

Association of indoor air quality and preschool children's respiratory symptoms

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Abstract

Background: This study aimed to assess the association between exposure to indoor air pollution in DCCs (Child Day Care Centers) and the respiratory symptoms of children under 6 years old.

Methods: Air quality data were collected three times regarding to seasonal variation. Pollutants measured in 11 DCCs included PM_{10} , CO, NO₂, SO₂, O₃, benzene, bacteria, fungi, and dust mite. The frequencyof respiratory symptoms including coughing, rhinitis, and dyspnea were recorded via teacher and parent-report questionnaires. Fractional exhaled nitric oxide (FENO) levels were measured to assess airway inflammation.

Results: 436 children participated in the study. 83% completed data collection in all 3 seasons. The frequency of rhinitis correlated with PM_{10} (IRR=70.3, 95%CI=12.4-399.7, p<0.001), CO (IRR=3.2, 95%CI=2.4-4.2, p<0.001), benzene (IRR=2.3, 95%CI=1.8-3.2, p<0.001) and *D.Pteronyssinus* level (IRR=2.1, 95%CI=1.7-2.7, p<0.001). The frequency of coughingcorrelated with levels of PM_{10} (IRR=15.2, 95%CI=3.0-78.2, p<0.001), CO (IRR=2.8, 95%CI=2.1-3.7, p<0.001), benzene (IRR=1.4, 95%CI=1.1-1.9, p=0.02). The frequency of dyspnea correlated with *D.Pteronyssinus* level (IRR=3.9, 95%CI=1.7-9.1, p=0.001). FENO levels associated with high benzene levels (OR=5.9, 95%CI=1.5-22.9, p=0.01). The majority of DCCs had at least one PM_{10} measurement above the standard level, and these high levels were noted in all 3 seasons. Three DCCs had PM₁₀ levels above the standard level in all seasons. 64% of the DCCs had indoor bacterial counts above the standard level in all seasons.

Conclusions: PM_{10} and bacterial count is a significant problem in Bangkok metropolitan DCCs. The respiratory symptoms of children positively associated with PM_{10} , CO, benzene and dust mite levels.

Keywords: Cough; Indoor air pollution; Particulate matter; Preschool, Children; Rhinitis

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Introduction

Clean air is a fundamental requirement for human health and well-being. Children are more susceptible to air pollution than adults. Having higher minute ventilation and lower body weight, they are exposed to proportionately larger doses of pollutants. Immature lungs and immune systems also make children more vulnerable to pollutant exposure.¹ Bangkok is the most highly populated city in Thailand and is also a highly polluted city. There are approximately 300 day care centers (DCCs) in Bangkok and increasing numbers of preschool children attend DCCs. Preschool children usually spend at least 8 hours per day in the DCCs; therefore, DCCs could be a significant source of pollution exposure. **Corresponding author:** Pongtong Puranitee Department of Pediatrics, Ramathibodi Hospital, Mahidol University, Bangkok, Thailand E-mail: pongtongung@gmail.com

Indoor air can be contaminated by both chemical and biological pollutants. Serious chemical pollutants include: particulate matters with diameter smaller than 10 μ m (PM₁₀), carbon monoxide (CO), nitrogen dioxide (NO₂), ozone (O₃), sulfur dioxide (SO₂), and benzene. PM₁₀ are particles small enough to enter the human respiratory tract and can originate from sources such as traffic, construction, and agriculture. The World Health Organization's (WHO) standard level for short term exposure to indoor PM₁₀ is 50 μ g/m³ (24 hours).² PM₁₀ is associated with both upper and lower respiratory symptoms including coughing, bronchitis, phlegm, and hay fever.³⁻⁴ It is also associated with decreased lung function in asthmatic



