



UNIVERSITI PUTRA MALAYSIA

***ASSOCIATION OF EXPOSURE TO PM₁₀ AND PM_{2.5} WITH DNA
DAMAGE IN EXFOLIATED BUCCAL MUCOSA CELL AMONG
PRIMARY SCHOOL CHILDREN LIVING NEAR PALM OIL ACTIVITY
AT SEMENYIH, SELANGOR***

SYHIDATUL FARHANA OTHMAN

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FPSK4 2019 23**

**ASSOCIATION OF EXPOSURE TO PM₁₀ AND PM_{2.5} WITH DNA DAMAGE
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CHILDREN LIVING NEAR PALM OIL ACTIVITY AT SEMENYIH,
SELANGOR**



**BY
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**Thesis submitted in fulfilment of the requirement for the degree of Bachelor
Science (Environmental and Occupational Health) from the Faculty of Medicine
and Health Sciences, Universiti Putra Malaysia.**

ACKNOWLEDGEMENTS

First and foremost, I would like to thank the Almighty for giving me strength and a good health to complete this thesis as part of the requirement to graduate as a student of Bachelor Environmental and Occupational Health at University Putra Malaysia.

I would like to express my special thanks of gratitude to my supervisor, Associate Prof Dr Juliana Jalaludin for guiding, helping and advising me along the process in accomplishing this research. Huge thanks to my coordinator of final year project course, Dr. Ho Yu Bin who willing to guide and helping me in writing journal, thesis and data analysis. A lot of thanks for keeping me on the right track and being considerate in every single thing during coordinating this course.

Next, the completion of this research also could not have been possible without the cooperation facilities provided by school management, Laboratory of Environmental Health Science and Department of Environmental and Occupational Health, Universiti Putra Malaysia.

Last but not least, I would like to thank to all my family and friends who always giving me a moral support in order for me not to give up and keep on doing the best in every process during this research.

ABSTRACT

ASSOCIATION BETWEEN PM₁₀ AND PM_{2.5} WITH DNA DAMAGE IN EXFOLIATED BUCCAL MUCOSA CELL AMONG PRIMARY SCHOOL CHILDREN LIVING NEAR PALM OIL ACTIVITY AT SEMENYIH, SELANGOR

SYHIDATUL FARHANA OTHMAN

Introduction: The combustion of biomass material such as fibre and shell of fresh fruit bunch will release particulate matters PM₁₀, PM_{2.5}, carbon monoxide, nitrogen oxide, and sulphur dioxide which contributes to air pollution problem. Exposure of PM_{2.5} released from combustion of biomass activity caused respiratory health among primary school children. **Objective:** This research aimed to determine the association between exposure of PM_{2.5} and PM₁₀ with DNA damage in exfoliated buccal mucosa cells among primary school children living nearby palm oil combustion activity at Semenyih. **Methodology:** The study was conducted at two different primary schools where Sekolah Kebangsaan A was chosen as studied group and Sekolah Kebangsaan B as a comparative group. Those schools were selected based on the distance of school from palm oil factory. A total of 167 primary school children aged 9-11 years old were randomly selected from the list name for both groups to participate in this study. A standardized questionnaire was distributed to respondent's parents. Concentrations of PM_{2.5} and PM₁₀ were collected by using Dust Trak DRX Aerosol Monitor Model 8534 and Escort LC Personal Sampling Pump. Measurement of indoor and outdoor air pollutants were conducted in schools and home. Buccal mucosa cells were collected, which then followed by micronucleus assay. **Result:** Mean±SD concentration of PM₁₀, (104.29±58.70µg/m³) among studied group were significantly higher compared to comparative group, (29.29±16.69 µg/m³); (p=0.07). PM_{2.5} at homes of studied group was higher (74.29±42.57 µg/m³) compared to comparative group with (Median±IQR) (28.57±32.11 µg/m³); (p=0.018). PM₁₀ and PM_{2.5} of studied schools were significantly higher (105.21±46.11); (68.83±13.03) compared to comparative schools (53.92 ±12.37); (50.80 ±17.45). MN frequencies of studied group were significantly higher (Median±IQR) (4.00±3.00) compared to comparative group (0.50±1.00); (p=0.001). **Discussion:** There were significant difference of respiratory symptoms between two groups of respondents with cough, phlegm, wheezing and chest tightness as p=0.001. There were significant correlation between PM₁₀ with MN frequency of studied group and comparative group which indicated by $r = 0.562$; p=0.001. **Conclusion:** This research indicated that the exposure of PM₁₀ and PM_{2.5} would increase the risk of having respiratory health symptoms among primary school children who lived near palm oil activity area. In the other hand, the exposure of particulate matters might induce the micronuclei formation among affected children. Lastly, this study would contribute to the baseline data on air quality and the health effects of primary school children based on frequency of micronuclei and respiratory health symptoms.

Keywords: Primary school, Children, Particulate matters, Respiratory health, Micronuclei Frequency (MN)

ABSTRAK

HUBUNG KAIT DI ANTARA PENDEDAHAN KEPADA $PM_{2.5}$ DAN PM_{10} DAN KEROSAKAN DNA DALAM SEL BUKAL MUKOSA DI KALANGAN KANAK-KANAK SEKOLAH RENDAH BERHAMPIRAN KILANG MINYAK KELAPA SAWIT DI SEMENYIH, SELANGOR

SYHIDATUL FARHANA OTHMAN

Pengenalan: Pembakaran bahan biomas seperti serat dan cangkerang daripada gugusan buah segar akan mengeluarkan bahan partikel PM_{10} , partikel halus $PM_{2.5}$, karbon monoksida, nitrogen oksida, dan sulfur dioksida yang menyumbang kepada masalah pencemaran udara. Pendedahan terhadap $PM_{2.5}$ yang dikeluarkan daripada pembakaran aktiviti biomas menyebabkan masalah kesihatan pernafasan di kalangan kanak-kanak sekolah rendah. **Objektif:** Tujuan penyelidikan ini adalah untuk menentukan perkaitan antara pendedahan $PM_{2.5}$ dan PM_{10} dengan kerosakan DNA dalam sel bukal mukosa di kalangan kanak-kanak sekolah rendah yang tinggal berdekatan kawasan aktiviti pembakaran kelapa sawit di Semenyih. **Kaedah:** Kajian ini dijalankan di dua buah sekolah rendah yang mana Sekolah Kebangsaan A dipilih sebagai kumpulan kajian dan Sekolah Kebangsaan B sebagai kumpulan perbandingan. Sekolah-sekolah itu dipilih berdasarkan jarak sekolah dari kilang minyak kelapa sawit. Sebanyak 167 kanak-kanak sekolah rendah berumur 9-11 tahun dipilih secara rawak dari senarai nama bagi kedua-dua kumpulan untuk mengambil bahagian dalam kajian ini. Soal selidik yang diseragamkan telah diedarkan kepada ibu bapa responden. Kepekatan $PM_{2.5}$ dan PM_{10} telah diukur dengan menggunakan DRX Aerosol Dust Trak 8534 dan pam persampelan udara Escort LC. Pengukuran bahan pencemar udara dalaman dan luaran telah dijalankan di sekolah dan di rumah. Sel pipi diambil, dan kemudian diikuti oleh “micronucleus assay”. **Keputusan:** Purata $\pm$ SD kepekatan PM_{10} , ($104.29 \pm 58.70 \mu\text{g} / \text{m}^3$) di kalangan kumpulan kajian adalah lebih tinggi berbanding kumpulan perbandingan, ($29.29 \pm 16.69 \mu\text{g} / \text{m}^3$); ($p = 0.07$). $PM_{2.5}$ di rumah kumpulan kajian lebih tinggi ($74.29 \pm 42.57 \mu\text{g} / \text{m}^3$) berbanding kumpulan perbandingan dengan (Median $\pm$ IQR) ($28.57 \pm 32.11 \mu\text{g} / \text{m}^3$); ($p = 0.018$). PM_{10} dan $PM_{2.5}$ bagi sekolah kajian adalah lebih tinggi (105.21 ± 46.11); (68.83 ± 13.03) berbanding dengan sekolah perbandingan (53.92 ± 12.37); (50.80 ± 17.45). Frekuensi MN kumpulan kajian jauh lebih tinggi (Median $\pm$ IQR) (4.00 ± 3.00) berbanding dengan kumpulan perbandingan (0.50 ± 1.00); ($p = 0.001$). **Perbincangan:** Terdapat perbezaan yang signifikan bagi simptom pernafasan antara dua kumpulan responden iaitu batuk, kahak, berdesir dan sesak dada dengan nilai $p = 0.001$. Terdapat perkaitan yang beerti antara PM_{10} dengan frekuensi MN kumpulan kajian dan kumpulan perbandingan yang dapat ditunjukkan oleh $r = 0.562$; $p = 0.001$. **Kesimpulan:** Kajian ini menunjukkan bahawa pendedahan PM_{10} dan $PM_{2.5}$ akan meningkatkan risiko mengalami simptom penyakit pernafasan di kalangan kanak-kanak sekolah rendah yang tinggal berhampiran kawasan aktiviti pembakaran bahan biomas kelapa sawit. Sebaliknya, pendedahan terhadap partikel mungkin mendorong pembentukan mikronuklei di kalangan kanak-kanak yang terjejas. Hasil kajian ini akan menyumbang kepada data asas mengenai kualiti udara dan kesan kesihatan

kanak-kanak sekolah rendah berdasarkan kekerapan mikronuklei dan peningkatan gejala-gejala kesihatan pernafasan.

Kata kunci: Sekolah rendah, Kanak-kanak, Bahan partikulat, Kesihatan pernafasan, Frekuensi Micronuclei (MN)



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LIST OF ABBREVIATIONS

MN	Micronuclei
US EPA	United State Environmental Protection Agency
PM₁₀	Particulate matter 10
PM_{2.5}	Particulate matter 2.5
WHO	World Health Organization
CO	Carbon Monoxide
NOX	Nitrogen oxide
NO₂	Nitrogen dioxide
SO₂	Sulphur dioxide
MAAQS	Malaysia Ambient Air Quality Standard
ATS	American Thoracic Society
NIOSH	National Institute of Occupational Safety and Health
PBS	Phosphate Buffer Solution
HCl	Hydrochloric Acid
SPSS	Statistical Package for Social Sciences
DOE	Department of Environmental
APCT	Air Pollution Control Technology

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CHAPTER 1

INTRODUCTION

1.1 Background

Air pollution has become the most significant concern, and many epidemiological studies were carried out related to this issue. The pollutants in the ambient air can threaten human health. It is crucial to combat pollutant emissions by evaluating the characteristics of particulate matters at the different sources to protect both human and the environment (Aldabe et al., 2011).

According to Environmental Protection Agency (2013), the source of particulate matter 10 (PM₁₀) which also known as inhalable dust can be found near roadways and industries with diameters larger than 2.5 micrometres and smaller than 10 micrometres. Meanwhile, particulate matter 2.5 or fine particles can directly be emitted from sources such as forest fire or from when gaseous emitted from industries and they react with air.

World Health Organizations stated that, the polluted environment that occurred in term of physical, chemical and biological risk factors caused an estimated total of 12.6 million deaths recorded in the world. From the numerous deaths, 23% of total global mortality and 26% of deaths were among young children (Landrigan et al., 2018); Prüss-Ustün et al., 2016).

Industrial area was an area that consists of a lot of factories. It became one of the sources of pollutants toward population living nearby which can create air pollution. For instance, palm oil activities also generate air pollutants instead of liquid effluent and solid waste. Oil palm is one of the major crops that are planted in Malaysia. The plantation of oil palm was aggressively growing from early 1875. Malaysia also has become the second largest producer and importer of palm oil in the world (May, 2012). According to the Malaysia Palm Oil Industry, currently, Malaysia contributes 39% of world palm oil production and 44% of world exports. The crude palm oil production shows an increase of 15% reaching 19.92 million tonnes in 2017 compared to 17.32 million tonnes in the year 2016. Since it is one of the largest economic sectors in Malaysia, palm oil mill industry employed numerous numbers of people for working at the plantation and also palm oil mill factory.

Since Malaysia needs to produce the oils and fats according to world demand, every factory needs to increase its production which it will operate during the day and night. Each palm oil mill will have one boiler to carry out combustion process (Rashid et al., 2013). According to Rashid et al. (2013), approximately one ton of fresh fruit bunch create about 14%-15% fibre and 6-7% shell to be burned as to generate energy for the palm oil mill. Despite the excellent benefit of waste produced, the combustion of biomass material released particulate matters which contribute to the air pollution problem (Kun & Abdullah, 2013).

Particulate matters such as PM_{10} and $PM_{2.5}$ are one of the contributing factors that can cause adverse effect toward human health especially to our respiratory system. Respiratory and cardiovascular morbidity such as aggravation of asthma, respiratory symptoms and an increase in hospital admissions are some of the health effects due to exposure of particulate matter in short and long term (WHO, 2013).

1.2 Problem Statement

Air pollution has become a significant threat towards human life especially children who lived near the palm oil industry. Particulate matters become the greatest harmful pollutant compared with the others. Particulate matters were comprised of all sums of solid and liquid particles which suspended in the air. It will form a complex mixture of organic and inorganic particles such as dust, pollen and liquid droplets (US EPA, 2003). Based on the United States Environmental Protection Agency (US EPA) (2003), these particles were inhaled; it can cause an adverse effect on heart and lung. The impact can be worsening depending on the duration of exposure and concentration

of inhalable coarse particles (PM₁₀) and fine particles (PM_{2.5}). Since Malaysia is one of the largest producers of palm oil, the production rate of mills will increase, and the emission of particulate matter subsequently will elevate. These particulate matters may enter the human body mainly through inhalation, dermal absorption and ingestion.

Particulate matter emission consists of PM₁₀ and PM_{2.5}, where it small enough to penetrate the thoracic region of respiratory systems. It will become more dangerous if it persists in the ambient air for an extended period. A susceptible group such as elderly, pregnant women, the person who is suffering from any respiratory health problem and children are prone to have higher chances of getting any respiratory problems when inhaling those particulate matters. The person who had a history of suffering respiratory problem is also faced difficulty in breathing and even worsening their health condition. Exposure to air pollutant at early age can caused impair of lung development, reduce lung function and increase risk of choric lung disease in adulthood (WHO, 2018). These are due to immune system and under-develop organs of children. Children breathe more air relative to their weight and lung surface than the adult does. Exposure to air pollutants concentration such as particulate matters gave an association with prevalence of respiratory health symptoms among children living in high polluted area (Langkulsen, Jinsart, Karita, & Yano, 2006). Therefore, children become an interest group in this study as it believed that this group were most affected.

Based on Industrial Processes & The Environment (Handbook No.3) Crude Palm oil Industry, (1999), palm oil mill can become a serious source of public

complaint when the mill located close to the residential area, and the emission of air pollutants was poorly controlled. Palm oil mill at the study location was nearly located in the residential area as well as the schools. Thus, the activities from the palm oil mill might become one of the contribution factors of causing respiratory problems especially among children.

Although the palm oil mill was established first before the residential area, due to poor control of air emission, it will affect the residential area. This will cause the concentration level of outdoor air pollutant at the study area become a great question. The burning of biomass material from the palm oil mill resulted in a decrease in the quality of ambient air at the nearby location. Children who are exposed to the particular level of PM_{10} and $PM_{2.5}$ that some time would be higher than stipulated in the regulation can show some respiratory symptoms such as chest tightness, wheezing, coughing and phlegm. So, it is essential to find out the respiratory health status of the children who live in the affected area, since the condition is still unknown.

From the previous study by Neri et al., (2006), the exposure of particulate matter such as PM_{10} and $PM_{2.5}$ can be indicated by the frequency of micronuclei (MN) as the biomarkers of exposure. MN assay in children is increasingly used because of its public health significance for monitoring environmental exposure such as pollution, radiation, industrial toxicants and various medical treatments (Holland et al., 2011). It has also been used to assess genotoxicity associated with diseases such as leukaemia, Down's syndrome, genetic conditions with DNA repair deficiencies, as well as chronic conditions such as asthma and inflammatory bowel disease (Holland, et al., 2011).

From this study, the concentration of PM₁₀ and PM_{2.5} was measured, and at the same time, the frequency of micronuclei (MN) was determined by collecting of buccal mucosa cells from cheeks of each respondent. Therefore, the result of this study will increase awareness among the community regarding health implication due to air pollutant exposure. Other than that, the authorised people need to take a quick and appropriate action to tackle this problem before it becomes worst.

1.3 Study Justification

This study is vital to carry out as the particulate matter PM_{2.5}, and PM₁₀ pose a threat to the health of the vulnerable group which is children. In this study, the exposure of PM_{2.5} and PM₁₀ will be determined and its association with DNA damage in exfoliated buccal mucosa cells among primary school children in Semenyih, Selangor. The parents should be more aware of the exposure that their children faced during school time. Besides, awareness among the community is hoped to be increased through this study. Therefore, to increase the awareness and also to fill the gap on knowledge about the health implication, the information should be widely disseminated to the community. Other authorities such as Department of Environment is hoping can organise conferences, workshops, talks and campaigns on the importance of clean environment toward the industrial company as well as to the community.

From the previous study by Farah and Juliana (2016), prove that children among the studied group are more prone to get the cough, phlegm, wheezing and chest tightness compared to the control group. Thus, this study will be conducted to find the

evidence that the palm oil is emitting particulate matters that are hazardous towards the health of children and determine the genotoxicity effects from the particulates matters. Besides, this study may create baseline data and reference for the future use of researchers when confronting the same problems.



