ORIGINAL ARTICLE

INDOOR AIR QUALITY AND ITS ASSOCIATION WITH RESPIRATORY HEALTH AMONG PRESCHOOL CHILDREN IN URBAN AND SUBURBAN AREA

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ABSTRACT

Indoor air quality (IAQ) has become a major concern nowadays because of the universality of exposure and its potential negative impact on human health especially on children. This study is intended to explore the association between IAQ and the respiratory health among preschool children in urban and suburban area. A cross-sectional comparative study was carried out among Malay preschool children in urban (N= 60, Puchong) and suburban (N=60, Hulu Langat) areas. An indoor air quality assessment was conducted in 12 preschools and 60 houses which include parameters of $PM_{2.5}$, PM_{10}, VOCs, mold, bacteria, Gram-negative bacteria and physical parameters. A set of standardized questionnaire was distributed to obtain respondents' background information, exposure history and respiratory health symptoms. Spirometry test was carried out and the data obtained were analyzed to determine the lung function of the respondents. There was a significant difference between IAQ in urban and suburban preschools for all parameters measured ($p<0.05$). Most of the pollutants were significantly associated with respiratory health symptoms. There was a significant association between the level of indoor pollutants with the lung function abnormalities among the respondents. Even though this study is the first to take Gram-negative bacteria as an indoor air pollutant, the finding also shows that there is a significant association between exposure of Gram-negative bacteria with lung function impairment and higher reported respiratory symptoms among the respondents. The finding concluded that exposures to indoor air pollutants, especially $PM_{2.5}$ increases the risk of getting lung function abnormality and respiratory health symptoms among respondents.

Keywords: *Indoor air quality, mold, bacteria, lung function, respiratory health symptoms*

INTRODUCTION

Airborne pollution has become a major concern nowadays because of the universality of exposure and its potential negative impact on human health. As the average human spends approximately 90% of their time indoors[1] the indoor environment is highly influenced the total daily exposure to air pollutants. According to United State Environmental Protection Agency (USEPA), Indoor Air Quality (IAQ) refers to the air quality within and around buildings and structures, especially as it relates to the health and comfort of building occupants[2]. IAQ is determined by exposure from a combination of countless pollutants that originated from various sources such as indoor smoking, cooking activities, paint, wall, air cooling system and even outdoor air toward building residents. On the other hand, common indoor air pollutants detected in schools varied from particulate matter, volatile organic compound (VOC), aldehydes, bacteria and molds[3].

Among the different age groups, IAQ is believed to affect the health of preschool children significantly[4]. Children's lungs are still growing and they breathe differently from adults. Their immune systems are not fully developed, and so infections are common[5]. Thus, exposure to indoor air pollutants can contribute to various short-term and long-term health effects such as respiratory infections, allergies and asthma[6]. In Malaysia, most of the parents send their children to kindergarten or preschool as a preparation of learning before going to the real school. The Malaysian statistics on preschool education shows the existence of 15,627 preschool classes in government preschools and 17,899 in government agencies preschools and the average class ratio of 1 teacher to 24 children with a class size of 23 children[7]. Children that enrolled in these classes spent at least half a day every day of their time during weekdays. Within the classes, children can be exposed to various types of indoor air pollutants including other types of illnesses carried and transmitted by their own classmate.

Therefore, the aim of this study is to determine the association between indoor air quality and respiratory health among preschool children in Puchong and Hulu Langat, Selangor. The finding of this study is relevant to public health as it focuses on association of air pollutants with children's lung condition and can be useful for improving the indoor environmental setting in Malaysian preschools. This study was also the first to assess Gram-negative bacteria as an indoor air pollutant because of their unique ability to produced specific toxin called endotoxin. The inhaled endotoxin has been associated with many pulmonary diseases. More than 10 ng/m^3 can cause airway inflammation, 100 ng/m^3 gives systemic effects and 200 ng/m^3 shows toxic pneumonitis[8]. The finding from this study can be used as a baseline data for exposure of Gram-negative bacteria towards preschool children in Selangor for future research.

METHODS

This cross-sectional comparative study was carried out in six preschools from Puchong as the urban (exposed) area and another six from Hulu Langat as representing the suburban (comparative) area. A total of 120 preschool children aged 5 to 6 years old from preschools in Puchong and Hulu Langat was included in this study. From that, both male and female students were randomly selected and from the Malay ethnic background. Only Malay preschool children were included in this study to homogenize the sample and to avoid genetic as a confounding factor. This study applies a stratified random sampling method to determine the preschool that being studied. Then all students in the selected preschool are choosing based on willingness to participate after parents' approval and after taking consideration of several inclusive criteria such as 5-6 years old, Malays and free from any respiratory illnesses. Incomplete questionnaire and children without parents' approval will be excluded from this study.

