Determination of PCDDs in spider webs: preliminary studies

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Abstract. The application of spider webs for determination of polichlorinated dibenzo-*para*-dioxins (PCDDs) has been studied for the first time. The aim of the studies was to find out if spider webs are suitable for such examinations as it was proved in the previous research they are excellent indicators of air pollutants. Spiders are ubiquitous, thus collection of samples is easy and non-invasive. Studies were conducted within the city of Wrocław and surroundings, one of the biggest and at the same time heaviest polluted city in Poland. Five research sites have been chosen, where spider webs were collected after 60 days of continuous exposure time. Webs belonging to two genera *Tegenaria sylvestris* and *Tegenaria ferruginea* (family Agelenidae) have been chosen as they are large and very dense, thus they are very suitable for such examinations. Webs were found to retain dioxins probably mainly by external exposure. These promising results should be continued and expanded in the future research.

1 Introduction

Air pollutants make people sick, causing breathing problems and increasing the likelihood of cancer [1]. Polichlorinated dibenzo-*para*-dioxins (PCDDs) belong to the group of chlorinated polycyclic aromatic hydrocarbons which are persistent environmental contaminants and as a result they are hazardous to the environment because of their toxic, mutagenic and carcinogenic character. They are by-products of industrial and combustion processes or industrial compounds [2].

Environmental exposure to dioxins contamination has caused health concerns as tetrachloro dibenzo-*p*-dioxin (TCDD) was classified as carcinogenic to humans by the International Agency for Research on Cancer [2, 3]. PCDDs have been identified as residues in hair, wool and human milk [4]. Spider webs have never been studied. They proved to be perfect indicators of air pollutants as web samples are easily collected from different areas. They are widely available and they are usually woven in secluded places preventing them from destroying by weather conditions. It is the universal, organic, environment friendly tool as webs do not need any preparation before exposition to pollutants and do not need to be degraded. Also the exact time of exposition to pollutants can be easily controlled as they can be obtained from rearing of spiders under laboratory conditions. Spider webs were

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successfully used to evaluate heavy metals and trace element pollution as well as polycyclic aromatic hydrocarbons (PAHs) content in the air [5,6].
The aim of this study was to find out if the spider webs are suitable to determine the environmental exposure to PCDDs.

2 Material and Methods

2.1 Material

The studies were conducted in 2012 at five sites in Wrocław (Fig. 1.). Genera of *Tegenaria ferruginea* (Panzer, 1804) and *Tegenaria silvestris* (L. Koch, 1872) from Agelenidae family have been chosen for studies as their web is very dense and large. *T. ferruginea* occurs in hedgerows, fences, tree crevices and on walls of buildings. Males are mature in summer, whereas females are present all year [7]. *T. silvestris* is usually associated with woodland, but occasionaly it can be found in fences or even houses where it weaves sheet webs. Adults are found most of the year [7]. Spiders belonging to Agelenidae do not eat their own webs when destroyed as such behaviour could influence the further pollution level in the web.
Samples were collected from secluded locations, from buildings and walls. All sites were protected from weather conditions e.g. severe rain. Webs were collected after 60 days from the creation of the new construction (after the removal an old web if such previously existed). Webs of similar sizes, age, and weight were used for analyses [8].

2.2 Sampling preparation

Sample preparation and analysis of PCDDs contained in the samples were made in the Chemistry Department of University of Wrocław and University of Environmental and Life Sciences in Wrocław. Webs were extracted with n-hexane/acetone (50/50, v/v) in a Soxhlet apparatus. The extract was concentrated on the rotary evaporator. Samples were analysed for the 7 PCDD EPA-designated toxic congeners with high resolution gas chromatograph/high resolution mass spectrometer (HRGC/HRMS) following EPA Method 8290 [9]. The recovery of analytes was calculated on the basis of stable isotopically labeled $_{13}$C-PCDDs which was introduced into samples of spider webs after extraction to measure the recovery. Mean recoveries ranged from 53% to 87%, depending on the congener. We calculated the TEQ in spider webs using World Health Organization TEFs.

2.3 Sampling sites

Five sites have been chosen for studies according to the pollution level.

Site 1.
51°04’46.6” N, 17°05’16.5”E Located in the south-east part of the city where tenement houses with domestic solid fuel furnaces predominate. The inhabitants also burn trash occasionally. The area lies distantly from the main communications trails. Spider silk was collected from buildings.
Site 2.
51°06’03.2” N, 17°05’40.9”E Biskupin, the housing estate which is situated in the eastern part of Wrocław, in Śródmieście district near Odra River. Heating system differs according to the wealth of inhabitants. Rarely houses still are still equipped with old furnaces where inhabitants use not only coal as a fuel, but occasionally burn trash. Surroundings are

diversified including areas of allotments, parks, high and low buildings. Spider silk was collected from building walls.

