobarake Steel factory soils on diversity of Collembolan Arthropods. The concentration of heavy metals and petroleum hydrocarbons were measured using atomic absorption and Soxhlet methods, respectively. Soil Collembola were collected using burl funnel, and finally, eleven species belonging to 6 families from 2 suborder of collembola were identified. In heavy metals contaminated soils, three species including *Isotomiella minor*, *Ceratophysella stercoraria* and *Hypogastura sp.1*, have the highest percentages of frequency respectively. As well as, the highest frequency of collembola in contaminated soil with petroleum hydrocarbons were related to *Hypogastura sp.*, *Isotomiella minor* and *Ceratophysella stercoraria* species respectively. In this research, the *Folsomia candida* showed highly susceptibility against soil pollutions, was not collected from heavy metals and oil hydrocarbons contaminated soils and can be used as a biomarker. This is the first report of collembolan fauna in soils containing environmental pollutants in Iran.

Keywords: Biomarkers, Environmental pollutions, Oil hydrocarbons, Cadmium

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Extended Summary

Introduction

Soil pollution and accumulation of heavy metals such as lead, cadmium, silver and mercury in agricultural products of industrial areas is one of the most important environmental issues that threaten the lives of plants, animals and humans. Harmful effects of these pollutants on living organisms, including disruption of biological activities, adverse effects on plants and humans due to their entry into the food chain have been proven. As well as, oil pollution is one of the most common of pollution in terrestrial and aquatic ecosystems. The development of the petrochemical industry and the non-observance of environmental requirements, have led to the introduction of large amounts of hydrocarbon pollutants into the environment in recent decades.

Biomarkers include biological processes, species, or communities of living organisms that are used to assess the quality of the environment and how the environment changes over time. In recent years, the use of biomarkers to identify environmental pollutants, especially heavy metals and petroleum hydrocarbons, has increased. Collembolan arthropods are one of the most important soil decomposers with high species diversity and density, especially in forest ecosystems. These organisms are widely used organisms as biomarkers due to their short life cycle, high density and high susceptibility to various contaminants in terrestrial ecosystems. This research was aimed to identify the collembola fauna in soils contaminated with heavy metals and petroleum hydrocarbons in two industrial areas of Isfahan and to investigate the impact of these contaminants on the diversity and population abundance of these organisms as a biological indicator of soil contamination.

Materials and Methods

Soil samples were taken from the lands around Isfahan Oil Refinery and the area around Mobarakeh Steel factory, as contaminated areas to petroleum hydrocarbons and heavy metals respectively. Also, a sample of pristine soil (non-contaminated soil) was taken from the test areas as a control. Heavy metals concentration was measured by atomic absorption method and the amount of petroleum hydrocarbons in the samples was measured by Soxhlet method. The terrestrial arthropods were collected using Berlese funnel. To identify the Collembola species, microscopic slides were prepared and identified according to the taxonomic keys.

Discussion of Results

Diversity of Collembola in contaminated soils

The results of chemical analysis of soils showed that there were different degrees of contamination with petroleum hydrocarbons and heavy metals including lead, cadmium and nickel in the sampled areas, and the soil of the control area was free of any contamination. The highest concentration of heavy metal in the studied soils was related to cadmium with 21.92 mg/kg and the lowest concentration of nickel was related to 42.85 mg/kg. Sampling was performed to collect collembolan arthropods throughout the year and a total of 11 species of collembola from 6 families and 2 suborders were collected (table 1).

As shown in Figure 1a, *Istomiella minor*, *Ceratophysella stercoraria*, *Hypogastrura sp.1*, *Entomobrya sp.1* and *Onychiurus sp.2* species are more abundant in heavy metal contaminated area than uncontaminated soil (control). Also, *Folsomia candida* was not found in the soil samples around the steel factory. On the other, the diversity and abundance of Collembola species collected in soils contaminated with petroleum compounds around the oil refinery were similar to soils contaminated with heavy metals (Figure 1-b). In these soils, the highest abundance of collembolan communication is related to *Hypogastura sp.1*, *I. minor* and *C. stercoraria*, respectively. Also, the frequency percentage of *F. candida*, *Pseudosinella sp.2* and O*nychurus sp.1* was zero and no samples of these species were collected, which indicates the high sensitivity of these species to petroleum products and hydrocarbon compounds.

Sampling was performed to isolate terrestrial Collembola in different seasons of the year. The results showed that these arthropods were active only in spring and summer. In both autumn and winter, no samples of Collembola were collected in either of the two areas contaminated with heavy metals and petroleum compounds. The interesting point in this study was that no significant difference was observed between the collembola population of contaminated soils and non-contaminated soils in spring.

However, in summer, the population of non-contaminated soils was much higher than soils contaminated with heavy metals and petroleum compounds and a significant difference ($p \leq 0.05$) was observed between them.

