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SHORT COMMUNICATION

AN ASSESSMENT OF THE ALVEOLAR MACROPHAGE ACTIVITY IN DOGS

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SUMMARY

The phagocytic and intracellular killing activities of alveolar macrophages (AM) were assessed in five dogs of varying health status. The results indicated that the percentage of phagocytic activity remained unchanged irrespective of age and health status. However, the intracellular killing activity was much higher ($p<0.05$) in diseased dogs. The findings indicated the importance of AM activities as an aid in the diagnosis and research of pulmonary diseases.

Keywords: Alveolar macrophage, phagocytosis, intracellular killing

INTRODUCTION

By functioning as scavengers, the alveolar macrophage (AM) acts as the major defender of the lower respiratory tract against potentially noxious particle (Ohmann and Babuik, 1986). As member of the mononuclear phagocyte system, the AM play the role as accessory cells in immunologic responses (Shaw and Anderson, 1984).

Pulmonary fluid samples obtained by lavage have enabled studies of the humoral and cellular elements of the lower respiratory tract of humans (Hunningkake *et al.*, 1979). Similarly, several infectious and non-infectious respiratory diseases of dogs can be used as models in medical and comparative disease studies.

Our earlier findings have demonstrated the AM phagocytic and intracellular killing activities in dogs with respiratory disease (Hazilawati *et al.*, 1998). Thus, the purpose of the present study is to further verify the role of AM in healthy and diseased dogs.

MATERIALS AND METHODS

Animals

A total of five adult dogs were obtained from the Dog Unit, Dewan Bandaraya, Kuala Lumpur. The selection led to a total of three healthy (Y1, Y2, Y3) and two diseased (Y4, Y5) dogs being chosen. Dogs Y4 and Y5 were chosen based on the evidence of respiratory symptoms such as coughing and nasal discharge while the other three were clinically normal.

Bronchio-alveolar lavage and AM assay

The dogs were euthanised with an overdosage of barbiturate and the lungs were removed immediately under sterile condition. Prior to the procurement of the lavage, an assessment was made on the gross pathology of the lungs of all five dogs. The bronchio-alveolar lavage and acridine orange chemiluminescense AM assay was done as described earlier (Hazilawati *et al.*, 1998).

Data were subjected to a one-way ANOVA using the SPSS statistical package (Windows Release 7.5). Only means with a level of $p<0.05$ or less were considered as significant.

RESULTS AND DISCUSSION

Pathology

The lungs of dogs Y1, Y2 and Y3 appeared normal. The lungs of dog Y4 showed evidence of moderate anthracosis at the right apical lobe. A dark red and firm consolidated area was found at the right apical lobe of the lung of dog Y5.

While the lung lavage fluid of dogs Y1, Y2 and Y3 were normal, significant changes were observed in the lung lavage fluid of dogs Y4 and Y5. The upper layer of the sediment of Y4 was covered with a black sediment, indicating the presence of carbon. In addition to the lavaged PBS, approximately 5 mL of mucopurulent exudate was also retrieved from the lung of dog Y5, indicating presence of a subacute bacterial infection.

Alveolar macrophage phagocytic and intracellular killing activities

Figs. 1 and 2 show the AM intracellular killing activities. A greenish AM indicated a viable AM or bacterial cell while yellow denoted dead cells.

The mean percentages of phagocytic and intracellular activities of AM are presented in Table 1. While the phagocytic activity was insignificantly higher in the diseased dogs, the intracellular killing activity in was significantly ($p<0.05$) surpassed in diseased dogs.

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The findings of this study are comparable to those documented in humans exposed to complex smoke (Lantz *et al.*, 1994) and in animals exposed to wood smoke (Judith, 1999). In both studies, no significant changes were observed on the AM phagocytic activity during and post-induction. However, the intracellular killing activity was significantly suppressed at the time of induction and became activated thereafter. This led to a conclusion that the air pollutant, as seen in dog Y4, is able to suppress the AM intracellular killing activity at the time of induction. Since dog Y4 was only used after the induction period (based on the lesions observed at post-mortem) the AM had probably returned to its active stage. Ironically, continuous induction of marijuana smoke, interstitial lung disease (ILDS), pulmonary fibrosis, acute extrinsic allergic alveolitis and AIDS in humans resulted in the impairment of both activities of AM (Luppi *et al.*, 1998).