Measurements of $PM_{2.5}$, PM_{10}, VOC, fungi, bacteria and Gram-negative were taken at 0.6-1.0 meter above the floor to represent the breathing zone level of the preschool children and more than 1 meter away from the wall, door and any active heating system. The number of sampling point was 1 for each classroom plus one point from outdoor. The equipments were placed at a location that the children spent time the most and assuming even distribution of pollutants. The measurements were taken every one hour from 8.00 am till 12.00 pm except for biological parameters. All methods were based on standard method by Industry Code of Practice on Indoor Air Quality 2010[9]. The IAQ monitoring instruments used in this study including DustTrak™ DRX Aerosol Monitor 8534 for $PM_{2.5}$ and PM_{10}, ppbRAE Portable VOC Monitor model PGM-7240 for VOCs, TSI Q-Trak™ Indoor Air Quality Monitor 7575 for temperature and relative humidity and TSI VelocicalC Plus Model 8386 for air velocity.

For biological parameters, pbi DuoSAS Super 360 was used to drawn air from the surrounding area onto culture agar to get the mold, bacteria and Gram-negative bacteria. The assessment was based on 500 liters of air sample with duplicate measurement: one reading around 9.00 am and another reading around 11.00 am. The measurement was taken simultaneously for mold, bacteria and Gram-negative bacteria. Sabaroud Dextrose Agar (SDA) was used to isolate mold, Tryptic Soy Agar (TSA) to isolate bacteria and MacConkey agar for Gram-negative bacteria. After sampling, all agar media plates were sealed and transported to the laboratory for incubation. TSA and MacConkey agar were incubated at 37ºC for 24 hours before calculation for colony forming. Whereas for SDA, samples were left at room temperature for 5 days before conduct the colony counting[10]. Quantification of mould and bacteria levels was performed by naked eye count according to EN 13098: 2000 and ISO 4833:2013[11].

This study used modified questionnaire based on two internationally standardized and recognized questionnaires which are questionnaire set by the American Thoracic Society (ATS) and questionnaire on asthma, allergies and respiratory symptoms from the International Study of Asthma and Allergies in Childhood (ISAAC) study[12]. The ATS questionnaire was used to obtain data on demographic and socioeconomic background of the respondents as well as data on preschool and home environment. Besides that, ISAAC questionnaire was used to gather data on the respondents reported respiratory symptoms.

The lung function or spirometry test was conducted by using Chestgraph HI-101 spirometer. Prior to the test, anthropometric measurement was first obtained where the height of respondents was measured by using height scale model "SECA 208 Body Meter". The respondents were instructed to stand in a straight position and barefooted. Then, body weight was measured by using an electronic weighing scale. The weighing scale was placed on a flat surface during measurements.

All data were analyzed by using Statistical Package for Social Science (SPSS) Version 21.0. The descriptive test was used to calculate mean, median, standard deviation and interquartile range. Meanwhile, Kolmogorov Smirnov test and skewness was used to determine the normality of the data. Besides that, t-test, Mann-Whitney U test and chi-square test were used to determine the differences and associations between indoor air pollutants and respiratory health among respondents. Multiple logistic regression test was used to determine the main factors in influencing the respiratory health among the preschool children.

The modified questionnaire used in this research was pre-tested on 10% of the total respondents to ensure its reliability and validity. The result was tested by using Cronbach's Alpha Test where a value of 0.7 and above shows acceptable reliability and validity. Besides that, all equipment used in this research were calibrated first according to the manufacturer's standard to avoid any error or bias. As for biological sample, mask and glove were used by researcher in handling the agar plate and samples to avoid direct contamination. The incubation and colony counting procedure was following European Standard (2000); EN 13098: 2000 and ISO

4833:2013[10,11]. Laboratory media blank and field blank also were prepared for every sample set for quality control measures.