1.4 Conceptual Framework

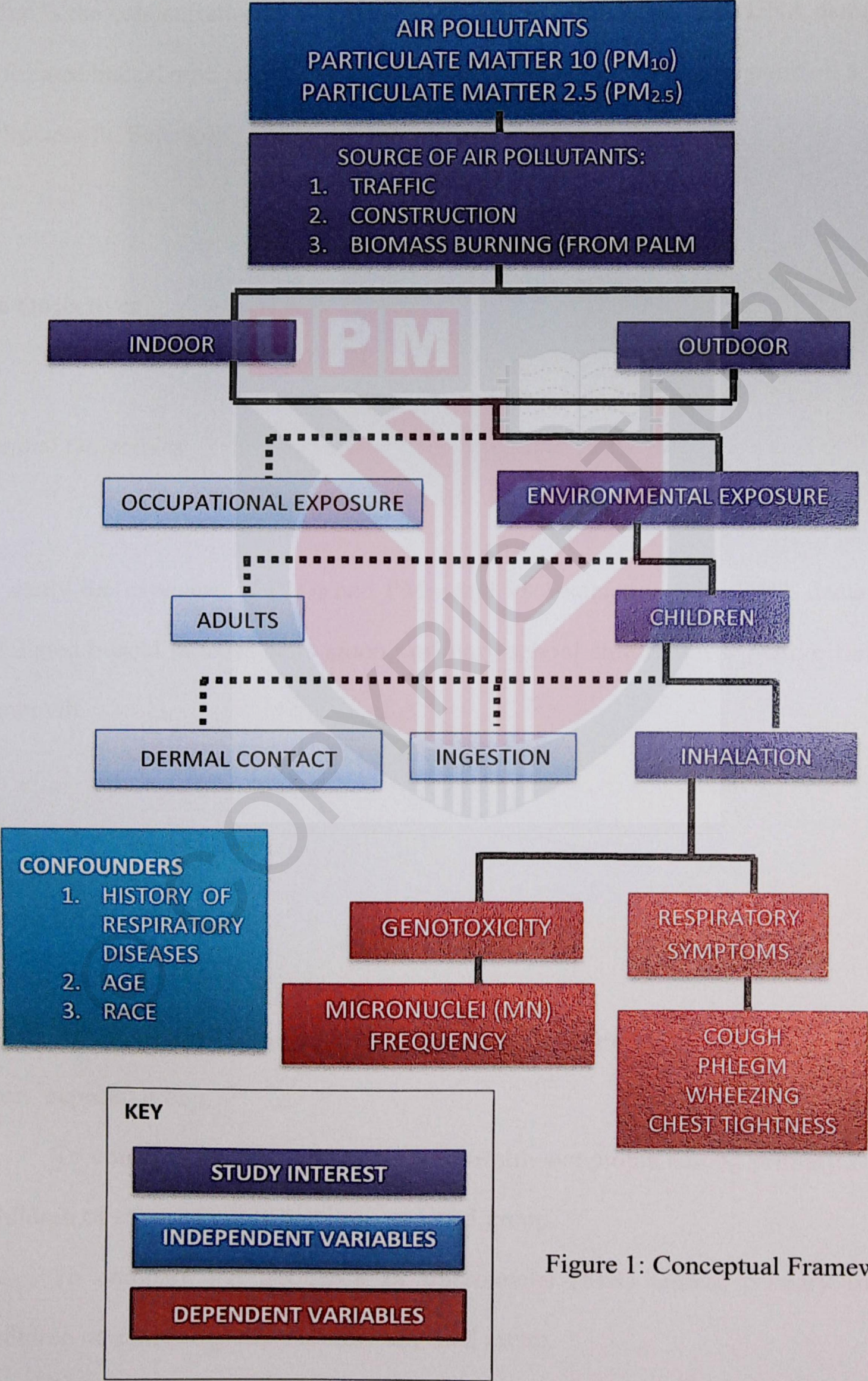


Figure 1: Conceptual Framework

1.5 Research Question

What is the concentration of PM₁₀ and PM_{2.5} and its association with DNA damage in exfoliated buccal mucosa cells among primary school children nearby palm oil activity at Semenyih, Selangor?

1.6 Objectives

General Objectives

To study the exposure of PM₁₀ and PM_{2.5} and its association with DNA damage in exfoliated buccal mucosa cells among primary school children near palm oil mill at Semenyih.

Specific Objectives

- 1. To compare the concentration of PM₁₀ and PM_{2.5} exposure among exposed and non- exposed group.**
- 2. To compare the reported respiratory health symptoms among primary school children of exposed group and non- exposed group.**
- 3. To compare the frequency of micronuclei (MN) among primary school children of exposed group and non- exposed group.**

4. To determine the association between PM₁₀ and PM_{2.5} with respiratory health symptoms among studied group.
5. To determine the association between exposure of PM₁₀ and PM_{2.5} with the micronuclei (MN) frequency among exposed and non- exposed group.

1.7 Hypothesis

1. The concentration of PM₁₀ and PM_{2.5} exposure is significantly higher among exposed than non- exposed group.
2. The reported respiratory symptoms among primary school children of exposed group are significantly higher than non- exposed group.
3. The primary school children of exposed group having higher micronuclei (MN) frequency than non- exposed group.
4. There is a significant association between exposure of PM₁₀ and PM_{2.5} with respiratory health symptoms among studied group.
5. There is a significant association between exposure of PM₁₀ and PM_{2.5} with the micronuclei (MN) frequency among exposed and non- exposed group.

1.8 Definition of Variables

Conceptual Definition

Particulate Matter 10 (PM₁₀)

Particulate matter 10 is also known as inhalable particles with aerodynamic diameter less than 10 micrometres and smaller. According to EPA (2013), the source of inhalable dust can be found near roadways and industries which are larger than 2.5 micrometres and smaller than 10 micrometres in diameters.

Particulate matter 2.5 (PM_{2.5})

Particulate matter 2.5 is also known as fine particles with aerodynamic diameter less than 2.5 micrometres. These particles can be easily penetrated deep for a long period of time into lung and bloodstream and led to serious health problems. These particles are directly emitted from sources such as forest fire or they form when gaseous are emitted from industries and they react with air. (United States Environmental Protection Agency, 2013).

Primary school children

Primary school children are referring to children who are between the ages of 7 to 12 years old (Education in Malaysia, 2015).

Buccal mucosa cells

The inner lining of the cheeks and lips, which is an anatomic region that includes all the mucous membrane lining of the inner surface of the cheeks and lips, from the line of contact of the opposing lips to the line of the attachment of mucosa to the alveolar ridges and pterygomandibular raphe that occupies an area of 50-60cm² (Mosby's Medical Dictionary, 9 th Edition, 2009).

Micronuclei (MN)

Micronuclei formed after irradiation represents genetic material which will lost from the genome of the cells. (Midander and Revesz,1980)

Operational Definition

Particulate Matter 10 (PM₁₀)

The exposure of particulate matter 10 was measured by using personal air sampling pump. The particulate matter can be filtered by using Poly Vinyl Chloride (PVC) filter with the aid of Aircheck 52 personal sampling pump. The flow rate of pump is depending on the type of cyclone (nylon or aluminium), 1.7L/min or 2.5L/min respectively. The concentration of PM₁₀ can be determined by using published formula such as NIOSH Manual of Analytical Method. The concentration of PM₁₀ will be obtained in µg/m³ after calculated by using the respective formula.

Particulate matter 2.5 (PM_{2.5})

The exposure of particulate matter 2.5 was measured by using personal air sampling pump. The particulate matter can be filtered by using Mixed Cellulose Ester (MCE)

filter with the aid of Aircheck 52 personal sampling pump. The flow rate of pump is depending on the type of cyclone (nylon or aluminium), 1.7L/min or 2.5L/min respectively. The concentration of PM_{2.5} can be determined by using published formula such as NIOSH Manual of Analytical Method. The concentration of PM₁₀ will be obtained in $\mu\text{g}/\text{m}^3$ after calculated by using the respective formula.

Primary school children

Primary school children who study at exposed and non-exposed area between the ages of 9 to 11 years old were recruited in this study.

Buccal mucosa cells

The biological samples (buccal mucosa cells) were collected in the inner surface of both cheeks by using toothbrush. This buccal mucosa collection was performed by medical laboratory technologist.

Micronuclei frequency

The collected buccal mucosa cell will be smeared and stained and then, the slides will be examined at 1000X magnification for microscope analysis (Ceretti et al. 2014). Micronuclei frequency was calculated by using scoring method. (Thomas, Holland, Bolognesi et al, 2009).

CHAPTER 2

LITERATURE

2.1 PARTICULATE MATTER 10 (PM₁₀) AND PARTICULATE MATTER 2.5 (PM_{2.5})

According to the United State Environmental Protection Agency (USEPA), particulate matter can be classified based on their sizes such as particulate matter 10 and particulate matter 2.5. Particulate matter (PM₁₀) is also known as inhalable particles with aerodynamic diameters less than 10 micrometres. These particles are emitted from a source such as construction sites, smokestacks and unpaved roads. However, most of the particles in the atmosphere are produced from complex reactions of chemicals which are emitted from industries, power plant and automobiles. Particulate matter (PM_{2.5}) can also be called as fine, inhalable particles, with aerodynamic diameters smaller than 2.5 micrometres.

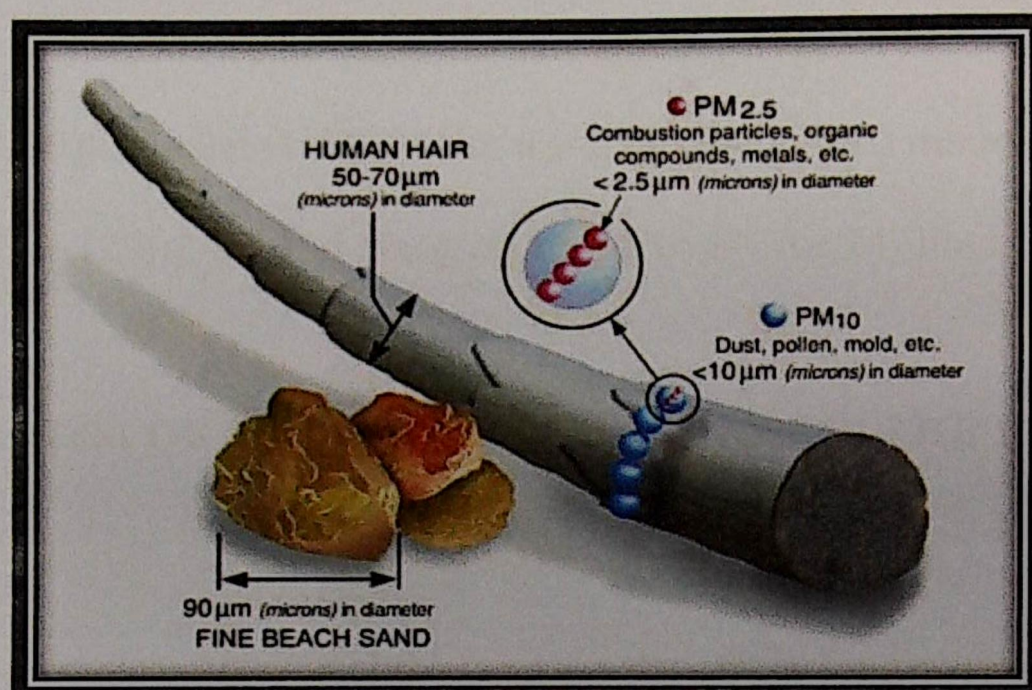


Figure 2: Size of Particulate Matter

Source: EPA, 2013

These particles can penetrate deeply into our lung and even into our bloodstream. According to (Whyand, Hurst, Beckles, & Caplin, 2018), the penetration of particulate matter into the thoracic regions or lower respiratory tracts can cause asthma and respiratory morbidity.

The particulate matter released from several sources of air pollutants especially $PM_{2.5}$, which can penetrate profoundly into the respiratory tract, increasing genotoxicity action and it can be transported for thousands of kilometres through the atmosphere and it is persistent in the ambient air for a long time (Silva da Silva et al., 2015). Based on Pierse et al. (2006), it showed that there was a significant association between exposures to PM_{10} at home with the prevalence of coughing. Exposure to particulate matters was associated with increased respiratory symptoms (Tiittanen et al., 2006).

Based on New Malaysia Ambient Air Quality Standard (MAAQS), the air pollutants limit of particulate matter with the size less than 10 micron and particulate matter (PM_{2.5}) are 120µg/m³ and 50µg/m³ respectively for 24- hours.

2.1.1 MECHANISM OF PARTICLES DEPOSITION IN THE LUNG

The mechanism of particles deposition can be classified into three main mechanism which are inertial impaction, gravitational sedimentation and Brownian diffusion (Darquenne, 2006).

Inertial impaction

The inhaled particles will follow the complex path through respiratory tract. Inertial impaction is the initial deposition for the particles which is larger than 5 micrometres in diameter, it also an important deposition for the particles as small as 2 micrometres. The impact of deposition are likely to deposited at upper respiratory tract (that extends from the nasal and mouth cavities to the entrance of trachea) and also at the conducting airway bifurcations (Darquenne, 2006).

Gravitational Sedimentation

This mechanism refers to the settling of particles because of the action of gravity. Terminal settling velocity of particles is reached when the gravitational force are equals to the opposing viscous resistive force of the air. Particles with diameter range

between 0.5 and 5 micrometres will be deposited at the upper respiratory tract and also at alveoli (Darquenne, 2006).

Brownian Diffusion

Deposition by Brownian diffusion is resulted from the random motions of the particles that caused by their collision with gas molecules. The deposition by Brownian diffusion involves particles with diameter less than 0.5 micrometres. It mainly occurs in the acinar region of the lung and also significantly deposited in the nose, mouth and pharyngeal airways (Darquenne, 2006).

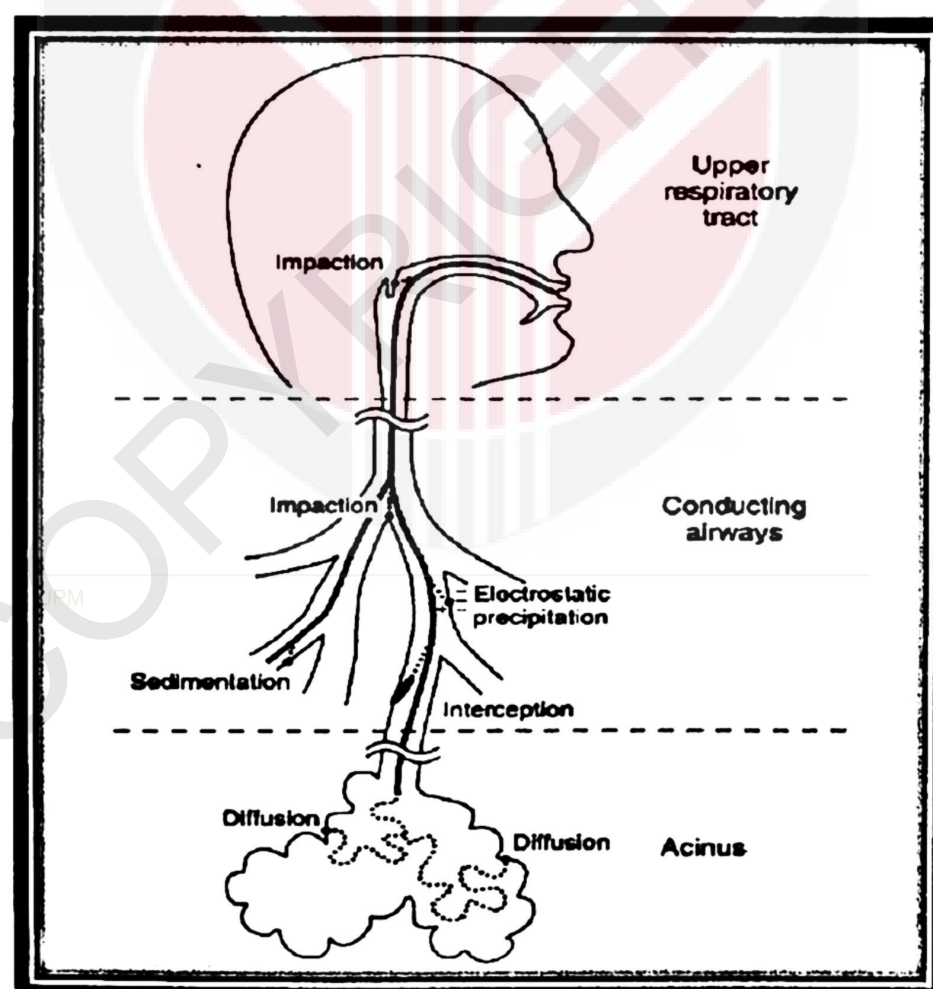


Figure 3: Mechanisms of deposition of inhaled particles in the respiratory tract. The mechanisms of deposition are not restricted to the zone of the lung as shown in the figure. For example, deposition by sedimentation can also occur in the acinus.

Source:(Darquenne, 2006)

2.2 PARTICULATE MATTER 10 (PM₁₀) AND PARTICULATE MATTER 2.5 (PM_{2.5}) FROM PALM OIL ACTIVITY

The production of palm oil in Malaysia is running smoothly without having any issues of generating electrical energy. In order to produce electricity, they are using energy efficient approach by which a million tonnes of waste comprise of fruit bunches, fibre and palm shell act as sources of fuel. However, the combustion of waste as a boiler fuel created a harmful emission in the industry (Kun & Abdullah, 2013). Rashid, Ramli and Rozainee (1998) stated that the combustion from palm fibre could release a massive amount of particles in the form of fly ash to the air.

The recent study reveals that the emissions of pollutants from the boiler give significant health and environmental impact. Based on Hanafi, Hashim and Yusof, (2016) they stated that particulate matters (PM), carbon monoxide (CO), nitrogen oxide (NOX) and sulphur dioxide (SO₂) are the example of pollutants emitted from the combustion of biomass material which could pose a dangerous threat toward human and environment. This shows that the composition of pollutants bring harmful effect especially to the human health. Moreover, the incomplete combustion of solid fuels also creates a problem as it releases dark smoke and the carryover of artificially carbonised fibrous particulates (Yusoff, 2006).

According to the World Health Organization (2013), the black carbon which is part of PM_{2.5}, produced from incomplete combustion was identified as a contributing factor on air quality to detrimental effects on health and also climate.

2.3 CHILDREN AS A VULNERABLE GROUP

The exposure to air pollution in children was different from the adults. Children are particularly vulnerable to exogenous factors such as air pollution (Liu et al., 2018). A study by Heft-Neal et al., (2018) stated that children that exposed to high risk of pollution even at low level air pollutants could affect their health for the whole time of their life. This was because based on Pedersen et al. (2006), they have higher levels of physical activity and spend their time more on outdoor air which pose higher tendency of exposure toward outdoor air pollution.

Moreover, children are more vulnerable than adults due to physiological differences based on body size, growth, development and immaturity of organs as well as body functions (Pedersen et al., 2006). The routes of exposure of air pollution are significantly through inhalation. Based on (Holland et al., 2008), the first barrier for the inhalation or ingestion is buccal cells which capable of metabolizing proximate carcinogens to reactive products.

