

URBAN CONTAMINATION ASSESSMENT OF POLYCYCLIC AROMATIC HYDROCARBONS RELEASED FROM AN OIL REFINERY IN RAWALPINDI

by

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*Polycyclic aromatic hydrocarbons are among persistent organic pollutants and incomplete combustion of hydrocarbons during petroleum processing activities may cause polycyclic aromatic hydrocarbons contamination in the surrounding area. For the first time in Morgah, Rawalpindi, Pakistan, three targeted polycyclic aromatic hydrocarbons i.e. naphthalene, phenanthrene, and pyrene were estimated from surroundings of an oil refinery. Samples were collected simultaneously in soil, water, and leaves of *Melia azedarach L.*, a widely grown tree species in the sampling area, along different selected sites of main drainage coming out of the refinery. Among three polycyclic aromatic hydrocarbons, pyrene was observed in maximum concentration (24.31 mg/kg in soil) among all selected environmental media and studied compounds showed a dynamic behavior in context of accumulation at different sampling points. There was a significant effect of distance, from petroleum refining area on polycyclic aromatic hydrocarbons levels in soil, water and plant leaves. Our findings provide conclusive evidence that location of an oil refinery has attributed to high polycyclic aromatic hydrocarbons contamination in selected study area which can pose significant risk to general environment and to the public.*

Key words: oil refinery, urban contamination, naphthalene, phenanthrene, pyrene, petroleum processing, soil, water environmental risk, vegetation

Introduction

Polycyclic aromatic hydrocarbons (PAH) belong to a group of organic compounds that have semi-volatility, photosensitivity, lipophilicity, and are also less soluble in water. Originally, PAH have various congeners that exist in the atmosphere, among them, USEPA [1] declared 16 PAH as priority pollutants. Some PAH are reported to have toxicity, mutagenicity, and carcinogenicity potential for humans and as well as for the ecosystem, as PAH are

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persistent compounds and ubiquitously exist in the environment. Moreover, these compounds when bounded with air-borne particles can be transported to far distances, away from their point of origin [2]. Various compartments of the environment are reported to have high levels of PAH and final deposition of these organics is based on weight of molecules, ring number and as well as on their congener's phases. The PAH are commonly present in the environment with fused rings of benzene (2 or >2) [3]. Congeners can be classified as low (2 to 3-rings), medium (4-rings), and high molecular weight (5 to 6-rings) [4]. The two main sources for PAH production are natural and anthropogenic, and former source includes sedimentary rocks, volcanic eruption, and forest fires, *etc.*, and the later source includes burning of biomass and fossil fuels. Yet, there is a negligible contribution from natural sources as compared to anthropogenic sources for PAH release into the environment and furthermore, anthropogenic sources can be divided into pyrogenic and petrogenic [4].

The PAH, both in gaseous or particulate phase are released directly into the environment, and then deposit on the soil that later on serve as a pool for PAH. The deposition of these compounds on the soil occurs through dry and wet deposition processes [5]. Sorption of PAH or other contaminants on the soil preferably occur in carbonaceous geo-sorbents, as well as in amorphous organic compounds (called black carbon and organic carbon, respectively). In fate and distribution of organic pollutants, soil plays a vital role either by adsorption on the black carbon or absorption in organic matter [6]. Based on physicochemical characteristics and environmental conditions, PAH deposited in the soil will experience multiple soil-air exchange processes. From soil, gaseous phase PAH will be subjected to re-volatilization as the temperature goes high (summer season) and re-deposit on the soil particles as the temperature decreases during winter season [7].

In urban environment, when organic substances experience exposure to elevated temperatures and also to low or non-oxidizing conditions, pyrogenic PAH are formed in high amount and almost in every compartment of the environment. Moreover, PAH are formed/released into the environment either intentionally or unintentionally. Most common examples include thermal cracking of petroleum residuals that yield low molecular hydrocarbons or destructive distillation of coal produce coal tar and coke and incomplete combustion of motor fuels in cars and trucks or fuel oils in heating systems produce such compounds. The PAH are abundantly available in the environment and can remain there for decades. Also, more PAH are being continuously added to the environment from direct sources (industrial or vehicular) [8] and will be deposited on the soil or in water, and from there eventually will be taken up by plants and enter into the human bodies [3, 5].