RESULTS AND DISCUSSION

Physical parameters, temperature was recorded higher in studied preschools as compared with the comparative preschools as tabulated in Table 1. However, parameter of humidity and air velocity were slightly higher in comparative area instead of the studied area. This might be due to the differences of the use of mechanical ventilation such as fan in both areas. As outdoor environment also influences the indoor environment by 10%, humidity and velocity in comparative area were slightly higher due to less tall building and more plantations around this area. For building occupants' comfortability, Industry Code of Practice on Indoor Air Quality 2010 stated that, for air temperature, relative humidity and air velocity, the acceptable range would be 23-26^{o}C, 40-70% and 0.15-0.50 m/s respectively. However, this range is less practical to be applied in preschools setting as the standard was designed specifically for air conditioned and commercial building.

On the other hand, for indoor air pollutants, $PM_{2.5}$, PM_{10}, VOCs, mold, bacteria and Gram-negative bacteria were significantly different between preschools in studied and comparative areas as (Z= -4.213, p= <0.001), (Z= -6.847, p= <0.001), (Z= -5.286, p= <0.001), (Z= -5.793, p= <0.001), (Z= -8.705, p= <0.001) and (Z= -8.471, p= <0.001) respectively. This result shows that, the concentration of indoor air pollutants was significantly higher in preschools of the studied area as compared to the comparative area. However, when compared to Malaysia Ambient Air Quality Guidelines for 24 hours averaging time, the value of $PM_{2.5}$ and PM_{10} in studied preschool with median 67 $\mu g/m^3$ and 74 $\mu g/m^3$ respectively were still complying the standard ($PM_{2.5}$ 75 $\mu g/m^3$ and PM_{10} 150 $\mu g/m^3$). The same results were also obtained for VOCs where the value was still within the Canada Residential Indoor Air Quality Guidelines for 24 hours averaging time which is 1 ppm.

For mold and bacteria, WHO IAQ guidelines set standard 500 CFU/m^3 (8 hour exposure) for both biological pollutants. Measurement of bacteria exceeds the WHO standard (Median= 550 CFU/m^3). However, this measurement was based on a 4-hour measurement and the value might be lower as for 8-hour measurement. A study has shown that mold and bacteria have a positive relationship with dampness and settled dust[13]. These two parameters are the main problem in the studied preschools as most of the studied preschools have toilet and kitchen in it that serve as damp area and particulate matter that relatively high in urban areas.

Nevertheless, the level of Gram-negative bacteria also show a significant difference in both studied and comparative areas as (Z= -8.471, p= <0.001). As this is the first study to include Gram-negative bacteria as an indoor air pollutant, the value was hardly to be compared with other local study as most of the study assessing on endotoxin; the toxin produced by the Gram-negative bacteria. However, study by Traversi shows that, the level of Gram-negative bacteria was associate with the level of PM_{10} in urban, rural and farm areas[14]. As the level of PM_{10} and Gram-negative bacteria in studied area were significantly higher compared to the comparative area, it is suggested that, the sources of PM_{10} may trigger the growth of Gram-negative bacteria in the air.

For exposure of pollutants in house setting (Table 2), only $PM_{2.5}$, PM_{10} and mold that show significant difference in both studied and comparative groups as (Z= -2.883, p= 0.004), (Z= -3.149, p= 0.002) and (Z= -2.198, p= 0.028) respectively. Nonetheless, as compared with the value of pollutants in schools, level of $PM_{2.5}$ (median= 119 $\mu g/m^3$), PM_{10} (median= 131 $\mu g/m^3$) and VOCs (median= 0.04 ppm) in the houses of the studied group is higher compared to studied preschools with median 67 $\mu g/m^3$, 74 $\mu g/m^3$ and 0.0054 ppm respectively. It is suggested that, housing locations have contributed to the concentration of the particulates. The location of houses as well as outdoor and indoor combustion activities is the major contributor to the high level of indoor $PM_{2.5}$ and PM_{10} in the houses of the studied area. Besides that, USEPA stated that, indoor air pollutants such as PM_{10} and VOCs may come from sources such as carpet, house paints, paved or unpaved roads from outside[15].

All the findings above were parallel with the local study by Wesley and Jalaludin[16] and Nur Azwani et al[17] in the urban area which the level of $PM_{2.5}$, PM_{10}, VOCs, mold and bacteria were said to be higher in studied group as compared to the comparative group. Another study by Choo et al[18] has further support the findings of this study where, Choo et al[18] highlighted that the mean concentrations of $PM_{2.5}$, PM_{10} and VOCs were highest in the urban area preschools. The higher concentrations of pollutants in urban area preschools could be explained by higher pollution in this environment. As location was influences the level of pollutants inside the preschools, unique characteristics to preschools such as abundant use of sorptive material (drapes, toys, bedding and carpets) as part of its furnishings, children's habits and activities also greatly influence the concentrations of indoor air pollutants[19].

