

## ORIGINAL ARTICLE



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#### \*Corresponding author.

Samantha Sivaswamy

Research Scholar, Department of Environmental Health Engineering, Sri Ramachandra Institute of Higher Education and Research, Porur, 600116, Chennai, India  
[samantha\\_1412@yahoo.com](mailto:samantha_1412@yahoo.com)

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## Urinary 1-Hydroxypyrene levels among children with asthma in Chennai, South India

Samantha Sivaswamy<sup>1\*</sup>, Sankar Sambandan<sup>2</sup>, Naveen Puttusamy<sup>3</sup>, Padmavathi Ramasamy<sup>4</sup>, M Surianarayanan<sup>5</sup>, Sudhakar Rao Saidam<sup>6</sup>

<sup>1</sup> Research Scholar, Department of Environmental Health Engineering, Sri Ramachandra Institute of Higher Education and Research, Porur, 600116, Chennai, India

<sup>2</sup> Professor and Head, Department of Environmental Health Engineering, Sri Ramachandra Institute of Higher Education and Research, Porur, 600116, Chennai, India

<sup>3</sup> Assistant Professor, Department of Environmental Health Engineering, Sri Ramachandra Institute of Higher Education and Research, Porur, 600116, Chennai, India

<sup>4</sup> Associate Dean of PG studies, Department of Physiology, Sri Ramachandra Institute of Higher Education and Research, Porur, 600116, Chennai, India

<sup>5</sup> Principal Scientist, Cell for Industrial Safety and Risk Analysis, Chemical Engineering Department, Central Leather Research Institute, Chennai, 600020, India

<sup>6</sup> Field/Lab Manager, HAPIN Project, Department of Environmental Health Engineering, Sri Ramachandra Institute of Higher Education and Research, Porur, 600116, Chennai, India

### Abstract

**Aim:** To explore the potential relationship between exposure to Polycyclic Aromatic Hydrocarbons (PAHs) and asthma exacerbation among children and also to explore the potential exposure sources of PAHs at the household level.

**Methods:** Case-control study was conducted at a tertiary care hospital in Chennai among 60 participants after obtaining approval from the Institutional Ethics Committee. The study was conducted from March till May 2018. Informed consent was obtained from the participants. The participants were included based on their clinical diagnosis of asthma and few participants were used as control based on no documented history of wheeze and PEF levels. All the participants were male children aged between 9-12 years and exclusion was done if they were reported to having congenital heart disease, surgery or chronic lung disease. They were interviewed with a standardized questionnaire to ascertain their exposure to PAHs sources. Urinary 1-OHP levels were analyzed using HPLC to determine their exposure. **Results:** Participants with asthma showed a higher concentration of 1-OHP (7.56 µg/g) in their urine. Second hand exposure to tobacco smoke, consumption of charred or grilled foods, use of indoor air pollutants and exposure to traffic fumes were identified as contributive factors to elevated 1-OHP levels. **Conclusion:** Exposure sources at household levels may play a major role in exacerbating asthma. Significant PAHs exposure was contributed through dietary habits. Further exposure monitoring studies are needed to quantify the impact of PAH exposure on respiratory health.

**Keywords:** Polycyclic Aromatic Hydrocarbons; Asthma; 1-Hydroxypyrene



## 1 Introduction

Polycyclic aromatic hydrocarbons (PAHs) are formed due to incomplete combustion of organic substances. Diesel exhaust particles (DEP) emitted from vehicles are major sources of PAHs in urban areas<sup>(1)</sup>. Blood and urine PAHs levels are biomarkers of interest as they can represent PAHs exposure through inhalation and ingestion of contaminated foods<sup>(2)</sup>. Chronic exposure to air pollution has been associated with exacerbation of asthma. These studies mainly focus on traffic related emissions which are made up of particulate matter, DEP, oxides of Nitrogen and Sulphur, volatile organic compounds and PAHs in the initiation of respiratory symptoms and disorders<sup>(3)</sup>. Recent studies have revealed that chronic exposure to PAHs can be associated with adverse respiratory health outcomes such as increased cough and wheeze in infants that may lead to development of breathing problems<sup>(4)</sup>. In a recent survey by the U.S. National Health and Nutrition Examination Survey (NHANES) (1999–2000, 2001–2002), higher levels of PAHs were detected among children, suggesting that they may face a higher risk for negative health effects<sup>(5)</sup>. Another significant factor that may contribute to PAHs exposure is the usage of biomass fuels. Burning of biomass fuels releases dense fumes into the air that consists of particulate matter, oxides of carbon and poly organic and polycyclic aromatic hydrocarbons<sup>(6)</sup>. Recent literature has showed evidence that there exists a negative association between PAHs exposure and asthma in children without prenatal exposure to environmental tobacco smoke<sup>(7)</sup>. In another study by Awasthi et al.<sup>(8)</sup>, a higher blood level of phenanthrene was observed in cases of bronchial asthma. Childhood asthma is believed to be associated with an increased generation of Reactive Oxygen Species (ROS) in the body leading to oxidative stress, which in turn can lead to development of respiratory distress. This study aims to identify the relationship between PAHs and asthma among children from selected parts of Chennai.