children.⁵ A study in Thailand⁶ comparing exposure to high and low PM_{10} levels (170 versus 36 μ g/m³) in high school children found that the prevalence of eye and airway symptoms were increased in high PM₁₀ group. FEF(25-75%) was also decreased in the high PM10 group while school absence and medical expense increased. CO mainly comes from incomplete combustion such as in cooking and traffic. It causes asphyxiating effects and also aggravates ischemic heart disease symptoms and respiratory symptoms.² Short term exposure is defined in the WHO standards for indoor CO level as 10 mg/m³ (8 hours) and 7 mg/m³ (24 hours).² NO₂ is an oxidant and corrosive which causes airway injury and inflammation. Primary NO₂ sources are traffic, tobacco smoke, fuel combustion, and also mosquito coils. According to WHO standards, short term indoor exposure to NO, should not exceed 200 µg/m3 (24 hours).2 NO, has been found to be associated with respiratory symptoms both in healthy and asthmatic children,⁷⁻⁹ and also with a reduction in children's peak expiratory flow (PEF).9 Ozone sources mainly come from outdoors. It is irritative and associated with increased respiratory symptoms¹¹ and decreased PEF in children.^{7,11} The WHO standard level for short term indoor exposure to O₂ is 100 μ g/m³ (8 hours).² Major sources of SO₂ are sulfur-containing fuel combustion such as in power plants and other industrial processes. It is associated with upper respiratory symptoms¹² and exacerbation of asthma symptoms.7 The WHO standard level for short term indoor exposure to SO₂ is 20 µg/m³ (24 hours).² For benzene, it generally has higher indoor levels than outdoor levels, because the main sources of benzene are from indoors including building material, paints, smoking, and combustion. It is known to affect respiratory systems, including asthma, respiratory tract infection and impaired lung function.13 It is associated with an increased risk of leukemia and aplastic anemia. The dose related effect of benzene exposure is well-established, with it causing death at the level of 20,000 ppm in up to 15 minutes. Due to its carcinogenic effects, the WHO has stated that no safe level of exposure can be recommended and indoor exposure should be as low as possible.² In Hong Kong, an airborne benzene level of less than 0.0161 mg/m³ is used as an objective cut-point for good indoor air quality.

Biological pollutants consist of airborne bacteria, airborne fungi, and house dust mites. Airborne bacteria come from humans and also human activities such as sneezing, coughing, and toilet flushing. They can cause infections such as respiratory tract and skin infections. Exposure to high endotoxin levels is associated with respiratory illnesses and wheezing in childhood,14-15 but some studies have found that exposure to endotoxins during infancy seems to prevent atopic diseases and wheezing.¹⁶⁻¹⁷ Airborne fungi can produce mycotoxins which can cause infection and immunoglobulin E mediated allergic reactions.¹⁸ Exposure to indoor molds can lead to respiratory symptoms, respiratory infections, and exacerbation of asthma.19 The standard recommendation for microbial count by the American Industrial Hygiene Association is 500 CFU/m³ for both bacterial and fungi counts, and standards in many countries such as Taiwan, Singapore, and Hong Kong range from 500-1000 CFU/m3.

House dust mites are usually found in mattresses, pillows, carpets, or cushions. The most common are *Dermatophagoidespteronyssinus* and *Dermatophagoidesfarinae*. Exposure to *Dermatophagoides* in infancy is associated with an increased prevalence of positive skin prick tests and an increased concentration of immunoglobulin E specific to dust mite by age 5 years old in children of atopic parents.²⁰ Exposure to *Dermatophagoides* levels higher than 10 μ g/g in infancy is associated with 4.8 fold increase in the relative risk of asthma by age 11.²¹ The WHO has recommended that exposure to dust mite level above 2 μ g/g of dust is a risk for sensitization and asthma.²²

Studies have previously been performed on indoor air pollutants in DCCs in different parts of the world.²³⁻²⁶ A study in Paris found that NO₂ and airborne endotoxin levels were significantly higher in DCCs compared to homes.²³ In Nigeria, a study of particulate matter burden in 48 DCCs found that mean indoor PM₁₀ levels significantly exceeded WHO guidelines.²⁴ In Norway, an inspection of 175 DCCs found that 51% of DCCs had problems with dampness, such as sign of moldand water leakage, but no significant effect on children's respiratory symptoms was found.²⁵ In Taiwan, a study of 2 DCCs revealed that indoor microbial levels were above the recommendation of the Taiwan Environmental Protection Administration.²⁶