Table 1- Comparisons of indoor air quality between preschool in study areas (n=120)

Variable	Urban (n=60) Median (IQR)	Comparative (n=60) Median (IQR)	Z value	P value
$PM_{2.5}$ (μg/m^3)	67.00 (16.1)	48.00 (7.3)	-4.213	<0.001**
PM_{10} (μg/m^3)	74.00 (19.6)	51.00 (5.0)	-6.847	<0.001**
VOCs (ppm)	0.005 (0.089)	0.0 (0.0)	-	-
Mold (CFU/m^3)	402.0 (260.0)	252.0 (135.0)	-5.793	<0.001**
Bacteria (CFU/m^3)	550.0 (350.0)	221.0 (200.0)	-8.705	<0.001**
Gram-negative bacteria (CFU/m^3)	60.0 (45.0)	25.0 (20.0)	-8.471	<0.001**
Temperature (°C)	31.19 (3.0)	30.64 (2.3)	-2.374	0.018*
Relative Humidity (%)	70.5 (4.6)	76.3 (3.5)	-4.213	<0.001**
Air Velocity (m/s)	0.216 (0.028)	0.297(0.087)	-6.847	<0.001**

***Abbreviations and Notes**: IQR = Inter-quartile range, Bold p-values = significant findings*
* = Significant at p<0.05, ** = Significant at p<0.001

Table 2 – Comparisons of indoor air quality between houses in study areas (n=60)

Variable	Urban (n=60) Median (IQR)	Comparative (n=60) Median (IQR)	Z value	P value
$PM_{2.5}$ (μg/m^3)	119.0 (79.5)	73.5 (56.0)	-2.883	0.004*
PM_{10} (μg/m^3)	131.0 (78.8)	93.0 (67.9)	-3.149	0.002*
VOCs (ppm)	0.04 (0.13)	0.03 (0.08)	-1.136	0.256
Mold (CFU/m^3)	275.0 (235.0)	247.5 (70.0)	-2.198	0.028*
Bacteria (CFU/m^3)	142.5 (280.0)	137.5 (70.0)	-0.873	0.383
Gram-negative bacteria (CFU/m^3)	17.5 (22.5)	10.0 (18.3)	-1.146	0.252
Temperature (°C)	29.42 (1.57)	29.87 (1.84)	-0.473	0.636
Relative Humidity (%)	72.95 (15.67)	70.27 (9.12)	-1.752	0.080
Air Velocity (m/s)	0.215 (0.118)	0.214 (0.123)	-1.014	0.310

***Abbreviations and Notes**: IQR = Inter-quartile range, Bold p-values = significant findings*
* = Significant at p<0.05, ** = Significant at p<0.001

The results of comparison of reported respiratory symptoms between the studied and comparative group are presented in Table 3. Eight respiratory symptoms were assessed in this study, including cough, phlegm, wheezing, chest tightness, runny nose, blocked nose, sneezing and sore throat by using constructed questionnaire based on American Thoracic Society (ATS) and International Study of Asthma and Allergy in Childhood (ISAAC) standard questionnaire. From the chi-square test, only cough, phlegm, runny nose and blocked nose show significant difference between both studied and comparative groups as p= 0.026, p= 0.015, p= 0.001 and p= 0.002 respectively. For cough (PR= 4.34, 95% CI= 2.06-9.11), 31 (51.7%) respondents from studied group having the symptoms with just only 19 (38.3%) respondents from the comparative group. It is suggested that, the preschool children in studied area have 4.34 times more likely to experience respiratory health symptom of cough than preschool children in comparative area. Besides that, for phlegm (PR= 3.61, 95% CI= 1.57-8.33), 23 (38.3%) respondents from the studied group having the symptoms with just only 11 (18.3%) respondents from the comparative group, thus, the preschool children in studied area have 3.61 times more likely to experience respiratory health symptom of phlegm than preschool children in comparative area.

On the other hand, for runny nose (PR= 2.27, 95% CI= 1.25-6.13), 30 (50.0%) respondents from studied group having the symptoms with just only 13 (21.7%) respondents from the comparative group. So, it is suggested that, the preschool children in studied area have 2.27 times more likely to experience respiratory health symptom of runny nose than preschool children in comparative area. For symptom of blocked nose (PR= 2.74, 95% CI= 1.20-6.28), 27 (45.0%) respondents from studied group having the symptoms with just only 11 (18.3%) respondents from the comparative group, thus, the preschool children in studied area have 2.74 times more likely to experience respiratory health symptom of blocked nose than preschool children in comparative area.