## 2 Materials and Methods

### 2.1 Study population

This was a case-control study conducted among 60 participants recruited from a tertiary care hospital in Chennai, between March and May 2018. Only male children of age 9-12 years were recruited to avoid gender confounding<sup>(9)</sup>. The participants included 30 asthmatic children and 30 non-asthmatic children. Recruitment of cases was done based on physician diagnosed asthma. Controls were screened and included based on the ISAAC questionnaire, which assess prevalence of wheeze and asthma symptoms. After obtaining informed consent, the child was subjected to a preliminary lung function test using a hand-held PEFR instrument. Children with normal PEFR rates, no history of clinical diagnosis of asthma and reported no respiratory symptoms to the ISAAC questionnaire were recruited to be the controls. Participants were excluded if they reported having congenital heart disease, chronic lung disease like suspected cystic fibrosis, bronchiectasis or lung abscess. Urine samples were collected from all the participants. Urinary concentration of 1-hydroxypyrene was analyzed as a biomarker of PAH exposure. The questionnaires were administered to the parent or guardian of the child below the age of 13. Anthropometric measurements were then obtained for all participants. The parents were interviewed briefly to collect data regarding exposure to traffic source, indoor air pollution sources, industrial or commercial sources of pollution, food habits, fuel use characteristic and time of activity patterns. A spot urine sample of minimum 30 ml of volume was collected after providing proper instructions to the participants.

### 2.2 Analysis of urinary 1-Hydroxypyrene

Urine samples collected were immediately quantified for creatinine using a semi-automated analyzer (ERBA Mannheim). After creatinine analysis, samples were aliquoted and stored at -20° C, within 4 h after sampling. They were thawed at room temperature and taken for analysis. The samples were centrifuged at 5000 rpm for 5 mins. An amount of 2 ml of urine sample was then added with 3 ml of sodium acetate buffer and 10  $\mu$ l  $\beta$ -glucuronidase enzyme for hydrolysis. After 16 hours of incubation, analytes were extracted using C18 cartridges packed in 3 cc tubes of 500 mg each, at room temperature. After extraction, the samples were concentrated using nitrogen gas. It was reconstituted with methanol. The extract was then loaded into HPLC-2010 AHD Shimadzu with fluorescence detector RF-NAXL detector. Analyte separation was performed on a C18 column (250 mm). The samples were creatinine corrected before further analysis<sup>(10)</sup>.

## 3 Results

The demographic characteristics of the participants are provided in Table 1. All participants were comparable with regard to age and anthropometry.



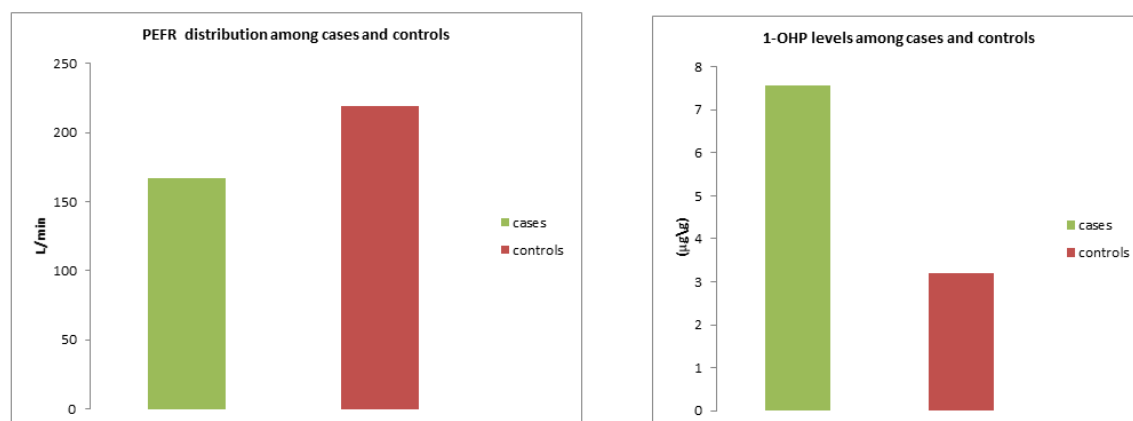
**Table 1. Demographic features of the participants**