In Thailand, only a few studies have been conducted on this subject.²⁷⁻²⁸ A 2011 study measured the indoor levels of PM₁₀ in 25 Bangkok DCCs in high-pollution areas and 25 in low-pollution areas and found both areas had higher than standard levels of PM₁₀ and significant difference in PM₁₀ concentrations. The study also found a positive correlation between PM₁₀ concentrations and the number of children complaining of coughs in the high-pollution area.²⁷ Another study was done in a child home care center in Bangkok, microbial count and PM₁₀ levels were measured in 20 households and found that 47.2% and 47.6% had bacterial and fungi counts above standard levels and 47.0% had higher PM₁₀ than standard level.²⁸

From literature review, although many studies have examined the effects of indoor air pollution on adult and older children's health, few studies have been done on the preschool age group, especially in tropical climate regions. Therefore, this study's primary objective was to assess the level of indoor air pollutants in DCCs in Bangkok and the association between exposure to indoor air pollution in DCCs and the respiratory symptoms of children under 6 years old. Fractional exhaled Nitric Oxide (FENO) was used to assess airway inflammation in children. Also, to determine the environmental characteristics of DCCS which influence indoor air pollution.

Methods

11 Bangkok Metropolitan DCCs were enrolled by computer -based stratified cluster random sampling from different areas. All children under 6 years old who had attended a DCC for at least one year and whose parents gave signed informed consent were included. The study was reviewed and approved by the Institutional Review Board of the Faculty of Medicine, Ramathibodi Hospital, Mahidol University (IRB number 05 -57-12). Demographic and home environment data were collected by questionnaires regarding age, gender, ethnicity of children, and socio-economic status, as well as history underlying diseases, immunization for influenza, exposure to cigarette smoke, and solid fuel use at home. Due to the possible seasonal variations in air pollution, data were collected in three different seasons: the rainy season (June-July, 2014), the winter (November-December, 2014), and the summer (May, 2015). Pollutants were measured while the DCCs were open, from 9.00 to 12.00 am on weekdays. In winter and summer, one DCC was temporarily closed and data collection was discontinued in that DCC.Pollutants measured in this study included PM₁₀, CO, NO₂, SO₂, O₃, benzene, bacteria count, fungi count, and dust mite level. Pollutants were measured using standard laboratory techniques at the Office of Public Health and Environmental Technology Services, Faculty of Public Health, Mahidol University. The techniques used in the measurement were gravimetric for PM₁₀, gas chromatography for benzene, CO and O₃, and ion chromatography for NO₂ and SO₂. Bacterial and fungal counts were achieved using a culture-based method and the specimens were obtained by impactor samplers. For dust mite antigen detection, specimens were collected from children's bedding materials by vacuum dust collector and analyzed for Dermatophagoidespteronyssinus (Dp) and Dermatophagoidesfarinae (Df) antigen levels using two-site monoclonal antibody based ELISA (INDOOR Biotechnologies, Ltd.) at Siriraj House Dust Mite Center for Services and Research, Faculty of Medicine, Siriraj Hospital, Mahidol University.

The environmental characteristic data of the DCCs were recorded by investigators on total indoor area, type of building material, year of construction, ventilation system, number of children, presence of pets indoors, indoor cooking, proximity to traffic or water sources, visible signs of dampness or mold. Respiratory symptoms frequencies (episodes/month) during one month after DCCs pollution measurement were assessed by parent-report and teacher-report questionnaires on symptoms including coughing, rhinorrhea, sneezing, congestion, itching nose, and dyspnea (breathing difficulty or tachypnea). Also, the questionnaires were asked weekly so the respiratory symptoms frequencies range was 0-4 episodes/month. Rhinitis is defined as presence of at least 2 symptoms of rhinorrhea, sneezing, congestion and itching nose. Other outcomes including absence from school, hospital visits and admission due to respiratory symptoms were also collected by the questionnaires. The questionnaires were sent weekly for 4 consecutive weeks during each season. Ten children were randomly selected from each DCC in each season for FENO measurement by multiple breath testing to represent airway inflammation. Statistical analysis, using Poisson regression was performed to analyze the association among the pollutant levels and clinical outcomes. The multiple logistic regression analysis was used to analyze the association between benzene, fungi and FENO levels. Mann-Whitney U test was used to analyze the difference of clinical outcome between air pollutant level exceeded standard and within standard level. All analyses were performed by STATA version 14 (College Station, TX: StataCorp LP).