Various studies have been conducted on PAH uptake into the plant bodies [9, 10]. A direct association was found between PAH content of the soil and plant tissues in a work conducted by Meudec *et al.* [11] and suggested that probably, there is a pathway for these compounds from polluted soil to plant roots. The PAH enter into plant tissues through roots or cuticle [12], as they are lipophilic in nature and can be bioaccumulated in plant bodies and may be partition into the epidermis of the roots or on the soil [13].

In developing countries like Pakistan with a two hundred million population, emissions from industrial and vehicular sources are recognized as major contributors of PAH [14]. Also, this country is at the seventh number in contributing PAH releases on the global scale [15, 16]. Moreover, out of total population of Pakistan, more than 70% population lives in rural areas. These people acquire their energy needs (around 94%) through burning of biomass. If biomass combustion is incomplete then this is also a source of PAH release into the environment [17].

In past, there is scarcity of information and work about levels of PAH in Pakistani environments, and first detailed work on PAH levels in the atmosphere and dust matrices was reported by Smith *et al.* [18] in Lahore. The average Σ PAH content was found between $0.35 (0.35 \cdot 10^{-4} \text{ ug/m}^3)$ and $14.64 \text{ ng/m}^3 (1.46 \cdot 10^{-2} \text{ ug/m}^3)$ with BghiP (benzo-ghi-perylene), as a major contributor to average Σ PAH content. With a span of nearly two decades, Kamal *et al.* [19] carried a comparative, intracity exploration in Lahore, Rawalpindi and Gujranwala, for profile of atmospheric PAH level. It was found that low molecular weight PAH were the prevailing isomers. A high concentration of $327 \text{ pg/m}^3 (3.27 \cdot 10^{-4} \text{ ug/m}^3)$ and $316 \text{ pg/m}^3 (3.16 \cdot 10^{-4} \text{ ug/m}^3)$ of naphthalene as an individual PAH was found in Lahore and Rawalpindi, respectively [19]. Irrespective of atmospheric studies on PAH content, biomonitoring studies associated with PAH exposure can also be found in Pakistan [20-22]. Most recently, PAH levels and sources, and associated health risks, as a result of changing energy demand have been reported [23]. Moreover, in soils and sediments along Jhelum riverine system of lesser Himalayan region of Pakistan, assessment of levels and sources of PAH has been studied by Riaz *et al.* [24].

After extensive literature review, consequently, no reported work has been done to assess PAH contamination in the surrounding environment of the Morgah oil refinery in Rawalpindi, Pakistan. This refinery is located near residential area and other public health care facilities are also present in the vicinity. Therefore, PAH contamination assessment is necessary for future pollution control and for the application of remediation techniques. The unique aspect of this investigation is simultaneous evaluation of selected PAH concentration in different media of the environment *i.e.* soil, water and plants (*Melia azedarach L.*; most abundant specie) in Morgah oil refinery. Three low molecular weight PAH congeners, *i.e.* naphthalene, phenanthrene and pyrene are selected for PAH assessment in the study area. The objectives of the current work include: quantification of selected PAH concentrations in soil, water and leaf samples from selected sampling area, and to assess the severity of PAH contamination with respect to the distance from petroleum hydrocarbon activity. Further, a correlation analysis has been carried out to find a relationship between soil PAH levels and soil organic matter and pH.

Methodology

Sampling area

For PAH assessment, soil, water and leaf samples of *Melia azedarach L.* are obtained in the locality of an oil refinery in Morgah, Rawalpindi, along a water drainage that flows directly from the refinery towards a populated area. This city is in Pothwar region and located in the northern part of Punjab, Pakistan. Sampling was carried out at four different points. Waste-water treatment plant located inside the refinery was chosen as Site 1 and the exit point of main drainage of the refinery was chosen as Site 2, about 0.5 km away from Site 1. Sampling Site 3 was selected near a small medical waste incinerator (1.0 km away from Site 1) along the drainage.