To simplify, the prevalence of respiratory symptoms was generally higher among urban preschool children as compared to suburban preschool children. Only four respiratory symptoms (cough, phlegm, runny nose and blocked nose) show a significant difference between the studied and comparative groups with cough has the highest prevalence rate followed by phlegm, runny nose and blocked nose. It is suggested that there is an association between respiratory symptom occurrence in preschool children and the location of the preschool they attend. Preschool children from highly polluted areas (urban preschools) have shown a higher prevalence of respiratory symptoms compared to preschool children from a less polluted area (suburban preschools). According to Chen et al[20], the adverse respiratory health impact experienced by children who live in urban areas may be attributed to their exposure to an assortment of air pollutants produced by mobile sources.

The finding of this study that the location of the preschool significantly contributed in the prevalence of respiratory health symptoms also supported by the findings from the local studies by Wesley and Jalaludin[16], Choo et al[18], Nazariah et al[21] and Kamaruddin et al[22], where prevalence of respiratory health symptoms was higher among children in the urban area as compared to the suburban or rural area.

Table 3 - Prevalence of respiratory symptoms among preschool children (n=120)

Variable	Urban (n=60)	Suburban (n=60)	χ^2 value	p value	PR	95% CI
	Median (IQR)					
Cough						
Yes	31 (51.7)	19 (38.3)	4.937	0.026*	4.34	2.06-9.11
No	29 (48.3)	41 (61.7)				
Phlegm						
Yes	23 (38.3)	11 (18.3)	5.910	0.015*	3.61	1.57-8.33
No	37 (61.7)	49 (81.7				
Runny Nose						
Yes	30 (50.0)	13 (21.7)	10.474	0.001*	2.77	1.57-8.33
No	30 (50.0)	47 (78.3)				
Blocked Nose						
Yes	27 (45)	11 (18.3)	9.859	0.002*	2.74	1.20-6.28
No	33 (55)	49 (81.7)				

Abbreviations and Notes*: IQR = Inter-quartile range, PR = Prevalence rate. Bold p-values = significant findings.*
*** = Significant at p<0.05*

Lung function of preschool children was assessed based on forced vital capacity (FVC%) predicted, forced expiratory volume in 1 second (FEV_1%) predicted and ratio FEV_1/FVC% predicted. FVC% predicted and FEV_1% predicted were obtained based on the prediction equation for the normal value of lung function parameters among children in Malaysia developed by Azizi and Henry[23]. The result is tabulated in Table 4. T-test analysis was used to compare the values of FVC (liter) and FEV_1 (liter) between the studied and comparative group. The test reveals that FVC (liter) and FEV_1 (liter) among the studied group were significantly lower compared to the comparative group as (t= -3.710, p= <0.001) and (t= -4.027, p= <0.001) respectively. Meanwhile, Mann-Whitney U test was used to compare the values of FVC%, FEV_1% and FEV_1/FVC % between the studied and comparative group. Thus, FVC% and FEV_1% among the studied group were significantly lower compared to the comparative group as (Z= -2.866, p= 0.004) and (Z= -3.139, p= 0.002) respectively. However, only the value of FEV_1/FVC % between the groups was not significantly difference (Z= -1.205, p= 0.228).

From the value of FVC%, FEV_1% and FEV_1/FVC %, lung function abnormalities among preschool children was then determined by categorizing them in normal or abnormal status based on American Thoracic Society (2005) classification. The result is tabulated in Table 5. Chi-square test was used to compare the lung function abnormalities between the studied and comparative groups. Only FVC% and FEV_1% show a significant difference as p= <0.001 for both groups. For FVC%, 40 (66.7%) respondents from studied group having abnormal status with just only 15 (25.0%) respondents from the comparative group. For FEV_1%, 38 (63.3%) respondents from studied group having abnormal status with just only 12 (20.0%) respondents from the comparative group. FEV_1/FVC % values were normal for all respondents. It is suggested that exposure to a pollution from heavy traffic may contribute to lower FVC and FEV_1 among urban preschool children. Brunekreef et al[24] stated that, children who stay close by highways may suffer from impaired lung function because of being exposed to road vehicle related air pollution.The finding above was also supported by the local studies by Nur Azwani et al[17], Choo et al[18], and Kamaruddin et al[22] that shows the location of the preschool primarily in urban areas or near to the industry was significantly contributed in lower lung function among the preschool children.

