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The Effects of Indoor Pollutants Exposure on Allergy and Lung Inflammation: An Activation State of Neutrophils and Eosinophils in Sputum

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Abstract

Background: To explore the inflammation phenotypes following indoor pollutants exposure based on marker expression on eosinophils and neutrophils with the application of chemometric analysis approaches. Methods: A cross-sectional study was undertaken among secondary school students in eight suburban and urban schools in the district of Hulu Langat, Selangor, Malaysia. The survey was completed by 96 students at the age of 14 by using the International Study of Asthma and Allergies in Children and European Community Respiratory Health Survey questionnaires. The fractional exhaled nitric oxide was measured, and an allergic skin prick test and sputum induction were performed for all students. Induced sputum samples were analysed for the expression of CD11b, CD35, CD63, and CD66b on eosinophils and neutrophils by flow cytometry. The particulate matter PM_2, NO_2, CO_2, and formaldehyde were measured inside the classrooms. Results: Chemometric and regression results have clustered the expression of CD63 with $PM_{2.5}$, CD11b with NO_2, CD66b with FeNO levels, and CO_2 with eosinophils, with the prediction accuracy of the models being 71.88%, 76.04%, and 76.04%, respectively. Meanwhile, for neutrophils, the CD63 and CD66b clustering with $PM_{2.5}$ and CD11b with FeNO levels showed a model prediction accuracy of 72.92% and 71.88%, respectively. Conclusion: The findings indicated that the exposure to $PM_{2.5}$ and NO_2 was likely associated with the degranulation of eosinophils and neutrophils, following the activation mechanisms that led to the inflammatory reactions.

Keywords:

lung inflammation; **allergy**; **indoorpollutants**; **biomarkers**; **FeNO** ; **eosinophil**; **neutrophil**

Introduction

Exposure to indoor pollutants are strongly associated with increased morbidity and mortality, mainly among school children who spend most of their time in the classrooms [1]. Reports have shown that exposure to high concentration of particles, nitrogen dioxide , carbon dioxide , ozone , volatile organic compounds and fibres induce persistent airway inflammation, which is mediated by the immune system [2,3]. Biomarkers, such as fractional exhaled nitric oxide (FeNO), cytokines, chemokines, lipid mediators, enzymes, adhesion molecules, and other growth factors, have been considered as the indicators of allergic airway inflammation [4]. The indicators that underlie the complex molecular pathways that regulate inflammation have not been fully elucidated; however, numerous studies have reported that the process certainly involves the activation of eosinophils and neutrophils [5,6]. Extensive investigation of the mediators that are implicated in allergy, lung inflammation, and asthma has been documented but there was insufficient evidence available for the multi-dimensional characters of the activation and degranulation markers expression of CD11b, CD35, CD63, and CD66b on eosinophils and neutrophils.

Materials and Methods

2.1. Study Population

The study population was randomly sampled from eight secondary schools in Hulu Langat, Selangor, Malaysia. The researchers of this study targeted school children at the age of 14 of which they were randomly selected from four classrooms in each school. The total number of students who received their guardian's consent and was thus recruited in the study was 470. Among them, only 50 (10.6%) students were diagnosed with asthma by a doctor based on the survey questionnaire. Another 46 students out of the remaining 420 students were randomly selected as a potential control group. The control group was selected among students who produced an adequate sputum cell count from the same class and school. In total, 96 students were included in the final study group. Students with a history of smoking in the last 12 months and students who received antibiotic treatments in the past four weeks were excluded from this study. The school areas were classified as urban and suburban by the Ministry of Education, Malaysia, based on the locale classification of the ecological measures.

Results

3.1. Data Analysis for the Personal and Clinical Characteristics of School Children

The study population was well-balanced between the doctor-diagnosed asthmatic children (52%) and healthy children (48%), in which 60% of them were from urban schools. The majority of asthmatic children tested positive for at least one of the allergens, with 74.0% of them sensitised towards house dust mites, followed by cat dander (30.0%). The FeNO levels were statistically higher among the doctor-diagnosed asthmatic children than healthy children ($p < 0.001$). The researchers observed that the total percentage of eosinophil count in the sputum samples was slightly higher in the doctor-diagnosed asthmatic children but not statistically different between the two groups ($p > 0.05$)

Discussion

This work is partly presented at Joint Event on 24th World Congress on Pharmacology & 7th World Heart Congress, August19- 20, 2019 at Vienna, Austria

The role of biomarkers in airways is complex and specific, which is helpful in evaluating the aetiology, characterisation of phenotyping, and treatment of allergy and lung inflammation [37]. In this study, the FeNO levels were significantly higher among asthmatic school children, which are similar with the studies conducted in China [38], Terengganu, Malaysia [39], and Penang, Malaysia [40]. The result showed that there was inflammation in the airways and the average value was above the threshold of 50 ppb, which could reflect a high degree of inflammation. Liu et al. [41] and Carlsen et al. [42] reported that there was a significantly positive relationship between the FeNO levels and almost all pollutants, namely PM_{10}, $PM_{2.5}$, SO_2, NO_2, CO, and VOCs. This advocates a relationship between the high levels of all pollutants measured inside the classroom of urban schools and the high levels of FeNO among school children in this study. The cluster approach used in this study, which is aimed at improving the interpretability of the data, interestingly revealed that the formaldehyde and NO_2 concentrations were in the same factor, with a total variance of 26.0%. This finding confirmed that the formation of formaldehyde was through the photochemical reactivity of NO_2 in the air with VOCs generating different aldehydes [84].

Conclusions

In conclusion, the chemometric analysis methods produced robust and immunologically meaningful results, which clustered the degranulation markers (CD11b and CD63) expressed on eosinophils with the concentration of NO_2 and $PM_{2.5}$. Besides that, the degranulation markers expressed on the neutrophils, CD63 and CD66b, were clustered together with the $PM_{2.5}$ concentration. A further prospective study is now obligatory to validate the models generated from this current study.

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