Polish Journal of Environmental Studies Vol. 12, No. 4 (2003), 431-437

Degradation of Individual Polycyclic Aromatic Hydrocarbons (PAHs) in Soil Polluted with Aircraft Fuel

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Received: 26 August, 2002
Accepted: 22 October, 2002

Abstract

Degradation of polycyclic aromatic hydrocarbons (PAH) such as phenanthrene, anthracene, fluoranthene, pyrene, benzo[a]anthracene, chryzene, benzo[b]fluoranthene, benzo[e]pyrene, benzo[k]fluoranthene and benzo[a]pyrene) in the surface layer (5 cm) of soil polluted with aircraft fuel in an amount of 10 ml/kg was analyzed. The direct influence of solubility in water on the degree of its loss was proven. The clear influence of the soil type – directly related, among other things, to organic matter content and contribution of alluvial fraction – on the decomposition of 5-ring PAHs. Half-lives ranged from 152 to 313 for 3-ring PAHs and from 117 to 492 for 4- and 5-ring PAHs.

Keywords: polycyclic aromatic hydrocarbons, aircraft fuel, degradation of PAHs, soil contamination

Introduction

Soil pollution with crude oil is most difficult and most expensive to remove [1]. This problem is especially visible in the vicinity of airfields. Aromatic hydrocarbons are composed of numerous substances originating from crude oil [2], and their contribution can vary from 4 to 22% depending on substance type [3]. One of the most harmful groups of aromatic hydrocarbons is that of polycyclic aromatic hydrocarbons (PAHs). Their negative influence on animal and plant organisms has been discussed in numerous research works [4, 5]. Due to their properties, PAHs are characterised by their high durability in the environment, which allows them to accumulate in the soil for many years, and degrade with difficulty. However, as shown by some researchers [6, 7], decomposition of the above pollutants depends to a large extent on soil properties, and the scope of their harmful impact on organisms is directly related to soil type [5].

In the present study, the ability of natural soil microflora to degradation-selected PAHs in soil polluted with aircraft fuel in the 0-5 cm soil layer was evaluated.

Materials and Methods

Preparation of Soil Samples

The study material consisted of soils collected from two areas endangered by pollution with PAHs (layer 0-20 cm) (Table 1). Containers (7 kg) made of plastic with a volume of 10 l were filled with the soil. The soil was kept wet for 10 days at room temperature in order to activate the microflora. Then the soil was polluted with aircraft fuel in an amount of 10 ml/kg. The first samples (about 50 g for each container) were taken from around the 5 cm layer 7 days after the introduction of aircraft fuel. Four samples from each container were taken with plastic pipe (2 cm in diameter) and all four were mixed to obtain a representative sample. The remaining samples were taken, respectively, after 25, 105, 191 and 362 days. The

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Table 1. Some properties of investigated soils.

Properties		Soil F	Soil L
pH		6.5	4.7
P^* [g/kg]		66.9	12.6
TOC [g/kg]		10.50	8.80
Total N [g/kg]		1.26	0.70
Soil texture [%]	1-0.1	33	57
	0.1-0.02	36	25
	<0.02	35	18
Σ PAH (μg/kg)		343.6	1077.2

* - available forms

experiment was carried out in two repetitions. For each repetition independent TOC, N, P and PAH content was determined.

Determination of PAH Content

A sample (20 g) was extracted in an ultrasonic bath (Sonic-3, Polsonic, Poland) with two batches of dichloromethane (2 x 40 ml). The extract was centrifuged and evaporated to dryness. The residue was then dissolved in a mixture 4 ml of acetonitrile:water (1:1 v/v) and purified by solid phase extraction (SPE) using C_{18} Octadecyl columns (3 ml, 500 mg) (JT Baker-Mallinckrodt, Germany). The column was conditioned with one portion of methanol (1x3 ml), followed by two portions (2x3 ml) of 2-propanol:water mixture (1: 9). The resulting extract was then transferred to the column without allowing it to dry out. After drying, the column was washed with one dose (1x3ml) of methanol:water mixture (1:1), dried (5 min, light vacuum) and the PAHs were eluted two portions (2x1.5 ml) of acetonitrile.