Risk is dependence in both present of sensitivity and also exposure. Children of all ages pose a greater risk than adults even if they are exposed to chemical at below level than an adult (Scheuplein, Charnley, & Doursont, 2002).

2.4 MICRONUCLEI (MN) STUDIES IN CHILDREN

Micronucleus test was an efficient cytogenetic method by providing a reliable index of chromosomal break or loss (Fenech & Bonassi, 2011). Moreover, it was

widely being used to investigate environmental samples and proved to be an effective tools for the assessment of airborne particulate matter.

The formation of extra- nuclear bodies from breakage of chromosome or the whole chromosome does not reach the spindle poles during cell division are called as micronuclei (MN). After failure of mitotic spindle, the micronuclei will be formed, thus it will lead to misattachments of microtubules. (Gina, Azeddine, Lukamowicz, & Gonzalez, 2011). The micronuclei can be identify based on (Thomas & Fenech, 2011). First and foremost, it contains both main nucleus and micronucleus; micronuclei are round or oval with similar intensity of stain as main nucleus. Next, the micronuclei can be determined by its size, where it have $1/3$ - $1/16$ diameter of main nucleus and lastly it must be located in the cellular cytoplasm.

One of the benefits of this method is non-invasively toward children. The collection of buccal cells is arguably the least invasive method available for measuring DNA damage in humans, especially in comparison to obtaining blood samples for lymphocyte and erythrocyte assays or tissue biopsies (Holland et al., 2008).

Studied performed by using approach of genetic monitoring and mutagenicity have become a major tool, especially in finding the area of references. This studied aim to establish baseline values which can be used as comparison with the studied performed in area where there are affected from anthropic activities (Silva da Silva et al., 2015).

Based on Thomas et al. (2009), the cells with multiple MNi was common in person who are exposed to radiation or other genotoxic agents but rare in healthy individuals. The MN index in rodent and/or human cells has become one of the

standard cytogenetic endpoints and biomarkers used in genetic toxicology *in vivo* or *ex vivo*. Besides, in human MNi can be easily assessed in erythrocytes, lymphocytes, and exfoliated epithelial cells (e.g. oral, urothelial, nasal) to obtain a measure of genome damage induced *in vivo* (Holland et al., 2008).

Based on Sisenando et al., (2012), it stated that, they found significant evidence in which increased exposure to PM_{2.5} can generates an increased micronuclei frequency in oral epithelial cells from school children.

Studied from Silva da Silva et al.,(2015), found that there were significant difference between the induction of MN and passive smoking with value of $p=0.021$, where this can be conclude that children who live with smokers are at increased risk of presenting a higher MN value through the analysis of generalised linear models with Poisson distribution.

Particulate matter (PM₁₀) was associated with mutagenic activities, as it presence in gas and particulate phase. It gave a significant health impact as the chemical was deposited in an organ that may cause *in vivo* induction of somatic cell mutation and carcinogenesis (Soares et al., 2003). Based on study by Coronas (2009), there were increased level of DNA damage was detected in the comet assay in subjects who are working or living near an oil refinery compared with people living in area with restricted traffic and industries.

CHAPTER 3

METHODOLOGY

3.1 STUDY DESIGN

This study was a cross sectional comparative study. This study was aimed to determine the association of exposure to PM₁₀ and PM_{2.5} in the ambient air with DNA damage in exfoliated buccal mucosa cells among primary school children who lived near to the palm oil factory while, the comparative group was among the primary school children who lived far away from the industrial area.

3.2 STUDY LOCATION

This study was taken place at two areas where the polluted area is at Semenyih while the comparative area is at Hulu Langat. At the polluted area, a primary school denoted as Sekolah Kebangsaan A was selected in this study, where it located near to the palm oil factory. For the comparative area, Sekolah Kebangsaan B was selected where it located far from palm oil factory. Residents who lived at the residential area located

near palm oil factory were expected to have high risk of exposure toward PM_{10} and $PM_{2.5}$ released from palm oil activities itself.

3.3 STUDY DURATION

This study was conducted from February 2019 until June 2019. The important things that need to be considered during data collection are the weather. This was because if the data collected during rainy day, the data obtained was not accurate and not represents the actual concentration of particulates in that area.

3.4 STUDY POPULATION

The study population were among primary school children who studied at Sekolah Kebangsaan A which located within 2.7 km from the palm oil. According to Department of Environment (2009), the zone of impact taken an area of 5km radius from the site and it depend on the source strength and also the pollutant dilution effect. $PM_{2.5}$ can persist at the atmosphere for a few days and even for a few weeks. The radius dispersion of particulate matter can travel up to hundreds to thousands kilometre. In addition, PM_{10} can travel in the atmosphere for less than one kilometre to hundreds kilometre (Dalglish et al., 2007). Meanwhile, the population for comparative group were among primary school children who studied at Sekolah Kebangsaan B which

located about 40km away from the palm oil area. Figure 3 and 4 showed a location of Sekolah Kebangsaan A.

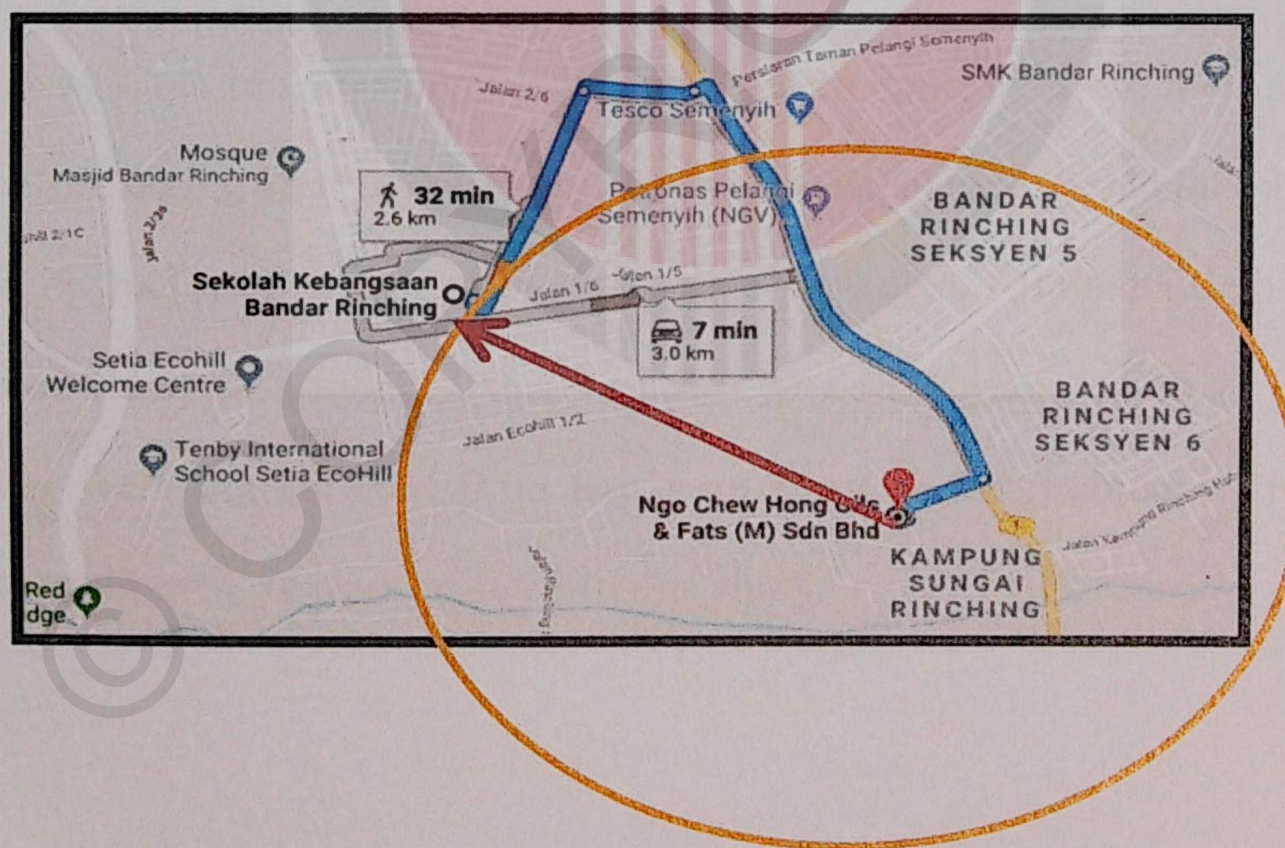
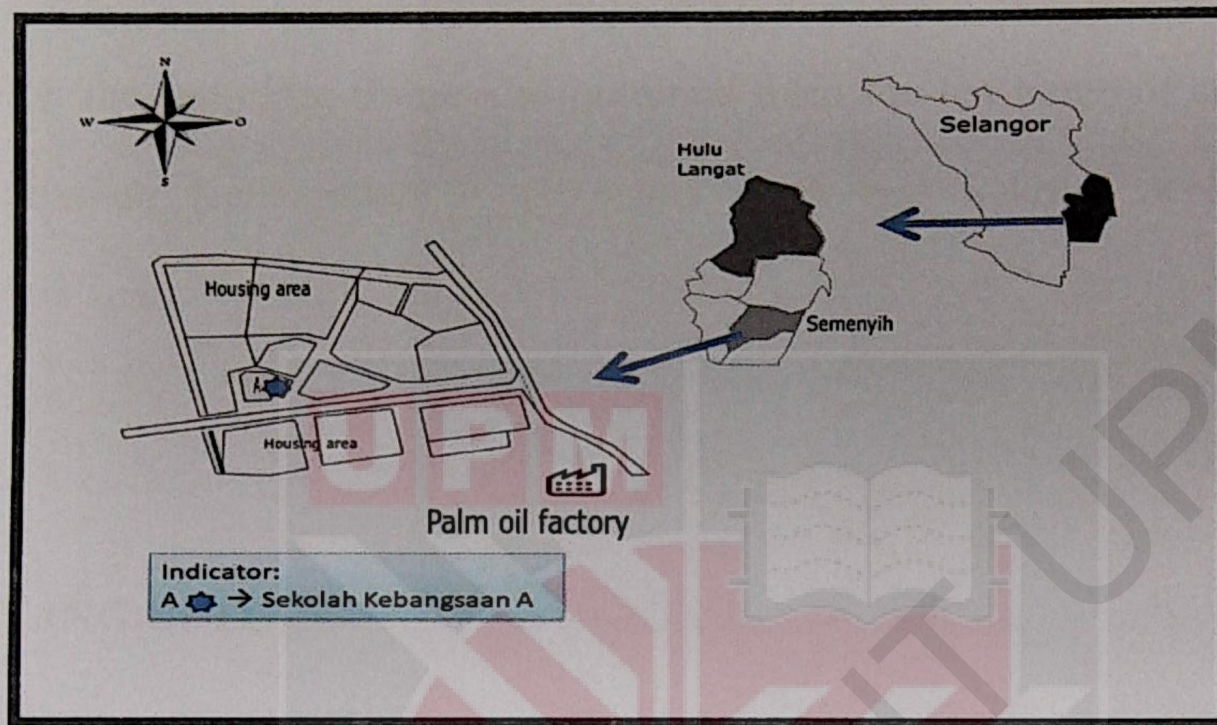


Figure 4 and 5: Location of Sekolah Kebangsaan A.

Source: Google maps

3.5 SAMPLING FRAME

In this study, the sampling frame was obtained from the list name of students who attended Sekolah Kebangsaan A (Exposed area) and Sekolah Kebangsaan B (Comparative area).

3.6 SAMPLING UNIT

The study samples were among students at Sekolah Kebangsaan A and Sekolah Kebangsaan B. Children who involved were given an approval from their parents or guardian before taking parts in this study. The students that involved in this study were fulfilled all the inclusion criteria as to meet the requirement. All the participants were the citizen of Malaysia. Secondly, their age was between 9 years old to 11 years old. Finally, they were not suffered from any respiratory health problems. Those who fulfilled all of the criteria were selected in this study.

3.7 SAMPLE SIZE

Based on Lemeshow, Klar and Lawanga (1990), the sample size was calculated by using the following formula:

$$n = \frac{\{z_{1-\frac{\alpha}{2}}\sqrt{2\bar{p}(1-\bar{p})} + z_{1-\beta}\sqrt{P_1(1-P_1)} + P_2(1-P_2)\}^2}{(P_1 - P_2)^2}$$

Where,

$$\bar{p} = (P_1 + P_2)/2$$

P_1 = estimated proportion (larger)

P_2 = estimated proportion (smaller)

$P_1 = 0.66$ (Rinne et al., 2006)

$P_2 = 0.45$ (Rinne et al., 2006)

$$\bar{p} = 0.555$$

$$Z_{1-\alpha} = 1.96$$

$$z_{1-\beta} = 0.842$$

$$n = \frac{\{1.96\sqrt{2(0.555)(1-0.555)} + 0.842\sqrt{0.66(1-0.66)} + 0.45(1-0.45)\}^2}{(0.66 - 0.45)^2}$$

$$n = 77$$

The sample size for each group (children in Semenyih and Hulu Langat) would be 77.

Therefore, the total number of sample size is 154.

3.8 SAMPLING METHOD

The schools were selected using purposive sampling. The students were selected randomly from the list name of student of each school. There were 85 respondents represented from each exposed group and also control group, which gave a total of 1170 respondents (boy and girl). Based on the list name provided by the teachers, every student has an equal potential to be selected in this study.

However, they were only 167 respondents who were participated in this study. The recruitment of all respondent was based on the inclusion criteria while the respondents who meet the exclusion criteria were not selected in this study. The inclusion and exclusion criteria were listed below;

Inclusion criteria:

- ✦ Malaysian citizen
- ✦ Primary school children aged between 9 to 11 years old
- ✦ Do not have any reported disease related to respiratory system such as asthma, bronchitis, emphysema and pneumonia.
- ✦ Primary school children without malignant tumours and who not undergone radiotherapy or chemotherapy in the previous 12 months or X-rays in the previous 3 months (Ceretti et al., 2014).

Exclusion criteria:

- ✚ Primary school who does not get the consent from parents or guardian to participate in this study.

3.9 INSTRUMENTATION

3.9.1 QUESTIONNAIRE

Parents answered the questionnaire so that the data regarding the history of respiratory symptoms and disease, demographic data, as well as the frequency and extent of the physical activity of children were obtained. The questionnaire was a structured questionnaire and it was adapted from American Thoracic Society (Questionnaire ATS –DCD-78C for children) and questionnaire from American Academy of Paediatrics. The ATS-DLD-78 was based on the Medical Research Council questionnaire with extensive question on asthma and wheezes were included. Other than that, to avoid information bias pre-test was performed on 10% of sample size.

3.9.2 MEASUREMENT OF PM₁₀ AND PM_{2.5} AT SCHOOL

Measurement of PM₁₀ and PM_{2.5} was taken at the back of classrooms of Sekolah Kebangsaan A and Sekolah Kebangsaan B. The measuring procedure was conducted during school hours. The measurement of PM₁₀ and PM_{2.5} was measured using DustTrak Aerosol Monitor Model 8534. The DustTrak was placed on the desk with the same level of children's breathing zone, where 1 meter above from ground and 1.5 meter away from windows and door. The measurement was taken from 7.20 am until 2.00 pm (6hours).

Measurement Points were selected at points that can avoid any disruption due to movement and also to avoid attraction from the children. Based on TSI Incorporate, the DustTrak was using scattering technology in order to determine the mass concentration in real- time. The DustTrak monitors have laser diode which has wavelength 780 nanometres (nm) and can limit the detection of particle until 0.1micrometer (µm).

3.9.3 MEASUREMENT OF PM₁₀ AND PM_{2.5}, AT RESPONDENTS' HOUSE

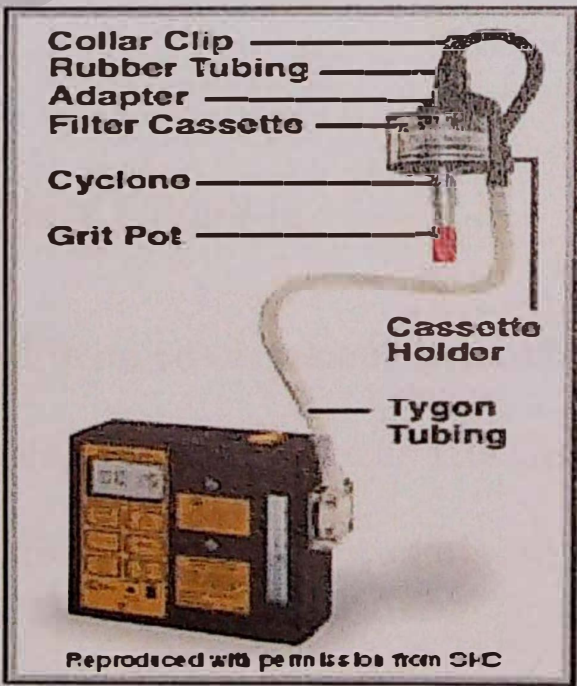
Based on NIOSH method 0600, the concentration of PM₁₀ and PM_{2.5} was done by using gravimetric technique. This technique was performed at the children's house. For this measurement, personal sampling pump by Zefon was used.

The pump was operated for 24 hours to measure PM₁₀ and PM_{2.5} in the air, and it was placed at the same level of children's breathing zone. Filter paper of Poly Vinyl Chloride (PVC) was used to measure PM₁₀ with pore size of 5µm. In addition, filter paper of Mixed Cellulose Ester Membrane (MCE) filter was used to measure PM_{2.5} with pore size of 5µm. The flow rate was set at 1.7 L/min. (NIOSH Manual of Analytical Method, fourth edition).

PARTICULATES NOT OTHERWISE REGULATED, RESPIRABLE			0000
DEFINITION: aerosol collected by sampler with 4-µm median cut point			CAS: None
			RTECS: None
METHOD: 0600, Issue 3		EVALUATION: FULL	Issue 1: 15 February 1984 Issue 3: 15 January 1990
NIOSH: 5 mg/m ³ NIOSH: no RD ACGM: 3 mg/m ³		CONFOUNDS: residues on solutions and quantity less than 1% nonrespirable nonregulated particles of respiratory irritants	
SYNONYMS: nonrespirable dusts, particulates not otherwise classified			
SAMPLING		MEASUREMENT	
SAMPLER:	CYCLONE + FILTER (10-mm nylon cyclone, Higgins-Dewitt (HD) cyclone, or Aluminum Oxide-coated 5-µm PVC membrane)	TECHNIQUE:	GRAVIMETRIC (FILTER WEIGHT)
		ANALYTE:	mass of respirable dust fraction
		BALANCE:	0.001 mg sensitivity; use same balance before and after sample collection
		CALIBRATION:	National Institute of Standards and Technology Class E 11 or ASTM Class 1 weights
FLOW RATE:	nylon cyclone: 1.7 L/min HD cyclone: 2.2 L/min Al cyclone: 2.8 L/min	RANGE:	0.1 to 2 mg per sample
VOLUME: -MAX:	20 L @ 5 mg/m ³ 400 L	ESTIMATED LOD:	0.03 mg per sample
SHIPMENT:	routine		

Figure 6: NIOSH Method of Particulates not Otherwise Regulated, Respirable 0600.