Results

Children characteristics:

The number of children included in this study was 436. The mean age was 50 months (S.D. = 9.4, range 29-72 months) and 52% were male. During the study period, one DCC was closed down. Although some children left the DCCs during the year, 83% of children completed data collection in all 3 seasons. Children, family and home environmental characteristics are shown in **Table 1**.

Table 1. Children, family and home environment characteristics

Children characteristics	
Mean age (months)	50.1 (SD = 9.4)
Mean duration of DCC attendant (months)	17.3 (SD = 7.5)
Boys	229 (52%)
Girls	207 (48%)
Underlying diseases Allergic rhinitis Asthma G6PD deficiency Anemia Hepatitis B Epilepsy	$\begin{array}{c} 42 \ (9.6\%) \\ 32 \ (7.3\%) \\ 6 \ (1.4\%) \\ 4 \ (0.9\%) \\ 2 \ (0.5\%) \\ 1 \ (0.2\%) \end{array}$
Influenza vaccination during 1 year	113 (25.9%)
Care taker not parents	105 (24.08%)
Family characteristics	
Father education Below primary school Primary school Secondary school Bachelor degree Above Bachelor degree Mother education Below primary school Primary school Secondary school Bachelor degree Above Bachelor degree Above Bachelor degree Above Bachelor degree Family income <10.000 Babt/month	4 (0.9%) 88 (20.2%) 289 (66.3%) 52 (11.9%) 3 (0.7%) 11 (2.5%) 129 (29.6%) 258 (59.2%) 37 (8.5%) 1 (0.2%) 145 (33.3%)
<10,000 Baht/month 10,000-30,000 Baht/month >30,000 Baht/month	145 (33.3%) 251 (57.6%) 40 (9.2%)
Home characteristics	
Indoor smoking More than one indoor smokers Father smoking Mother smoking	224 (51.4%) 63 (14.4%) 189 (43.3%) 5 (1.1%)
Cooking fuel Gas Electricity Solid fuel	390 (89.4%) 76 (17.4%) 27 (6.2%)



Pollutants levels:

The majority of DCCs had at least one PM₁₀ measurement above the standard level, and these high levels were noted in all 3 seasons. 3 out of 11 DCCs had PM₁₀ levels above the standard level in all seasons.The mean PM₁₀ level was 70 µg/m³ (range 20-205). O₃ levels exceeded WHO standards in 80% of DCCs only in winter. The mean level of O₃ in winter (123 µg/m³) was significantly higher than in the rainy season (36 µg/m³) and summer (45 µg/m³) (p < 0.001).This finding correlated with the finding of the Thai Pollution Control Department, which showed O₃ levels in Bangkok to be higher between November and March. The mean level of SO₂ was 16 µg/m³ (10-12). Three DCCs had a single measured SO₂ level higher than the WHO standard level. Benzene was higher than the Hong Kong standard in 2 DCCs during the rainy season (0.105 and 0.054 mg/m³). In other seasons, benzene levels were within the standard in all day care centers. NO₂ and CO were within WHO standard levels in all seasons.

64% of the DCCs had higher indoor bacterial counts than the standard level in all seasons. Fungi levels were within standard levels in all DCCs in the summer, and higher than standard levels in 40% of DCCs in the rainy season and 70% of DCCs in the winter. For dust mite antigens, Dp antigen level was above the sensitization threshold level of 2 μ g/g of dust in only 1 DCC in the rainy season (3.13 μ g/g of dust) and winter (4.25 μ g/g of dust), while the rest were below the sensitization threshold. Df antigen levels were below the sensitization threshold in all seasons. PM₁₀, O₃ levels, and bacterial counts are shown in **Figure 1-3**.



Figure 1. The study flow of stratified cluster random sampling method.



Figure 2. PM₁₀ levels measured from DCCs in each season.

*DCC5 was closed in winter and summer





Figure 3. O₃ levels measured from DCCs in each season.