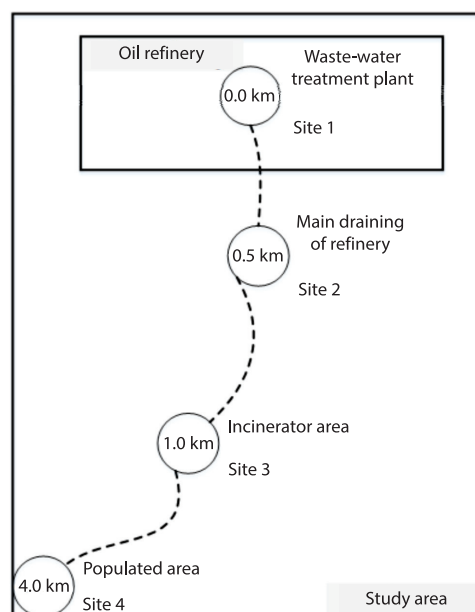


Figure 1. Lay-out for soil, water and leaf (*Melia azedarach L.*) sampling sites

Site 4 (at 4.0 km distance from Site 1) was the populated area. The difference between distances of first three sampling sites was consistent *i.e.* 0.5 km, while Site 4 was selected at about 4.0 km away from Site 1 along the waste-water drainage, when water flow greatly reduced, fig. 1.

Chemicals and materials

Solvents of HPLC grade *i.e.* acetonitrile, ethyl acetate, n-hexane, and dichloromethane (DCM) and naphthalene, phenanthrene, and pyrene standards for GC chromatograph were procured from sigma-Aldrich, USA. Solvents used in mobile phase were degassed by sonication before use. Anhydrous sodium sulphate, Na₂SO₄, was procured from Merck, Pakistan. Silica gel (100-200 mesh) for column preparation and sample cleaning was also purchased from sigma-Aldrich, USA.

Sample collection

Samples of soil, water, and plant leaves were collected according to standard procedures. For soil samples, upper 10 cm layer of the soil with an area approximately 20 cm by 20 cm was collected in ziplock polyethylene bags with a spade and immediately transferred to the box containing ice, so that biodegradation of the samples will be reduced due to any bacterial or chemical activity. When there was vegetation or leaf litter (about 0.5 cm deep), then these materials were carefully scraped away before sampling. The four to five samples were taken to make one composite sample at each sampling site and each sampling point was about 5 m away from each other. To ensure complete extraction of selected PAH and to prevent volatilization of temperature sensitive analytes like naphthalene, samples of soil were freeze dried in a freeze drier (Telstar CRYOOS- 50, Spain) and lastly kept at 4 °C. From the same sampling points, water samples (composite samples) were collected from surface of wastewater drainage with sterilized sample bottles of 500 ml. In order to avoid any microbial activity, concentrated HCl was added at the time of water samples collection. *Melia azedarach L.* was abundantly grown tree species in the study area and leaves (weight > 8 g) of this tree were collected at 1 m height and after that immediately enfolded in aluminum foil. Like soil and water, leaf samples were stored at 4 °C for pretreatment.

Pretreatment of samples

Soil, water, and leaf samples were pretreated by adopting the following methods. For soil samples, 1 g freeze dried soil sample was transferred into 100 ml teflon tube, and DCM was added in a ratio of 1:3 (1 g soil: 3 ml DCM). Next, samples were extracted for two hours in an ultrasonic bath, and temperature of water inside the ultrasonic bath was set at 40 °C. After sample extraction, centrifugation was performed for five minutes on this mixture at 4000 rpm in order to isolate the supernatant from soil. Silica gel column was prepared and wetted with n-hexane prior to sample cleaning. 0.5 ml extract from isolated supernatant was added into the column and 25 ml DCM: n-hexane (*v/v* 50:50) mixture was used to elute the extract and finally sample was totally dried under gentle nitrogen stream. Solid residue was re-dissolved in 1 ml acetonitrile and mixed well with vortex mixture for HPLC analysis [25]. For preparation of water samples, liquid liquid extraction method was adopted. Extraction of water samples of 1 L was performed in a separatory funnel with 2 × 30 ml n-hexane by using a shaking incubator (SI-100R, Korea) at 120 rpm and at a temperature of 20 °C, for 15 minutes. After extraction, combined extracts were dried by-passing through anhydrous sodium sulfate and later on reduced to 0.5 ml by using a rotary evaporator (Rotavapor Büchi RE-210, coupled with a water bath Büchi 491, BUCHI, Flawil, Switzerland). Nitrogen stream was used to completely dry the sample and

remaining solid residue was re-dissolved in 1 ml acetonitrile and mixed well at vortex mixture for further analysis [26]. For pretreatment of leaf samples, 7.5 g leaves were extracted with ethyl acetate (75 ml) at 38 °C for 20 minutes in an ultrasonicator. Insoluble materials and leaves were separated by decantation through a membrane filter [27], next steps were same as previously described for soil and water samples. Walkley-Black acid digestion method was used to estimate soil (organic matter) and multimeter (CRISON MM 40⁺) was used to calculate soil pH in a 1:1 suspension (soil: water).