	Controls (N= 30)	Cases (N=30)
Age	10.37±2.06	9.33±3.09
Height (cm)	135.45±16.8	132.59±19.7
Weight (Kg)	30.33±9.71	30.18±17.56
BMI	16.26±2.91	15.901±4.40
PEFR (L/Min)	218.74	164.44

*Mean + SD*

### 3.1 Prevalence of 1-Hydroxypyrene levels in urine and potential sources of PAHs exposure

The results were analyzed to examine the relationship between PEFR and 1-OHP levels among the two groups of participants as shown in Figure 1. PEFR levels were lower in children with asthma as compared to healthy children. The mean PEFR of cases and controls were observed to be 165.56 L/min and 218.74 L/min respectively. Mean 1-OHP levels were slightly higher in children with diagnosed asthma (7.56  $\mu\text{g/g}$ ) than healthy children (3.19  $\mu\text{g/g}$ ).

**Fig 1.** Comparison of mean PEFR levels and urinary 1-OHP levels among cases and controls

The difference in 1-hydroxypyrene levels between Cases and Control can depend on a range of exposures sources in the indoor and ambient environments. Table 2 examines the role of potential PAH exposure sources that may influence elevation in 1-OHP levels.

**Table 2. 1-OHP levels with respect to possible sources of PAHs exposure**

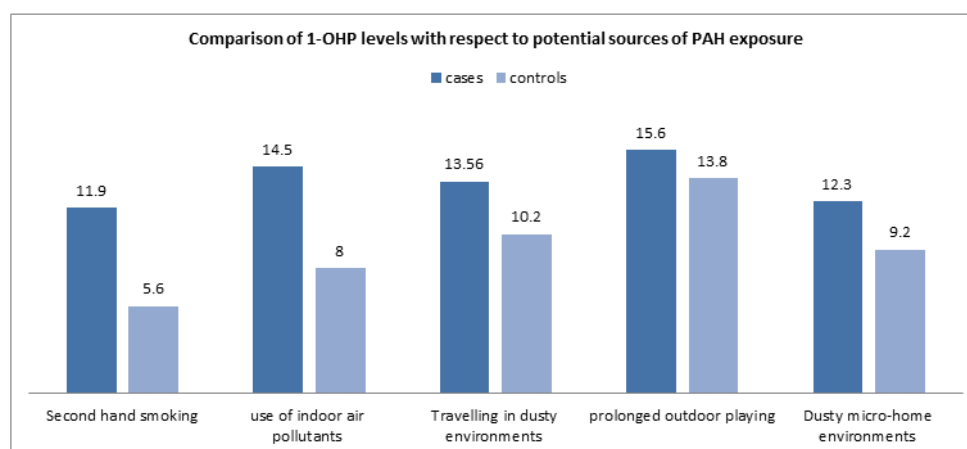
	Controls ( $\mu\text{g/g}$ )	Cases ( $\mu\text{g/g}$ )	P-Value
Presence of a smoker in the household	5.86±3.13	11.9±12.66	0.038*
Household vicinity to highways	2.73±1.36	9.99±12.33	0.08
Heavy traffic density	2.82±1.11	13.32±14.06	0.23
Vicinity to industry	6.07±9.62	10.6±14.9	0.55
Waste burning near household	2.4±1.13	5.37±4.35	0.15
Indoor usage of kerosene lamps / agar-bathis	3.6±3.93	14.02±17.4	0.043*
Consumption of grilled foods	3.3±2.56	9.06±12.97	0.032*
Using indoor air pollutants.	4.28±9.82	6.74±9.52	0.046*

*Mean + SD*

Certain exposure variables were observed to be significant at 0.05 level. Other exposure variables that showed considerable



difference between the groups are represented in Figure 2.