Pollutant levels and association with respiratory symptoms:

Frequency of parent-reported rhinitis correlated with PM₁₀ (IRR = 70.26, 95%CI = 12.35 - 399.69, p-value < 0.001), CO (IRR = 3.15, 95%CI = 2.36 - 4.21, p-value < 0.001), benzene (IRR = 2.33, 95%CI = 1.71 - 3.18, p-value < 0.001) and Dp level (IRR = 2.14, 95%CI = 1.67 - 2.74, p-value < 0.001). In addition, frequency of teacher-reported rhinitis correlated with CO (IRR = 3.04, 95%CI = 1.61 - 3.84, p-value < 0.001) and benzene levels (IRR = 2.49, 95%CI = 1.61 - 3.83, p-value < 0.001). Frequency of parent-reported coughing correlated with levels of PM₁₀, (IRR = 15.20, 95%CI = 2.95 - 78.23, p-value = 0.001) CO (IRR = 2.76, 95%CI = 2.06 - 3.69, p-value < 0.001), and benzene (IRR = 1.41, 95%CI = 1.06 - 1.89, p-value = 0.02). Frequency of teacher -report coughing also correlated with CO (IRR = 3.51, 95%CI = 2.52 - 4.91, p-value < 0.001) and benzene levels (IRR = 3.16, 95%CI = 2.21 - 4.54, p-value < 0.001). Frequency of parent -report dyspnea correlated with dust mite level (IRR = 3.13, 95%CI = 1.36 - 7.21, p-value = 0.008), and frequency of parent -reported dyspnea correlated with benzene levels (IRR = 37.64, 95%CI = 8.96 - 158.11, p-value < 0.001). The correlations found in this study are shown in Table 2. The confounding factors including influenza vaccination, indoor smoking and underlying diseases such as asthma and allergic rhinitis had no significant association with respiratory symptoms, therefore these factors were not used to adjust during statistical analysis of the correlation between pollutants and respiratory symptoms.

When comparing DCCs with pollutant levels higher than standard levels for indoor air quality and those within standard levels, we found that children in DCCs with high PM_{10} levels had significantly higher parent and teacher -reported rhinitis symptoms, parent-reported and teacher -reported cough. Children in DCCs with high PM_{10} levels also had higher frequency of school absences and hospital visits due to respiratory symptoms. Alternatively, the DCCs with high airborne bacterial levels, children had lower frequency of parent-reported rhinitis and cough symptoms when compared to DCCs with airborne bacterial counts within standard. In DCCs with high Dp levels, children also had significantly higher parent-reported cough frequency, as shown in **Table 3**.

Table 2. Correlation between pollutants and respiratory symptoms

Pollutants	Respiratory symptoms	IRR	p-value
PM ₁₀	Parent-report rhinitis frequency	70.26 (12.35 - 399.69)	< 0.001
	Parent-report cough frequency	15.20 (2.95 - 78.23)	0.001
СО	Parent-report rhinitis frequency	3.15 (2.36 - 4.21)	< 0.001
	Teacher-report rhinitis frequency	3.04 (1.61 - 3.84)	< 0.001
	Parent-report cough frequency	2.76 (2.06 - 3.69)	< 0.001
	Teacher-report cough frequency	3.51 (2.52 - 4.91)	< 0.001
High Benzene	Parent-report rhinitis frequency	2.33 (1.71-3.18)	< 0.001
	Teacher-report rhinitis frequency	2.49 (1.61 - 3.83)	< 0.001
	Parent-report cough frequency	1.41 (1.06 - 1.89)	0.02
	Teacher-report cough frequency	3.16 (2.21 - 4.54)	< 0.001
	Teacher-report dyspnea	37.64 (8.96 - 158.11)	< 0.001
	Number of absent days due to respiratory	2.85 (2.29 - 3.55)	<0.001
	symptoms		
DP	Parent-report rhinitis frequency	2.14 (1.67 - 2.74)	<0.001
	Parent-report dyspnea	3.13 (1.36 - 7.21)	0.008

PM, Particulate Matter; CO, carbon monoxide; DP, *Dermatophagoidespteronyssinus*.

