UNIVERSITI PUTRA MALAYSIA

INDOOR AIR POLLUTANTS EXPOSURE AND EOSINOPHIL CATIONIC PROTEIN (ECP) AS UPPER AIRWAY INFLAMMATORY BIOMARKER AMONG MALAY PRESCHOOL CHILDREN IN SELANGOR

ANDREW DANA ANAK WESLEY

Ip
FPSK4 2014 4

INDOOR AIR POLLUTANTS EXPOSURE AND EOSINOPHIL CATIONIC PROTEIN (ECP) AS UPPER AIRWAY INFLAMMATORY BIOMARKER AMONG MALAY PRESCHOOL CHILDREN IN SELANGOR

BY

ANDREW DANA ANAK WESLEY

Research thesis submitted in fulfillment for the degree Bachelor of Science (Environmental and Occupational Health) from Faculty of Medical and Health Sciences, University Putra Malaysia

© COPYRIGHT UPM

ACKNOWLEDGEMENT

First and foremost, I would like to express my highest gratitude to God Almighty for the strengths and blessing He has given me in completing this final year project. Then, my special appreciation goes to my supervisor, Dr Juliana Jalaludin for her excellent technical guidance, attentive supervision and endless support throughout this process until the completion of this thesis. With her vast knowledge and wise thought, she has contributed greatly to the success of this research. Not forgetting my appreciation to my course coordinator, Dr Saliza for her advices, knowledge sharing and encouragement all this while which has given me the opportunity and time to complete my thesis successfully.

Next, I would like to thank my study respondents and their parents for the utmost cooperation and support into this research. They have made the data collection process convenient and enjoyable throughout the research period. In addition, my sincere thanks also goes to the teachers and preschool managements where this study has been conducted. Thank you for aiding the process and welcoming us to be in your present.

Last but not least, my deepest gratitude goes to my beloved family especially my mother for their support, encouragement, advices and prayers all these while. To those who directly or indirectly contributed to data collection and analysis process, moreover for the completion of the whole project, your kindness, time and help means a lot to me. Thank you very much.

© COPYRIGHT UPM

ABSTRACT

INDOOR AIR POLLUTANTS EXPOSURE AND EOSINOPHIL CATIONIC PROTEIN (ECP) AS UPPER AIRWAY INFLAMMATORY BIOMARKER AMONG MALAY PRESCHOOL CHILDREN IN SELANGOR

ANDREW DANA WESLEY

Background: Upper airway and lower airway are functionally-linked, with inflammation in the former play a vital role in the pathogenesis of asthma and allergy. Studying the association between indoor air pollutants with upper airway inflammation in children will help to improve childhood asthma and allergy management related poor indoor air quality. **Method:** A cross-sectional study was carried out among Malay preschool children in industrial (N=53, Kelana Jaya and Shah Alam) and suburban (N=45, Semenyih and Hulu Langat) areas in Selangor. Questionnaire adapted from American Thoracic Society (ATS) and International Study on Asthma and Allergy in Children (ISAAC) was distributed to obtain respondents' background information, exposure history and reported respiratory symptoms. Eosinophil Cationic Protein (ECP) concentration in nasal swab sample was collected and analysed to determine the prevalence of upper airway inflammation. Indoor Air Quality (IAQ) assessment was also carried out in a total of 7 preschools in both study areas which include parameters of PM_{10}, VOCs, total mould and total bacteria, relative humidity (RH) and air temperature. **Results:** Results from statistical analysis show significant difference between areas for PM_{10}, $z = -1.976$, $p = 0.048$; total mould, $z = -2.420$, $p = 0.016$; total bacteria, $z = -1.981$, $p = 0.048$; and RH, $z = -5.587$, $p = 0.001$. There was significant difference for ECP level among respondents between study areas $t = 8.473$, $p<0.001$. ECP level was consistent and statistically significant with reported upper airway symptoms of phlegm, runny nose, blocked nose and sneezing, χ^2 (1, N=98)=6.437, $p<0.05$; χ^2 (1, N=98)=10.516, $p<0.001$; χ^2 (1, N=98)=9.130, $p<0.05$; χ^2 (1, N=98)=10.516, $p<0.001$. Logistic regression shows significant association between VOC and ECP level, $\beta=42.596$, $p<0.05$ (PR=6.410, 95% CI=1.268 – 32.394) after controlling all the confounding factors in this study. **Conclusion:** This study concludes that exposure to indoor air pollutants increase the risk of respiratory problems and might have an impact on the inflammatory and secretory response of the nasal mucosa.

Keywords: Indoor air quality (IAQ), respiratory health, upper airway inflammation, eosinophil cationic protein (ECP)

© COPYRIGHT UPM

ABSTRAK

PENDEDAHAN KEPADA PENCEMAR UDARA DALAMAN DAN 'EOSINOPHIL CATIONIC PROTEIN (ECP)' SEBAGAI PENANDA BIOLOGI BAGI RADANG SALUR PERNAFASAN ATAS DI KALANGAN KANAK-KANAK TADIKA BERBANGSA MELAYU DI SELANGOR

ANDREW DANA WESLEY

Pengenalan: Salur pernafasan atas dan bawah saling berkait fungsinya dengan radang yang berlaku pada bahagian atas memainkan peranan penting dalam patogenesis asma dan alergi. Dengan memahami hubungkait antara pencemar udara dalaman dan radang salur pernafasan atas di kalangan kanak-kanak akan membantu menambahbaik pengurusan penyakit asma dan alergi akibat kualiti udara dalaman yang tercemar teruk pada peringkat awal. **Metodologi**: Kajian keratan rentas telah dijalankan di kalangan kanak-kanak tadika berbangsa Melayu di kawasan industri (N=53, Kelana Jaya dan Shah Alam) dan luar Bandar (N=45, Semenyih dan Hulu Langat) di Selangor. Borang kaji selidik yang disesuaikan dari kajian oleh 'American Thoracic Society (ATS)' dan 'International Study on Asthma and Allergy in Children (ISAAC)' telah diedarkan untuk mendapatkan latar belakang responden, sejarah pendedahan dan laporan simptom pernafasan. Kepekatan 'Eosinophil Cationic Protein (ECP)' dalam sampel sapuan hidung telah diambil dan dianalisa untuk menentukan kelaziman radang salur pernafasan atas. Penilaian Kualiti Udara Dalaman (IAQ) telah dijalankan di 7 buah tadika di kedua-dua kawasan kajian termasuk parameter seperti PM_{10}, VOCs, kulat dan bakteria, relatif kelembapan (RH) dan juga suhu udara. **Hasil Kajian**: Analisa statistik menunjukkan perbezaan yang signifikan antara kawasan untuk PM_{10}, $z = -1.976$, $p = 0.048$; kulat, $z = -2.420$, $p = 0.016$; bakteria, $z = -1.981$, $p = 0.048$; dan RH, $z = -5.587$, $p = 0.001$. Terdapat perbezaan yang signifikan untuk paras ECP di kalangan responden antara dua kawasan, $t = 8.473$, $p<0.001$. Paras ECP juga konsisten dan signifikan dengan kadar laporan simptom radang salur penafasan atas iaitu kahak, hidung berair, hidung tersumbat dan bersin, χ^2 (1, N=98)=6.437, $p<0.05$; χ^2 (1, N=98)=10.516, $p<0.001$; χ^2 (1, N=98)=9.130, $p<0.05$; χ^2 (1, N=98)=10.516, $p<0.001$. Regresi logistik menunjukkan yang VOCs merupakan faktor utama mempengaruhi paras ECP, $\beta=42.596$, $p<0.05$ (PR=6.410, 95% CI=1.268 – 32.394). **Kesimpulan**: Kajian ini telah menunjukkan bahawa pendedahan kepada pencemar udara dalaman akan meningkatkan risiko masalah pernafasan dan mungkin penyebab kepada radang di kawasan mukosa hidung.

Kata Kunci: **Kualiti udara dalaman, kesihatan pernafasan, radang salur pernafasan atas, eosinophil cationic protein (ECP)**

© COPYRIGHT UPM

TABLE OF CONTENT

© COPYRIGHT UPM

© COPYRIGHT UPM

© COPYRIGHT UPM

© COPYRIGHT UPM

LIST OF TABLES

© COPYRIGHT UPM

© COPYRIGHT UPM

LIST OF FIGURES

© COPYRIGHT UPM

LIST OF APPENDICES

© COPYRIGHT UPM

LIST OF ABBREVIATIONS

>	More than
<	Less than
IAQ	Indoor Air Quality
ECP	Eosinophil Cationic Protein
ATS	American Thoracic Society
ISAAC	International Study on Asthma and Allergy in Children
PM_{10}	Particulate Matter to the size of 10 microns and smaller
VOCs	Volatile Organic Compounds
RH	Relative humidity
CO	Carbon monoxide
CO_2	Carbon Dioxide
NO_2	Nitrogen dioxide
USEPA	United States Environment Protection Agency
WHO	World Health Organisation
NHLBI	National Heart, Liver, and Blood Institute

© COPYRIGHT UPM

SCAQMD	South Coast Air Quality Management District
NAL	Nasal lavage
ELISA	Enzyme-Linked Immunosorbent Assay
$\mu g/m^3$	Microgram per meter cube
ppm	Part Per Million
CFU/m^3	Colony Forming Unit per meter cube
ng/mL	Nanogram per milliliter
µL	Microliter
mL	Milliliter
df	Degree of freedom
PR	Prevalence Ratio
CI	Confidence Interval
IQR	Interquartile Range

© COPYRIGHT UPM

CHAPTER 1

INTRODUCTION

1.1 Background

Indoor air is the air that circulates human life that was breathe in inside of buildings be it houses, offices, private establishment, shopping complexes and so on. Meanwhile, indoor air quality (IAQ) is a measure of how clean the air that man breathe in which may not give negative implications to human health. A good indoor air is very important because we spend much of our time in a building, about 80-95% of our life (Decarro *et al*, 2003).

There are many parameters or determinant which can be used to determine the level of indoor air quality. The most common of measurement are taking into consideration indoor air velocity, relative humidity and also temperature. Meanwhile,

© COPYRIGHT UPM

in more advanced studies were often interested in the presence of indoor air pollutants such as particulate matter, VOCs, mould or fungi, bacterial contaminants and also formaldehyde (Yoon *et al*, 2010 & Lignell *et al*, 2005). Level of pollution in indoor air may come from various sources contributed from human activities and characteristics of the building itself. These sources may include human activities such as cooking, smoking, maintenance and cleaning activity with frequency, hobby such that likes to keep pet indoors and personal and so on. Meanwhile, building materials can also contribute to pollution in indoor air with building materials containing asbestos, wet carpet, house paints, furniture made of pressed wood and building cooling and heating system. Outdoor sources of pollutants may also contribute to high level of indoor air pollutants for example house location near industrial areas and construction activities (EPA, 2013).

Many studies have been conducted to determine the relationship of exposure to indoor air pollutants with health effects, particularly which involved respiratory illnesses such as asthma and allergy. However, as indoor air quality research has been well established, there are still very few studies that focus on its health effects among children of lower age; preschool children. Compared to adults and children of older age, this group is more vulnerable to compromised IAQ due to their immature immune systems, greater food intake and inhaled breath per unit mass while also experiencing rapid growth (US EPA, 1995). Moreover, fewer studies have been done to understand their exposure in preschools as most focus on exposure at home. In

© COPYRIGHT UPM

reality, by attending preschools, these children are of increased risk to exposure to many pollutants that may be the culprit of asthma and allergy cases in them (Yoon *et al*, 2010). All these negative effects of poor IAQ in preschool has been found to reduced school attendance, respiratory problems and compromised performance (Yoon *et al*, 2010). All these while, epidemiological studies on paediatric asthma are based on extrapolation from children of older age; 7 years old and above acting as the working definition of asthma (Tan *et al*, 2006).

The issue of IAQ in preschool is now becoming increasingly importance with more studies that try to relate this group of age exposure to indoor air with more acute effect such as airways inflammation. This has become an attractive research to develop as study of the role of airway inflammation in adults has demonstrated association between level of biomarkers with the prevalence of asthma (Frischer and Baraldi, 2000). By understanding the importance of IAQ in preschool setting, this study will be able to jumpstart more researches in the area. This is because exposure in preschool determined the major part of personal exposure for this age of children (Yoon *et al*, 2010).

© COPYRIGHT UPM

1.2 Problem Statement

Children at the age of 4 to 6 years old arguably spend most of their time indoors thus suggesting that exposure to indoor air pollutants is higher. It is understand that preschool children will attend classes in the morning which will last up to 5 hours before their parents pick them up for homes. Then, most of the remaining hours of the day will be spend at homes. Generally, children of that group of age will spend 80% of their time indoor. This will increase their exposure to potential indoor air pollutants.

Poor indoor air quality expose children to the potential harm of particulate matter, biological contaminants such as fungi and bacteria, volatile chemicals or volatile organic compounds (VOCs) and also formaldehyde (Yoon *et al*, 2010 & Lignell *et al*, 2005). Studies also found that concentration of these pollutants are higher indoor than outdoor. Other than that, it is agreed that exposure is higher at home compare to in school.

© COPYRIGHT UPM

15 DEC 2014 PERPUSTAKAAN PERUBATAN DAN SAINS KESIHATAN
UNIVERSITI PUTRA MALAYSIA

Corresponding to these theory, studies in recent decades also seen that the prevalence of asthma and allergies cases increased particularly among children (Broms *et al*, 2013) affirming the statistics proposed by the International Study on Asthma and Allergies in Children (ISAAC). Much research on potential environmental causes of asthma and allergies has been conducted worldwide in correspond to the increase of such cases (Chan-Yeung and Becker, 2006). Most of these researches focused on the exposure at home where majority of children spend their time (Mendell, 2007).

A study of association also found numerous health implications due to exposure to indoor pollutants among this group of children. This suggest that extrapolation of data from study in other children is becoming less relevant to determine the preschool children age group exposure and implications. Factors being use in such studies include hospital admission, school absences, physician diagnosed cases, prevalence of respiratory symptoms, and increased infants mortality (Barnett *et al,* 2005). In most common of studies, exposure to indoor air pollutants often derived from manifestation of respiratory symptoms such as coughing, sneezing, wheezing, breathlessness, runny nose and many others which can relate to allergic reaction. Newer studies also look in association with inflammation of airways to exposure to indoor air pollutants by assessing concentration of biomarkers. All of this helps to also determine the acute and maybe relation to long term effects of

© COPYRIGHT UPM

respiratory diseases in children as a results of exposure to indoor allergens and other pollutants.

Children are a susceptible group in a population. In term of growth, children are still experiencing the development of parts of their bodies and organs making their immune system weaker as compare to adults or even teenagers. Thus, when it comes to exposure of indoor pollutants, they are more sensitive. Their environment will directly influence the growth of their lung function and immune systems (Gordon, 2004). The World Health Organisation (2008) stated that children may exhibit a severe response to exposure of air pollutants as compare to adults.

1.3 Study Justification

The purpose of the study was to determine the upper airways inflammation among preschool children in response to indoor air pollutants exposure in preschools. The study relates upper airway inflammation to the prevalence of reported respiratory symptoms in study areas.

© COPYRIGHT UPM

The study of exposure to indoor air pollutants and its health effect among preschool age children is less established as compare to other groups of age. Often, management of cases such as asthma and allergy among preschool age children is based on extrapolation of data from studies involving children of older age. Study involving preschool age children with exposure to indoor air pollutants should be considered as a step of primary approach towards better avoidance of asthma and allergy in later life; excluding the fact of genetic influences. This is because at this stage in life, their body immune system is weak and still developing. This is also true for other parts of their body and organs. Thus, they need extra protection when it comes to exposure to indoor air pollutants. This study helps to provide insights on asthma and allergy avoidance in preschools.

Other than that, studies involving preschool children exposure in this sense will be advantageous in term of control of confounders. This is because there is low possibility that they are smokers, presence of exposure to occupational chemicals or pollutants; seldom spend time away from the area of their home and schools. The study will be able to determine the real environmental exposure to indoor air pollutants and its health effects. Furthermore, as mention earlier on the developing organs in particular the respiratory system in preschool children tend to be more sensitive towards air pollutants.

© COPYRIGHT UPM

Most study confirmed that concentration of pollutants in indoor air is higher than outdoor air. To add up to the risk, an individual spend most of the time indoor at home, offices, shopping complex, school and others. This is also true in the case of preschool children who spend around 4-6 hours in preschools and much of the remaining hours at home. It is no surprise that the number of asthmatic and atopic continue to increase among this age group. However, not many studies that go as far as measuring sera level of immunoglobulin E (IgE) for example in assessing allergy in preschool children. So far, studies on allergy and asthma among preschool age children in Malaysia are done based on questionnaires alone. Future research will be lacking of data published on biological specimen collection for inflammation of airways for this age group.

As such, the study provides a baseline data for measurement of biomarkers for airway inflammation among preschool children. In addition, this study was the first of its kind in Malaysia that use the collection of nasal swab for analysis of inflammatory biomarkers such as Eosinophil Cationic Protein (ECP), albumin, lysozyme, myeloperoxidase (MPO) and others. Hopefully, the study helped to provide a new insight in evaluation and management of asthma and allergy in preschool age children while also acts as a jumpstart for more research in the area in the future.

© COPYRIGHT UPM

On a bigger scale, the researcher hope the study findings will be of good use to the related Ministry; especially Ministry of Education and Ministry of Health in formulating a better guidelines for preschool setting as it is very important to manage the risk of asthma, allergy and other diseases which may result from exposure to indoor air pollutants in preschool context. Lastly, the study will serves as a public awareness call regarding the issues. This study aimed to educate parents and preschool managements in particular in matters that concern asthma and allergy avoidance and management at homes and preschools as that are where the primary exposure to indoor air pollutants starts.

© COPYRIGHT UPM

1000718924

1.4 Conceptual Framework

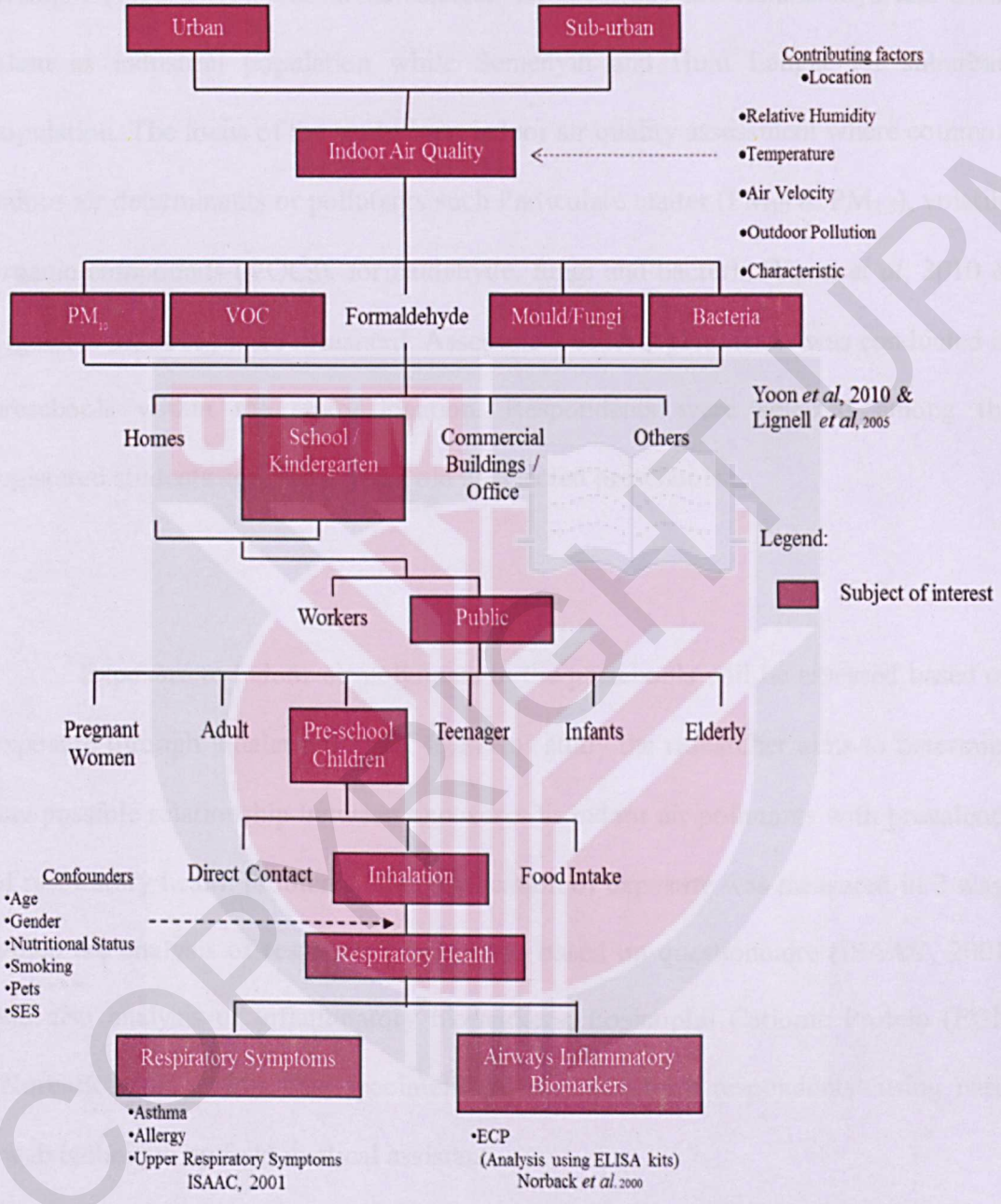


Figure 1.1 Conceptual Framework of indoor air pollutants exposure among preschool children

© COPYRIGHT UPM

This study includes the population within industrial and suburban areas in Selangor (Figure 1.1). The areas selected for the study are Kelana Jaya and Shah Alam as industrial population while Semenyih and Hulu Langat for suburban population. The focus of the study is an indoor air quality assessment where common indoor air determinants or pollutants such Particulate matter (PM_{10} & $PM_{2.5}$), volatile organic compounds (VOCs), formaldehyde, fungi and bacteria (Yoon *et al*, 2010 & Lignell *et al*, 2005) were measured. Assessment of IAQ parameters was conducted in preschools within the study location. Respondents were selected among the registered students age 5 to 6 years old at selected preschools.

Exposure to indoor air pollutants in the preschools will be assessed based on exposure through inhalation. Thus, from this study the researcher aims to determine any possible relationship between exposures to indoor air pollutants with prevalence of respiratory health problems. The implication of exposure was measured in 2 ways which are analysis of respiratory symptoms based on questionnaire (ISAAC, 2001) and also analysis of inflammatory biomarkers; Eosinophil Cationic Protein (ECP) (Norback *et al*, 2000). The specimen was sampled from respondents' using nasal swab technique by field medical assistant.

In addition, there are few contributing factors that were included in the assessment that contributed to the level of pollutants in preschools. These factors

© COPYRIGHT UPM

were location of preschool, relative humidity, temperature, and also the characteristics of the preschool. Lastly, there were also confounders outlined that influenced study findings especially in determining the relationship between exposure to indoor pollutants and their implications. These confounders are age factor, respondents' gender, social economic status, nutritional status, present of pets and smokers at home.

1.5 Research Objectives

1.5.1 General Objective

To study the association between exposure to indoor air pollutants and upper airway inflammation among Malay preschool children in Selangor .

1.5.2 Specific Objectives

1. To determine the demographic and socio-economic status of preschool children in study areas.
2. To compare the concentration of indoor air pollutants concentration in preschool between study areas.
3. To compare ECP level among preschool children between study areas.
4. To compare reported respiratory symptoms between study areas.

© COPYRIGHT UPM

5. To determine the association between indoor air pollutants level with ECP level of the nasal mucosa among preschool children.
6. To determine the association between indoor air pollutants with reported respiratory symptoms among preschool children.
7. To determine the association between ECP level and reported symptoms among preschool children.
8. To determine association between indoor air pollutants exposure, home exposure and ECP level among preschool children.

1.6 Research Hypotheses

1. There is a significant different in indoor air pollutants concentration between preschool between study areas.
2. There is a significant different in ECP level among preschool children between study areas.
3. There is a significant different in number of reported respiratory symptoms between study areas.
4. There is a significant association between indoor air pollutants level with ECP level of the nasal mucosa among preschool children.
5. There is a significant association between indoor air pollutants with reported respiratory symptoms among preschool children.

© COPYRIGHT UPM

6. There is a significant association between ECP level and reported symptoms among preschool children.
7. There is a significant association between indoor air pollutants exposure, home exposure and ECP level among preschool children.

1.7 Study Variables

1.7.1 Independent Variable

Indoor Air Pollutants Exposure via inhalation in preschools setting

1.7.2 Dependent Variable

Eosinophil Cationic Protein (ECP) presence in nasal mucosa sample of the preschool children

© COPYRIGHT UPM

1.8 Definition of Term

1.8.1 Conceptual Definition

a) Particulate Matter 10 (PM_{10})

Particulate matter is a complex mixture of extremely small particles and liquid droplets. They are made up of many components such as duct particles, chemicals, metals and others. PM_{10} and $PM_{2.5}$ are component of these particles which are varying in size. PM10 is also known as inhalable coarse particles with size of more than 2.5 micron but smaller than 10 micron (EPA, 2013).

b) Volatile Organic Compounds (VOCs)

VOCs are emitted gases from certain solid and liquids which include variety of chemicals present indoors or outdoors. However, study found that concentration of indoor VOCs are about 10 times higher than outdoor. Organic chemicals are commonly present in household products such as paints, varnishes, cleaning detergents, and many others (EPA, 2012).

c) Biological Contaminants

The biological contaminants consist of groups such as bacteria, mould or fungi, viruses, house dust, mite and others. In indoor air quality (IAQ),

© COPYRIGHT UPM

present of these contaminants indicates poor IAQ and often associated with allergic diseases. Sources of these contaminants may come from plants pollen, bacteria and viruses carried by human and also animal allergens (EPA, 2012).

d) Respiratory Symptoms

Respiratory symptoms include cough, shortness of breath, body ache and difficulty in breathing (Ministry of Health, Malaysia).

e) Airways Inflammation

Airway inflammation is defined by infiltration of the airway by inflammatory cells which contribute to elevated levels of inflammatory mediators and procontractile stimulants (Khan, 2013).

f) Inflammatory Biomarkers

These are inflammatory cells such as eosinophils, mast cell, monocytes, lymphocytes, active complement fragments (C3a and C5a), and neutrophils (Khan, 2013)

g) Preschool Children

Children of the age of 4 to 6 years old, preparing to enter primary school (Ministry of Education, 2013).

© COPYRIGHT UPM

h) Industrial Area

Industrial areas are land designated by the local authority for the purpose of sitting industrial, manufacturing and processing plants, factories or facilities (The Planning Guidelines for Environmental Noise Limits and Control). All these facilities are built to contribute to production of products to support our daily life and demands as the result of increasing number of population (Department of Statistics, Malaysia. 2010).

i) Suburban area

Suburbs are medium density areas outside a town or city with population of 75 to 200 persons per acre (The Planning Guidelines for Environmental Noise Limits and Control).