Site 3.

51°06'40.7" N, 17°03'30.7"E Grunwaldzki Square– the city centre in the south east part of Wrocław, one of the main and most important place, main communication node with high traffic load day and night. Trucks and other vehicles cross this section as the ring road of Wrocław is still missing. Webs were collected from walls of the parking neighbouring with main road.

Site 4

51°05'03.8" N, 17°01'18.2"E Fences and hedges along Wiśniowa Avenue, one of the most polluted are in Wrocław, because of heavy traffic which is constantly monitored by local pollution agency. Webs were collected from hedges surrounding the street.

Site 5.

51°31'04.6" N, 17°15'42.3"E Quiet rural area situated near the Landscape Nature Park "Dolina Baryczy" where medical and veterinary solid waste incinerator was built by ECO ABC company. Company is relying on the old equipment and is still working despite inhabitants complaints and severe protests as it burns the waste exceeding recommended norms and standards. Spider silk was collected from hedges and bushes at distance up to 500 m from incinerator.

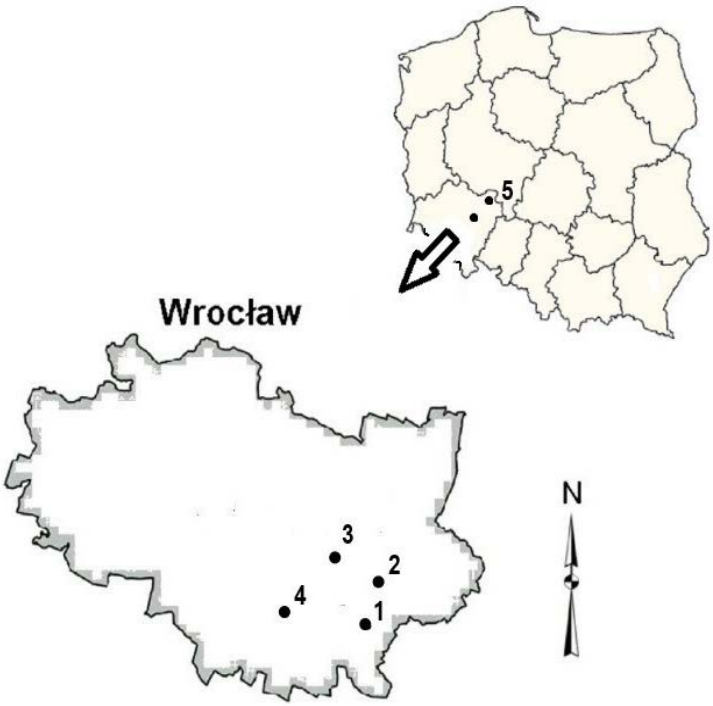

Fig. 1. The map of study area.

3 Results

We found measurable PCDD in spider webs at all sites. All congeners were present at levels above the LOD (limit of detection) at studied sites (Table 1 and 2). The most toxic congeners, 2,3,7,8-TCDD and 1,2,3,7,8-P_5CDD (TEFs=1), had concentrations of 6.89 pg/g and 10.88 pg/g, at sites 4 and 5 and 7.23 and 24.9 at sites 1 and 5, respectively. At site 5 the most abundant congeners were 2,3,7,8-TCDD and 1,2,3,7,8-P_5CDD, contributing 30.2%, and 67.3% to the TEQ. At site 1 the most abundant congeners were 1,2,3,7,8-P_5CDD and 1,2,3,7,8,9-H_6CDD contributing 39.9%, and 19.2% to the TEQ. At site 2 the most abundant congeners were 1,2,3,7,8-P_5CDD and 1,2,3,7,8,9-H_6CDD, contributing 36.7%, and 20.8% to the TEQ. At site 3 the most abundant congeners were 1,2,3,7,8-P_5CDD and 1,2,3,6,7,8-H_6CDD, contributing 28%, and 24.1% to the TEQ. At site 4 the most abundant congeners were 2,3,7,8-TCDD and 1,2,3,7,8-P_5CDD, contributing 45.2%, and 41.1% to the TEQ. The web samples from site 5 had the total TEQ (36.95 pg/g).

Table 1. Concentration of PCDD in spider webs at sites 1, 2 and 3 (pg/g).