Source: NIOSH Method, Fourth Edition



A typical sampling train

Figure 7: Personal Air Sampler

Source: General Introduction to Occupational Health: Occupational Hygiene
Website.



Figure 8: TSI DustTrak DRX 8534 Handheld Particulate Monitors for Mass
Concentration of Aerosols and Total PM Size.

Source: TSI Website

Procedure

Preparation of Sampler before Sampling

- i. The filters were weighed in an environmentally controlled area.
- ii. The filters were assembled in the cassettes. The Cassettes was closed firmly to avoid leakage around the filter. A plug was placed in each opening of the filter cassettes.

- iii. Before used the cassettes, the cyclone's gift cap was removed and the cyclone interior was inspected. The interior of the cyclone will be cleaned to prevent re-entrainment of large particles.
- iv. The sampler head was assembled. The alignment of filter holder and cyclone in the sampling head was checked in order to prevent leakage.

Sampling

- i. Each personal sampling pump was calibrated to the appropriate flow rate (1.7 L/min) with a representative sampler in line.
- ii. Sample was taken for 24 hours at the respondent's house and it was placed at the living room and children's bedroom where the usual place for children stayed most of their time.
- iii. The top and bottom cap was removed from the filter cassette. It was equilibrated for at least 2 hours in an environmentally controlled area or chamber.

Quality control

- i. According to the procedure, each of the filter paper was weighed before it placed inside the cassette; the weight of filter papers was then recorded.
- ii. Electrostatic discharged use
- iii. d on the filter papers to eliminate electrostatic bias.
- iv. The microbalance was re-zeroed before all weighing. The same microbalance was used to weight the filter papers before and after sample collection.

- v. The set of replicate field samples was exposed to the same dust environment. The quality control samples were taken with the same equipment, procedures and personnel used in the routine field samples.
- vi. Each sampling pump was calibrated by using calibration jar before sample collection.

Measurement

- i. Each filter, including field blanks was weighed. Beside its corresponding tare weigh, its post-sampling was weighed and recorded. Anything remarkable about a filter was recorded (example: visible, overloading, leakage, wet, ton, etc).

Calculation

The concentration of respirable particulate was calculated, C ($\mu\text{g}/\text{m}^3$), in the air volume sampled, V (L):

$$C = \frac{(W2 - W1) - (B2 - B1)}{V} \cdot 10^3, \text{ mg}/\text{m}^3$$

Where: $W1$ = tare weigh of filter before sampling (mg)

$W2$ = Post sampling weight of sample-containing filter (mg)

$B1$ = Mean tare weight of blank filters (mg)

$B2$ = mean post-sampling weight of blank filter (mg)

V= Volume as sampled at the nominal flow-rate

3.9.4 Anthropometry Measurement

Anthropometry measurement involves the measurement of height and weight of the respondents. Each of the respondent's height and weight was measured.

3.9.5 SECA Body Meter

SECA Body Meter was used to measure the height of the respondents. In order to measure the height of the respondents, they were asked to stand in a straight position and with bare footed.

3.9.6 SECA Weighing Scale

SECA Weighing Scale was used to measure the weight of the respondents in kilogram. The respondents were required to take off their shoes so that the measurement can be done on a flat surface area, so that the measurement was accurate.

3.10 COLLECTION OF BIOLOGICAL SAMPLES

According to (Villarini et al., 2018), the children need to rinse their mouth twice with natural mineral water before sample collection. The sample of buccal mucosa cells was collected using cytobrush which has been autoclaved and it was performed by medical laboratory technologist.

Epithelial buccal cells were gently scraped for 10 times in a circular motion by using small-headed toothbrush in the inner surface of cheeks. Then, the head of the brush was immediately dipped into micro-centrifuge tubes containing 0.1M phosphate buffer solution (PBS) (pH7.5). The samples was stored at -20°C and rotated repeatedly in order to dislodge and release the cells into the buffer.

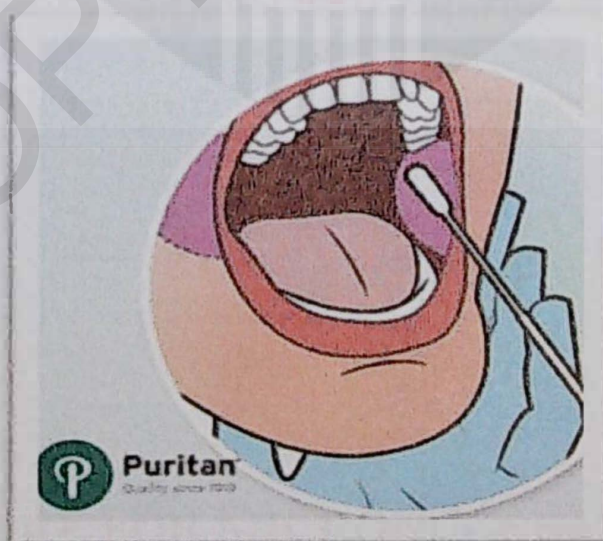


Figure 9: Collection of epithelial buccal cells

Source: Puritan Medical Product Website

The epithelial buccal cells in 0.1M phosphate buffer solution (PBS) (pH7.5) were centrifuged and washed twice with PBS. Then the samples was smeared on the glass slides and leaved overnight in room temperature (27°C) at the Environmental Laboratory, Universiti Putra Malaysia, for the next processing such as staining preparation and scoring.

3.10.1 STAINING OF BUCCAL MUCOSA SMEAR PREPARATIONS

The cell smears were stained with the Feulgen reaction according to the following procedure, the cell were fixed with 1% gluteraldehyde in cold 0.1M PBS for 20 minutes and pre-treated with 5N HCl for 30 minutes. After that, the cell smears were rinsed in distilled water and put into Schiff reagent for 90min. The cell smears were counterstained for 20 seconds with 0.1% fast green (Fisher Scientific Co., Chemical Manufacturing Division, Fairlawn, NJ) and rinsed twice in distilled water. The slides were examined at 40X magnification for microscope analysis (Ceretti et al. 2014).

3.10.2 SCORING PROCEDURE FOR MICRONUCLEATED CELLS

Slides was coded before scoring by a person who does not involved in the experiment so that the person who scored the slides were not aware of the treatment conditions, individuals or groups to which the cells on the slides belong. The slides were examined

at ×40 magnifications using a good quality of microscope which is Nikon Eclipse E200. Firstly, the frequency of all various cells types was determined in a minimum of 1,000 cells where it was the optimal way to score slides in the BMCyt assay. According to Thomas (2009), the frequency of DNA damage biomarkers of the MN and NBUDs was scored in a minimum of 2,000 differentiated cells. Scoring of micronuclei frequency was performed by using formula by (Montero et al., 2006);

where, X = Micronuclei score (MN)

2000 is referring to 2000 binucleated cells analysed for presence of MN

$$\frac{\text{Number of } X}{2000} \times 1000$$

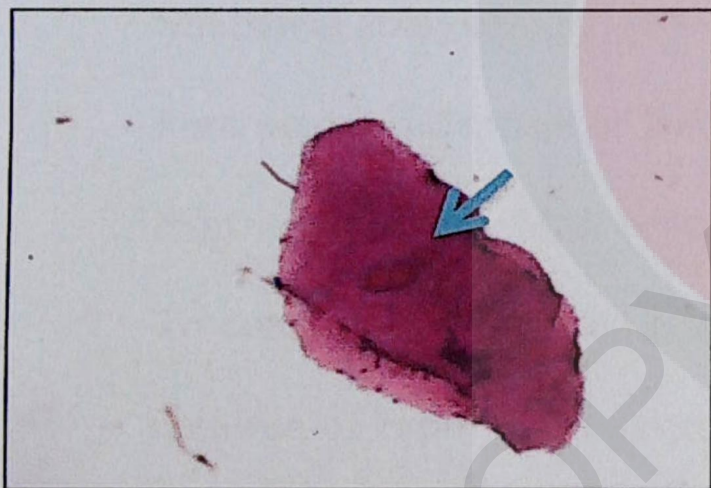


Figure 10: Buccal mucosa cell with micronucleus under x40 magnification

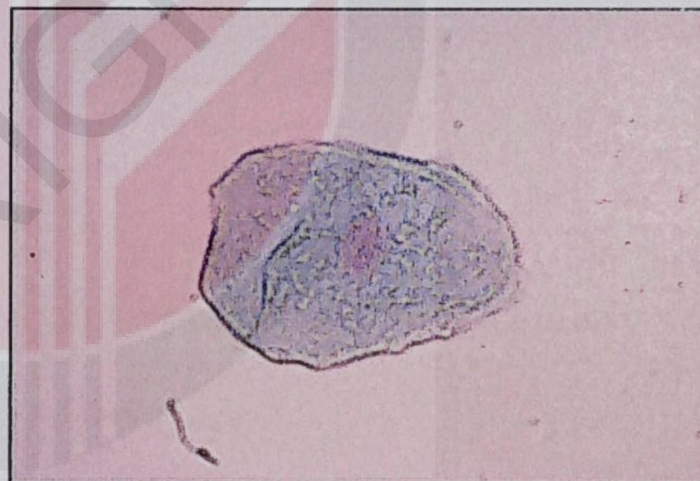


Figure 11: Buccal mucosa cell without micronucleus under x40 magnification

3.11 STATISTICAL ANALYSIS

Obtained data were analysed by using Statistical Package for Social Sciences (SPSS) version 23. The data was analysed by using univariate and bivariate. The significant level of this study was set at $p < 0.05$. Mean, standard deviation, minimum and maximum was determined by performing univariate analysis. In the other hand, bivariate analysis was conducted in order to find the significant difference between variables in this study. Following are the statistical analysis used based on specific objectives:

1. To compare the concentration of PM_{10} and $PM_{2.5}$ exposure among exposed and non- exposed group.

Statistical analysis;

Parametric: Independent T-test

Non – parametric: Man Whitney U -Test

2. To compare the reported respiratory health symptoms among primary school children of exposed group and non- exposed group.

Statistical analysis:

Non – parametric: Chi-Square Test

3. To compare the frequency of micronuclei (MN) among primary school children of exposed group and non- exposed group.

Statistical analysis:

Parametric: Independent T-test

Non – parametric: Man Whitney U –Test

4. To determine the association between PM_{10} and $PM_{2.5}$ with respiratory health symptoms among studied group.

Statistical analysis:

Non – parametric: Chi-Square Test

5. To determine the association between exposure of PM₁₀ and PM_{2.5} with the micronuclei (MN) frequency among exposed and non- exposed group.

Statistical analysis:

Parametric: Pearson correlation

Non – parametric: Spearman correlation

3.12 ETHICAL ISSUES

Children who participated in this study were given an approval from their parents or guardian. Brief explanation was explained by the researcher regarding to the research objective and procedure. All of the participant's information in this study was kept as private and confidential. Before the research begun, this research was given an approval from the Ethics Committee of Faculty Medicine and Health Sciences, Universiti Putra Malaysia, Reference number: UPM/TNCPI/RMC/1.4.18.2 (JKEUPM).

CHAPTER 4

RESULTS

4.1 Information of Respondents

From table 4.1, there were 82 and 85 children of studied and comparative group recruited respectively in this study. Therefore, a total of 167 respondents involved in this study with all of them were Malay. Both studied and comparative group shows the majority of selected respondents were among girls with 43(52.4%) and 53(62.4%).

Table 4.1: Total number of respondents according to studied group and comparative group.

Group	Total	Gender	
	N=167	Boy (%)	Girl (%)
Studied group	82	39 (47.6)	43 (52.4)
Comparative group	85	32 (37.6)	53 (62.4)

Table 4.2: Education level of respondent's parent among studied and comparative group.

Variables	Studied group N=82	Comparative group N=85	χ^2	p-value
	Total (%)	Total (%)		
Father's education level				
Primary school	3 (3.7)	6 (7.1)		
PMR/SRP	1 (1.2)	12 (14.1)		
SPM	28 (34.1)	28 (32.9)	40.206	0.001*
Qualification	-	18 (21.2)		
STPM/Diploma	26 (31.7)	10 (11.8)		
Degree/Master/PhD	24 (29.3)	11 (12.9)		
Mother's education level				
Primary school	2 (2.4)	7 (8.2)		
PMR/SRP	1 (1.2)	1 (1.2)		
SPM	20 (24.4)	36 (42.4)	28.056	0.001*
Qualification	.4)	13 (15.3)		
STPM/Diploma	1 (1.2)	12 (14.1)		
Degree/Master/PhD	25 (30.5)	16 (18.8)		
	33 (40.2)			

*significant at $p < 0.05$ Pearson's Chi-Square

The highest education level obtained by parents between studied and comparative group was SPM with 36 numbers of people came from mother's educational level. Chi- square test was used to analyse parent education level which gave the significant p-value of less than 0.05.

4.2 Respondent's environment

Table: 4.3 Indoor Air Pollutant Source (Home)

Variables	Exposed group N=82 Total (%)	Comparative group N= 85 Total (%)	χ^2	PR	p- value	95%CI
Indoor home						
Smoking						
Yes	23(28)	21(24.7)	0.240	1.188	0.726	0.596-
No	59(72)	64 (75.3)				2.367
Fuels for cooking						
Gas	76(93)	64(76)	8.544	3.958	0.005*	1.499-
Electric and gas	6(7)	20(24)				10.453
Open windows during cooking						
Yes	82(100)	82(97)	2.947	1.037	0.086	0.995-
No	-	3(3)				1.080
Air conditioning system						
Air conditioner	5(6.1)	4(4.6)	3.317	-	0.190	-
Fan	58(70.7) 19(23.2)	70(82.4) 11(12.9)				

Air conditioner and fan						
Mosquito repellent						
Yes	55(67.1)	58(68.2)	0.260	0.948	1.00	0.496-
No	27(32.9)	27(31.8)				1.814
Carpets						
Yes	42 (51.2)	37 (43.5)	0.990	1.362	0.354	0.741-
No	40 (48.8)	48 (56.5)				2.505
Open burning						
Yes	18 (22.0)	20 (23.5)	0.059	0.914	0.855	0.443-
No	64 (78.0)	65 (76.5)				1.886
*significant at p<0.05 Pearson's Chi-Square						

From Table 4.3, for studied group it shows higher number of family members who are smoking in their house compared to comparative group with 23(28%) and 21(24.7%) respectively. The p-value was 0.726, ($p>0.05$). There was no significant difference between two groups of respondents with indoor home smoking, (PR= 1.188, 95%CI 0.596-2.367).

Majority of studied and comparative group using gas as source of fuel during cooking, while only 31% using electric and gas. The p-value was 0.005 where $p<0.05$. There was a significant difference between two groups of respondents with source of fuels during cooking, (PR= 3.958, 95%CI 1.499-10.453).

164 of respondents opened their window during cooking, while only 3 of them were not. The p-value was 0.246; ($p > 0.05$). There was no significant difference between two groups of respondents with opening of windows during cooking, (PR= 1.037, 95%CI 0.995-1.080).

A total of 128 respondents are using fan as their air conditioning system at home, while 30 of them using both fan and air conditioner. According to the table, the Pearson's Chi-Square value was 3.317. The p-value was 0.005; ($p < 0.05$). There was a significant difference between two groups of respondents with air conditioning system.

113 respondents were using mosquito repellent, while 54 of them were not. The value of Pearson's chi-square was 0.260. The p-value was 1.00; $p > 0.05$. There was no significant difference between two groups of respondents with mosquito repellent, (PR= 0.848, 95%CI 0.496-1.814).

From Table 4.3 also, for studied group it shows slightly high number of respondents who used carpets in their house compared to comparative group with 42 (51.2%) and 40(45.5%) respectively. The p-value was 0.354; ($p > 0.05$). There was no significant difference between two groups of respondents with carpets, (PR= 1.362, 95%CI 0.741-2.505).

Only 38 respondents who performing open burning at their house with 18 of them was from studied group and 20 of them from comparative group. The p-value was 0.855 ;(p >0.05. There was no significant difference between two groups of respondents with open burning, (PR= 0.914, 95%CI 0.443-1.886).

Table 4.4: Background of respondent’s house

Variables	Studied group N=82	Comparative group N= 85	χ^2	p-value
	Total (%)	Total (%)		
Distance from main road				
<100 meters from house	5 (6.1)	3 (3.5)		
>100-500 meters from house	5 (6.1)	9 (10.6)		
>500-1000 meters from house	28 (50.9)	27 (31.8)	1.652	0.648
> 1000 meters from house	44 (53.7)	46 (54.1)		
Distance from palm oil factory				
<500 meters	5 (6.1)	-		
>1-1.5 km	18 (22.0)	-		
>1.5-3 km	18 (22.0)	6 (7.1)	40.993	0.001*
>3 km	41 (50.0)	79 (92.9)		

Distance from power station

<500 meters	-	-		
>1-1.5 km	-	-		
>1.5-3 km	6 (7.3)	10 (11.8)	0.953	0.329
>3 km	76 (92.7)	75 (88.2)		

Perception on surrounding area

Very dusty	5 (6.1)	2 (2.4)	5.124	0.077
Moderate dusty	59 (72.0)	52 (61.2)		
Less dusty	18 (22.0)	31 (36.5)		

***significant at p<0.05, Pearson Chi-square**

Based on Table 4.4, there were higher number of respondent 46(54.1%) who lived more than 1000 meters from main road in comparative group while, 44(53.7%) of the respondent were from studied group. Only 5(6.1%) of respondent among studied group who lived less than 500 metres from palm oil factory and most of them live more than 3km from it. The respondents were asked about the perception of their surrounding area whether they were dusty or not and the results showed moderate dusty with 59(72%) and 52(61.2%) from studied and comparative group respectively.

4.3 Respondent’s exposure

Table 4.5: PM₁₀ and PM_{2.5} concentration level between studied and comparative schools.