1.8.2 Operational Definition

a) Particulate Matter 10 (PM_{10})

Particulate Matter 10 (PM10) in this study was measured based on exposure in preschools. The concentration of particles were measured using DustTrak™ DRX Aerosol Monitor 8534 which is a handheld battery-operated, data-logging, light-scattering laser photometer that gives real-time aerosol mass readings. It uses a sheath air system that isolates the aerosol in the optics clean for improved reliability and low maintenance. The DustTrak™ DRX Aerosol Monitor 8534 is capable of

© COPYRIGHT UPM

measuring aerosol concentrations corresponding to PM_1, $PM_{2.5}$, Respirable or PM_{10} size fractions with concentration range from 0.001 to 150 mg/m^3. Measurement of PM_{10} for this research was done in mg/m^3.

b) Volatile Organic Compounds (VOCs)

VOCs were other determinants of indoor air quality that were measured in this study in preschools environment. Concentrations of volatile chemicals were measured using ppbRAE Volatile Organic Compound (VOC) Monitor (Model PGM-7240) which is an extremely sensitive Photo-ionization Detector (PID) for real-time monitoring of volatile organic compounds (VOCs) at part per billion (ppb) levels. The device is capable of measuring VOCs concentration ranging from 1 part per billion (ppb) up to 10,000 part per million (ppm). As for this study, the measurement was taken as part per million.

c) Biological Contaminants

In this study, fungi or mould and bacteria are measure based on airborne presence and sample taken using air sampler which is the pbi DuoSAS Super 360. The equipment uses surface air system where air is aspirated at a fixed speed for variable time. The resulting laminar air flow was directed onto the agar surface containing medium consistent with the microbial examination being made. Airborne fungal/ mould bodies will

© COPYRIGHT UPM

be collected on sabaroud dextrose agar while airborne bacterial will be collected on Tryptic soy agar. Total colony forming of both samples will be calculated in the measurement of colony forming unit (cfu/m^3).

d) Respiratory Symptoms

Presence of respiratory symptoms among preschool children selected for the study was determined based on a set of questionnaire which was answered by parents. The questionnaire was a modified set derived from internationally standardized question by the International Study of Asthma and Allergies in Childhood (ISAAC, 2001) and also American Thoracic Society (ATS). The symptoms are cough, phlegm, runny nose, blocked nose, sneezing, and etc. Statistical analysis will be done to determine relationship with exposure to indoor air pollutants in preschools while also comparing the prevalence between 2 different study areas.

e) Airways Inflammation

The association between exposure to indoor air pollutants and presence of respiratory problem was also indicated by occurrence of upper airway inflammation. In this study, sample of nasal swab was obtained from the respondent's by inserting cotton swab stick, immersed beforehand in normal saline solution into middle of inferior turbinate and was gently

© COPYRIGHT UPM

rotated 90-180° on the nasal wall. The swab was then inserted in collecting tube containing 2ml of 0.9% NaCl solution. The same process was done for both nostrils. Biomarker found in sample was detected to measure the occurrence of nasal inflammation.

f) Inflammatory Biomarkers

Nasal swab specimen that was collected to measure upper airway inflammation was centrifuged at 500g and 1000g to separate the cell in the specimen. Inflammatory biomarker analysed in this study was Eosinophil Cationic Protein (ECP) present in the sample. Analysis was done using commercial enzyme-linked immunosorbent assay (ELISA) kit with concentration measure at ng/mL.

g) Preschool Children

Preschool children in this study were those in the age of 5 to 6 years old and registered at the preschools within the study areas of Shah Alam and Kelana Jaya (industrial) and also in Semenyih and Hulu Langat (suburban). The preschool children are of Malay ethnicity and healthy; free from respiratory illnesses.

© COPYRIGHT UPM

h) Industrial

Industrial areas were categorised by dense activity of factories and warehouses in the area. Any preschools located within the 5 kilometre radius of that area were included in the sample population. For representation of this population, Shah Alam and Kelana Jaya were selected and 42 respondents were included from that areas.

i) Sub-urban

Suburbans are areas at the outskirt of big city where in this study, Semenyih and Hulu Langat were selected to represent suburban population. Any preschools within the radius of 5 kilometres of the area were included as suburban preschool. 42 preschool children were selected as representation of this area.

© COPYRIGHT UPM

CHAPTER 2

LITERATURE REVIEW

2.1 Indoor Air Pollutants and Respiratory Health Effects

Indoor air quality (IAQ) is a term which refers to the air quality within and around buildings and structures, especially as it is related to the health and comfort of building occupants. Nowadays, indoor air-borne pollution is of concern due to its universality of human exposure especially those who are spent most of their time in indoors. It is not surprisingly that most people in the industrialized world spent about 60% of their time in the dwelling and about 90% could be spent outdoors (World Health Organization, 1999).

Poorer indoor air quality possesses threat to the building occupants. In the United States, the Environmental Protection Agency lists poor indoor air quality as the fourth largest environmental threat (EPA, 2013). The elements within our home

© COPYRIGHT UPM

and workplace have been increasingly recognized as threats to our respiratory health. These elements can be derived directly either from natural resources, combustion or man-made sources.

There are various types of airborne pollutants that may play a substantial role in the development and morbidity of asthmatic respiratory illness and allergies. The major indoor pollutants include both chemicals (nitrogen dioxide, ozone, sulfur dioxide, particulate matter, and VOCs) and biological parameters (dust mites, pet allergens, and mould) (Kim *et al,* 2013; Bernstein *et al*, 2008; Duhme *et al*, 1998).

Particle's behaviour for each indoor pollutant depends on its size. Larger particles are rapidly settling down compared to smaller particles where they remain airborne for long periods in the atmosphere which can be inhaled directly. The particle size of indoor pollutants also affects the rate of penetration and deposition in the airways and lungs. In this matter, ultrafine particles (UFP) play major roles in causing negative health impacts to human respiratory health where the rate of penetration and deposition of particles is deeper in the alveolar region. Figure 2.1 below indicates the common sources of pollutants which can be found inside buildings.

© COPYRIGHT UPM

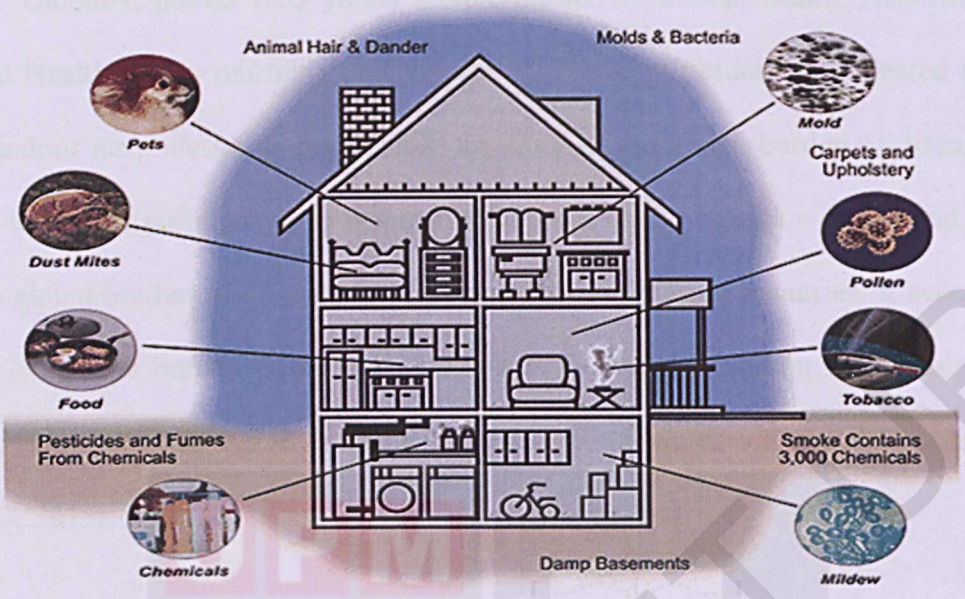


Figure 2.1 Common sources of indoor pollutants.

Source: Florida Health, Department of Health (2013)

The indoor environment in any building is a result of the interaction between the site, climate, building system (original design and later modifications in the structure and mechanical systems), construction techniques, contamination sources (building materials and furnishings, moisture, processes and activities within the building, and outdoor sources), and building occupants. Apart from that, other contributing factor that affects the indoor environment are such as temperature, relative humidity, air velocity as well as the location of building.

© COPYRIGHT UPM

Globally, poorer IAQ yields a huge burden on human health. According to Global Health Risks, mortality and burden of disease attributable to selected major risks indoor air pollution is responsible for 2.7% of the global burden of disease. In 2000, indoor air pollution was responsible for more than 1.5 million deaths and 2.7% of the global burden of disease. In high mortality developing countries, it accounted for 3.7% of the burden of disease, making it the most important risk factor after malnutrition, the HIV/AIDS epidemic and lack of safe water and adequate sanitation (WHO, 2013).

Indoor allergen exposure may be important in childhood atopic disease development (Kim *et al,* 2013, Gold, 2000, McHugh, 2011) and influence morbidity (Ownby, 2010). Asthma severity in children can be related to the level of exposure to common indoor allergens such as dust mite and cat allergens (Gent, 2009). A review article concluded that allergen exposure may cause asthma, be protective, or have no effect, depending on the type of allergen, age of exposure, route of exposure, dose of exposure and underlying genetic susceptibility (Arshad, 2010).

Some biological contaminants may trigger allergic reactions, including hypersensitivity pneumonitis, dyspnea, wheeze, allergic rhinitis, and some types of asthma. Several symptoms of health problems caused by biological pollutants may include sneezing, watery eyes, coughing, and shortness of breath, dizziness, lethargy,

© COPYRIGHT UPM

fever, and digestive problems. This is supported by large number of studies in many geographical regions which have found consistent associations between evident indoor dampness or mold and respiratory or allergic health effects in infants, children, and adults (WHO, 2009).

Children are particularly vulnerable to respiratory conditions because of their developmental stage and physical differences from adults (Faustman *et al*, 2000). Children's lungs and airways are immature and especially susceptible to insults from pollution. The developing lungs present a large surface area through which pollutants may be easily absorbed (Landrigan, 1998). Children breathe faster and therefore inhale and absorb a relatively greater volume of contaminants compared to adults (Faustman *et al*, 2000).

2.2 Children Respiratory System and Deposition of Air Pollutants

The respiratory system is made up of organs and tissues that help human breath. This system involved in the interchanges of gases and consists of the nose, mouth (oral cavity), pharynx (throat), larynx (voice box), and trachea for the upper respiratory tract. While for the lower respiratory tract include bronchi and bronchioles (airways), alveoli and lungs.

© COPYRIGHT UPM

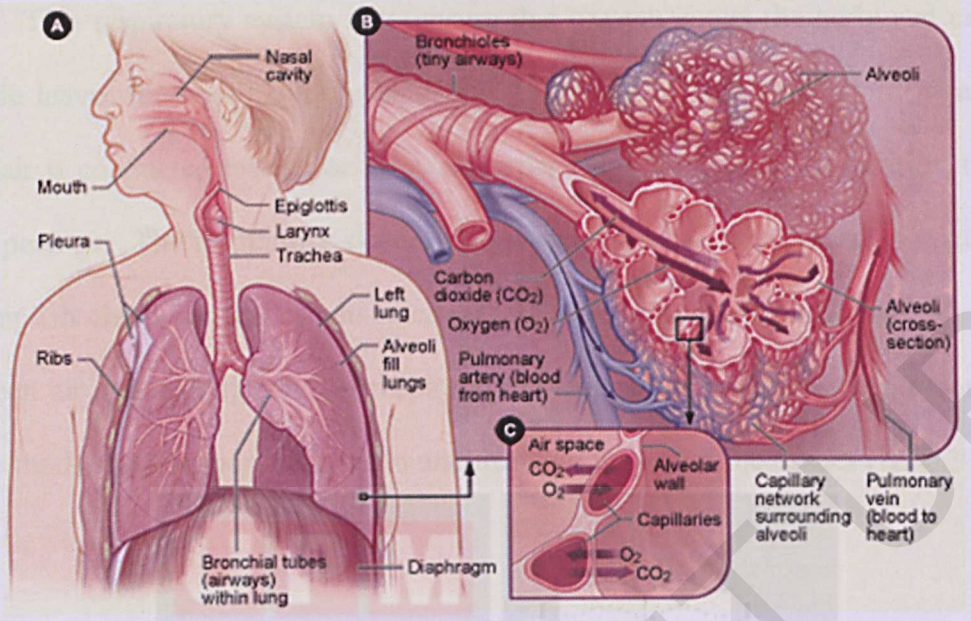


Figure 2.2 the Respiratory System

Source: NHLBI (2012)

Based on the Figure 2.2, Figure A shows the location of the respiratory structures in the body which includes the nasal cavity, mouth, pharynx, larynx, epiglottis, trachea, bronchial tubes, pleura, ribs, lungs, alveoli and diaphragm. For the Figure B, it is an enlarged view of the airways, alveoli (air sacs), and capillaries. Figure C is a close-up view of gas exchange between the capillaries and alveoli (NHLBI, 2012).

© COPYRIGHT UPM

This respiratory system is to ensure that oxygen enters the body and carbon dioxide leaves the body. During inhalation (breathing in) and exhalation (breathing out), air is conducted toward or away from the lungs by a series of cavities, tubes, and openings. The respiratory system also works with the cardiovascular system to accomplish these several events include external respiration (exchange of gases between air and blood), internal respiration (exchange of gases between blood and tissue fluid) and transport of gases to and from the lungs and the tissues (Hill, 2005).

Respiration begins when oxygen enters into the body through the nose and the mouth. The oxygen then travels through the trachea and pharynx where the trachea divides into two bronchi. Here the bronchi are divided into bronchial tubes, in the chest cavity, so air can be directly moved into the lungs. The nose is the primary upper respiratory organ in which air enters into and exits from the body. Cilia and mucus line the nasal cavity and traps bacteria and foreign particles that enter in through the nose. In addition, air that passes through the nasal cavity will be filtered, warm and moisten. For the pharynx, it act as a connection to surrounding region which allow air to pass from the mouth to the lungs. It contains three parts which are the nasopharynx (connects the upper part of the throat with the nasal cavity), the oropharynx (positioned between the top of the epiglottis and the soft palate), and the laryngopharynx (located below the epiglottis). From the pharynx, air enters into the larynx, commonly called the voice box. The role of larynx are passage for air to enter into the lungs, and a source of sound production (Hill, 2005).

© COPYRIGHT UPM

Trachea is the one of the lower respiratory tract that act as a passage of air to bronchi. The mucous membrane that lines the trachea has an outer layer of pseudostratified ciliated columnar epithelium. The cilia that project from the epithelium keep the lungs clean by sweeping mucus, produced by goblet cells, and debris toward the pharynx. Trachea is a flexible tube that connects larynx with the bronchi. The bronchi allow the passage of air to the lungs. The lungs are paired, cone-shaped organs that occupy the thoracic cavity, except for the central area that contains the trachea, the heart, and esophagus. The lungs have about 300 million alveoli, with a total cross sectional area of 50 to 70 m^2 (meter square). Each alveolar sac is surrounded by blood capillaries. The wall of the sac and the wall of the capillary are largely simple squamous epithelium (thin flattened cells) and this facilitates gas exchange. Gas exchange occurs between air in the alveoli and blood in the capillaries (Figure 2.2 C) (NHLBI, 2012). Oxygen diffuses across the alveolar wall and enters the bloodstream, while carbon dioxide diffuses from the blood across the alveolar wall to enter the alveoli. The alveoli of human lungs are lined with a surfactant, a film of lipoprotein that lowers the surface tension and prevents them from closing. The lungs collapse in some newborn babies, especially premature infants, who lack this film. The condition, called infant respiratory distress syndrome, is now treatable by surfactant replacement therapy (Hill, 2005).

Respirable particles and gases affect different parts of the respiratory tree depending upon their inherent characteristics (Figure 2.3). For gases, relative

© COPYRIGHT UPM

solubility is important. Note that sulfur dioxide, because it is highly water soluble, initially affects the upper airway, whereas ozone, which has medium solubility, initially affects the middle airways, and nitrogen dioxide, which has low solubility, initially affects the lower airways.

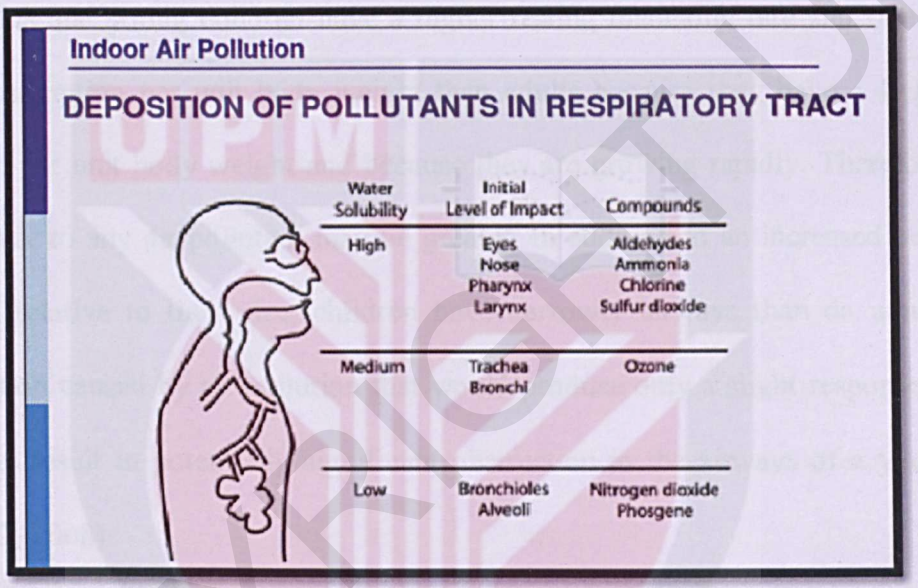


Figure 2.3 Deposition of pollutants in respiratory tract

Source: WHO (2008)

In addition, particle size is the most important factor in determining where particles are deposited in the lung. Compared with large particles, fine particles can remain suspended in the atmosphere for longer periods and be transported over

© COPYRIGHT UPM

longer distances. Some studies suggest that fine particles have stronger respiratory effects in children than large particles. The particles greater than 10 micrometres rarely make it past the upper airways, whereas fine particles smaller than 2 micrometres can make it as far as the alveoli (WHO, 2008).

Infants and young children have a higher resting metabolic rate and rate of oxygen consumption per unit body weight than adults because they have a larger surface area per unit body weight and because they are growing rapidly. Therefore, their exposure to any air pollutant may be greater. In addition to an increased need for oxygen relative to their size, children have narrower airways than do adults. Thus, irritation caused by air pollution that would produce only a slight response in an adult can result in potentially significant obstruction in the airways of a young child (WHO, 2008).

2.3 Mechanism of Upper Airway Inflammation

When inhale, human will breathe in air along with any particles that are in the air. The air and the particles will travel into the respiratory system (lungs and airway). Along the way, the particles can stick to the sides of the airway or travel deeper into the lungs. Smaller particles can pass through the smaller airways. While

© COPYRIGHT UPM

bigger particles are more likely to stick to the sides or get wedged into one of the narrow passages deep in the lung. Other factors that affect how deep into the lungs particles can penetrate are by way of mouth or nose breathing which breathing through mouth allows particles to travel deeper into the lungs than nose. Besides, exercising allow particles to travel deeper in the respiratory system (WHO, 2008). High exposure to particle can affect the respiratory, cardiovascular, circulatory, and nervous system through multiple potential pathways, some of which might be appropriate for linking air pollution with birth outcomes (Xiaohui Xu, 2011).

Additionally, a number of pollutants, including ozone, Sulfur Dioxide (SO_2) and Nitrogen Dioxide (NO_2), have proinflammatory actions. Each of these gases has been shown to induce proinflammatory cell influx into the airway, and also cause a number of proinflammatory changes in epithelial cells. Indeed, initial signals from the epithelium, such as the release of various cytokines and lipids, may be fundamental in mediating the proinflammatory action of pollutants (Figure 2.4).

© COPYRIGHT UPM

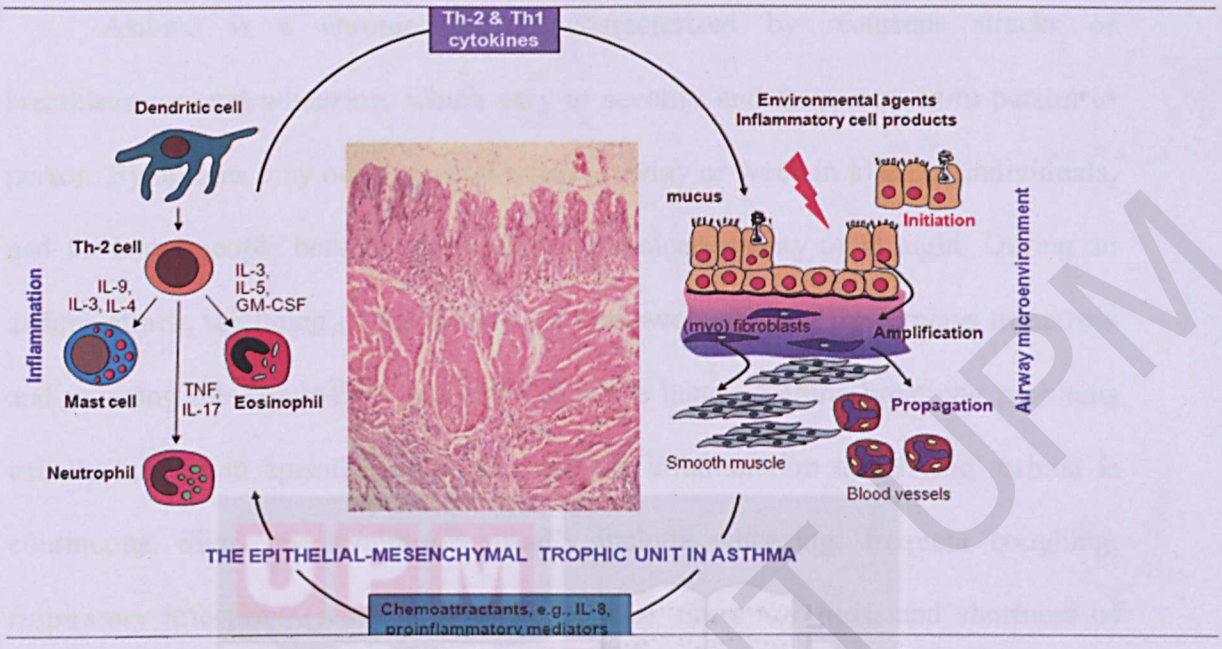


Figure 2.4 Inter-relationship between immunological and inflammatory mechanisms and the structural elements of the airways in asthma pathogenesis.

Source: Holgate (2010)

Asthma is a disease characterized by increased airway sensitivity to inhaled irritants, increased airway inflammation, and injury to the epithelium. Given the effect of ambient air pollutants, such as ozone, SO_2, and NO_2, on lung mechanics, airway inflammation, and proinflammatory changes in epithelial cells, it seems likely that asthmatics may be more sensitive than the general population to the effects of these gases. The proinflammatory effect of these agents suggests that they may not only exert direct effects in asthma, but that they may prime the airways, such that the response of extrinsic asthmatics to inhaled allergen is also enhanced (Peden, 1997).

© COPYRIGHT UPM

Asthma is a chronic disease characterized by recurrent attacks of breathlessness and wheezing, which vary in severity and frequency from person to person. Symptoms may occur several times in a day or week in affected individuals, and for some people become worse during physical activity or at night. During an asthma attack, the lining of the bronchial tubes swell, causing the airways to narrow and reducing the flow of air into and out of the lungs. Asthma breathing problems usually happen in episodes or attacks but the inflammation underlying asthma is continuous. Signs and symptoms usually include wheezing, frequent coughing, respiratory infections (pneumonia or bronchitis), chest tightness, and shortness of breath. Recurrent asthma symptoms frequently cause sleeplessness, daytime fatigue, reduced activity levels and school and work absenteeism (WHO, 2013).

2.4 Nasal Swab

The nasal sampling using lavage, swab or simply blowing the nose is an easy technique to be used, non-invasive, and non-traumatic to the sample subject, and multiple sequential samples are able to be collected from the same person (Frischer and Baraldi, 2000). For these particular reasons and because there is no special equipment is required, the nasal swab is an attractive and cost effective approach for epidemiologic and occupational studies. It also applicable in determining which air pollutants are capable of causing an inflammatory reaction in the human respiratory

© COPYRIGHT UPM

tract. It is important in determining the risk of normal subjects or susceptible groups such as asthmatics (Hillel *et al*, 1990).

Besides the advantages mentioned, this method was found to be more significant in sampling dust for allergen exposure assessment if compared to other methods (Renstrom, 2002) (Figure 2.5). This method is also able to be safely applied to almost every child as it is well tolerated and simple. Because of its simplicity, it can be used in field studies involving children and even infants as young as 4 weeks old (Frischer and Baraldi, 2000).

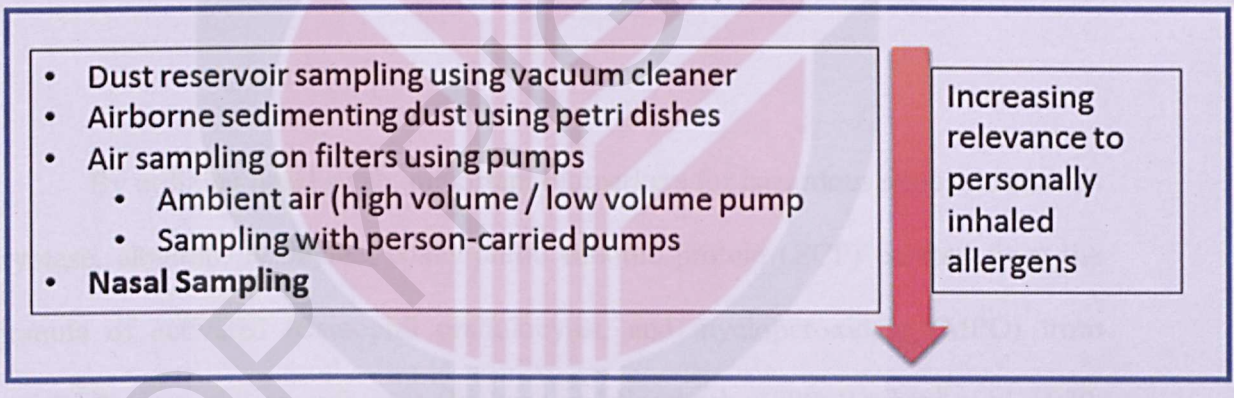


Figure 2.5 Relevance to personally inhaled allergens sampling methods

Source: Renstrom (2002).

The nasal swab was done by adapting the method from a previous study (Norback *et al*, 2000, Boot *et al*, 2008, Renstrom, 2002 and Klimek & Rasp, 1999).

© COPYRIGHT UPM

Normal saline solution (0.9% NaCl) was prepared prior to nasal sampling. Cotton swab stick was immersed in the normal saline solution before being inserted into respondent's nostril to the middle of the inferior turbinate. The cotton swab was gently rotated 90-180° on the nasal wall. The swab was then put into collecting tube containing 2ml of normal saline solution. The process was repeated for the other nostril. Sample was analyzed using the ELISA kit for human ribonuclease 3 gene specific for ECP. The detailed description on the collection and analysis method were clearly discussed in Chapter 3.

2.5 Eosinophil Cationic Protein (ECP) As Inflammatory Biomarker

By utilizing nasal swab, important biomarkers for hazardous exposure such as tryptase, albumin, lysozyme, eosinophilic cationic protein (ECP) emitted from the granula of activated eosinophil granulocytes, and myeloperoxidase (MPO) from activated neutrophil granulocytes (Pipkorn and Enerback, 1989; Raphael *et al*, 1989; Walinder, 1999) can be found. In a previous study, significant effects for ECP or lysozyme in NAL (Wieslander, 1999) was found. This is a similar trend as in another previous studies applying nasal sampling in indoor air epidemiology (Walinder *et al*, 1998, 1999).

© COPYRIGHT UPM

Eosinophil plays an important role in the inflammatory process. Eosinophil number will increase in the blood and tissues of patients with allergic disease. In addition, eosinophils recruited massively in reaction to specific allergen in several organ and biological fluids such as in the NAL. Among inflammatory mediators of human eosinophils, granule cationic proteins have been well characterized (Klimek & Rasp, 1999). The reports of increase ECP in biological fluids (NAL) or in serum of patients with allergic disease suggesting that the measurement of ECP level might be useful as a clinical test in monitoring disease activity and the efficacy of therapeutic agents. ECP secretion commonly associated with allergic rhinitis but can also be used for acute exposure effects in healthy individuals trigger by irritants and pollutants (Chawes, 2011).

© COPYRIGHT UPM

CHAPTER 3

METHODOLOGY

3.1 Study Design

This research was a cross sectional study design to determine the association of preschools exposure to indoor air pollutants with upper airway inflammation using detection of inflammatory biomarker, Eosinophil Cationic Protein (ECP) in nasal swab sample among preschools children in different areas in Selangor. The study was also able to determine the prevalence of respiratory symptoms in study areas and determine relationship with upper airway inflammation.

