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Fluoride Accumulation in Different Earthworm Species near an Industrial Emission Source in Southern Germany

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The local pollution of landscapes with fluorides (F) around industrial works like aluminium smelters, steel works and coal fired power plants is well known and documented (e.g. Drury et al., 1980). Potentially toxic airborne F compounds may not only injure the vegetation directly but also accumulate in soils, soil microflora and -fauna are exposed to these chronic hazards, but sofar little is known on F- effects.
The information on F-pollution of soil invertebrates is sparse and only a few recent publications deal with F accumulation in some taxonomic groups of soil fauna (Garrec and Plebin, 1984; Buse, 1986; Walton, 1987; Vogel et al., 1989; Vogel, 1990). Earthworms in particular become the focus of soil- soil fauna interactions in F- polluted sites, the more since a significant relationship between soil pollution and F load in earthworms was observed (Garrec and Plebin, 1984; Vogel et al., 1989). Earthworms coat their burrowings and this may be a mechanismn of F-dissemination and subsoil contamination (Breimer et al., 1989). Evidence is growing that fluorides pass through foodchains (Groth, 1975; Murray, 1981). Earthworms as the prefered prey of a wide range of animals (MacDonald, 1983; Lee, 1985) are therefore in the centre of interest as a possible way of F- bioaccumulation in higher trophic levels. For a risk assessment of F- pollution and pathways of F through organisms and ecosystems, detailed knowledge of F- accumulation in soil fauna, and in earthworms in particular is required.

MATERIALS AND METHODS

The investigations were carried out in the surroundings of the chemical factory Kali Chemie AG in Bad Wimpfen/ Neckar, northern Baden-Württemberg (FRG). The factory started F- emission in 1921 with cryolith production and diversified its F- chemistry from 1960 on (including HF, refrigerants and spray propellants). In the meantime, F- emission and -pollution of the surrounding landscape have a 70 year old history.

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The soils surrounding the site of emission were examined for F-pollution (total F, HCl extractable F and water soluble F) in detail and grouped in a heavily polluted zone (acid soluble F >200 $mgF.kg^{-1}$), a moderately polluted zone (100 - 200 $mgF.kg^{-1}$) and a non-polluted area (< 100 $mgF.kg^{-1}$) (Breimer et al., 1989). The earthworms were extracted from the heavy and the non polluted sites with 5 L formalin solution (0,5 %) per 0,25 m^2. The worms were sacrified in 70 % ethanol. Just half the worms were dissected to remove gut content in order to differentiate between the F- content of animals with gut and F in tissues only. All animals were cleaned with CDTA-(Cyclo-hexandiamin-tetra-acetic acid)solution (0,25 %) and rinsed carefully with distilled water to remove surface contaminations. The dissected earthworms were cleaned also inside with CDTA- solution and distilled water. After cleaning, the samples were oven dried (72 h, 60 °C) and then ground to a fine powder in an agate mortar. The F- content was determined by the oxygen combustion method after Levaggi et al. (1971). The F- determinations were carried out with a F- sensitive electrode (WTW F 500) and ionmeter (WTW pMX 2000) as described previously (Breimer et al., 1989; Vogel et al., 1989). For statistical evaluation the Kruskal-Wallis H-test and Student´s t-test were used.

RESULTS AND DISCUSSION

In table 1 the F- content of the different earthworm species are presented. The F- content in the species without gut revealed a clear F- accumulation whereas the values of the whole animals well reflect the contamination of soil and litter. In the control group no significant difference in F load between the various species was found. Opposite, the polluted earthworms showed highly significant differences in the F- accumulation of the various species (H= 24,94*** with gut and H= 44,91*** without gut).

The significance of differences in the F- accumulation between the species examined are listed in table 2. A sequence of decrease in F- content in the contaminated species without gut content followed the order: Octolasion cyaneum and lacteum > Lumbricus castaneus > Aporrectodea rosea > A.longa > A.caliginosa > L. rubellus > L.terrestris.

Different life-forms (epigeics, endogeics and anecics) and food requirements (geophagous- detritophagous) of the earthworms may cause variable F- contamination and may reflect the various contamination levels discussed by Kühle (1983). The ecological groups of the investigated Lumbricidae are listened in table 3.