A qualitative and quantitative analysis of PAHs was carried out on the liquid chromatograph with UV detection (TermoSeparation Products). The mobile phase was acetonitrile:water mixture (80:20) (isocratic condition). A Spherisorb S5 PAH (250x4.6 mm I.D., 5μm) (Schambeck SFD GmbH, Germany) column was used for the separation of 16 PAHs. Detection was carried out at 254 nm. The column was installed in a thermostated oven at 31°C (LCO 101, ECOM, Czech Republic).

Quantitative determination was performed using the absolute calibration curve method. The correlation coefficients of calibration functions in the intervals of linearity were higher that 0.9982-0.9998.

The detection limits calculated with a signal to noise ratio of three (IUPAC criterion), for 20 μl loop injection, were less than than 0.05 μg/ml and 0.5 ng for the concentration and amount of the PAHs.

Determination of Soil Properties

The physical-chemical properties of the soils studied were determined by methods generally used in chemical-soil laboratories: pH in KCl, the total of the exchangeable bases (TEB) and cation exchange capacity (CEC) [8, 9]. Total carbon content (TOC) was determined by the Tiurin method as modified by Simakov [10], and the content of phosphorus available forms according to the Egner-Riehm method [8, 9]. Granulometric composition by the aerometric method of Casagrade as modified by Prószyński [8, 9].

Data Analysis

The rate of decomposition of some chosen PAHs (Table 2) was determined on the basis of the comparison of the kinetic equation of the pseudo-first order [6]:

$$\ln \frac{C_0}{C_t} = {}^*k \cdot t \tag{1}$$

where:
*k – apparent constant reaction rate of the pseudo first order (1/day),
t- time [days],
C_o – initial PAH contents in soil,
C_t – PAH content in soil after time t.

Half-live ($T_{1/2}$) [6] was determined from equation (2):

$$T_{\frac{1}{2}} = \frac{\ln 2}{{}^*k} \tag{2}$$

Statistical analysis was performed using MS Excel 2000 (Microsoft) and ARSTAT (AR Lublin). Standard deviation (SD), relative standard deviation (RSD, %) and Pearson correlation coefficient were calculated. Statisical evaluations were performed using a t-Student test. Differences were considered as significant at $p<0.05$.

Results and Discussion

Soil Characteristics

The physical-chemical properties and PAH total sum in the soils used in our research are presented in Table 1. The data obtained clearly showed that the studied soils differed clearly in their properties. Soil *F* (proper brown soil originating from loess) was characterised by a neutral pH, the mean content of phosphorus available and the mean total of nitrogen and carbon content characteristic of most soils in Poland. Further to BN-78/9180-11 classification it is light silt loam. Soil *L* (proper podzolic soil originating from sand) showed an acidic pH and a low content of phosphors available, as well as nitrogen and total carbon (Table 1). Further to the aforementioned classification it is loamy, sandy, strong and silty.

Also in respect to PAH content, the soils studied were also clearly differentiated. In soil *F*, an amount of 343.6 μg/kg (RSD=16%, n=3) was determined, whereas in soil *L*

Table 2. Some properties of investigated PAHs.

PAHs	MW	S	log K_{oc}	log K_{ow}
Phenanathrene	178	1.29	4.36	4.6
Anthracene	178	0.07	4.42	4.5
Fluoranthene	202	0.26	6.38	5.2
Pyrene	202	0.14	4.80	5.2
Benz[a]anthracene	228	0.014	7.30	5.9
Chrysene	228	0.002	3.66	5.9
Benzo[b]fluoranthene	252	0.0012	5.74	5.8
Benzo[k]fluoranthene	252	0.00055	5.74	6.0
Benzo[e]pyrene	252	0.004	7.20	-
Benzo[a]pyrene	252	0.0038	8.30	6.0

* - made on the basis of data from Department of Environmental Science, Lancaster University.
MW – molecular weight ($g \cdot mol^{-1}$); S – aqueous solubility (g/m^3).