© COPYRIGHT UPM

3.2 Study Location

The study locations were chosen based on the potential of surrounding environment to contribute towards indoor air pollution. Registered preschools in the industrial and suburban areas of Selangor were selected in order to find variations and pattern of indoor pollutants and upper airway inflammation among preschool children. The research seeks to determine the influence of different socioeconomic activities between the study areas with concentration of indoor air pollutants. Thus, 1 preschool in Shah Alam and 2 more in Kelana Jaya were selected to represent the industrial population. 3 preschools in Semenyih and another 1 preschool in Hulu Langat were selected for suburban population after carrying out site survey studies. (Refer Figure 3.1, Figure 3.2, Figure 3.3 and Figure 3.4)

© COPYRIGHT UPM

Figure 3.1 Area location map for study group in Shah Alam (Industrial)
Source: Google Map

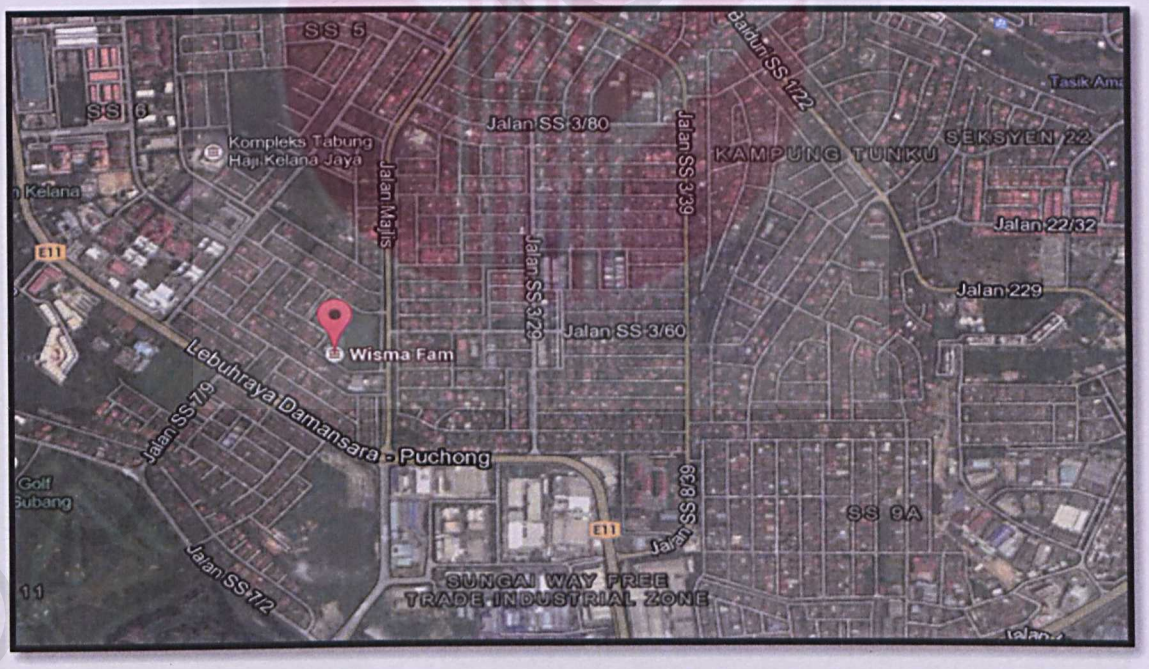


Figure 3.2 Area location map for study group in Kelana Jaya (Industrial)
Source: Google Map

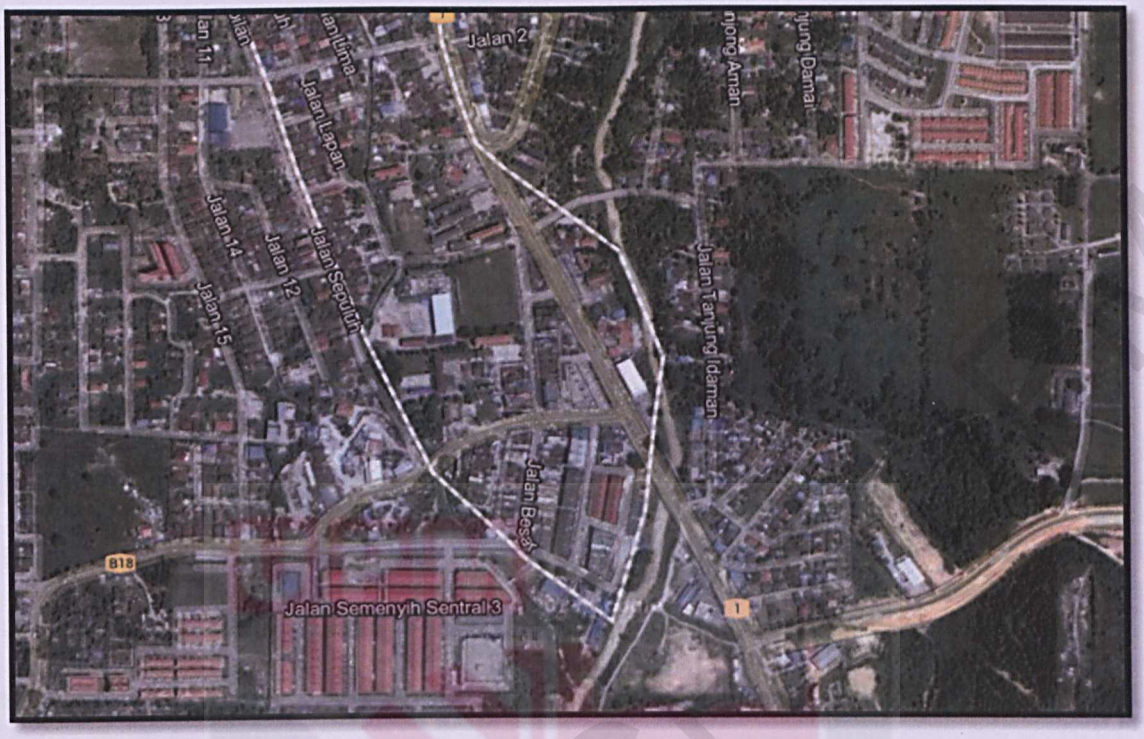


Figure 3.3 Area location map for study group in Semenyih (Suburban)
Source: Google Map

Figure 3.4 Area location map for study group in Hulu Langat (Suburban)
Source: Google Map

3.3 Sampling

3.3.1 Sample Population

A total of 98 preschool children aged 5 to 6 years old from preschools in Shah Alam, Kelana Jaya, Semenyih and Hulu Langat was included in this study. From that, both male and female students were selected of Malay ethnic background in all study areas. Shah Alam and Kelana Jaya were selected as industrial area due to concentrated present of industrial setting of factories and warehouses. Besides that, school were only selected despite randomly, based on an approved list given by Jabatan Kemajuan Masyarakat Negeri Selangor. For suburban, there were less or no industrial setting found in Semenyih and Hulu Langat thus justifying the choice made to select the area. Only Malay respondents were included in this study to homogenise the sample in order to avoid confounding by genetic factor.

3.3.2 Sampling Frame

Sampling frame included all boys and girls students registered at selected preschools in Shah Alam, Kelana Jaya, Semenyih and Hulu Langat which had their parents' consent to participate in the study. All respondents were of age 5 to 6 years

© COPYRIGHT UPM

old. List of registered students was obtained from the teachers of their respective preschools with inclusion of healthy children without evidence of diagnosed asthma, allergy and other respiratory illness.

3.3.3 Study Sample

In this research, there were 53 preschools children selected in industrial study areas while 45 preschool children selected for suburban areas. The total number of study sample was 98 preschool children. The parents of the respondents must approve the permission to include their children in the study in addition to fulfilling the inclusive criteria.

3.3.4 Sample Size Calculation

The objective of the study was to estimate the mean difference of respiratory health implication of exposure to indoor air pollutants in preschools at 2 different areas. The symptoms in study was coughing among healthy respondents and those who reported to have asthma based on questionnaire, the estimation was calculated with reasonable level of precision. It is confident that 95 percent (%) of the case

© COPYRIGHT UPM

sample estimates will fall within 1.96 standard errors (Z1-?) of the specified population value, if it was true value. Therefore, the sample size calculation was based on Lemeshow et.al (1990) formula for group comparison study using the combined (or pooled) standard deviation for the two groups as follows:

Formula,

$$n = \frac{2 \text{ x} 2 \sigma^2 [Z_{1-\alpha/2}+Z_{1-\beta}]^2}{(\mu 1 - \mu 2)^2}$$

Where,

$2\sigma^2$ = Estimated standard deviation (assumed to be equal to each group) $\mu 1$ = Estimated mean (larger) $\mu 2$ = Estimated mean (smaller)

Z1-? = Standard error associated with confidential interval, 95% CI=1.96

Z1-? = Standard error associated with power, 80% of power =0.84

Prevalence of reported coughing among study group:

$$n = \frac{4(0.68)^2[1.96+0.84]^2}{(0.792-0.146)^2}$$

n = 35 (Yahaya and Jalaludin, 2013)

© COPYRIGHT UPM

Based on the formula, 35 respondents were needed for each study areas (industrial and suburban). Thus, the total numbers of needed respondents were 70. The number of respondents were increased by 20% for the strength of analysis of the study and to take into account non responsive respondents, missing data and errors. Therefore, the total number of samples included in this study were 84 samples.

3.3.5 Sampling Method

This study applied stratified random sampling method to determine the preschools into areas in Selangor. Then, random sampling was used where respondents were recruited based on willingness to participate after parents' approval. This was done after obtaining students' name list and distribution of questionnaires. The purpose and procedures of the study were explained to preschool teachers and parents prior to distribution of consent form and questionnaires. Through the questionnaires, the preschool children that fulfilled the inclusive criteria were selected as respondents. Meanwhile, incomplete questionnaires or children who have their parents' disapproval to participate in the study will be excluded.

© COPYRIGHT UPM

Inclusive Criteria:

a) Preschool children aged 5 to 6 years old

- ❖ Both boy and girl students
- ❖ Registered at preschools within the study areas

b) Malay ethnic

- ❖ To homogenised the respondents so that no confounding by genetic factors which may influence the results validity

c) Free from respiratory illnesses

- ❖ Respondents have no history of doctor-diagnosed respiratory illnesses
- ❖ This is to ensure that results are based on studied exposure and not attributed to medical history status

Exclusive Criteria:

a) Preschool children aged below 5 years old

b) Preschool children aged above 6 years old

c) Respondents of other ethnicity than Malay

d) Respondents with history of doctor-diagnosed respiratory illnesses

© COPYRIGHT UPM

3.4 Study Instrumentation

3.4.1 Questionnaire

This study used modified questionnaire for preschool children based on 2 internationally standardised and recognised questionnaires. Along with the informed consent form, parents who were willing to allow their children to participate in the study filled in a questionnaire set by American Thoracic Society (ATS) and also questionnaire on asthma, allergies and respiratory symptoms obtained from International Study of Asthma and Allergies in Childhood (ISAAC, 2000) study.

The ATS questionnaire was used to obtain data on 2 parts which were; Part 1-Data on demographic and socioeconomic background of the respondents while Part 2-Data on school and home environment as well as respondent's respiratory health. From the data of the ISAAC questionnaire, we were able to determine preschool children with recurrent respiratory symptoms and those without the symptoms.

© COPYRIGHT UPM

3.4.2 Preschools Survey and Inspection

The survey and inspection was done to determine the characteristics of preschools which could well influence the exposure to indoor air pollutants. Example of the characteristics investigated were preschools location with information of activities and potential source of pollutants emission in the area, ventilation type of the building, materials use for building construction, buildings age and history of major maintenance or rebuilding works, hygiene and cleaning history, number of occupants, location of kitchen and bathroom and others.

Verbal and telephone interviews with preschools teacher and children's parents were done to obtain the necessary information.

3.4.3 Indoor Air Quality (IAQ) Assessment

IAQ assessment was done to measure the quality of air and presence of indoor pollutants in preschools environment. Approval to enter and take measurement at preschools was obtained prior to data collection date.

© COPYRIGHT UPM

IAQ parameters that were measured in this study are concentration of PM_{10}, VOCs, microbial contaminants such as mould and bacteria. For measurement of parameter, researcher was using TSI DustTrak™ DRX Aerosol Monitor 8534, ppbRAE Portable VOC Monitor (ppbRAE 3000), TSI Velocalc Plus Model 8386, and pbi DuoSAS Super 360.

Measurement of PM_{10}, VOCs, and microbial contaminants was taken at 0.6-1.0 meter above the floor to imitate the breathing zone level of the children. Number of sampling point was 1 for each classroom where the instrument will be placed at location that the children most common at. For example, at the middle of the classroom to represent average exposure point, assuming even distribution of pollutants. The selected sampling point was also more than 1 meter from the wall, a door or an active heating system.

Meanwhile, the measurement of temperature and relative humidity were taken periodically and spread throughout many areas in building to be sure that they will be distributed evenly.

© COPYRIGHT UPM

Assessment of PM_{10}

The DustTrak™ DRX Aerosol Monitor 8534 (Figure 3.5) is a handheld battery-operated, data-logging, light-scattering laser photometer that gives real-time aerosol mass readings. It uses a sheath air system that isolates the aerosol in the optics clean for improved reliability and low maintenance. It is suitable for clean office settings as well as harsh industrial workplaces, construction and environmental sites, and other outdoor applications. The DustTrak™ DRX Aerosol Monitor 8534 measures aerosol contaminants such as dust, smoke, fumes, and mists.

The DustTrak™ DRX Aerosol Monitor 8534 is capable of measuring aerosol concentrations corresponding to PM_1, $PM_{2.5}$, Respirable or PM_{10} size fractions with concentration range from 0.001 to 150 mg/m^3. In this study, researcher was measuring PM_{10} concentrations only in preschool. The flow rate of the instrument was set at 1.7 litre per minute (L/min) fixed for 4 hours inside the preschools (continuous measurement) and 30 minutes measurement done for particulate matter concentration in outdoor air at the preschools to observed influences by outdoor environment into indoor. TRAKPRO™ Data Analysis Software was used to analyse and extract logged data from the equipment.

© COPYRIGHT UPM

Figure 3.5 DustTrak™ DRX Aerosol Monitor 8534 (Source: TSI Website)

Assessment of VOCs

The ppbRAE parts per billion (ppb) Volatile Organic Compound (VOC) Monitor Model PGM-7240 (Figure 3.6) is an extremely sensitive Photo-ionization Detector (PID) for real-time monitoring of volatile organic compounds (VOCs) at ppb levels. With its highly compact design, it is used as a broadband VOC gas monitor and data logger for work in hazardous environments. The new RAE patented PID sensor has an increased sensitivity down to a few ppb, with reduced humidity interference, improved linearity, and an easily accessible lamp and sensor. The device is capable of measuring VOCs concentration ranging from 1 part per billion (ppb) up to 10,000 part per million (ppm).

© COPYRIGHT UPM

Again, using similar method of measurement, concentration of VOCs in preschool classroom was taken continuously for 4 schooling hours. For logged data extraction and analysis, ProRAE Remote Software was used.

Figure 3.6 ppbRAE Portable VOC Monitor Model PGM-7240 (Source: RAE System Website)

Assessment of Microbial Contamination

The pbi DuoSAS Super 360 (Figure 3.7) is a twinhead surface air sampler for simultaneous sampling of mould and bacteria or can also be used for one type of sampling at one time. Using the principle of surface air system (SAS), certain volume of air is aspirated at a fixed speed for variable time through a cover which has been machined with a series of small holes of a special design. The resulting

©COPYRIGHT UPM

luminar air flow is directed onto agar surface of a contact plate containing desired medium suitable for microbial being assessed.

During sampling at preschools, the microbial contaminants were measured based on 500 liters of air sampled with measurement was done in duplicate with one reading was done around 9am while anther at 11am. Measurement was done simultaneously for both fungi and bacterial contaminants.

The method used to measure for colony culture using collection of microbial contaminants on Tryptic soy agar with 500mg cycloheximide for bacterial isolation and sabaroud dextrose agar with 100mg cycloheximide to isolate fungi. Sample collected is then sealed and transported to the lab for incubation. The agar plates for bacterial sample were incubated at 37°C for 24 hours before calculation for colony forming (cfu/m^3) while for mould samples; they were left at room temperature for 5 days before colony counting was done.

© COPYRIGHT UPM

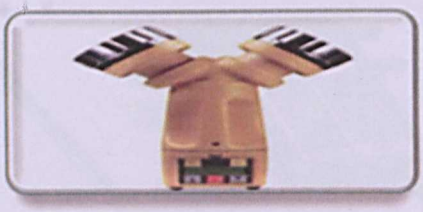


Figure 3.7 pbi DuoSAS Super 360. (Source: International pbi Spa Website)

Assessment of Temperature and Relative Humidity

TSI Q-Trak™ Indoor Air Quality Monitor 7575 (Figure 3.8) is one of the instruments that are used in IAQ assesment. This instrument is capable of measuring humidity, temperature , CO_2 and CO in the ambient air of indoor air. In this study, periodic measurement of temperature and relative humidity were taken and spread throughout many areas in preschools to be sure that they will be distributed evenly. To increase accuracy, readings were taken 3 times and mean readings is obtained.

© COPYRIGHT UPM

Figure 3.8 TSI Velocalc Plus Model 8386 (Source: TSI Website)

3.4.4 Collection of Nasal Swab

Nasal swab was collected from each respondent to study the inflammation of nasal mucosa. Selection of nasal lavage was chosen because the sample is easily accessible and the procedure is non-invasive. Adopting the techniques done in previous studies such as Norback *et al* (2000), Boot *et al* (2008), Renstrom (2002) and Klimek & Rasp (1999), the researcher was able to establish a collection method suited to Malaysian preschool children's setting.

The procedures for collection of nasal swab were as follows; the child will be asked to sit on a chair with his head hyper-extended or flexed about 30° forward, which is to the back of the neck. A cotton swab stick (HmbG brand) immersed in

© COPYRIGHT UPM

normal saline solution (0.9% NaCl) beforehand was then inserted into the middle of the inferior turbinate of one nostril (Figure 3.9). The cotton swab was gently rotated 90-180° on the nasal wall. Immediately after the sampling, the swab was placed into a collection tube containing 2ml of normal saline (0.9% NaCl). All sampled was kept in an ice box with temperature between 0-4°C for transferred to the lab. The saline solution containing the sample was then transferred into Eppendorf 1.5ml centrifuge tubes for centrifugation at 500g for 10 minutes. The supernatant was then transferred into another tube and re-centrifuged at 1000g for further 10 minutes to obtain a purer sample. Centrifugation was done within 5 hours after collection. The supernatant was then immediately frozen at -70 to -80°C for later analyses of biomarkers.

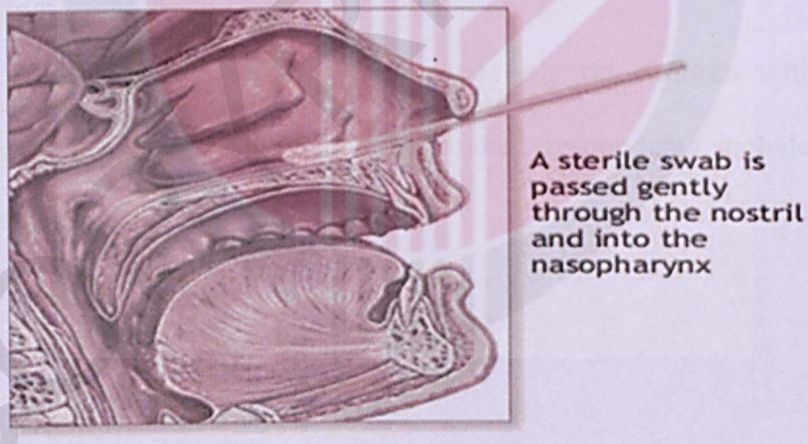


Figure 3.9 Collection of nasal swab

Source: US National Institute of Health Website

© COPYRIGHT UPM

The saline solution prepared beforehand is a mixture of 9.1% distilled water and 0.9% sodium chloride (NaCl) solution. All sample was analysed for the present of ECP using commercial enzyme-linked immunosorbent assay (ELISA) kits for human RNase following manufacturer's manual. Duplication of test was done.

In order to fulfil ethical requirement, the collection of nasal sample was performed by medical assistant supervised by a medical doctor. The medical doctor assisting researcher in the field for nasal swab collection is Dr Titi Rahmawati Hamedon from the Department of Community Medicine, Faculty of Medicine and Health Sciences, Universiti Putra Malaysia

*Nasal swab sampling was done only on children with parents' consent prior to the study. Not all of the studied respondents included for the sampling with only 70 of them participated.

© COPYRIGHT UPM

3.4.5 Analysis of Nasal Swab Sample

All collected specimens were transferred to the lab in ice box of 0-4°C. Within 2 to 5 hours, the specimens must reach the lab for centrifugation procedure (Norback *et al*, 2000). The 2ml sample was transferred into 1.5ml Eppendorf tube for centrifugation using Eppendorf Microcentrifuge 5415C. Sample was first centrifuged at 500g for 10 minute followed by second centrifugation of supernatant at 1000g for another 10 minute. 2 ELISA Kits was used in this analysis to produce duplication of test. The kit used was Cloud-Clone Corp SEB758Hu 96 Tests, Enzyme-linked Immunosorbent Assay Kit for Ribonuclease A3 (RNASE3) for human ECP sample. This analysis method of nasal swab sample was adopted from Renstrom, 2002 and carried out based on standard manual by the manufacturer.

a) Standard solutions were prepared by diluting the standard stock with 0.5mL standard diluent. 7 double dilution series were done with 0.25mL standard diluent in each tube. Each time, 0.25mL was drawn from the previous tubes starting from the stock solution. The eighth tube was a blank with only the standard diluent in it. The final concentrations of the standards were 2.5ng/mL, 1.25ng/mL, 0.625ng/mL, 0.312ng/mL, 0.156ng/mL, 0.078ng/mL, 0.039ng/mL, and 0ng/mL respectively.
b) Detection Reagent A and B were then prepared by diluting with Assay Diluent A and B respectively at working concentration of 1:100.

© COPYRIGHT UPM

c) 600mL of wash solution was also prepared by diluting 20 mL of Wash Solution concentrate (30x) with 580 mL of distilled water. The final concentration of Wash Solution was (1x).

d) After all reagents were prepared, then the assay procedure was carried out.

e) 100µL of standards, blank and sample was added into the wells and covered with plate sealer. This was done for 2 microplate, labelled 1 and 2 respectively for duplication of samples. Each standards, blank and samples well number was recorded. The plates were then incubated at 37°C for 2 hours.

f) After the first incubation, the liquid in each well was removed without washing of well. 100 µL of Deyection Reagent A was added into each well for both plates and were incubated again at 37°C for 1 hour after covering with new plate sealer.

g) The solution was aspirated from the plates and washed with 350µL of 1x Wash Solution to each well, and let it sit for 1 to 2 minutes. The liquid was then removed completely by snapping the plate onto absorbent paper. Washing was done 3 times for each well.

h) Next, 100µL of Detection Reagent B working solution was added into each well and incubated for 30 minutes at 37°C after covering using plate sealer. Then, washing procedure was repeated for 5 times as conducted in (g).

© COPYRIGHT UPM

i) After that, 90µL of TMB substrate solution was added into each well and incubated again at 37°C for 15-25 minutes. The liquid that turned blue indicated the present of ECP in the well.

j) Lastly, reaction was stopped by the addition of 50µL of Stop Solution into each well where the liquid solution turned yellow. Any drop of water or fingerprint was removed from the bottom of the plates and confirmed that there was no bubble on the surface of the liquid.

k) The microplate reader (ChroMate Reader) was run and measurement was conducted at 450nm immediately. Readings were printed out for calculation of concentration.

Test Principle

Each well of the microtiter plate was pre-coated with biotin-conjugated antibody specific to RNASE3. Next, Avidin conjugated to Horseradish Peroxidase (HRP) was added into each well and incubated. Upon addition of TMB substrate, only the wells that contained RNASE3, biotin-conjugated antibody and enzyme conjugated Avidin will changed to blue. Lastly, the enzyme-substrate reaction was terminated by the addition of sulphuric acid solution and the yellow colour changed was measured spectrophotometrically at a wavelength of 450nm. The concentration

© COPYRIGHT UPM

of ECP in the samples were determined by comparing the optical density of the samples to the standard curve.

Calculation of results

The duplicate readings were averaged for each standards, control and samples and subtract the average zero standard optical density. From here, the standard curve was constructed by plotting the mean optical density with known concentration for each standard. Equation of the curve was obtained and the concentrations of samples were calculated by substituting the optical density values into the formula:

$$y = 1153.7x - 204.53$$

*Pre-test for ELISA analysis was conducted on 16 nasal samples collected from the students of the Faculty of Medicine and Health Sciences, roughly 20% of 70 respondents (preschool children) included for nasal sampling prior to respondents sample collection.

© COPYRIGHT UPM

of ECP in the samples were determined by comparing the optical density of the samples to the standard curve.

Calculation of results

The duplicate readings were averaged for each standards, control and samples and subtract the average zero standard optical density. From here, the standard curve was constructed by plotting the mean optical density with known concentration for each standard. Equation of the curve was obtained and the concentrations of samples were calculated by substituting the optical density values into the formula:

$$y = 1153.7x - 204.53$$

*Pre-test for ELISA analysis was conducted on 16 nasal samples collected from the students of the Faculty of Medicine and Health Sciences, roughly 20% of 70 respondents (preschool children) included for nasal sampling prior to respondents sample collection.

© COPYRIGHT UPM

3.5 Data Collection Procedure

Permission was obtained from the related department (Jabatan Kemajuan Masyarakat Negeri Selangor) to carry out this research in preschools in Selangor. The purpose of the study was explained to the preschool teachers and parents to seek their cooperation and approval in conducting the research at their respective preschools with inclusion of their children as respondents.

Then, consent letter and questionnaires were distributed. Parents who approved their children participation in the study had to fill in the questionnaire and returned it to the researcher. Based on inclusive and exclusive criteria outline in the research, respondents were determined. However, the research will based on volunteerism and any respondents who wish not to participate or pull off during the period of research were excluded from the sample population.

Assessment was done prior to approval by the preschools management and parents. Site survey and inspection were done simultaneously with IAQ assessment. Specimen's collection was done at the selected preschools by a medical assistant supervised by a medical doctor. All sample collected were only used for the purpose

© COPYRIGHT UPM

outline in the study objectives. Lastly, data analyses were done before thesis writing.

Data collection workflow was summarised in Figure 3.10.