In the present study, the endogeic species showed the highest mean F- contamination. Since these species are essentially geophagous, the soil water and soil colloids during intestine passage may be the most important contamination source. In addition, endogeic forms ingest dead roots, which have always higher F-

Table 1. F- content ($\mu gF.g^{-1}$) in different earthworm species with and without gut content near the pollution source in southern Germany

species	X 1)	non polluted min- max 2)	Ni:Na 3)	X	polluted min - max	Ni: Na
	earthworms with gut content					
Lumbricus rubellus	64	15 - 221	81:43	323	145 - 563	97: 81
L.castaneus	82	26 - 217	13: 5	280	157 - 525	39: 21
L.terrestris	90	44 - 177	21:33	254	43 - 521	237:137
Aporrectodea caliginosa	175	-	1: 3	322	76 - 515	19: 29
A.rosea	-	-	-	518	-	1: 1
A.longa	-	-	-	408	-	1: 1
Octolasion lacteum + cyaneum	-	-	-	449	353 - 556	18: 24
Lumbricus spp.	86	15 - 331	19: 9	303	93 - 724	121: 61
Aporrectodea spp.	111	78 - 175	56:19	355	113 - 588	147: 72
	earthworms without gut content					
species	X	non polluted min- max	Ni:Na	X	polluted min - max	Ni: Na
Lumbricus rubellus	21	1 - 62	62:47	65	18 - 142	91: 75
L.castaneus	21	6 - 46	12: 3	119	29 - 435	19: 8
L.terrestris	17	3 - 47	48:47	46	7 - 120	199:232
Aporrectodea caliginosa	23	5 - 56	11:10	83	15 - 239	33: 46
A.rosea	40	-	3: 2	108	70 - 123	5: 5
A.longa	21	3 - 40	4: 2	88	-	2: 3
Octolasion lacteum + cyaneum	11	-	3: 2	127	51 - 231	32: 32
Lumbricus spp.	18	4 - 44	13:11	37	20 - 100	19: 18
Aporrectodea spp.	15	8 - 22	8: 4	90	54 - 236	26: 18

1)mean F- content, 2) minimum and maximum F load in the earthworms analysed, 3) Ni= number of individuals tested and Na= number of analyses

contents then other vegetation components (Andrews et al., 1989). Finally, endogeic earthworms preferentially colonise mineral-horizons, which contained the highest F load (especially water soluble F) in the soils studied (Breimer et al., 1989).

Table 2. Significant differences in F-content of the single earthworm species studied from the polluted sites

earthworms without gut content			earthworms with gut content		
L.terrestris	- L.rubellus	**	O.cyaneum + lacteum	- L.terrestris	***
"	- A.caliginosa	***	"	- L.rubellus	**
"	- A.rosea	***	"	- A.caliginosa	*
"	- O.cyaneum + lacteum	***	L.rubellus	- L.terrestris	*
A.rosea	- L.rubellus	**			
"	- O.cyaneum + lacteum	**			

For F- content and numbers of individuals and analyses see table 1 above. Significance of Student´s t-test: ***= P 0,001; **= P 0,01; *= P 0,1

Table 3. Life- forms and food requirements of the investigated earthworms. After accounts from Bouche´and Gardner (1984), Lamparski (1985) and Lee (1985)

	life-form endogeic	epigeic	anecic
species:	A.rosea A.caliginosa O.lacteum O.cyaneum	L.rubellus L.castaneus	L.terrestris A.longa
horizon:	humic mineral horizon	humus layer	soil surface-subsoil
food:	geophagous microorganismns dead roots	litter microorganismns casts	litter microorganismns

The epigeic worms obtained their F from the litter, which showed a heavy F- contamination at the sites in question (Vogel et al., 1989). The high mean F- content of L.castaneus may reflect the small sampling number and the maximum load of 435 µgF.g-1 (table 1). The anecic L.terrestris showed always the lowest F-accumulation. Perhaps its temporary stay in deeper mineral horizons, which were not contaminated even in heavy F- polluted soils (Breimer et al., 1989), allow these organisms to release F in this clean environment. In the case of earthworms, soils become apparently not only a sink for F, but also a direct source in a way Davison (1987) mentioned for vegetation and for animals via plants.

Based on the present study, a differentiation between the various earthworm species is essential, if these organisms are used as indicators for F- pollution. Consequently, a comparison of our data with the findings of other authors is only of little significance if data do not distinguish even between the different genera of earthworms (Garrec and Plebin, 1984; Walton, 1986). Undoubtedly, the elimination of the gut content is a prerequisite if fluoride accumulation in earthworm tissues should be proved.

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