Variable	Studied schools N=3 Mean±SD	Comparative schools N=3 Mean±SD	t-value	p-value
PM ₁₀ (µg/m ³)	105.21±46.11	53.92 ±12.37	2.843	0.014*
PM _{2.5} (µg/m ³)	68.83±13.03	50.80 ±17.45	2.287	0.040*

Independent T-test, *significant at p<0.05

Table 4.5 showed that the concentration of PM₁₀ and PM_{2.5} of studied schools were higher (105.21±46.11); (68.83±13.03) compared to comparative schools (53.92 ±12.37); (50.80 ±17.45). The data were analysed using Independent T- test because both groups were normally distributed, p>0.05. It showed that the p value was less than 0.05, where it indicates that there was a significant difference of mean between studied and comparative schools.

Table 4.6: PM₁₀ and PM_{2.5} concentration level between studied and comparative respondents' house

Variable	Studied group N=30 Mean±SD/Median± IQR	Comparative group N=32	t- value/z- value	p-value
PM ₁₀ ^a (µg/m ³)	104.29±58.70	29.29±16.69	3.25	0.007*
PM _{2.5} ^b (µg/m ³)	74.29±42.57	28.57±32.11	-2.37	0.018*

*significant at p<0.05

^a= T-test

^b= Mann-Whitney U Test

Based on data in Table 4.6, the normality test gave p-value of p=0.200 for concentration of PM₁₀ at studied and comparative group, while the data of PM_{2.5} at studied group was p=0.200 and p=0.020 for comparative group. Therefore, PM₁₀ concentrations of studied and comparative group were normally distributed. Meanwhile, only PM_{2.5} concentration of studied group showed the data was normally distributed but not for comparative group. From Table 4.6, the mean and standard deviation concentration of PM₁₀,(104.29±58.70µg/m³) among studied group was higher compared to comparative group, (29.29±16.69 µg/m³). The mean and standard deviation concentration of PM_{2.5} at home of studied group was higher (74.29±42.57 µg/m³) compared to comparative group with (Median±IQR) (28.57±32.11 µg/m³). The

data of PM₁₀ between two groups were analysed using Independent T-test since the data from both group were normally distributed. The p- value showed significant difference of mean between studied group and comparative group p= 0.007. Meanwhile, PM_{2.5} data of two groups were analysed using Man – Whitney U test. The result showed that there were significant difference of mean between both groups (p=0.018).

4.4 Comparison of MN Frequency among respondents for studied and comparative group

Table 4.7: MN Frequency among studied and comparative group

Variables	Studied group N=82 Median (IQR)	Comparative group N=85 Median (IQR)	z-value	p-value
MN Frequency	4.00(3.00)	0.50(1.00)	-9.473	0.001*

N= 167, Man - Whitney U Test, *significant at p<0.05

Since the data of MN frequency for both groups were not normally distributed where the normality value was less than 0.05, thus, non- parametric test which are Man-Whitney U test was performed. As shown in Table 4.7, MN frequency of studied group was higher (Median±IQR) (4.00±3.00) compared to comparative group (0.50±1.00).

The results showed that there were significant difference of mean between studied and comparative group (p=0.001).

4.5 Reported Respiratory Health Symptoms

Table 4.8: Reported respiratory symptoms among studied and comparative group.

Variables	Studied group n=82	Comparative group n=85	χ^2	p-value	PR	95% CI
Total (%)						
Cough						
Yes	24 (29.3)	1 (1.2)	25.875	0.001*	34.76	4.57-264.17
No	58 (70.7)	84 (98.8)				
Phlegm						
Yes	13 (15.9)	1 (1.2)	11.706	0.001*	15.83	2.02-124.01
No	69 (84.1)	84 (98.8)				
Wheezing						
Yes	22 (26.8)	1 (1.2)	23.127	0.001*	30.80	4.04-234.80
No	60 (73.2)	84 (98.8)				
Chest tightness						
Yes	13 (15.9)	2 (2.4)	9.305	0.002*	7.82	1.71-35.84
No	69 (84.1)	83 (97.6)				

^aFisher Exact test

***significant at $p < 0.05$**

Table 4.8 showed the reported respiratory symptoms among studied and comparative group. Based on the table, most reported respiratory health symptoms were cough 24(29.3%) comes from respondents of the studied group and only 1(1.2%) from comparative group. There was significant difference between two groups of respondents with cough ($p=0.001$) Respondents in studied group were 34 times as likely to get cough, (PR= 34.76, 95%CI 4.57-264.17).

There were 14 respondents who experienced phlegm where 13(15.9%) of them are respondents from studied group while only 1(1.2%) are from comparative group. There was a significant difference between two groups of respondents with phlegm ($p < 0.05$). Chi square result revealed that respondents in studied group were 15 times as likely to get phlegm, (PR= 15.83, 95%CI 2.02-124.01).

28% of respondents were having wheezing where 22(26.8%) of them are respondents from studied group while only 1(1.2%) are from comparative group. There was a significant difference between two groups of respondents with wheezing ($p < 0.05$). Respondents in studied group were 30 times as likely to get wheezing, (PR= 30.800, 95%CI 4.040-234.799).

28.3% of respondents were having chest tightness where 13(15.9%) of them are respondents from studied group while only 2(2.4%) are from comparative group. There was a significant difference between two groups of respondents with chest tightness ($p < 0.05$). Respondents in studied group were 7 times as likely to get chest tightness, (PR= 7.819, 95%CI 1.706-35.841).

4.6 Correlation between PM₁₀ and PM_{2.5} (Home) with MN Frequency among studied group and comparative group.

Table 4.9: Correlation between PM₁₀ and PM_{2.5} (Home) with MN Frequency of studied group and comparative group

Variables	MN Frequency (N=62)	
	r-value	p-value
PM ₁₀	0.562	0.001*
PM _{2.5}	-0.041	0.830

N=62, Spearman Correlation test, *Significant at p<0.05

Based on Table 4.9, there was a significant association between PM₁₀ with MN frequency of studied and comparative group as the p- value was less than 0.05, p= 0.001. Therefore, the correlation of PM₁₀ showed that r-value was r= 0.562, where there was a moderate positive correlation between PM₁₀ and MN frequency.

4.7 Association between PM₁₀ and PM_{2.5} (Home) with reported respiratory health symptoms.

Table 4.10: PM₁₀ concentration level with respiratory symptoms

	High PM ₁₀ (≥150µg/ m ³) N=30	Low PM ₁₀ (<150µg/ m ³) N=32	χ ²	P-value	PR	95% CI
Total (%)						
Cough^a						
Yes	9(30)	1(3.1)	8.267	0.005*	13.28	1.565-
No	21 (70)	31 (96.9)			6	112.803
Phlegm^a						
Yes	5 (16.7)	1 (3.1)	3.248	0.099	6.200	0.680-
No	25 (83.3)	31 (96.9)				56.559
Wheezing^a						
Yes	8 (26.7)	3 (9.4)	3.172	0.101	3.515	0.835-
No	22 (73.3)	29 (90.6)				14.806
Chest tightness^a						
Yes	5 (16.7)	2 (6.3)	1.677	0.249	3.000	0.535-
No	25 (83.3)	30 (93.8)				16.814

*Significant at p<0.05

^aFisher Exact test

The association between concentration of PM₁₀ at home and respiratory health symptoms among respondents are shown in Table 4.10. The table revealed that the association between PM_{2.5} with cough (PR=13.286, 95%CI 1.565-112.803), phlegm (PR= 6.200, 95%CI 0.680-56.559), wheezing (PR= 3.515, 95%CI 0.835-14.806), and chest tightness (PR= 3.00, 95%CI 0.535-16.814). Statistical analysis showed that there was a significant association between PM₁₀ with cough (p=0.005). Respondents who exposed to high level of PM₁₀ were 13 times as likely to get cough, (PR= 13.286, 95%CI 1.565-112.803).

Table 4.11: PM_{2.5} concentration level with respiratory symptoms

	High	Low	χ^2	P-value	PR	95% CI
	PM _{2.5} (≥75	PM _{2.5} (<75				
	μg/m ³)	μg/m ³)				
	N=32	N=33				
	Total (%)					
Cough^a						
Yes	9 (30)	2 (6.3)	5.984	0.020*	6.429	1.259-
No	21 (70)	30 (93.8)				32.827
Phlegm^a						
Yes	6 (20)	1 (3.1)	4.402	0.050	7.750	0.873-
No	24 (80)	31 (96.9)				68.768
Wheezing^a						
Yes	5 (16.7)	2 (6.3)	1.677	0.249	3.000	0.535-
No	25 (83.3)	30 (93.8)				16.814

Chest

tightness^a

Yes	6 (20)	2(6.3)	4.402	0.050	7.750	0.873-
No	24 (80)	30 (93.8)				68.768

***Significant at p<0.05**

^aFisher Exact test

Since the dependent and independent variables were categorical data, thus, Chi-square test was used to determine the association between it. The association between concentration of PM_{2.5} at home and respiratory health symptoms among respondents are shown in Table 4.11. The table revealed that the association between PM_{2.5} with cough (PR=6.429, 95%CI 1.259-32.827), phlegm (PR= 7.750, 95%CI 0.873-68.768), wheezing (PR= 3.000, 95%CI 0.535-16.814), and chest tightness (PR= 7.750, 95%CI 0.873-68.768). Statistical analysis showed that there was significant association between PM_{2.5} with cough (p=0.020). Respondents exposed to high level of PM_{2.5} were 6 times as likely to get cough, (PR=6.429, 95%CI 1.259-32.827).

CHAPTER 5

DISCUSSION

5.1 Discussion

5.1.1 Respondent's Background

This study was a cross sectional comparative study where it involves two groups of respondents. The studied group were among primary school children that exposed to particulate matters (PM_{10} and $PM_{2.5}$) which are released from palm oil activity at Semenyih. The comparative group was among school children who lived at Hulu Langat. Based on sample size calculation, the number of sample required was 170; however after the distribution of questionnaire, a total of 167 respondents were volunteered to participate in this research. All of the respondents were meet the inclusion criteria and all of them got the approval from their parents/ guardians. There are two school are selected in this study, one from studied area and the another one from comparative group. All of the respondents were Malay.

Apart from 167 respondents, 82 of them were from exposed group, whereas 85 were from comparative group. The respondents were selected based on list name provided by schools management. The aged of selected respondents were among 9-11 years old. The selected respondents must do not have any respiratory illness such as

asthma, bronchitis, emphysema and pneumonia. In the others hand, the respondents also must do not undergone radiotherapy or chemotherapy in the previous 12 months or X-rays in the previous 3 months. Studied group are among those who living near to palm oil activity, while for comparative group are among those who are living far from palm oil activity.

5.1.2 Respondent's Socio-demographic Information

After comparison between two groups of respondents, the median age for each group was 10.0 years. The median weight of respondent between studied and comparative group was 28.25kg and 27.20kg respectively. Meanwhile, the mean height of respondents between studied and comparative group was 133.02cm and 128.39cm. The p-value when comparing mean of height showed significant difference between studied and comparative group with $p=0.008$, less than 0.05. For the mean height and median weight, the p-value was more than 0.05 where it indicates there was no significant difference between two group; $p=0.922$, $p=0.605$ respectively. Therefore, it can be concluded that, the availability of respondents also depend on the permission of school management, whereby they only allowed to have only 10 years old children who are from Standard 4 in studied group.

In this study, the highest education level obtained by parents was SPM, 42.4% (mothers) and 32.9% (fathers) where both are from comparative group. Meanwhile, there were 33(40.2%) mothers from studied group who obtained Degree/Master/PhD and 28(34.1%) fathers who obtained SPM. After performing Chi-Square test, the p -

value is 0.001, less than 0.05. It can be concluded that, there was a significant difference between father's and mother's education level.

5.1.3 Respondent's environment

As presented in Table 4.3, there were 140 of respondents using gas as sources of fuel for cooking with 93% of them are from studied group and 76% from comparative group. The p-value shows significant difference with $p = 0.005$. There were a few factors which contribute to the production of indoor air pollutants such as ventilation and also the source of fuel when cooking at home. This can be proven with a study by Rojas-Bracho et al.,(2002) as the ventilation status in houses was one of the factors that contribute to the production of indoor particulate matters concentration by which the particles are generated from the outdoor. Particulate emission from palm oil refinery also catching the attention of local authorities due to the combustion process as it caused air pollution (Abdullah et al.,2007). According to the World Health Organisation (2008), $PM_{2.5}$ produced from incomplete combustion was identified as a contributing factor on air quality to detrimental effects on health and also climate.

Based on Table 4.4, only 5(6.1%) of respondent among studied group who lived less than 500 metres from palm oil factory and most of them live more than 3km from it. The distance of houses from factory can be the factors due to high PM_{10} and $PM_{2.5}$ at home, where half (50%) of the studied group lived less than 3km from the factory as the house were located within zone of impact. The zone of impact taken an area of 5km radius from the site and it depends on the source strength and also the pollutant dilution effect (Department of Environment, 2012). Air pollutants such as $PM_{2.5}$ can persist at the atmosphere for a few days and even for a few weeks as well as the radius dispersion of particulate matter can travel up to hundreds to thousands

kilometre. Meanwhile, PM₁₀ can travel in the atmosphere for less than one kilometre to hundreds of kilometre (Dalglish et al., 2007).

5.1.4 Respondent's exposure (School)

Based on the results presented, the concentrations of PM₁₀ and PM_{2.5} of studied schools were significantly higher 105.21µg/m³ compared to comparative schools 53.92µg/m³. Based on the previous study Hussin & Jalaludin (2016), the concentration of indoor air pollutants (PM₁₀; 68.52 µg/m³ and PM_{2.5}; 41.33µg/m³) are higher in school which located near the palm oil factory. In the other hand, vulnerable groups such as elderly, pregnant women and children who were residing near palm oil were exposed to high indoor and outdoor air pollutants by the emission of air pollutants released from the factory. One of the activities performed by the palm oil factory was the combustion of biomass material such as fibre and shell from fresh fruit bunch as the source of energy for the factory. Unfortunately, the burning of waste as a boiler fuel created a harmful emission in the industry, and it released a massive amount of particles in the form of fly ash to the air (Kun & Abdullah, 2013); (Rashid, Ramli, & Rozainee, 1998).

5.1.5 Comparison of PM₁₀ and PM_{2.5} at Respondent's Houses between Studied and Comparative Area

From this study, personal air sampling was placed in respondent's house, 30 from studied group and 32 from comparative group. Based on Table 4.6, the mean concentration of PM₁₀ at home within 24-hours was 104.29µg/m³ for the studied group

were higher compared to the comparative group, $29.29\mu\text{g}/\text{m}^3$. Based on study by Gholampour et al., (2014) in urban location, found that mean in 24-hours of PM_{10} was $85.3\mu\text{g}/\text{m}^3$. Previous local study by Hussin & Jalaludin (2016) revealed that the mean concentration of PM_{10} in 24-hours located near palm oil mill was lower than present study with concentration $72.45\mu\text{g}/\text{m}^3$.

The concentration of PM_{10} ($104.29\mu\text{g}/\text{m}^3$) was lower compared to $150.0\mu\text{g}/\text{m}^3$ in 24-hours measurement. Moreover, for the concentration of $\text{PM}_{2.5}$ at home, the studied group recorded higher $74.29\mu\text{g}/\text{m}^3$ compared to the comparative group $28.57\mu\text{g}/\text{m}^3$. However, $74.29\mu\text{g}/\text{m}^3$ was lower when comparing with Ambient Air Quality Standard Malaysia (AAQS), $75.0\mu\text{g}/\text{m}^3$ within 24-hours period. As compared to previous local study by Hussin & Jalaludin (2016) the mean concentration of $\text{PM}_{2.5}$ in 24-hours located near palm oil mill was lower than present study with $42.26\mu\text{g}/\text{m}^3$ among studied group and comparative group, $23.54\mu\text{g}/\text{m}^3$.

5.1.6 Comparison of MN Frequency among respondents for studied and comparative group

MN assay in children was increasingly used because of its public health significance for monitoring environmental exposure such as pollution, radiation, industrial toxicants and various medical treatment (Holland et al., 2011). The MN frequency of the studied group was higher compared to comparative group where it increases by eight fold from median MN frequency of comparative group. However, the mean MN frequency of this study (4.00%) was slightly lower compared to baseline mean MN frequency (5.7%) from pooled analysis by Neri et. al (2005) among children

aged 0- 18 years old. MN frequency of this study also lower when compared to MN frequency (6.6%) of children with aged 5 -11 years old who exposed to air pollutant resulted from study by (Pedersen et al., 2006). Another study by Sisenando et al., (2012) it stated that, they found significant evidence in which increased exposure to PM_{2.5}, it can generate an increased micronuclei frequency in oral epithelial cells from school children. From this study, it can be conclude that the MN frequency was still does not exceed the baseline mean MN frequency from the pooled analysis. However, the exposure of particulate matter would induced the formation of micronuclei.

5.1.7 Reported Respiratory Health Symptoms

Most reported respiratory health symptoms were coughing 29.3% from respondents of the studied group followed by wheezing, phlegm and also chest tightness. Meanwhile, for the comparative group, only 1.2% of the respondents were experienced cough, phlegm, wheezing and 2.4% of the respondents had reported having chest tightness. Respondents in studied group were 34 times as likely to get cough, 15 times likely to get phlegm, 30 times likely to get wheezing and 7 times likely to get chest tightness, (PR= 34.76, 95%CI 4.57-264.17); (PR= 15.83, 95%CI 2.02-124.01); (PR= 30.800, 95%CI 4.040-234.799); (PR= 7.819, 95%CI 1.706-35.841). These outcomes were parallel with a study by Farah and Juliana (2016) where the reported respondents who were having a cough, phlegm, wheezing has a significant difference ($p<0.01$) between studied and comparative group. This was because the location of studied area was near to palm oil factory. Particles inhaled into our respiratory tract can penetrate deeply into our lung and even into our bloodstream.

According to (Whyand et al., 2018), the penetration of particulate matter into the thoracic regions or lower respiratory tracts can cause asthma and respiratory morbidity.