			DCC	S with high F	vollutant leve	F			DCCs	with normal	pollutant le	vel		P-value
		R	kespiratory sy	mptom frequ	uency (episod	de/month)		E	kespiratory sy	mptom frequ	uency (episoo	de/month)		
lutants	Symtoms			N (%)			Median			N (%)			Median	
		0	1	2	3	4	(IQR)	0	1	2	ю	4	(1QR)	
e	Parent-report rhinitis symptoms	6(15.4)	10(25.6)	9(23.1)	10(25.6)	4(10.3)	2 (2)	689(59.6)	271(23.4)	112(9.7)	45(3.9)	4(3.4)	0 (1)	< 0.001
	Teacher-report rhinitis symptoms	19(48.7)	6(15.4)	7(17.9)	3(7.7)	4(10.3)	1 (2)	991(85.7)	91(8.0)	33(2.8)	28(2.4)	13(1.1)	(0) (0)	< 0.001
	Parent-report cough	7(17.9)	10(25.6)	12(30.8)	4(10.3)	6(15.4)	2 (2)	611(52.8)	326(28.2)	119(10.3)	60(5.2)	40(3.5)	0(1)	< 0.001
	Teacher-report cough	16(41.0)	10(20.6)	5(12.8)	3(7.7)	5(12.8)	1 (2)	853(73.8)	222(19.2)	59(5.1)	13(1.1)	9(0.8)	0(1)	< 0.001
	Absent school caused by	13(33.3)	26(66.7)	ı	I	ı	2 (5)	774(64.4)	412(35.6)	I	I	I	0(1)	< 0.001
	respiratory symptoms* Hospital visit*	19(48.7)	20(51.3)				1 (1)	961(83.1)	195(16.9)			ı	(0) 0	< 0.001
al count	Parent-report rhinitis symptoms Parent-report cough	520(62.3) 451(54.1)	176(21.1) 232(27.8)	80(9.6) 81(9.7)	42(5.0) 44(5.3)	16(1.9) 26(3.1)	$\begin{array}{c} 0 \ (1) \\ 0 \ (1) \end{array}$	175(48.5) 167(46.3)	105(29.1) 104(28.8)	41(11.4) 50(13.8)	13(3.6) 20(5.5)	27(7.5) 20(5.5)	$\begin{array}{c} 1 \ (1) \\ 1 \ (1.5) \end{array}$	< 0.001 < 0.001
lite ronyssinus)	Parent-report cough	168(45.4)	114(30.8)	49(13.2)	25(6.8)	14(3.8)	1 (2)	450(54.5)	222(26.9)	82(9.9)	39(4.7)	32(3.9)	0 (1)	< 0.001

* All of statistical analysis performed by Mann-Whitney U Test, except absent school caused by respiratory symptoms (0=present, 1=absent) and Hospital visit (0= no, 1=yes) was analyses by Wilcoxon Rank Sum Test. The statistically significant is p-value < 0.05.



DCC environmental characteristics:

that recommended by the Thai Ministry of The median indoor area of the DCCs was 240 m² (IQR = 56, 400), and the median child density was 0.63 person/m² (IQR = 0.33, 0.99). Child density was higher than Public Health (0.5 person/m^2) in 64% of the DCCs. The median distance from DCCs to Water sources were found nearby 64% of the DCCs. Open ventilation was used exclusively in 7 DCCs, with the others using Visible signs of indoor dampness or mold the DCC's environment characteristics and pollutant levels found wasfor child density 2 DCCs cooked without a separate kitchen. were seen in 55% of the DCCs. The only both open ventilation and air-conditioner. between road traffic was 20 meters (IQR = 2, 100). and bacterial count ($\beta = 0.952$, p < 0.001). significant positive association







Fractional exhaled nitric oxide (FENO) measurement:

We randomly enrolled 110 children in therainy season and 100 children in winter and summer (due to one DCC was closed) for FENO measurement by using computer-based sampling for 10 children/DCC in each season (four months apart). The test was successfully performed on 102, 93 and 86 children in the rainy season, winter, and summer respectively. The FENO measurement failed in some children due to non -cooperation. We found that the majority of children hadlow FENO levels (lower than 20 ppb). In this study, there were no children withhigh FENO levels (above 35 ppb). Intermediate FENO levels (between 20 and 35 ppb) were found in nine children in the rainy season, three children in winter, and eleven children in summer. The mean FENO levels were 8.10 (95%CI = 6.65 - 9.54) in rainy season, 6.69 (95%CI = 5.44 - 7.94) in winter, and 9.32 (95%CI = 7.69 - 10.96) in summer. These mean levels were statistically different in each season (p < 0.001). FENO levels hadastrong association with high benzene levels (OR 5.9 95%CI 1.5 - 22.9, p-value = 0.01).