Table 4 - Comparison of lung function among preschool children. (n=120)

Variable	Urban (n=60)	Suburban (n=60)	Z/t value	p value
	Median (IQR)			
FVC (Liter)[a]	0.61 ± 0.20	0.75 ± 0.19	-3.710	<0.001**
FEV_1 (Liter)[a]	0.59 ± 0.19	0.74 ± 0.19	-4.027	<0.001**
FVC %[b]	67.69 ± 42.64	88.35 ± 19.10	-2.866	0.004*
FEV_1 %[b]	71.54 ± 40.87	93.72 ± 20.14	-3.139	0.002*
FEV_1/FVC %[b]	107.08 ± 2.23	107.73 ± 1.31	-1.205	0.228

***Abbreviations and Notes**: IQR = Inter-quartile range, a = T-test, b = Mann-Whitney U Test*
** = Significant at p<0.05, ** = Significant at p<0.001*

Table 5 - Lung function abnormalities among preschool children (n=120)

Lung Function	Urban (n=60)		Suburban (n=60)		χ^2	p value
	Normal (%)	Abnormal (%)	Normal (%)	Abnormal (%)		
FVC %	20 (33.3)	40 (66.7)	45 (75.0)	15 (25.0)	20.979	<0.001**
FEV_1 %	22 (36.7)	38 (63.3)	48 (80.0)	12 (20.0)	23.177	<0.001**
FEV_1/FVC %	60 (100.0)	0 (0.0)	60 (100.00)	0 (0.0)	-	-

***Abbreviations and Notes**: χ^2 = chi square, Bold p-values indicate significant findings. ** = Significant at p<0.001*

In order to associate indoor air pollutants with lung function, all pollutants were categorized based on their median value of the studied group. A value that was higher than median was categorized as high while the value that was lower than median was categorized as low. Table 6 shows the association between indoor air pollutants in schools and lung function (FVC%) among preschool children in both studied and comparative areas. The chi-square result shows a significant association between all pollutants ($PM_{2.5}$, PM_{10}, VOCs, mould, bacteria and Gram-negative bacteria) with FVC% abnormality among the respondents. It is suggested that, odds of preschool children who exposed to high level of $PM_{2.5}$ and PM_{10} to get abnormal FVC% was 2.37 times higher than those who did not expose. Odds of preschool children who exposed to high level of VOCs to get abnormal FVC% was 3.27 times higher than those who did not expose. Furthermore, for biological pollutants, odds of preschool children who exposed to high level of mould, bacteria and Gram-negative bacteria to get abnormal FVC% were 2.59, 3.14 and 4.75 times higher than those who did not expose respectively.

Table 7 shows the association between indoor air pollutants in schools and lung function (FEV_1%) among preschool children in both studied and comparative areas. From the result, $PM_{2.5}$, PM_{10}, VOCs, mould, bacteria and Gram-negative bacteria shows a significant association with FEV_1% abnormality among the respondents. It is suggested that, odds of preschool children who exposed to high level of $PM_{2.5}$ and PM_{10} to get abnormal FEV_1% was 2.66 times higher than those who did not expose. Odds of preschool children who exposed to high level of VOCs to get abnormal FEV_1% was 3.67 times higher than those who did not expose. Besides that, for biological pollutants, odds of preschool children who exposed to high level of mould, bacteria and Gram-negative bacteria to get abnormal FEV1% were 2.72, 3.29 and 4.91 times higher than those who did not expose respectively.

The finding of this study was consistent with the local study by Choo et al[18] and Nur Azwani et al[17] in urban area of Selangor, Malaysia where significant association was found between indoor $PM_{2.5}$, PM_{10} and VOCs with lower lung function among children in urban area. It is suggested that, due to a very small size of these pollutants, they can reach deeper inside the children's lung and interfere with normal function of the lung in gaseous exchange[25].