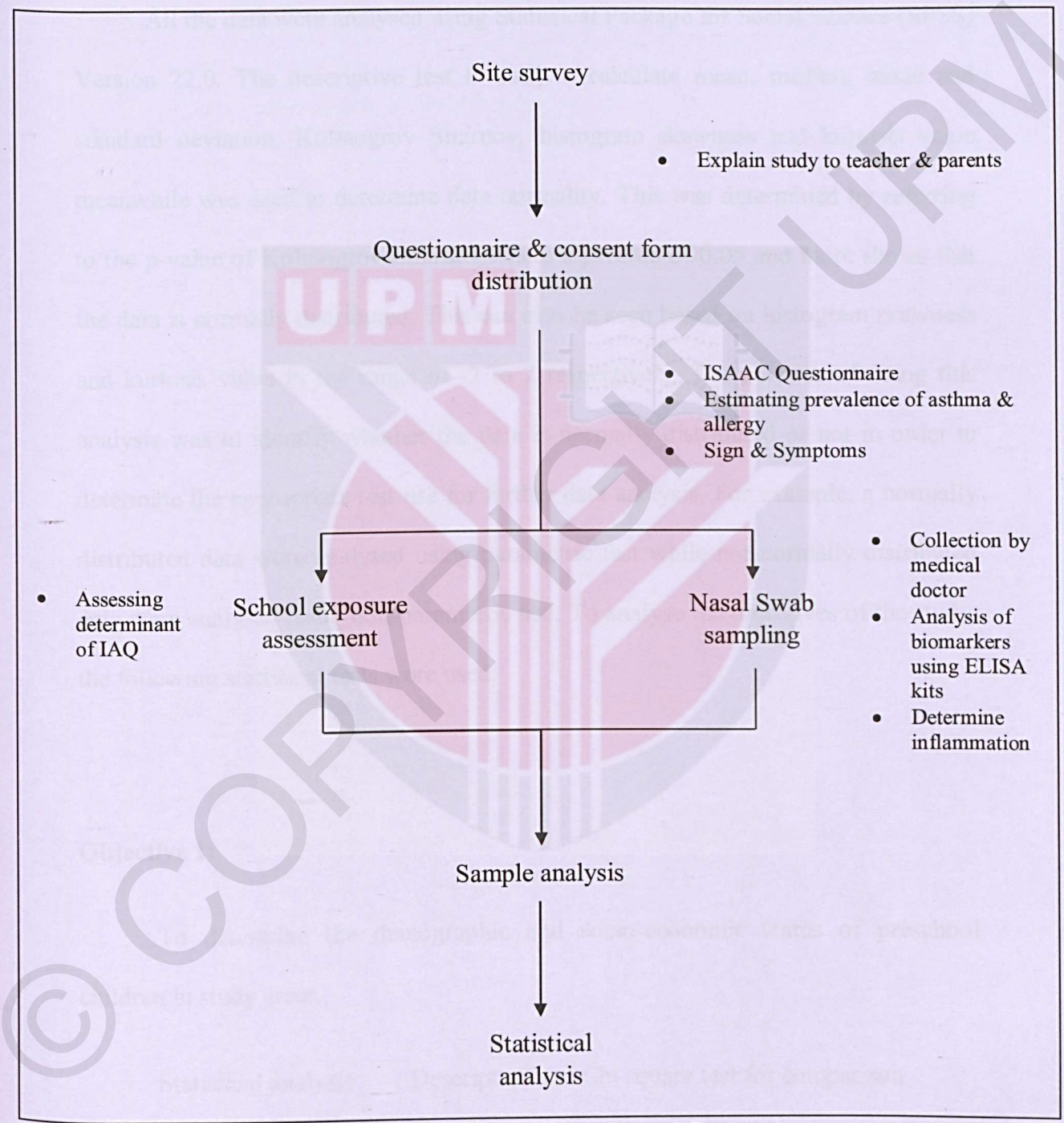


Figure 3.10 Data collection workflow

© COPYRIGHT UPM

3.6 Data Analysis

All the data were analysed using Statistical Package for Social Science (SPSS) Version 22.0. The descriptive test is used to calculate mean, median, mode and standard deviation. Kolmogrov Smirnov, histogram skewness and kurtosis value meanwhile was used to determine data normality. This was determined by referring to the p-value of Kolmogrov Smirnov where a p-value of 0.05 and more shows that the data is normally distributed. This can also be seen based on histogram skewness and kurtosis value in the range of -2 to 2 respectively. The purpose of doing this analysis was to identify whether the data is normally distributed or not in order to determine the appropriate test use for further data analysis. For example, a normally distributed data were analysed using parametric test while not normally distributed data were analysed using non-parametric test. To analyse the objectives of the study, the following statistical tests were used:

Objective 1:

To determine the demographic and socio-economic status of preschool children in study areas.

Statistical analysis : Descriptive and Chi square test for comparison

© COPYRIGHT UPM

Objective 2:

To compare the concentration of indoor air pollutants concentration in preschool between study areas.

Non- parametric test : Mann-Whitney U test

Objective 3:

To compare ECP level among preschool children between study areas.

Parametric : Indepent t test

Objective 4:

To compare reported respiratory symptoms between study areas.

Categorical data : Chi square test

© COPYRIGHT UPM

Objective 5:

To determine the association between indoor air pollutants level with ECP level of the nasal mucosa among preschool children.

Categorical data : Chi square test

Objective 6:

To determine the association between indoor air pollutants with reported respiratory symptoms among preschool children.

Categorical data : Chi square test

Objective 7:

To determine the association between ECP level and reported symptoms among preschool children.

Categorical data : Chi square test

© COPYRIGHT UPM

Objective 8:

To determine association between indoor air pollutants exposure, home exposure and ECP level among preschool children.

Categorical data : Logistics Regression

3.7 Quality Control

3.7.1 Questionnaire

The questionnaire used in this study was pre tested on 10% of the total respondents to ensure reliability and validity of the question set. Result of the pre-test was tested with Cronbach's Alpha test where value of 0.7 and above shows acceptable realiability and validity.

3.7.2 Indoor Air Quality Assessment

To ensure that all measurement data is free from error and bias, all equipment used in the IAQ assessment were first calibrated according to manufacturer's standard.

© COPYRIGHT UPM

3.7.3 Nasal Swab Sample

All equipment for collection of specimen was sterile. Collected samples were transported to the lab for centrifugation in ice box with temperature of 0-4°C within 2 to 5 hours after collection. All supernatant were kept frozen in temperature of -70 to -80°C to preserve the specimen for later analyses.

3.7.4 ELISA test kits for human Eosinophil Cationic Protein (ECP)

All analyses using the test kits were carried out following the Standard Operating Procedure (SOP) or manual provided with the kits suited to number of specimens collected.

© COPYRIGHT UPM

3.8 Ethical Approval

The ethical approval for this study was obtained from Research Ethic Committee of Universiti Putra Malaysia (**Appendix 2**). Approval to conduct research in preschools was obtained from related department and preschool's management (**Appendix 3**). Besides that, consent letter and information (**Appendix 4**) on the research were distributed and briefed to the preschools teacher and parents prior to data collection. This study was done on voluntary basis and researcher ensured that all parties involved understand the purpose of the research. All data collected during the research was kept confidential and used only for research purposes.

© COPYRIGHT UPM

CHAPTER 4

RESULTS

4.1 Socio-demographic data of respondents between study areas

Distribution of respondents' socio-demographic data and socio-economic background between 2 study areas of industrial and suburban of Selangor are shown in Table 1. In industrial area, a total of 41 (77.4%) of 5 years old preschoolers participate in the study with the remaining 22.6% are 6 years of age (N=12). As for suburban area, the age of respondents distributed normally between 5 years old preschoolers (N=23, 51.1%) and 6 years old preschoolers (N=22, 48.9%).

© COPYRIGHT UPM

Table 1: Distribution of Respondent's Demographic and Socio-Economic Background between Study Areas

Variables	Industrial (n=53)	Suburban (n=45)	$\chi 2$ value	p value
	Number (%)			
Age				
5 years old	41 (77.4)	23 (51.1)	7.400	0.007*
6 years old	12 (22.6)	22 (48.9)		
Gender				
Boy	22 (41.5)	12 (26.7)	2.366	0.124
Girl	31 (58.5)	33 (73.3)		
Education Level (Father)				
Primary Education	2 (3.8)	2 (4.8)	0.051	0.975
Secondary Education	36 (69.2)	29 (69.0)		
Higher Education	14 (26.9)	11 (26.2)		
Education Level (Mother)				
Primary Education	1 (1.9)	3 (6.8)	2.119	0.347
Secondary Education	30 (56.6)	27 (61.4)		
Higher Education	22 (41.5)	14 (31.8)		

N=98

*Significant at $p<0.05$

Overall gender distribution in both areas are not equal as more girl preschoolers (N=64) participate in the study as compared to boy preschoolers (N=34). Girl preschoolers made up the 58.5% of preschoolers in the industrial area and boy with the remaining percentage. A large difference was observed in the suburban area with girl preschoolers (73.3%) compared to boy (26.7%).

© COPYRIGHT UPM

The data which was obtained from the distributed questionnaires show that most respondents' father in both areas completed their secondary education (N=65) with a small portion (N=4) has only primary school qualification and the remaining 25 fathers did their higher education. The same pattern was observed with respondents' mother education level. Within the industrial group, 22 mothers have pursued their higher education (41.5%) with secondary education (N=30, 56.6%) and primary education (N=1, 1.9%) respectively.

Statistical analysis using chi-square test was performed to evaluate whether respondents' socio-demographic distribution differ between study areas. The chi-square test was statistically significant only for their age distribution, χ^2 (1, N=98) =7.400, $p<0.01$, with the effect was small, $\phi=0.275$.

Meanwhile, the study also took into account respondents' outdoor residential background to compare with respondents' distribution in both areas. Variables included in this analysis are types of housing area, respondents' house distance from the main road and distance from the nearest factory. The results of the analysis are as tabulated in Table 2.

© COPYRIGHT UPM

Most respondents' in the industrial area live in terrace housing (N=23, 43.4%) and quite a large number living in flat housing (N=16, 30.2). Almost the same pattern was observed for respondents' in the suburban with the highest number living in terrace housing (N=26, 57.8). However, the second highest housing area for suburban respondents is village settlement (N=11, 24.4) instead of flat housing in the industrial area.

Respondents in the industrial area mostly living in a house located near to the main road as suggested by the frequency of respondents living within 100 meters to 500 meters off the main road (N=35, 66.0%) with quite a large number living within 500 meter to 1 kilometer (N=14, 26.4%) barrier. Surprisingly a number of suburban respondents living very near within 100 meter (N=12, 29.3) to the main road. However, a higher number living more than 1 kilometer barrier (N=7, 17.1) when compare to those in the industrial area.

Chi-square test was also performed on the variables whether they have any relation to respondents' distribution between study areas. The test for types of housing area was statistically significant, χ^2 (5, N=98) =15.131, $p<0.05$ with medium effect, $\phi=0.393$. Respondents' house distance from main road was also significantly different between the study areas, χ^2 (3, N=98) =21.978, $p<0.001$ with medium effect,

© COPYRIGHT UPM

ϕ=0.484. Meanwhile, there is no significant different of house distances from factory with distribution between study areas.

Table 2: Distribution of Respondent's Outdoor Residential Background between Study Areas

Variables	Industrial (n=53)	Suburban (n=45)	$\chi 2$ value	p value
	Number (%)			
Types of Housing Area				
Terrace House	23 (43.4)	26 (57.8)	15.131	0.010*
Villa	2 (3.8)	1 (2.2)		
Apartment	7 (13.2)	1 (2.2)		
Flat	16 (30.2)	5 (11.1)		
Village Settlement	3 (5.7)	11 (24.4)		
Other	2 (3.8)	1 (2.2)		
Distance from Main Road				
< 100 meters	0 (0.0)	12 (29.3)	21.978	0.001**
> 100-500 meters	35 (66.0)	15 (36.6)		
> 500-1000 meters	14 (26.4)	7 (17.1)		
> 1000 meters	4 (7.5)	7 (17.1)		
Distance from Factory				
< 500 meters	0 (0.0)	4 (11.4)	6.765	0.080
> 500-1500 meters	11 (20.8)	8 (22.9)		
> 1500-3000 meters	24 (45.3)	12 (34.3)		
> 3000 meters	8 (34.0)	11 (31.4)		

N=98

*Significant at p<0.05; **Significant at p<0.001

© COPYRIGHT UPM

4.2 Home sources for possible exposure to indoor air pollutants

From the questionnaires distributed to respondents' parents, data on possible indoor air pollutants sources at home was obtained and statistical analysis results on the use of mosquitoes coil at home, fuel use for cooking, presence of indoor smoking and pets, carpet usage, mould stain presence on house wall or ceiling and any house painting activity within the past 12 months are tabulated in Table 3.

© COPYRIGHT UPM

Table 3: Indoor Air Pollutants (PM_{10}, VOCs & Microbiological Contaminants) Sources at home among Preschool Children between Study Areas

Variables	Industrial (n=53)	Suburban (n=45)	$\chi 2$ value	p value
	Number (%)			
Mosquitoes Coil				
Yes	33 (62.3)	39 (86.7)	7.435	0.006*
No	20 (37.7)	6 (13.3)		
Fuel for Cooking				
Gas	51 (96.2)	44 (97.8)	0.197	1.000***
Gas and Electric Cooker	2 (3.8)	1 (2.2)		
Indoor Smoking				
Yes	22 (41.5)	22 (48.9)	0.536	0.464
No	31 (58.5)	23 (51.1)		
Pets				
Yes	10 (18.9)	13 (28.9)	1.361	0.243
No	43 (81.1)	32 (71.7)		
Carpet Usage				
Yes	30 (56.6)	21 (46.7)	0.963	0.326
No	23 (43.4)	24 (53.3)		
Painting of House within 12 Months				
Yes	25 (47.2)	24 (53.3)	0.370	0.543
No	28 (52.8)	21 (46.7)		
Mould Stain on Wall/ Ceiling				
Yes	13 (24.5)	6 (13.3)	1.952	0.162
No	40 (75.5)	39 (86.7)		

N=98

*Significant at $p<0.05$

***Fisher exact test for value <5

© COPYRIGHT UPM

Chi-square test results show that only the use of mosquitoes coil at home is statistically significant between the two study areas, χ^2 (1, N=98) =7.435, $p<0.05$ but with small effect, ϕ= -0.275. Large number of respondents in the suburban used mosquitoes coil at home (N=39, 86.7%) with only 6 respondents not using it (13.3%). Meanwhile, almost half the respondents in the industrial area opt against the choice (N=20, 37.7).

Though there is no significant different for presence of indoor smoking at home between the study areas, the large number of respondents living with at least one smoker at home is worrisome with 22 respondents in each area respectively.

In term of carpet usage, more is used at the industrial area (N=30) as compare to home in the suburban area (N=21) but not statistically significant. Within the industrial area itself, larger number of carpet usage at home (N=30, 56.6%) with the remaining 43.4% do not use any carpet (N=23). The choice for carpet usage at homes of suburban respondents has an inverse outcome as compare to industrial area. Larger number of respondents home do not use carpet (N=24, 53.5%) compare to homes which used carpet (N=21, 46.7%).

© COPYRIGHT UPM

4.3 Indoor air quality in preschool between industrial and suburban area of Selangor

Analysis of indoor air pollutants level was done based on IAQ measurement from all the preschools participated in this study and are tabulated in Table 4. The parameters being measured were PM_{10}, VOCs, microbiological contaminants (mould and bacteria), relative humidity (RH) and air temperature. The results were then compared between study areas using Mann-Whitney U statistical analysis as all the data were not normally distributed.

Table 4: Indoor Air Parameters Distribution and Comparison of PM_{10}, VOCs, Microbiological Contaminants, Relative Humidity and Air Temperature between Study Areas

Variables	Industrial (n=53)	Suburban (n=45)	z value	p value
	Median (IQR)			
PM_{10} ($\mu g/m^3$)	126.00 (36.00)	85.00 (224.00)	-1.976	0.048*
VOC (ppm)	0.02 (0.00)	0.02 (0.02)	-0.926	0.355
Mould (CFU/m^3)	361.00 (0.00)	344.00 (95.00)	-2.420	0.016*
Bacteria (CFU/m^3)	275.00 (19.00)	286.00 (65.00)	-1.981	0.048*
Relative Humidity	64.50 (8.20)	64.00 (8.40)	-5.587	0.001**
Temperature	30.1 (0.00)	30.2 (1.20)	-1.422	0.155

N=98

*Significant at $p<0.05$; **Significant at $p<0.001$

IQR=Interquartile

© COPYRIGHT UPM

The level of PM_{10} measurement of preschools in the industrial area (Mean Rank=54.57, N=53) were significantly higher than PM_{10} level in the suburban (Mean Rank=43.53, N=45), U=924.00, z = -1.976, p = 0.048 (2-tailed). Meanwhile, the median readings for PM10 are 126.00μg/m^3 and 85.00μg/m^3 in preschools in the industrial and suburban respectively.

Besides that, Mann-Whitney U statistical test also indicated that there are significant differences for mould and bacteria level between study areas. Mould level (CFU/m^3) in the industrial preschools (Mean Rank=55.70, N=53) is significantly higher than suburban preschools (Mean Rank=42.20, N=45) with U=864.00, z = -2.420, p = 0.016 (2-tailed). Same pattern was observed for bacteria level (CFU/m^3) where it is significantly higher in the industrial preschools (Mean Rank=54.47, N=53) than suburban preschools (Mean Rank=43.53, N=45) with U=924.00, z = -1.981, p = 0.048 (2-tailed).

Lastly, the significant different was observed on RH readings between the two areas. The statistical analysis shows that industrial preschools (Mean Rank=63.85, N=53) are significantly more humid than suburban preschools (Mean Rank=32.60, N=45), U=432.00, z = -5.587, p = 0.001 (2-tailed). The median humidity level in the industrial preschool was 65.5% while in the suburban preschools was 64.0%.

© COPYRIGHT UPM

There are no significant differences observed for VOCs level (ppm) and air temperature level (°C) in both study areas.

Further analysis was done to determine any correlation between different types of indoor air pollutants being measured in this study. Thus, with believe that these pollutants level are related, a statistical analysis was done using Spearman's rho test for non-parametric data.

Table 5 show results for correlation analysis between PM_{10} level ($\mu g/m^3$) with mould and bacteria level (CFU/m^3). Spearman's rho indicated the presence of positive correlation between PM_{10} level ($\mu g/m^3$) with mould level (CFU/m^3), $r = 0.322$, $p<0.001$ (2-tailed), $N=98$ and also PM_{10} level ($\mu g/m^3$) with bacteria level (CFU/m^3), $r = 0.275$, $p<0.05$ (2-tailed), $N=98$. These mean that as PM_{10} level ($\mu g/m^3$) increase, so do mould and bacteria level (CFU/m^3).

Besides that, the correlation between mould level (CFU/m^3) with relative humidity (%) and air temperature (°C) was tested and the results are as tabulated in Table 6. Strong positive correlation was indicated for both test on mould level (CFU/m^3) with relative humidity (%), $r = 0.540$, $p<0.001$ (2-tailed), $N=98$ and mould level (CFU/m^3) with air temperature (°C), $r = 0.767$, $p<0.001$ (2-tailed), $N=98$.

© COPYRIGHT UPM

Table 5: Correlation between different Indoor Air Pollutants; PM_{10} with mould and bacteria level

Variables	PM_{10} Level ($\mu g/m^3$)	
	r value	p value
Mould (CFU/m^3)	0.322	0.001**
Bacteria (CFU/m^3)	0.275	0.006**

**Correlation significant at the 0.01 level (2-tailed); r=Correlation Coefficient

Table 6: Correlation between different Indoor Air Pollutants; mould level with Relative Humidity and Air Temperature

Variables	Mould Level (CFU/m^3)	
	r value	P value
Relative Humidity (%)	0.540	0.001**
Temperature (°C)	0.767	0.001**

**Correlation significant at the 0.01 level (2-tailed); r=Correlation Coefficient

4.4 Factors affecting indoor air pollutants level in preschools

A set of preschool characteristics were observed and recorded during field measurement and interview session. Some of the characteristics such as preschool distance from main road and factory, and the usage of carpet in the classroom were tested for association with level of indoor air pollutants.

© COPYRIGHT UPM

Table 7 contains results for chi-square test statistical analysis performed on pollutants sources and PM_{10} level ($\mu g/m^3$) which were recoded as categorical data prior to the test. The newly recoded PM_{10} data categorized its level into high and low based on the median of all PM_{10} readings in the study areas (Median=126$\mu g/m^3$). Any values fall below the median were considered as low level while values of median and above were categorized as high level.

The chi-square test indicated that there are significant associations of all the three variables with level of PM_{10}. Most respondents (N=43) attending preschools with high level of PM_{10} which were located within 500 meters to 1 kilometer from the main road while there were 15 respondents in preschools with low level of PM_{10} which are more than 1 kilometer away from the main road. Level of PM_{10} is significantly associated with preschools distance from the main road, χ^2 (3, N=98)=50.998, $p<0.001$ with large effect, $\phi = 0.721$.

There is also a significant association between PM_{10} level with school distance from the nearest factory, χ^2 (2, N=98)=8.883, $p<0.05$ with medium effect, $\phi = 0.301$. Lastly, chi-square test indicated that carpet usage in preschools also associated with PM10 level with significant association, χ^2 (1, N=98)=42.129, $p<0.001$ and large effect, $\phi = 0.656$.

© COPYRIGHT UPM

Table 7: Association of Indoor Air Pollutants Sources with PM_{10} Level in Preschools

Variables	High PM_{10} (>126 $\mu g/m^3$)	Low PM_{10} (<126 $\mu g/m^3$)	$\chi 2$ value	p value
	Number (%)			
School Distance from Main Road (km)				
< 100 meters	18 (25.4)	0 (0.0)	50.998	0.001**
> 100-500 meters	10 (14.1)	0 (0.0)		
> 500-1000 meters	43 (60.6)	12 (44.4)		
> 1000 meters	0 (0.0)	15 (55.6)		
School Distance from Factory (km)				
> 1000-3000 meters	17 (23.9)	0 (0.0)	8.883	0.012*
> 3000-5000 meters	36 (50.7)	15 (55.6)		
> 5000 meters	18 (25.4)	12 (44.4)		
Carpet Usage				
Yes	52 (73.2)	0 (0.0)	42.129	0.001**
No	19 (26.8)	27 (100.0)		

*Significant at $p<0.05$; **Significant at $p<0.001$

© COPYRIGHT UPM

There are more respondents attending preschools with high VOCs level (>0.02 ppm) as compare to those attending preschools with low VOC level (<0.02 ppm). There were 7.6% of respondents in high VOCs preschools which were located less than 100 meters from the main road (N=6) while most attending preschools with high VOCs level which are 500 meters to 1 kilometer away from the main road (N=48, 60.8). For preschools with low level of VOCs, there were 63.2% respondents attending classes which were located less than 100 meters from main road (N=12). Chi-square test results as in Table 8, indicates a significant association of VOCs level with the preschool location to the main road, χ^2 (3, N=98)=33.318, p<0.001 and large effect, $\phi = 0.583$.

Besides that, a significant association was observed for preschool distance from the nearest factory with level of VOCs in classroom, χ^2 (2, N=98)=25.585, p<0.001 and large effect, $\phi = 0.511$. Lastly, the chi-square test also indicated a significant association of carpet usage in classroom with VOCs level in preschools, χ^2 (1, N=98)=26.644, p<0.001 with large effect, $\phi = 0.521$.

© COPYRIGHT UPM

Table 8: Association of Indoor Air Pollutants Sources with VOCs Level in Preschools

Variables	High VOC (>0.02 ppm)	Low VOC (<0.02 ppm)	χ2 value	p value
	Number (%)			
School Distance from Main Road (km)				
< 100 meters	6 (7.6)	12 (63.2)	33.318	0.001**
> 100-500 meters	10 (12.7)	0 (0.0)		
> 500-1000 meters	48 (60.8)	7 (36.8)		
> 1000 meters	15 (19.0)	0 (0.0)		
School Distance from Factory (km)				
> 1000-3000 meters	10 (12.7)	7 (36.8)	25.585	0.001**
> 3000-5000 meters	51 (64.6)	0 (0.0)		
> 5000 meters	18 (22.8)	12 (63.2)		
Carpet Usage				
Yes	52 (65.8)	0 (0.0)	26.644	0.001**
No	27 (34.2)	19 (100.0)		

*Significant at $p<0.05$; **Significant at $p<0.001$

4.5 Eosinophil Cationic Protein (ECP) in nasal swab sample

ECP concentration was determined from a subset sample of respondents from both study areas obtained using nasal swab technique with analysis of the swab sample showed 100% positive detection. The assumption of normality was not violated (with neither Shapiro-Wilk statistic was significant) for ECP level distribution across the two study areas. Hence, an independent samples *t* test was

© COPYRIGHT UPM

used to compare ECP level among industrial respondents (N=37) to the ECP level among suburban respondents (N=33).

From the test, it is found that Levene's test was significant thus equal variances are not assumed. The *t* test as tabulated in Table 9 was statistically significant, with the industrial group (Mean=1.37, SD=0.45) some of which are 0.708 higher than the suburban group (Mean=0.66, SD=0.22), *t* (53.44)=8.473, p<0.001 (2-tailed).

Table 9: Eosinophil Cationic Protein (ECP) Sample among Preschool Children and Comparison between Study Areas

Variables	**Industrial (n=37) Number (%)**	**Suburban (n=33) Number (%)**	**t value**	**p value**
	Mean (SD)			
ECP level (ng/mL)	1.37 (0.45)	0.66 (0.22)	8.473	0.001**

N=70

**Significant at p<0.001

© COPYRIGHT UPM

Further analysis was done to determine any association between ECP levels in respondents nasal swab sample with measured pollutants level in the preschools for both study areas as shown in Table 10. For the purpose of this statistical test, the ECP level was recoded into categorical data of high and low level based on it median level for both study areas, 0.87ng/mL. Any values equal or higher than 0.87ng/mL will be categorized as high level and others lower than that as low level. The same was method was use to recode pollutants level into category of high and low level such as mould (Median=361 CFU/m^3), bacteria (Median=275 CFU/m^3), PM_{10} (Median=126 μg /m^3) and VOC (Median=0.02 ppm).

Chi-square test indicated that ECP level was significantly associated with 2 of the pollutants which are PM_{10}, χ^2 (1, N=70)=6.873, $p<0.05$ with medium effect, ϕ = 0.313 and bacteria, χ^2 (1, N=70)=15.306, $p<0.001$ with medium effect, ϕ = 0.468. There is no significant association with VOC and mould.

© COPYRIGHT UPM

Table 10: Association between Eosinophil Cationic Protein (ECP) Concentrations found in Preschool Children's Nasal Sample with Level of Indoor Air Pollutants

Variables	High ECP Level (>0.86 ng/mL)	Low ECP Level (<0.86 ng/mL)	χ^2 value	p value	PR (95% CI)
	Number (%)				
PM_{10} ($\mu g/m^3$)					
High (>126)	32 (91.4)	23 (65.7)	6.873	0.009*	5.565 (1.409-21.987)
Low (<126)	3 (8.6)	12 (34.3)			
VOC (ppm)					
High (>0.02)	28 (80.0)	23 (65.7)	1.806	0.179	2.087 (0.707-6.165)
Low (<0.02)	7 (20.0)	12 (34.3)			
Mould (CFU/m^3)					
High (>361)	22 (62.9)	17 (48.6)	1.447	0.229	1.792 (0.690-4.650)
Low (<361)	13 (37.1)	18 (51.4)			
Bacteria (CFU/m^3)					
High (>275)	32 (91.4)	17 (48.6)	15.306	0.001**	11.294 (2.909-43.847)
Low (<275)	3 (8.6)	18 (51.4)			

N=70

*Significant at $p<0.05$; **Significant at $p<0.001$

© COPYRIGHT UPM

4.6 Prevalence of parental reported respiratory symptoms among preschool children

Table 11 shows the respondents' parental reported respiratory symptoms. The symptoms studied were cough, wheezing, phlegm, chest tightness, runny nose, blocked nose and sneezing with data obtained from distributed questionnaire constructed based on standard questionnaire by American Thoracic Society (ATS) and International Study of Asthma and Allergy in Childhood (ISAAC). Symptoms such as runny nose, blocked nose and sneezing were included to complement the study subset on upper airway inflammation by determining ECP production in nasal region.

Respondents from industrial (N=53) area reported a larger number for each of the symptoms as compared to suburban respondents (N=45). For cough, 18 respondents were reported of having the symptom (34.0%) while the remaining 66.0% (N=35) of respondents in the industrial area were not. In the suburban, only 1(2.2%) respondent was reported by the parent to have the symptom. The 18 (34.0%) respondents in the industrial area were parental reported of having phlegm symptom with a lower number reported in the suburban (N=1, 2.2%). The same pattern of reporting for the symptoms of runny nose and blocked nose in both study area with 32 respondents (60.4%) in the industrial area have both symptoms while 21

© COPYRIGHT UPM

respondents (39.6%) do not. For the suburban group, only 2 respondents (4.4%) reported to experience both symptoms of runny nose and blocked nose with the remaining 43 respondents (95.6%) do not. Lastly, 33 respondents (62.3%) in the industrial area were parental reported of having the symptom of sneezing while 20 respondents (37.7%) do not. A smaller number of parental reported sneezing (N=2, 4.4%) in the suburban with the remaining respondents (N=43, 95.6%) do not experience the symptom.

Statistical analysis was done using chi-square test to compare parental reported symptoms pattern between the 2 study areas. Based on the test, significant differences were found in 5 of the reported symptoms between the 2 study areas.