– 1077.2 μg/kg (RSD=13%, n=3), which should be related to automotive pollution (soil collected in the vicinity of motorways). A difference in the composition of individual PAHs were also noted. In soil *F*, Fluo (22.2%) and B[e]P (19.8%) were predominant, whereas in soil *L* B[b]F (22.1%) and B[k]F (21.7%). After the introduction of aircraft fuel, a rapid increase in all PAHs was observed. The degree and scope of this increase was clearly differentiated and depended not only on the type of hydrocarbons but also on soil type (Fig. 1).

PAH Degradation in Relation to the Number of Rings

Based on the model of organic pollutant distribution in soils enriched with organic matter of anthropogenic origin as proposed by Beck et al. [11], it can be assumed that most of the PAHs studied behaved according to curve *C*, less frequently *B*, and only in one case, according to *E*. Model *C* is characteristic of most organic pollutants with semi-volatility and average sorption properties (3-4 rings PAHs). The scope and degree of degradation of individual PAHs differed clearly both in the framework of properties of these compounds (number of rings, molecular weight, solubility in water) (Table 2), and also soil type.

The influence of PAH properties (first of all solubility in water - SW) was clearly visible in soil F (Figs. 2-4). This is understandable since it is well known that SW is a factor which determines both PAH leaching into the depth

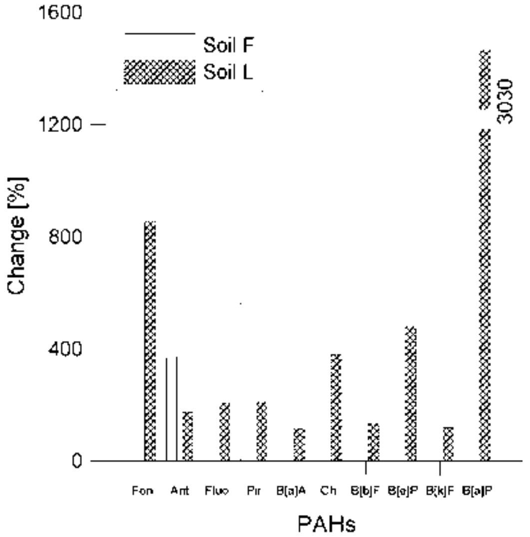

Fig. 1. Changes in PAH content [%] in the soil after contamination with aircraft fuel.

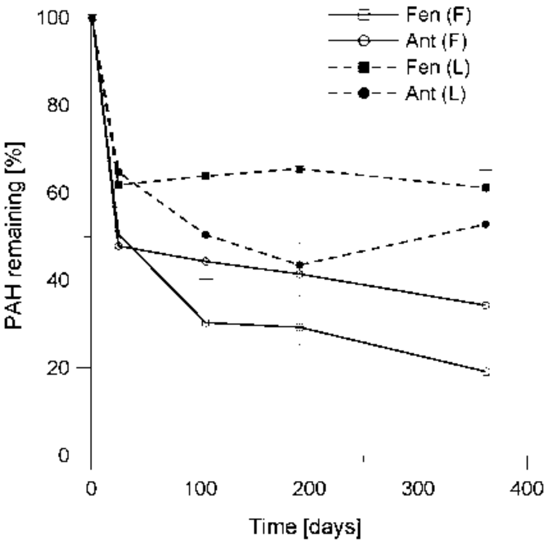

Fig. 2. Degradation of 3-ring PAHs in investigated soils. Error bars represent standard deviation, n=2.

of the soil profile and its susceptibility to biological and physical degradation [6].

In the case of 3-ring PAHs (Fig. 2), a 50% decrease in their presence was observed within the first 25 experimental days. In the following days, their content continued decreasing; however, this decrease was not as intensive as in the initial period. Finally, 20 and 38% of Fen and Ant which were originally present in the soil were determined.