Table 6 The association between exposure of indoor air pollutants in schools and lung function (FVC%) among preschool children. (n=120)

Variables	Lung Function		x^2 value	p value	PR	95% CI
	Abnormal	Normal				
$PM_{2.5}$						
High (>67µg m^{-3})	24	16	4.850	0.028*	2.37	1.09-5.15
Low (<67µg m^{-3})	31	49				
PM_{10}						
High (>74µg m^{-3})	24	16	4.850	0.028*	2.37	1.09-5.15
Low (<74µg m^{-3})	31	49				
VOCs						
High (>0.0051ppm)	26	14	8.878	0.003*	3.27	1.48-7.22
Low (<0.0051ppm)	29	51				
Mold						
High (>402CFU m^{-3})	19	11	4.434	0.026*	2.59	1.10-6.09
Low (>402CFU m^{-3})	36	54				
Bacteria						
High (>550CFU m^{-3})	20	10	6.993	0.008*	3.14	1.32-7.49
Low (>550CFU m^{-3})	35	55				
Gram Negative Bacteria						
High (>60CFU m^{-3})	22	8	12.185	<0.001*	4.75	1.90-11.87
Low (>60CFU m^{-3})	33	57				

Abbreviations and Notes: χ^2 = chi square, *95% CI = 95% Confidence Interval, PR = Prevalence rate. Bold p-values indicate significant findings. **Significant at p<0.001*

Table 7. The association between exposure of indoor air pollutants in schools and lung function (FEV_1%) among preschool children (n=120)

Variables	Lung Function		X^2 value	p value	PR	95% CI
	Abnormal	Normal				
$PM_{2.5}$						
High (>67µg m^{-3})	23	17	6.189	0.013*	2.66	1.22-5.79
Low (<67µg m^{-3})	27	53				
PM_{10}						
High (>74µg m^{-3})	23	17	6.189	0.013*	2.66	1.22-5.79
Low (<74µg m^{-3})	27	53				
VOCs						
High (>0.0051ppm)	25	15	10.714	0.001*	3.67	1.66-8.13
Low (<0.0051ppm)	25	55				
Mold						
High (>402CFU m^{-3})	18	12	5.531	0.019*	2.72	1.16-6.35
Low (>402CFU m^{-3})	32	58				
Bacteria						
High (>550CFU m^{-3})	19	11	7.726	0.005*	3.29	1.39-7.77
Low (>550CFU m^{-3})	31	59				
Gram Negative Bacteria						
High (>60CFU m^{-3})	21	9	13.211	<0.001*	4.91	2.00-12.04
Low (>60CFU m^{-3})	29	61				

Abbreviations and Notes: χ^2 = chi square, *95% CI = 95% Confidence Interval, PR = Prevalence rate. Bold p-values indicate significant findings. **Significant at p<0.001*

Another study conducted in four cities in China shows that, an increase 10 µg/m^3 of $PM_{2.5}$ and PM_{10} was associated with decreases of 3.5 ml FVC and 2.7 ml FEV_1 in lung function of school children[26]. A study in southern California by Gauderman et al[27] stated that, exposure to particulate matter especially $PM_{2.5}$ affects the growth of lungs during the rapid lung development between the ages of 10 and 18 years old. The results also were supported by international study among school children in Portugal, where significant association was found between indoor bacteria and fungi with lower FEV_1 value although quantitatively determined concentration of microbiological agents do not show a consistent association with respiratory health outcomes in different studies[3]. Study among healthy adults by Henberg et al[28] in Finland indicated that mold odor exposure was related to lower lung function levels among nonasthmatic adults, especially among women. It is suggested that, microbial contaminant such as fungi and bacteria also produce toxins and irritants with suspected effects on respiratory health[29].

For Gram-negative bacteria, many Gram-negative bacteria are in the range of size 1-5 µm and when inhaled, it can penetrate deeper into human lungs[30]. As the only agent that can produce endotoxin, higher house-dust endotoxin level has been found to correlate with increased medication used and greater airflow obstruction in asthmatic adults, in children both with and without asthma and even with infant wheezing in the first year of life[31,32,33]. On the other hand, the association between indoor air pollutants inside 60 respondents' houses and their lung function (FVC%) test also been carried out and from the result, only $PM_{2.5}$ (p= 0.032, PR= 2.56, 95% CI= 1.553-7.473) and PM_{10} (p= 0.018, PR= 3.23, 95% CI= 1.240-8.421) shows a significant association with FVC% abnormality as the concentration is higher compared to the concentration in schools.