© COPYRIGHT UPM

Table 11: Prevalence of Reported Respiratory Symptoms between Study Areas

Variables	Industrial (n=53)	Suburban (n=45)	χ^2 value	p value	PR	95% CI
	Number (%)					
Cough						
Yes	18 (34.0)	1 (2.2)	15.687	0.001**	22.629	2.878-
No	35 (66.0)	44 (97.8)				177.902
Wheezing						
Yes	7 (13.2)	2 (4.4)	2.241	0.173***	3.272	0.644-
No	46 (86.8)	43 (95.6)				16.624
Phlegm						
Yes	18 (34.0)	1 (2.2)	15.687	0.001**	22.629	2.878-
No	35 (66.0)	44 (97.8)				177.902
Chest Tightness						
Yes	7 (13.2)	1 (2.2)	3.917	0.066***	6.696	0.791-
No	46 (86.8)	44 (97.8)				56.663
Runny Nose						
Yes	32 (60.4)	2 (4.4)	33.604	0.001**	32.762	7.159-
No	21 (39.6)	43 (95.6)				149.925
Blocked Nose						
Yes	32 (60.4)	2 (4.4)	33.604	0.001**	32.762	7.159-
No	21 (39.6)	43 (95.6)				149.925
Sneezing						
Yes	33 (62.3)	2 (4.4)	35.437	0.001**	35.475	7.738-
No	20 (37.7)	43 (95.6)				162.635

N=98

*Significant at $p<0.05$; **Significant at $p<0.001$

***Fisher exact test for value <5

PR=Prevalence Ratio/ Odds Ratio

©COPYRIGHT UPM

Reported cough symptom was significantly difference, χ^2 (1, N=98)=15.687, p<0.001 with medium effect, ϕ = 0.400 between industrial and suburban with prevalence ratio, PR = 22.629 (95% CI=2.878-177.902). Phlegm reporting was significantly difference, χ^2 (1, N=98)=15.687, p<0.001 with medium effect, ϕ = 0.400 between the 2 study areas with prevalence ratio, PR=22.629 (95% CI=2.878-177.902). Chi-square test also indicated that the symptoms of runny nose and blocked nose are both statistically significant, χ^2 (1, N=98)=33.604, p<0.001, (PR=32.762, 95% CI=7.159-149.925) with large effect, ϕ = 0.586 which is also the same for sneezing symptoms, χ^2 (1, N=98)=35.437, p<0.001(PR=35.475, 95% CI=7.738-162.635) with large effect, ϕ = 0.601.

Further analysis was done to determine any association between the parental reported symptoms with exposure to pollutants, based on high and low level of exposure. Table 12 contains results of statistical analysis between reported symptoms and PM_{10} level; Table 13 shows results on VOC level and association with reported symptoms; Table 14 and Table 15 show reported symptoms association with the pollutants mould and bacteria level respectively.

A chi-square test was used to evaluate whether pollutants level is related to the prevalence of parental reported symptoms among the preschool children in both study areas. In Table 12, the statistical analysis was significant for the association of

© COPYRIGHT UPM

PM_{10} with 5 of the symptoms which are cough, χ^2 (1, N=98)=5.866, p<0.05; phlegm, χ^2 (1, N=98)=8.963, p<0.05; runny nose and blocked nose, χ^2 (1, N=98)=12.247, p<0.001; and sneezing, χ^2 (1, N=98)=13.006, p<0.001.

Meanwhile, statistical result tabulated in Table 13 indicates that only the symptom of phlegm is statistically significant, χ^2 (1, N=98)=5.669, p<0.05 with VOCs level. The rest of the symptoms are not significantly associated with VOCs level in the preschools.

For association of mould level in preschools with the reported symptoms as in Table 14, chi-square test indicate that mould level is statistically significant with 4 of the reported symptoms in phlegm, χ^2 (1, N=98)=7.551, p<0.05; and runny nose, χ^2 (1, N=98)=6.406, p<0.05. Significant association was also observed for the symptoms of blocked nose, χ^2 (1, N=98)=8.754, p<0.05; and lastly sneezing, χ^2 (1, N=98)=7.294, p<0.05.

Table 15 shows only 3 of the reported symptoms have significant association with level of bacteria in preschool based on statistical analysis using chi-square test. Bacteria level is statistically significant with the symptoms of runny nose, χ^2 (1,

© COPYRIGHT UPM

N=98)=7.473, p<0.05; blocked nose, χ^2 (1, N=98)=7.473, p<0.05; and also symptoms of sneezing, χ^2 (1, N=98)=7.985, p<0.05.

© COPYRIGHT UPM

Table 12: Association of PM_{10} Level with Prevalence of Reported Respiratory Symptoms among Preschool Children

Variables	High PM_{10} (>126 $\mu g/m^3$)	Low PM_{10} (<126 $\mu g/m^3$)	χ^2 value	p value	PR	95% CI
	Number (%)					
Cough						
Yes	18 (25.4)	1 (3.7)	5.866	0.015*	8.830	1.117-
No	53 (74.6)	26 (96.3)				69.812
Wheezing						
Yes	7 (9.9)	2 (7.4)	0.141	1.000***	1.367	0.266-
No	64 (90.1)	25 (92.6)				7.034
Phlegm						
Yes	19 (26.8)	0 (0.0)	8.963	0.003*	-	-
No	52 (73.2)	27 (100.0)				
Chest Tightness						
Yes	8 (11.3)	0 (0.0)	3.313	0.102***	-	-
No	63 (88.7)	27 (100.0)				
Runny Nose						
Yes	32 (45.1)	2 (7.4)	12.247	0.001**	10.256	2.256-
No	39 (54.9)	25 (92.6)				46.625
Blocked Nose						
Yes	32 (45.1)	2 (7.4)	12.247	0.001**	10.256	2.256-
No	39 (54.9)	25 (92.6)				46.625
Sneezing						
Yes	33 (46.5)	2 (7.4)	13.006	0.001**	10.855	2.389-
No	38 (53.5)	25 (92.6)				49.331

N=98

*Significant at $p<0.05$; **Significant at $p<0.001$;***Fisher exact test for value <5

PR=Prevalence Ratio/ Odds Ratio

- Prevalence Ratio can only be calculated for 2x2 table

© COPYRIGHT UPM

Table 13: Association of VOC Level with Prevalence of Reported Respiratory Symptoms among Preschool Children

Variables	High VOC (>0.02 ppm)	Low VOC (<0.02 ppm)	χ^2 value	p value	PR	95% CI
	Number (%)					
Cough						
Yes	16 (20.3)	3 (15.8)	0.195	1.000***	1.354	0.351-5.223
No	63 (79.7)	16 (84.2)				
Wheezing						
Yes	8 (10.1)	1 (5.3)	0.434	1.000***	2.028	0.238-17.277
No	71 (89.9)	18 (94.7)				
Phlegm						
Yes	19 (24.1)	0 (0.0)	5.669	0.020***	-	-
No	60 (75.9)	19 (100.0)				
Chest Tightness						
Yes	7 (8.9)	1 (5.3)	0.264	1.000***	1.750	0.202-15.144
No	72 (91.1)	18 (94.7)				
Runny Nose						
Yes	29 (36.7)	5 (26.3)	0.730	0.393	1.624	0.530-4.972
No	50 (63.3)	14 (73.7)				
Blocked Nose						
Yes	29 (36.7)	5 (26.3)	0.730	0.393	1.624	0.530-4.972
No	50 (63.3)	14 (73.7)				
Sneezing						
Yes	30 (38.0)	5 (26.3)	0.907	0.341	1.714	0.561-5.242
No	49 (62.0)	14 (73.7)				

N=98

*Significant at $p<0.05$; **Significant at $p<0.001$;***Fisher exact test for value <5

PR=Prevalence Ratio/ Odds Ratio

- Prevalence Ratio can only be calculated for 2x2 table

© COPYRIGHT UPM

Table 14: Association of Mould Level with Prevalence of Reported Respiratory Symptoms among Preschool Children

Variables	High Mould (>361 CFU/m^3)	Low Mould (<361 CFU/m^3)	χ^2 value	p value	PR	95% CI
	Number (%)					
Cough						
Yes	11 (20.0)	8 (18.6)	0.030	0.862	1.094	0.397-3.012
No	44 (80.0)	35 (81.4)				
Wheezing						
Yes	5 (9.1)	4 (9.3)	0.001	1.000***	0.975	0.245-3.875
No	50 (90.9)	39 (90.7)				
Phlegm						
Yes	16 (29.1)	3 (7.0)	7.551	0.006*	5.470	1.476-20.265
No	39 (70.9)	40 (93.0)				
Chest Tightness						
Yes	5 (9.1)	3 (7.0)	0.144	1.000***	1.333	0.300-5.919
No	50 (90.9)	40 (93.0)				
Runny Nose						
Yes	25 (45.5)	9 (20.9)	6.406	0.011*	3.148	1.272-7.793
No	30 (54.5)	34 (79.1)				
Blocked Nose						
Yes	26 (47.3)	8 (18.6)	8.754	0.003*	3.922	1.543-9.969
No	29 (52.7)	35 (81.4)				
Sneezing						
Yes	26 (47.3)	9 (20.9)	7.294	0.007*	3.387	1.369-8.377
No	29 (52.7)	34 (79.1)				

N=98

*Significant at $p<0.05$; **Significant at $p<0.001$

***Fisher exact test for value <5

PR=Prevalence Ratio/ Odds Ratio

© COPYRIGHT UPM

Table 15: Association of Bacteria Level with Prevalence of Reported Respiratory Symptoms among Preschool Children

Variables	High Bacteria (>275 CFU/m^3)	Low Bacteria (<275 CFU/m^3)	χ^2 value	p value	PR	95% CI
	Number (%)					
Cough						
Yes	18 (23.4)	1 (4.8)	3.658	0.066***	6.102	0.765-
No	59 (76.6)	20 (95.2)				48.673
Wheezing						
Yes	7 (9.1)	2 (9.5)	0.004	1.000***	0.950	0.182-
No	70 (90.9)	19 (90.5)				4.953
Phlegm						
Yes	18 (23.4)	1 (4.8)	3.658	0.066***	6.102	0.765-
No	59 (76.6)	20 (95.2)				48.673
Chest Tightness						
Yes	7 (9.1)	1 (4.8)	0.412	1.000***	2.000	0.232-
No	70 (90.9)	20 (95.2)				17.228
Runny Nose						
Yes	32 (41.6)	2 (9.5)	7.473	0.006*	6.756	1.469-
No	45 (58.4)	19 (90.5)				31.070
Blocked Nose						
Yes	32 (41.6)	2 (9.5)	7.473	0.006*	6.756	1.469-
No	45 (58.4)	19 (90.5)				31.070
Sneezing						
Yes	33 (42.9)	2 (9.5)	7.985	0.005*	7.125	1.550-
No	44 (57.1)	19 (90.5)				32.751

N=98

*Significant at $p<0.05$; **Significant at $p<0.001$

***Fisher exact test for value <5

PR=Prevalence Ratio/ Odds Ratio

© COPYRIGHT UPM

Again, in order to complement the subset of the study to determine the occurrence of upper airway inflammation by assessing ECP level, symptoms related to the upper respiratory airway were tested for association with ECP level found in nasal swab.

Table 16 shows the result of the test where ECP level is statistically significant with all the upper respiratory airway symptoms with phlegm, χ^2 (1, N=98)=6.437, $p<0.05$ with medium effect, $\phi = 0.303$; runny nose, χ^2 (1, N=98)=10.516, $p<0.001$ with medium effect, $\phi = 0.388$; blocked nose, χ^2 (1, N=98)=9.130, $p<0.05$ with medium effect, $\phi = 0.361$; and sneezing, χ^2 (1, N=98)=10.516, $p<0.001$ with medium effect, $\phi = 0.388$.

© COPYRIGHT UPM

Table 16: Association of Reported Respiratory Symptoms with Eosinophil Cationic Protein (ECP) Concentrations among Preschool Children

Variables	High ECP (>0.86 ng/mL)	Low ECP (<0.86 ng/mL)	χ^2 value	p value	PR	95% CI
	Number (%)					
Phlegm						
Yes	10 (28.6)	2 (5.7)	6.437	0.011*	6.600	1.326-32.843
No	25 (71.4)	33 (94.3)				
Runny Nose						
Yes	19 (54.3)	6 (17.1)	10.516	0.001**	5.740	1.906-17.282
No	16 (45.7)	29 (82.9)				
Blocked Nose						
Yes	18 (51.4)	6 (17.1)	9.130	0.003*	5.118	1.702-15.389
No	17 (48.6)	29 (82.9)				
Sneezing						
Yes	19 (54.3)	6 (17.1)	10.516	0.001**	5.740	1.906-17.282
No	16 (45.7)	29 (82.9)				

N=70

*Significant at $p<0.05$; **Significant at $p<0.001$

PR=Prevalence Ratio/ Odds Ratio

© COPYRIGHT UPM

4.7 Factor influencing Eosinophil Cationic Protein (ECP) concentration among preschool children after controlling confounders

Logistic regression was performed in order to determine the factors which have significant association with ECP level after controlling the confounders. Table 17 shows main variables; PM_{10}, VOCs, mould and bacteria with cofactors such as indoor smoking, mosquito's coils usage and others.

Result from the test found that VOCs is the only statistically significant regression, $\beta=42.596$, $p<0.05$ (PR=6.410, 95% CI=1.268 – 32.394 with ECP level. Based on the prevalence ratio, respondents who are exposed to VOCs are six times more likely to experience upper airway inflammation, in this case determine by the presence of ECP in the nasal sample.

© COPYRIGHT UPM

Table 17: Logistic Regression for Association between Indoor PM_{10}, VOCs, Mould and Bacteria with ECP Concentration after Controlling the Confounders

Independent Variables	β	S.E.	p value	PR	95% CI
Constant	-6.221	3.433	0.070	0.002	
PM_{10}	-19.649	19865.088	0.999	0.000	-
VOCs	1.858	0.827	0.025*	6.410	1.268–32.394
Mould	-19.866	12562.005	0.999	0.000	-
Bacteria	42.596	23503.740	0.999	3.15E+18	-
Distance house from road	0.453	0.488	0.354	1.573	0.604–4.096
Distance house from factory	0.057	0.422	0.898	1.058	0.445–2.518
Indoor smoking	0.181	0.764	0.813	1.198	0.268–5.360
Mosquito's coils	-0.480	0.847	0.570	0.619	0.118–3.252
Carpet usage	0.090	0.750	0.904	1.904	0.252–4.761
House painting within 12 months	-0.687	0.836	0.412	0.503	0.098–2.593

N=98

*Significant at $p<0.05$

95% CI= 95% Confidence Interval; B= Regression Coefficient; S.E= Standard Error

Nagelkerke R Square= 0.532

© COPYRIGHT UPM

CHAPTER 5

DISCUSSION, CONCLUSION AND RECOMMENDATION

5.1 Discussion

5.1.1 Background of Respondents

This study began in the end of January 2014 and finished in early May of the same year. The study was carried out with the objective to study the association between exposure to indoor air pollutants and upper airway inflammation among Malay preschool children in 4 areas in Selangor. The schooling hour of the preschools was 8.00 a.m. till 12.00 p.m.

© COPYRIGHT UPM

In the early stage, a total of 238 questionnaires were distributed to 7 preschools in the 2 study areas. After exclusion of respondents who do not fulfilled the criteria and by taking account unresponsive respondents, only 98 preschools students were included in the study; a number exceeded the minimum required sample size of 84. There were 34 boy preschooler from the total number of respondents with the remaining 64 respondents were girl preschooler. Out of 98 included respondents, 71.4% was given parents' consent for nasal sample collection while the remaining 29.6% only agreed to fill in the questionnaires. The situation may attribute to lack of understanding about the health issue being raised and importance of the study among parents. Thus, low response rate may introduce selection bias in the study. Selection process before the inclusion of respondents has been done to reduce bias at later stage of data analysis.

In term of respondents' age distribution, there was significant different (χ^2=7.400, $p<0.05$) between study areas of industrial and suburban of Selangor. However, most of the respondents were 5 years of age (N=64) with 6 years old respondents were only 34 preschoolers.

Other socio-demographic information such as parents' education and type of housing areas were also included in the questionnaire. Most of the parents regardless of study areas at least had attended secondary education (N=65) with roughly a

© COPYRIGHT UPM

quarter of the respondents' parents had attended higher education (N=25). In between areas, parents' education level was not statistically significant. Thus, the study suggested that educational background was no more predetermined by residential distribution. However, there was a significant different between study areas in term of type of housing area. Respondents' in the suburban mostly living in village settlement although terrace house was also a common dwelling. In the industrial setting meanwhile, respondents' were living mostly in and apartment and flat with terrace house still a common dwelling even in this area.

5.1.2 Comparison of Indoor Air Pollutants Level in Preschools between Study Areas of Industrial and Suburban

Measurement of indoor air quality was done in every of the 7 preschools included in this study. The parameters being measured were level of PM_{10}, VOCs, mould, bacteria, RH and temperature. All measurements were done for 4 hours of schooling period to represent the environmental exposure of the children during class except for mould and bacteria measurement. Mould and bacteria were measured based on volume air sampled (500L) and were done twice, one in the morning at 9.00 a.m. while another towards the end of class at 10.30 a.m.

© COPYRIGHT UPM

The results from this study indicate that indoor air pollutants may affect respiratory health and biomarkers in nasal mucosa. This can be seen with the high concentration indoors pollutant such PM_{10} with median readings of 126µg/m^3, total mould (Median=361CFU/ m^3) and total bacteria (Median=275CFU/ m^3) present in preschools indoor air (Table 4). However, the PM_{10} level in preschools is within the permissible limit (150µg/m^3) suggested by the 24 hour exposure Recommended Malaysia Ambient Air Quality Guidelines (DOE, 1998) and EPA 24 hour PM_{10} guideline (USEPA, 2012b). There were significant different of PM_{10}, mould, bacteria and RH level between study areas (Table 4). Level of PM_{10}, mould and bacteria were significantly higher in the industrial preschools as compared to suburban preschools. However, RH in the suburban was higher than industrial area.

High level of pollutants indoor in the preschools may be contributed by the surrounding factors such as preschools distance to main road and to factory and also the used of carpet inside preschool's building. Statistical analyses were done on PM_{10} and VOCs level with the above mentioned factors and found that there were significant associations. Based on Table 7, most of the preschools with high level of PM10 are located near to the main road with distance less than 500m and 100m. This was also similar in terms of carpet usage where high used of carpet in schools contribute to high indoor PM_{10} readings. The same patterns were observed for VOCs readings with significant association with the three factors.

© COPYRIGHT UPM

The high levels of pollutants indoor suggest that outdoor environmental factors also play important roles in distribution of pollutants between study areas. USEPA (2013) stated that indoor air pollutants such as PM_{10} and VOCs may come from sources such as carpet, house paints, outdoor souces like combustion activities from the road, and many more. Previous studies by Nazariah *et al* (2013) also suggested that local environment factors may influenced pollutants readings and contributed to significant different between study areas. This is further supported by Ismail *et al* (2010) that surrounding human activities influenced the differences of indoor concentrations of pollutants between selected schools in Terengganu. In addition, each pollutant may have positive effect towards each other as shown in previous study that focus on reactive indoor chemistry, with possible reaction between ozone NO2, and VOCs and other biologically potent compound (Wolkoff *et al*, 1997). Furthermore, statistical analysis performed to determine any correlation between pollutants level in study areas showed that PM_{10} level is significantly correlate with positive relationship with mould ($r = 0.322$, $p<0.01$) and bacteria ($r = 0.275$, $p<0.05$) level (Table 5). Study has shown that mould and bacteria have positive relationship with settled dust (Wickman *et al*, 1999).

© COPYRIGHT UPM

5.1.3 Level of ECP among Respondents in Industrial and Suburban Preschools

ECP concentration was determined in the nasal swab sample from 70 respondents with parents' approval using ELISA Kit. From the lab analyses, ECP level in nasal mucosa was detected in all of the respondents (N=70, Table 9) where count is higher in the industrial area (N=37) as compare to the suburban (N=30). This may be due to a higher level of indoor air pollutants presence in the industrial area as supported by results tabulated in Table 4. Based on Chi-square test, PM_{10} and bacteria level were significantly associated with ECP level in the nasal mucosa. Respondents exposed to high level of PM_{10} were 5 times more likely to have higher ECP as compared to those exposed to low PM_{10} (PR=5.565, 95% CI=1.409-21.987). Respondents were 11 times more likely to have a higher ECP level if they were exposed to high level of bacteria compared to those exposed at a lower bacteria's level. Study by Norback *et al* (2000) found that particulate pollutants and indoor concentrations of various types of gaseous pollutants had significant relationships with nasal effects. Another study also found that exposure to high level of formaldehyde; which one of VOCs caused concentration of eosinophilic granulocytes and albumin in NAL to increase (Pazdrak *et al*, 1993). This also supported the result from logistic regression model which found that VOCs (B=42.596, $p<0.05$, PR=6.410, 95% CI=1.268 – 32.394) were the main contributing factor in this study after controlling confounders. This suggests possible

© COPYRIGHT UPM

inflammatory and allergic mechanism where exposure to VOCs increases the risk at about 6 times higher. In this study, VOCs level in preschools may be contributed to the high used of carpet as tabulated in Table 8 which was significantly associated. However, the possible effects of mould on ECP level should be neglected completely as findings in Norback *et al* (2000) found there were significant association between the presence of *Aspergillus* spp.; a type of mould with the increased of ECP level in NAL.

ECP has been known as the best standardized marker protein for eosinophil which can be measured in blood and local body fluid (Klimek and Rasp, 1999). However, present of ECP in nasal sample is normally associated with allergic rhinitis but not restricted to; while there is possible production during acute upper airway inflammation in non-allergic rhinitis cases (Chawes, 2011). Nevertheless, this study only exclude respondent based on known or diagnosed respiratory illnesses. The findings in this study may be camouflaged by underreported or undertreated allergic rhinitis among the respondents.

© COPYRIGHT UPM

5.1.4 Reported Respiratory Symptoms among Respondents between Study Areas

Inflammation of the upper airway in this study has been categorized by the presence of nasal symptoms such as runny nose, blocked nose, phlegm and sneezing. Consistent association of indoor air pollutants primarily PM_{10}, total mould, total bacteria and VOCs with prevalence of reported symptoms (Table 12-15) can be observed. . In both study areas, consistent reporting of the same nasal symptoms can be observed although prevalence is higher in the industrial area. The symptoms of phlegm, runny nose, blocked nose and sneezing were significantly associated with level of PM_{10}, mould and bacteria. This is supported by Norback et al 2000 where they found significant relationship between nasal effects and indoor concentration of various types of gaseous and particulate pollutants. They even found the relationship where level of formaldehyde, NO_2, and presence of mould affects biomarkers in nasal lavage. Association of these symptoms was found significant with ECP level in the nasal mucosa of the respondents (Table 16). The relation between ECP levels in nasal lavage was also found significant with nasal obstruction (blocked nose) in study by Norback *et al* 2000.

Other symptom reported such as cough is significantly associated with PM_{10} ($p<0.05$, PR=8.830, 95% CI=1.117-69.812) which also reported in study by Nazariah

© COPYRIGHT UPM

et al, 2013. However, the result may be affected by recall bias on symptoms reporting although clinical sign unlikely to be affected (Norback *et al*, 2000).

5.1.5 Possible Sources of Indoor Air Pollutants in Preschool and at Home

From the preschool environment and homes environment information obtained from the questionnaire, there were many possible sources of exposure to indoor air pollutants. This study found that one of the most prominent factors is distance of preschool from road which is statistically significant in relation to PM_{10}, χ^2 (3, N=98)=50.998, $p<0.001$ with large effect, $\phi = 0.721$ and VOCs, χ^2 (3, N=98)=33.318, $p<0.001$ and large effect, $\phi = 0.583$; level in classroom. Rather than building associated sources of emission, indoor and pollutants level also attributed to outdoor concentration of pollutants and their migration patterns indoor (Flynn *et al*, 2000). Students attending schools located near to heavy traffic are at higher risk of respiratory problems as compare to those with large distance from busy roads. Motor vehicles was found as major contributor of toxic air pollutant with emission of PM_{10} from brake and re-entrained road dust are common (SCAQMD, 2005). Studies in California also found that attending school within close proximity to high traffic road and exposure to associated pollutants may lead to adverse health effects (SCAQMD, 2005). In addition, factory combustion also identified as one of the main producers of

© COPYRIGHT UPM

particulates where it can contribute to higher indoor pollutants level, especially in school or home within close proximity (Flynn *et al*, 2000, Janssen, 1999).

Other than that, this study found that a large usage of mosquito's coil at home (N=72, 73.5%) may be a confounding factor to respiratory symptoms reported and level of ECP in nasal mucosa because Liu W. *et al* (2003) found that it pose potential threats to health because some emits formaldehyde while the coil also emits particulate. Other sources of exposure at homes and in school that cannot be ignore though may not be significant in this study are second-hand tobacco smoke, spraying of insecticides, accumulation of pollutants in carpets, and others (Reigart *et al*, 2001).

Findings from this research have been valuable but further research to study mix exposure in both home and school should be carried out. Many similar studies in overseas can be referred to for extra understanding of this study such as Norback *et al* (2000) and Walinder *et al* (1998). However, there are few differences that need to be pointed out between this study and the study by Norback *et al* (2000). Norback et al conducted their study in a primary schools setting in Uppsala, Sweden. Norback *et al* chose the respondents among working personnel in the schools instead of preschool children as conducted in this study. The sample collection technique used by Norback *et al* was nasal lavage with more biomarkers being measured which

© COPYRIGHT UPM

included ECP, lysozyme, albumin and myeloperoxidase (MPO). They did measurement for CO_2 and NO_2 which did not included in this study. In addition, Norback *et al* also did the identification of bacteria and mould species whereas in this study both parameters were measured for colony forming alone. The findings from the study in Uppsala concluded that the pollutants found in the indoor schools environment have an inflammatory and secretory response of the upper airway which was similar to this study. The study done by Walinder *et al* (1998) was also conducted on adults (school personnel) but measured the significance of air exchange rate and ventilation types with effects to upper airway.

5.1.6 Limitations

There are few limitations to this study where sample size was small which may differed in statistical significant if it was done with a larger sample size. This is very true when it comes to ECP sampling where not all respondents are included due to parents disapproval which may be due to lack of understanding towards the purpose of the research and also may due to protection of certain concerns. Besides that, the population samples are only of Malay ethnicity and children of age 5 to 6 years old which means that the population characteristics are specific. Thus, results of the study cannot be generalized for other populations. Information bias may have occur in term of recall bias and measurement bias where parents have trouble to

© COPYRIGHT UPM

recall previous exposure and having problems to understand the question asked in the questionnaire.

In term of indoor air assessment, a limited or no standards specifically established for domestic buildings and houses in Malaysian setting limit the researcher in the case where we need to determine the severity of exposure at a certain level. Most of the standards for exposure to indoor air pollutants were designed for occupational setting. Ambient air guidelines available may not be a suitable measure for this study as measurements are based on 24 hour exposure whereas the preschool duration is only 4 hour.

Lastly, due to limited time to complete the study, researcher was unable to find more time to carry out extra assessment on other sources for exposure. With more time, the study should be able to evaluate also exposure at homes supported with IAQ assessment measurement. However in this study, home exposure to indoor air pollutants were evaluated based on reporting in the questionnaire.

© COPYRIGHT UPM

5.2 Conclusion

In conclusion, this study suggest that indoor air pollutants in school especially VOCs are most likely to have an impact on the inflammatory and secretory response of the nasal mucosa but, the definite causal agent cannot be elucidated as the study was done for mixed exposure with neglecting the effects of other pollutants such as PM_{10} and microbiological contaminants. Symptoms of upper airway inflammation and other respiratory symptoms were consistently observed in the present of exposure to pollutants. In addition, ECP in nasal sample is consistent with present of some of the nasal symptoms suggest that ECP can be useful in evaluating upper airway inflammation in healthy person while the nasal swab method prove to be a reliable technique in term of ECP reproducibility and also suitable for use in small children for non-invasive effect.