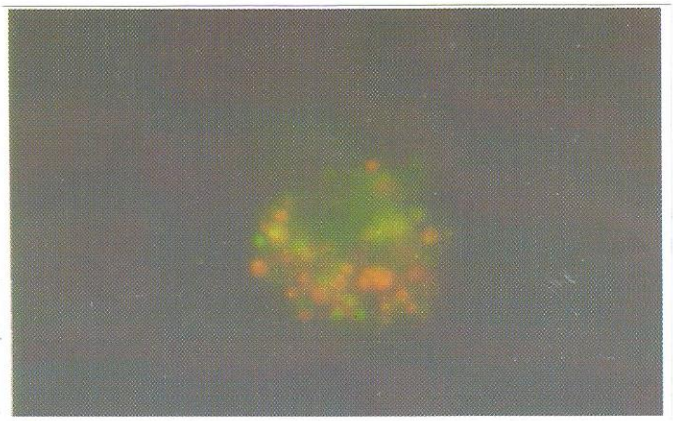

Fig. 1. Photomicrograph showing green macrophages and bacteria denotes viability while those in yellow are dead

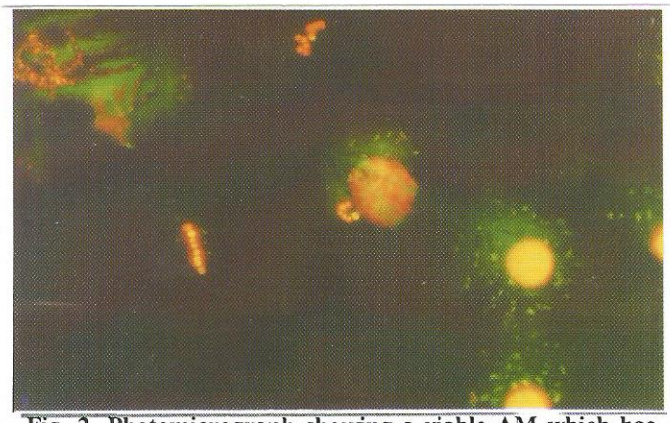

Fig. 2. Photomicrograph showing a viable AM which has successfully engulfed and killed bacteria

We believed that the activity of AM depends on the aetiology and time of onset of a particular condition. The discrepancy between the findings in humans (Baldwin *et al.*, 1996; Luppi *et al.*, 1998) to that reported here could be due to several reasons. Firstly, it was assumed that dog Y4 was not suffering from an infectious disease. Thus, the severity of suppression on the AM might not be fully expressed leading to a minimal effect on AM killing activity.

Table 1. The percentage of phagocytic and intracellular killing of alveolar macrophages

Animal	Status	Phagocytosis (%)	Intracellular killing (%)
Y1, Y2, Y3	Healthy	73.0 ± 4.5^{a}	37.6 ± 18.7^{a}
Y4, Y5	Diseased	66.3 ± 4.2^{a} ($p<0.19$)	81.3 ± 4.6^{b} ($p<0.05$)

Values bearing similar superscript do not differ significantly

Secondly, even though dog Y5 was infected, it was in the subacute stage of infection while the above mentioned conditions are chronic in nature (Baldwin *et al.*, 1996; Luppi *et al.*, 1998). This makes it difficult to compare with findings. Thirdly, the marijuana, ILDS, AIDS and others might have direct immunosuppressive effects on the pulmonary system *per se* (Baldwin *et al.*, 1996; Luppi *et al.*, 1998). This will further suppressed the AM to perform at its fullest in the latter condition. The lesions observed in dog Y5 appeared to be well under control and possibly the immunosuppressive effects were minimal, leading to an un-compromised AM functions. Finally, it is possible that AM of the diseased dogs used on the study were already in a sensitised state. In such a state, AM are always on the alert and will react promptly and effectively.

Factors such as the dose and particle size will also determine the efficacy of the AM intracellular killing ability. Continuous administration or exposure to the agent pollutant can cause suppression of both AM activities. Nevertheless, a single exposure to a high dose will lead to the impairment of the AM intracellular killing ability, or even killing the AM directly. However, exposure to low dose for a prolonged period can lead to suppression of both AM activities.

The study showed that the determination of AM activities can be a useful diagnostic tool to diagnose the state of lung diseases in both human and animals especially in the field of environmental pathology.

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RINGKASAN

PENILAIAN AKTIVITI MAKROFAJ ALVEOLUS DALAM ANJING

Aktiviti pembunuhan fagosit dan intrasel makrofaj alveolus (AM) telah dinilaikan dalam lima ekor anjing pelbagai status kesihatan. Hasil kajian menunjukkan peratusan aktiviti fagosit kekal tidak berubah, tidak kira umur atau status kesihatannya. Bagaimanapun, aktiviti pembunuhan intrasel adalah lebih tinggi ($p < 0.05$) dalam anjing berpenyakit. Penemuan ini menunjukkan pentingnya aktiviti AM dalam membantu diagnosis dan penyelidikan penyakit pulmonari.