Nevertheless, for the association between indoor air pollutants inside respondents' houses with their lung function (FEV_1%) only $PM_{2.5}$ (p= 0.003, PR= 3.02, 95% CI= 1.59-15.59), PM_{10} (p= 0.003, PR= 3.55, 95% CI= 1.98-16.69) and Gram-negative bacteria (p= 0.016, PR= 3.79, 95% CI= 1.26-11.46) shows a significant association with FEV_1% abnormality. According to Yahaya et al[34] indoor air pollutants in homes are an important influencing factor of lung function. Prolonged exposure to particulate matter in ambient air has been associated with a noticeable decrease in lung function[35]. Although very limited study had been done about airborne Gram-negative bacteria on human, this study shows a significant association between the exposures of Gram-negative bacteria with lung function abnormalities among the respondents. In line with that, a type of Gram-negative bacterium known as *L. pneumophila* is suggested can cause Legionellosis in human. Most people with Legionellosis will have pneumonia since the Legionella bacteria grow and thrive in the lungs.

Logistic regression was performed to determine the main factor that influenced the abnormality of FVC% and FEV_1% among the preschool children in study areas. From Table 8 and Table 9, exposure of preschool children toward $PM_{2.5}$ was significantly associated with both abnormality of FVC% (ß=1.403, p= 0.013, PR= 4.07, 95% CI= 1.34-12.33) and FEV_1% (ß=1.858, p= 0.025, PR= 6.41, 95% CI= 1.27-12.39). Based on the result, respondents that significantly exposed with $PM_{2.5}$ is four times more likely to have lung function (FVC%) abnormalities compared to those who did not expose. On the other hand, respondents that exposed with $PM_{2.5}$ is also getting six times more likely to have lung function (FEV_1 %) abnormalities compared to those who did not expose. Gemenetzis et al[36] reported that, increase in particulate matter concentration had significant with related lung disorder and with reduction in lung function, while $PM_{2.5}$ were strongly associated with cardiopulmonary diseases and lung cancer. It is suggested that, these problems could be due to reaction of polycyclic aromatic hydrocarbon contained in particulate matter on human body tissues[3].

Table 8. Factors influenced the lung function (FVC%) abnormality among respondents. (n=120)

Variables	**B**	**S.E**	**p value**	**PR**	**95% CI**
Constant	-1.698	0.570	0.003		
$PM_{2.5}$	1.403	0.566	**0.013***	**4.07**	**1.34-12.33**
PM_{10}	0.634	0.730	0.386	1.88	0.45-7.89
VOCs	-0.949	1.277	0.457	0.38	0.03-4.73
Mold	-0.431	0.660	0.513	0.65	0.17-2.37
Bacteria	0.109	0.878	0.901	1.12	0.20-6.24
Gram-negative Bacteria	1.792	1.300	0.168	6.00	0.47-76.70

Abbreviations and Notes: *95% CI = 95% Confidence Interval, ß= Regression Coefficient, PR = Prevalence rate, S.E= Standard Error, Nagelkerke R Square= 0.284. * = Significant at p<0.05*

Table 9: Factors influenced the lung function (FEV_1%) abnormality among respondents. (n=120)

Variables	ß	S.E	P value	PR	95% CI
Constant	-1.533	0.557	0.006		
$PM_{2.5}$	1.858	0.827	**0.025***	**6.41**	**1.27-12.39**
PM_{10}	0.799	0.736	0.277	2.22	0.53-9.41
VOCs	0.067	1.302	0.959	1.07	0.08-13.74
Mold	-0.422	0.652	0.518	0.66	0.18-2.36
Bacteria	0.802	0.919	0.383	2.23	0.37-13.51
Gram-negative Bacteria	0.758	1.316	0.565	2.13	0.16-28.16

*Abbreviations and Notes:, 95% CI = 95% Confidence Interval, ß= Regression Coefficient, PR = Prevalence rate, S.E= Standard Error, Nagelkerke R Square= 0.289. * = Significant at p<0.05*

CONCLUSION

Finding from this study indicated that the exposures to poor indoor air quality or high level indoor pollutants might increase the risk of getting respiratory symptoms and reduction in lung function among the study respondents. $PM_{2.5}$ was found to be the most significant with the decreased of lung function among the preschool children. Even though very limited study had been done on airborne Gram-negative bacteria towards human, this study proved that; there was a significant association between exposures of Gram-negative bacteria with lung function abnormalities and higher reported respiratory symptoms among the respondents.

Based on the result of this study, it is recommended for preschools management to implement a housekeeping program such that the ventilation in the classroom is well-maintained and cleaned regularly. As $PM_{2.5}$ is the most significant in predicting the lung function abnormalities among preschool children, it is advisable to clean the classroom by using wet vacuum cleaner as it proved to be more effective in eliminating small particulate instead of disperse them as compared to regular floor sweeping.

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COMPETING INTERESTS
There is no conflict of interest.

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