Analysis of samples

High performance liquid chromatography (HPLC) SPD- 10A VP- Shimadzu with UV detection (254 nm), was used for quantitative analysis of soil, water and leaf samples. Acetonitrile and double deionized water in the ratios of 6:4 (v/v), respectively, was used as mobile phase at a flow rate of 1.25 ml per minute and oven temperature of 40 °C, and sample injection volume was 10 ul. Within a total run time of 25 minutes, all the three PAH could be selectively quantified. Concentrations of PAH in the target samples were calculated by calibration curve method, which was developed by running the reference standards and the recovery rates obtained during validation were in range from 80-91%. Main statistical parameters, obtained from experimental data were generated with SPSS 16.0 for windows. In analysis of variance (ANOVA), mutual relationship between mean concentrations of selected PAH in the selected media (soil, water, and plant leaves) and between selected sites is established, further mean concentrations of soil PAH and organic matter and pH of the soil are also correlated.

Results and discussion

The PAH assessment in soil

Naphthalene, phenanthrene, and pyrene are quantified by HPLC in soil and mean values (mg/kg) of the contaminants with standard error bars are presented in fig. 2. The ANOVA test showed a significant difference between the mean values of naphthalene, phenanthrene, and pyrene in soil at different sampling sites, tab. 1. However, a non-significant difference (P -value = 0.36) among mean concentrations of naphthalene, phenanthrene and pyrene in soil was recorded at four sites, tab. 2.

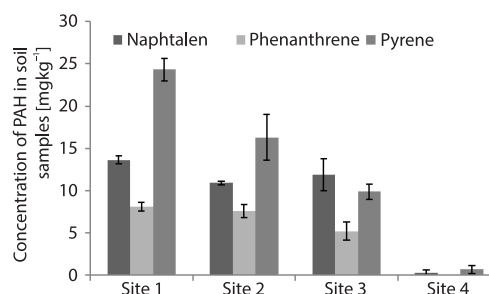


Figure 2. Comparison of selected PAH (mg/kg) in soil samples at four sampling sites

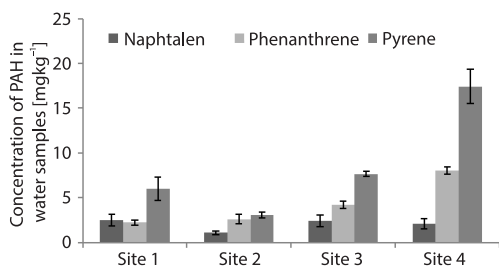
Table 1. One-way ANOVA results for selected PAH in soil, water and plant samples

PAH at different sampling sites	P -value	PAH at different sampling sites	P -value
Naphthalene in soil	0.00013	Pyrene in water	$1.5 \cdot 10^{-5}$
Phenanthrene in soil	0.06391	Naphthalene in plant	0.11402
Pyrene in soil	0.00465	Phenanthrene in plant	0.00534
Naphthalene in water	0.34714	Pyrene in plant	0.00044
Phenanthrene in water	$5.11 \cdot 10^{-8}$		

Table 2. One-way ANOVA results for selected PAH at different sampling sites

		Sum of squares	DoF	Mean square	P-value
Soil	Between groups	114.23	2	57.11	0.36
	Within groups	451.10	9	50.12	
	Total	565.33	11		
Water	Between Groups	087.83	2	43.91	0.11
	Within groups	138.50	9	15.39	
	Total	226.33	11		
Plant	Between groups	095.21	2	47.61	0.35
	Within groups	364.84	9	43.54	
	Total	460.06	11		