Poor IAQ readings also called for the need to improve indoor environment and exposure prevention in preschools. This is supported with results of high prevalence of reported respiratory symptoms among respondents with children in poor indoor environment are at higher risk. There is no clear middle line to separate the difference of exposure between study areas. In addition, ECP was positive in all of the included respondents' samples suggesting that suburban children are also at risk of upper airway inflammation.

© COPYRIGHT UPM

From the results for the statistical analysis, the following hypotheses were answered:

1. Indoor air pollutants concentrations were significantly higher in the industrial area as compared to the suburban.
2. ECP level among preschool children in the industrial areas was significantly higher compared to suburban.
3. There is a significantly higher number of reported respiratory symptoms in the industrial areas compared to suburban.
4. There is a significant association between indoor air pollutants level with ECP level of the nasal mucosa among preschool children.
5. There is a significant association between indoor air pollutants with reported respiratory symptoms among preschool children.
6. There is a significant association between ECP level and reported symptoms among preschool children.
7. VOCs were the main factor that influenced the ECP level in nasal mucosa among study respondents after controlling confounders.

© COPYRIGHT UPM

5.3 Recommendations

It is recommended that guidelines on IAQ management in domestic building and preschool in particular should be developed by the related authority or ministry with consultation with several agencies. The standard should outline for indoor air pollutants limit for reference to maintain good air quality. This standard should also include guidelines for design of preschool building, material for construction and also furniture used inside the school which may play a part in contributing to indoor air pollutants. In addition, a system for refurbishment, cleaning and good hygiene practice should be made known for all school operators.

For pollutants specific control, the researcher recommends regular cleaning of classroom using vacuum cleaner as it prove more effective in eliminating small particulate as compare to floor sweeping. Besides, dust accumulates on fan blades are source of particulate matter, mould and bacteria. Weekly cleaning of fan blades and curtains washing should be done. Periodically and sparingly, use house bleach for cleaning the floor and furniture surface in the school to eliminate bacteria and other allergens. The bleach must then be stored away from classroom area as it also a source for VOCs. Kitchen should be separated from the classroom and door to the kitchen should be closed when cooking to avoid downwind movement of fumes into the classroom. Mould stain on the wall and ceiling must be removed if present.

© COPYRIGHT UPM

Where necessary, replacing the ceiling plywood with new one in the situation of bad mould issue. Repairs any water leaks immediately if present.

Next, we need to address the student health issue. Based on the results of the study, it is found that ECP was present in the entire children nasal sample. Thus, parents should seek medical treatment immediately to confirm presence of allergic rhinitis or asthma among the children and determine the need for anti-inflammatory drugs intervention. In the case where both problems are absent, the researcher would like to advice parent to perform nasal irrigation at home whenever symptoms appear. This method has been proven effective to control the severity of airway inflammation and also commonly use in medical treatment of rhinitis.

In term of the study itself, the researcher recommend that more study to be done to evaluate exposure of indoor air pollutants among children to serve as early understanding in preventing long term effects of poor indoor air quality towards health. The researcher hope that this study will be a baseline for more study to look into the immunologic level of effects as data is very limited especially with regards to study in Malaysia as this most probably the first of its kind in the country or even in Asia itself due to its nature of environmental health study. Even so, similar study in overseas mostly carried out in adults and children of older age with limited data for children of lower age. Besides, studies done in preschool age children focus more

© COPYRIGHT UPM

on medical values for asthma and allergic rhinitis knowledge instead of environmental health impacts. In addition, future studies in the country may look to use a different method for analysis of sample as studies overseas have proved that nasal lavage has better reproducibility of biomarkers found in nasal mucosa.

© COPYRIGHT UPM

REFERENCES

Arshad, S.H., (2010) Does exposure to indoor allergens contribute to the development of asthma and allergy? *Current Allergy Asthma Rep, 10(1):* 49-55.63.

Barnett *et al.* (2005). Air Pollution and Child Respiratory Health. *American Journal of Respiratory and Critical Care Medicine*, 1272–1278.

Bernstein, J.A., N. Alexis, H. Bacchus, I.L. Bernstein, P. Fritz, E. Horner, et al., (2008). The health effects of non-industrial indoor air pollution. *Journal of Allergy Clinical and Immunology 121(3):* 585-91.

Boot J. D., Chandoesing P., Kam de M. L., Mascelli M.A et al (2008) Applicability and Reproducibility of Biomarkers for the Evaluation of Anti-Inflammatory Therapy in Allergic Rhinitis. *J Investig Allergol Clin Immunol; Vol 18(6)*: 433-442

Bousquet J, Khaltaev N, Cruz A. A *et al.* (2008) Allergic Rhinitis and its Impact on Asthma (ARIA) 2008 update (in collaboration with the World Health Organization, GA(2) LEN and Allergen). *Allergy*; 63 (Suppl 86):8–160.

Bousquet J. Fokkens W. Burney P. Dunham S. R. Bachert C. Akdis C. A. et al (2008) Important Research Questions in Allergy and Related Diseases: Non Allergic Rhinitis: a GA2LEN Paper. *Allergy*; 63(7): 842-53

Bousquet J. van C. P. and Khaltaev N. (2001) Allergic Rhinitis and Its Impact on Asthma. *J Allergy Clin Immunol*; 108 (5 Suppl): S147-S334

Broms, K. D. N., Margaretha Erikkson, Claes Sundelin, Kurt Svardsudd (2013). Prevalence and co-occurrence of parentally reported possible asthma and

© COPYRIGHT UPM

allergic manifestations in pre-school children. *BioMed Central Public Health*, 13, 764.

Chan Yeung M, B. A. (2006). Primary prevention of childhood asthma and allergic disorders. Current Opinion in Allergy and Clinical Immunology, 6, 146-151.

Chawes B. L. (2011) Upper and Lower Airway Pathology in Young Children with Allergic and Non-Allergic Rhinitis. *Dan Med Bull; 58(5)*:B4278

Cooley, JD., Wong, WC, Jumper, CA, Straus, D,. (1998). Correlation between the prevalence of certain fungi and sick building syndrome. *Occup Environ Med, 55*, 579-584

DB, P. (1997). Mechanisms of pollution-induced airway disease: *in vivo* studies. *Allergy, 52*, 37-44.

Decarro C, Picco AM, Grisoli P, Redolfi M. (2003). Determination of aerial microbiological contamination in scholastic sports environment. *J Appl Microbiol* 95:904-912.

Dennis K. Ledford, Richard F. Lockey. (2013) Asthma and Comorbidities. *Curr Opin Allergy Clin Immunol*; 13(1): 78-86

Department of Environmental, DOE (1988) Recommended Malaysian Air Quality Guidelines (RMAQG). *Department for Environmental*, Malaysia.

Duhme, H., S.K. Weiland, & U. Keil, (1998). Epidemiological analyses of the relationship between environmental pollution and asthma. *Toxicology Letter, 102-103:* 307-16.

© COPYRIGHT UPM

EPA. (2012, July 10). Volatile Organic Compounds (VOCs). Retrieved from United States Environmental Protection Agency: http://www.epa.gov/iaq/voc.html

EPA. (2012, June 22). Biological Pollutants. Retrieved from United States Environmental Protection Agency: http://www.epa.gov/iaq/biologic.html

EPA. (2013, June 13). An Introduction to Indoor Air Quality (IAQ). Retrieved from United States Environmental Protection Agency: http://www.epa.gov/iaq/ia-intro.html

EPA. (2013, March 18). Particulate Matter. Retrieved from United States Environmental Protection Agency: http://www.epa.gov/ncer/science/pm/

Faustman, E.M., S.M. Silbernagel, R.A. Fenske, T.M. Burbacher, & R.A. Ponce, (2000). Mechanisms underlying Children's susceptibility to environmental toxicants. *Environmental Health Perspective 108 Suppl1:* 13-21.

Florida Health (2013) Indoor Air Quality. Department of Health, Florida. Retrieved 19th November, 2013, from http://www.floridahealth.gov/chdlee/eh/iaq.html

Flynn E, Matz P, Woolf A. Wright R. (2001) Indoor Air Pollutants Affecting Child Health. American College of Medical Toxicology. Retrieved May, 2014. From acmt.net/_Library/docs/IndoorAirPolution.pdf.

Frischer, T. and Baraldi, E. (2000). Upper airway sampling. *American Journal of Respiratory and Critical Care Medicine, 162*, S28-S30.

Gent, J.F., K. Belanger, E.W. Triche, M.B. Bracken, W.S. Beckett, & B.P. Leaderer, (2009). Association of pediatric asthma severity with exposure to common household dust allergens. *Environ Res, 109(6):* 768-74.

© COPYRIGHT UPM

Gold, D.R., 2000 Environmental tobacco smoke, indoor allergens, and childhood asthma. *Environmental Health Perspective,* 108 Suppl 4: 643-51.

Gordon, D. (2004). The Environment & Children's Health: A Journalist's Resource for In-Depth Reporting. Environmental. *Washington*: Washington.

Gui-Hong Cai, J. H. H., Zailina Hashim, Faridah Ali, Erica Bloom, Lennart Larrson, Erik Lampa, Dan Norback (2011). Fungal DNA, allergens, mycotoxins and associations with asthmatic symptoms among pupils in schools from Johor Bahru, Malaysia. *Pediatric Allergy and Immunology*, 22, 290-297.

Henretig FM, K. C. (1997). Textbook of Pediatric Emergency Procedures, Chapter 123: Obtaining Biological Specimens. Williams and Williams.

Hill, M. (2005). Chapter 8: Respiratory system pp. (18). Available from http://highered.mcgrawhill.com/sites/dl/free/0072841869/234727/mad41869ch08pg141158.pdf

Hillel S, K., Gary E Hatch, Delores E Graham (1990). Nasal lavage as a tool in assessing acute inflammation in response to inhaled pollutants. *Toxicology, 60*, 15-25.

Holgate S. T. (2010) A Look at the Pathogenesis of Asthma: The Need for A Change in Direction. *Discovery Medicine*; 9 (48): 439-447.

ISAAC Steering Committee (2000). ISAAC Phase Three Manual. *ISAAC International Data Centre,* Auckland, New Zealand.

Janssen N. A., Hoek G., Brunekreef B., Harssema H. (1999). Mass Concentration and Elemental Composition of PM10 in Classroom. *Occ Environ Med*: 56: 482-487

© COPYRIGHT UPM

Jessen, M., Malm, L. (1997). Prevalence and development of nasal obstruction. Allergy, 52 (40), 3-6

Jonas, B. (2002). Eosinophil cationic protein, expression levels and polymorphisms. Comprehensive Summaries of Uppsala Dissertations from the Faculty of Medicine, 1161, 11-13

Khan, M. A. (2013). Inflammation signals airway smooth muscle cell proliferation in asthma pathogenesis. *Multidisciplinary Respiratory Medicine*, 8:11.

Kim, K.H., S.A. Jahan, & E. Kabir, (2013). A review on human health perspective of air pollution with respect to allergies and asthma. *Environ Int, 2013. 59C*: 41-52.

Klimek, L. and Rasp, G. (1999) Norm Values for Eosinophil Cationic Protein in Nasal Secretions: Influence of Specimen Collection. *Clinical and Experimental Allergy, 29*: 367-374

Koren, HS., Hatch, GE., Graham, ED. (1990). Nasal lavage as a tool in assessing acute inflammation in response to inhaled pollutants. *Toxicology, 60*, 15-25

Landrigan, P.J., (1998). Environmental hazards for children in USA. *Int Journal of Occupational Medicine and Environmental Health, 11(2)*: 189-194.

Lemeshow *et al.* (1990) Adequacy of Sample Size in Health Studies. World Health Organisation, *John Wiley and Son,* Chischester.

Lignell, U., T. M., Tuula Puius, Asko Vepsalainen, Marjut Roponen, Eila Torwinen, Morten Reeslev, Sirpa Pennanen, Maija-Riitta Hirvonen, Pentti Kalliokoski, Aino Nevalainen (2005). Microbial exposure, symptoms and inflammatory mediators in nasal lavage fluid of kitchen and clerical personnel in schools.

© COPYRIGHT UPM

International Journal of Occupational Medicine and Environmental Health, 18(2), 139-150.

Liu W et al. (2003) Mosquito coil emissions and health implications. *Environ Health Perspect. 111*:1454-1460.

Lwanga, S. K. and Lemeshow, S. (1991). Sample Size Determination in Health Studies: A Practical Manual. World Health Organisation, Geneva.

M. Ismail *et al* (2010) Indoor Air Quality in Selected Samples of Primary Schools in Kuala Terengganu, Malaysia. *Environment Asia 3 (special issue)*, 103-108, 2010

McHugh, B.M. & A.J. MacGinnitie, 2011 .Indoor allergen sensitization and the risk of asthma and eczema in children in Pittsburgh. *Allergy Asthma Proc 32(5)*: 372-6

Mendell M. J. (2007). Indoor resential chemical emissions as risk factors for respiratory and allergic effects in children: a review. *Indoor Air*, 17, 259-277.

MOE. (2013, November 19). Prasekolah. Retrieved from Malaysia Ministry of Education: http://www.moe.gov.my/my/prasekolah

Munir, AKM, Einarsson, R., Schou, C. (1993). Alergens in school dust. I. The amount of the major cat (Fel d I) and dog (Can f I) allergens in dust from Swedish schools is high enough to probably cause perennial symptoms in mot children with asthma who are sensitized to cat and dog. *J Allergy Clin Immunol, 91*, 1067-1074

© COPYRIGHT UPM

National Heart, L., and Blood Institute (2012). The respiratory system Retrieved 19th November, 2013, from http://www.nhlbi.nih.gov/health/health-topics/topics/hlw/system.html

Nazariah S.S.N., Juliana J. and Abdah M.A. (2013) Interleukin-6 VIA Sputum Induction as Biomarker of Inflammation for Indoor Particulate Matter among Primary School Children in Klang Valley, Malaysia. *Global Journal of Health Science;* 5 (4): 93-105.

Norback D, W. R., Wieslander G, Smedje G, Erwall C, Venge P (2000). Indoor air pollutants in schools: nasal patency and biomarkers in nasal lavage. *Allergy, 55*, 163-170.

Norback, D., Torgen, M., Endling, C. (1990). Volatile organic compounds, respirable dust and incidence of sick building syndrome in primary schools. *Br J Ind Med, 74*, 733-741

Ownby, D.R. (2010), Pet dander and difficult-to-control asthma: The burden of illness. *Allergy Asthma Proc, 31(5)*: 381-4.

Pazdrak K, Gorski P, Krakowiak A, and Ruta U (1993). Changes in nasal lavage fluid due to formaldehyde inhalation. Int Arch Occup Environ Health; 64: 515-519.

Pazdrak, K., Gorski, P., Krakowiak, A., Ruta, U. (1993). Changes in nasal lavage fluid due to formaldehyde inhalation. *Int Arch Occup Environ Health, 64*, 515-519

Peatce N, A.-K. N., Beasly R, Mallol J, Keil U, Mitchell E, Robertson C (2008). Worldwide trends in the prevalence of asthma symptoms: phase III of the international study of asthma and allergies in childhood (ISAAC). Thorax, 62(9), 758-766.

© COPYRIGHT UPM

Peden, DB. (1997). The use of nasal lavge for objective measaurement of irritant induced nasal in inflammation. *Regul Toxicol Pharmacol, 24*, 76-78

Pipkorn N, K. G., Enerback L (1989). Nasal mucosal response to repeated challenges with pollen allergen. *American Review of Respiratory Disease, 140*, 729-736.

Raphael GD, J. E., Baraniuk JN, Kim I, Meredith SD, Kaliner MA (1989). Pathophysiology of rhinitis, lactoferrin and lysozyme in nasal secretions. *Clinical Investigation, 84*, 1528-1535.

Reigart R et al. (2011) Pesticides in children. *Pediatric Clinics of North America. 48*:1185.

Renstrom, A. (2002). Exposure to airborne allergens: a review of sampling methods. *Journal of Environmental Monitoring, 4*, 619-622.

Smedge, G., Norbaeck, D. (2000). New ventilation systems at selected schools in Sweden- effects on asthma and exposure. *Arch Environ Health* 2000 (in press)

Smedhe, G., Norbaeck, D. Edling, C. (1997). Asthma among secondary school pupils in relation to the school environment. *Clin Exp Allergy, 27*, 1270-1278

Smedje, G., Norback, D., Edling, C. Subjective indoor air quality in schools in relation to exposure. *Indoor Air, 7*, 143-150

South Coast Air Quality Management District (SCAQMD) Air Quality Issues in School Site Selection. Guidance Document June 2005 (revised May 2007)

© COPYRIGHT UPM

Tan T.N, Shek L.P, Goh D.Y.T, Chew F.T, Lee B.W. (2006) Prevalence of asthma and comorbid allergy symptoms in Singaporean pre-schoolers. *Asian Pacific Journal of Allergy and Immunology 24*: 175-182

Taskinen, T., Meklin, T., Nousiainen, M., Husman, T., Nevalainen, A., Korppi, M., (1997). Moisture and mould problems in schools and respiratory manifestations in school children: clinical and skin test. *Acta Pediatr, 56*, 1181-1187.

USEPA (1995) Indoor air quality tools for schools. EPA 402-K-95-001, Washington, DC, pp 3-4.

USEPA (2012b) Revision to National Air Quality Standards for Particle Pollution. Webinar for States and Local Agencies. US Environmental Protection Agency. Retrieved May, 2014, from http://www.4clean.org/documents/12192012PMNAAQSWebinar.pdf

Waelinder, R., Norbaeck, D., Wieslander, G., Smedje, G., Erwall, C. Venge, P. (1998). Nasal patency and biomarkers in nasal lavage- the significance of air exchange rate and type of ventilation in schools. Int Arch Occup Environ Health, 71, 479-486

Walinder R, N. D., Wieslander G, Smedje G, Erwall C, Venge P (1999). Nasal patency and lavage biomarkers in relation to settled dust and cleaning routines in school. *Scandinavian Journal of Work, Environment, and Health, 45*, 137-143.

Wang, JH., Devalia, JL., Duddle, JM., Hamilton, SA., Davies, RJ. (1995). Effect of six-hour exposure to nitrogen dioxide on early-phase nasal response to allergen challenge in patients with a history of seasonal allergic rhinitis. *J Allergy Clin Immunol,* 96, 669-676.

© COPYRIGHT UPM

WHO (2008). WHO training package for the health sector, Children's health and the environment Available from http://www.who.int/ceh/capacity/Indoor_Air_Pollution.pdf

WHO (2013). Asthma Retrieved 19th November, 2013, from http://www.who.int/respiratory/asthma/en/

WHO. (2008). Indoor Air Pollution. Retrieved from The World Health Organization: www.who.int/ceh

Wickman M, Egmar, A. C., Emenius G, et al (1999) Fel d 1 and Can f 1 in Settled Dust and Airborne Fel d 1 in Allergen Avoidance Day-Care Centres for Atopic Children in Relation to Number of Pet-Owners, Ventilation and General Cleaning. *Clin Exp Allergy; 29*:626-632

Wieslander G, N. D., Walinder R, Erwall C, Venge P (1999). Inflammation markers in nasal lavage, and nasal symptoms in relation to relocation to a newly painted building: a longitudinal study. *International Archives of Occupational and Environmental Health, 72*, 507-515.

Willers, S., Andersson, S., Andersson, R., Gratten, J., Sverdrup, C., Rosell, L. (1996). Sick building syndromes among the staff in schools and kindergartens: are levels of volatile organic compounds and carbon dioxide raesponsible?. *Indoor Built Environ, 5*, 232-235

Wolkoff, P., Clausen, P. A., Jensen, B. Nielsen G. D., Wilkins, C. K (1997) Are we Maesuring the Relevants Indoor Pollutants? *Indoor Air; 7:* 92-106

World Health Organization (WHO), Strategic approaches to indoor air policy-making: WHO European centre for environment and health, *Bilthoven*, 1999.

© COPYRIGHT UPM

Xiaohui Xu, H. K., Sandie Ha (2011). *The impact of air pollution on health, economy, environment and agricultural sources*: InTech.

Yahaya NA and Jalaludin J (2014) Exposure to Indoor PM10 and Volatile Organic Compounds (VOCs) and Its Association to Respiratory Health among Preschool Children in Urban and Rural Area From Sources to Solution. 3: 13-16. *Springer*: IBSN 978.981-4560-69-6

Yoon, C., K. L., Daeguk Park (2011). Indoor air quality differences between urban and rural preschools in Korea. *Environmental Science and Pollution Research*, 18, 333-345.

© COPYRIGHT UPM

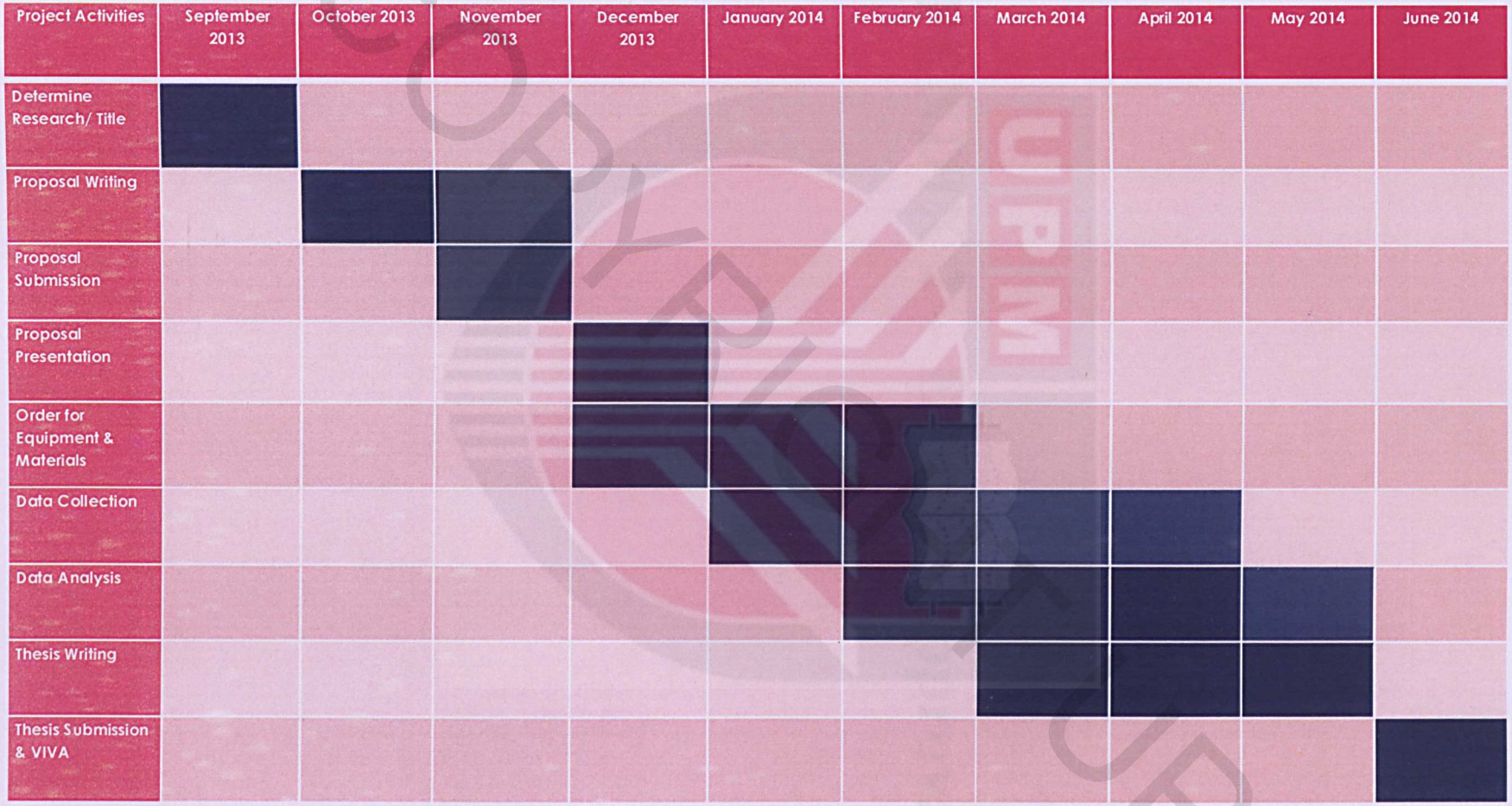
APPENDICES

© COPYRIGHT UPM

APPENDIX 1 – Gantt Chart

© COPYRIGHT UPM

GANTT CHART



© COPYRIGHT UPM

APPENDIX 2 – Ethical Approval

© COPYRIGHT UPM

APPENDIX 3 – Preschool

Approval

© COPYRIGHT UPM

APPENDIX 4 – Information Sheet and Consent Form (Parents)

© COPYRIGHT UPM

JAWATANKUASA ETIKA UNIVERSITI UNTUK PENYELIDIKAN MELIBATKAN MANUSIA (JKEUPM)
UNIVERSITI PUTRA MALAYSIA, 43400 UPM SERDANG,
SELANGOR, MALAYSIA

FORM B2: RESPONDENT'S INFORMATION SHEET AND GUARDIAN'S/ PARENT'S CONSENT

Please read the following information carefully. Do not hesitate to discuss any questions you may have with the researcher.

1. STUDY TITLE :

Indoor Air Pollutants Exposure and Eosinophil Cationic Protein (ECP) as Upper Airway Inflammatory Biomarker among Malay Preschool Children in Selangor.

2. INTRODUCTION:

Children at the age of 4 to 6 years old arguably spend most of their time indoors thus suggesting that exposure to indoor air pollutants is higher. Poor indoor air quality expose children to the potential harm of particulate matter, biological contaminants such as fungi and bacteria, volatile chemicals or volatile organic compounds (VOCs) and also formaldehyde (Yoon *et al*, 2010 & Lignell *et al*, 2005). Corresponding to these theory, studies in recent decades also seen that the prevalence of asthma and allergies cases increased particularly among children (Broms *et al*, 2013). We believe that, more time spend indoor with high level of pollutants induce respiratory symptoms among children of preschool aged. The study will assess the quality of indoor air in preschools environment to establish the association with upper airways inflammation among study respondents.

In this study, researcher is trying to measure the prevalence of upper airways inflammation due to exposure to indoor air pollutants by assessing the concentration of inflammatory biomarkers collected in nasal swab.

3. WHAT WILL YOU HAVE TO DO?

The guardian/parents are asked to answer the questionnaire to obtain the data information regarding on the study. 20 minutes will be given to complete the questionnaire.

4. WHO SHOULD NOT PARTICIPATE IN THE STUDY?

All students age below than 5 years old or those who are above 6 years old and not of Malay ethnicity. Students with severe allergic rhinitis problems, who have the history of severe injury to the nose and someone with history of other allergy and asthma problems.

© COPYRIGHT UPM

5. WHAT WILL BE THE BENEFITS OF THE STUDY:

(a) TO YOU AS THE SUBJECT?

This study will clarify whether the status of indoor air quality can cause problems to respiratory system or not to preschool children. If the concentrations of indoor air pollutants are higher and permission is given effect to the respondent, further action can be done based on the results of this research. Through this study, you will be able to determine whether your child is having respiratory problems not without incurring any charges.

(b) TO THE INVESTIGATOR?

Data will help investigator to determine the relationship between exposures to indoor pollutants with prevalence of respiratory symptoms such as upper airways inflammation among preschool children. Besides that, investigator will also be able to determine factors that contributing to level of indoor air pollutants in study areas.

6. WHAT ARE THE POSSIBLE RISKS?

All procedure is non-invasive and safe to be performed on all respondents.

7. WILL THE INFORMATION THAT YOU PROVIDE AND YOUR IDENTITY REMAIN CONFIDENTIAL?

The information and identity used in this study will remain confidential.

8. WHO SHOULD YOU CONTACT IF YOU HAVE ADDITIONAL QUESTIONS DURING THE COURSE OF THE RESEARCH?

If you have any additional questions, you may contact to Dr. Juliana Jalaludin, supervisor of the study research at 017-6834103 (M)/ 03-89472401 (O) or Andrew Dana anak Wesley, the researcher at 019-8828792 or email to andrew.dana18@gmail.com.