Fluo and Pyr (4-rings), characterised by an identical molecular weight and a similar solubility in water (Table 2), behaved in an almost identical way during the whole experimental period. On the other hand, B[a]A and Ch, with considerably lower SW values than the aforementioned hydrocarbons, showed markedly lower losses both in the initial stage of the experiment, and after its completion (25 days) (finally, 60% of PAHs which were initially in the soil, remained). In the case of 5-ring PAHs, a similar tendency was also observed. However, similarities resulted from similarities in the molecular composition (homologues).

In soil *L* no clear influence of PAH properties on their decomposition was noted (Figs. 2-4). PAH losses of a similar level were observed in individual groups. Only in the case of 4-ring PAHs were differences in the initial (up to 100 days) stage of the experiment seen (Fig. 3).

PAH Degradation in Relation to Soil Type

Differences in PAH decomposition related to soil type are seen in Figures 2-4. Half-lives (Fig. 5) determined on the basis of equations (1) and (2) confirm the above. The clearly better decomposition of 4- and 5-ring PAHs was noted in soil *L*, and the relatively weak decomposition of 3-ring PAHs. Data presented in Figure 5 shows that starting from B[a]A in soil *L*, better decomposition of PAHs was observed. It is especially distinct for B[b]F and B[k]F (Fig. 5).

The role of autochthonic microorganisms in PAH degradation of the soils studied is not without impact. It is known [12, 13] that bacteria, fungi and *Actinomycetes* take part in biological PAH degradation. Cases of the degradation of various pollutants by microorganisms naturally occurring in the polluted environment are known in literature [14, 15]. PAHs with a low number of rings (<4) are especially prone to biodegradation [12]. For example, half-lives given in literature for Fen (3-rings) in soils and sediments are from 16 to 123 days, whereas for B[a]P (5-rings) from 229 to over 1,400 days [16].

Biodegradation of 4-rings PAHs and PAHs with higher number of rings is difficult but possible [17]. As shown in literature [12, 18, 19], biodegradation takes place by co-metabolism. For example, Kanaly and Bartha [18] showed the distinct positive influence of crude oil on the decomposition of B[a]P. Jimenez and Bartha [20] suggested that it was probably related to an increase of B[a]P bioavailable forms. Studies by Juhasz, et al. [17], on the other hand, showed a visible increase of dibenz[a,h]anthracene and benzo[a]pyrene mineralisation in the presence of phenanthrene.

Considering the above information, the role played by all PAHs in the experiment could not be omitted. It can be concluded that the quick decomposition of 3-ring PAHs in the initial stage of the experiment (Fig. 2) limited their amount and hence the later degradation of PAHs >4 rings was slow (Figs. 3 and 4).

Attention should also be drawn to the fact that soil *L* – proper podzolic soil in which a better distribution of xenobiotics studied was observed, especially carcinogenic 5-ring compounds, was characterised by PAH content on a level three times higher than soil *F* (Table 1). Hence, it can be presumed that groups of micro-organisms present

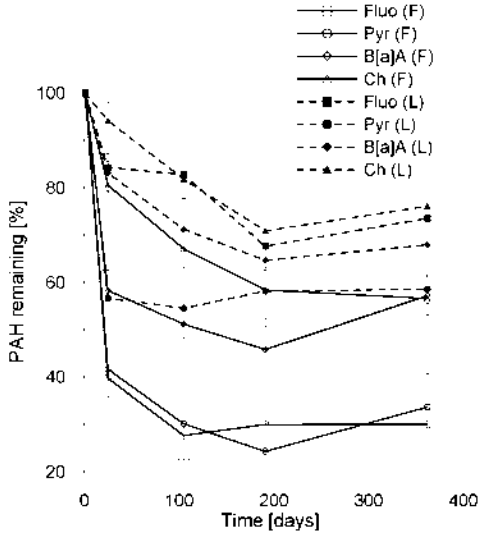

Fig. 3. Degradation of 4-ring PAHs in investigated soils. Error bars represent standard deviation, n=2.