5.1.8 Association between PM₁₀ and PM_{2.5} (Home) with reported respiratory health symptoms.

This study revealed that there was significant association between PM₁₀ and PM_{2.5} with prevalence cough, $p=0.005$ and $p=0.020$ respectively. Exposure to particulate matters can increase the prevalence of respiratory symptoms in young children. Based on Pierse et al.(2006), it showed that there was a significant association between exposures to PM₁₀ at home with the prevalence of coughing. Exposure to particulate matters also was associated with increased respiratory symptoms (Tiittanen et al.,2006). Study by Darquenne (2006) stated that, the initial deposition for the particles which is larger than 5 micrometres are likely to deposited at upper respiratory tracts which are nasal and mouth cavities to the entrance of the trachea. A study conducted in Delhi, identified that there was significantly higher prevalence of respiratory symptoms in urban areas compared to rural area because the effect of air pollution.

5.1.9 Correlation between PM₁₀ and PM_{2.5} (Home) with MN Frequency among studied group and comparative group.

Based on Table 4.9, there was a significant association between PM₁₀ and MN frequency ($p<0.01$). Study by Soares et. al, (2003) found that micronuclei frequency

was associated with PM₁₀, NO₂ and CO where it comes from an automotive emission. Particulate matter (PM₁₀) was associated with mutagenic activities, as its presence in gas and particulate phase. It gave a significant health impact as the chemical was deposited in an organ that may cause in vivo induction of somatic cell mutation and carcinogenesis (Soares et al., 2003).

Based on study by Coronas (2009), there were increased levels of DNA damage was detected in the comet assay in subjects who are working or living near an oil refinery compared with people living in areas with restricted traffic and industries.

CHAPTER 6

CONCLUSION, RECOMMENDATIONS AND STUDY LIMITATION

6.1 Conclusion

In a nut shell, findings from this study revealed that the studied group has a higher exposure to PM_{10} and $PM_{2.5}$. The exposure to high level of PM_{10} and $PM_{2.5}$ was a contributing factor for the reported respiratory health symptoms and also might induce the formation of micronuclei among affected primary school children who lived near palm oil factory.

Based on the result presented in this study, children among studied group were prone to get cough (29.3%), phlegm (15.9%), wheezing (26.8%) and chest tightness (15.9%) compared to comparative group. The mean concentration of PM_{10} at respondent's house among studied group was higher compared to comparative group. The p- value showed significant difference of mean between studied group and comparative group ($p= 0.007$). Meanwhile, the mean concentration of $PM_{2.5}$ at home of studied group was also higher compared to comparative group. The p-value of $PM_{2.5}$ was 0.018, showed that, there were significant difference of mean between both

groups. It also can be concluded, there was a significant difference in reported respiratory symptoms (cough, phlegm, wheezing, chest tightness), as it were significantly higher among children in studied group than comparative group. On the other hand, respondents in studied group who were exposed to high level of PM_{10} were 13 times as likely to get cough and 3 times as likely to experience wheezing. Meanwhile, respondents in studied group who were exposed to high level of $PM_{2.5}$ were 6 times likely to experience cough and 7 times likely to get chest tightness. The findings of this study also portrayed, there were significantly higher mean MN frequency among studied group compared to comparative group ($p < 0.01$). There were also significant association between MN frequency with PM_{10} in studied and comparative group ($p = 0.001$). Last but not least, the result showed that, there were association between PM_{10} and MN frequency.

6.2 Recommendations

Further comprehensive investigation was needed as this study only provides the preliminary results which represent the data of school and respondent's house that lived near palm oil factory. Moreover, the best study design suggested was prospective cohort study, as the main factors of respiratory health symptoms and induced formation of MN frequency can be determined.

As for the management of palm oil factory, there was a need for the palm oil management to revise again the effectiveness of existing control measure implemented by the factory as the emission released contains harmful air pollutants where it can

cause serious health problem. There are a few ways that can be performed by the management as result to reduce the impact of biomass fuel to public health community and ambient air quality. The fabric filter baghouses can be used to control and reduce the primary fine particulate emissions from the combustion of biomass fuels. As the time pass by, the technologies on controlling of particulate emissions has been revolving. The Air Pollution Control Technology Centre (APCT Centre) was a system that assists vendors and users in demonstrating technologies for air pollution control and it was programmed on the baghouse filtration products. The best material for filter baghouses was believed can improve the protection of human health and the environment. If the management of palm oil factory would consider the suggested recommendation, the researcher believe that the concentration of PM_{10} and $PM_{2.5}$ at studied area can be reduced, therefore the number of reported respiratory symptoms (especially cough and wheezing) and the frequency of micronuclei formation among studied group can be controlled.

The parents of respondents are advised to seek for doctor's consultation immediately if any reported respiratory health symptoms experienced by their children. Moreover, the parent need to monitor their children from prolong doing outdoor activities.

Last but not least, the researcher would like to suggest the Department of Environmental to organize a conference, talks or campaign on the alarming issues regarding clean environment. This would create awareness among community in that area especially parents of respondents. The information also should be widely

disseminated to the community in variety of platforms such as mass media, pamphlets and poster.

6.3 Study Limitation

Based on this research, there are some limitations that can be spotted which can be improved for future research. First and foremost, the sample size of studied and comparative group only involved the comparison of one to one school. Therefore, there should be compare to more than one school to make this study are representable for that area. Other than that, the results of this study cannot be generalised for other ethnicity as this study only involved Malay respondents.

Next, the number of house's respondent for dust sampling at home. The number of respondent for dust sampling at home was insufficient because the parents only allowed their children participated in collection of buccal mucosa sample, but not gave a permission to do sampling at home. The instruction of sampling dust at home was given earlier, however only a few of them agree.

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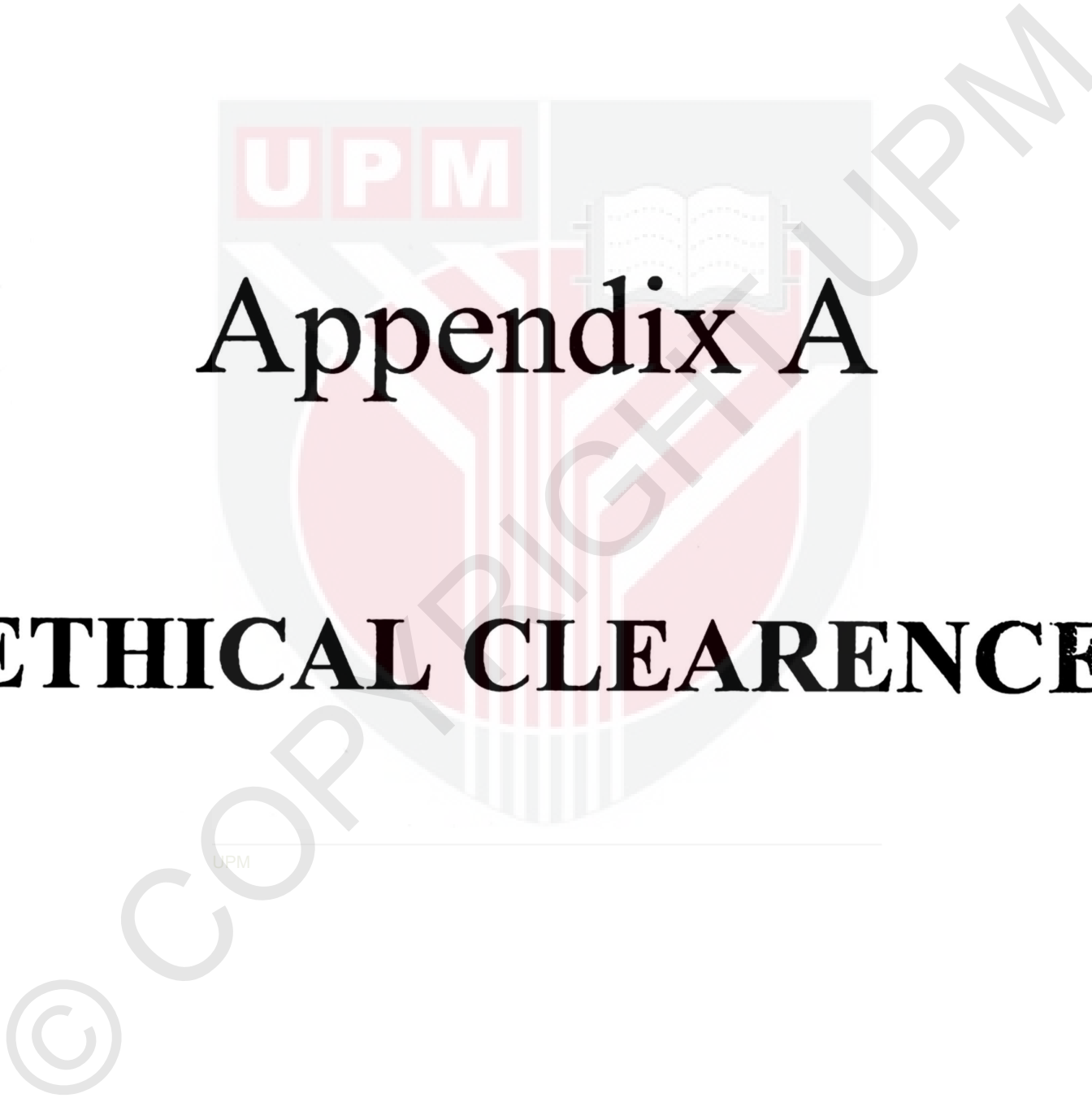
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A large, light gray watermark of the UPM logo is centered on the page. The logo features a shield with a red and white striped field, a red chief with the letters 'UPM' in white, and a red base with a white open book. The text 'UPM' is also written diagonally across the shield. Below the shield, the text '© COPYRIGHT UPM' is written in a light gray font.

Appendix A

ETHICAL CLEARANCE

**ETHICS COMMITTEE FOR RESEARCH INVOLVING HUMAN SUBJECTS
(JKEUPM)
UNIVERSITI PUTRA MALAYSIA**

Research title	: Association Between Exposure to PM2.5 And PM10 With DNA Damage in Exfoliated Buccal Mucosa Cells Among Primary School Children Nearby Palm Oil Mill at Semenyih
Study Site	: Semenyih
JKEUPM Ref No.	: JKEUPM-2018-374
Researcher	: Syhidatul Farhana binti Othman
Supervisor	: Assoc Prof. Dr. Juliana Jalaludin

Documents received and reviewed with reference to the above study:

1. Ethics Application Form, Version 1 dated 29/10/2018
2. Respondent Information Sheet & Guardian's/Parent's Consent (Malay), Version 2 dated 5/12/2018
3. Proposal (English), Version 1 dated 15/11/2018
4. Questionnaires Interviews (Malay), Version 1 dated 15/11/2018
5. Curriculum Vitae of:
 - a. Assoc Prof. Dr. Juliana Jalaludin

The University Research Ethics Committee, Universiti Putra Malaysia (JKEUPM) operates in accordance to the ICH-GCP Guidelines.

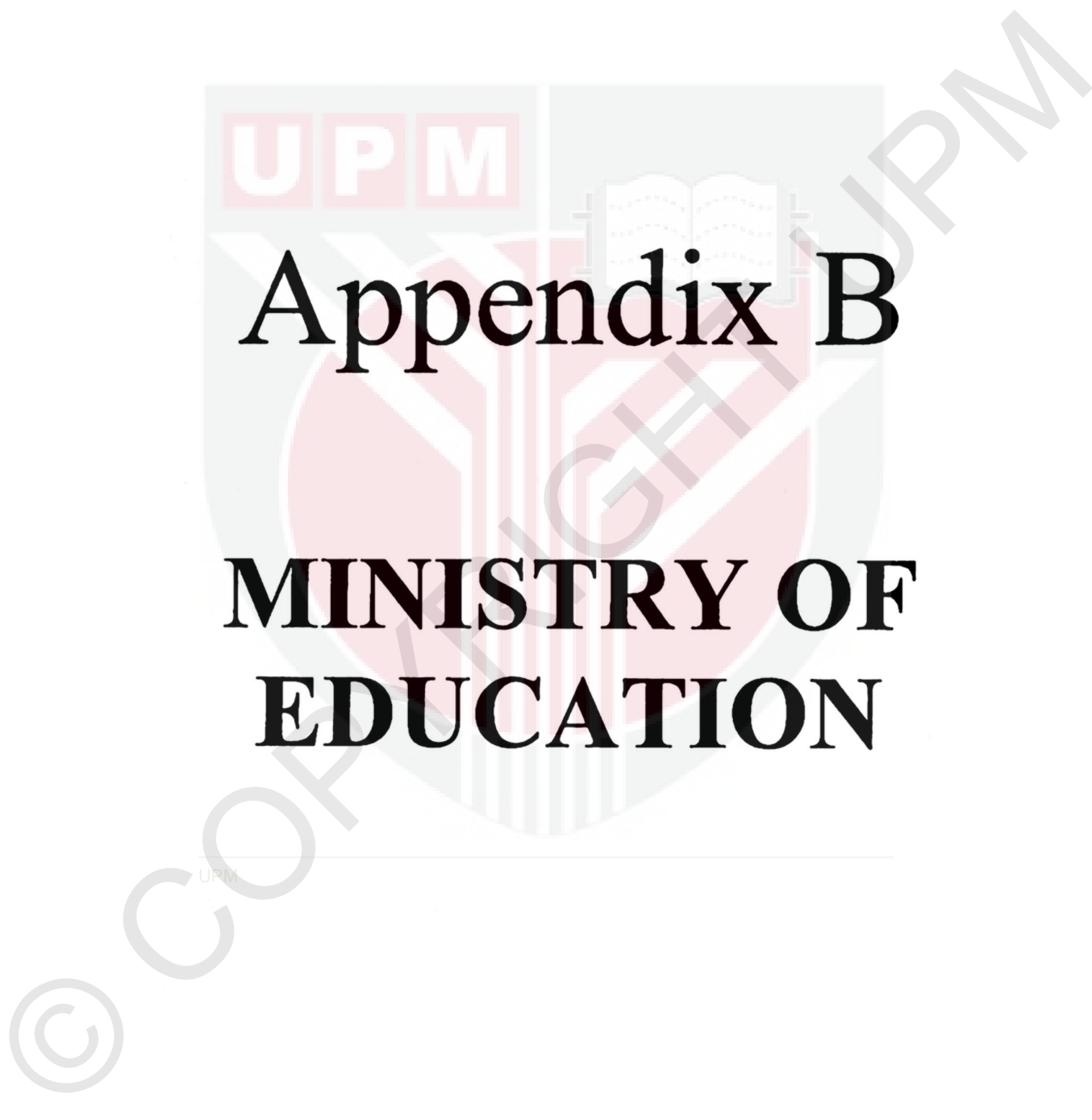
Decision by JKEUPM:

- ☒ Approved
- ☒ Permission **MUST BE OBTAINED** from the respective hospitals/ institutions before conducting the research
- ☐ Disapproved

Please note that the approval is **VALID UNTIL 18 DECEMBER 2019**

Researchers should comply with the following:

- I. Complete a Study Final Report upon study completion (Form 3.2).
- II. Ethical approval is required in the case of amendments/ changes to the study documents/ study sites/ study team.
- III. Applicable for Clinical Trial Studies and Clinical interventional Studies only: Progress Report has to be submitted to JKEUPM at every 6 months from the date of approval (Form 3.1). Report

A large, light gray watermark of the UPM logo is oriented diagonally across the page. The logo features a shield with a red and white geometric design, a book, and the letters 'UPM' in a red box.

Appendix B

**MINISTRY OF
EDUCATION**



KEMENTERIAN PENDIDIKAN MALAYSIA
BAHAGIAN PERANCANGAN DAN PENYELIDIKAN DASAR PENDIDIKAN
ARAS 1-4, BLOK E8
KOMPLEKS KERAJAAN PARCEL E
PUSAT Pentadbiran Kerajaan Persekutuan
62604 PUTRAJAYA

TEL : 0388846591
FAKS : 0388846579

Ruj. Kami : KPM.600-3/2/3-eras(2764)
Tarikh : 7 Januari 2019

SYHIDATUL FARHANA BINTI OTHMAN
NO. KP : 960331015812

SBK 77 SUNGAI BALANG KECIL
SEMERAH BATU PAHAT JOHOR 83600 MUAR
JOHOR

Tuan,

KELULUSAN UNTUK MENJALANKAN KAJIAN DI SEKOLAH, INSTITUT PENDIDIKAN GURU, JABATAN PENDIDIKAN NEGERI DAN BAHAGIAN DI BAWAH KEMENTERIAN PENDIDIKAN MALAYSIA

Perkara di atas adalah dirujuk.

2. Sukacita dimaklumkan bahawa permohonan tuan untuk menjalankan kajian seperti di bawah telah diluluskan.

" HUBUNG KAIT DI ANTARA PENDEDAHAN KEPADA PM2.5 DAN PM10 DAN KEROSAKAN DNA DALAM SEL BUKAL MUKOSA DI KALANGAN KANAK-KANAK SEKOLAH RENDAH BERHAMPIRAN KAWASAN PERINDUSTRIAN DI SEMENYIH "

3. Kelulusan adalah berdasarkan kepada kertas cadangan penyelidikan dan instrumen kajian yang dikemukakan oleh tuan kepada bahagian ini. Walau bagaimanapun kelulusan ini bergantung kepada kebenaran Jabatan Pendidikan Negeri dan Pengetua / Guru Besar yang berkenaan.

4. Surat kelulusan ini sah digunakan bermula dari 2 Januari 2019 hingga 2 Mei 2019 .

5. Tuan dikehendaki menyerahkan senaskhah laporan akhir kajian dalam bentuk *hardcopy* bersama salinan *softcopy* berformat pdf dalam CD kepada Bahagian ini. Tuan juga diingatkan supaya mendapat kebenaran terlebih dahulu daripada Bahagian ini sekiranya sebahagian atau sepenuhnya dapatan kajian tersebut hendak diterbitkan di mana-mana forum, seminar atau diumumkan kepada media massa.

Sekian untuk makluman dan tindakan tuan selanjutnya. Terima kasih.