*Please initial here if you have read and understood the contents of this page*_____

© COPYRIGHT UPM

9. GUARDIAN'S/PARENT'S CONSENT

I …… Identity Card No. ……

address……..

……..hereby voluntarily agree to allow my *son / daughter / ward..... to take part in the research stated above *(clinical/ questionnaire/drug trial/video recording/ focus group/interview).

I have been informed about the nature of the research in terms of methodology, possible adverse effects and complications (as written in the Respondent's Information Sheet). I understand that my *son / daughter / ward has the right to withdraw from this research at any time without giving any reason whatsoever. I also understand that this study is confidential and all information provided with regard to the identity of my* son / daughter / ward will remain private and confidential.

I* wish / do not wish to know the results related to my my *son's / daughter's / ward's participation in the research

I agree/do not agree that the images/photos/video recordings/voice recordings related to my son/daughter/ward be used in any form of publication or presentation. (if applicable).

* delete where necessary

Signature …….. Signature ……..
(*Parent/Guardian*) (*Witness*)

Date :…….. Name :……..

I/C No. :……

I confirm that I have explained to the respondent's parent/guardian the nature and purpose of the above-mentioned research.

Date …….. Signature ……..
(*Researcher*)

© COPYRIGHT UPM

JAWATANKUASA ETIKA UNIVERSITI UNTUK PENYELIDIKAN MELIBATKAN MANUSIA (JKEUPM)
UNIVERSITI PUTRA MALAYSIA, 43400 UPM SERDANG, SELANGOR, MALAYSIA

BORANG B2: PENERANGAN DAN PERSETUJUAN IBUBAPA/PENJAGA

Sila baca maklumat berikut dengan teliti. Sekiranya anda mempunyai sebarang pertanyaan, sila kemukakan kepada penyelidik.

1. TAJUK KAJIAN

Pendedahan Kepada Pencemar Udara Dalaman Dan Eosinophil Cationic Protein (ECP) Sebagai Penanda Biologi Bagi Radang Salur Penafasan Atas Di Kalangan Pelajar Prasekolah Berbangsa Melayu di Selangor.

2. PENGENALAN

Kanak-kanak berumur sekitar 4 hingga 6 tahun dipercayai banyak menghabiskan masa mereka di dalam bangunan; di rumah atau di sekolah. Hal ini menunjukkan kadar pendedahan kepada pencemar udara dalaman bagi kanak-kanak tersebut adalah tinggi. Kualiti udara dalaman yang teruk akan mendedahkan seseorang kepada risiko kesihatan akibat pencemaran zarah-zarah, pencemar biologi seperti kulat dan bakteria, kimia organik yang meruap (VOCs), dan juga formadehid (Yoon et al, 2010 & Lignell et al, 2005). Sejajar dengan teori ini, kajian beberapa dekad ini telah menunjukkan peningkatan kes-kes asma dan alergi terutamanya di kalangan kanak-kanak (Broms et al, 2013). Jumlah masa yang banyak dihabiskan di dalam bangunan mampu menyebabkan keadaan masalah respiratori di kalangan kanak-kanak berumur lingkungan prasekolah. Kajian ini akan menilai kualiti udara dalaman bangunan tadika untuk mewujudkan perkaitan antara pendenddahan kepada pencemar udara dalaman dengan masalah radang salur penafasan atas.

Dalam kajian ini, penyelidik cuba menilai kadar kejadian kes radang salur penafasan atas dan hubungkait dengan pendedahan kepada pencemar dalaman udara menerusi analisa kepekatan penanda biologi yang didapati dari pensampelan sapuan hidung.

3. APAKAH YANG PERLU ANDA LAKUKAN?

Anda sebagai ibu/bapa/penjaga kepada anak anda dikehendaki menandatangani borang penyertan responden dan menyatakan minat anda untuk menyertai kajian ini. lanya boleh dilakukan setelah anda membaca dan memahami isi kandungan penerangan ini. Borang penyertaan responden harus dikembalikan kepada pengkaji sebelum temubual dan ujian yang akan dijalankan.

4. SIAPA YANG TIDAK BOLEH MENYERTAI KAJIAN INI?

Pelajar atau kanak-kanak yang berumur di bawah 5 tahun atau berumur melebihi 6 tahun dan bukan berbangsa Melayu tidak dibenarkan menyertai kajian ini. Pelajar atau kanak-kanak yang mempunyai masalah alahan rhinitis atau resdung, pernah mengalami kecederaan teruk pada hidung, dan mempunyai sejarah alergi dan asma yang disahkan oleh pihak perubatan/ doktor juga tidak dibenarkan untuk menjadi responden.

© COPYRIGHT UPM

5. APAKAH FAEDAH MENYERTAI KAJIAN INI?

a) Kepada Anak/Jagaan Saya Sebagai Peserta?

Kajian ini akan menjelaskan sama ada status kualiti udara dalaman boleh mengakibatkan masalah kepada sistem respiratori atau tidak kepada kanak- kanak prasekolah. Sekiranya kepekatan pencemar udara dalaman adalah tinggi dan memberi kesan kepada responden, tindakan selanjutnya boleh dilakukan berdasarkan hasil daripada maklumat kajian ini. Melalui kajian ini juga, anda akan dapat menentukan sama ada anak anda mengalami masalah dari segi sistem penafasan ataupun tidak tanpa dikenakan sebarang bayaran. Selain itu, kajian ini akan membantu anda untuk menentukan langkah terbaik bagi mengelakkan penyakit asma dan alergi pada peringkat awal di kalangan ahli keluarga terutama anak-anak sekiranya kualiti udara dalaman yang diperiksa tidak memuaskan.

b) Kepada Penyelidik?

Data yang diambil akan membantu penyelidik untuk mengkaji hubungkait pendedahan kepada pencemar udara dalaman dengan kadar kejadian masalah respiratori seperti radang salur penafasan atas di kalangan kanak-kanak prasekolah. Selain itu, penyelidik juga dapat mengkaji faktor-faktor yang menyebabkan wujudnya pencemaran udara dalaman. Hasil kajian ini boleh digunakan sebagai rujukan dan panduan kasar dalam membantu mengatasi masalah asma dan alergi pada peringkat umur yang awal melalui kawalan kualiti udara dalaman di rumah dan tadika.

6. ADAKAH IA BERISIKO?

Semua prosedur adalah selamat dan tidak berisiko.

7. ADAKAH MAKLUMAT DAN IDENTITI ANAK/JAGAAN SAYA KEKAL RAHSIA?

Maklumat dan identiti yang digunakan dalam kajian ini akan kekal sulit.

8. SIAPA YANG SAYA PERLU HUBUNGI SEKIRANYA SAYA MEMPUNYAI SOALAN TAMBAHAN SEPANJANG PENYELIDIKAN INI?

Jika anda mempunyai sebarang soalan tambahan, anda boleh menghubungi Dr. Juliana Jalaludin, penyelia kajian di 017-6834103 (M)/ 03-89472401 (O) atau Andrew Dana anak Wesley, penyelidk di 019-8828792 atau emel kepada andrew.dana18@gmail.com.

Sila tandatangan di sini sekiranya anda telah membaca dan memahami kandungan halaman ini

© COPYRIGHT UPM

9. PROSEDUR KAJIAN

Kajian ini mengandungi 2 peringkat secara keseluruhannya. Di sini, prosedur di setiap peringkat akan dijelaskan untuk kefahaman ibababpa/ penjaga bagi kanak-kanak yang akan terlibat dalam kajian ini. Adalah dimaklumkan bahawa kajian ini dijalankan atas dasar sukarela. Anda **digalakkan** untuk menyertai dan menyiapkan kedua-dua peringkat kajian ini untuk membantu mendapatkan data yang lebih tepat dan sesuai untuk digunakan oleh penyelidik dan anda sendiri berkaitan kesihatan anak-anak di bawah tanggungan. Berikut adalah peringkat-peringkat dalam kajian ini:

a. Borang Kaji Selidik

Peringkat ini wajib disiapkan oleh ibubapa/ penjaga yang bersetuju untuk anak mereka menyertai kajian ini. Borang Kaji Selidik akan diedarkan melalui guru tadika yang mengajar di tadika Kemas yang dihadiri oleh anak-anak tuan/ puan. Borang tersebut akan dibawa balik oleh pelajar prasekolah untuk diisi oleh ibubapa/ penjaga di rumah dan harus dikembalikan kepada guru tadika masing-masing mengikut tempoh yang diberitahu kemudian. Semua borang yang diisi sama ada bersetuju atau tidak bersetuju untuk membenarkan anak-anak tuan/ puan untuk menyertai kajian harus dikembalikan. Borang Kaji Selidik tersebut haruslah mengandungi:

I. Borang Penerangan dan Persetujuan Ibubapa/ Penjaga
II. Borang Penerangan dan Persetujuan Responden/ Peserta Kajian
III. Borang Soal Selidik Kajian

Borang I dan Borang II akan menerangkan tentang kajian ini; tujuan dan keperluannya. Borang-borang ini juga akan menerangkan sifat dan prosedur kajian dan akhirnya bahagian jawapan untuk persetujuan menyertai kajian.

Borang III pula merupakan soalan-soalan yang harus diisi oleh ibubapa/ penjaga peserta kajian mengenai maklumat responden, tahap kesihatan lampau dan semasa yang diperhatikan oleh penjaga atau yang disahkan oleh doctor perubatan serta simptom-simptom radang salur pernafasan atas yang mungkin disebabkan oleh bahan pencemar udara dalaman. Borang ini juga mengandungi soalan mengenai tahap pendedahan peserta kajian kepada kualiti udara dalaman dan juga ciri-ciri persekitaran di sekeliling kanak-kank prasekolah ini. Siap semua borang diisi, ianya haruslah dikembalikan kepada guru tadika dalam masa yang ditentukan pada awalnya.

Maklumat dalam Borang Kaji Selidik ini akan dirahsiakan dan hanya digunakan untuk mendapatkan data pendedahan peserta kajian kepada kualiti udara dalaman di tadika dan mencari hubungkait dengan hasil analisis bagi peringkat kajian yang seterusnya seperti dinyatakan di bawah.

b. Pengambilan Sampel Sapuan Hidung di kalangan Peserta Kajian

Peringkat kedua kajian ini memerlukan persetujuan dan kebenaran ibubapa/ penjaga dan juga responden bagi pengambilan sampel sapuan hidung. Pengambilan sampel basuhan hidung akan dilakukan oleh doktor perubatan atau jururawat kesihatan yang membantu dalam kajian ini. Dengan ini, setiap prosedur pengambilan sampel basuhan hidung ini mematuhi kehendak etika penyelidikan yang melibatkan manusia dan tidak berisiko kepada peserta kajian. Proses pengambilan sampel ini telah disahkan keberkesanannya dalam kajian-kajian sebelum ini termasuk di negara Eropah.

© COPYRIGHT UPM

Tujuan pengambilan sampel sapuan hidung ini adalah untuk menentukan kadar kepekatan/ konsentrasi sel protein iaitu Eosinophil Cationic Protein (ECP) dalam salur penafasan atas kanak-kanak tersebut. Sampel sapuan hidung tersebut akan dibawa ke makmal untuk dianalisa untuk kehadiran sel Eosinophil Cationic Protein (ECP). Jika sampel sapuan hidung peserta kajian didapati wujud kepekatan sel protein tersebut di dalamnya, ini menunjukkan peserta kajian mengalami radang salur penafasan atas. Justeru, ibubapa/ penjaga dan peserta kajian **digalakkan** untuk menyertai peringkat ketiga ini kerana hasil analisa sapuan hidung boleh membantu untuk menentukan hubungkait radang salur penafasan atas di kalangan kanak-kanak dengan pendedahan kepada bahan pencemar udara dalaman terutama di rumah dan tadika. Kejadian masalah radang salur penafasan atas dibuktikan berhubungkait dengan penyakit asma peringkat awal oleh kajian-kajian yang terdahulu. Persampelan ini akan dilakukan di rumah atau di sekolah mengikut permintaan ibubapa/ penjaga dan akan ditentukan kemudian bersama penyelidik selepas bersetuju untuk membenarkan anak di bawah jagaan menyertai kajian ini.

***Keputusan sampel sapuan hidung tidak boleh diambil kira sebagai penentu penyakit asma secara 100% dan ujian perubatan lanjut di hospital perlu dilakukan untuk mengesahkan sesuatu penyakit. Hasil analisa sampel sapuan hidung ini hanya untuk tujuan kajian ini dan hanya boleh dianggap penanda awal kepada masalh respiratori yang lain.**

Berikut adalah proses bagaimana sampel sapuan hidung ini akan dijalankan. Diingatkan lagi bahawa proses ini hanya akan dilakukan oleh doktor perubatan atau jururawat kesihatan yang terlibat dengan kajian ini.

Batang kapas yang telah direndam dalam air garam terlarut akan dimasukkan ke dalam kedua-dua lubang hidung untuk disapu pada dinding salur hidung. Sampel ini akan dibawa ke makmal untuk dianalisa.

Demikianlah prosedur kajian ini dijalankan. Diingatkan sekali lagi kajian ini dijalankan atas dasar sukarela, walau bagaimanapun kebenaran untuk kanak-kanak ini menyertai ketiga-tiga peringkat di atas amatlah digalakkan. Sekirannya ibubapa/ penjaga bersetuju untuk anak di bawah jagaan untuk menyertai kajian ini, borang kaji selidik di belakang ini wajib dilengkapkan namun boleh memilih untuk membenarkan atau tidak pensampelan sapuan hidung.

Sila tandakan pilihan anda di bawah ini. Tandakan semua jika setuju untuk membenarkan anak di bawah jagaan untuk menyertai semua peringkat kajian tersebut.
Dengan ini saya bersetuju untuk:

a. Membenarkan anak saya terlibat dengan kajian ini dan melengkapkan Borang Kaji Selidik ☐

b. Membenarkan sampel sapuan hidung diambil pada anak saya ☐

© COPYRIGHT UPM

10. PERSETUJUAN

Saya..... No Kad Pengenalan. .....

beralamat..... .....

..... dan bernombor telefon .....(bagi tujuan temujanji jika bersetuju) dengan ini secara sukarela bersetuju/ tidak bersetuju membenarkan *anak / jagaan

saya ..... menyertai **penyelidikan tersebut di atas *(klinikal/percubaan ubat-ubatan/rakaman video/kumpulan sasaran/temuduga/ soal selidik).**

Saya telah diberi penjelasan secara menyeluruh mengenai penyelidikan ini dari segi metodologi, risiko dan komplikasi (seperti yang tercatat dalam Helaian Penerangan). Saya memahami bahawa *anak / jagaan saya berhak menarik diri dari penyelidikan ini pada bila-bila masa tanpa memberi sebarang alas an. .Saya juga memahami bahawa sebarang maklumat yang berkaitan identiti *anak / jagaan saya akan dirahsiakan.

Saya* berminat / tidak berminat untuk mengetahui keputusan kajian yang **melibatkan** *anak / jagaan saya.

I setuju/tidak bersetuju untuk imej/gambar/rakaman video/ rakaman suara berkaitan dengan anak/ jagaan saya digunakan dalam apa jua bentuk penerbitan atau pembentangan. (sekiranya berkaitan).

*potong yang tidak berkenaan

Tandatangan ..... Tandatangan .....
(*Ibubapa/ Penjaga*) (*Saksi*)

Nama:..... Nama :.....

Tarikh :..... No. K/P: .....

Saya mengesahkan bahawa saya telah menerangkan kepada ibubapa/penjaga responden mengenai sifat dan tujuan penyelidikan tersebut di atas.

© COPYRIGHT UPM

Tarikh ……..…… Tandatangan ……..……

(*Penyelidik*)

© COPYRIGHT UPM

APPENDIX 5 – Information Sheet and Consent Form (Respondents)

© COPYRIGHT UPM

JAWATAN KUASA ETIKA UNIVERSITI UNTUK PENYELIDIKAN MELIBATKAN MANUSIA (JKEUPM)
UNVIVERSITI PUTRA MALAYSIA, 43400 UPM SERDANG, SELANGOR, MALAYSIA

FORM B1: RESPONDENT'S INFORMATION SHEET AND CONSENT

Please read the following information carefully and do not hesitate to discuss any questions you may have with the researcher

1. **STUDY TITLE:**

   Indoor Air Pollutants Exposure and Eosinophil Cationic Protein (ECP) as Upper Airway Inflammatory Biomarker among Malay Preschool Children in Selangor.

2. **INTRODUCTION:**

   Children at the age of 4 to 6 years old arguably spend most of their time indoors thus suggesting that exposure to indoor air pollutants is higher. Poor indoor air quality expose children to the potential harm of particulate matter, biological contaminants such as fungi and bacteria, volatile chemicals or volatile organic compounds (VOCs) and also formaldehyde (Yoon *et al*, 2010 & Lignell *et al*, 2005). Corresponding to these theory, studies in recent decades also seen that the prevalence of asthma and allergies cases increased particularly among children (Broms *et al*, 2013). We believe that, more time spend indoor with high level of pollutants induce respiratory symptoms among children of preschool aged. The study will assess the quality of indoor air in preschools environment to establish the association with upper airways inflammation among study respondents.

   In this study, researcher is trying to measure the prevalence of upper airways inflammation due to exposure to indoor air pollutants by assessing the concentration of inflammatory biomarkers collected in nasal swab.

3. **WHAT WILL YOU HAVE TO DO?**

   Nasal swab specimens will be collected from each respondent perform by medical assistant and supervise by medical doctor. You will have to cooperate with the medical assistant while the procedure of inserting cotton swab into both nostrils in process. Detail explanation of the procedure will be briefed to you from time to time during the collection. The specimens will be taken to the lab for analysis.

© COPYRIGHT UPM

4. WHO SHOULD NOT PARTICIPATE IN THE STUDY?

All students age below than 5 years old or those who are above 6 years old and not of Malay etnicity. Students with severe allergic rhinitis problems, who have the history of severe injury to the nose and someone with history of other allergy and asthma problems.

5. WHAT WILL BE BENEFITS OF THE STUDY:

a. TO YOU AS THE SUBJECT?

The respondent will be able to understand his health status regarding suspected respiratory problems resulted from exposure to indoor air pollutants.

b. TO THE INVESTIGATOR?

Data will help investigator to determine the relationship between exposures to indoor pollutants with prevalence of respiratory symptoms such as upper airways inflammation among preschool children. Besides that, investigator will also be able to determine factors that contributing to level of indoor air pollutants in study areas.

6. WHAT ARE THE POSIBLE RISKS?

All procedure is non-invasive and safe to be performed on all respondents.

7. WILL THE INFORMATION THAT YOU PROVIDE AND YOUR IDENTITY REMAIN CONFIDENTIAL?

The information and identity used in this study will remain confidential.

8. WHO SHOULD YOU CONTACT IF YOU HAVE ADDITIONAL QUESTIONS DURING THE COURSE OF THE RESEARCH?

If you have any additional questions, you may contact to Dr. Juliana Jalaludin, supervisor of the study research at 017-6834103 (M)/ 03-89472401 (O) or Andrew Dana anak Wesley, the researcher at 019-8828792 or email to andrew.dana18@gmail.com.

© COPYRIGHT UPM

9. CONSENT

I ..... Identify Card No. .....
address.....
..... hereby voluntarily agree to take part in the research stated above *(clinical/drug trial/video recording/focus group/interview-based/questionnaire-based).

I have been informed about the nature of the research in terms of methodology, possible adverse effects and complications (as written in the Respondent's Information Sheet). I understand that I have the right to withdraw from this research at any time without giving any reason whatsoever. I also understand that this study is confidential and all information provided with regard to my identity will remain private and confidential.

I* wish / do not wish to know the results related to my participation in the research

I agree / do not agree that the images/photos/video recordings/voice recordings related to me be used in any form of public or presentation (if applicable)

*delete where necessary

Signature ..... Signature .....

(Respondent) (Witness)

Date :..... ..... Name :.....

I/C No. :.....

I confirm that I have explained to the respondent the nature and purpose of the above-mentioned research.

Date..... Signature.....

(Researcher)

© COPYRIGHT UPM

JAWATAN KUASA ETIKA UNIVERSITI UNTUK PENYELIDIKAN MELIBATKAN MANUSIA (JKEUPM)
UNVIVERSITI PUTRA MALAYSIA, 43400 UPM SERDANG, SELANGOR, MALAYSIA

BORANG B1: HELAIAN KEBENARAN DAN MAKLUMAT RESPONDEN

Sila baca maklumat berikut dengan teliti. Sekiranya anda mempunyai sebarang pertanyaan sila kemukan kepada penyelidik.

1. TAJUK KAJIAN:

Pendedahan Kepada Pencemar Udara Dalaman Dan Eosinophil Cationic Protein (ECP) Sebagai Penanda Biologi Bagi Radang Salur Pernafasan Atas Di Kalangan Pelajar Prasekolah Berbangsa Melayu di Selangor.

2. PENGENALAN:

Kanak-kanak berumur sekitar 4 hingga 6 tahun dipercayai banyak menghabiskan masa mereka di dalam bangunan; di rumah atau di sekolah. Hal ini menunjukkan kadar pendedahan kepada pencemar udara dalaman bagi kanak-kanak tersebut adalah tinggi. Kualiti udara dalaman yang teruk akan mendedahkan seseorang kepada risiko kesihatan akibat pencemaran zarah-zarah, pencemar biologi seperti kulat dan bakteria, kimia organic yang meruap (VOCs), dan juga formadehid (Yoon et al, 2010 & Lignell et al, 2005). Sejajar dengan teori ini, kajian beberapa dekad ini telah menunjukkan peningkatan kes-kes asma dan alergi terutamanya di kalangan kanak-kanak (Broms et al, 2013). Jumlah masa yang banyak dihabiskan di dalam bangunan mampu menyebabkan keadaan masalah respiratori di kalangan kanak-kanak berumur lingkungan prasekolah. Kajian ini akan menilai kualiti udara dalaman bangunan tadika untuk mewujudkan perkaitan antara pendedahan kepada pencemar udara dalaman dengan masalah radang salur penafasan atas.

Dalam kajian ini, penyelidik cuba menilai kadar kejadian kes radang salur penafasan atas dan hubungkait dengan pendedahan kepada pencemar dalaman udara menerusi analisa kepekatan penanda biologi yang didapati dari pensampelan sapuan hidung.

3. APAKAH YANG PERLU ANDA LAKUKAN?

Spesimen sapuan hidung akan diambil dari setiap responden yang akan dilakukan oleh pembantu perubatan dengan diselia oleh pegawai perubatan. Anda dimohon untuk memberikan kerjasama sepenuhnya ketika prosedur sapuan hidung dengan batang kapas ke dalam kedua-dua belah lubang hidung. Penerangan lanjut mengenai proses tersebuat akan

© COPYRIGHT UPM

dilakukan dari semasa ke semasa ketika pensampelan. Spesimen yang dikumpul kemudian akan dibawa ke makmal untuk dianalisa.

4. SIAPA YANG TIDAK BOLEH MENYERTAI KAJIAN INI?

Pelajar atau kanak-kanak yang berumur di bawah 5 tahun atau berumur melebihi 6 tahun dan bukan berbangsa Melayu tidak dibenarkan menyertai kajian ini. Pelajar atau kanak-kanak yang mempunyai masalah alahan rhinitis atau resdung, pernah mengalami kecederaan teruk pada hidung, dan mempunyai sejarah alergi dan asma juga tidak dibenarkan untuk menjadi responden.

5. APAKAH FAEDAH MENYERTAI KAJIAN INI:

a. KEPADA ANDA SEBAGAI PESERTA?

Responden boleh mendapatkan informasi dan mengetahui status kesihatan berkaitan masalah respiratori yang dikaitkan dengan pendedahan kepada pencemar udara dalaman.

b. KEPADA PENYELIDIK?

Data yand diambil akan membantu penyelidik untuk mengkaji hubungkait pendedahan kepada pencemar udara dalaman dengan kadar kejadian masalah respiratori seperti radang salur penafasan atas di kalangan kanak-kanak prasekolah Selain itu, penyelidik juga dapat mengkaji faktor-faktor yang menyebabkan wujudnya pencemaran udara dalaman.

6. ADAKAH IA BERISIKO?

Semua prosedur adalah selamat dan tidak berisiko.

7. ADAKAH MAKLUMAT IDENTITI RESPONDEN KEKAL RAHSIA?

Maklumat dan identiti yang digunakan dalam kajian ini akan kekal sulit.

8. SIAPA YANG SAYA PERLU HUBUNGI SEKIRANYA SAYA MEMPUNYAI SOALAN TAMBAHAN SEPANJANG PENYELIDIKAN INI?

Jika anda mempunyai sebarang soalan tambahan, anda boleh menghubungi Dr. Juliana Jalaludin, penyelia penyelidikan kajian di 017-6834103 (M)/ 03-89472401 (O) atau Andrew Dana anak Wesley, penyelidk di 019-8828792 atau emel kepada andrew.dana18@gmail.com.

© COPYRIGHT UPM

9. PERSETUJUAN

Saya ..... No Kad Pengenalan.....
beralamat.....
..... dan nombor telefon .....
dengan ini secara sukarela bersetuju/ tidak bersetuju menyertai penyelidikan tersebut di atas *(klinikal/percubaan ubat-ubatan/rakaman video/kumpulan sasaran/temuduga/soal selidik).

Saya telah diberi penjelasan secara menyeluruh mengenai penyelidikan ini dari segi metodologi, risiko dan komplikasi (seperti yang tercatat dalam Helaian Penerangan). Saya memahami bahawa saya berhak menarik diri dari penyelidikan ini pada bila-bila masa tanpa member sebarang alasan. Saya juga memahami bahawa sebarang maklumat yang berkaitan identiti saya akan dirahsiakan.

Saya* berminat / tidak berminat untuk mengetahui keputusan kajian

Saya setuju / tidak bersetuju untuk imej/gambar/rakaman video/rakaman suara berkaitan dengan saya digunakan dalam apa jua bentuk penerbitan atau pembentangan. (sekiranya berkaitan)

*potong yang tidak berkenaan

Tandatangan ..... Tandatangan .....

(Responden) (Saksi)

Tarikh :..... ..... Nama :.....

No. K/P :.....

Saya mengesahkan bahawa saya telah menerangkan kepada responden mengenai sifat dan tujuan penyelidikan tersebut di atas.

Tarikh..... Tandatangan.....

(Penyelidik)

© COPYRIGHT UPM

APPENDIX 6 – Questionnaire

Form

© COPYRIGHT UPM

JABATAN KESIHATAN PERSEKITARAN DAN PEKERJAAN
FAKULTI PERUBATAN DAN SAINS KESIHATAN
UNIVERSITI PUTRA MALAYSIA

BORANG SOAL SELIDIK

TAJUK:

INDOOR AIR POLLUTANTS EXPOSURE AND EOSINOPHIL CATIONIC PROTEIN (ECP) AS UPPER AIRWAY INFLAMMATORY BIOMARKER AMONG MALAY PRESCHOOL CHILDREN IN SELANGOR.

ID RESPONDEN: ☐☐☐☐

TARIKH SOAL SELIDIK: ☐☐☐

SOAL SELIDIK DILENGKAPKAN OLEH:

Ibu kanak-kanak ☐

Bapa kanak-kanak ☐

Ibu dan bapa kanak-kanak ☐

Terima kasih atas kesudian anda menyertai penyelidikan saintifik ini. Kerjasama dari anda adalah sangat penting dalam menjayakan kajian ini. Jawapan yang jujur dan tepat amat diperlukan untuk melaksanakan projek ini.

Kesemua maklumat yang diperolehi dalam kajian ini akan dirahsiakan dan hanyalah untuk tujuan kajian kesihatan sahaja.