**Fig 2.** Distribution of urinary 1-OHP levels ( $\mu\text{g/g}$ ) between cases and controls with respect to exposure sources

The participants were between 9-12 years of age. They were distributed among the lower middle (48%), upper middle (32%) and upper lower classes (20%), of the socio-economic strata. The family type most commonly observed was nuclear (82%). Family size observed were mostly 4-5 people per household. It was observed that single-child household was very rarely present among the participants (4%). Siblings of participants who were in the criteria age range were included in the study or were selected as controls in some households.

This gave both the groups of participants a shared exposure environment. About 30% of the participants had an active smoker in the household. It was reported during the study that about 24% were actively exposed to second-hand smoke in their households or neighborhood.

It was observed that cases who had a smoker in the household exhibited higher urinary 1-OHP concentration, similar to a study conducted in Taipei<sup>(11)</sup>.

Previous studies report smoking, effects of grilled food consumption and proximity to roadways as a major source of PAHs exposure for non-occupational exposure<sup>(12)</sup>. About 20% of the participants reported having contact with pet or stray animals. About 16% of the households in the cases category reported using kerosene and biomass fuel for bathing and other domestic purposes, which may also be an attributing factor for the elevated levels observed. Similar results were seen in a study in Nigeria which showed an association between biomass fuel usage and severe symptoms of asthma with an odds of ratio of 2.37. About 40% of the houses were situated less than 10 meters from the highways and reported of deposition diesel exhaust particles on window screens and curtains. An elevated level of 1-OHP was observed in these households though the difference was not proved significant. They reported heavy duty vehicles and light duty commercial vehicles plying near their homes frequently at moderate volume. These results were similar to a study done in Italy, where there was a weak association between moderate traffic and asthma symptoms with an odds ratio of 1.24<sup>(13)</sup>. Elevated levels of urinary PAHs were seen in participants who reported burning their household waste regularly in open areas near their homes. Few of the parents of the participants reported sneezing and wheezing, due to their exposure to construction debris and dust nearby. About 60% of the participants reported playing outdoors at schools, near roads and open grounds for an average of 30 to 45 mins every day. This may be a contributing factor for asthma exacerbation as discussed in a similar study in Sweden, where house dust was associated with asthma symptoms in children<sup>(14)</sup>. About 70% of the participants reported commuting to school and other places for an average duration of 15 -30 mins every day in moderate traffic density.

In a study at Minnesota, asthma exacerbations were observed to rise with increase in traffic density and proximity<sup>(15)</sup>. Consistent higher concentrations of 1-OHP were seen in cases who consumed charred or grilled food on an average of 3-4 times a week, compared to their controls. This was similar to a study in Baltimore where ingestion of charred and boiled foods were associated with elevated levels of urinary 1-OHP<sup>(15)</sup>. Participants from households who used indoor air pollutants like mosquito coils, repellants and various types of incenses were observed to show elevated levels of 1-OHP. This may be due to higher exposure to PAHs emissions in a closed environment as observed in a similar study in Taiwan<sup>(16)</sup>. Time activity of the participants



plays a major role in identifying potential exposure sources. The participants who spent significant amount of their time in dusty environment during playing or commuting were observed to have higher levels of 1-OHP concentration than participants who spent less than an average of 60 minutes outdoors every day. Dietary consumption of charred food may have been a confounding factor because of its positive influence on 1-OHP. This may have been responsible for the absence of attributable associations to air pollution. Exposure to second-hand smoke, consumption of charred food, traffic-related pollution in urban areas and usage of indoor air pollutants were observed to cause an elevation in 1-OHP levels. They may serve as predictors to assess the exposure to PAHs with additional evidence<sup>(17,18)</sup>.

## 4 Conclusion

Traffic fumes, ETS and dietary intake of contaminated food have been reported as having an adverse impact on respiratory health in this study. This study strengthens the belief that PAHs adsorbed to dust particles may have an association with the exacerbation of asthma symptoms in children. Though the association between PAH and asthma is not definitively clear, this study provides an insight into various contributing factors that may play a role in the etiology of asthma. Further molecular based research is needed to quantify the exact effect of the reported factor that have shown promise in this study, as potential exposure sources at the household. This can lead to development of source based control strategies to reduce the impact of asthma on children.