Table 1- Collembola species collected from heavy metals and petroleum hydrocarbons contaminated soils

No.	Suborder	Family	Species	Heavy metals	petroleum hydrocarbons	Non contaminated soil
1	Entomobryomorpha	Isotomidae	*Isotoma sp.*	+†	+	+
2	Poduromorpha	Hypogastruridae	*Hypogastrura sp.1*	+	+	+
3	Entomobryomorpha	Entomobyridae	*Pseudosinella octopunctata*	+	+	+
4	Entomobryomorpha	Entomobyridae	*Entomobrya sp.1*	+	+	+
5	Entomobryomorpha	Isotomidae	*Isotomiella minor*	+	+	+
6	Poduromorpha	Onychiuridae	*Onychiurus sp.2*	+	+	+
7	Poduromorpha	Onychiuridae	*Onychiurus sp.1*	+	-	+
8	Poduromorpha	Hypogastruridae	*Ceratophysella stercoraria*	+	+	+
9	Entomobryomorpha	Entomobyridae	*Pseudosinella sp.2*	+	-	+
10	Poduromorpha	Tullbergidae	*Mesaphorura sp.1*	+	+	+
11	Entomobryomorpha	Poduridae	*Folsomia candida*	-*	-	+

†: The species was collected from soil sample
*: The species was not collected from soil sample

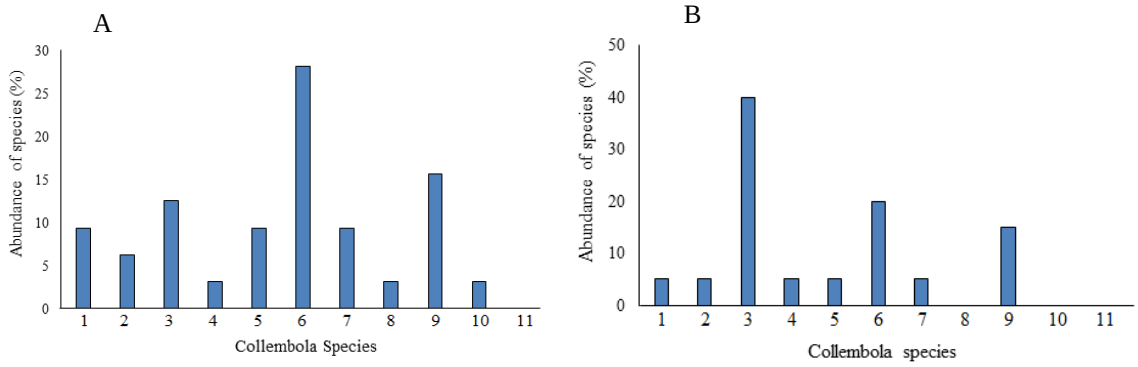

Fig. 1: Abundance percentage of Collembola species collected from heavy metals (A) and petroleum hydrocarbons (B) contaminated soils. 1: *Mesaphorura sp.1*; 2: *Isotoma sp.1*; 3: *Hypogastrura sp.1*; 4: *Pseudosinella octopunctata*; 5: *Entomobrya sp.1*; 6: *Onychiurus sp.1*; 7: *Isotomiella minor*; 8: *Onychiurus sp.2*; 9: *Ceratophysella stercoraria*; 10: *Pseudosinella sp.2*; 11: *Folsomia candida*.

As shown in Figure 2, the frequency of *Hypogastrura sp.1* and *Pseudosinella octoppuncta* species in oil-contaminated soils is higher than their frequency in heavy metal-contaminated soils. The frequency of *Hypogastrura sp.1* in oil-contaminated soils (40%) was significantly higher ($p \leq 0.05$) than the frequency of this species in heavy metal-contaminated soils (12.5%) (Fig. 2). Therefore, *Hypogastrura sp.1* is the most resistant species to petroleum products among the species studied in this research. Also, the *I. minor* showed the highest frequency (28.13%) in soils contaminated with heavy metals. Therefore, this species is the most resistant to heavy metals among the studied species.

The main purpose of this study was to compare the Collembola fauna in soils contaminated with heavy metals and petroleum compounds as a biological indicator in the some industrial regions of Isfahan. In this study, a total of 11 species belonging to 6 families from 2 suborders of Collembola were collected. *F. candida* was not isolated from contaminated soils with heavy metals, while this species has acceptable population abundance in pristine and non-polluted soils. These results indicate the high sensitivity of this species to heavy metals. Lead and cadmium have very destructive effects on the reproduction of *F.*

candida and cause a significant reduction in reproduction of this species. According to Sandifer and Hopkin (1996), when the cadmium concentration in the soil reaches to 1.2 µg / kg, the reproduction of *Folsomia* species decreases dramatically. The soils studied in this study were also highly contaminated with cadmium and the concentration of this element in some samples was 21.92 mg / kg, which is several hundred times than the global pollution standard, this is why *F. candida* was not collected from these soils.

Also the species *F. candida*, *Pseudosinella sp.2* and *Onchiusus sp.1* were not collected from soils contaminated with petroleum hydrocarbons. Studies by Fountain and Hopkin (2005) show the very high susceptibility of these species, especially *F. candida*, to chemical contaminants and introduce this species as a reliable biomarker against soil pollutants. In contrast, populations of some species such as *I. minor*, *C. stercoraria* and *Hypogastrura sp.1* was higher in soils contaminated with heavy metals and petroleum compounds than in non-contaminated soils. These results may be due to the fact that some species of Collembola have the ability to detoxify and remove of poisonous chemicals from their bodies with repeated molting.

Conclusions

The genus *Folsomia* is one of the most well-known soil collembolan which whose species, due to non-migratory living and its low dependence on sub-habitats, make them important organisms for determining air and soil quality and tracking pollution. Among the identified species of this genus, *F. candida* has been considered as an ecological criterion and biological indicator of soil. In this study, this species was not isolated in soils contaminated with heavy metals and petroleum compounds and was introduced as the most sensitive species to chemicals. On the other hand, *I. minor* with a population frequency of about 25% in the studied soils was introduced as the most resistant species to chemical compounds.