"BERKHIDMAT UNTUK NEGARA"

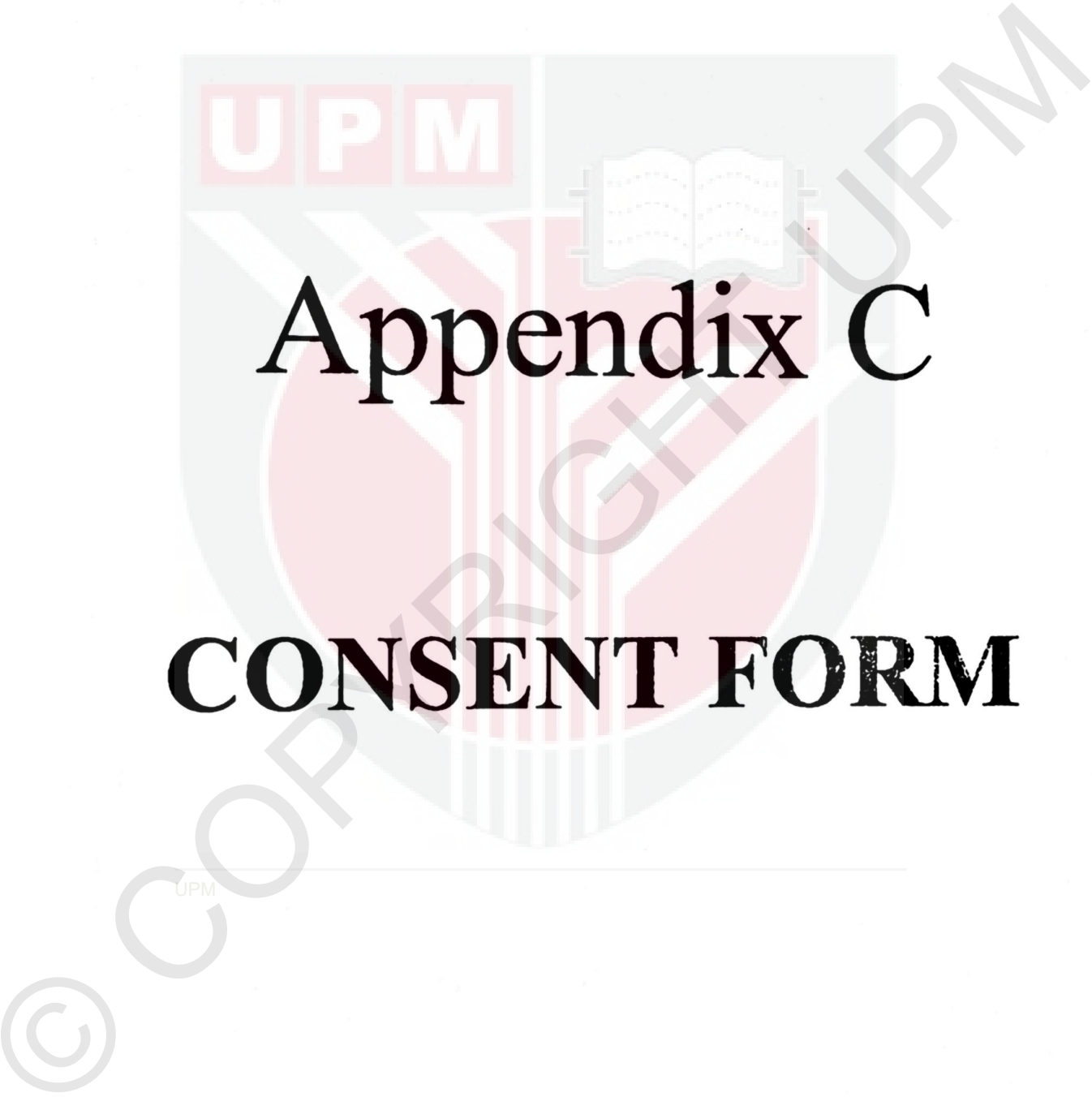
Saya yang menjalankan amanah,

Ketua Sektor
Sektor Penyelidikan dan Penilaian
b.p. Pengarah
Bahagian Perancangan dan Penyelidikan Dasar Pendidikan
Kementerian Pendidikan Malaysia

salinan kepada:-

JABATAN PENDIDIKAN SELANGOR

* SURAT INI DIJANA OLEH KOMPUTER DAN TIADA TANDATANGAN DIPERLUKAN *

The image features a large, faint watermark of the Universiti Putra Malaysia (UPM) logo in the background. The logo is a shield-shaped emblem with a red and white color scheme. It includes a book icon at the top, a stylized 'U' and 'P' in the center, and the letters 'UPM' in a red box at the top left. A diagonal banner across the shield contains the word 'BERKHIDMAT' in white. Below the shield is a banner with the motto 'BERSAMA SAMA MELAKSANAKAN TRANSFORMASI'. A copyright symbol and the word 'UPM' are also visible at the bottom left of the watermark.

Appendix C

CONSENT FORM



BORANG 2.5: PENERANGAN DAN PERSETUJUAN IBUBAPA/PENJAGA

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

1.TAJUK KAJIAN

Hubung Kait di antara Pendedahan kepada $PM_{2.5}$ dan PM_{10} dan Kerosakan DNA dalam Sel Bukal Mukosa di kalangan Kanak-Kanak Sekolah Rendah Berhampiran Kilang Minyak Kelapa Sawit di Semenyih.

2. PENGENALAN

Kelapa sawit merupakan salah satu tanaman utama yang ditanam di Malaysia. Perladangan minyak secara agresif berkembang dari awal tahun 1875. Malaysia juga telah menjadi pengeluar minyak kelapa sawit kedua terbesar di dunia. Menurut Industri Minyak Sawit Malaysia, Malaysia kini menyumbang 39% pengeluaran minyak sawit dunia dan 44% eksport dunia. Pengeluaran minyak sawit mentah menunjukkan kenaikan sebanyak 15% mencapai 19.92 juta tan pada tahun 2017 berbanding 17.32 juta tan pada tahun 2016. Oleh kerana ia merupakan salah satu daripada sektor ekonomi terbesar di Malaysia, industri kilang minyak sawit telah menggunakan ramai tenaga buruh untuk bekerja di sektor perladangan dan juga kilang kelapa sawit.

Memandangkan Malaysia perlu menghasilkan minyak mengikut permintaan dunia, setiap kilang perlu meningkatkan pengeluaran dan beroperasi pada waktu siang dan malam. Setiap kilang pasti mempunyai satu dandang untuk menjalankan proses pembakaran. Menurut Rashid et.al, kira-kira satu tan brunch buah segar menghasilkan serat 14% -15%. 6-7% serat tersebut akan dibakar untuk menjana tenaga pemprosesan minyak sawit. Walaupun sisa yang dihasilkan digunakan semula, namun hasil pembakaran daripada bahan biomas telah menyumbang kepada masalah pencemaran udara. Hal ini kerana, hasil pembakaran tersebut mengeluarkan partikel yang berbahaya terhadap kesihatan manusia.

Berdasarkan Industri dan Alam Sekitar (Buku Panduan No.3) Industri Minyak Sawit Mentah, (1999), kilang minyak kelapa sawit sebenarnya mencemarkan persekitaran kita melalui penjaan efluen cair, sisa pepejal dan juga pelepasan gas. PM_{10} dan $PM_{2.5}$ adalah salah satu faktor penyumbang yang boleh menyebabkan kesan buruk terhadap kesihatan manusia terutama kepada sistem pernafasan. Bahan partikel 10 juga dikenali sebagai zarah-zarah yang tidak dapat dilihat dengan diameter aerodinamik kurang daripada 10 mikrometer dan lebih kecil. Menurut *Environmental Protection Agency* (2013), sumber debu dapat dijumpai berhampiran jalan raya dan industry, di mana saiz partikel lebih besar daripada 2.5 mikrometer dan lebih kecil

daripada diameter 10 mikrometer. Manakala, bahan partikel 2.5 juga dikenali sebagai zarah halus dengan diameter aerodinamik kurang daripada 2.5 mikrometer. Zarah-zarah ini, mudah ditembusi dalam tempoh yang lama dalam paru-paru dan aliran darah sekaligus membawa kepada masalah kesihatan yang serius. Zarah-zarah ini secara langsung dihasilkan dari sumber seperti kebakaran hutan, atau ia terbentuk apabila gas dihasilkan dari industri dan mereka bertindak balas dengan udara (Agensi Perlindungan Alam Sekitar Amerika Syarikat, 2013). Morbiditi pernafasan dan kardiovaskular seperti gangguan asma, gejala pernafasan dan peningkatan kemasukan ke hospital adalah beberapa kesan kesihatan akibat pendedahan bahan zarah dalam jangka pendek dan jangka panjang (*World Health Organization*, 2013). Objektif kajian ini adalah untuk mengkaji pendedahan PM_{10} dan $PM_{2.5}$ dan perkaitannya dengan kerosakan DNA dalam sel mukosa buccal di kalangan kanak-kanak sekolah rendah berhampiran kilang minyak sawit di Semenyih.

3. APAKAH YANG PERLU ANDA LAKUKAN?

Kajian ini akan dijalankan di Sekolah Kebangsaan Jenis Tamil (SJKT) Ladang Rinching, Semenyih dan Sekolah Kebangsaan Bandar Rinching di kalangan kanak-kanak sekolah rendah yang terdiri daripada kumpulan terdedah dan kumpulan perbandingan yang belajar di Sekolah Kebangsaan Dusun Tua Hulu Langat dan Sekolah Kebangsaan Kelubi Hulu Langat. 77 sampel pelajar akan dipilih secara rawak dari senarai nama setiap kawasan yang tercemar dan kawasan perbandingan. Para penjaga atau ibu bapa pelajar akan ditemuramah dengan menggunakan satu set soal selidik untuk mendapatkan maklumat seperti latar belakang sosio, status kesihatan, sejarah kesihatan pernafasan, pendedahan kepada asap rokok dan kebiasaan pemakanan kanak-kanak. Soal selidik itu telah diadaptasi daripada *American Thoracic Society* (Soal Selidik ATS -DCD-78C untuk kanak-kanak) dan soal selidik daripada *American Academy of Pediatrics*. Prosedur pengukuran akan dilakukan semasa waktu sekolah. Kepekatan PM_{10} dan $PM_{2.5}$ akan diukur dengan menggunakan DustTrak DRX 8534 Model. DustTrak tersebut akan diletakkan pada tahap pernafasan yang sama dengan jarak 1 meter dari pelajar untuk mengelakkan sebarang gangguan semasa pengumpulan data. Poin pengukuran dipilih untuk mengelakkan sebarang gangguan akibat pergerakan dan juga untuk mengelakkan tarikan daripada kanak-kanak. Selain itu, instrumen-instrumen tersebut akan diletakkan di belakang sudut bilik darjah untuk mengelakkan gangguan pelajar semasa pembelajaran. Kepekatan PM_{10} dan $PM_{2.5}$ akan diukur dengan menggunakan pam persampelan udara. Pam persampelan udara tersebut akan diletakkan di rumah kanak-kanak dan dibiarkan selama 24 jam. Kemudian, sampel biologi yang merupakan sel mukosa buccal akan dikumpulkan dari setiap responden dengan bantuan juruteknik makmal perubatan untuk menganalisis kekerapan micronuclei (MN). Sel mukosa buccal ialah lapisan dalam pipi dan bibir, di mana ia merangkumi semua lapisan membran mukus permukaan dalaman pipi dan bibir, dari garis sentuhan bibir lawan ke garis lampiran mukosa ke alveolar ridges dan pterygomandibular raphe yang merangkumi kawasan antara 50-60cm². (Kamus Perubatan Mosby, Edisi ke-9, 2009). Sel mukosa buccal yang

dikumpul akan diwarnakan dan kemudian, slaid akan diperiksa pada pembesaran 1000X untuk analisis mikroskop (Ceretti et al., 2014). Kekekapan Micronuclei akan dikira dengan menggunakan kaedah pemarkahan. (Thomas, Holland, Bolognesi et al, 2009).

4. SIAPA YANG TIDAK BOLEH MENYERTAI KAJIAN INI?

Kanak-kanak sekolah rendah yang bukan warganegara Malaysia, didiagnos menghidap penyakit kanser dan mengalami sebarang penyakit pernafasan. Selain itu, kanak-kanak yang telah menjalani radioterapi atau kemoterapi dalam 12 bulan sebelumnya atau X-ray dalam 3 bulan sebelumnya tidak akan diambil dalam kajian ini.

5. APAKAH FAEDAH MENYERTAI KAJIAN INI?

a) KEPADA ANAK/JAGAAN SAYA SEBAGAI PESERTA?

Subjek ini akan mendapat maklumat tentang status kesihatan mereka dan kemungkinan risiko yang timbul daripada pendedahan $PM_{2.5}$ dan PM_{10} yang dilepaskan ke udara oleh kilang minyak kelapa sawit yang berdekatan. Risiko akan ditentukan oleh kekerapan micronuclei (MN) daripada sel bukal mukosa.

b) KEPADA PENYELIDIK?

Maklumat dan data yang dikumpul akan digunakan oleh penyelidik untuk menganalisa data. Data daripada kajian ini juga boleh digunakan sebagai rujukan untuk penyelidikan masa depan. Selain itu, kajian ini juga boleh membuat data asas dan rujukan untuk penggunaan penyelidik masa depan apabila menghadapi masalah yang sama.

6. ADAKAH IA BERISIKO?

Terdapat risiko yang rendah apabila mengumpul sampel biologi, oleh itu juruteknik makmal perubatan akan membantu apabila mengumpul sel mukosa buccal di kalangan kanak-kanak sekolah rendah.

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

Ya, maklumat yang diberikan dan identiti akan disimpan sebagai sulit. Kajian ini adalah bebas dan terbuka kepada semua kanak-kanak berumur 9 tahun hingga 11 tahun yang akan di jalankan di Sekolah Kebangsaan Jenis Tamil (SJKT) Ladang Rinching, Semenyih dan Sekolah Kebangsaan Bandar Rinching di mana terdiri daripada kumpulan terdedah dan kumpulan perbandingan yang belajar di Sekolah Kebangsaan Dusun Tua Hulu Langat dan Sekolah Kebangsaan Kelubi Hulu Langat.

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

Jika terdapat sebarang pertanyaan dan kekeliruan, anda boleh menghubungi:

- i). Penyelia Kajian : Associate Professor Dr Juliana Jalaludin
Jabatan Kesihatan Persekitaran dan
Pekerjaan
Fakulti Perubatan dan Sains Kesihatan
No. Pejabat : 03 8947 2401 / 03 8947 2397
H/P No : 017-6834103
Email : juliana@upm.edu.my
- ii). Penyelidik Kajian: Syhidatul Farhana binti Othman
H/P No : 0182946300
Email : syhidatulfarhana@gmail.com

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

9. PERSETUJUAN

Saya..... No Kad Pengenalan.
.....beralamat.....
.....d
engan ini secara sukarela 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 alasan. 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**

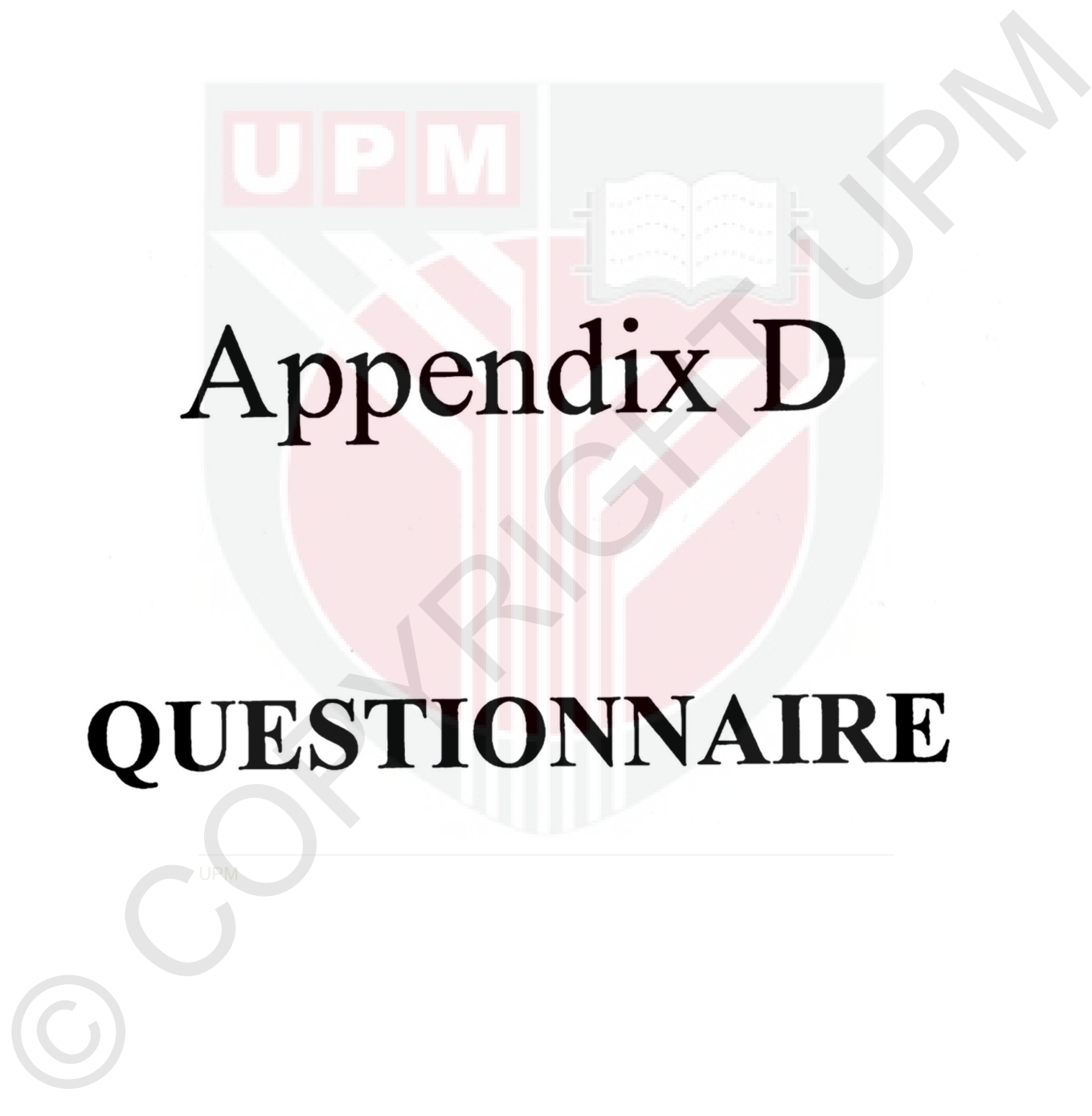
Tandatangan Tandatangan

.....
(Ibubapa/ Penjaga)
(Saksi)

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

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

**Tarikh Tandatangan
(Penyelidik)**

The image features a large, faint watermark of the Universiti Putra Malaysia (UPM) logo in the background. The logo is a shield-shaped crest with a red and white color scheme. It includes a book icon at the top, a stylized 'UPM' acronym in the center, and a banner at the bottom. The text 'Appendix D' and 'QUESTIONNAIRE' are prominently displayed in the foreground, centered over the watermark.