© COPYRIGHT UPM

BAHAGIAN I: MAKLUMAT PERIBADI KANAK-KANAK DAN KELUARGA

1. No. Responden: ☐☐☐
2. Nama kanak-kanak: _____
3. Nama ibu/bapa/penjaga* kanak-kanak: _____
4. Alamat: _____

   _____
5. No. Telefon: _____(pejabat)_____(rumah)
6. Tinggi kanak-kanak: ☐☐☐ cm Berat kanak-kanak: ☐☐ ☐
7. Umur: ☐☐ tahun
8. Tarikh lahir: ☐☐ hari ☐☐ bulan ☐☐ tahun
9. Bilangan adik-beradik: _____(orang)
10. Tahap Persekolahan: ☐☐☐☐
11. Nama sekolah: _____
12. Jantina: ☐ Lelaki ☐ Perempuan
13. Bangsa: ☐ Melayu ☐ Cina ☐ India ☐ Lain-lain
14. Tahap pendidikan bapa:

    ☐ Sekolah rendah ☐ PMR/SRP ☐ SPM ☐ STPM/Diploma

    ☐ Ijazah/Master/PHD
15. Tahap pendidikan ibu:

    ☐ Sekolah rendah ☐ PMR/SRP ☐ SPM ☐ STPM/Diploma

    ☐ Ijazah/Master/PHD
16. Pekerjaan bapa: _____
17. Pekerjaan ibu: _____

© COPYRIGHT UPM

18. Pendapatan bapa: RM _____._____ sebulan

19. Pendapatan ibu: RM _____. _____ sebulan

20. Pendapatan isi rumah: RM_____.____ sebulan

21. Tempat di mana kanak-kanak dilahirkan, sila nyatakan negeri dan bandar:

_____

22. Sudah berapa lama kanak-kanak tinggal di alamat sekarang: _____tahun _____ bulan

23. Sila senaraikan tempat-tempat dimana kanak-kanak ini pernah tinggal selama enam bulan atau lebih sejak lahir hingga sekarang:

a) _____

b) _____

© COPYRIGHT UPM

BORANG SOAL SELIDIK

BAHAGIAN B: SEKOLAH DAN PERSEKITARAN RUMAH SERTA KESIHATAN RESPIRATORI

B1: SESAK NAFAS DAN NAFAS BERBUNYI

1. Pernahkah anak anda mengalami sesak nafas ataupun nafas berbunyi di dada pada bila-bila masa yang lepas? 1 ☐

1 ☐ Ya 0 ☐ Tidak

Jika anda menjawab 'TIDAK', sila terus ke soalan 6.

2. adakah anak anda mengalami sesak nafas ataupun nafas berbunyi di dada 12 bulan yang lepas? 2 ☐

1 ☐ Ya 0 ☐ Tidak

Jika anda menjawab 'TIDAK', sila terus ke soalan 6

3. berapa banyakkah serangan sesak nafas yang dialaminya pada 12 bulan yang lepas? 3 ☐

☐ Tiada
☐ 1-3 episod
☐ 4-12 episod
☐ Lebih dari episod

4. Pada 12 bulan yang lepas, berapa kerapkah secara puratanya, tidur anak anda diganggu oleh masalah sesak nafas ? 4 ☐

☐ Tidak pernah terjaga disebabkan sesak nafas
☐ Kurang dari satu malam seminggu
☐ Satu atau lebih malam dalam seminggu

5. Pada 12 bulan yang lepas, adakah masalah kesesakan nafas anak anda berada di tahap yang serius, sehingga membataskan percakapan pada hanya satu atau dua patah perkataan sahaja diantara nafas? 5 ☐

1 ☐ Ya 0 ☐ Tidak

6. Pernahkah anak anda menghidapi asma (pengesahan oleh doktor/ pihak hospital)? 6 ☐

1 ☐ Ya 0 ☐ Tidak

Jika anda menjawab 'TIDAK', sila ke bahagian B2.

© COPYRIGHT UPM

7. Pada 12 bulan yang lepas, pernahkah dada anak anda berbunyi kuat selepas bersenam? 7 ☐

1 ☐ Ya 0 ☐ Tidak

8. pada 12 bulan yang lepas, pernahkah anak anda mengalami batuk tanpa kahak pada waktu malam? 8 ☐

1 ☐ Ya 0 ☐ Tidak

B2: SIMPTOM-SIMPTOM KULIT

1. Pernahkah anak anda mengalami ruam-ruam gatal yang tidak konsisten pada sekurang-kurangnya dalam tempoh 6 bulan? 9 ☐

1 ☐ Ya 0 ☐ Tidak

Jika anda menjawab 'TIDAK', sila terus ke soalan 7.

2. Adakah anak anda mengalami ruam-ruam gatal ini pada bila-bila masa dalam bulan yang lepas ? 10 ☐

1 ☐ Ya 0 ☐ Tidak

Jika anda menjawab 'TIDAK', sila terus ke soalan 7.

3. Adakah ruam-ruam gatal ini menyerang mana-mana bahagian ini?

1.	lipatan siku	1 ☐	Ya	0 ☐	Tidak	11	☐
2.	belakang lutut	1 ☐	Ya	0 ☐	Tidak	12	☐
3	dihadapan pergelangan kaki	1 ☐	Ya	0 ☐	Tidak	13	☐
4.	di bawah punggung	1 ☐	Ya	0 ☐	Tidak	14	☐
5.	sekelililing leher,telinga atau mata	1 ☐	Ya	0 ☐	Tidak	15	☐

4. Pada anak anda umur berapakah ruam-ruam gatal ini mula berlaku? 16 ☐

☐ Bawah 2 tahun

☐ 2-4 tahun

☐ 5 atau lebih

5. Adakah ruam ini hilang sepenuhnya pada bila-bila masa dalam 12 bulan yang lepas? 17 ☐

1 ☐ Ya 0 ☐ Tidak

© COPYRIGHT UPM

6. Pada 12 bulan yang lepas, berapa kerapkah secara puratanya, tidur anak anda diganggu disebabkan oleh ruam-ruam gatal ini? 18 ☐

☐ Tidak pernah
☐ Kurang dari satu malam seminggu
☐ Satu atau lebih malam dalam seminggu

7. Pernahkah anak anda menghidapi penyakit kulit (pengesahan oleh doktor/ pihak hospital)? 19 ☐

1 ☐ Ya 0 ☐ Tidak

B3: SIMPTOM-SIMPTOM HIDUNG

1. Pernahkah anak anda mempunyai masalah bersin, hidung berair, serta tersumbat apabila anda TIDAK menghidapi selsema ataupun demam? 20 ☐

1 ☐ Ya 0 ☐ Tidak
Jika anda menjawab 'TIDAK', sila terus ke soalan 6.

2. Dalam 12 bulan yang lepas, adakah anak anda mempunyai masalah bersin, ataupun hidung berair, serta tersumbat apabila anda TIDAK menghidapi selsema ataupun demam? 21 ☐

1 ☐ Ya 0 ☐ Tidak

3. Dalam 12 bulan yang lepas, pernahkah masalah hidung anak anda ini disertai dengan mata berair yang gatal? 22 ☐

1 ☐ Ya 0 ☐ Tidak

4. Pada bilakah dalam 12 bulan yang lepas ,masalah hidung ini terjadi? 23 ☐

Januari	☐	Mei	☐	September	☐
Februari	☐	Jun	☐	Oktober	☐
Mac	☐	Julai	☐	November	☐
April	☐	Ogos	☐	Disember	☐

5. Dalam 12 bulan yang lepas, masalah hidung ini menganggu aktiviti harian anak anda? 24 ☐

☐ Tidak sama sekali
☐ Sedikit
☐ Sederhana
☐ Banyak

© COPYRIGHT UPM

BAHAGIAN C: LAGI SOALAN TENTANG ASMA DAN ALAHAN

C1: SESAK NAFAS DAN NAFAS BERBUNYI DI DADA

1. Pernahkah anak anda mengalami sesak nafas atau nafas berbunyi di dada pada bila-bila masa dalam 12 bulan yang lepas? **Jika 'TIDAK', sila terus ke soalan di bahagian C2.**	**Ya** ( )	**Tidak** ( )
2. Pernahkah anak anda tercungap-cungap apabila berkeadaan sesak nafas?	**Ya** ( )	**Tidak** ( )
3. Pernahkah anak anda mengalami sesak nafas atau nafas berbunyi ini apabila anda tidak menghidap selsema?	**Ya** ( )	**Tidak** ( )
4. Pernahkah anak anda terjaga dengan rasa tegang di dada dalam 12 bulan yang lepas?	**Ya** ( )	**Tidak** ( )

C2: SESAK NAFAS

1. Pernahkah anak anda mendapat serangan sesak nafas yang datang tiba-tiba ketika berehat dalam 12 bulan yang lepas?	**Ya** ( )	**Tidak** ( )
2. Pernahkah anak anda mendapat serangan sesak nafas yang datang selepas melakukan aktiviti berat pada bila-bila masa dalam 12 bulan yang lepas?	**Ya** ( )	**Tidak** ( )
3. Pernahkah anak anda terjaga kerana sesak nafas dalam 12 bulan yang lepas?	**Ya** ( )	**Tidak** ( )

C3: ASMA

1. a. Pernahkah anak anda menghidap asma? **JIKA 'TIDAK', SILA KE BAHAGIAN D.**	**Ya** ( )	**Tidak** ( )
b. Adakah asma itu dikesan oleh doktor?	**Ya** ( )	**Tidak** ( )
c. Jika ya, berapakah umur anak anda ketika itu?	_____ **tahun.**	
d. Berapakah umur anak anda apabila mendapat serangan asma pertama?	_____ **tahun.**	
e. Berapakah umur anak anda ketika mendapat serangan yang terbaru?	_____ **tahun.**	
2. Pernahkan anak anda mendapat serangan asma dalam 12 bulan yang lepas?	**Ya** ( )	**Tidak** ( )
3. Adakah anak anda sekarang menggunakan apa-apa ubat untuk asma? (semburan- inhaler, ubat biji, dan sebagainya)	**Ya** ( )	**Tidak** ( )

©COPYRIGHT UPM

BAHAGIAN D: SOALAN TENTANG KESIHATAN DAN ALAHAN SEMASA

1. Berapa banyakkah jangkitan pada paru-paru yang anak anda alami semasa 3 bulan yang lepas?

   _____ ***(nombor)***

2. Pernahkah anak anda menggunakan antibiotic untuk melawan jangkitan pada paru-paru semasa 12 bulan yang lepas?

   Tidak pernah ( ) **Ya, sekali** ( ) **Ya, lebih dari sekali** ( )

3. Adakah anak anda mempunyai apa-apa penyakit yang memerlukan anda berjumpa dengan doctor? **Ya** ( ) **Tidak** ( )
   Jika ya, penyakit apa?
   _____

4. Adakah bapa anak anda seorang perokok? **Ya** ( ) **Tidak** ( )

5. Adakah ibu anak anda seorang perokok? **Ya** ( ) **Tidak** ( )

6. Adakah anak anda terlalu sensitif /alah kepada kucing? **Ya** ( ) **Tidak** ( )

7. Adakah anak anda terlalu sensitif /alah kepada anjing? **Ya** ( ) **Tidak** ( )

8. Adakah anak anda terlalu sensitif /alah kepada objek yang berkulat? **Ya** ( ) **Tidak** ( )

9. Adakah anak anda terlalu sensitif/alah kepada debunga? **Ya** ( ) **Tidak** ( )

10. Adakah anak anda mempunyai alahan kepada makanan? **Ya** ( ) **Tidak** ( )
    Jika ya, apa penyebab reaksi alahan itu?
    _____

11. Lain-lain alahan: _____

D1: PENYAKIT ALAHAN DI KALANGAN AHLI KELUARGA YANG LAIN

1. Bilangan jumlah adik beradik yang lebih tua, kesemuanya: _____
2. Jumlah adik-beradik yang lebih tua yang tinggal di rumah sekarang: _____
3. Jumlah adik beradik yang lebih muda,kesemuanya: _____
4. Jumlah adik-beradik yang lebih muda yang tinggal di rumah sekarang: ____
5. Adakah terdapat penyakit alahan di dalam keluarga? Tandakan X pada yang berkenaan walaupun simptom itu sudah tiada.

© COPYRIGHT UPM

	Bapa	Ibu	Adik-beradik
Asma	( )	( )	( )
Alahan simptom hidung	( )	( )	( )
Alahan pada kulit	( )	( )	( )

BAHAGIAN E: TENTANG ZAMAN KANAK-KANAK

1. Adakah anak anda minum susu ibu ketika kecil? **Ya** **Tidak**
   Jika ya, sehingga umur berapa ? _____ ( ) ( )
2. Adakah sesiapa di dalam keluarga yang merokok dari anak anda lahir sehingga berumur setahun? **Ya** **Tidak**
   Bapa merokok ( ) ( )
   Ibu merokok ( ) ( )
   Ahli keluarga lain merokok ( ) ( )

3. Adakah anak anda dihantar ke tempat jagaan/nurseri? **Ya** **Tidak**
   ( ) ( )

   Jika ya, pada umur berapakah anak anda mula dihantar ke tempat jagaan/nurseri?

   _____

BAHAGIAN F: PERSEKITARAN RUMAH SEKARANG

1. Apakah jenis rumah yang didiami sekarang? Sila tandakan dia kotak sebelah:
   Teres ( )
   Banglo ( )
   Apartment ( )
   Flat ( )
   Kampung/felda ( )
   Jenis-jenis lain ( )

2. Adakah anda tinggal di kediaman yang sama sejak anak anda lahir? **Ya** **Tidak**
   ( ) ( )
   Jika TIDAK, pada tahun berapakah anda pindah ke kediaman sekarang? _____
3. Berapakah keluasan tempat kediaman anda? **Ya** **Tidak**
   ( ) ( )

4. Bahan apakah yang digunakan dalam pembinaan rumah anda?
   Batu ( )
   Konkrit ( )

© COPYRIGHT UPM

Batu-bata ( )
Kayu ( )
Bahan-bahan lain ( )

	Ya	Tidak
5. Adakah bahagian dalam rumah anda dicat dalam 12 bulan yang lepas?	( )	( )
6. Adakah lantai rumah anda ditukar sejak 12 bulan yang lepas?	( )	( )
7. Adakah terdapat binatang peliharaan didalam kediaman anda?	( )	( )

Jika ya, binatang jenis apakah ia?
_____

8. Adakah mana-mana yang berikut dikenalpasti di tempat kediaman anda dalam 12 bulan yang lepas?

	Ya	Tidak
Air bocor dan merosakkan dinding,lantai atau siling	( )	( )
Buih atau luntur kuning pada pelapik plastik lantai	( )	( )
ataupun luntur hitam pada lantai karpet	( )	( )
Kulat tumbuh di dinding,lantai, atau siling	( )	( )
Bau kulat di mana-mana bilik (kecuali bilik bawah tanah)	( )	( )
Bau-bau lain di dalam rumah.jika YA, nyatakan bau itu	( )	( )

_____

	Ya	Tidak
9. Adakah masalah lembap/kerosakan air berlaku di tempat kediaman anda dalam 5 tahun yang lepas?	( )	( )

10. Adakah terdapat asap rokok di dalam rumah?
Ya, setiap hari ( )
Ya, biasanya 1-4 kali seminggu ( )
Ya,kadang-kadang 1-3kali sebulan ( )
Tidak pernah ( )

BAHAGIAN F: LAGI MAKLUMAT BERKAITAN PERSEKITARAN RUMAH

1. Berapa buah bilikkah yang terdapat di dalam rumah ini? _____ Bilik
2. Berapa orangkah yang tinggal di dalam rumah ini? _____ orang
3. Kanak-kanak ini tidur/tinggal di dalam bilik

☐ Sendiri ☐ Berkongsi dengan 2 orang

☐ Berkongsi dengan 3 orang ☐ Berkongsi dengan 4 orang

© COPYRIGHT UPM

4. Apakah bahan api yang digunakan untuk memasak?

   ☐ Elektrik ☐ Minyak tanah ☐ Arang

   ☐ Gas ☐ Kayu api

5. Berapa kali dalam sehari anda gunakan untuk memasak ? _____ kali

6. Semasa anda memasak, adakah anda membuka tingkap atau pintu atau alat penyedut asap keluar untuk membenarkan pengaliran udara di dalam rumah ?

   ☐ Ya ☐ Tidak

7. Alat apakah yang digunakan untuk menyejukkan udara di dalam rumah ?

   ☐ Penyaman udara ☐ Kipas

   ☐ Lain-lain _____ (sila nyatakan)

8. Adakah anda mempunyai haiwan peliharaan di dalam rumah ?

   ☐ Ya ☐ Tidak

9. Jika 'Ya', sila nyatakan : _____

10. Adakah anda menggunakan bahan tertentu untuk mengelakkan serangan nyamuk ?

    ☐ Ya ☐ Tidak

10a. Jika ya, jenis apakah yang selalu digunakan ?

    ☐ Lingkaran biasa ☐ Semburan Aerosol

    ☐ Elektrik ☐ Lain-lain _____ (sila nyatakan)

10b. Berapa kerapkah anda menggunakannya dalam seminggu ? _____ Kali seminggu

10c. Dimanakah ianya ditempatkan di dalam rumah ?

    ☐ Diruang tamu sahaja ☐ Di bilik tidur

    ☐ Bilik tidur dan ruang tamu

11. Apakah alat yang digunakan untuk membersihkan rumah anda?

    Sila nyatakan : _____

12. Berapa kerapkah dalam seminggu anda membersihkan rumah anda ?

    _____ Kali seminggu

© COPYRIGHT UPM

13. Adakah anda menggunakan karpet di kediaman anda ?

☐ Ya ☐ Tidak

14. Jenis kawasan perumahan

☐ Kampung ☐ Flat

☐ Rumah teres setingkat ☐ Banglo

☐ Rumah teres dua tingkat

15. Lokasi rumah dari jalanraya

☐ < 100 meter dari jalanraya

☐ > 100-500 meter dari jalanraya

☐ > 500-1000 meter dari jalanraya

☐ > 1000 meter dari jalanraya

16. Lokasi rumah anda dari kawasan ladang

☐ < 500 meter dari kilang

☐ > 1-1.5 kilometer dari kilang

☐ > 1.5 – 3 kilometer dari kilang

☐ > 3 kilometer dari kilang

17. Apakah pendapat anda mengenai persekitaran rumah anda ?

☐ Sangat berhabuk

☐ Sederhana berhabuk

☐ Kurang berhabuk

18. Apakah kenderaan yang digunakan oleh anak anda ke sekolah ?

☐ Kereta ☐ Basikal

☐ Berjalan kaki ☐ Bas ☐ Motorsikal

© COPYRIGHT UPM

BAHAGIAN G: MAKLUMAT TENTANG TABIAT PEMAKANAN SEKARANG

	Tidak pernah	Jarang sekali	Sekali seminggu	Lebih sekali seminggu	Hampir setiap hari
Berapa kerapkah anak anda makan daging?	( )	( )	( )	( )	( )
Berapa kerapkah anak anda makan ikan?	( )	( )	( )	( )	( )
Berapa kerapkah anak anda makan makanan laut?	( )	( )	( )	( )	( )
Berapa kerapkah anak anda makan buah-buahan?	( )	( )	( )	( )	( )
Berapa kerapkah anak anda makan ulam-ulaman?	( )	( )	( )	( )	( )
Berapa kerapkah anak anda makan sayur yang dimasak?	( )	( )	( )	( )	( )
Berapa kerapkah anak anda meminum susu?	( )	( )	( )	( )	( )
Berapa kerapkah anak anda mengambil makanan tenusu? (yogurt,keju,mayonis dll)	( )	( )	( )	( )	( )
Berapa kerapkah anak anda makan makanan segera? (burger,pizza,hotdog,nugget,dll)	( )	( )	( )	( )	( )
Berapa kerapkah anak anda minum jus buah-buahan?	( )	( )	( )	( )	( )
Berapa kerapkah anak anda minum air bergas?	( )	( )	( )	( )	( )

2. Apakah jenis minyak masak yang sering digunakan di rumah anda?

Mentega	( )
Margerin	( )
Minyak jagung	( )
Minyak kelapa sawit	( )
Minyak sayuran	( )
Lain-lain	( )

© COPYRIGHT UPM

BAHAGIAN H: SIMPTOM SEMASA

1. Adakah anak anda mengalami simptom-simptom berikut dalam masa 3 bulan yang lepas?

	Ya, setiap hari	Ya, 1-4 kali seminggu	Ya, kadang-kadang 1-3 kali sebulan	Tidak pernah
1. Ruam di tangan	( )	( )	( )	( )
2. Ruam di muka/tekak	( )	( )	( )	( )
3. Sakit kulit. Jika ya, di mana? _____	( )	( )	( )	( )
4. Gatal-gatal di muka/tekak	( )	( )	( )	( )
5. Gatal-gatal di tangan	( )	( )	( )	( )
6. Radang mata (merah,kering, gatal)	( )	( )	( )	( )
7. Bengkak kelopak mata	( )	( )	( )	( )
8. Sakit kepala	( )	( )	( )	( )
9. Loya	( )	( )	( )	( )
10. Hidung berair/selsema	( )	( )	( )	( )
11. Hidung tersekat/tersumbat	( )	( )	( )	( )
12. Kering tekak	( )	( )	( )	( )
13. Sakit tekak	( )	( )	( )	( )
14. Radang batuk	( )	( )	( )	( )
15. Susah bernafas	( )	( )	( )	( )
16. Penat dan tak berdaya	( )	( )	( )	( )

	Ya	Tidak	Tidak tahu
2. Adakah simptom-simptom ini bertambah baik bila balik dari sekolah? Jika YA, simptom yang mana? _____	( )	( )	( )
3. Adakah simptom-simptom ini bertambah baik bila berjauhan dari tempat tinggal? Jika YA, simptom yang mana? _____	( )	( )	( )

© COPYRIGHT UPM

TERIMA KASIH

SULIT DAN PERIBADI

© COPYRIGHT UPM

APPENDIX 7 – School

Environment Evaluation Form

© COPYRIGHT UPM

MAKLUMAT PERSEKITARAN SEKOLAH

A. MAKLUMAT PERSEKITARAN DALAM SEKOLAH

1. Alat apakah yang digunakan untuk menyejukkan udara di dalam sekolah?

   ☐ Penyaman udara ☐ Kipas

   ☐ Lain-lain _____ (nyatakan)

2. Berapa orangkah kanak-kanak yang bersekolah di dalam sekolah ini?

   _____ orang

3. Jika anda memasak, apakan bahan api yang digunakan untuk memasak?

   ☐ Elektrik ☐ Minyak tanah ☐ Arang

   ☐ Gas ☐ Kayu api

4. Berapa kali dalam sehari anda gunakan untuk memasak?

   _____ kali sehari

5. Semasa anda memasak, adakah anda membuka tingkap atau pintu untuk membenarkan pengaliran udara di dalam sekolah?

   ☐ Ya ☐ Tidak

6. Berapa kerapkah dalam seminggu anda membersihkan kawasan dalam kelas?

   _____kali seminggu

7. Apakah alat yang digunakan untuk membersihkan kawasan dalam kelas?

   Sila nyatakan: _____

8. Adakah anda menggunakan karpet di dalam kelas?

   ☐ Ya ☐ Tidak

© COPYRIGHT UPM

B. MAKLUMAT PERSEKITARAN LUAR SEKOLAH

1. Bahan binaan sekolah kanak-kanak:

☐ Batu/simen ☐ Kayu/papan ☐ Lain/lain _____(sila nyatakan)

2. Jenis kawasan sekolah:

☐ Bandar ☐ Luar Bandar

3. Lokasi sekolah dari jalan raya:

☐ < 100 meter dari jalan raya

☐ > 100-500 meter dari jalan raya

☐ > 500-1000 meter dari jalan raya

☐ > 1000 meter dari jalan raya

4. Lokasi sekolah dari kilang:

☐ < 1 kilometer dari kilang

☐ > 2-3 kilometer dari kilang

☐ > 3-4 kilometer dari kilang

☐ > 5 kilometer dari kilang

6. Apakah pendapat anda mengenai persekitaran sekolah anda?

☐ Sangat berhabuk

☐ Sederhana berhabuk

☐ Kurang berhabuk

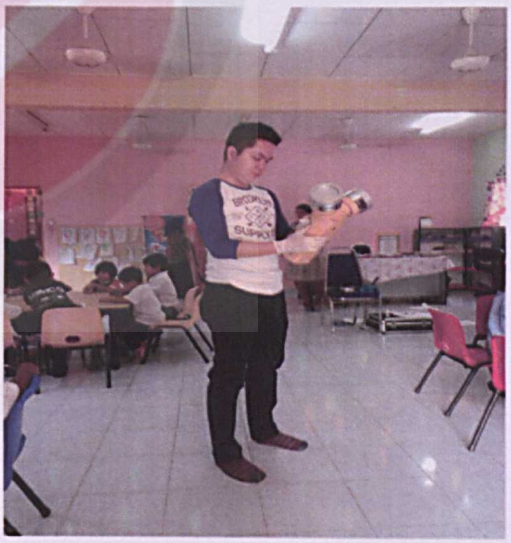
©COPYRIGHT UPM

APPENDIX 8 – Sampling and Analysis Pictures

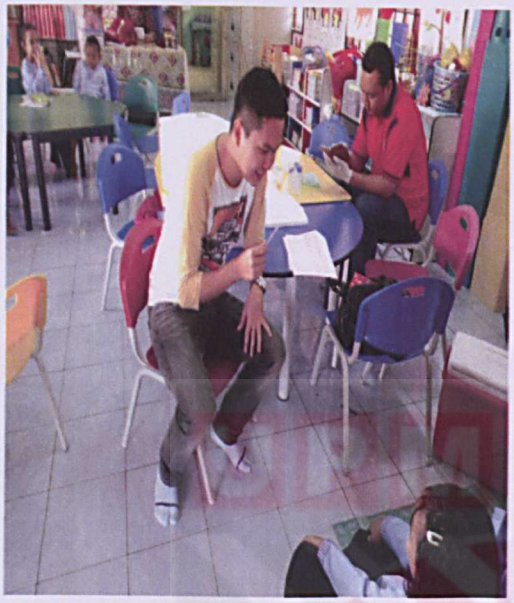
© COPYRIGHT UPM

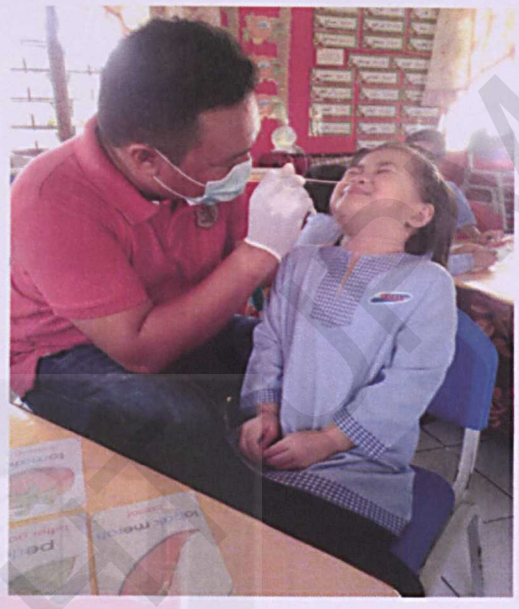
SAMPLING & ANALYSIS PICTURES

Distribution of questionnaires

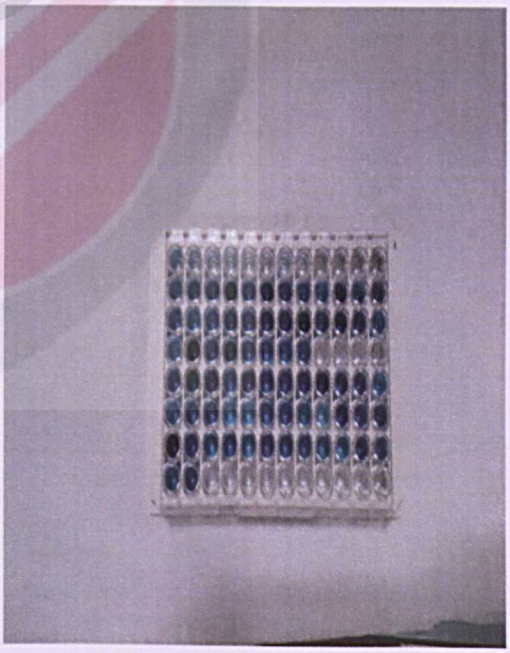


Indoor Air Quality (IAQ) monitoring





Collection of nasal sample



ELISA Test for ECP