Discussion

In comparison with previous studies of DCCs in Bangkok,²⁷⁻²⁸ this study found similarly high results of PM_{10} levels and airborne microbial counts in the majority of DCCs. SO_2 , O_3 , CO, NO₂, and dust mite antigen levels had never been studied in DCCs in Bangkok before. As such, this was the first study to provide data on indoor levels of these pollutants.

In this study, PM_{10} was found to be a major pollutant in most DCCs and had a correlation with the frequency of coughing and rhinitis. This is similar to previous studies which found PM_{10} to be associated with respiratory symptoms, including coughing, phlegm, and wheezing in children and also to correlate with interleukin-6 level in children's sputum - a biomarker for respiratory inflammation.^{33,34}

CO levels associated with the frequency of coughing and rhinitis, as found in a previous study between CO and coughing.³³ In this study, we also found that indoor CO significantly correlated with the frequency of respiratory symptoms (rhinitis and coughing) in preschool children despite being within WHO standard levels.

In recent studies, indoor benzene levels were found to associate with respiratory symptoms particularly asthma symptoms and pulmonary infection in children.¹³ The results are relevant to this study. We found that indoor benzene associated with rhinitis, coughing, dyspnea, and fractional exhaled nitric oxide level representing airway inflammation.

In this study, house dust mite antigen levels associated with rhinitis and dyspnea. This was also similar to other studies in which exposure to house dust mite antigen increased allergic sensitization and risk of asthma.^{20,21}

The finding of children in DCCs with high airborne bacterial levels having a lower frequency of rhinitis and cough could be explained by the hygiene hypothesis. Other studies have also found that higher exposure to bacterial endotoxins might be a protective factor for respiratory allergy.^{16,17}

In our study, The FENO levels were measured in 3 seasons because pollutants and allergens have possible seasonal variations and therefore might effects the airway inflammation. Although the mean FENO levels were different statistically, they were far below 20 ppb, and therefore assumed not to be clinically significant.

The limitations of this study was that only 11 DCCs were included in this study, meaning there was a relatively small sample size. Therefore, the power to determine the association between DCC environmental characteristics and pollutant levels may have been inadequate. Further studies to identify the sources of indoor air pollutants in DCCs should be conducted for the proper intervention and establishment of a public policy in the future. Also, this study was not aimed to determine the causal relationship between air pollutants and respiratory symptoms, only the association between those data could be examined. Research to confirm the association and identify the causal relationship between air pollutants and respiratory symptoms of preschool children is suggested. Also, indoor pollutant levels could notbe continuously monitored in this study, and a single-day measurement might not represent the actual average exposure to indoor pollutants in a season. Moreover, respiratory symptom frequency assessment by parent and teacher-reported questionnaires could suffer from recall bias. The duration of stay in DCCs wasn't found to be associated with respiratory symptoms. To understand more on the effect of duration of pollutants exposure on respiratory symptoms, a longitudinal study might be needed.

The correlation between child density and indoor airborne bacterial count found in this studywas similar to results in previous studies that found an association between occupancy rate and bacterial and $\rm CO_2$ levels.^{29,30} Although this study found no association between ventilation systems and pollutant levels, another study³¹ identified a positive correlation between air-conditioning with mixed ventilation and higher levels of indoor pollutants compared to naturally ventilated DCCs. Also, this study found no significant correlation between visible signs of dampness or mold and levels of airborne fungi, while a positive correlation was reported in another study.³²

Aside from the objectives of this study, we found that a number of Bangkok DCCs suffered from over crowding, and the cigarette-smoking at home rate was high. These issue should receive attention and intervention. A further recommendation would be that a separate kitchen should be present in all DCCs with cooking activities to limit possible sources of air pollution.

Conclusion

 PM_{10} and airborne bacterial counts were above WHO standard levels in DCCs in all seasons. The respiratory symptoms of children associated with PM_{10} , CO, benzene, and dust mite levels, even though the CO level was within the WHO recommendation level. Further study on the safety level of the air pollution specifically for preschool children might be suggested.

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