The highest mean concentration of naphthalene (13.63 mg/kg) in soil was recorded for samples collected from Site 1, whereas lowest mean concentration of naphthalene (0.17 mg/kg) was observed for Site 4 with a 98.75% less concentration compared with Site 1. Similarly, average concentration of phenanthrene in soil among four sites decreased from Sites 1-4 (99.75 % less content compared with Site 1). Highest mean value of phenanthrene (8.11 mg/kg) was obtained near wastewater treatment plant within refinery, whereas average values of 7.57 mg/kg and 5.21 mg/kg were obtained for exit point of main drainage of oil refinery and near the incinerator area, respectively. The lowest mean value (phenanthrene: 0.02 mg/kg) was obtained for populated area in Morgah. For pyrene, the highest mean concentration (24.31 mg/kg) in soil was recorded for samples collected from Site 1, whereas lowest mean concentration of pyrene (0.66 mg/kg) was observed in the soil samples of Site 4 (99.75% less content compared with Site 1). All PAH concentrations at Site 4 were lower than the safe limit of sum of PAH in soil *i.e.* is 1.5 mg/kg for residential areas. The highest mean values of three measured PAH (naphthalene, phenanthrene, and pyrene) observed in the soil samples of Site 1, pertained to continuous influx of these PAH from petroleum processing and deposition at the nearest point and in concord with the findings of Bakker *et al.* [28]. At Sites 2 and 3, almost equal content of selected PAH was found in the samples of soil, indicating direct deposition from wastewater of the main drainage of the oil refinery, on the surrounding soil [29]. At Site 3, incineration of medical waste from a local hospital produce organic pollutants including PAH, that could increase concentration of naphthalene, phenanthrene and pyrene in soil [30, 31]. Furthermore, atmospheric deposition of the selected contaminants may influence their concentration in soils at Sites 2 and 3 [32], and almost negligible concentrations of these compounds at Site 4 could be attributed to their re-evaporation from the soil or biodegradation within soil [33].

**Figure 3. Comparison of selected PAH (mg/kg) in water samples at four sampling sites**

The PAH Assessment in water samples

Figure 3 shows mean values of naphthalene, phenanthrene, and pyrene in water samples, at selected sites and ANOVA results show a non-significant relationship among them, tab. 1. The recorded values of naphthalene level in water samples for Site 1 (2.50 mg/L), Site 2 (1.08 mg/L), Site 3 (1.07 mg/L), and Site 4 (2.05 mg/L) did not show any significance difference (tab. 2, p -value = 0.11).

Mean values of phenanthrene in water samples show highly significant difference among four sites. Highest mean value of phenanthrene was observed at Site 4 (8.0 mg/L) and it was found that mean concentration of phenanthrene has significantly increased from Sites 1-4 in water samples, as compared to soil and plant samples. Similarly, highest mean value of pyrene of 17.4 mg/L was also assessed at Site 4, while lowest water pyrene level of 3.0 mg/L was observed at Site 1. It was observed that mean water pyrene level at selected sites vary significantly (p -value = 0.00). These values are higher than the maximum concentration limit for benzo(a)pyrene in water which is 0.0002 mg/L (US EPA).

Higher mean values of phenanthrene and pyrene in water samples at Site 4 is accredited to the decrease in the flow of wastewater that resulted in the high concentration of assessed PAH [34]. Moreover, high content of PAH, may be attributed to their high resistance to biodegradation and high persistence in water bodies [35]. Naphthalene is highly volatile dicyclic compound among selected PAH, so might be re-evaporated at the same time it was deposited on the water surface, as its concentration is lower than pyrene and phenanthrene concentrations, at Site 1. Interestingly, almost persistent concentration of naphthalene obtained in water samples from the all selected sites, indicate atmospheric deposition of naphthalene from refinery and also from vehicular emissions as selected study area is profoundly populated.

The PAH assessment in plant samples

The ANOVA results show non-significant difference (p -value = 0.11) in the mean concentration of naphthalene in leaves of *Melia azedarach L.*, tab. 2, at different sampling sites. Highest mean concentration of naphthalene was found for Site 1 *i.e.*, 13.48 mg/kg. Leaf samples of Sites 2 and 3 showed average concentration of 2.91 mg/kg and 2.01 mg/kg, respectively, and lowest average content of naphthalene 0.12 mg/kg was observed at Site 4 (99.11% less content compared with Site 1), fig. 4.