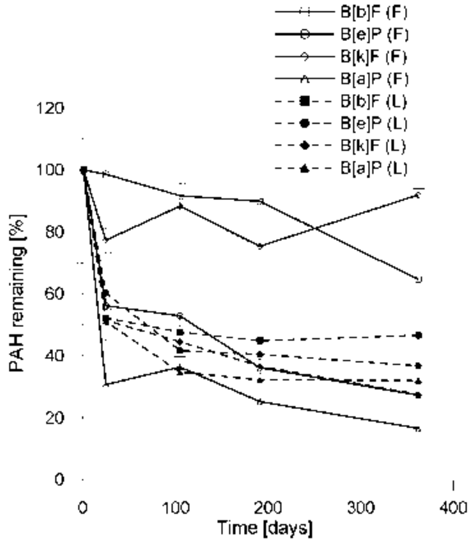

Fig. 4. Degradation of 5-ring PAHs in investigated soils. Error bars represent standard deviation, n=2.

in this soil, probably mostly fungi when judged by the soil's reaction [13], adapted to the existing conditions and were capable of biodegradation pollutants introduced to the soil. At the same time, it should be noted that biological degradation with a fungal contribution is unfavourable as toxic intermediate compounds can result from it [21] such as, for example, highly carcinogenic intermediate epoxy forms [6].

When analysing the data presented in Fig.1, it is clearly seen that soil *L* "survived a smaller shock" after its contamination with fuel than soil *F*. Only Fen, Ch, B[e]P and B[a]P showed an increase comparable to their initial content in the soil. Whereas in soil *F*, as much as seven PAHs increased their content more than three times. From the above information it seemed that soil *L* was microbiologically better prepared for the decomposition PAHs introduced than soil *F*.

The suggested adaptation of microorganisms to PAH degradation was also confirmed by the fact that predominant PAHs in soil *L* were B[b]F and B[k]F which quickly underwent decomposition, whereas in soil *F*, Fluo was predominant, whose half-life in this soil was clearly shorter than in soil *L* (Fig.5).

Organic matter plays an important role in biodegradation intensity [5-7, 11]. A higher content of TOC in soil *F* can significantly limit the availability of pollutants for micro-organisms, which can explain their poorer degradation. It is especially visible in the case of 4-ring B[a]A and 5-ring homologues: B[b]F and B[k]F (Fig. 5) which strongly react with organic matter as indicated by high levels of *log* K_{oc} coefficient (Table 2). The above has been confirmed by other authors, who found that organic compound sorption, PAH included, by organic matter can significantly limit their bioavailability [11, 22]. It is especially visible in soil enriched with organic matter of anthropogenic origin (i.e. compost) [23, 24]. Limitation of bioavailability depends first of all on the character of organic matter and type of PAHs. Laor et al. [25] showed differentiation in phenanthrene sorption related to the type of humic acids. The aforementioned authors did not find limitations in the biological degradation of phenanthrene adsorbed on acid particles. However, it is known that in the sorption of organic pollutants and bound-residue formation which can limit PAH degradation, the remaining part of organic matter components, first of all humines [26], play an important role beside humic acids. It was interesting to get to know the character of organic matter and contribution of its individual forms (fulvic, humic acids and humines) of the soils studied. Taking soil type into consideration it can be concluded that organic matter content differs, and hence type of impact and its strength in the soil studied was varied.

Considerable amounts of clay fractions (Table 1) might also have an impact on the limitation of PAH degradation in soil *F*. Clay minerals have reactive surfaces that are normally coated with organic matter [27]. Their clear influence on MCPA sorption was observed by Haberhauer, et al. [28] in soil with a low content of organic matter (1.1-1.3%), and the content of clay minerals above 25%. Also Maliszewska-Kordybach [29] noted the role of clay minerals in the limitation of PAH availability in her formula for determining the related sum of PAH content.

Besides biodegradation, photodegradation and evaporation also account for PAH losses in the surface soil layer (up to 5 cm) [6]. However, these processes play an insignificant role and are often neglected when evaluating PAH degradation degree in soils.