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## References

- 1) Strickland P, Kang D, Sithisarankul P. 1996.
- 2) van Schooten FJ, Moonen EJC, van der Wal L, Levels P, Kleinjans JCS. Determination of Polycyclic Aromatic Hydrocarbons (PAH) and Their Metabolites in Blood, Feces, and Urine of Rats Orally Exposed to PAH Contaminated Soils. *Archives of Environmental Contamination and Toxicology*. 1997;33:317–322. doi:10.1007/s002449900259.
- 3) Balakrishnan K, Ramaswamy P, Sambandam S, Thangavel G, Ghosh S, Johnson P, et al. Air pollution from household solid fuel combustion in India: an overview of exposure and health related information to inform health research priorities. *Global Health Action*. 2011;4:5638–5638. doi:10.3402/gha.v4i0.5638.
- 4) Leem JH, Kim JH, Lee KH, Hong YC, Lee KH, Kang D, et al. Asthma attack associated with oxidative stress by exposure to ETS and PAH. *Journal of Asthma*. 2005;42(6).
- 5) Burney PG, Chinn S, Rona RJ. Has the prevalence of asthma increased in children? Evidence from the national study of health and growth 1973–86. *BMJ*. 1990;300(6735):1306–1310. doi:10.1136/bmj.300.6735.1306.
- 6) Awasthi S, Agarwal S. Determinants of childhood mortality and morbidity in urban slums in India. *Indian pediatrics*. 2003;40:14722365–14722365.
- 7) Huang X, Zhou Y, Cui X, Wu X, Yuan J, Xie J, et al. Urinary polycyclic aromatic hydrocarbon metabolites and adult asthma: a case-control study. *Scientific reports*. 2018;8:1–8. Available from: <https://doi.org/10.1038/s41598-018-26021-3>.
- 8) Awasthi S, Mahdi AA, Patel DK, Singh VK, Rita M, Suresh R. Assessment of association of exposure to polycyclic aromatic hydrocarbons with bronchial asthma and oxidative stress in children: A case control study. *Indian Journal of Occupational and Environmental Medicine*. 2009;13:33–33. doi:10.4103/0019-5278.50722.
- 9) Almqvist C, Worm M, Leynaert B. working group of GA2LEN WP 2.5 ‘Gender’. Impact of gender on asthma in childhood and adolescence: a GA2LEN review. *Allergy*. 2008;63(1):47–57.
- 10) Jongeneelen FJ, van Leeuwen FE, Oosterink S, Anzion RB, van der Loop F, Bos RP, et al. Ambient and biological monitoring of cokeoven workers: determinants of the internal dose of polycyclic aromatic hydrocarbons. *Occupational and Environmental Medicine*. 1990;47(7):454–461. doi:10.1136/oem.47.7.454.
- 11) Wang IJ, Karmaus WJJ, Yang CC. Polycyclic aromatic hydrocarbons exposure, oxidative stress, and asthma in children. *International Archives of Occupational and Environmental Health*. 2017;90:297–303. doi:10.1007/s00420-017-1198-y.
- 12) & Yuan TH, & Shie RH, & Chin YY, Chan, Chang-Chuan. 2015.
- 13) Migliore E, Berti G, Galassi C, Pearce N, Forastiere F, et al. Respiratory symptoms in children living near busy roads and their relationship to vehicular traffic: results of an Italian multicenter study (SIDRIA 2). *Environmental Health*. 2009;8. doi:10.1186/1476-069x-8-27.
- 14) Bornehag CG, Sundell J, Weschler CJ, Sigsgaard T, Lundgren B, Hasselgren M, et al. The Association between Asthma and Allergic Symptoms in Children and Phthalates in House Dust: A Nested Case–Control Study. *Environmental Health Perspectives*. 2004. Available from: <https://dx.doi.org/10.1289/ehp.7187>. doi:10.1289/ehp.7187.
- 15) Lindgren P, Johnson J, Williams A, et al. Asthma exacerbations and traffic: examining relationships using link-based traffic metrics and a comprehensive patient database. *Environmental Health*. 2016;15. doi:10.1186/s12940-016-0184-2.
- 16) Tang CS, Lung SC, Chang TY, Tu HH, Chang LT. 2019.
- 17) Sun Z, Zhu D. Exposure to outdoor air pollution and its human health outcomes: A scoping review. *PloS one*. 2019;14(5).
- 18) Freire C, Abril A, Fernández MF, Ramos R, Estarlich M, Manrique A, et al. Urinary 1-hydroxypyrene and PAH exposure in 4-year-old Spanish children. *Science of The Total Environment*. 2009;407(5):1562–1569. doi:10.1016/j.scitotenv.2008.10.068.