Level of phenanthrene vary significantly in leaf samples for four selected sites (p -value = 0.00). Mean values of phenanthrene successively decreased from Site 1 (17.64 mg/kg) to Site 4 (1.54 mg/kg) in leaf samples with a 91.11% less phenanthrene than mean value at Site 1. In case of mean concentration of pyrene in leaf samples, a highly significant difference was found among four sites (p -value = 0.00). Pyrene showed highest mean concentration in plant samples at Site 1 (18.88 mg/kg) and lowest mean concentration (3.11 mg/kg) was found at Site 4, with a 89.39% less pyrene than Site 1. At Site 1, selected PAH profile of plant samples (leaves) was similar to soil samples and its reason is the presence of a point source, *i.e.*, petrochemical activities in the oil refinery and also bioconcentration potential of selected plant species, *i.e.*, *Melia azedarach L.* However, selected PAH levels were higher than the safe limits in plants according to US EPA (0.0002 mg/kg). High concentration of three selected PAH obtained in plant leaves can also be attributed to the absorption of these lower molecular weight PAH, both via stomatal openings and diffusion through cuticle layer on the leaf surface [36]. Bakker *et al.* [28] also reported high concentration of PAH near an oil refinery in plants, though they focus on seven PAH congeners and a total of 2 mg/kg was obtained for all selected compounds. Lower mean values of naphthalene in *Melia azedarach L.* at Sites 2-4 was ascribed to its volatility as naphthalene can easily dissipate into the atmosphere, away from

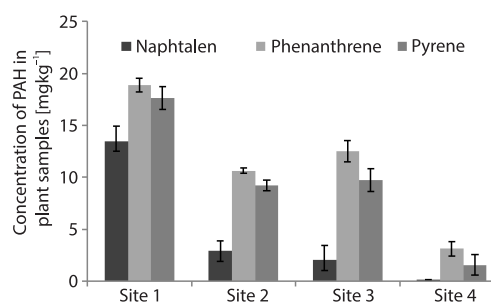


Figure 4. Comparison of selected PAH (mg/kg) in plant samples at four sampling sites

the point source and therefore, less available to accumulate in the plant [37]. Other than *Melia azedarach L.*, various other plant species were grown in the study area, might be attributed in biodegradation of this contaminant in the soil and less available to the plant [38]. At Site 3, slightly higher mean content of phenanthrene and pyrene obtained in leaf samples than at Site 2, can be accredited to incineration of medical waste at Site 3 [33]. Unexpectedly, high mean concentration of pyrene obtained at Site 4, could be attributed to the fact that this compound has more aromatic rings (four fused rings) compared with two other compounds (naphthalene and phenanthrene has two and three rings, respectively). It is speculated that; pyrene is more lipophilic and plants must absorb pyrene from particles of soil and destined in the plant tissues [13].

Relationship of soil organic matter and pH with PAH

The average values obtained for soil organic matter at selected sites were in range of 4.99-2.91% and soil pH average values observed at selected sites were 8.06 (Site 1), 8.09 (Site 2), 7.51 (Site 3), and 7.41 (Site 4). Relationship of soil organic matter and pH with mean concentration of naphthalene, phenanthrene and pyrene is shown in tab. 3. The PAH levels of soil are not significantly correlated with organic matter of the soil; likewise, Kordybach *et al.* [39] reported non-significant correlation between PAH and soil organic matter. However, mean concentration of phenanthrene and pyrene in soil is significantly correlated with soil pH.

Table 3. Correlation of soil organic matter and pH with PAH along drainage of oil refinery

	OM**	pH	Naphthalene	Phenanthrene	Pyrene
OM**	1.00				
pH	0.68	1.00			
Naphthalene	0.27	0.55	1.00		
Phenanthrene	0.56	0.83	0.73*	1.00	
Pyrene	0.57	0.69	0.85*	0.77	1.00

*Correlation is significant at the 0.05 level (2-tailed); **OM – organic matter

Conclusion

Petroleum processing activity greatly influences the environment in the vicinity of the oil refinery. To assess three targeted PAH contaminants near oil refinery, soil, water, and plant leaf samples (*Melia azedarach L.*) are collected. Results of present study reveal that, distance plays a key role in the contamination level of naphthalene, phenanthrene, and pyrene within soil, water, and plant samples, which gradually decreased as distance increased from the point source. However, naphthalene showed higher concentration in water samples at Site 4. The concentration of selected PAH was found to be higher in soil and plant samples than in water samples and pyrene was the most abundant PAH among selected congeners. Present study focused on three PAH analytes and for the future work, it is recommended that work should be expanded to develop sixteen priority PAH emission inventory data of the study area for control of such compounds at the source. Moreover, installation of proper pollution control equipment is highly recommended for oil refinery to reduce PAH emissions in the surrounding area and exposure to public.

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