Isomers	TEF	Site 1	TEQ (pg/g)	Site 2	TEQ (pg/g)	Site 3	TEQ (pg/g)
2,3,7,8-TCDD	1	2.21	2.21	1.05	1.05	6.02	6.02
1,2,3,7,8-P5CDD	1	7.23	7.23	3.21	3.21	8.6	8.6
1,2,3,4,7,8-H6CDD	0.1	14.3	1.45	7.54	0.75	27.4	2.74
1,2,3,6,7,8,-H6CDD	0.1	33.7	3.37	15.6	1.56	74.1	7.41
1,2,3,7,8,9-H6CDD	0.1	34.5	3.47	18.2	1.82	49.7	4.97
1,2,3,4,6,7,8-H7CDD	0.01	35.9	0.36	28.3	0.28	89.8	0.89
OCDD	0.0003	223	0.06	210	0.06	181	0.05
Total TEQ	-	-	18.09	-	8.73	-	30.68

Table 2. Concentration of PCDD in spider webs at sites 4 and 5 (pg/g).

Isomers	TEF	Site 4	TEQ (pg/g)	Site 5	TEQ (pg/g)
2,3,7,8-TCDD	1	6.89	6.89	10.88	10.88
1,2,3,7,8-P5CDD	1	6.27	6.27	24.9	24.9
1,2,3,4,7,8-H6CDD	0.1	23.8	0.24	63.4	0,63
1,2,3,6,7,8,-H6CDD	0.1	67	0,67	114	0,11
1,2,3,7,8,9-H6CDD	0.1	46.9	0,47	110	0,11
1,2,3,4,6,7,8-H7CDD	0.01	66.5	0,66	234	0,23
OCDD	0.0003	172	0,05	311	0.09
Total TEQ	-	-	15.24	-	36.95

4 Discussion

This is the first study to examine concentrations of PCDDs in spider webs in different sites across Lower Silesia region in Poland. PCDDs were detectable in spider webs, and we noticed relationships between high values of PCDDs and certain sites. Proximity to medical solid waste incinerator at site 5 was associated with highest concentrations of PCDDs (total TEQ 36.95 pg/g). Proximity to major roadways (site 3) was also associated with higher concentrations of PCDDs (total TEQ 30.68 pg/g). Sites 1 and 3 had lower values of PCDDs but not very low which is probably connected with the proximity of major roadways (site 4; total TEQ 15.24 pg/g) and possibility of trash burning at the houses (site 1; total TEQ 18.09 pg/g). The lowest values of PCDDs were obtained in spiders webs collected at site 2 (total TEQ 8.73 pg/g) where trash burning at houses occurs only occasionally. However, results of these analyses are based on small numbers of samples and need to be explored in a larger number of spider webs at studied sites.

What is interesting, other studies [10] did not observe the association between PCDD in dust (spider webs have never been analysed) and proximity to medical waste incinerators, because such incinerators are usually small, thus emission level is rather low. In our studies site 5 was characterised by elevated level of PCDDs which is probably related with the proximity of incinerator, but on the other hand samples at site 5 were collected in the rural area where trash burning and heating with coal or waste are usually applied by inhabitants, what is more, the backyard trash burning and agriculture burning are also very popular. These emissions could significantly contribute to the obtained results of PCDD concentration.

The emissions of PCDD from petrol and diesel powered vehicles were well documented in the past (sites 3 and 4) as well as the existing relationship between trash burning and elevated dioxins levels (sites 1 and 2) [10, 11].

TCDD is perceived as a human carcinogen according to the International Agency for Research of Cancer. Many studies have demonstrated that exposure to dioxins cause various diseases including endocrine disruption, reproductive problems, neurotoxicity and cancers [12]. The obtained concentrations of PCDDs suggest the possible humans' exposure as they are probably exposed to resuspended, contaminated dust which is was found in spider webs. Because webs showed elevated levels of dioxins there is the evidence that inhabitants have been and are exposed to dust containing dioxins.

5 Conclusion

The spider webs were found to absorb dioxins from ambient air. The recorded high concentrations of PCDDs were highly combustion oriented.The highest values were recorded near the medical solid waste incinerator (site 5) which is not a surprise. The high content of PCDDs was also related with personal furnaces and vehicle emissions. The presented prelimnary results proved that webs can serve as an effective, natural tool for the indication of PCDDs contained in silk. Compared with studies based on other bioindicators such as mosses, lichens and vascular plants research based on spider webs are rare. Especially, webs of Agelenidae show excellent accumulative properties which is a result of their high density, and large size comparing to the webs of other spider families. What is more, representatives of Agelenide are active and present all year which is an additional advantage comparing to other bioindicators which can grow and develop only within the vegetative season. The obtained results suggest the need for further investigation of the possibilities of the broader application of spider webs in PCDDs determination.

The work was realized within the allocation No. 0401/0068/16 awarded for Faculty of Environmental Engineering Wroclaw University of Science and Technology by Ministry of Science and Higher Education.

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