Appendix D

QUESTIONNAIRE

ID NO:	
TARIKH :	



**JABATAN SAINS KESIHATAN
PERSEKITARAN DAN PEKERJAAN,
FAKULTI PERUBATAN DAN SAINS
KESIHATAN, UNIVERSITI PUTRA
MALAYSIA, 43400 UPM SERDANG,
SELANGOR MALAYSIA**

BORANG KAJI SELIDIK

TAJUK KAJIAN	HUBUNG KAIT DI ANTARA PENDEDAHAN KEPADA PM _{2.5} DAN PM ₁₀ DAN KEROSAKAN DNA DALAM SEL BUKAL MUKOSA DI KALANGAN KANAK-KANAK SEKOLAH RENDAH BERHAMPIRAN KILANG MINYAK KELAPA SAWIT DI SEMENYIH.
NAMA PENYELIDIK	SYHIDATUL FARHANA BINTI OTHMAN

Arahan:

Borang kaji selidik ini mempunyai 8 bahagian:

1. Bahagian A: Maklumat Peribadi Kanak-Kanak Dan Keluarga
2. Bahagian B: Maklumat Persekitaran Dalam Rumah
3. Bahagian C: Maklumat Persekitaran Luar
4. Bahagian D: Maklumat Taraf Kesihatan Kanak-Kanak
5. Bahagian E: Penyakit-Penyakit Lain
6. Bahagian F: Tabiat Pemakanan Kanak-Kanak
7. Bahagian G: Sejarah Kesihatan Keluarga
8. Bahagian H: Kebenaran Mengambil Sampel Di Rumah Kanak-Kanak

Kanak

Semua maklumat yang diberikan adalah rahsia dan akan digunakan untuk tujuan pengajian sahaja. Sila jawab semua soalan yang berkaitan. Terima kasih atas kerjasama anda.

BAHAGIAN A: MAKLUMAT PERIBADI KANAK- KANAK DAN KELUARGA

1. No. Responden:

2. Nama kanak-kanak :

3. Nama Ibu kanak- kanak:

4. Alamat :

5. No. Telefon (Pejabat): _____ No. Telefon (Rumah): _____

6. Tinggi: cm Berat: . kg

7. Umur : tahun

8. Tarikh Lahir : hari bulan tahun

9. Bilangan adik beradik: _____ (orang)

10. Jantina : ☐ Lelaki ☐ Perempuan

11. Bangsa : ☐ Melayu ☐ Cina ☐ India ☐ Lain-
Lain

12. Tahun persekolahan: _____

13. Nama Sekolah: _____

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: _____
18. Pendapatan Bapa: RM _____
19. Pendapatan Ibu: RM _____
20. Jumlah pendapatan isi rumah : RM _____
21. Tempat kanak- kanak dilahirkan:
Negeri _____ Bandar _____
22. Sudah berapa lama anda tinggal di alamat sekarang? _____
tahun _____ bulan _____
23. Sila senaraikan tempat- tempat di mana anda pernah tinggal selama 6 bulan atau lebih sejak lahir hingga sekarang.
- a) _____

- b) _____

BAHAGIAN B: MAKLUMAT PERSEKITARAN DALAM 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

☐

Berkongsi dengan 3 orang ☐

Berkongsi dengan ≥

4 orang

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 sehari

6. Semasa anda memasak, adakah anda membuka tingkap atau pintu 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 _____(nyatakan)

8. Adakah anda mempunyai binatang 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 _____(nyatakan)

- 10b. Berapaka kalikah anda menggunakannya dalam seminggu?

_____ kali seminggu

- 10c. Dimanakah ianya ditempatkan di dalam rumah?

☐ Di ruang tamu sahaja ☐ Di bilik tidur ☐ Bilik tidur dan ruang tamu

11. Adakah terdapat sesiapa/ahli keluarga yang merokok di persekitaran dalam rumah? (bermaksud sekurang kurangnya 1 batang rokok sehari atau 1 aun tembakau dalam masa satu bulan) jika 'TIDAK', teruskan ke no. 14, jika 'YA', teruskan ke no.12 .

☐ Ya ☐ Tidak

12. Senaraikan individu yang merokok di dalam rumah

☐ Suami ☐ Bapa
☐ Anak Lelaki ☐ Bapa Mertua ☐ Lain-lain

13. Berapa batang rokokkah yang dihisap oleh individu di atas (di persekitaran dalam rumah anda sahaja)?

_____ batang sehari

14. Apakah alat yang digunakan untuk membersihkan rumah anda?

Sila nyatakan: _____

15. Berapa kerapkah dalam seminggu anda membersihkan rumah anda?

_____ kali seminggu

16. Adakah anda menggunakan karpet di kediaman anda

☐ Ya ☐ Tidak

BAHAGIAN C: MAKLUMAT PERSEKITARAN LUAR

Arahan: Sila tandakan(/) pada kotak ☐ tersebut.

1.	Bahan binaan rumah anda.		
	Batu/simen	☐	Kayu/papan

	Lain-lain (sila nyatakan)			
2.	Jenis kawasan perumahan:			
	Kampung		Rumah Teres dua tingkat	
	Rumah Teres Setingkat		Banglo	
	Flat			
3.	Lokasi rumah dari jalanraya:			
	Kurang 100 meter dari jalanraya		Lebih 500 - 1000 meter dari jalanraya	
	Lebih 100- 500 meter dari jalanraya		1000 meter dari jalanraya	
4.	Lokasi rumah dari kawasan industri			
	Kurang 500 meter dari kawasan industri		Lebih 1.5 – 5 kilometer dari kawasan industri	
	Lebih 1- 1.5 kilometer dari kawasan industri		Lebih 5 kilometer dari kawasan industri	
5.	Lokasi rumah anda dari kilang kelapa sawit			
	Kurang 500 meter dari kilang		Lebih 1.5- 3 kilometer dari kilang	
	Lebih 1 – 1.5 kilometer dari kilang		Lebih 3 kilometer dari kilang	
6.	Apakah pendapat anda mengenai persekitaran rumah anda?			
	Sangat berhabuk		Kurang berhabuk	

	Sederhana berhabuk		
7.	Apakah kenderaan yang sering anda gunakan sebagai pengangkutan utama anda?		
	Kereta		Berjalan Kaki
	Basikal		Motosikal
	Bas		

Soalan –soalan berikut merupakan soalan – soalan mengenai taraf kesihatan di bahagian dada anda, sila beri jawapan samada **YA** atau **TIDAK** jika anda tahu jawabannya. Jika didapati soalan tersebut tidak merujuk kepada anda, sila tandakan pada bahagian **TIDAK BERKENAAN**. Jika sekiranya anda ragu-ragu samada jawabannya **YA** atau **TIDAK**. Sila tindakan **TIDAK**

BAHAGIAN D: MAKLUMAT TARAF KESIHATAN KANAK-KANAK

	BATUK	YA	TIDAK
1.	Adakah anak anda selalu mengalami batuk berserta selesema?		
2.	Adakah anak anda mengalami batuk sahaja?		
	2a. Jika YA, adakah anak anda batuk pada keseluruhan hari (4 hari atau lebih dalam masa seminggu atau selama 3 bulan berturut-turut dalam masa setahun)		
	2b. Sudah berapa tahunkah anak anda mengalami batuk seperti ini? _____ tahun		
	KAHAK	YA	TIDAK
1.	Adakah anda selalu mengalami kesesakan nafas serta mengeluarkan kahak dan juga mengalami selesema?		
	1a. Jika YA, adakah anak anda mengalami kesesakan nafas dan mengeluarkan kahak pada keseluruhan (4 hari atau lebih dalam masa		

	seminggu) selama 3 bulan berturut-turut dalam masa setahun?		
	1b. Sudah berapa lamakah anak anda mengalami masalah batuk seperti ini? _____ Bulan /tahun		
2.	Adakah anak anda pernah mengalami serangan batuk, kesesakan nafas atau kahak dalam masa seminggu atau lebih dalam masa setahun?		
	Jika YA , untuk soalan 3 diatas, sila jawab soalan berikut:		
	2a. Sudah lamakah masalah ini berlaku? _____ Bulan /tahun		
	2b. Secara purata, berapa kalikah masalah diatas dalam masa setahun? _____ Bulan /tahun		
	DADA BERBUNYI/WHEEZING	YA	TIDAK
1.	Adakah anda mengalami masalah pernafasan berbunyi di dada?		
	1a. Apabila anda mengalami selsema?		
	1b. Kadangkala walaupun tidak mengalami selsema		
	1c. Hampir setiap hari (waktu siang dan juga malam)		
	Jika jawapan anda YA untuk soalan 1b dan 1c di atas sila jawabkan soalan berikut:		
	1d. Sudah berapa lamakah anda mengalami masalah ini(dada berbunyi) tahun ☐ Tidak berkenaan		
2.	Adakah anda pernah mengalami masalah dada berbunyi yang menyebabkan anda mengalami sesak nafas?		

	Jika YA bagi soalan 2 di atas sila jawab soalan berikut:		
	2a. Adakah anda memerlukan rawatan dan ubatan untuk masalah ini?		
	2b. Adakah anda boleh bernafas secara normal semasa mengalami serangan ini?		
3.	Adakah anda mengalami masalah ini setelah melakukan aktiviti seperti senaman atau latihan?		
	KESAKITAN DADA	YA	TIDAK
1.	Sejak 3 tahun lepas, adakah anda pernah mengalami kesesakan di bahagian dada yang menghalang anda daripada melakukan aktiviti biasa selama 3 hari?		
	Jika YA bagi soalan 1 sila jawab soalan berikut:		
	1a. Adakah anda mengeluarkan kahak atau mengalami kesesakan nafas lain daripada keadaan biasa selain daripada mengalami penyakit ini?		
	1b. Berapa kali anda mengalami penyakit seperti berikut dalam 3 tahun lepas?		
	Kurang daripada sekali dalam setahun ☐ Sekali dalam setahun ☐ 2-5 kali dalam masa setahun ☐ Lebih 5 kali dalam setahun ☐ Tidak berkenaan		
	1c. Berapa kalikah penyakit seperti ini berlaku sekurang-kurangnya selama 7 hari? kali ☐ Tidak berkenaan		
2.	Adakah anda pernah dimasukkan ke hospital kerana mengalami masalah jangkitan di dada sebelum berumur 2 tahun?		

--	--	--	--

BAHAGIAN E: PENYAKIT-PENYAKIT LAIN

1.	Adakah anda mengalami penyakit-penyakit seperti berikut? jika YA, pada umur berapakah anda didiagnoskan mengalami penyakit berikut?	YA	TIDAK
	a) Campak Umur didiagnoskan.....		
	b) Bronkitis Umur didiagnoskan.....		
	c) Emfisema Umur didiagnoskan.....		
	d) Asma(lelah) Umur didiagnoskan.....		
	e) Pneumonia (jangkitan paru-paru) Umur didiagnoskan.....		
	f) Lain-lain nyatakan..... Umur didiagnoskan.....		
2.	Adakah doktor pernah mengatakan bahawa anda menghidap asma?		
	Jika jawapan anda YA bagi soalan 2 sila jawab soalan-soalan berikut		
	2a. Pada umur berapakah anda mengalami asma?tahun		
	2b. Adakah sekarang anda masih mengalami asma?		
	2d. Pada umur berapakah anda disahkan tidak mengidap asma lagi?bulan/tahun		

ALLERGI/ALAHAN

1. Adakah doktor pernah menyatakan bahawa anda mengalami alahan kepada makanan atau ubatan tertentu?

☐ Ya hanya kepada makanan tertentu sahaja, nyatakan.....

☐ Ya hanya kepada ubatan tertentu sahaja, nyatakan.....

☐ Ya kepada ubatan dan makanan tertentu, nyatakan.....

☐ Tidak

2. Adakah doktor pernah menyatakn bahawa anda mengalami alahan kepada debu?

☐ Ya

☐ Tidak

3. Adakah doktor pernah menyatakan bahawa kulit anada mengalami alahan kepada detergen atau bahan kimia tertentu?

☐ Ya

☐ Tidak

4. Adakah anda mengambil suntikan untuk mengurangkan masalah alahan tersebut?

☐ Ya

☐ Tidak

BAHAGIAN F: TABIAT PEMAKANAN KANAK-KANAK

Berapa kerapkah anak anda makan makanan yang diberikan?	Tidak pernah	Jarang sekali	Sekali seminggu	Lebih dari sekali seminggu	Hampir setiap hari
Berapa kerapkah anak anda makan daging?					
Berapa kerapkah anak anda makan ikan?					
Berapa kerapkah anak anda makan daging yang diproses?					
Adakah anak anda makan makanan					

bertin? (canned food)					
Berapa kerapkah anak anda makan makanan yang bergoreng ?					
Berapa kerapkah anak anda makan kerepek kentang/ kentang goreng?					
Berapa kerapkah anda makan makanan segera?					
Berapa kerapkah anak anda makan coklat/gula-gula?					
Berapa kerapkah anak anda minum air bergas/ bikarbonat/ soda?					
Adakah anak anda mengambil makanan tambahan seperti vitamin atau jus. Jika YA nyatakan:					

BAHAGIAN G: SEJARAH KESIHATAN KELUARGA

Adakah diantara ahli keluarga anda yang lain mengalami masalah-masalah berikut

	SUAMI	YA	TIDAK	TIDAK TAHU
a)	Bronkitis kronik			
b)	Emfisema			
c)	Asma(lelah)			
d)	Barah paru-paru			
e)	Lain-lain masalah paru-paru			
	ANAK-ANAK			
a)	Bronkitis kronik			
b)	Emfisema			
c)	Asma(lelah)			
d)	Barah paru-paru			
e)	Lain-lain masalah paru-paru			

BAHAGIAN H: KEBENARAN MENGAMBIL SAMPEL DI RUMAH KANAK KANAK

Sampel dua jenis habuk (inhalable dust PM10 dan fine particle PM2.5) akan diambil di rumah kanak-kanak untuk mengukur pendedahan kanak-kanak di rumah. Pengambilan sampel ini akan dibuat dengan menggunakan Escort LC Personal Sampling Pump dan kertas turas dengan liang bersaiz 5µm. Pam tersebut akan ditinggalkan selama 24 jam di rumah kanak-kanak dan akan diletakkan pada paras bernafas bagi kanak-kanak tersebut. Pengambilan sampel di rumah kanak-kanak tidak akan mengganggu aktiviti seharian di dalam rumah kanak-kanak.

Persetujuan Ibu Bapa / Penjaga

Saya..... No Kad Pengenalan.
.....beralamat.....
.....d
engan ini secara sukarela membenarkan / tidak membenarkan penyelidik
untuk mengambil sampel di rumah saya.

Tandatangan
(Ibubapa/ Penjaga)

No. Telefon:

Tarikh :

*Terima kasih atas kerjasama anda...../Thank you for your
cooperation.....*

Appendix E
GANTT CHART

UPM

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GANTT CHART AND MILESTONE

[illegible]

A large, light gray watermark of the UPM logo is centered on the page. The logo features a shield with a red and white striped field, a red cross, and a red banner at the top with the letters 'UPM'.

Appendix F

RESEARCH

PHOTOGRAPH



Figure 12: Sampling of particulate matters (PM₁₀ and PM_{2.5}) at respondent's house

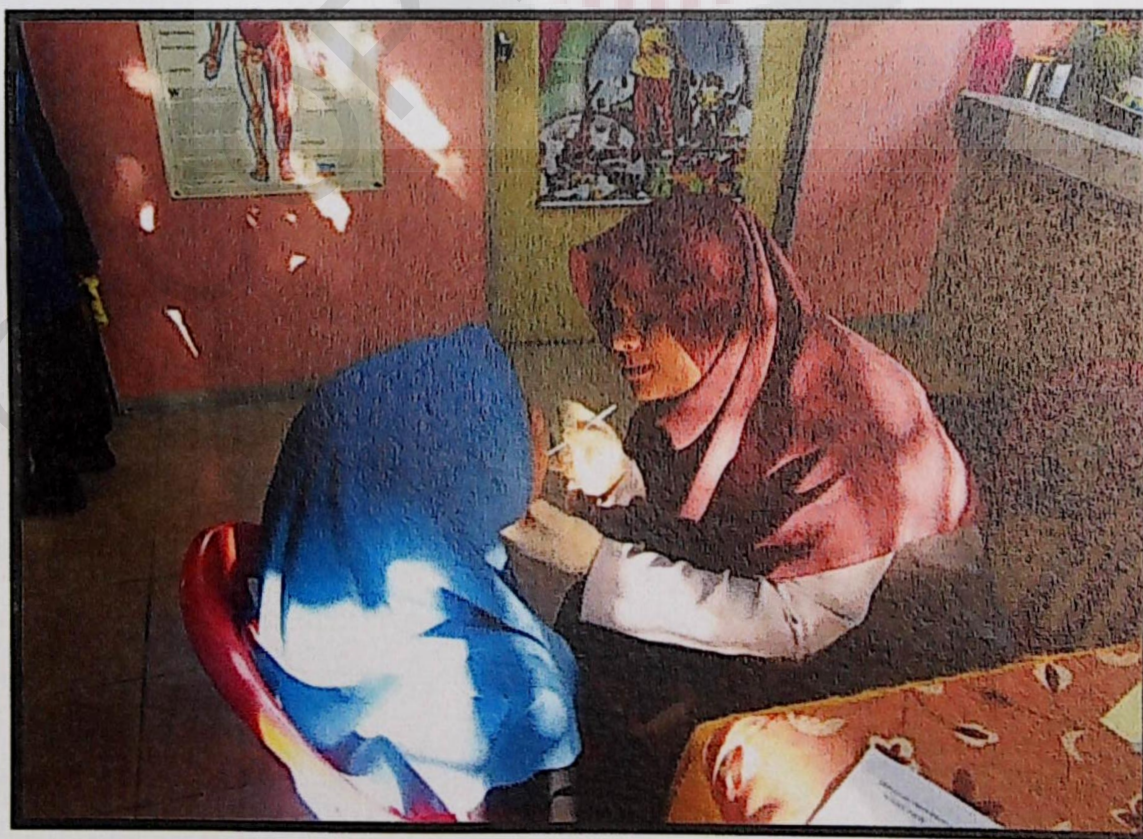


Figure 13: Collection of buccal mucosa cells