Analysis of standard deviation (SD) (Figs. 2-4), showed clear differences in SD values only in the case of 3-ring PAHs and its dependence on soil type (Fig. 2). In remaining cases (4 and 5-ring PAHs) individual standard

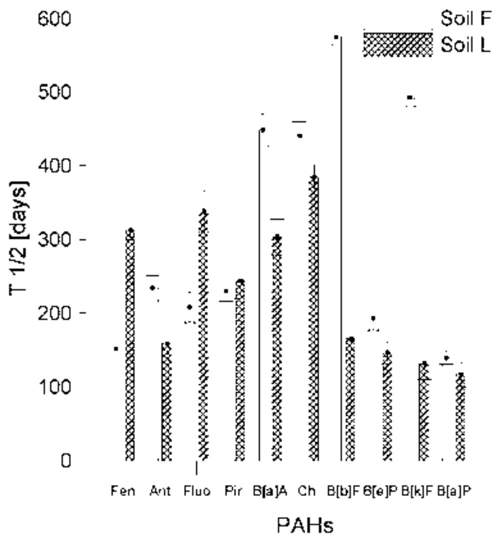

Fig. 5. Half-life time (T1/2) of PAHs in investigated soils.

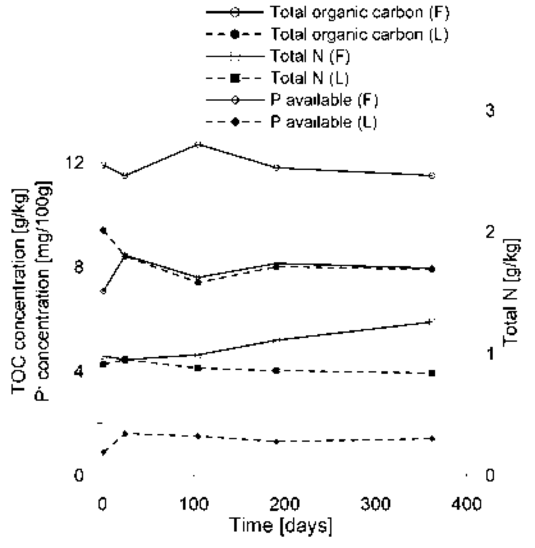

Fig. 6. Changes of the total organic carbon, total nitrogen and available phosphorous during research.

deviation was clearly dependent on sample collection time and most samples did not exceeded 10%.

Changes in PAH Content and Chemical Properties of Soils

Contents of TOC, available phosphorus and total nitrogen was also monitored during the present research (Fig. 6). The content of the above-mentioned components did not undergo any significant changes. However, in the case of *L* soil, a high positive correlation between the content of Σ PAH and TOC amounting to r=0.919 (α=0.05), as well as the negative correlation between PAH and available phosphorus (r=-0.791, α=0.05) which may prove that phosphorus availability influenced PAH degradation. A relation on a similar level between Σ PAH and available phosphorus was also noted in soil *F* (r=-0.760, α=0.05).

Conclusions

The following conclusions were drawn on the basis of the results obtained:

1. PAHs decomposition in soils belonging to various types clearly depends on both PAH properties (solubility in water, *log* K_{oc}), and the properties of the subject soils (content of organic matter, content of clay fractions);
2. Groups of microorganisms occurring naturally in the soil are able to degrade pollutants introduced into the soil, and the scope of degradation is clearly better in soils characterised by increased content of these microorganisms, which can result from the earlier formation of cultures of microorganisms which are capable of degrading them;
3. Favourable conditions for the development of microorganisms (neutral soil pH – Soil *F*) and the high content of nutrients (available phosphorus, and also nitrogen) play an important role in the case of degradation of 3-ring PAHs, and the degradation of PAHs with a higher number of rings (>4) is determined by their bioavailability. The presence of organic matter and a considerable share of alluvial fraction limits these processes;
4. The rapid decomposition of 3-ring PAHs may significantly influence the limitation of PAH degradation with higher numbers of rings (first of all 5-rings ones), which is related to the phenomenon of cometabolism.

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